Developing through relationships with the physical and social environment: disentangling the transition from co-regulation to self-regulation

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Abstract

The present thesis centres its attention on early regulatory skills of physiological arousal and attention. These abilities play a central role during early infancy and are crucial for achieving autonomy and establishing the foundation for later social, behavioural, and cognitive development.

Early in development, infant's self-regulatory capacities are thought to be immature, poorly coordinated, and limited. Because of this, co-regulation with the caregiver is particularly important. Much of the infant/ child research emphasizes the developmental increase in self-regulation and highlights a gradual transition from co-regulation (where regulatory processes are shared between child and caregiver) to self-regulation. Research on this transition, however, is scarce and complex and remarkably little is known on how these co-regulatory dynamics between infant-caregiver change and evolve over time.

Taking a novel, multi-method approach that integrates neural, physiological, and behavioural techniques and uses a mixture of home- and naturalistic lab-based research, the present thesis examines the development of self- and co- regulatory processes in infancy. More specifically, it explores whether infants' physiological and attentional states gradually become less dependent on others as they get better at self-regulation over developmental time.

Evidence is presented showing developmental changes in the way environmental factors (both physical and social) influence infants' regulation of physiological arousal and attention. Evidence also shows that, contrary to our hypothesis, dyadic strategies, rather than being phased out or replaced, seem to continue to play an important role. Discussion is focused on the contribution of the findings to theories of the development of dyadic regulatory process, and in identifying new and more naturalistic ways to study them.

Key words: regulation, early development, parent-infant dyad, dynamic system theory, naturalistic research, physiology, arousal, attention, EEG.

Declaration

This thesis focusses mainly on data collected in one large longitudinal study, "Oscillatory Neural and Autonomic Correlates of Social Attunedness (ONACSA)", headed by Sam Wass and funded by the European Research Council Horizon 2020 853251. In chapter 4, however, the data from the 10-months-old infants belong to another study, "Studying the Microdynamics of Social Interaction (SMSI)", again headed by Sam Wass but funded by the Leverhulme Trust RPG-2018-281.

During my first year as a PhD student, together with another PhD student, James Ives, we planned and piloted the design of the lab paradigms intended for data collection in the ONACSA project. The Ethics application was written and submitted by James Ives (see APPENDIX D). I, instead, was responsible for the production, design, and piloting of the wearable device described in the relevant Methods section (3.3). I also organised the questionnaires and consent forms that needed to be administered throughout the ONACSA project and created our main data management documents.

We started data collection at the end of our first year. After this, three new PhD students joined the project, Emily Greenwood, Pierre Labendzki and Narain Viswanathan. Alongside James, I was responsible for onboarding and training them on data collection and management. In February 2023, after having managed more than 100 lab and home visits, I stopped collecting data as two new PhD students, Tom Northrop and Giovanni Esposito, joined the project and assumed responsibility for it.

In chapter 3, Giovanni Esposito, Emily Greenwood, Pierre Labendzki and Einari Vaaras were responsible for training and running the Voice Type Classifier I employed in my analyses. In chapter 4, the data from the 10-months-old infants was from the SMSI project. These data were collected by Emily Phillips, Megan Whitehorn and Ira Marriott Haresign. The 10-months-old EEG data in chapter 4 was pre-processed by Ira Marriott-Haresign. I then used their scripts to pre-process the 5-months-old EEG data in chapter 4. The coding of the behavioural data for 10-months-old infants was overviewed by Megan Whitehorn. I, instead, organised the rest of the behavioural coding included in chapters 4 and 5, and was responsible for the training and supervision of the coders.

All the data analyses, as well as the formulation and testing of the hypotheses and the writing of the text, was entirely my own work.

Impact of this thesis

Chapter 3 is undergoing the review process for potential publication in *Child Development*:

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- Ives, J., Labendzki, P., Perapoch Amadó, M., Greenwood, E., Viswanathan, N., Northrop, T., & Wass, S. [preprint]. At which low amplitude modulated frequency do infants best entrain? A frequency tagging study. bioRxiv. https://www.biorxiv.org/content/10.1101/2022.12.08.519576v1
- Wass, S. V., Perapoch Amadó, M., & Ives, J. (2022). Oscillatory entrainment to our early social or physical environment and the emergence of volitional control. *Developmental Cognitive Neuroscience*, 54. https://doi.org/10.1016/j.dcn.2022.101102

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Abbreviations

ACF	Autocorrelation Function
ADHD	Attention Deficit Hyperactivity Disorder
BH	Benjamini and Hochberg
BPM	Beats Per Minute
CBP	Cluster Based Permutation
ECG	Electrocardiography
EEG	Electroencephalography
ESCS	Early Social-Communication Scales
FDR	False Discovery Rate
Fig	Figure
fMRI	Functional Magnetic Resonance Imaging
fNIRS	Functional Near-Infrared Spectroscopy
fps	Frames Per Second
GPS	Global Positioning System
HR	Heart Rate
Hz	Hertz
IBI	Inter Beat Interval
ICA	Independent Component Analysis
IJA	Initiating Joint Attention
IRR	Inter Rater Reliability
JA	Joint Attention
LC	Locus Coeruleus
М	Mean
min	Minutes
p-val	p-value
PSD	Power Spectral Density
r	Correlation coefficient
RF	Radio Frequency
RJA	Responding Joint Attention
sec	Seconds
SEM	Standard Error of the Mean
SER	Speech Emotion Recognizer
SM	Supplementary Materials

std	Standard Deviation
SWC	Sliding Window Correlation
UK	United Kingdom
UTC	Coordinated Universal Time
VTC	Voice Type Classifier
WCC	Window Cross Correlation
β	Beta coefficient
5m	5 months
10m	10 months
15m	15 months

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Chapter 1. Thesis overview

This thesis aims to examine the development of self- and co-regulatory processes in infancy. The primary goals were twofold: (1) to contribute to the existing literature by addressing current gaps, such as recognising a more active role of the infant, shifting away from static methodologies, and enhancing the ecological validity of the research, and (2), to expand our understanding of the early developmental trajectories of self- and co-regulation, with a specific emphasis on the regulation of physiological and of attentional states. To do so, the research presented in this thesis employs a multi-method and longitudinal approach that integrates neural, physiological, and behavioural techniques and uses a mixture of home- and naturalistic laboratory-based research. The thesis incorporates three separate empirical analyses (chapters 3 to 5) preceded by a General Introduction (chapter 2) and followed by a General Discussion (chapter 6).

The General Introduction in Chapter 2 presents a review on the current status of understanding of the development of these early physiological arousal and attentional regulatory skills, outlining the areas of theoretical and empirical debate and highlighting gaps in knowledge. The chapter concludes with a summary of the research objectives that drove the work undertaken for this thesis, reflecting on the unique contributions this work will bring to the field.

Chapter 3 focuses on regulation of physiological arousal and presents research conducted in the home environment. More specifically, it examines how early parent–infant interpersonal regulatory processes occur and develop across the first year of life. Historically, our understanding of caregiver-infant dynamics has been held back by methodological challenges (see DePasquale, 2020; Hollenstein et al., 2017; Morales et al., 2018; Taipale, 2016; Wass & Jones, 2023). To address these limitations, wearable technologies capable of recording a range of parameters including heart rate, proximity, activity, and vocal data from infants and their mothers within their home environment, were developed. This chapter presents the data obtained from full-day recordings conducted with 57 mother-infant dyads at 5 months and 32 dyads at 10 months using these devices. It investigates how early inter-relationships between infants and their caregivers varied in response to negative and neutral vocalizations, evolved across developmental time, and associated with infant calming. Results are discussed in relation to the ongoing debate regarding whether these developmental variances primarily stem from infants' improved self-regulation or, rather, from enhanced caregiver-infant co-regulation.

Chapter 4 and 5, instead, focus on early attentional regulatory skills. Chapter 4 examines how infants develop the capacity to regulate their attention in real-world settings, and how it changes over developmental time. Our understanding in this area is limited because most of the research has studied how the infants passively respond to stimuli as they appear and disappear on-screen, following an experimenter-determined sequence. This differs in several important ways from how we pay attention in dynamic, interactive, real-world settings (Wass & Jones, 2023; Wass, 2014; Wass & Goupil, 2022). This chapter presents data collected from 58 infants (aged 5- or 10-months) while they played alone with toys in a naturalistic play setting while concurrent gaze behaviour, physiology and electroencephalography (EEG) were recorded. It examines how moment-by-moment changes in physiology and cortical neural activity inter-relate in determining how infants spontaneously allocate attention in real-world settings. The chapter ends with a discussion on how these inter-relationships changed across development.

Chapter 5 follows from these findings to study the influences of social partners on such developing ability of the infants. More specifically, it examines how the ability to share attention with a social partner is established between parents and infants in free-flowing interactions. To do so, it presents behavioural data from N=48 infants at 5 and 15 months and their mothers that was collected during a free-flowing tabletop toy play. The chapter

explores how leader-follower dynamics within the dyad, as well the sensitivity of one partner to changes in behaviours generated by the other partner, evolve over time to create moments of shared attention. Results are discussed in relation to an increase in symmetrical patterns of attention co-regulation at the expense of unilateral.

In the General Discussion of Chapter 6, the three empirical chapters are summarised and integrated within the broader literature. Next, it follows a discussion of both the limitations and strengths of the work and concluding remarks are considered at the end of the chapter.

Chapter 2. General introduction

The present thesis centres its attention on early regulatory skills of physiological arousal and of cognition, more especially, attention. These abilities are thought to play a central role during early infancy and are crucial for achieving autonomy and establishing the foundation for later social, behavioural, and cognitive development. The main aim of this chapter is to present the current status of understanding of the development of these early arousal and attentional regulatory skills, outline areas of theoretical and empirical debate and highlight gaps in knowledge. The chapter concludes with an outline of the research objectives that stimulated the work conducted for this thesis.

2.1 Studying regulation

Scholars over time have increasingly focused their attention on regulation (Bell & Wolfe, 2004; Frick et al., 2017; Rothbart et al., 1992). Research has gradually acknowledged the importance of it in many key developmental milestones and it has been shown to be a positive predictor of a range of cognitive and socio-emotional relevant behaviours (Frick et al., 2019). Such behaviours include, but are not confined to, the ability to regulate negative emotions in response to distress, sustain attention, comply with demands, delay specific activities, and monitor one's own behaviour (e.g. Calkins, 2007; Rueda et al., 2021; Cheese et al., 2008). More broadly, it has been considered essential for life-long healthy as well as competent social and academic functioning (S. Calkins, 2007; Cole et al., 2019; N. Fox & Calkins, 2003; McClelland & Cameron, 2011; Ursache et al., 2012, 2013; Williford et al., 2013).

Because of its importance, the consequences of regulatory failures have been studied widely. Poor regulation has been linked to developmental problems such as, for example, attentional deficits (Nigg, 2017), behavioural maladjustment (S. Calkins, 2007), sleep

problems (Degangi et al., 2000), socio-emotional and academic difficulties (Dollar & Calkins, 2019) and psychopathology (Degangi et al., 2000; Eisenberg et al., 2009; Thompson, 2019). Accordingly, failures in adequate regulation can constrain subsequent development and compromise the child's response to later developmental challenges.

But what does the term 'regulation' mean? Despite its importance, the literature can sometimes be confusing and different authors have conceptualised it in different ways without an apparent consensus. Over time, the term has been understood through concepts like physiological arousal-, emotion-, mood- and affect-regulation (S. D. Calkins, 1997; Thompson, 2019); effortful control (Eisenberg et al., 2009; Rothbart et al., 2003), executive functioning and attentional control (Cole et al., 2019; Johansson et al., 2015; Rueda et al., 2005), cognitive control (Kopp, 1982), behavioural inhibition and impulsivity (Eisenberg et al., 2009), and temperament (Posner & Rothbart, 2000), amongst others (see Nigg, 2017, for more).

2.1.1 Narrowing down the concept

The current lack of consistency and conceptual integration makes it challenging to reach a shared definition. As seen above, some definitions of it are domain-specific (either distinguishing it from, or else equating it with, for example, regulation of emotion) or narrow (e.g. referred primarily to inhibitory processes). For the purposes of this thesis, we chose to consider regulation broadly as "the ongoing, dynamic, and adaptive modulation of internal states or behaviour" (Nigg, 2017). Importantly, the primary objective is "to attempt to enhance adaptation to changing circumstances (i.e. to achieve an explicit or implicit goal or goal state)" (Nigg, 2017).

There are three reasons why a definition like this is interesting. First, it allows us to differentiate regulation of and by oneself, also known as *self-regulation*, from regulation

of and by others, also known as *co-regulation*. Second, it avoids defining it exclusively as goal-directed behaviour (unlike e.g. Diamond & Aspinwall, 2003; Karoly, 1993; Thompson, 1994). This is especially important in infancy when individual goals are difficult to infer. Additionally, it introduces a challenge to the study of co-regulation as others (e.g. caregivers) may be able to infer the infants' state but not necessarily their goals. And third, it allows us to extend the term to both regulation *of* physiological arousal and *of* cognition. Of note, the idea of cognitive regulation, as derived from Nigg (2017), is not widespread. Nevertheless, in this thesis, it serves as a framework for understanding the "modification of attention to enhance adaptation (or achieve a goal) in the absence of salient emotion" (Nigg, 2017).

In the following sections, we focus on regulation *of* physiological arousal and *of* cognition, specifically focusing on attention. We consider these as two distinct components or subdomains of regulation and provide a review of the existing literature and current understanding of these.

2.2 Early regulatory skills of physiological arousal

"When there is a discrepancy between the current level of activation and the optimal level or range for the given situation, the organism will typically engage in behaviour designed to shift activation to reduce the discrepancy" (Fiske & Maddi, 1961 in Wass, 2021). Here we focus on the ability to modulate and to manage internal physiological arousal states for: (a) affective biological and social adaptations, and/ or (b) to achieve individual goals (Kopp and Neufeld (2003) in Eisenberg & Zhou, 2015) (though see limitations of studying point (b) in infancy above). Such skill is believed to play a critical role in supporting the development of academic and social-emotional skills and is thought to undertake rapid development throughout the first years of life. The next sections offer an overview of such rapid development but first, it is essential to establish a clear understanding of what is physiological arousal.

2.2.1 Understanding physiological arousal

In this thesis, physiological arousal, also referred to as 'arousal', is defined as the overall levels of activity within the autonomic nervous system (similar to Smith, 2021; Wass, 2017; Wass et al., 2016). Generally, research has suggested that there is an optimal (usually intermediate) level of activation towards which organisms strive, and any deviation from it (whether it be hypo- or hyper-activation) can result in aversive states for which individuals need to regulate (e.g. Aston-Jones & Cohen, 2005; Brazelton, 1978; Karmel et al., 2006; or see Wass (2017) for a review in the topic). Most of the research (including the current thesis) focuses on how individuals down-regulate arousal in response to hyperstimulation of the autonomic nervous system. More specifically, research has focused on the responses to higher ratios of sympathetic (activated for 'fight or flight') compared to parasympathetic (activated to 'rest and digest') autonomic nervous system activity. Nevertheless, it is noteworthy that regulating from hypo-arousal is equally significant because, as mentioned above, the optimal state is frequently identified as an intermediate level.

Throughout this thesis, heightened psychological arousal is generally treated as a proxy for "stress" and understood as an increased activity in the sympathetic nervous system compared to the parasympathetic nervous system. Similarly, regulation of physiological arousal is predominantly conceptualized as down-regulation.

In the context of developmental research with infants, electrocardiography (ECG) has overwhelmingly been the method of choice to collect heart rate data. Electrodes are placed on the infant's body to detect the electrical signals generated by the heart. ECG is

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particularly useful in developmental research because it is well tolerated and provides precise and reliable heart rate activity measurements.

2.2.2 Self-regulation of physiological arousal

Even from birth, infants show some capacity to act by themselves to maintain an optimal intermediate critical state. For example, even neonates are thought to have a tendency to close their eyes when overstimulated (Brazelton, 1978) or to respond to visual stimuli of varying complexity, movement, or novelty in ways that correspond to their individual arousal levels (Gardner & Karmel, 1984). Other behaviours such as thumb-sucking, proximity seeking, withdrawal, calming self-talk, gaze aversion, looking to mother or distraction with other objects have all been observed in early infancy and hypothesised as potential arousal regulatory behaviours (see Buss & Goldsmith, 1998; Feldman et al., 2011; Suata, 2023). Of note, only few studies have actually demonstrated that these regulatory behaviours alter arousal states (Buss & Goldsmith, 1998; Wass, 2021) (more on this in 2.2.5.1).

Overall, there is evidence of potential arousal regulatory behaviours being present at a very early age, however, infants are not born fully equipped to manage internal physiological arousal and accomplish their goals on their own. Instead, infants' capacities to regulate arousal are immature and poorly coordinated (Taipale, 2016; Tronick, 1989). Moreover, they are also limited. Firstly, because the effectiveness of their strategies is closely bound by the infant's arousal state. That is, heightened levels of arousal can easily compromise the infant's ability to manage discomfort (Eisenberg & Zhou, 2015; Kopp, 1989). And secondly, because infants can attempt to change an arousal state, but they can very rarely alter the cause of their own distress (Kopp, 1989). Considering these limitations and disruptions, a question arises: "why don't infants typically fail to achieve their goals and continuously experience dysregulation?" (Tronick, 1989).

2.2.3 Introducing the concept of co-regulation of physiological arousal

Luckily, infants do not develop in isolation (Cox et al., 2010; Diamond & Aspinwall, 2003; Wass, 2021). Instead, they are part of a wider system wherein their efforts towards managing physiological arousal and achieving goals are supported and complemented by other adults, such as caregivers. Many researchers have conceptualised this as coregulation, a process where one person serves an *'external regulatory'* function to the physiological systems of another through timely adaptation to distress and social cues (Feldman, 2007; Taipale, 2016). Co-regulation is particularly important during early development (Bridgett et al., 2013; Butler & Randall, 2013; Kopp, 1982; E. Z. Tronick & Cohn, 1989), when infant's self-regulatory capacities are thought to be immature and under-developed. Thus, the role of the caregiver is fundamental for the infant's appropriate regulation (Bell & Wolfe, 2004; Kopp, 1989; Sroufe, 2000) as well as for learning the skills to do so independently (Butler & Randall, 2013).

2.2.4 Developmental trajectories

Over the course of the first year, infants become more deliberate in their efforts to selfregulate (Atkinson et al., 2021). Emerging motor, attentional and social skills permit higher-order and more sophisticated forms of arousal regulation responses (Atkinson et al., 2021; Buss & Goldsmith, 1998). For example, infants become more skilled at social signalling as they come to recognize that parents' behaviour may assist them in regulating their states (Atkinson et al., 2021; S. D. Calkins & Hill, 2007). Infants also become better at controlling their attention, which allows them to disengage from the source of distress (Feldman, 2009; Posner & Rothbart, 1998, 2000). When infants' regulation of arousal is examined under laboratory paradigms such as the arm restraint or the still-face paradigms,
developmental changes in the use of regulatory strategies are found. Atkinson et al., (2021), for example, found that older infants used a lower proportion of more rudimentary self-soothing behaviours (e.g., mouthing, self-touch) than younger infants during the Still-Face procedure. Rothbart et al., (1992), showed that hand-to-mouth - a measure assessing oral self-soothing thought to be rather basic - was at its peak at three months and gradually decreased to its lowest level at thirteen months of age. Generally, older infants were more active stimulus seekers and showed more gestural communication with fewer self-soothing and avoidance behaviours. The authors interpreted these results as suggesting a developmental change from more palliative methods of self-regulation to more active coping (Rothbart et al., 1992). More recently, Suata (2023), using the toy retraction task, found that 12-month-old infants displayed more negative reactivity but also exhibited a greater number of regulatory behaviours than 6-month-olds during the challenge condition, indicating bidirectional relations between reactivity and regulatory systems. Such developmental shifts suggest that, during the first year of life, infants become more efficient in how they use regulation strategies to modify their immediate responses to arousal-eliciting situations (e.g. Calkins & Hill, 2007; Ekas et al., 2013; Kopp, 1989). More specifically, regulatory strategies are thought to evolve from reflexive or more passive attempts to regulate arousal states (e.g., gaze aversion) to more proactive forms of regulatory behaviour (e.g., self-distraction) (Santucci et al., 2008; Wu et al., 2021).

Collectively, these findings have led researchers to believe that infants become increasingly reliant on themselves rather than exclusively on others and to consider this transition from dyadic to independent regulation a normative part of development (Calkins et al., 1998; Ekas et al., 2013; Granat et al., 2017; Kopp, 1982, 1989). Challenging this view, some findings offer support for the opposite. That is, with time, infants' abilities for dyadic regulation, rather than diminishing, become increasingly

refined. For example, Rothbart et al., (1992) found that looking from the stimulus to other aspects of the environment (excluding the mother) significantly decreased across age, whereas looking from the stimulus to the mother increased between 6 and 10 and 13 months. Braungart-Rieker & Stifter (1996) showed lower levels of orienting and avoidance and greater levels of communication at 10 months compared to 5 months during an arm restraint task (Braungart-Rieker & Stifter, 1996). More recently, Atkinson et al., (2021) found an increase in dyadic strategies such as attention seeking behaviours over time. Together, these results challenge conventional perspectives by suggesting that over time, infants become progressively skilled in employing both independent but also dyadic strategies. Of note, most of these longitudinal studies have measured regulation in situations in which artificial constraints have been imposed on the mother-child interaction. Consequently, meaningful changes in dyadic regulation behaviours over time have been overlooked (for additional constraints in the literature, refer to 2.2.5).

2.2.5 Methodologies used to study self- and co-regulation of arousal

In this section, we explore the most commonly employed methods for investigating these processes while highlighting some of their main limitations.

2.2.5.1 Measures for self-regulation

In the field of behavioural science, researchers have commonly assessed infants' ability to self-regulate through controlled experiments like the toy removal task (Stifter & Braungart, 1995), the still face paradigm (Tronick et al., 1978), the mask task (Goldsmith & Rothbart, 1996), and similar laboratory paradigms. Briefly, these experiments involve subjecting infants to mild stressors or challenges such as having their mothers holding a neutral expression without speaking nor responding to any of the infants' signals (Tronick et al., 1978). These setups enable researchers to record behaviours thought to be regulatory such as gaze aversion or thumb-sucking (e.g., Buss & Goldsmith, 1998; Granat et al., 2017; Kopp, 1989; Rothbart et al., 1992; Tronick et al., 1978; Ursache et al., 2013) and document typical developmental changes in infants' choice of regulatory strategies. Although useful, there are also some limitations worth outlining. First, most research has focused exclusively on behaviours that are assumed to down-regulate physiological arousal, but very few of them have tested whether they actually serve as a downregulatory function or not (e.g. Rothbart et al., 1992) (Buss & Goldsmith, 1998; Morales et al., 2018; Wass, 2021). Similarly, very few studies have tested whether the environmental stressors defined by the experimenter genuinely function as intended. Second, the statistical models used in much of this work consist, mainly, in using summary/ static statistics (e.g. total number of times a child uses a certain strategy, presence or absence of certain behaviours, change between a baseline period and the stimulus, etc.) and have not appropriately accounted for neither the temporal dynamics nor dyadic influences (Cole et al., 2019; DePasquale, 2020; Hollenstein et al., 2017; Morales et al., 2018).

In response to these limitations, some researchers have argued in favour of an approach that views regulation as the product of constant, dynamic interactions between regulatory behaviours and prepotent responses such as an emotion primed by a laboratory task. Consequently, they have shifted away from using static variables and focused on modelling the intrinsic dynamics and reciprocal relations embedded in the behavioural time series data of the infants (e.g. Cole et al., 2017; Morales et al., 2018; Suata, 2023). Such dynamic approaches offer opportunities to investigate in detail important elements of the development of self-regulation like, for example, how and when attempts at certain strategies become effective. Despite these advances, some limitations remain. First, the ecological validity of the experimental paradigms is largely compromised with the use of

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these laboratory-tasks (Wass & Jones, 2023). A stressor suddenly appearing and disappearing after a short interval, with no control from the participant, rarely occurs in the real world (Wass, 2021). Accordingly, it is possible that spontaneous distress in infants may vary from the distress exhibited in stress-inducing tasks created by researchers (de Barbaro et al., 2023). And second, most of these studies are largely built on data that has been gained when infants are observed and examined during moments of so-called "alert inactivity" (i.e. when an infant is neither sleeping, hungry, eating, fussing, crying nor engaged in full activity) (Taipale, 2016), but these moments are relatively brief periods and, by no means are the infant's prevalent way of being (Taipale, 2016). Of note, recent work has used more ecologically valid settings (e.g. Madden-Rusnak et al., 2023); demonstrating the feasibility of studying physiological self-regulation in the context of spontaneous and unstructured infant crying outside the laboratory. Nevertheless, more efforts in this direction are required to broaden our currently limited understanding of self-regulation.

Importantly, other studies have used neuroimaging tools like functional magnetic resonance imaging (fMRI), functional near-infrared spectroscopy (fNIRS), or electroencephalography (EEG) to examine brain activity associated with self-regulation of arousal (e.g. Perone et al., 2020; Reindl et al., 2018). These methods help researchers understand the neural underpinnings of these processes but share many, if not all, of the limitations mentioned above. Similarly, parental or caregiver reports have also been commonly used in the field (e.g. IBQ; Rothbart, 1981). Although these observations can offer valuable insights into infants' regulatory behaviours in everyday life, along with the presence of negatively valence arousal, they are limited in that they only provide us with a "snapshot" of one moment in time (Wass & Jones, 2023).

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2.2.5.2 Measures for co-regulation

Research on co-regulation in parent–infant dyads has typically focused on the temporal coordination and interdependency of physiological arousal states during parent-child interactions (for review, see Davis et al., 2018). Although other measures have also been used to study co-regulation such as behavioural synchrony of gaze, affect, gestures and touch (e.g. Aureli et al., 2022; Beebe et al., 2016; Feldman, 2007; Feldman et al., 1999; MacLean et al., 2014), reviewing them falls beyond the scope of the current thesis as many of them have not tested whether they modulate physiological arousal. Similarly, it is important to emphasize that co-regulation, as per our definition (see 2.1.1), does not always require temporal coordination of physiological arousal between caregivers and infants. For instance, it may manifest as caregivers adjusting the infants' environment based on their state (e.g. moving them to a quieter room when they are over-stimulated). However, this thesis specifically concentrates on the coordination and interdependence of caregiver-infant physiological arousal states.

We know that interpersonal processes can influence our physiological arousal states and associated affect (Smith et al., 2022; Wass et al., 2019). Generally, it has been assumed that a highly sensitive and attuned caregiver is better able to meet the infant's needs in any given moment (Hoehl et al., 2021). Research in the lab has shown that increases in child arousal are down-regulated faster in the presence of a caregiver than in their absence (Calkins & Hill, 2007; Ham & Tronick, 2009; Thompson et al., 2008), and identified synchronous patterns of change in physiological arousal following the administration of experimental stressors that related to greater synchrony in mother-infant engagement behaviours (Ham & Tronick, 2009). It seems plausible that, after a stressor, parents increase their efforts to attend to their infants and this increased effort leads to a greater coherence in physiological states which, in turn, may help regulate infant's physiological arousal responses to stress. However, findings are divergent and vary according to a

number of factors including the physiological index being examined, the type of task used, and broader contextual factors such as maternal psychopathology (e.g. de Barbaro et al., 2023; or see Davis et al., 2018 for a review). Several empirical findings now support an 'optimum midrange model of contingency' in caregiver-infant interactions (Beebe, 2006; Beebe et al., 2008; Jaffe et al., 2001) in which the ideal interaction is not of absolute synchrony and coordination, but it rather involves the mismatch of responses and their subsequent repair (Beebe et al., 2011; Ham & Tronick, 2009; Jaffe et al., 2001). Most of this research has been documented during short time frames, such as single laboratory visit in a controlled environment (Bridgett et al., 2015; Ham & Tronick, 2009). Consequently, these studies are subject to most, if not all, of the previously discussed limitations (refer to 2.2.5.1). Additionally, during these visits, parents are, most of the time, on their "best behaviour", aware of being watched by multiple researchers and recorded by cameras (Wass, 2021). This makes it difficult to elicit certain important aspects of real-world caregiver-child interactions, such as child-caregiver oppositionality (Wass et al., 2023) which, in turn, limits but also compromise our understanding on what happens in real life.

In more naturalistic settings, emerging findings are also complex. Some research has shown that greater adult arousal responses to peak arousal from the infant lead to faster infants' subsequent recovery (Smith et al., 2022; Wass et al., 2019). They also showed short-term increases in concurrent parent-infant arousal synchrony following infants' negative but not positive vocalisations (Wass et al., 2019), suggesting that synchronous responses may be constrained to stressful events to help infants regulate faster. Other researchers have, however, found stronger physiological synchrony in the context of greater risk (Davis et al., 2018). For example, Smith et al., (2022) found higher physiological synchrony in anxious parent-child dyads. While non-anxious parents responded primarily to peaks in their infant's arousal, anxious parents responded also to

small fluctuations of their infant's arousal. Collectively, this adds to the increasing body of evidence indicating that too much physiological synchrony in caregiver–child interaction might be detrimental (Bornstein & Manian, 2013; Smith et al., 2022).

Overall, despite variations in experimental settings, methodologies, and operationalization of co-regulatory processes within the dyad, there are some important agreements between the findings obtained from both laboratory-controlled studies and more naturalistic observations worth outlining. First, relational influences within a caregiver-infant dyad interact with psychobiological systems in the process of coregulation (Thompson et al., 2008). Second, arousal synchrony is generally stronger in interactive contexts (Feldman, Magori-Cohen, et al., 2011) and seems to play a role in co-regulatory processes. And third, parent arousal appears to relate to child arousal in terms of both magnitude and timing, but the relationship with child soothing is unclear and unlikely to follow a simple, linear pattern.

To date, our understanding of the development of early parent-infant interpersonal regulatory processes during the first year of life remains limited (Evans & Porter, 2009). The idea that older children rely less on caregivers for co-regulation is often accepted (Thompson et al., 2008) (although also challenged, refer to 2.2.4) but, to our knowledge, no research has tested it. Nevertheless, it seems plausible that, as infants' capacity to self-regulate improves, the nature and implications of these bi-directional influences may also differ across development.

2.2.6 Summary

In infancy, regulation of physiological arousal can be split in two different key components. One extrinsic, defined as *"regulation of and by others"*, and another one intrinsic, defined as *"regulation of and by oneself"* (Nigg, 2017). The former one, also

known as *co-regulation*, is key in early infancy when the capacities for self-regulation of arousal are thought immature, poorly coordinated, and limited. This suggests that early arousal regulation should be better understood as a joint action, where both partners play an active role rather than - as it is often studied – an isolated construct within the infant. By measuring regulation in situations in which artificial constraints have been imposed on the mother-infant interaction, we have obscured meaningful changes in dyadic regulation behaviours that happen over time. Similarly, the common use of lab-based paradigms has largely compromised the generatability of the results. Consequently, more research is needed to understand how early parent–infant interpersonal regulatory processes occur and develop across the first years of life.

2.3 Early regulatory skills of attention

This section provides an overview of the research in the development of regulation of attention. It discusses the most used methods to assess it in infants and highlights relevant gaps.

2.3.1 Developmental trajectories of infant attention

The ability of an infant to effectively allocate, maintain, and, when necessary, reorient visual attention is a fundamental skill to development. It is not only a core component of learning (Mundy & Newell, 2007; Oakes et al., 2002; Schroer & Yu, 2022; Yu & Smith, 2013) but, especially in developmental populations, is considered one of the essential skills for arousal regulation (Rothbart et al., 1992; refer to 2.4 for more).

At birth, newborn "attention" is believed to be primarily involuntary, exogenously driven, and exclusively under the control of a reflexive system and subcortical structures (Gardner & Karmel, 1995; Reynolds & Romano, 2016; Richards, 1997). With time, it is

hypothesised that the rapid maturation in all cortical attentional systems enables infants to regulate their attentional states more and more flexibly and quickly and to respond to objects and events in a more controlled way. The posterior orienting system is involved in this voluntary control of eye movements and shows considerable development from 3 to 6 months of age (Johnson et al., 1991; Reynolds & Romano, 2016). For example, a study from Johnson et al., (1991) showed that only 4 months old infants, but not 2- nor 3-months old, were able to disengage from an attractive central stimulus to orient toward a simultaneously presented target. Other studies have shown a decline in infant look duration to a wide range of stimuli from 3 to 6 months of age (Courage et al., 2006). Together, these results – and others (see Hood & Atkinson, 1993; Kulke et al., 2015; Rose et al., 2004) – have been interpreted as a reflection of the improvements in the infants' ability to disengage from a stimulus. This improved ability to exert rudimentary selective attention has important implications to the development of regulation of arousal - infants are now able to shift their attention away from a distressing stimulus (e.g. McConnell & Bryson, 2005; Rothbart et al., 1992, 2011; Wu et al., 2021; refer to 2.4 for more) - and it is, perhaps, one of the most significant advances in the development of attention regulation. Before that, infants have great difficulty in disengaging from highly novel or salient stimuli.

As infants enter the second half of the first year, a seemingly opposite skill of disengagement of attention emerges with the maturation of the anterior attention system. That is, infants around the age of 9 months start to show the first signs of focused or sustained attention (Hendry et al., 2016). For example, Courage (2006), showed that, when 6-months old infants viewed Sesame Street material or faces (but, importantly, not static or less complex stimuli), their look durations increased markedly with age. The authors, - and others (e.g. Richards & Anderson, 2004; Ruff & Capozzoli, 2003) - interpreted this increase in look duration as the emergence of a more endogenous ability

to regulate attention that involves two important functions: 1) the apparently volitional direction of attention as a function of the tasks in which the individual is engaged (attention getting), and 2) the ability to inhibit attention, or "hold" one's attention to the stimulus, event, or task at hand (attention holding) (Colombo, 2001).

Generally, years of research in the field seem to agree that, early in development, "attention-getting", or how quickly infants look at the stimulus, seems to be related to salience (e.g., the size of the checks, Cohen, 1972) and "attention-holding", or how long infants look at a stimulus, seems to relate to the complexity of a stimulus (e.g., the number of checks in a checkerboard, Cohen, 1972). Over time, attentional states become gradually modulated by higher-level processes such as the infants' internal states, motivation, comprehensibility, and goals (Colombo & Cheatham, 2006; Oakes et al., 2002). In essence, a more mature type of attention, regulated by task demands, knowledge, and internal cognitive processes, begins to emerge (Orekhova et al., 2001; Pomaranski et al., 2021).

2.3.2 Co-regulation of attention

So far, we have presented the development of attention regulation studied in isolation and thus, only considered the properties of the individual who is attending (i.e. infants) and of the stimulus/ events being attended (e.g. salience, complexity, etc.). While this two-way model can explain some aspects of the development of attention regulation, it falls short, especially during early infancy when attention is predominantly social (i.e. occurs in a social setting) (Wass & Leong, 2016). Thus, in addition to properties that we see when we study attention in isolation, we need to consider properties of the social environment (Brandes-Aitken et al., 2019; Wass & Leong, 2016; Yu & Smith, 2016). In this section we briefly describe how coordinated processes with a social partner

(understood here as co-regulation of attention) can give rise to and/or boost higher order functions, such as joint and sustained attention.

Research in this topic has shown that caregivers provide opportunities and establish boundaries to steer children's attention and prevent distractions (Bakeman & Adamson, 1984; Graziano et al., 2011; Suarez-Rivera et al., 2019; Wass, Clackson, et al., 2018; Yu et al., 2019; Yu & Smith, 2016). More specifically, research has shown that when adults and infants jointly attend towards the same object during shared play, infant's attention durations are longer than towards objects that they attend to on their own (Yu & Smith, 2016). Relatedly, infant's attention spans are generally longer during joint play as opposed to solo play (McQuillan et al., 2020; Wass, Clackson, et al., 2018). One explanation could be that social context *per se* increases infants' endogenous (voluntary) attention regulation. Follow-up analyses have, however, indicated otherwise: rather, it seems that it is the exogenous attentional scaffolding (and capture) provided by the parent during social play the key factor leading this increase in infant's attentional levels (Wass, Clackson, et al., 2018). In other words, caregivers, by responding contingently with multi-modal behavioural inputs, regulate (or, in other words, co-regulate) their infants' attention, even before the infants are capable of such performances unaided.

As development progresses, these episodes of shared attention with social partners become more and more frequent. For example, Bakeman & Adamson (1984), found a developmental increase in coordinating attention to partner and to the object the two shared, but not an increase in solitary object play. Consistent with this, other studies have found that that "*unilateral coregulation*" in which one person (i.e. the infant) ignores the other's bids for attention, engaging instead in their own activity, largely prevail at the beginning of the first year of life and then decrease linearly to give way to a more "*symmetrical co-regulation*", when mother and child are engaged with each other and both contribute to the ongoing interactions (e.g. Aureli & Presaghi, 2010; Evans & Porter, 2009; Fogel et al., 2003). Together, these findings point to an interesting (yet underexamined) transition where, over time, infants take on a more active role in co-regulation of attention.

Being able to share the attention with others is a key developmental milestone as it is during these voluntary shared attentional engagement periods where most (if not all) of infants' information processing and learning takes place (e.g. Bornstein, 1985; Mundy et al., 2007; Mundy & Newell, 2007; Schroer & Yu, 2022; Yu & Smith, 2013). However, despite universal agreement about its importance, there is still little agreement on exactly how the *jointness* is achieved. As a result, the mechanisms through which caregivers influence and regulate infant real-world attention on a second-by-second basis and *vice versa*, and how these influences change over developmental time, remain relatively unexplored.

2.3.3 Methodologies used to study attention in infancy

Measurement of attention in infancy is difficult, with most studies using indirect methods of assessment to elicit and infer attentional processes. Broadly, there have been three approaches to the measurement of infant attention: some authors have indexed attention behaviourally by studying looking behaviour, some have used associated autonomic responses such as heart rate and others have turn to neuroimaging techniques. The following subsections offer a brief overview of them.

2.3.3.1 Attention measured through looking behaviour

Infant looking behaviour has been widely used as the main behavioural measure of infant attention. By manipulating the familiarity, the complexity, the comprehensibility, the salience, etc. of the stimuli presented (e.g. Cohen, 1972; Oakes et al., 2002) researchers

have been able to learn how "look duration", "number of fixations", "resistance to the distractor" etc. change not only over developmental time but also under different situations and neurodevelopmental disorders (Cohen, 1972; Colombo & Cheatham, 2006; Elsabbagh et al., 2013; Hendry et al., 2018; Johnson et al., 1991; Oakes et al., 2002).

While this research has been highly informative, our understanding of infants' attention remains limited. First, the majority of this research has studied how infants passively respond to stimuli as they appear and disappear on-screen, following an experimenterdetermined sequence, or to an experimenter performing a specific activity (e.g. Orekhova et al., 2006; Richards & Turner, 2001; Throm et al., 2023). This has allowed researchers to study infants' "pure" capacity for attention regulation, but it differs in several important ways from how we actually pay attention in complex, dynamic, interactive, real-world settings (Brandes-Aitken et al., 2019; Wass & Jones, 2023; Wass & Goupil, 2022; Wass & Smith, 2014). For example, whereas attention in screen-based tasks is largely passive, the real-world is interactive and manipulable, and so how we interact with the world determines what information we, in turn, receive from it: experiences generate behaviours (Anderson et al., 2022; Smith & Gasser, 2005). Second, as mentioned above, attention in infancy is predominantly social and paradigms like these have usually overlook changes in dyadic influences (Wass, Clackson, et al., 2018; Wass & Leong, 2016). Third, behaviour alone has its limitations as it is not unusual for infants to maintain their gaze on a stimulus even when they are no longer actively paying attention. Consequently, similar behaviours labelled as "attention" could occur across varying levels of attentional engagement (Phillips et al., 2023; Reynolds & Romano, 2016).

More recently, there have been some efforts in studying attention as it occurs spontaneously during more naturalistic unstructured paradigms such as object exploration, free play, or parent-child interactions (e.g. Begus et al., 2015; Wass, Clackson, et al., 2018; Yu & Smith, 2013). This has allowed researchers to understand not only how infant attention "in isolation" (i.e. self-regulation of attention) works but also to explore how changes in the focus of one social partner's (e.g. mothers) attention associate, on a second-by-second scale, with changes in the other social partner (e.g. infants) (i.e. co-regulation of attention). As of today, however, we know very little on how these bi-directional processes between infants and their social and physical environment develop and change over time.

2.3.3.2 Attention measured through autonomic physiology

To overcome the constraint of exclusively studying attention through looking behaviour, which may not distinguish between passive looking and active attention, some researchers have chosen to explore infant attention through the examination of associated autonomic responses. Richards and colleagues (e.g. Courage et al., 2006; Lansink & Richards, 1997; Richards, 1997), have developed a model that identifies changes in heart rate that coincide with different phases of infant attention. Their research has shown that within a single glance, infants go through four attention phases: stimulus orienting, sustained attention, pre-attention termination, and attention termination. Among these phases, the most relevant ones are sustained attention and attention termination. Sustained attention is manifested as a significant and sustained decrease in heart rate from its initial level that occurs when infants are actively engaged in an attentive state. Attention termination follows sustained attention and is manifested as a return of heart rate to its initial level. This research has proven useful to differentiate times when infants are engaged in attentive states from times when infants might still be looking at the stimulus but are no longer in an attentive state. Importantly, this research has been carried out within the context of extended looking at television (Richards, 2010, 2011) and screen-based tasks (e.g. Xie et al., 2019) which limits its applicability in more real-world situations where attention spans are typically shorter.

2.3.3.3 Attention measured through neural activity

The use of neuroimaging techniques has been another popular approach to study infant attention beyond overt attention (i.e. looking behaviour). Amongst these, electroencephalography (EEG) has been the preferred method in infant populations. Briefly, EEG involves placing electrodes on the scalp to detect and amplify the electrical signals generated by neurons within the brain. EEG is particularly useful for studying the temporal dynamics of brain activity, as it provides high temporal resolution, meaning it can detect changes in brain activity with millisecond precision. EEG not only offers a non-invasive technique to study sub-second changes in brain function (Phillips et al., 2023), surpassing all other neuroimaging methods in their temporal resolution, but it is also developmentally sensitive for use with infants from birth and more pragmatic (e.g. less expensive, and portable) than other methods (e.g. MRI, MEG) (Norton et al., 2021). This measure is interesting because it is a more direct measure of neural activity than it is heart rate. As a result, infant visual attention has been studied using both measures of changes in the power spectral density (PSD) of EEG oscillations in different frequency bands and measures of event-related potentials (ERP). The latter falls beyond the scope of the present thesis but see De Haan (2002) for a review of this literature.

Measuring power within certain frequency bands can provide important information about child development and different cognitive processes (Norton et al., 2021). A substantial body of research has focused mainly on two infant EEG rhythms to study infant attention. These are theta (3 - 6Hz) and alpha (6 - 9Hz) rhythms. Increases in the 3 - 6Hz theta band have been observed during both anticipatory and sustained attention. Orekhova et al. (1999) found an increase in the theta PSD when infants were anticipating a person (internally regulated attention) as compared with the "more passive" attention to the real sensory object, considered as the baseline state (Orekhova et al., 1999). In another study, Orekhova et al. (2006) found increases in theta activity during exploratory behaviour in both infants and children. Similarly, Xie et al. (2018), found that infants started to show a pattern of increased theta PSD during sustained attention as early as 8 months. This effect became well established from 10 months of age (Xie et al., 2018). Together, these studies - and similar others (see Braithwaite et al., 2020; Brandes-Aitken et al., 2023; Jones et al., 2020; Meyer et al., 2019, 2023) - suggest that the expression of theta could signify the engagement of neural networks related to attention such as the anterior attention system (Xie et al., 2019).

On the other hand, other studies have found that infants, as well as adults, typically exhibit decreases in alpha band activity under conditions of increased attention (Libertus et al., 2009). For example, Xie et al. (2019) found greater attenuation of the alpha PSD during sustained attention than inattention in the frontal, central and posterior electrodes of 12 months-old infants watching a Sesame Street movie. Such effects were not found in 6 months-old infants or 10 months-old infants, when these effects of attention on alpha are thought to only start to emerge (Xie et al., 2019). Another study, however, did find a reduction in the PSD in the alpha band at midline parietal and central electrodes in younger infants (7-month-old) in a context of increased numerical novelty (Libertus et al., 2009). The authors interpreted such decreases in alpha power as reflecting increased attention. Conversely, Orekhova et al. (2001), examined relative alpha amplitudes and the duration of internally regulated attention and found an increase in alpha over the posterior parietal region in infants who maintained their attention for a relatively longer period. One possible explanation to such inconsistent findings within the alpha activity could be that different brain regions might play different roles in these attentional processes.

While the study of theta and alpha activity can offer insights into infants' intrinsically guided attention beyond its behavioural manifestations, caution should be taken when interpreting the functional significance of both frequency power bands. Most researchers, however, would agree that both theta and alpha bands reflect some aspect of infant attention. As such, both theta and alpha effects are now widely known in the literature as "theta synchronization" and "alpha desynchronization" (Xie et al., 2018). This has led several authors to both interpret theta activity as an index of active regulation of attention and cognitive effort (Begus & Bonawitz, 2020; Bosseler et al., 2013; Orekhova et al., 1999; Xie et al., 2018), and consider "alpha desychronization" as an electrophysiological sign of enhanced cortical excitability and potentially increased attention allocation and/ or engagement (Jones et al., 2015; Xie et al., 2019).

The scope of these findings on infant's attention is vast and well replicated, however, there are also some limitations. First, the statistical models used in much of this work consist, mainly, in analysing the EEG PSD data during whole episodes of looking but are not time locked to the start of it (e.g. Braithwaite et al., 2020; Jones et al., 2020; Libertus et al., 2009; Meyer et al., 2019; Orekhova et al., 2006). Second, similar to 2.3.3.1, most of the experimental paradigms used are very structured lab-based tasks that rely on a type of attention that is intentionally elicited. To our knowledge, there are only a limited number of studies (Phillips, 2023; Phillips et al., 2023; Wass, Noreika, et al., 2018) that looked at time-resolved changes in brain activity linked to particular attentional events using more naturalistic paradigms (e.g. free play). The exploration of how specific fluctuations in brain activity time-locked to specific naturalistic attentional events (e.g., onset of a self-initiated attentional period) change during the first year of life, however, remains unknown.

2.3.4 Summary

The regulation of attention in developmental populations should be conceptualized as the result of the "interaction among different systems at different levels of maturity"

(Colombo, 2001). Rudimentary forms of various attentional functions are present at birth but, within the first years of life, significant changes occur to give way to a more endogenous, voluntarily regulated, attentional ability. Understanding how infants develop this ability is foundational to understanding subsequent cognitive development and learning, as well as for identifying and intervening in atypical development, and conditions such as Attention Deficit Hyperactivity Disorder (ADHD).

Studying infant attention, however, is challenging and our understanding on how infants regulate attention in real-world settings (e.g. what determines where, and for how long infants spontaneously allocate attention), and how this changes over developmental time is limited. Likewise, it is recognized that attention during infancy is predominantly social, and external environmental influences, especially those from the social environment (e.g., social partners), play a significant role in shaping the development of these early regulatory attention skills in infants. However, little is known on how influences from the social environment change and develop over time to influence infant attention.

2.4 Linking the two: regulation of physiological arousal and attention in infancy

In the past two sections we have reviewed two primary domains of regulation separately. First, we presented "regulation of physiological arousal", which consists in managing inner physiological states (more in 2.2). Next, we introduced the concept of "regulation of attention", which consist in the ability to control attention endogenously according to goals and intentions (Conejero & Rueda, 2018; Feldman, 2009) (more in 2.3). Although they are discussed separately, and while examining the connections between the two is beyond the scope of the current thesis, it is worth noting that there is substantial evidence indicating an overlap between the two (Coull, 1998), implying that arousal regulation can facilitate attention and *vice versa*, regulation of attention can facilitate arousal regulation.

Research using single cell recordings in animals has found a relationship between the activity levels in the locus coeruleus (LC) - a structure from the brainstem thought to project throughout the cerebral cortex and to areas associated with attentional processing in particular (Morrison and Foote 1986 in Aston-Jones G & Cohen J. D., 2005). At very low LC activity levels, animals struggled to engage adequately in the task, at higher levels of LC activity, arousal and performance increased and, finally, at the highest LC activities, performance decreased again with behaviour (and attention) relatively indiscriminate and labile (Aston-Jones et al., 1999; Aston-Jones G & Cohen J. D., 2005). These findings gave way to the Aston-Jones model that posits that different behavioural indices of attention (e.g. inattention, selective or focused attention and orienting or scanning attentiveness) are paralleled by the activation of the LC and each have adaptive advantages under different environmental circumstances (Aston-Jones et al., 1999). In human research, single cell studies are not feasible, so alternative approaches are used. Since LC activation is strongly associated with the activation of the sympathetic nervous system, researchers have employed autonomic measures like heart rate to investigate the relationship between autonomic arousal fluctuations and their impact on attention. Generally, evidence suggests that dynamic changes in arousal track the relative uncertainty of the environment, either promoting focused or selective attention over orienting or scanning attentiveness, or vice versa (Cohen et al., 2013; de Barbaro et al., 2017; Gardner & Karmel, 1984; Karmel et al., 2006; Kleberg et al., 2019; Wass, 2017). For example, de Barbaro et al. (2017) found that spontaneous increases in arousal were associated with a more vigilant attentional profile, characterized by shorter look durations, while lower arousal levels were linked to longer looks. Kleberg et al., (2019), on the other hand, found evidence for an inverted U-shaped relation between infants' pupil dilation – an index of arousal, more particular of LC activity - and the likelihood of a first fixation at the targets, with highest likelihood of a first fixation at the targets at

intermediate levels of arousal. Collectively, these findings indicate that arousal can influence attention. Therefore, when physiological arousal is not optimally regulated, the capacity to flexibly regulate attention is likely to be compromised (Eisenberg & Zhou, 2015).

On the other hand, the regulation of attention is also closely related to infant's physiological arousal. Ample evidence suggests that showing attention-eliciting, comprehensible stimuli can lead to decreases in physiological arousal (e.g. Richards, 2007, 2010). Similarly, the capacity to disengage from distressing stimuli and/or engage effectively with objects or caregivers has been widely hypothesized to associate with the regulation of arousal (though rarely measured directly; see Morales et al., 2018; Wass, 2021). More recent work has found a positive association between attention regulation at 6 months and physiological regulation at 15 months (Wu et al., 2021). Generally, it is likely that attention regulation skills allow young children to maintain concentration, stay on task, handle frustration, and resist distractions (Cuevas & Bell, 2014; Feldman, 2009; Wu et al., 2021). In this way, these skills empower children to navigate and master their environment which, in turn, might help reduce hypervigilance, often accompanied with higher physiological arousal states.

In sum, in line with some literature (e.g. see Feldman, 2009; Gardner et al., 1992; Karmel et al., 1996), arousal and attention regulatory systems are probably best understood within a framework of an inherent homeostatic regulatory mechanism that functions to preserve infants' balance with their environment and, most likely, allow them to accomplish their goals and/or goal states. Changes in arousal or attention might be both a cause, and a consequence, of changes in attention or arousal, respectively. Thus, learning how to regulate one is closely related to the other, and *vice versa*. Consequently, the study of these processes seems crucial for all the development that follows.

2.5 Aim of the current thesis and novel contributions

This section briefly highlights a couple of key concluding remarks drawn from the sections above. It follows with an overview of the principal aims of the thesis and finishes with a summary of the novel contributions this work will bring to the field.

2.5.1 Final remarks

2.5.1.1 Developmental assumptions

The development and mastery of the ability to both regulate our physiological arousal and attentional states is critical to all aspects of development (refer to 2.1). Much of the infant/ child research emphasizes the developmental increase in self-regulation while highlighting a gradual shift from the infant's initial dependency on direct regulatory support from the caregiver to the incremental internalization of regulation. Whilst this transition from co-regulation (where regulatory processes are shared between child and caregiver) to self-regulation seems reasonable from a developmental point of view, it is difficult to pinpoint exactly when this transition occurs (Atkinson et al., 2021). Moreover, some evidence suggests that dyadic strategies, instead of diminishing or being substituted, persist in playing a significant role in regulation (Atkinson et al., 2021; Braungart-Rieker & Stifter, 1996; Rothbart et al., 1992; Wass, Clackson, et al., 2018; Yu & Smith, 2013). Research on this is scarce and complex and remarkably little is known on how these co-regulatory dynamics between infant-caregiver change and evolve over time (Brandes-Aitken et al., 2019; Wass et al., 2023).

2.5.1.2 Dynamic systems' perspective

"Humans develop through relationships" (Fogel, 1993) - "how I move and where I attend influences what information I receive" and, similarly, "how I behave toward others influences what I receive from them in return" (Wass & Goupil, 2022). Following from Fogel's idea, in order to properly understand the behaviour and development of intricate systems, such as human infants, it is imperative to grasp not only their intrinsic qualities (as done by most pervious research) but also their interactions with the specific attributes of the environment (Cox et al., 2010). Consequently, there is a necessity for researchers to transition away from more conventional laboratory-based studies, where the events (and their exact timings and type) are decided by the experimenters, rather than the infants. Instead, there is call for more "naturalistic" or "real-world" research. In this thesis, we understand this as any effort to observe and understand infant behaviour as it naturally occurs, without interference or manipulation from the researchers. This type of research is conducted in unstructured settings - it can be done in the lab or in the home environment – but crucially, the events are self-generated spontaneous behaviours from the specific population under study, such as infants and their caregivers. As a consequence, the type of events and their specific timings, are based on the participants themselves (Wass et al., 2023). Such naturalistic approaches, however, do not come without challenges. Naturalistic research is time-consuming, it typically generates hundreds of variables, and the events (i.e. behaviours of interest) happen irregularly and in an uncontrolled manner. It is not uncommon for data collected in these unstructured paradigms to contain a higher volume of artifacts (see Georgieva et al., 2020; Haresign et al., 2021; Kayhan et al., 2022) compared to data collected in controlled environments where participants' behaviours are more constrained. As a result, researchers need suitable training in data management, statistical analysis, and analytic tools (Wass et al., 2023). Additionally, there is a considerable amount of manual coding of behaviours required at present, which consumes both time and resources (see also Tamis-LeMonda et al., 2017; Wass & Goupil, 2022 for more challenges in the field). Despite these, many researchers recognise the importance of adopting new approaches that can provide new insights into understanding how we develop through our everyday, moment-by-moment interactions

with our physical and social environments around us (e.g. Madden-Rusnak et al., 2023; Phillips et al., 2023; Wass & Goupil, 2022; Wass et al., 2018).

2.5.2 Aim of the thesis

The main aim of this project was to study the development of early regulatory abilities of physiological arousal and of attention in infancy, while considering the bidirectional, mutually influential regulatory processes between infants and their physical and social environment. The main research questions of this thesis were:

- Does the regulation of physiological and attentional states transition towards greater endogenous control as time progresses? In other words, does self-regulation increase over time?
- And can we show that both our physiological and attentional states become progressively less contingent on others over developmental time? In other words, does co-regulation decrease over time?

2.5.3 Novel contributions

This thesis applies an integrated and longitudinal approach that combines home- and naturalistic lab-based research to study how bidirectional influences between the infants and their physical and social environment contribute to the development of physiological arousal and attention regulation.

First, this project offers a new methodological approach on the study of physiological arousal regulation by bringing its study in the home environment. We have developed a new wearable device that allowed us to record naturalistic data on a variety of parameters such as heart rate, audio data, proximity between the members of the mother-infant dyad, activity levels and location. These devices can be worn by infants as young as 5-month-

old and their mothers and can record continuous data for up to 10 hours. Technologies like this can help enhance our understanding of, for example, how instances of infant distress are corrected in the home environment (and throughout development) and clarify the caregiver's involvement in infants' physiological regulation as well as how it evolves and changes over development (chapter 3).

Second, understanding how infants develop the capacity to regulate attention is foundational to understand subsequent cognitive development and learning. However, our understanding in this area is limited (refer to 2.3). The current thesis presents a variety of state-of-the-art time series analyses to examine how moment-by-moment changes in physiology and cortical neural activity inter-relate in determining how infants spontaneously allocate attention in real-world settings and considers how these inter-relationships change across development (chapter 4). In a separate analysis, it explores the dynamic interactive contingencies between infants and their caregivers that drive inter-personal attention coordination on a moment-by-moment basis and assesses how these processes change over time (chapter 5). Importantly, it brings the research of attention regulation to a more naturalistic setup (i.e. free play).

Methodological advancements and shifts towards more naturalistic paradigms like the ones mentioned above are crucial because they not only offer greater generatability of the results, but also enable researchers to work with events defined by the participants themselves (e.g. negative vocalisations from the infant or spontaneous attentional redirections during play), rather than pre-determined events defined by experimenters (e.g. a sudden restriction of the arm, or stimuli appearing and disappearing on screen).

Third, it provides a longitudinal view on how parental behaviours and infant regulatory skills interact and evolve over time. These studies are scarce in the literature, however, only through a longitudinal approach that expands across multiple levels of analysis we

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can gain new insights into (co-)regulatory dynamics and advance our understanding of non-normative developmental pathways.

Overall, this work is important not only to advance our understanding of the development of infant (co-)regulatory abilities, but also for identifying new and more naturalistic ways to study it.

Chapter 3. Do shorter infant distress episodes with age result from improvements in caregiver-infant co-regulation or infant self-regulation?

This chapter is a pre-print article submitted to *Child Development* for publication. It examines developmental changes in the dynamics between infants and their caregivers around infant negative and neutral affect vocalisation. Subheadings and figure placement and style have been adapted to conform to the thesis format. The supplementary materials for this chapter are available in Appendix A.

3.1 Abstract

We examined developmental changes in naturalistic co-regulatory processes during early life by recording day-long ECG, proximity, vocal and activity data using wearable devices from 89 caregiver-infant dyads at 5 and10-months (73% white). First, we found that infants' negative vocalisations were shorter at 10 months. Second, we examined whether these changes were due to improved self-regulation or more effective co-regulation. At 5 months, caregivers were more active partners in synchronizing to infants' affect, but increased caregiver involvement did not associate with faster quieting. Instead, caregiver responses became more predictive of infant quieting at 10 months. Based on this, we argue that shorter infant distress periods and reduced caregiver responses at 10 months imply more efficient co-regulation rather than better infant self-regulation alone.

3.2 Introduction

Infants are heavily reliant on caregivers for the regulation of physiological arousal (Calkins, 2007; DePasquale, 2020; Feldman, 2009; Thompson et al., 2008). Early in development, stress-regulatory systems are still developing, and caregivers play a key

role in working jointly together with the child to regulate their infant's stress response and to help them respond adaptively (DePasquale, 2020; Feldman et al., 2006). There is now considerable evidence that caregivers, by providing contingent stimulation and soothing, help infants and children towards appropriate physiological, behavioural and affective regulation (Bell & Wolfe, 2004; Donald Woods, 1957; Somers & Luecken, 2021; Tronick, 1982). This phenomenon is known as co-regulation, which refers to regulatory processes that operate through the dynamic, bidirectional coordination between two interacting partners (Beebe et al., 2016; Feldman, 2007; Fogel, 1993, 2017; Taipale, 2016; Wass et al., 2023 for a recent review).

Previous research has suggested that the benefits of dyadic co-regulation processes are multiple. Although bidirectional, early coregulation is asymmetric, driven largely by the caregiver adapting to the child's needs (Beebe et al., 2016; Wass et al., 2023). The parent, by adapting to the child, can respond contingently to the child's needs. This contingent parental responsive behaviours, in turn, help infants to understand that their behaviour effects the behaviour of others from an early age and thus contributes to the development of self-awareness (Bigelow & Power, 2016). On the other hand, the child, by adapting to the parent, gains self-control (Brown et al., 2022; Feldman et al., 1999). Evidence suggests that these early co-regulatory processes within the caregiver–child dyad drive the development of self-regulation within the individual (Butler & Randall, 2013). By co-constructing optimal emotional states, the caregiver extends and scaffolds the infant's emerging self-regulatory capacity (Butler & Randall, 2013; Lobo & Lunkenheimer, 2020).

We know that interpersonal processes can influence our physiological arousal states and associated affect (Smith et al., 2022; Waters et al., 2017). Because of this, research on co-regulation in parent–infant dyads has typically focused on the temporal coordination and interdependency of physiological arousal states during parent-child interactions (Butler

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& Randall, 2013). Generally, it has been assumed that a highly sensitive and attuned caregiver is better able to meet the infant's needs in any given moment (Bornstein & Manian, 2013; Somers & Luecken, 2021).

Research in the lab has shown that externally induced increases in child arousal are downregulated faster in the presence of a caregiver than in their absence (Calkins, 2007; Thompson et al., 2008); and has suggested that children and their caregivers show synchronous patterns of change in arousal following the administration of experimental stressors (e.g. DePasquale, 2020; Ham & Tronick, 2009). Research in more naturalistic settings has also shown that adults tend to experience an increase in their own arousal in the time window following an increase in infant arousal, along with a transient increase in caregiver-child arousal synchrony (Smith et al., 2022; Wass et al., 2019). These changes were particularly marked following infants' negative but not positive vocalisations (Wass et al., 2019); and moments where caregivers showed increased arousal responses to peak arousal moments from the infant led to faster infant recovery. Other researchers have, however, found elevated arousal synchrony in the context of greater risk (Davis et al., 2018), such as increased caregiver-child synchrony in anxious caregivers (Smith et al., 2022). This is consistent with lab-based studies which support an 'optimum midrange model' of contingency in caregiver-infant interactions (Beebe et al., 2016; Jaffe et al., 2001) in which the ideal interaction is not of absolute synchrony and coordination, but rather it involves the mismatch of responses and their subsequent realignment (Beebe et al., 2011; Ham & Tronick, 2009; Hubbard & Van Ijzendoorn, 1991; Jaffe et al., 2001).

Collectively, despite variations in experimental settings, methodologies, and operationalization of co-regulatory processes within the dyad, there are, then, some important agreements between the findings obtained from both laboratory-controlled studies and naturalistic observations. First, interactions within the caregiver-infant dyad

interact with psychobiological systems to facilitate the process of co-regulation (Thompson et al., 2008). Second, arousal synchrony is generally stronger in interactive contexts (Feldman et al., 2011) and seems to play a role in co-regulatory processes. And third, parent arousal appears to relate to child arousal in terms of both magnitude and timing, but the relationship with child soothing is unclear and unlikely to follow a simple, linear pattern.

According to most classic models, development shows a gradual transition from coregulation, where changes in the child's arousal are managed jointly across an interacting dyad, to self-regulation, where changes in the child's arousal are managed by the child increasingly on their own/ without external support (Bronfenbrenner, 1977; Feldman, 2007; Kopp, 1982; Stern, 1985; Tronick, 1982). Some studies have tested this by observing changes in facial affect and attention during short lab-based caregiver-child interactions. For example, Cohn & Tronick (1987) documented changes in facial affect during a dyadic interaction at 3, 6 and 9 months, and found that caregiver positive affect preceded infant positive affect at 3 and 6 months, but that sequential links were not observed at 9 months. This suggests that the child's positive affect displays become increasingly independent of the caregiver with age (Aureli et al., 2022; Doiron et al., 2022). Other research has suggested, though, that the lead-lag structure of caregiver-child arousal synchrony shifts over time. Early in development, the caregiver synchronises primarily with the infant by adjusting and adapting to them. Later in development, the child adapts to the caregiver as well as *vice versa*, such that both partners are actively engaged in maintaining coordination across time (Feldman et al., 1999; Porter et al., 2022).

These findings were obtained from lab studies that observed relatively small bouts of child-caregiver lab-based interaction. In consequence, these studies showed variability over a relatively narrow range of affect. Do similar principles guide real-world behaviours? Here, we wished to examine this question, by tracking the development of self- and co-regulation in everyday naturalistic child-caregiver interactions. Our initial aim was to consider: do infant negative affect displays become more short-lived over developmental time? Based on previous research (Hubbard & Van Ijzendoorn, 1991) we predicted that they would. We then planned to ask: is this because infants become better at self-regulation (i.e., does infant recovery become less dependent on caregiver responses over time)? Or is it because co-regulatory processes are becoming more effective?

To examine this, we developed small wearable devices that allowed us to obtain day-long recordings across a variety of data modalities such as heart rate, proximity, activity and vocal data from infants and their caregivers in their home environment (see Figure 3.1, and Methods for more details on the devices). We focused our analysis on physiological arousal changes around vocalisations and temporal coordination within the dyad. In particular, we examined heart rate, which is thought to reflect overall autonomic state (Palumbo et al., 2017) and is more robust to physical movement than other measures (Thorson et al., 2018). We also used machine learning classifiers to identify vocal signals from the infant and to differentiate between vocal displays of negative affect from positive-to-neutral affect. This is important because by 5 months of age, infants have already learnt that their vocalisations influence the behaviour of social partners (Goldstein et al., 2009) and can produce speech-like vocalisations in conjunction with both positive and negative valence (Oller et al., 2013). The way infants express and perceive emotions changes throughout development, however, it is well established that human caregivers exhibit remarkable sensitivity in perceiving the information conveyed through infants' vocalisations from very early on (Lindová et al., 2015). Similarly, infant cry and laughter show functional stability across development, with cry overwhelmingly expressing negative and laughter positive emotional states (Oller et al., 2013). Accordingly, the integration of physiological arousal with vocal affect allowed us to explore early physiological influences in mother-infant dyads around different emotional moments.

Previous research has used similar methods to show that 12-month-old-infants experience peaks in their own physiological arousal around speech-like vocalisations and (especially) cries; caregivers also experience increases in their own physiological around infant speech-like vocalisations and (especially) cries, but do not experience arousal changes around their own vocalisations (Wass et al., 2022, 2019). These studies also showed that different types of vocalisations elicit different patterns of change across the dyad. Cries occur following reduced infant arousal stability and are followed by increased child-caregiver arousal coupling, and subsequent decreases infant arousal. Speech-like vocalisations also occur at elevated arousal, but lead to longer-lasting increases in arousal, and elicit more parental verbal responses (Wass et al., 2022, 2019).

Our aim was to examine how these patterns of arousal and infant-caregiver arousal coupling that are time-locked to infant vocalisations change during the first year of life. First, we examined whether infant negative affect vocal displays become more short-lived with infant age, as expected based on previous research (Hubbard & Van Ijzendoorn, 1991). If this was observed, we planned to test two hypotheses. The first (Hypothesis 1) is that infants become better at self-regulation (i.e. that infant recovery becomes less dependent on caregiver responses over time). We predicted that, if Hypothesis 1 were true and older children rely less on caregivers for (or are less susceptible to) co-regulation, then infant arousal should become progressively less contingent on others over developmental time. Thus, we hypothesised that dyads with younger infants would show higher synchrony in arousal around and after negative vocalisations (see Methods for formal definitions of synchrony), and that recovery times would correlate more strongly with their caregivers' responsiveness at younger ages.

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An alternative hypothesis (Hypothesis 2) is that improvement over developmental time might lie not only in improvements in the infant's capacity for self-regulation *per se*, but rather in the quality of the dyadic relationship, where both members of the dyad become more attuned to each other (e.g. infants learn to respond to their caregivers' contingent caregiving and use it more efficiently for co-regulation, while caregivers learn to adapt to their infants faster/ better). If developmental changes are attributable to enhanced caregiver-infant co-regulation rather than solely to improvements in infant self-regulation, then our prediction would be the opposite. That is, dyads with older infants would show higher concurrent and sequential synchrony in arousal around and after negative vocalisations and faster recovery times would correlate more strongly with caregivers' responsiveness at 10 months compared to 5 months. We chose to concentrate on the first year of life, as it marks the initial stages of the development of self-regulation (Aktar & Pérez-Edgar, 2020), and research has demonstrated that early individual differences can be linked to later developmental outcomes (Stifter et al., 1999).

3.3 Materials and methods

3.3.1 Experimental design

Participating caregivers were invited to select a day during which they would be spending the entire day with their child, and which was, as far as possible, typical for them and their child. The researcher visited the participants' homes in the morning to fit the equipment and returned later to collect it. The mean recording time per day was 6.84 hours (std = 1.62) at 5 months and 6.31 hours (std = 1.9) at 10 months. The average duration did not differ significantly between ages (t(73)= 1.25, p=0.21).

The equipment consisted of two wearable devices, for both infant and caregiver (see Figure 1A and B). These devices have been developed specifically for the current project

and contain a built-in Electrocardiogram (ECG) (recording at 250Hz), a microphone (recording at 48kHz), a proximity sensor (recording at 1Hz), an accelerometer (recording at 40Hz) and a GPS (recording at 1Hz). Data were synchronised and timestamped using UTC and stored to an internal micro-SD card (see 3.3.3.1 for more details).

For the infant, we designed a one-piece bodysuit with a pocket around the chest area that held the devices, with apertures which allowed the ECG and microphone wires to pass through the bodysuit and be placed on the infant's skin (ECG wires) or clipped to the top right of the vest (microphone) (see Figure 1D). For the caregivers, we designed an elastic belt to be worn around their torso that contained a pocket to hold the device (see Figure 1C). Similar to the infants, caregivers were instructed to clip their microphones onto their clothes. The ECG electrodes were attached using standard Ag-Cl electrodes, placed in a modified lead II position.





A) Picture of the wearable device showing the microphones, ECG leads and their housing.

B) Picture of the inside of the wearable devices showing the printed circuit board, the

space for the SD card and the port to charge the battery (both circled in blue). C) Picture showing the elastic belts we designed for the caregivers to wear the devices. There was a pocket sewn onto the belts to house the devices and some Velcro that allowed the caregivers to adjust the belts around their chest. D) Picture of the vests designed to be worn by the infants. The devices were fit inside the inner pocket situated at the height of the infant's chest. Four holes were made on the vest to let the ECG wires through and be able to stablish contact with the skin of the infants. The extra hole was used for the microphone – this was passed in and back out through the hole situated on the top right of the vest and clipped next to the neck. The Velcro straps made the vest suitable to different bodies.

3.3.2 Participants

Participants were typically developing infants and their mothers. The catchment area for this study was East London, including boroughs such as Tower Hamlets, Hackney and Newham. Further demographic details on the sample are given in APPENDIX A, Table S1. Participants were recruited postnatally through advertisements at local baby groups, local preschools/nurseries, community centres and targeted social media campaigns aimed at all caregivers in the area. We also operated a word-of-mouth approach, asking caregivers who got involved to ask if their local networks would be interested in participating. Ethical approval was obtained from the University of East London ethics committee (application ID: ETH2021-0076, see APPENDIX D).

Initial exclusion criteria included: complex medical conditions (e.g. heart rate condition, neurological/ genetic abnormality), known developmental delays, prematurity, uncorrected vision difficulties and caregivers below 18 years of age. Further exclusion criteria as well as final numbers of data included in each of the analyses for both samples are summarised in APPENDIX A, Table S2. The final samples included 23 infant females and 26 infant males at 5 months and 15 infant females and 11 infant males at 10 months. Data was analysed in a cross-sectional manner. Average age for infants was 5.91 months

(std = 0.85) and 10.89 months (std = 0.75) respectively. Average age for caregivers was 35.76 years (std = 4.09) at 5 months and 35.68 years (std = 4.23) at 10 months. This is the first time that any of these data have been analysed and reported.

3.3.3 Data processing and statistical analysis

3.3.3.1 Raw data structure

ECG data were recorded at 250Hz and contained the second of each reading, the value of the ECG trace, and two data quality confidence estimates. Proximity data were sampled at 1Hz and, like the ECG dataset, contained information on whether proximity was in range or not (i.e. devices detected each other or not) and the distance between the paired devices in metres with an accuracy of centimetres. Actigraphy data were sampled at 40Hz. Audio data was recorded at 48kHz and saved as an .MP3 file.

Two steps were key for the correct synchronisation between devices (i.e. caregiver' and infant' devices). First, the devices would need to be turned on (on but not recording) to find the GPS signal. The GPS acquisition not only allowed the devices to acquire the real date and UTC time but also to find and pair with each other. After successful pairing, the devices were ready to start recording (on and recording). The instruction for initiating recording (on and recording) always originated from the master device (the caregivers'), which was programmed to transmit a signal to the infants' device, ensuring synchronized data recording down to the millisecond for both infant and maternal datasets.

3.3.3.2 Vocal affect coding

To classify audio segments into vocalisations produced by the key child (i.e. infant wearing the wearable device), we used a Voice Type Classifier (VTC) for child-centred daylong recordings (Bredin et al., 2020; Lavechin et al., 2020). Briefly, this is an open-

source speech processing model that classifies audio segments into key-child vocalisations, other child vocalisations, adult male speech, and adult female speech. The general architecture of the classifier as well as the process followed to train the model are presented in more detail in Lavechin et al., (2020). Importantly, the model was trained with 250+ hours of recordings of children growing up in multilingual settings – covering 10 different languages - with a wide variety of typological characteristics.

In the current study, we focused on key-child vocalisations. To obtain these, we segmented each recording into one-hour segments and analysed each segment of each recording with the VTC which returned a percentage confidence estimate, indicating the probability the key child was vocalising (or not) for every 10ms frame. Next, we hand coded six hours of data, covering both timepoints, and labelled the speaker every 500 milliseconds. Following this, we compared the estimates from the VTC against the hand coding, which allowed us to ascertain the precision/recall we could expect from each choice of VTC confidence estimate threshold (see APPENDIX A, SM1). For this paper, we chose a relatively high confidence estimate threshold, which yield a precision of 0.8 (i.e. the proportion of identified vocalisations which really belong to our class of interest is roughly 80%, according to our hand coding) and a concomitant recall of 0.3 (i.e. our analyses are based on 30% of the total vocalisations present in the data). Of note, a precision of 0.8 aligns with that of other widely used systems such as LENA© (e.g. Cristia et al., 2021). This way, we were able to extract moments when the VTC was almost sure that the key-child was vocalising, accepting that our analyses are based only on a subset of the vocalisations present. In total, 49110 vocalisations were identified, with a mean (std) of 518.2 (255.59) per participant at 5 months and 546 (232.73) at 10 months. Of note, our results for how infant and caregiver arousal changes around infant vocalisations are very similar to those based on continuous recording, based on hand coding (Wass et al., 2022).
Once we identified the key-child vocalisations, we analysed their emotional characteristics using a Speech Emotion Recognizer (SER) developed by E.V (Vaaras et al., 2023). Details of the model are presented in detail in Vaaras et al., (2023). Briefly, the SER model is a support vector machine classifier, which was trained following Section 4.3.1. in Vaaras et al., (2023). The input features for the SER model were utterance-level eGeMAPS features (Eyben et al., 2016) extracted from the .MP3 files using the openSMILE toolkit (Eyben et al., 2013). For training data, 24 random one-hour segments were selected, and 7 different independent annotators performed affect labelling on all infant vocalisations occurring during these hours, labelling each one of five categories (very negative to very positive). These five categories were subsequently collapsed to three categories (negative, neutral, and positive) for training and testing the SER support vector machine model. Due to the fact that the SER classifier could not accurately distinguish positive from neutral affect, these were pooled into one class (i.e. neutral-to-positive affect, henceforth referred to as neutral affect). The five-fold crossvalidation process was carried out with a distinct testing set left out in each iteration, yielding a precision of 66% for infant negative vocalizations and 61% for neutral categories (similar to Cristia et al., 2021; Fan et al., 2021) (see APPENDIX A, SM1 for more details on the confusion matrix used for evaluating the performance of the SER classifier).

3.3.3.3 Pre-processing of vocalisations and autonomic measure

Each infant vocalisation was identified in the ECG, proximity and activity datasets by extracting the onset of each vocalisation (in milliseconds) and finding it in the respective datasets. For each vocalisation, we first checked the proximity levels between caregiver and infant and discarded any vocalisations that occurred when the proximity between the

caregiver-infant dyad was greater than 10 metres or when there was no proximity data (i.e. the proximity was not "in range"; see APPENDIX A, Table S2). This allowed us to make sure we measured co-regulatory processes within direct interpersonal interactions between the infants and their caregivers.

Second, we extracted the heart rate data before and after each vocalisation onset. We excluded all vocalisations that happened within the first and last 10 minutes of the recording as well as all the vocalisations that had more than or equal to 80% of the data missing (i.e. the ECG leads were not detected for more than 80% of the 20 minutes epoch around the vocalisation) (see APPENDIX A, Table S2). Next, we detrended the ECG signal and performed R-peak identification using the in-built MATLAB function 'findpeaks'. The minimum peak height was defined as a simple amplitude threshold. Minimum peak distance, instead, was set at 240ms for infants and 300ms for caregivers (corresponding to a maximum heart rate of 250 beats-per-minute (BPM) for infants and 150 BPM for caregivers) and used to improve the performance of 'findpeaks'. Following this, automatic artefact rejection was performed. First, we applied a maximum temporal threshold check to exclude those R peaks occurring within more than 900ms (for infants) and 1500ms (for caregivers) since the previous R peak (corresponding to a minimum heart rate of 66 BPM for infants and 40 BPM for caregivers). Second, we applied a maximum rate of change check and excluded those beats showing a rate of change of IBI greater than 3 std from the mean. Next, we converted IBI values into beats-per-minute (BPM) values and removed outliers. These were defined based on the derivative of the time series and identified using a moving window method from MATLAB ("isoutlier" with "movemedian"). If the number of consecutive outliers was lower than 30 samples, outliers were interpolated using the MATLAB in-built function 'fillmissing'. If the gap of missing data was greater, data was left as NaN. Following this, the BPM dataset was down sampled into one-second epochs and normalised (i.e. converted to z-scores).

To process the other two datasets (i.e. proximity and actigraphy) we selected all the preprocessed vocalisations and extracted the proximity and actigraphy data before and after the onset of each of these vocalisations. First, we interpolated missing values when the gap was equal or less than 20 seconds. Second, similar to Salo et al., (2021), to mitigate the impacts of random, brief fluctuations in the data, a moving average was calculated using a ± 3 s window. For the actigraphy data we first applied a low-pass filter with a cutoff of 0.1Hz to remove high-frequency noise (Wass et al., 2019). Next, we calculated the derivative (i.e. jerk) of all three-axis accelerometer recordings (i.e. X, Y and Z axes). We then calculated the average between the three axes and down sampled the data to 1Hz by calculating the mean from all the readings within each second. Finally, we normalised the data by converting it to z-scores.

Ultimately, to ensure good quality of the data, we applied one last filter and excluded from all datasets those vocalisations that had more than 60% of the ECG data missing (i.e. the ECG leads were not detected for 40%, or more, of the 20-minute time window). All analyses involving arousal are performed using normalised (i.e. z-scored) heart rate (BPM) data.

3.3.3.4 Calculation of significance: Cluster-based permutation

To estimate the significance of the time-series relationships in the results, we used two different types of cluster-based permutation tests: 1D and 2D cluster-based permutation tests.

3.3.3.4.1 1D cluster-based permutation

We employed this test in Analysis 2, Analysis 3.1 and Analysis 4.1. To do so, we used the function "ft_timelockstatistics" from FieldTrip (Maris & Oostenveld, 2007;

<u>http://fieldtriptoolbox.org</u>) to calculate the cluster-based test statistic (cfg.correctm = 'cluster') using the Monte Carlo method (cfg.method = 'montecarlo'). This nonparametric framework allowed us to both control for the multiple comparison problem that arises from the fact that the effect of interest (e.g. difference in arousal between different types of vocalisations) is evaluated many times, and to reduce the potential for false negative effects (Meyer et al., 2021).

3.3.3.4.2 2D cluster-based permutation

We employed this test in Analysis 4.2 (see section 3.3.3.8.2). This test builds on the assumption that effects are clustered along two dimensions of interests: (a) time (in our case, the 20 minutes around vocalisation onset), and (b) space (in our case, the continuum from negative to positive lags). Similar to 2.3.4.1, the 2D cluster-based permutation test realises its control of the multiple comparison problem while maximising power, using code adapted from Cohen (2014) (https://github.com/mikexcohen/ANTS youtube videos/tree/main/ANTS8 stats).

Briefly, we created two null-hypothesis distributions: 1. There is no difference between the windowed cross-correlation values at 5 months compared to 10 months; and 2. There is no difference between the windowed cross-correlation values contingent on timing (i.e. comparing positive and negative lags). First, we randomly swapped the data across the different 'conditions' (i.e. data at 5 and 10 months, or data at positive and negative lags) to create one null-hypothesis test statistic value. We then repeated this procedure 1000 times to create a distribution of test statistic values observed under the null hypotheses. To assess significance, we took the largest cluster in the observed data and the largest cluster in each random shuffle iteration. If the size of the cluster in the observed data fell below a critical level of 0.05, the null hypothesis of exchangeability of the data over the two conditions was very unlikely and rejected, hence the test result was considered significant (Cohen, 2014; Meyer et al., 2021).

3.3.3.5 Analysis 1: Descriptive analyses of the vocal data

For this and all analyses a rationale for why each analysis was conducted is given in Results section and has not been repeated here for reasons of space. For Analysis 1, our main aim was to explore between- and within-group differences in the frequency (i.e. count) and duration of the vocalisations overall and also split by affect (negative versus neutral). For visualisation purposes, we also calculated the percentage of negative and neutral vocalisations per hour at both time points. To test for differences in the frequency of neutral and negative vocalisations within and between age groups, we performed onesample (to test for differences within the same age group) and two-sample (to test for differences between the two age groups) t-tests. We used the R function "Ime" to perform a linear mixed effects analysis of the relationship between vocalisation durations (response variable), Affect, Age (predictor variables) and the interaction between Affect x Age, with random effects of subject (grouping variable) (refer to APPENDIX A, Table S3 for more details on the model specification). Briefly, linear mixed effect models are an extension of simple linear models to allow both fixed and random effects. These models are particularly useful when there is non-independence in the data as they allow us to account for both by-item variation (e.g. durations of all the vocalisations) as well as by-subject variation (e.g. the fact that some vocalisations belong to the same subject).

3.3.3.6 Analysis 2: Infant arousal around different types of vocalisations

We explored whether there were differences in arousal around negative compared to neutral vocalisations between and within the two ages. First, we examined differences in infant arousal *at the time* of the infant's vocalisations. To do this, we took the heart rate data at the time of the event (window length of 1 second) and performed a linear mixed effect model to examine whether there were significant differences between affect (i.e. arousal at the time of neutral compared to negative vocalisations) and across time (e.g. infant arousal at the time of negative vocalisations at 5 months versus 10 months) (refer to APPENDIX A, Table S7 for more details on the model specification). For visualisation purposes, we calculated the distribution of these data.

Second, we took the heart rate activity data *during* the time period before and after the vocalisations. We split the data by type of vocalisations and averaged at subject level. Next, we averaged across subjects to create a group average heart rate response to different types of vocalisations. We calculated these responses for both types of vocalisations (negative and neutral vocalisations) and groups (5 and 10 months). In order to test for differences within (e.g. infant arousal responses to negative compared to neutral vocalisations) as well as between groups (e.g. infant arousal responses to infants' negative vocalisations at 5 months compared to 10 months), we performed a cluster-based permutation test (see methods 3.3.3.4.1).

Third, we examined infant autonomic reactivity. We defined infant autonomic reactivity as the change in infants' heart rate relative to their own vocalisations and calculated it by taking the average heart rate during the 1-minute period following the vocalisation and subtracting it from a baseline period, taken as the average heart rate in the 10 minutes before the vocalisation onset. We calculated this measure for both categories of affect at both time points and tested for significance within- (i.e. infant autonomic reactivity to neutral compared to negative vocalisations) and between-groups (i.e. infant autonomic reactivity to neutral compared to negative vocalisations at 5 months compared to 10 months) using linear mixed effect models (refer to APPENDIX A, Table S8 for more details on the model specification). Prior to this, we calculated whether infant autonomic reactivity to different

types of vocalisations was significantly different to zero (i.e. no significant change) with a single-sample t-test.

3.3.3.7 Analysis 3: Caregivers' responsiveness to negative and neutral vocalisations and its association with infants' recovery

3.3.3.7.1 Analysis 3.1. Caregiver responsiveness to infants' vocalisations

First, we explored whether there were differences in caregiver arousal *at the time* as well as *during* negative compared to neutral infant vocalisations between and within the two time points. We followed the same procedure as in Analysis 2 (refer to APPENDIX A, Table S10 for more details on the model specified to test caregiver arousal *at the time* of a vocalisation as a function of Affect, Age and Affect x Age).

Second, we examined caregiver autonomic responsiveness. We defined caregiver autonomic responsiveness as the change in caregiver heart rate relative to infant vocalisations and calculated it by taking the average heart rate during the 1-minute period following the vocalisation and subtracting it from a baseline period, taken as the average heart rate in the 10 minutes before vocalisation onset. Next, we tested for significant changes following the same set of analyses as for infant autonomic reactivity (Analysis 2) (refer to APPENDIX A, Table S11 for more details on the model specified to test caregiver autonomic responsiveness as a function of Affect, Age and Affect x Age). We also calculated whether caregiver autonomic responsiveness to different types of vocalisations was significantly different to zero (i.e. no significant change) with a single-sample t-test.

We also looked at caregiver-child proximity around vocalisations as another proxy of caregivers' responsiveness. To do this, we selected the proximity data around all vocalisations, split them by affect and averaged the data at subject level. Next, we averaged across subjects to create a group average proximity response to the two types of vocalisations. We calculated these responses for both types of affect (negative and neutral vocalisations) and age groups (5 and 10 months). Finally, we examined for both significant differences within- (e.g. proximity responses to negative compared to neutral vocalisations) and between-groups (e.g. proximity responses to negative vocalisations at 5 months compared to 10 months). To do so, we performed a cluster-based permutation test by following the same procedure as in methods 3.3.3.4.1.

3.3.3.7.2 Analysis 3.2. Caregiver's autonomic responsiveness and infant's recovery

Here we asked whether caregiver autonomic responsiveness to negative vocalisations from the infant is associated with bigger decreases in infant arousal. To do so, we first selected all the negative infant vocalisations that occurred during the day. Next, we calculated infant heart rate changes relative to these vocalisations in the same way as we calculated infant autonomic reactivity (described above in Analysis 2) and extended it to five 1-minute windows that started after vocalisation onset. We treated this 5-minute window as our "recovery window". To assess how caregiver autonomic responsiveness related to infants' changes in heart rate, we calculated the correlation between the two measures across the recovery window. Results were subsequently corrected for multiple comparisons using the Benjamini and Hochberg (BH) method which controls the false discovery rate (FDR). Briefly, FDR is the expected proportion of false positives among all positives which rejected the null hypothesis and not among all the tests undertaken (Jafari & Ansari-Pour, 2019).

Finally, we examined infants' recovery across the 5-minute recovery window and asked whether there was any association between caregiver autonomic responsiveness and the speed at which the infants recovered. First, we compared the infant average heart rate across the recovery window within (i.e. from one 1-minute window to the next one) and between groups using one- and two-samples t-test respectively. Again, we adjusted the p-values following the BH-FDR method described above. Next, we calculated the recovery line. To do so, we calculated the difference between the infant average heart rate at the time of the vocalisation (i.e. 1st minute after vocalisation onset) and the time at which the average heart rate stopped decreasing within the recovery window (i.e. heart rate decreased significantly until the fourth minute at 5 months and until the fifth minute at 10 months). We then estimated the slopes of these recovery lines and tested whether there were significant differences as a function of age (refer to APPENDIX A, Table S13 for more details on the model specified to test speed of recovery of infants as a function of age). Finally, we explored the association between the slopes of the recovery line (i.e. speed of infants' recovery) and caregivers' autonomic responsiveness by performing a correlation between the two measures at both time points. We repeated this last analysis using a recovery line from the 1st to the 5th minute (instead of 4th) at 5 months to make sure our decision to use different end-times to calculate the recovery lines was not changing the results.

3.3.3.8 Analysis 4: Concurrent and sequential infant-caregiver arousal synchrony around different types of vocalisations from the infant

We examined the degree of concurrent (Analysis 4.1) and sequential (Analysis 4.2) infant-mother synchrony in physiological arousal. Concurrent synchrony ('when X is high, Y is high') is undirected (A \rightarrow B is indistinguishable from B \rightarrow A). Sequential synchrony ('changes in X forwards-predict changes in Y') is directional (A \rightarrow B \neq B \rightarrow A).

3.3.3.8.1 Analysis 4.1. Concurrent synchrony

First, we calculated the co-fluctuation (concurrent synchrony) of infant-caregiver arousal around vocalisation onset, using a sliding window correlation (SWC) method. Before computing the SWC, we detrended the 20-minute signal. In the SWC, a temporal window of a certain size is selected, and the zero-lag correlation coefficient between two signals of interest within that window is computed. Afterwards, the window is shifted (slided) by some offset, and this process is repeated for the entire length of the signal. In Analysis 4.1, we computed the SWC for every vocalisation separately using a window length of 120 seconds with an overlap of 60 seconds. Next, we averaged the results across subjects and averaged again at group level to obtain the final results around neutral and negative vocalisations respectively. Despite the popularity of the SWC method, results are strongly dependent on window length (Shakil et al., 2016), thus, we performed a sensitivity analysis for temporal sliding windows of different lengths and overlaps, which we report in the SM (APPENDIX A, Figure S4). To explore differences in concurrent synchrony in arousal around negative compared to neutral vocalisations, we performed a cluster-based permutation test (see methods 3.3.3.4.1).

3.3.3.8.2 Analysis 4.2. Sequential synchrony

Second, we also explored the sequential synchrony between infant and caregiver arousal around negative and neutral vocalisations. One way to examine how the strengths and lags of association between two time series are changing over time is to use short overlapping intervals of data from each time series and estimate the cross-correlation within these shorter windows (Boker et al., 2002). This method is known as window cross-correlation (WCC) and has the advantage of only making an assumption about local stationarity rather than assuming stationarity over the whole time series (Boker et al., 2002). Here we performed a WCC analysis to examine sequential synchrony in arousal between infants and their caregivers. We calculated the WCC for every vocalisation separately using a window length of 120 seconds with an overlap of 60 seconds and a lag of ± 30 seconds. We then estimated the lag at which the association between the two timeseries was at its highest (i.e. we found the lag that corresponded to the maximum crosscorrelation value) for each of the time windows and stored them on a separate matrix. Next, we averaged the results from the WCC and the peak lag analysis across subjects and averaged again at group level. Each of the parameters used in calculating the WCC has consequences (Boker et al., 2002). Thus, in the absence of previous research that could guide our decisions, we performed a sensitivity analysis for temporal sliding windows of different lengths and overlaps, which we report in the SM (APPENDIX A, Figure S5).

Finally, we wanted to compare for differences within- (e.g. whether the cross-correlation values were different at positive compared to negative lags) and between-groups (e.g. whether the cross-correlation values were higher at 5 months compared to 10 months). We did so by performing two 2D cluster-based permutation tests respectively (see methods 3.3.3.4.2).

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3.4 Results

Our results section is structured as follows. First, we explore how naturalistic vocalisations change across the first year of life, comparing the frequency and durations of negative affect and neutral affect vocalisations (Analysis 1). Our prediction based on prior research (see Introduction) was that negative infant vocalisations would become more short-lived with increasing age. Next, we examined arousal around negative compared to neutral infant vocalisations (Analysis 2). We predicted that, if negative infant vocalisations are intended to elicit co-regulation, then increases in arousal would be greater for negative affect compared with neutral affect vocalisations. We predicted that this would not change as a function of age.

Next, we examined caregiver responses to negative infant vocalisations, with the aim of differentiating between two hypotheses. Hypothesis 1 is that infants become better at self-regulation with age (i.e., that infant recovery becomes less dependent on caregiver responses over time). Hypothesis 2 is that developmental changes are attributable to enhanced caregiver-infant co-regulation rather than to improvements in infant self-regulation alone. To test this, we first examined caregiver responses to negative infant vocalisations (Analysis 3.1) and explored the association between caregiver autonomic responsiveness and subsequent infant recovery (Analysis 3.2). We predicted that, if Hypothesis 1 were true, then caregivers would show decreased responses to negative infant vocalisations, and that the association between caregiver synchrony in arousal around negative infant vocalisations. We predicted that, if Hypothesis 1 were true, then increases in concurrent and sequential synchrony around negative infant vocalisations would become less strong with increasing age. Alternatively, if the improvement over

developmental time lies in the quality of the dyadic relationship leading to enhanced caregiver-infant co-regulation (Hypothesis 2) our prediction would be the opposite.

3.4.1 Analysis 1: Descriptive analyses of the vocal data

At both time points, infants produced more negative vocalisations compared to neutral vocalisations ($t_{5m}(56) = 6.24$, p<0.001 and $t_{10m}(31) = 5.44$, p<0.001; APPENDIX A, Figure S1). We found no evidence that 10-month-old infants produced more vocalisations than 5-month-old infants overall (t_{all}(87)=-1.06, p=0.29), nor split by affect (t_{neg}(87)=-1.06, p=0.29, $t_{neutral}(87)=-0.62$, p=0.54). Next, we tested the effect of Affect and Age group as well as the interaction between the two on the average durations of infant vocalisations. The model resulted in a significant effect of Affect (β = -0.217, P < 0.001), a significant effect of Age (β = -0.183, P=0.02), and a significant Affect × Age interaction effect (β = 0.106, P < 0.001; refer to APPENDIX A, Table S3 for more details on the model specification and results). Following this, we explored the main effect of affect and age separately. We observed a main effect of *Affect* at 5 months, (β_{5M} = -0.111, p_{5M} <0.001), with negative vocalisations lasting longer than neutral vocalisations, but not at 10 months $(\beta_{10M} = -0.004, p_{10M} = 0.834)$. Second, we explored the effect of Age on the average duration of a vocalisation (irrespective of the affect). We found no significant Age effects $(\beta = -0.142, p = 0.07)$. Next, we examined differences in the average duration of the vocalisations split by affect. There were no significant differences in the duration of neutral vocalisations between 5 and 10 months ($\beta_{neutral} = -0.036$, $p_{neutral} = 0.652$). However, we did find a significant difference in the duration of negative vocalisations between ages $(\beta_{neg} = -0.193, p_{neg} = 0.024)$. Five-month-old infants produced longer negative vocalisations than 10-month-old infants. Overall, negative vocalisations at 5 months were not only longer than neutral vocalisations at 5 months but also significantly longer than

negative vocalisations at 10 months. Refer to APPENDIX A, Tables S4-S6 for more descriptive analyses on infant vocalisations.

3.4.2 Analysis 2: Infant arousal around different types of vocalisations

Next, we examined infant arousal around negative compared to neutral infant vocalisations. We did this in two ways. First, by examining infant arousal levels *at the time of* the vocalisation onset (Figure 3.2). Second, by examining infant arousal levels *during* the period before and after each vocalisation onset (Figure 3.3). Lastly, we examined infant autonomic reactivity to their own different types of vocalisations.

We tested the effect of Affect and Age group as well as their interaction effect on infant arousal *at the time* of a vocalisation. The model resulted in a significant effect of *Affect* (β = -0.068, *p*= 0.044) and a significant effect of *Age group* (β = -0.153, *p*= 0.004). There was no significant *Affect* × *Age group* interaction effect (β = 0.009, *p* < 0.699; refer to APPENDIX A, Table S7 for more details on the model specification and results). At 5 months and 10 months, infants had significantly higher arousal at the time of negative compared to neutral vocalisations (β_{5M} = -0.058, p_{5M} <0.001; and β_{10M} = -0.049, p_{10M}= 0.013; Figure 3.2A, C, E, G and I). For both negative and positive vocalisations, infant arousal was significantly higher at 5 months compared to 10 months ($\beta_{neutral}$ = -0.131, p_{neutral} = 0.012; and β_{neg} = -0.160, p_{neg}= 0.005).





A-B: show results from the linear mixed effect analyses comparing infant (A) and caregiver (B) arousal at the time of negative and neutral vocalisations for both age groups. Asterisks indicate significant differences between different groups (*p<0.05, **p<0.01, ***p<0.001). Data is plotted at the vocalisation (instead of subject) level. Vertical bars represent SEM. C-J: Histograms showing the distribution of arousal at the time of infants' negative and neutral vocalisations at 5 and 10 months. Infant data are represented in darker colours. Caregiver data are represented in shaded colours. The right side shows arousal levels relative to negative (C and D) and neutral (E and F) vocalisations at 5 months. The left shows arousal levels relative to negative (G and H) and neutral (I and J) vocalisations at 10 months.

Next, we examined arousal changes during the 10-minute period before and after each vocalisation (Figure 3.3) (This choice of time window was arbitrary, and our analyses were designed so that the significance of results could in no way depend on the length of time window used; see Methods). First, we compared arousal changes around negative versus neutral vocalisations, separately for the two age groups. At 5 months (Figure 3.3A), a cluster-based permutation test found significantly increased arousal right before negative compared with neutral vocalisations onsets, and significantly decreased arousal

during the periods after the vocalisation (p<0.025, two sided) (This finding that arousal decreases faster following negative vocalisations is consistent with previous findings (Wass et al., 2023), and likely to be because negative vocalisations elicit co-regulation). At 10 months (Figure 3.3C), increased arousal was found for positive compared to negative vocalisations around 6 minutes prior to the vocalisation, and increased arousal for negative compared to positive vocalisations was observed immediately before and after the vocalisation onsets (p<0.025, two sided). Next, we examined whether these arousal changes around negative and positive vocalisations differed by age. A cluster-based permutation test found no significant differences between ages for any of the two types of vocalisations.



Figure 3. 3. Arousal *during* the period from 10 minutes before to 10 minutes after specific infant vocalisations.

Infant (A, C) and caregiver (B, D) arousal around negative (in red) and neutral (in green) vocalisations at 5 months, and at 10 months respectively. Shaded areas show standard error. Straight lines are negative (in red), and positive (in green) significant clusters (p<0.025, two sided) identified using a cluster-based permutation test.

Overall, these results suggest that infant arousal increases are greater around negative compared with neutral vocalisations at both 5 months and 10 months. For both negative and positive vocalisations, infant arousal at the time of the vocalisation was higher at 5 months compared with 10 months.

Lastly, we examined infant autonomic reactivity (i.e. change in infant heart rate) to their own different types of vocalisations (see methods 3.3.3.6; Figure 3.4A). Infant autonomic reactivity was significantly different from zero for both affects and at both time points $(t_{5m neg}(48) = 9.14, p < 0.001; t_{5m neutral}(48) = 10.56, p < 0.001; t_{10m neg}(25) = 6.56, p < 0.001;$ $t_{10m neutral}(25) = 5.8$, p<0.001). Next, we tested the effects of Affect, Age group, and the interaction between the two on infant autonomic reactivity. Results showed no significant effect of Affect (β = 0.047, p= 0.085) or Age group (β = -0.079, p= 0.083), but a significant Affect \times Age group interaction effect (β = -0.05, p= 0.009; refer to APPENDIX A, Table S8 for more details on model specification and results). Autonomic reactivity to neutral vocalisations was significantly reduced at 10 months compared to 5 months (β_{Age} = -0.128, $p_{Age} < 0.002$; Figure 3.4A), and was lower compared to negative vocalisations at 10 months but not at 5 months (β_{10M} = -0.055, p_{10M} < 0.001; Figure 3.4A). Infant autonomic reactivity to negative vocalisations, instead, was comparable across ages. Importantly, this pattern in infant autonomic reactivity did not translate into significant changes in infant activity levels (see APPENDIX A, Table S9 where we explore whether changes in infant arousal were parallel to changes in infant activity levels).



Figure 3.4. Infant autonomic reactivity and caregiver autonomic responsiveness to different types of vocalisations from the infants at 5 and 10 months.

Plot A) Shows infant autonomic reactivity to infant vocalisations at 5 (left) and 10 months (right). B) Shows caregiver autonomic responsiveness to infant vocalisations at 5 (left) and 10 months (right). Asterisks indicate significant differences between different groups (*p<0.05, **p<0.01, ***p<0.001). Vertical bars represent SEM. Alpha symbols (α) indicate significant differences (<0.05) from zero (i.e. no change).

3.4.3 Analysis 3: Caregiver responsiveness to negative and neutral vocalisations and its associations with infant' recovery

Building on Analysis 2, we explored changes in caregiver arousal and child-caregiver proximity in response to infant vocalisations (Analysis 3.1) as well as the association between caregiver autonomic responsiveness with infant subsequent recovery (Analysis 3.2).

3.4.3.1 Analysis 3.1. Caregiver responsiveness to infants' vocalisations

First, we examined how caregivers responded to naturally occurring vocalisations from the infant. First, we examined caregiver arousal *at the time of* the vocalisation onset (Figure 3.2 B, D, F, H and J). Second, we examined caregiver arousal levels *during* the period before and after each vocalisation onset (Figure 3.3B and D). Third, we calculated

the change in caregiver arousal (Figure 3.4B) and reduction in child-caregiver proximity (Figure 3.5) during the time window before and after infant vocalisations.

We investigated how caregiver arousal *at the time of* an infant vocalisation was influenced by *Affect*, *Age group*, and the interaction between the two. Our results showed no significant effects of *Affect* (β = -0.032, p= 0.316), or *Age group* (β = -0.012, p= 0.666), and no significant *Affect* × *Age* interaction effect (β_{Affect} *Age = -0.007, p_{Affect}*Age = 0.759; refer to APPENDIX A, Table S10 for more details) (Figure 3.2B). When examining caregiver arousal changes *during* the time period before and after infant vocalisations, we observed increased caregiver arousal following negative compared with neutral infant vocalisations at both time points (Figure 3.3B and D). Interestingly, caregiver arousal was more elevated during the time period immediately preceding a negative compared to neutral vocalisations at 5 months but not at 10 months (Figure 3.3B). A cluster-based permutation test found no significant differences between ages for any of the two types of vocalisations.

Next, we examined caregiver autonomic responsiveness (i.e. change in caregiver heart rate) to infant vocalisations (see Methods 3.3.3.7.1). At 5 months, caregivers showed significant arousal increases following both negative and neutral vocalisations ($t_{5m_neg}(48)=4.46$, p<0.001; $t_{5m_neutral}(48)=2.34$, p<0.05). At 10 months, caregiver autonomic responsiveness did not show significant increases following either type of vocalisation ($t_{10m_neg}(25)=1.57$, p=0.13; $t_{10m_neutral}(25)=0.16$, p=0.87; Figure 3.4B). Next, we investigated how Affect, Age group, and the interaction between the two, influenced caregiver autonomic responsiveness to infant vocalisation. Our analysis showed no significant effects of *Affect* (β =-0.019, p=0.47), or *Age group* (β =-0.035, p=0.287) and no significant *Affect* × *Age* interaction effect ($\beta_{Affect}*_{Age}=-0.02$, p_{Affect*Age}=0.3; refer to APPENDIX A, Table S11 for more details on model specification and results). Caregivers did not show larger autonomic responses following vocalisations from

younger compared with older infants, for either negative or neutral vocalisations. Refer to APPENDIX A, Table S12 for changes in caregiver levels of activity relative to infant vocalisations.

Next, we examined changes in caregiver-child proximity around infant vocalisations (Figure 3.5). At 5 months, significantly (p<0.025, two sided) increased proximity (i.e. dyad was closer to each other) was observed for negative compared with neutral vocalisations from -0.7 to 10 minutes after infant vocalisations (Figure 3.5A). At 10 months, increased proximity was observed for negative compared with neutral vocalisations from 1.2 to 3.2 minutes (Figure 3.5B). Finally, we compared proximity levels between the two time points. At 5 months, infants and caregivers were significantly closer to each other than at 10 months throughout the 20-minute time window analysed, irrespective of affect (see APPENDIX A, Figure S2).





Proximity levels around negative (in red) and positive (in green) vocalisations at 5 months (A) and 10 months (B). Units are in metres. Shaded areas show SEM. Straight lines are negative (in red), and positive (in green) significant clusters (p<0.025, two sided) identified using a cluster-based permutation test.

Overall, these results suggest that caregivers did not show larger autonomic responses following vocalisations from younger compared with older infants, for either negative or neutral vocalisations. We did, however, observe more long-lasting increases in childcaregiver proximity at 5 months compared with 10 months.

3.4.3.2 Analysis 3.2. Caregiver's autonomic responsiveness and infant's recovery

Next, we asked whether greater increases in caregiver arousal following infant vocalisations were associated with bigger decreases in infant arousal during the recovery window (see Methods section 3.3.3.7.2 for details of how this analysis was conducted).

Greater increases in caregiver arousal were associated with greater increases in infant arousal during minute 1 of the recovery period at both 5 months (R= 0.094, BH adjusted p<0.001, Figure 3.6A) and 10 months (R= 0.053, BH adjusted p<0.001, Figure 3.6F) (Presumably driven by parents reacting more to episodes of greater infant distress). Later during the recovery window, however, this relationship reversed, so that greater increases in caregiver arousal were associated with greater decreases in infant arousal (replicating previous findings (Wass et al., 2019)). This finding was significant for the 3rd (R= -0.046, BH adjusted p<0.001) and 4th (R= -0.036, BH adjusted p<0.001) minute of the recovery window at 5 months (Figure 3.6C, D and K), and for the 5th minute of the recovery window at 10 months (R= 0.045, BH adjusted p<0.001) (Figure 3.6J and K).





Correlations between caregiver autonomic responsiveness around the onset of the vocalisation and changes in infant arousal at different time windows across the recovery window at 5 months (A-E) and 10 months (F-J). K) is showing the recovery window (minutes post-event) on the x axis and the R value of the correlation analysis between caregiver autonomic responsiveness and changes in infant arousal on the y axis. Asterisks indicate significant correlations (* BH adjusted p< 0.05, ** BH adjusted p<0.01, *** BH adjusted p<0.001).

Next, we examined infant arousal across the 5-minute recovery window. We found that both age groups showed a significant steady decrease in arousal throughout the recovery window (Figure 3.7A) but identified no significant age differences in the speed at which infants recovered (more in Methods 3.3.3.7.2, Figure 3.7B and C) (β = 0.075, p = 0.145; refer to APPENDIX A, Table S13 for more details on model and results).

Finally, we asked whether there was any association between caregiver autonomic responsiveness and infants' speed of recovery (see Methods 3.3.3.7.2). We found no significant relationship at 5 months ($R_{5m} = -0.181$, p = 0.214) (Figure 3.7D). However, at 10 months, there was a significant association, such that the more caregivers increased their arousal relative to the event (i.e. higher caregiver autonomic responsiveness) the steeper the recovery line was (i.e. infants recovered faster) ($R_{10m} = -0.545$, p = 0.004)

(Figure 3.7E). We repeated this last analysis using a recovery time window from the 1st to the 5th minute (instead of 4th) at 5 months (see Methods 3.3.3.7.2). Results stayed the same ($R_{5m} = -0.129$, p = 0.378) (APPENDIX A, Figure S3).



Figure 3.7. Infant recovery and its relationship with caregiver autonomic responsiveness. Plot (A) Average arousal across the 5-minute recovery window. Light red shows data at 5 months and dark red shows data at 10 months. Asterisks indicate significance (* BH adjusted p< 0.05, ** BH adjusted p<0.01, *** BH adjusted p<0.001). Slopes of the recovery lines at 5 months, calculated from the 1st to 4th minute from the recovery window (B), and at 10 months, calculated from the 1st to 5th minute from the recovery window (C). Results showing the association between caregivers' autonomic responsiveness relative to the onset of a negative infant vocalisation and infants' recovery indexed as the slope of the recovery line at 5 months (D), and at 10 months (E).

In the Supplementary Materials (APPENDIX A, Figure S4), we repeated the entirety of Analysis 3.2 using a random sample of 26 infants (matched gender) from the 5-month group to ensure that the mismatched number of participants in the 5-month and 10-month groups did not influence our results. The results of these control analyses were very similar to those presented here in the main text.

Overall, we found that both groups showed a significant decrease in arousal following negative vocalisations (Figure 3.7A) and that these decreases correlated significantly with caregiver autonomic responsiveness (Figure 3.6K). We found no evidence that older infants recovered faster (Figure 3.7B and C). However, increased caregiver responsiveness was associated with faster recovery for older, but not younger, infants (Figure 3.7D and E).

3.4.4 Analysis 4: Concurrent and sequential infant-caregiver arousal synchrony around different types of vocalisations from the infant

3.4.4.1 Analysis 4.1. Concurrent synchrony

To calculate the co-fluctuation of infant-caregiver arousal around infant vocalisations we used a sliding window correlation (SWC) (see Methods 3.3.3.8.1). Figure 3.8 shows the results from the SWC analysis using a window length of 120s with an overlap of 60s. In contrast with previous findings (Wass et al., 2019, 2023), we found no evidence of increased concurrent caregiver-infant arousal synchrony around negative vocalisations compared to positive-to-neutral vocalisations at any time point (Figure 3.8). These results were replicated using different temporal windows and overlaps (see APPENDIX A, Figure S5).



Figure 3.8. Concurrent synchrony in infant-caregiver arousal.

Results from the SWC analysis using a window length of 120s with an overlap of 60s at 5 months (A) and at 10 months (B). Shaded areas show standard error. Straight lines are significant clusters identified using a cluster-based permutation test.

3.4.4.2 Analysis 4.2. Sequential synchrony

We estimated the sequential synchrony between caregiver and infant arousal around negative vocalisations using a windowed cross-correlation (WCC) and peak picking method (Boker et al., 2002) (see Methods 3.3.3.8.2). Here we present the results from the WCC using a window length of 120s with an overlap of 60s (Figure 3.9A and D); in APPENDIX A we present additional analyses using different time window widths and overlaps (APPENDIX A, Figure S6). When the correlation values are stronger at negative lags, this indicates that changes in the infants' physiology forward-predict changes in caregiver arousal. Instead, when the correlation values are stronger at positive lags, this indicates that changes in the caregivers' physiology forward-predict changes in the infant arousal.

First, we compared the results from the WCC analysis between the two age groups. The cluster-based permutation test (see methods 3.3.3.4.2) revealed no significant clusters. This indicates that, overall, the lead-lag relationships of arousal changes around negative infant vocalisations did not vary significantly between age groups. Second, we tested for within-group differences and examined the strength of the correlation values at negative lags (infant \rightarrow caregiver) compared to positive lags (caregiver \rightarrow infant). The cluster-based permutation test found a significant cluster at 5 but not at 10 months, such that the correlation values at negative lags before the onset of the negative vocalisation were significantly stronger than the values observed at positive lags at 5 but not at 10 months (Figure 3.9C F). These results were replicated in all the analyses we ran using different time window widths and overlaps (see APPENDIX A, Figure S6).





Windowed cross-correlation and pick peaking (thick solid line) analyses at 5 months (A) and at 10 months (D). Caregivers' physiology forward-predicting infant arousal on the positive lags, infant's physiology forward-predicting caregiver arousal on the negative lags. Results from subtracting the correlation values at positive lags from the correlation values at negative lags at 5 months (B) and at 10 months (E). Significant clusters identified by the CBP test at 5 months (C) and at 10 months (F).

Overall, we were unable to replicate previous findings that concurrent infant-caregiver synchrony in arousal increased around negative vocalisations (Smith et al., 2022; Wass et al., 2019). However, we did identify changes in sequential synchrony, such that, during the time periods prior to negative vocalisations, caregiver-child arousal changes were significantly more infant-led than caregiver-led at 5 months.

3.5 Discussion

The present study examined bi-directional dynamics between infants and their caregivers around negative and neutral affect vocalisations and considered how these relationships change across the first year of life. To do this we developed wearable devices that allowed us to collect day-long recordings of ECG, proximity, audio, and activity data from 57 caregiver-infant dyads at 5 months and 32 dyads at 10 months.

Our first aim was to test our prediction that negative affect infant vocalisations would become more short-lived with increasing age. Then, if we observed this, we planned to investigate why this might be, by comparing two hypotheses. Hypothesis 1 was that infants would become better at self-regulation with age (i.e., that infant recovery would become less dependent on caregiver responses over time). Hypothesis 2 was that developmental changes would be attributable to enhanced caregiver-infant co-regulation rather than to improvements in infant self-regulation alone.

Analysis 1 revealed that negative vocalisations in our study were more frequent and lasted for longer than positive vocalisations at both time points. We found no age differences in the durations of neutral infant vocalisations, but negative affect vocalisations were shorter at 10 months compared to 5 months. Unexpectedly, we found no evidence that 10-montholds produced more vocalisations than 5-month-olds (Camp et al., 1987; Esteve-Gibert & Prieto, 2013). A possible explanation for these null results might reside in the stringent threshold we chose for the Voice Type Classifier (VTC), such that we only retained vocalisations with the highest confidence estimates returned (see Methods 3.3.3.2). It is also possible that the VTC exhibited higher confidence in detecting negative vocalisations (e.g. cries) compared to neutral ones. Consequently, it is important to interpret these findings within the scope of the present study and avoid using them to address broader developmental questions. Nevertheless, there is no indication to suggest that the classifier's accuracy differed between the two age groups (APPENDIX A, Figure S1), and so the disparities in the duration of negative vocalisations between the ages can meaningfully be interpreted.

Overall, our findings from Analyses 2, 3 and 4 suggest that caregiver involvement in arousal co-regulation following real-world spontaneous negative affect displays

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decreases through infancy. Caregiver arousal levels were greater during negative compared to neutral infant vocalisations at both time points (Figure 3.3B and D). Interestingly, caregiver arousal was more elevated immediately before and after negative vocalisations at 5 months (Figure 3.3B), while at 10 months, this increase was observed only afterwards (Figure 3.3D). We found no evidence that caregivers showed larger autonomic responses following vocalisations from younger compared with older infants (Figure 3.2). However, these responses following both negative and neutral infant vocalisations were significantly different from zero at 5 months but not at 10 months (Figure 3.4B). We also observed greater and more long-lasting increases in caregiverchild proximity following negative vocalisations at 5 months compared to 10 months (Figure 3.5). And when we examined changes in sequential synchrony (Analysis 4.2), we found that during the time periods prior to negative vocalisations, caregiver-child arousal changes were significantly more infant-led than caregiver-led at 5 months (Figure 3.9). This suggests that caregivers of younger but not older infants were more inclined to adjust their responses to their infants' during the time period leading up to a negative vocalisation. Together, these findings suggest that caregiver responses are more specific (and greater) to negative vocalisations at 5 months. Infant autonomic reactivity, on the other hand, was significantly different from zero at both time points for both types of vocalisations. Importantly, there were no differences between infant autonomic reactivity to negative vocalisations at 5 compared to 10 months. Together, these findings suggest that caregivers respond more to negative vocalisations from their infants at 5 months but that this increased caregiver responsiveness does not align with the magnitude of the infants' autonomic reactivity, which was comparable across the two age points (Figure 3.4).

Replicating previous research (Wass et al., 2019), we found that the more caregivers upregulated their arousal around child negative affect vocalisations, the greater the decreases in infant arousal were during the minutes following the event (Figure 3.6K). This relationship was observed at both ages, albeit over slightly different time-frames (3-4 mins post onset at 5 months; 5 mins post onset at 10 months). One possible interpretation to the peak association between increased caregiver autonomic responsiveness and decreases in infant arousal moving from the 3rd to the 5th minute with age might be that dyads with younger infants are more responsive to co-regulation.

In Analysis 4.1 we were unable to replicate previous findings that concurrent infantcaregiver synchrony in arousal increased around naturalistic negative vocalisations (Wass et al., 2019, 2023) (Figure 3.8). There are two possible explanations for these null findings. One is that previous research has defined their events based on certain "intensity" thresholds. For example, Smith et al., (2022) defined their events based on peaks in the infant's arousal. Similarly, Wass et al., (2019, 2023) defined their events based on peaks in both infant arousal and affect and examined only high intensity negative affect vocalisations. In the current study, our events are based exclusively on vocal affect data irrespective of the intensity of the infants' reaction. We know that caregivers respond to approximately 30-50% of infants' prelinguistic syllables (Goldstein et al., 2009) so, one hypothesis is that caregivers might react less to low-intensity infant' vocalisations (Smith et al., 2022; Yoo et al., 2018), allowing infants to attempt to self-regulate rather than intervening (Moore & Calkins, 2004). Thus, intensity could be modulating this relationship and mixing high- with moderate- and low- intensity moments could have blurred our findings. Another explanation might be the use of different time resolutions. In the current study, we worked on very specific and localised responses in time and, consequently, analysed the data on a second-by-second basis. Previous studies have, however, used lower time-resolutions (Wass et al., 2022, 2019). It is possible that selecting more extreme or intense moments and/or using wider time windows could change our results (Palumbo et al., 2017).

So far, we have provided evidence suggesting that, at 5 months, caregivers seem to respond more to their infants' signals than at 10 months. We observed increased caregiver responsiveness at both the physiological (e.g. they were likely the more active partners in synchronising and matching their infants' affect) and physical (i.e. they decreased the distance between them and their infants more) levels. Furthermore, we presented evidence indicating that caregiver responsiveness to infant negative vocalisations is linked to bigger subsequent reductions in infant arousal (Analysis 3.2) and found that the peak association between these two measures is postponed as developmental time progresses (Analysis 3.2). Collectively, these findings seem to suggest that caregiver involvement in co-regulation decreases with developmental time. However, this reduced responsiveness of caregivers to their older infants' signals does not imply that they are less significant in co-regulation. In fact, our findings revealed the strongest relationship between caregiver autonomic responsiveness and speed of infants' recovery at 10 months. The more caregivers increased their arousal relative to the event, the faster older infants recovered (Figure 3.7E).

Overall, then, our findings are more consistent with Hypothesis 2, that developmental changes would be attributable to enhanced caregiver-infant co-regulation rather than to improvements in infant self-regulation alone. This is consistent with other literature which provides relatively little evidence that self-regulation improves during the first few years of life. Questionnaire ratings suggest that parent-rated fear and negative emotionality in infants actually increases from 6 months to 12 months (Gartstein et al., 2018; Gartstein & Hancock, 2019), and experimental evidence suggests that infant negative reactivity to a toy removal task also increases from 6 months to 12 months to 12 months (Suata, 2023). As infants get older, they react more intensely, both positively and negatively, attributed to neurological, cognitive, and experimental growth during the first year of life (Braungart-Rieker et al., 2010; Dollar & Calkins, 2019; Thompson, 1991).

This may be context-specific, consistent with functionalist perspectives on emotion development which suggest that infants learn over time that negative affect displays are useful in certain situations (e.g. in ensuring that a toy that has been removed from them is returned) (Barrett & Campos, 1987; Miller & Sperry, 1987).

One possibility that might underpin our observation that real-world negative affect displays get shorter between 5 and 10 months is that, with time, the caregivers' perceptivity (i.e. the ability to understand their infants' states) improves (Thorson et al., 2018). Consequently, their responses may also become more effective. Similarly, it is also reasonable to think that the expressivity of older infants (i.e. the quality and quantity of the cues that the infants give off that reflect their psychological states) may be enhanced (Aktar & Pérez-Edgar, 2020; Thorson et al., 2018), making their cues also easier to interpret. Together, the combination of enhanced perceptivity and expressivity could give rise to a greater understanding between the dyad members over time that does not necessarily translate into more caregiver involvement nor attunement (more is not always better), but faster (or more efficient and selective) physiological co-regulation. Over time, caregivers changed their responses and learnt that by responding less, they were able to help their infants recover faster. Alternatively, it is also worth considering the possibility that older infants might have become more able to use their mothers' support more effectively.

In sum, the current study showed that during infancy, dyadic strategies, rather than being phased out, continue to play an important role in infants' regulation, even as caregivers' responses to infants' signals decrease over time. We discussed the possibility that caregiver responses become more effective, or infants become more able to use their support more effectively. It is likely that a combination of both factors leads to more efficient co-regulatory processes within the dyad over developmental time.

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3.5.1 Limitations

Our findings should be interpreted with consideration to a number of limitations of the study. First, our sample consisted of mothers and thus may not necessarily be representative of caregiver-child relations more generally.

Second, the threshold we employed to identify speech-like vocalisations from the infants was very stringent, meaning that while we might have identified key-child vocalisations to quite a high precision, it is also true that we might have missed many vocalisations that, due to low confidence estimates from the classifier, the classifier opted to disregard. This might have biased our results by giving us certain types of vocalisations but not others (e.g. vocalisations that happened in noisier environments, or vocalisations associated with more significant emotion). Similarly, to improve the performance of the SER we had to collapse positive with neutral affect vocalisations into one group. It is possible that the neutral affect could have attenuated the effects of the positive affect, and had we been able to exclusively compare negative versus positive affect, the outcomes might have differed. Relatedly, we were unable to assign intensity to these vocalisations nor code for the semantics of the caregiver responses, which would have broadened our understanding of the co-regulatory process occurring within the dyad.

Third, it is important to recognise the fact that cries, but also other types of vocalisations tend to cluster together in time, forming, for instance, crying episodes. The current study, however, focused on single vocalisations rather than whole episodes of, for example, crying. As a result, infants might have vocalised several times during the time frames we examined around each vocalization; when this happened, another event was inserted at the time of the subsequent vocalisation and the analyses were repeated. This is a fairly standard procedure when examining time series changes relative to irregularly spaced events - see e.g. these publications (Phillips et al., 2023; Wass et al., 2022, 2019) which

used the same approach. However, it is important to acknowledge the possibility that this might have extended the duration of heightened arousal.

Related to this, the findings from the windowed cross-correlation (Figure 3.9) exhibit a somewhat "burst-like" pattern at 5 months. One potential explanation for this observation could be a higher frequency of negative vocalisations (e.g. cries) within the 20-minute window analysed compared to the data from 10-months-old infants. However, we do not have evidence that 5-months-old infants produced more negative vocalisations than 10-months-old infants and we cannot identify a way in which this method (i.e. single vocalisations vs. whole episodes) might have artificially augmented the observed finding, but rather only attenuated it.

Fourth, we took a relatively reductionist approach by exclusively focusing our analysis on arousal changes. Such an approach is limited in that it overlooks other important factors such as cognitive and emotional development from the infant, as well as infant temperament, along with the social, environmental, and cultural contexts in which these processes occur.

Fifth, all our analyses were run at group level so individual differences within the group as well as within participants remain unexplored and results should only be interpreted as the tendency of a sample as a whole (Palumbo et al., 2017). Relatedly, it is worth noting that there was a substantial discrepancy in sample sizes, with the group of 10-month-olds being considerably smaller. As a result, our statistical power to identify significant effects at that age might have been limited.

Lastly, due to the naturalistic nature of our events, we combined various stressors from different external sources (e.g., a dog barking at the infant, a glass breaking and creating a loud noise) as well as internal sources (e.g., infant hunger, sleepiness, etc.) into the same analyses.

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3.5.2 Overall implications

Understanding how early parent–infant interpersonal regulatory processes occur and develop across the first year of life is key as they influence all the emotional and cognitive development that follows. Historically, our understanding of caregiver-infant dynamics has been held back by different methodological challenges (see 3.1). To address these limitations, we developed small wearable devices that allowed us to record a variety of parameters such as heart rate, proximity, activity and vocal data from infants and their mothers in their home environment.

Our results showed increased caregiver responsiveness at both the physiological (e.g. they were likely the more active partners in synchronising and matching their infants' affect) and physical (i.e. they decreased the distance between them and their infants more) levels at 5 months. However, the strongest relationship between caregiver' autonomic responsiveness and speed of infants' recovery was at 10 months. We argue that dyadic strategies, rather than being phased out, continue to play an important role in infants' regulation. We discuss the possibility that caregiver responses become more effective, or infants become more able to use their support more effectively. It is likely that a combination of both factors what leads to more efficient co-regulatory processes within the dyad over developmental time.

Considering the impact of ineffective early regulation on later-life socioemotional, behavioural, and psychological well-being, an understanding of normative and disrupted developmental patterns of regulation is central for promptly identifying and intervening with high-risk parent-infant pairs. Furthermore, this research introduces a novel methodological perspective that could interest researchers in the field.

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3.6 Acknowledgments

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Chapter 4. The neural and physiological substrates of real-world attention change across development

The following chapter is a publication of an original article investigating the mechanisms through which regulation of attention develops during early life in complex, real-world settings (Perapoch Amadó et al., in press). Subheadings, figure placement, figure and table numbers, and citation style have been adapted to conform to the general thesis format. The supplementary materials for this chapter are available in Appendix B. Of note, physiological arousal is here referred as autonomic arousal.

4.1 Abstract

The ability to allocate and maintain visual attention enables us to adaptively regulate perception and action, guiding strategic behaviour within complex, dynamic environments. This capacity to regulate attention develops rapidly over the early years of life and underpins all subsequent cognitive development and learning. From screen-based experiments we know something about how attention control is instantiated in the developing brain, but we currently understand little about the development of the capacity for attention control within complex, dynamic, real-world settings. To address this, we recorded brain activity, autonomic arousal, and spontaneous attention patterns in N=58 5- and 10-month-old infants during free play. We used time series analyses to examine whether changes in autonomic arousal and brain activity anticipate attention changes or follow on from them. Early in infancy, slow-varying fluctuations in autonomic arousal forward-predicted attentional behaviours, but cortical activity did not. By later infancy, fluctuations in fronto-central theta power associated with changes in infants' attentiveness and predicted the length of infants' attention durations. But crucially, changes in cortical power followed, rather than preceded, infants' attention shifts, suggesting that processes

after (but not before) an attention shift determine how long that episode will last. We also found that changes in fronto-central theta power modulated changes in arousal at 10 but not 5 months. Collectively, our results suggest that the regulation of real-world attention involves both arousal-based and cortical processes but point to an important developmental transition. As development progresses, attention control systems become dynamically integrated and cortical processes gain greater control over modulating both arousal and attention in naturalistic real-world settings.

4.2 Introduction

The ability to allocate and maintain visual attention enables the flexible regulation of perception and action that is characteristic of strategic behaviour (Hendry et al., 2019; Rueda et al., 2021). The capacity to pay attention develops rapidly over the early years of life (Richards, 2010), and individual differences in early attention predict long-term cognitive and clinical outcomes (Colombo & Mitchell, 2009; Wass, 2015). Recent new methodological advances such as naturalistic neuroimaging are allowing us to build on previous research using lab-based behavioural experiments and animal studies.

The development of attention is traditionally conceptualised as the product of "interactions among different systems at different levels of maturity" (Colombo, 2001; Hendry et al., 2019; Johnson, 2002). Traditionally, the earliest subcomponent of attention to develop is thought to be the arousal/ alertness subcomponent, mediated via brainstem reticular activating systems centred on the locus coeruleus (LC) and instantiated primarily via norepinephrine neurotransmitter systems (Colombo, 2001). In young infants, alertness is more readily initiated by exogenous events (Wolff, 1965); over time, infants gain the ability to both attain and maintain an alert state even in absence of external stimulation. Areas around the brainstem (including the LC) are thought to be some of the earliest to become functionally mature (Deoni et al., 2011; Johnson & Haan, 2015). Consequently,

the relative influence of this subcomponent of attention is thought to be strongest during early development (Colombo, 2001).

Behaviourally, the arousal/ alertness subcomponent of attention is thought to reflect a state of anticipatory readiness, or alertness for stimulus input (Colombo, 2001). Arousal is generally measured indirectly, via proxy measures of autonomic nervous system activity such as heart rate (Richards, 1980). Heart rate has been extensively studied in the context of infant attention (Richards, 2010, 2011; Wass, 2021). During anticipatory readiness, we know that reorientations of visual attention take place periodically, clustered around a preferred modal reorientation rate (Feldman & Mayes, 1999; Nuthmann et al., 2010; Robertson, 1993; Saez de Urabain et al., 2017; Wass & Smith, 2014). This may reflect rhythmic activity in the central nervous system (Mcauley et al., 1999).

With time, it is thought that looking behaviours become increasingly modulated by higher-level executive processes that reflect the infant's internal states, motivation, comprehension, and goals (Colombo & Cheatham, 2006; Oakes et al., 2002; Rueda et al., 2021). Behaviourally, this increase in endogenous or internally directed attention (i.e. self-regulation of attention) has been shown as: a developmental increase in the degree to which attentional engagement is accompanied by decreases in distractibility (Anderson et al., 1987; Lansink & Richards, 1997); an increase in selective attention as measured indirectly, using the blink reflex (Richards, 2000); and differences in the trajectory of how attention durations to simple vs complex stimuli change over developmental time (Courage et al., 2006).

Other research that used experimenter-controlled, screen-based tasks to examine neural correlates of attention has examined changes in the power spectral density (PSD) of electroencephalography (EEG) oscillations, in particular infants' theta (3-6Hz) rhythm, which increases during active, anticipatory, and exploratory behaviour (Begus &

Bonawitz, 2020; Braithwaite et al., 2020; Jones et al., 2020; Meyer et al., 2019; Orekhova et al., 2006; Xie et al., 2018). Together, these studies suggest that the expression of theta during attention-eliciting episodes could signify the engagement of neural networks related to executive attention (Braithwaite et al., 2020; Orekhova et al., 2006; Xie et al., 2018). Similarly, other studies have reported decreases in alpha band activity under conditions of increased attention (Libertus et al., 2009; Xie et al., 2019). Both theta and alpha effects are now widely known in the literature as "theta synchronization" and "alpha desynchronization" (Xie et al., 2018) and have commonly been associated with infant attention.

How children allocate their attention in experimenter-controlled, screen-based lab tasks differs, however, from actual real-world attention in several ways (Wass & Jones, 2023; Wass, 2014; Wass & Goupil, 2022). For example, the real-world is interactive and manipulable, and so how we interact with the world determines what information we, in turn, receive from it: experiences generate behaviours (Anderson et al., 2022). While lab-based studies can be made interactive (e.g., Meyer et al., 2023; Stahl & Feigenson, 2015), the way infants actively and freely initiate and self-structure their attention remains unexplored.

The present study aims to examine developmental changes in the relationship between autonomic arousal, cortical activity, and attention in real-world settings. To do this, we first explored how naturalistic attention patterns from a solo play interaction change between 5 and 10 months. Then, we explored temporal relations between changes in infant's arousal and attention episodes. Finally, we investigated changes in EEG theta power relative to attention episodes, and changes in EEG theta relative to arousal (see Figure 4.1). As attentional systems mature and brain regions become increasingly specialised (Grossmann et al., 2008; Jones et al., 2015), we expected to see both a developmental increase in visual attentiveness towards play objects and a developmental shift in the way different mechanisms (i.e. arousal/ alertness vs. executive attention subsystems) regulate attention.

Our first set of analyses examined attentional inertia (the phenomenon that, as individuals become progressively more engaged with an object, their attention progressively increases) as a measure of internally driven attentional engagement (Richards, 2010; Richards & Anderson, 2004; Wass, Clackson, et al., 2018). We tested whether attentional inertia influenced attentional behaviours more strongly at 10 months compared to 5 months. To do so, we calculated both the Autocorrelation Function (ACF) and the survival probability of spontaneously occurring attention episodes during play (analysis 1). The ACF allowed us to quantify the rate of change of spontaneous attention durations. A faster rate of change would indicate lower attentional inertia. The survival probability, on the other hand, allowed us to quantify the probability between looking (i.e. paying attention) and looking away. A slower decrease in the probability of an attention episode surviving would indicate increased attention engagement and decreased distractibility by other stimuli. We hypothesised that, as slow-varying fluctuations in endogenous interest or engagement start to influence looking behaviour more strongly over time, 10-monthold infants would show increased endogenous attention control indexed by a slower rate of change of attentiveness and slower decreases in the survival probability. We also predicted that we would be able to identify periodic attentional reorientations during early as well as later development (Robertson, 1993, 2013); later in development, however, we predicted that infants would be more likely to extend visual fixations beyond their modal periodic reorientation rate, possibly indicating a greater or more efficient integration of attention and gaze shifting (Robertson et al., 2012), and that attention duration episodes would be longer overall.

Next, to assess the link between lower-level mechanisms of autonomic arousal and attention, we calculated cross-correlations between autonomic arousal (indexed via heart

rate) and attention episodes across the entire play session for both 5- and 10-months olds. This allowed us to examine whether arousal changes tend to forward-predict changes in attention, or vice versa (analysis 2). Based on previous research (de Barbaro et al., 2017), we hypothesized that periods of elevated autonomic arousal would associate with, and forward-predict, shorter attention episodes. We also predicted that such relationship would weaken with time due to the maturation of cortical attentional systems.

We used a similar approach to examine developmental changes in the relationship between neural markers of executive attention and real-world attention behaviours. We were interested to examine whether neural changes (indexed by theta power) anticipate subsequent attentional behaviour shifts (Robertson et al., 2012); or, whether neural processes after the attention shift relate to increases in infants' attention engagement. To test this, we conducted three analyses. First, we analysed neural activity across a range of time windows both before and after the onsets of new attention episodes and performed linear mixed effect models to examine how neural activity before and after attention onset associated with the subsequent durations of those episodes (analysis 3). Second, we examined changes in neural activity during individual attention episodes (analysis 4). Finally, we used cross-correlations to examine whether, across the entire dataset, neural markers tend to forward-predict changes in attention, or vice versa (analysis 5). We predicted that the associations between neural markers of executive attention and realworld attention behaviours would become stronger with increasing age (i.e., theta activity would show a stronger predictive relation with infants' attentional behaviours at 10 months, as evidence of increased modulatory power from the executive attention system on infants' attention).

Finally, we examined whether there were any interdependencies between autonomic arousal and theta activity. We had no predictions for how this relationship would change over time.

4.3 Materials and Methods

4.3.1 Experimental Design

Looking behaviour, EEG and electrocardiography (ECG) data were collected from mothers and their infants at two age points: 5 and 10 months while playing alone. At 5 months, infants were seated either in a highchair or on a researcher's lap and a table was positioned in front so that toys on the table were within easy reach (see Figure 4.1). To reduce infant's stress, mothers were present in the room but moved to another smaller table on the right side of the original table and given an identical set of toys which they played with in parallel. A wooden divider was positioned between the two tables to prevent infants from seeing the objects with which their mothers were playing. At 10 months, the same procedure was used but the divider was positioned across the midline of the table and the adult participants were seated directly opposite the infants. In both situations, mothers and infants had direct line of sight to one another but neither could see the others' toys on the table.

The same three age-appropriate toys were always used for each age group. These were small and relatively engaging (see APPENDIX B, Figure S5). During the solo play interaction, one of the researchers sat behind the infant to collect the toys that fell on the floor (either because the infants threw them or because they fell from their hands) and brought them back on the table. Mothers were allowed to speak during the interaction but were instructed not to name the toys they were playing with to prevent infants from the influence of any exogenous parental' influence. In the Supplementary Materials, we present a set of analyses that preclude the possibility of maternal influence on infants' behaviour and demonstrate that the impact of the mothers on the infants' behaviour did not differ between age groups (APPENDIX B, SM3). The average duration of the interactions with usable EEG/ ECG data did not differ significantly between 5 and 10 months (interactions with EEG (average duration at 5m = 292.4s, and 10m = 250.1s,

t(46) = -1.85, p = 0.07); interactions with ECG data (average duration at 5m = 351.2s, and 10m = 317.9s, t(40) = -1.1, p = 0.27)).

The interactions were filmed using three Canon LEGRIA HF R806 camcorders recording at 50 frames per second (fps). At 5 months, one camera was placed in front of the infant and another one was placed in front of the mother. At 10 months, two cameras faced the infant: one placed on the left of the divider, and one on the right. The other camera faced the mother and was positioned just behind the right side of the divider. All cameras were placed so that the infant's and the mother's gaze, as well as the three toys placed on the table, were always visible.

Brain activity was recorded using a 64-channel at 5 months and a 32-channel at 10 months, BioSemi gel-based ActiveTwo system with a sampling rate of 512Hz with no online filtering using Actiview Software.

Heart rate activity was recorded using a BioPac[™] (Santa Barbara, CA) system recording at 2000Hz. ECG was recorded using disposable Ag-Cl electrodes placed in a lead II position.





Top figure shows the experimental set up for solo play. Below, on the left, is shown the procedure followed to parse the looking behaviour and create the variable "attention", and further cross-correlation analyses (analysis 2 and 5). On the right, instead, is shown the steps followed to identify attentional episodes in the EEG signal and further EEG analyses (analysis 3 and 4).

4.3.2 Participants

Participants were typically developing infants and their mothers. The catchment area for this study was East London, including boroughs such as Tower Hamlets, Hackney and Newham. Participants were recruited postnatally through advertisements at local baby groups and local preschools/ nurseries. We also operated a word-of-mouth approach, asking parents who got involved to ask if their local networks would be interested in participating. Ethical approval was obtained from the University of East London ethics committee (application ID: ETH2021-0076, see APPENDIX D).

Initial exclusion criteria included complex medical conditions (e.g., heart rate condition, neurological/ genetic abnormality), known developmental delays, prematurity, uncorrected vision difficulties, and parents below 18 years of age. Further exclusion criteria as well as final numbers of data included in each of the analyses for both samples are summarised in APPENDIX B, Table S7. The final sample included 12 infant females and 19 infant males at 5 months and 14 infant females and 15 infant males at 10 months. Average age was 5.32 months (std = 0.58) and 10.49 months (std = 0.87). This is the first time that any of this data has been analysed and reported.

4.3.3 Data processing and Statistical Analysis

4.3.3.1 Synchronisation between behavioural and EEG/ECG data

The cameras were synchronised to the EEG and ECG via radio frequency (RF) receiver LED boxes attached to each camera. The RF boxes received trigger signals from a single source (computer running Matlab) at the beginning and end of the play session, and concurrently emitted light impulses, visible from each camera. At the same time, triggers were sent and stored in the Actiview Software and recorded to the EEG data as well as to the Acknowledge Software and recorded to the ECG data. The video coding and EEG/ ECG data synchronisation was done by aligning the times of the LED lights and the EEG/ ECG triggers. We also checked for dropped/missing frames by checking that the time between the LED lights matched the times between the EEG/ ECG triggers.

4.3.3.2 Video coding

The looking behaviour of the infants was manually coded offline on a frame-by-frame basis, at 50fps. The start of a look was considered to be the first frame in which the gaze was static after moving to a new location. The following categories of gaze were coded: looks to objects (where the infant was focussing on one of the three objects), looks to partner (where the infant was looking at their partner), inattentive (where the infant was not looking to any of the objects nor the partner) and uncodable. Uncodable moments included periods where: 1) the infant's gaze was blocked or obscured by an object and/or their own hands, 2) their eyes were outside the camera frame, and/ or 3) a researcher was within the camera frame and the infant turned to them and/or realised a researcher was around. To assess inter-rater reliability, ~15% of our data (10 datasets) were double-coded by a second coder and Cohen's kappa was calculated. There was moderate agreement ($\kappa = 0.581$, std= 0.183) (Landis & Koch, 1977). Due to the unusual nature of our behavioural coding (with gaze coded across many 20ms bins) the interrater reliability is heavily contingent on how we calculate it. We chose to report the most stringent calculator of inter-rater reliability.

Looking behaviour data was then processed such that any look preceding and following an "uncodable" period was NaN-ed and excluded from further analyses. Similarly, both the first and the last look of every interaction were also NaN-ed and excluded from further analyses.

4.3.3.3 EEG artefact rejection and pre-processing

EEG data was pre-processed and cleaned from oculomotor and other contaminatory artefacts using a fully automatic artefact rejection procedure specially designed for naturalistic infant EEG data by Mariott Haresign (Haresign et al., 2021), building on previous related work (Haresign et al., 2021, Kayhan et al., 2022). Briefly, this involved the following steps: 1) data were high-pass filtered at 1Hz, 2) line noise at 50Hz was eliminated using the EEGLAB function clean line.m, 3) data were low-pass filtered at 20Hz, 4) the data were referenced to a robust average reference 5) noisy channels were rejected using the EEGLAB function pop rejchan.m, 6) the channels identified in the previous stage were then interpolated back, using the EEGLAB function eeg interp.m, 7) continuous data were automatically rejected (NaN-ed) in a sliding 1s epoch based on a percentage of bad channels (set here at 70% of channels) that exceed 5 standard deviations of the mean channel EEG power, and 8) ICAs were computed on the continuous data using the EEGLAB function runica.m. Only participants with fewer than 30% of channels interpolated at 5 months and 25% at 10 months (in step 6) made it to the final step (step 8, ICA) and final analyses. To compare the quality of the EEG data at 5 and 10 months we performed a series of analyses on percentage of channels interpolated, total segments removed (i.e., zeroed out) and total ICA components rejected (see APPENDIX B, Table S5-6 and Figure S4).

The higher density array was down sampled so that all the EEG analyses described below used only shared channels between the 32- and the 64- channel EEG systems. We selected three main clusters of electrodes for our analyses: Frontal channels ('Fp1', 'Fp2', 'AF3', 'AF4', 'Fz'), Central channels ('FC1', 'FC2', 'C3', 'Cz', 'C4', 'CP1', 'CP2'), and Occipital channels ('PO3', PO4', O1', 'Oz', 'O1') (see APPENDIX B, Figure S6).

4.3.3.4 Heart Rate – Beats per minute

R-peak identification was performed using the in-built Matlab function 'findpeaks'. The minimum peak height was manually defined as a simple amplitude threshold after visualising the raw data, minimum peak distance, instead, was set at 230msec. Following this, automatic artefact rejection was performed by excluding those beats showing an inter-beat interval (IBI) <300 or >750msec, and by excluding those samples showing a rate of change of IBI greater than 90msec between samples. Next, we converted IBI values into beats-per-minute (BPM) values and removed outliers in the BPM time series. These were defined as values falling 2.5 interquartile ranges above the upper quartile and below the lower quartile. Outliers were then interpolated using the Matlab in-built function 'fillmissing' with the 'spline' method. Finally, we epoched the data into one-second epochs by averaging all the BPM values comprised in each one-second epoch.

4.3.3.5 Analysis 1. Developmental changes in attention

4.3.3.5.1 Overt attention and inattention extraction

The aim of our analysis was to identify moments where the infants paid attention to any of the play objects as opposed to inattentive moments. Accordingly, all looks to object and inattentive looks were selected and categorised as attentional and inattentive episodes, respectively. Looks to partner were excluded from all analyses.

Following this, we extracted the first and last frame of all looks of interest (i.e. looks to objects and inattentive looks). To calculate attention and inattention durations, we subtracted the last frame from the first frame of each look of interest and divided it by the sampling rate (i.e. 50) to convert "duration in frames" to "duration in seconds". Attentive and inattentive reorientations were calculated by counting the occurrence of each of these

two attentional states, irrespective of their durations, and dividing it by the total duration of the interaction.

4.3.3.5.2 ACF of the attention duration

Here, we extracted the duration (in seconds) of all the attentional episodes that happened within the play session and zero-ed out all the non-attentional episodes. This allowed us to create a time series string with the durations of each consecutive attention episode. We then calculate the autocorrelation of that signal and repeated these steps for each behavioural dataset. Finally, we averaged the ACF values within each age group to obtain the ACF values reported in Figure 4.3A.

4.3.3.6 Analysis 2. Auto- and cross-correlation analyses between infant autonomic arousal and attention

4.3.3.6.1 Attention, one-second epochs.

To calculate the "attention" variable, we epoched the gaze data into one-second epochs and calculated the duration (in msec) of each attention episode relative to the one-second epochs. Most epochs were coded as either 1000 (epochs where the child was attending throughout) or 0 (epochs where the child was inattentive throughout). If an attention episode started halfway through a one-second epoch, then it was coded as 500. The other non-attention episodes (i.e., inattentive looks, looks to partner) were zeroed. See Figure 4.1 for a schematic view of the procedure we followed to parse the looking behaviour into one-second epochs.

4.3.3.6.2 Cross-correlation analyses

To investigate the relationship between autonomic arousal and fluctuations in attention we performed a cross-correlation analysis between the two variables. Importantly, these analyses are not time locked to specific moments (i.e., start of an attentional event) and are conducted on two time series (i.e., attention and heart rate fluctuations) as a whole. Because of this, the strength of the overall correlation is weakened by the fact that periods of expected stronger correlation are balanced by weaker correlations where we would not necessarily expect any correlation at all.

Additionally, we also computed the autocorrelation for autonomic arousal to assess how well it predicts itself over time and evaluate its stability. All analyses were computed at lags from -30 to +30s in 1s intervals. The cross-correlations values at each time-lag were computed individually and then averaged across all participants. The procedure was identical for the autocorrelation, except that instead of examining the relationship of two different time series at variable time intervals, we assessed the relationship of one time series to itself at variable time intervals.

To assess significance of the cross-correlations, we first used bootstrapping to generate confidence intervals, using an approach that controls for the level of autocorrelation in the data. To do this, the time series data of one participant (e.g. attention of participant 1) was randomly paired with the time series data of another participant (e.g. autonomic arousal of participant 13). If the time series datasets had different lengths (due to different participants having different session lengths), we appended zeros to the end of the shorter vector to match the length of the longest vector. We then computed the cross-correlation between all the unique combinations that could be found within each sample (e.g. in a sample N=23, the maximum of unique combinations is 529). Next, the cross-correlation results in the permuted data were randomly grouped in samples that were the same size as the original data (e.g., N= 23) and averaged together. This procedure was repeated

1000 times and used to generate the 95% confidence intervals. In this way, we identified whether the observed cross-correlation values at each time interval differed significantly from chance.

Next, to control for multiple comparisons across time intervals, we used a cluster-based permutation approach. On each iteration, one permutation was compared with the 999 other permutations, and significant time-points were identified as values falling above the 97.5th centile and below the 2.5th centile (corresponding to a significance level of 0.05). We then identified the two largest clusters of significance that occurred by chance: one for positive correlation values and the other for negative correlation values. We repeated this 1000 times. Following this, we created a distribution of cluster sizes for positive and negative correlation values and took the size value corresponding to the 95th percentile in each distribution to define our cluster-size threshold. Finally, we compared the cluster sizes obtained in the observed data against the cluster-size threshold and only considered significant the ones that exceeded such threshold.

Calculating the significance levels of the autocorrelation was more straightforward. This was done by first calculating the autocorrelations based on individual datasets, and then averaging the significance values of the Spearman's correlations at each time interval.

4.3.3.7 Analysis 3. Calculation of neural power changes around an attention episode

We examined the associations between the duration of infant attention episodes and infant theta changes around these looks using linear-mixed effects models. Infant attention episodes and attention duration were calculated as explained above in section 4.3.3.5.1.

To conduct these analyses, each infant attention episode onset (i.e. gaze shift to a different toy) was identified in the EEG signal by calculating the time from the start of the interaction (first LED) to the onset of the look (in the behavioural data) and adding it to the first EEG trigger. For each look, we extracted theta (3-6Hz) power for two timewindows immediately prior to the onset of each look (-2000 to -1000msec and -1000 to 0 msec pre-look onset) and three time-windows immediately after the onset of each look (0 to 1000ms, 1000ms to 2000msecand 2000 to 3000msec post look onset) (see Figure 4.1). To calculate the EEG power spectra, we use the 'mtmfft' method from the ft_freqanalysis function in FieldTrip, an open-source Matlab toolbox (Maris & Oostenveld, 2007); http://fieldtriptoolbox.org). Extreme power values that were 4 times greater than the interquartile range were treated as outliers and excluded from further analyses (similar to Xie et al., 2018). More detail on the amount of data available (i.e. average duration of the session per participant and number and duration of attentional episodes per minute) can be accessed in APPENDIX B, Figure S1.

For each epoch, we only selected power within our cluster of central channels (similar to Wass, S., Noreika, V. et al., 2018). Power at each bin was expressed as relative power, defined as the total power at a specific frequency band (e.g. 3 to 6Hz for theta) divided by the total power across all frequency bands (1 to 20Hz) during that epoch. After extracting the relative power in the theta band, we calculated separate linear mixed effects models for each of the five windows to examine the relationship between EEG power within that time window and attention duration (with participant ID as random effect).

4.3.3.8 Analysis 4. Calculation of neural power changes within an attention episode

In addition, we also wanted to look at power changes within attention episodes. Infant attention episodes and attention duration were calculated as explained above (section 4.3.3.5.1) and each infant look onset towards an object was identified in the EEG signal as described in analysis 3 (section 4.3.3.7). For each look, we extracted the first (0 to ± 1000 msec, "start") and third-to-fourth (3000 to 4000msec, "middle") second into the look, and the last second (-1000msec prior to look offset to look offset, "termination")

before look termination (see Figure 4.1). Looks that did not make it to the full second segment were excluded from further analysis. Similarly, only looks that were longer than 5 seconds were included to the "middle" group. This was done to avoid an overlap between the activity from the "middle" and the "termination" groups.

Frequency analysis was conducted to assess the power spectral density for both theta (3-6Hz) and alpha (6-9Hz) frequency rhythms for each of the three time-segments. These analyses were calculated for the three prespecified clusters of channels: Frontal, Central and Occipital (see APPENDIX B, Figure S6). Again, power at each time segment was expressed as relative power.

The selection of both theta (3-6Hz) and alpha (6-9Hz) frequency bands was led by previous work using this same approach (e.g., Orekhova et al., 2006; Xie et al., 2018, Meyer et al., 2019, Jones et al., 2015).

4.3.3.9 Analysis 5. Auto- and cross-correlations analyses between infant theta activity and attention

4.3.3.9.1 EEG relative power, one-second epochs

For this analysis, we parsed the EEG data into one-second segments and calculated the relative theta power for each one-second segment as described above (see 4.3.3.7).

4.3.3.9.2 Cross-correlation analysis

To explore whether modulations in endogenous theta activity related to fluctuations in infants' attention, we conducted a cross-correlation analysis between infants' relative theta and attention. Attention was calculated as described in analysis 2 (4.3.3.6.1, and Figure 4.1). Additionally, we also computed the autocorrelation for relative theta to assess

how theta predicts itself over time. Again, all analyses were computed at lags from -30 to +30s in 1s intervals. Significance was assessed following the steps described in analysis 2 (4.3.3.6.2).

Finally, to explore interdependencies between autonomic arousal and theta activity we conducted a cross-correlation analysis between infants' autonomic activity and relative theta.

4.4 Results

4.4.1 Analysis 1: Developmental changes in attention

Our first set of analyses examined attentional inertia as a measure of internally driven attentional engagement. Attention inertia is the phenomenon that, as individuals become progressively more engaged with an object, their attention progressively increases. We tested whether attention inertia is stronger at 10 months compared to 5 months. To do so, we calculated the ACF and the survival probability of spontaneously occurring attention episodes during play to quantify the rate of change of spontaneous attention durations and the probability between looking (i.e. paying attention) and looking away, respectively.

Initially, we conducted four descriptive analyses to test how attention and inattention durations and reorientations change over both the course of the solo play interactions and developmental time. First, we tested how many times per minute 5- and 10-months-old infants redirected their attention from one object to the other. We found that, on average, 5-month-old infants performed significantly more both attentive (t(10)=4.346, p=0.001) and inattentive (t(10)=4.202, p=0.002) reorientations during the solo play interaction than 10-months-old infants (Figure 4.2A). When we looked at how attention reorientations changed during the course of the solo play episode, we found that 5-month-old infants performed infants performed solo play episode, we found the time the interaction even

though the number of looks per minute decreased over the course of the interaction for both age groups (Figure 4.2B, and APPENDIX B, Figure S1A and B).

Second, we investigated the average duration that 5- and 10-month-old infants spent in attentive and in inattentive states during the solo play interaction and minute by minute (Figure 4.2C and D respectively). In general, infants' attention durations toward play objects at 10 months were longer (t(10)= -2.787, p=0.019). At 5 months, moments of inattention were longer than moments spent looking towards the object (t(10)= -3.749, p=0.003). Overall, at 10 months, but not 5 months, infants spent more time in attentive compared to inattentive states (t(58)= 10, p<0.001) (Figure 4.2E). We then calculated a best fit line, individual by individual, to look at how average attention duration changed within the session. We found no significant differences in the way attention duration changed during the interaction between the two age groups (APPENDIX B, Figure S1C and D).

Third, we explored the distribution of looks towards the objects (Figure 4.2F). At both ages, attention durations shorter than or equal to 5 seconds follow a positively skewed lognormal distribution, with modal attention durations in the 0.5 - 0.6 second range. Modal attention durations were significantly lower at 5 than at 10 months (t(58)= 2.211, p=0.03). Finally, the right plot in Figure 4.2F shows extended attention episodes. There was an increasing amount of such looks with increasing age.





(A) Average number of attentive and inattentive looks per minute at 5 months (left) and 10 months (right), (B) Average number of attentive and inattentive looks minute by minute. (C) Average duration spent in one of the two possible attentional states: attentive and inattentive, and (D) minute by minute. (E) Percentage of time infants spent in attentive vs. inattentive states, during the whole interaction. (F) Histogram showing the distribution of the proportion of all the looks that lasted less than or equal to 5 seconds (right) and more than 5 seconds (left) at 5 months (yellow) and 10 months (blue). Continuous black line indicates the mode of each distribution. Significance is indicated with asterisks where * = p < 0.05, ** = p < 0.01, and *** = p < 0.001. Error bars represent SEMs.

Following the descriptive statistics, we calculated both the ACF and the survival probability of the looking behaviour (Figure 4.3). First, we used time-series analyses to

examine the rate of change of attention durations, relative to itself. We calculated the ACF of the attention durations at both time points (more details in 4.3.3.5.2). The ACF indexes the cross-correlation of a measure with itself at different lag-intervals in time (Wass, Clackson, et al., 2018). ACF values were obtained from 0 to 10 seconds lag, in steps of 500 milliseconds. As shown in Figure 4.3A, the ACF of the time series looking behaviour fell off more sharply at 5 months than at 10 months. The ACF values were compared across ages using independent sample t-tests. From lag +500 milliseconds to 10 seconds to 10 seconds, 10-months-old infants showed significantly higher correlation values than 5-months-old infants.

Second, we performed a survival analysis by calculating the survival probability function of the looking behaviour towards the objects at both time points. The survival probability function is the probability that an attention episode survives longer than a certain time. As shown in Figure 4.3B, the survival probability of a look decreased abruptly at the beginning, for the very short looks, and flattened as looks got longer. The differences in the speed at which the survival probability decreased can be seen more clearly by calculating the derivative of the survival probability (Figure 4.3C). To compare survival among the two groups, we performed the log rank test using the Matlab function 'Logrank' (Wass et al., 2022). The results for the log-rank test rejected the null hypothesis (p<0.001) indicating that the survival curves for looking behaviour at 5 months and 10 months were significantly different. Notably, the likelihood of a look ending is more tightly clustered around the modal value of 0.5 seconds at 5 months.

Overall, our results showed that older infants demonstrated to have a slower-changing profile of attention with longer attention episodes overall (Figure 4.2A and B, Figure 4.3A). At both ages, there was evidence for a preferred modal reorientation rate in the 0.5-0.6 second range, which was slightly faster at 5 months than 10 months (Figure 4.2F). Attention durations were more tightly clustered around the modal value at 5 months. At

10 months, attention episodes were more likely to be extended beyond the preferred modal reorientation rate than at 5 months (Figure 4.2B and C).



Figure 4.3. ACF and survival probability analyses of the looking behaviour. (A) Autocorrelation of the time series looking behaviour at 5 months (in yellow) and 10 months (in blue). (B) Survival analysis. Survival probability function for looking behaviour toward object toys. The survival function is the probability that a look will survive a given time. Yellow line shows data from 5-months-old infants with confidence bounds (dotted yellow line) and blue line shows data form 10-months-old infants with confidence bounds (dotted blue line). (C) Derivative of the Survival Probability at 5 months (yellow) and 10 months (blue).

4.4.2 Analysis 2. Auto- and cross-correlation analyses between infant autonomic arousal and attention

In this section we investigated the relationship between changes in infant autonomic arousal indexed by heart rate activity and their associations with moment-to-moment changes in attention.

Figure 4.4 (A and C) shows the results of the autocorrelation analyses for autonomic arousal at 5 and 10 months of age respectively. Significant autocorrelations were observed at relatively short lags around t=0 (from -4 to +4s) at both ages. Figure 4.4 (B and D) shows the results of the cross-correlation analysis between autonomic arousal and attention at 5 and 10 months of age respectively. The negative values indicate that, at 5

months, lower heart rate forward-predicted increased likelihood of attention from lags between -9 to -2 seconds (i.e. lower heart rate at time *t* significantly associated with increased attention at time t+9 seconds). The same pattern was present but not significant at 10 months. The asymmetry of this cluster around the lag t=0 indicates that changes in heart rate tended to forward-predict changes in attention more than *vice versa*.



Figure 4.4. Relationship between infant autonomic arousal and attention.

Autocorrelation results for infant autonomic arousal at 5 months (A) and 10 months (C). Significant clusters are indicated by red dots. Cross correlation between infant autonomic arousal and attention at 5 months (B) and at 10 months (D). Infant autonomic arousal forward-predicting infant attention on the negative lags, infant attention forward-predicting infant autonomic arousal on the positive lags. Black lines show the cross-correlation values, shaded grey areas indicate the SEM. Shaded yellow areas show confidence intervals from the permuted data. Significant time lags identified by the cluster-based permutation analyses are shown by a thick red line.

4.4.3 Analysis 3. Calculation of neural power changes around an attention episode

We used linear mixed effects models to examine the associations between the length of each attention episode and relative theta power at different time windows relative to the onset of that attention episode (see Figure 4.5). At 10 months, relative theta power in the time window of 0 to \pm 1000msec and \pm 1000 to \pm 2000msec after onset of a new attention episode associated with the subsequent duration of that attention episode. At 5 months, the same relationships were not significant. We found no evidence of neural activity before the start of an attention episode associating with the length of that attention episode at any time point (Figure 4.5).

The final number of accepted trials (i.e. attention episodes) in the analyses varied across the three time-windows immediately after the onset of each look. More trials were obtained for the first window (total number of looks at 5 months was 790, and 411 at 10 months) than the second (total number of looks at 5 months was 473, and 336 at 10 months) and the third (total number of looks at 5 months was 301, and 277 at 10 months). All three conditions ended up with enough number of clean trials that was greater than the recommended number of trials in the infant EEG literature (De Haan, 2002; Monroy et al., 2021; Phillips et al., 2023). Thus, the differences between the number of trials for each time window are not expected to contribute to the results described above. However, we repeated this analysis by matching the number of attention episodes at 5 months to the ones analysed at 12 months. We found no differences in the results (see APPENDIX B, Figure S2).



Figure 4.5. Calculation of theta power changes around an attention episode.

Results of the linear mixed effects models conducted to examine whether individual looks accompanied by higher theta power are longer lasting. For each look, we calculated the association between the total duration of the look and relative theta power during five time-windows (-2000msec to -1000msec and -1000msec to 0 prior to the look, and 0 to 1000msec, 1000 to 2000msec and 2000 to 3000msec before the look), using a series of separate linear mixed effects models. (A) Shows results at 5 months, and (B) shows the results at 10 months. Asterisks (*) indicate p values < .05. Central channels include: 'FC1', 'FC2', 'C3', 'C2', 'C4', 'CP1' and 'CP2'.

4.4.4 Analysis 4. Calculation of neural power changes within an attention episode

In addition to the previous analyses, which examined the associations between the length of each attention episode and relative theta power at different time windows relative to the onset of that attention episode, we also wished to examine whether power at the theta and alpha band changed significantly during a look (Figure 4.6). Relative theta was analysed as a function of these three factors: time within an attentional episode, brain areas and age with a 3-way ANOVA (Figure 4.6). There was no statistically significant interaction between the three factors. However, the analysis revealed two simple two-way interactions: one between time within an attention episode and age, F(2) = 5.58, p < .005 and the other between channel cluster and age F(2) = 11.98, p < .001. Next, we performed a multiple comparison test to find out which groups of factors were significantly different. Results are shown in APPENDIX B, Table S1-S3. A follow up analysis showed a

significant effect of "time within an attentional episode": 10-months-old infants had greater theta during the third-to-fourth second into the look (middle) than the first second (start) in both the central and the frontal poles. These effects were not present in 5-months-old infants. Similarly, relative alpha was also analysed as a function of these three factors: time within and attentional, brain areas and age with a 3-way ANOVA. We found no statistically significant interactions. Results are shown in APPENDIX B, Figure S3, Table S4.

Again, the final number of accepted trials (i.e., attention episodes) in the analyses varied across the three time-windows into each look. More trials were obtained for the first-second window (total number of looks at 5 months was 791, and 415 at 10 months) than the third-to-fourth second (total number of looks was 172 at 5 months, and 194 at 10 months) and the last second before look termination (total number of looks was 476 at 5 months, and 337 at 10 months).





Bar plots for the average relative theta power throughout a look, at both time points (5 and 10 months) and at different brain networks (central, occipital, and frontal). Asterisks (*) indicate p<0.05. Error bars represent SEMs.

4.4.5 Analysis 5. Auto- and cross-correlation analyses between infant theta activity and attention

In this section we investigated the relationship between dynamic changes in infant endogenous brain activity and their associations with moment-to-moment changes in attention. Figure 4.7A and D shows the results of the autocorrelation analyses for infant theta activity. Figure 4.7B and E shows the results for the cross-correlation analyses between infant theta activity and infant attention. Cluster-based permutation analysis revealed a significant positive association between the two variables (marked with a red line) at 10 months around time lag=0. More specifically, increases in infant theta activity at 10 months were significantly correlated with fluctuations in infant attention (Figure 4.7E). No associations were found between theta activity and infant attention at 5 months of age. Interpreting the exact time intervals over which a cross-correlation is significant is challenging due to the autocorrelation in the data (Clifford et al., 1989; Clifford et al., 2013), but the fact that the significance window is asymmetric around time 0 indicates a temporally specific relationship between infant attention and theta power, such that attention forward-predicts theta power more than vice versa.

Finally, to test whether there were any interdependencies between autonomic arousal and brain activity, we performed a cross-correlation analysis between these two variables. We found a significant cluster at 10 months (Figure 4.7F) but not at 5 months (Figure 4.7D). The asymmetry of this cluster around t=0 indicated that changes in brain activity tended to precede changes in autonomic arousal more than vice versa.



Figure 4.7. Relationship between infant relative theta activity, infant attention, and infant autonomic activity.

Autocorrelation for infant theta activity at 5 months (A) and at 10 months (D). Crosscorrelation results between infant theta activity and infant attention at 5 months (B) and at 10 months (E). Infant theta activity forward-predicting infant attention on the negative lags, infant attention forward-predicting infant theta activity on the positive lags. Crosscorrelation results between infant autonomic activity (indexed by heart rate activity) and relative theta power at 5 months (C) and at 10 months (F). Infant autonomic activity forward-predicting infant theta activity on the negative lags, infant theta activity forwardpredicting infant autonomic activity lags. Black lines show the Spearman correlation at each time lag, shaded grey areas indicate the SEM. Shaded yellow areas show confidence intervals from the permuted data. Significant time lags identified by the cluster-based permutation analyses are shown by a thick red line.

4.5 Discussion

We examined developmental changes in the physiological and neural correlates of realworld attention patterns during early development. To do so, we measured attention durations (to an accuracy of 50Hz), along with cortical neural activity (EEG) and autonomic arousal (via ECG) from typical 5- and 10-month-old infants playing alone while seated at a tabletop with 3 toys. This age range is a key period for early cognitive development, as differential patterns of brain development (Johnson & Haan, 2015) drive a transition from primarily subcortical to cortical control (Johnson, 2005), and early-emerging atypicalities can have life-long consequences (Johnson et al., 2021; Shephard et al., 2021). However, many of the mechanisms that drive early development in attention regulation remain unclear.

From Analysis 1 we found that infants at both ages showed a preferred modal reorientation rate. The modal durations of attention episodes towards different play objects were in the 0.5-0.6 second range at both ages but were lower at 5 months (Figure 4.2F, 2C). This contrasts with analyses of micro-level fixation durations (time intervals between individual refoveating eye movements), which decreases from early infancy (~0.5 secs) through to later infancy (~0.4 secs) through to adulthood (~0.3 secs) (Saez de Urabain et al., 2017; Wass & Smith, 2014). Research with adults suggests that the minimum time necessary to plan and executive a saccade is ~80msecs in adults (Nuthmann et al., 2010). Although the equivalent figure is not known in infancy, the fact that modal attention durations towards objects were shorter at 5 months than 10 months, makes it likely that the figures we observed do not simply indicate that infants were reorienting at the fastest speed possible, but rather were reorienting according to a preferred modal reorientation rate (Saez de Urabain et al., 2017).

The survival analysis showed that, at both ages, looks were fragile early in their existence and most likely to terminate in the <1 second range (Richards & Anderson, 2004) but the speed at which the survival probability curve decreased was faster at 5 months, meaning that the probability of a look lasting longer than time t was lower at 5 months (Figure 4.3). Richards and colleagues have found similar relationships in infants in both lab-based and naturalistic settings (Richards & Anderson, 2004). Overall, attention durations were shorter at 5 months; this faster-changing pattern of attention to the object was also reflected in the ACF of their looking behaviour, which decreased significantly faster, showing lower overall self-similarity. Collectively, these data fit well with what we know about the development of attention regulation. With time, we seem to observe a higher-level control of attention that allowed older infants to prioritize the task at hand – learning about/ exploring the toys – as well as to inhibit the tendency to shift attention away from an interesting task (Colombo, 2001; Courage et al., 2006; Oakes et al., 2002). Alternatively, longer attention episodes might arise because children physically manipulate objects, bringing objects closer to themselves which makes them more exogenously salient (Anderson et al., 2022; Méndez et al., 2021). In this case, then the infant's increased looking behaviour would be the result of increased exogenous attentional capture rather than an increase in endogenous attention control (Wass, Clackson, et al., 2018).

Analysis 2 examined how dynamic fluctuations in autonomic arousal relate to momentto-moment changes in attention. Consistent with previous work (de Barbaro et al., 2017; Pfeffer et al., 2022; Wass, 2021), the average concurrent correlation between autonomic arousal and attention was negative at both age points, indicating that lower arousal was associated with increased likelihood of attention. Such links have been considered within the developmental attention regulation literature, where increases in arousal are thought to lead to distraction or difficulties focusing attention, and *vice versa* (Gardner & Karmel, 1995). We also found that arousal levels were significantly forward predictive of attention at 5 months but not at 10 months (Figure 4.4B and D). Theoretically, if attentional episodes drive decelerations in the heart rate (Richards, 2011), and older infants show longer attentional episodes on average, then one could hypothesise that older infants ought also to show a more stable pattern (i.e., higher autocorrelations) in their heart rate fluctuations than younger infants. However, this was not what we observed (Figure 4.4A and C). Overall, the much shorter attention durations observed in this setting, compared with screen-based TV viewing (Richards, 2011), means that heart rate decelerations relative to individual attention episodes were observed infrequently in our data. However, our data did suggest, consistent with previous research, that at 5 months, changes in autonomic arousal forward-predict subsequent changes in attention.

In Analysis 3 we examined the associations between attention episode durations and theta power either before, or after, onset of that attention episode. At 10 but not 5 months, increased theta during the period immediately after the onset of a new attention episode (0-2000msec) forward-predicted the subsequent length of that attention episode (Figure 4.5). At neither age, however, did cortical neural activity before the onset of an attention episode forward-predicted attention durations.

In Analysis 4 we examined whether cortical neural activity changed significantly during an attention episode. Consistent with previous research (Xie et al., 2018), theta power in central and frontal electrodes increased significantly during an attention episode at 10 months, but not at 5 months (Figure 4.6). Contrary to our expectations, we did not find a link between attenuated alpha during an attention episode at any age (see APPENDIX B, Figure S3).

In Analysis 5 we investigated the relationship between dynamic changes in infants' theta activity and moment-to-moment changes in attention. We identified a significant positive association between infant theta activity and infant attention at 10 months but not at 5 months (Figure 4.7B and E). Interpreting the exact time intervals over which a cross-correlation is significant is challenging (Clifford et al., 1989; Clifford et al., 2013), but the asymmetry of the cluster around time 0 indicates that attention forward-predicted theta power more than vice versa, consistent with the findings from Analysis 3.

These findings are consistent with previous research suggesting that, by 10 months, but not during early infancy, theta oscillations increase during sustained attention and encoding (Begus et al., 2016; Jones et al., 2015; Xie et al., 2018) and associate with longer

attentional periods (Wass, Noreika, et al., 2018). Importantly, though, we found no evidence that endogenous neural markers before the onset of an attention episode forward-predict the length of an attentional episode at either age. Instead, what we found suggests that neural activity shortly after the onset of an attention episode forwardpredicts the length of that episode. One possible interpretation of this is that neural activity associates with the maintenance more than the initiation of attentional behaviours (Cohen, 1972).

Finally, we examined the relationship between theta power and autonomic arousal (Figure 4.7C and F). A cross-correlation analysis found a negative forward-predictive relationship between the two, such that increases in theta forward-predicted decreases in autonomic arousal at 10 months, but not at 5 months. This suggests that changes in the brain activity could be modulating subcortical changes (i.e. changes in the heart rate) and may thus be able to initiate or maintain states of arousal that are common to vigilant or sustained attentional states (Colombo, 2001; Richards, 2011). Overall, it appears that, by 10 months, the different substrates of attention are more inter-linked, and stronger associations are emerging between behaviour, cortical activity, and autonomic arousal (Tardiff et al., 2021).

In summary, our results suggest that, earlier in development, attentional episodes are more influenced by lower-order endogenous factors such as a preferred modal reorientation timer – which characterises infants' attention shifting more strongly -, and a general arousal system (Richards, 2010, 2011) – that might reflect a stronger influence of subcortical structures over the regulation of attention. Such factors would also be present at older ages; however, their association with attention would weaken over developmental time due to the maturation in cortical attentional areas thought to take place throughout the first year of life. Later in infancy, cortical neural activity reliably changes during attention episodes, but does not forward-predict attention at either age;

rather, it seems that neural changes associate with the maintenance more than the initiation of attentional behaviours. Overall, the regulation of attention seems to involve both arousal-based and cortical processes. With developmental time, however, the latter increases its control over the modulation of both (i.e. overt attentional behaviours and arousal), resulting in a more inter-linked system where associations between attentional systems are stronger. Theoretically, this is consistent with what we know about the development of executive attention from experimental and neuroanatomical studies.

4.5.1 Limitations and strengths

Our findings should be interpreted with consideration to a number of limitations of the study. First, our events of interest are intrinsically linked with one of the biggest EEG artefacts (i.e. eye movements), and so it is possible that residual artifact in the EEG signal may have contaminated our data. However, our data were processed using algorithms specially designed to clean naturalistic EEG data (Haresign et al., 2021; Kayhan et al., 2022), and previous analyses suggest that the electrode locations and frequency bands that we examined should be least affected by artifact, compared with more anterior locations and higher and lower frequencies (Georgieva et al., 2020). Additionally, our analyses were carefully designed to preclude this potential confound. First, our analyses compare events that we know share the same level of artefact/ noise (i.e. saccades at 5 months old contribute to comparable noise levels than at 12 months old (Noreika et al., 2020)); second, analysis 3 and 4 are time-locked to a saccade to eliminate the possibility that saccadic frequency may have influenced our results; and third, other research (Marriott Haresign et al., 2023) suggests that artifact associated with saccades disappears within ~300msecs, whereas the associations between theta and look duration lasts longer than this. For all this, we consider that the possibility that our results may have been caused by infants' saccades is unlikely.

Second, the use of different EEG systems (32- vs. 64-channel BioSemi gel-based ActiveTwo) and age groups might have contributed to the differences we observed over time. However, we compared the EEG signal quality between groups and found no significant differences (APPENDIX B, Table S5-6, Figure S4).

Third, we used a different set of toys at the two ages (see APPENDIX B, Figure S5). Consequently, this introduced a new source of variation (i.e. toy characteristics) that might have contributed to any of the observed differences (Oakes et al., 2002). However, we chose to present developmentally appropriate stimuli at the two ages to ensure that the cognitive demands were similar at the two ages. Thus, while still possible, it is unlikely that the developmental differences observed in the current study might be due to differences in the amount of information processing on the part of the infant and/ or the "interestingness" of the toys.

Fourth, it is worth mentioning that, while infants gather information about their world through aggressive visual foraging, looking and attending are not synonymous. Previous research has shown that shifts of attention can occur without shifts of gaze by 4-6 months of age (Hood, 1993; Johnson et al., 1994). However, the current study has treated them interchangeably, focusing on the study of overt attention exclusively.

Finally, our laboratory setting was a novel environment for our participants and might have elicited behaviours that are different from the ones that develop at home. However, it still represents a significant advancement relative to other screen-based or highly controlled tasks, moving closer to more naturalistic research.

4.5.2 Overall implications

This work addresses a foundational question that is central to our understanding of how attention develops, namely: what determines where, and for how long, infants

spontaneously allocate attention in naturalistic settings? And how do these influences change over developmental time? Our understanding in this area is limited because almost all previous research has studied how infants passively respond to stimuli as they appear and disappear on-screen, following an experimenter-determined sequence.

Early in infancy, slow-varying fluctuations in autonomic arousal forward-predicted attentional behaviours. Later in infancy, fluctuations in fronto-central theta power associated with changes in infants' attentiveness and predicted the length of infants' attention durations. Studying the mechanisms through which attention control develops during early life is crucial for identifying and intervening in atypical development, and conditions such as ADHD. But it is also crucial for understanding how attention and learning development can best be facilitated for typical children in educational and home settings, and to inform future research.

Additionally, this manuscript also presents new technical and analytical approaches to study early attention development in complex real-world settings. We believe it will be relevant for other researchers interested in infant attention as it reflects the summated products of years of innovation and method development.

4.6 Acknowledgments

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Chapter 5. Who leads and who follows? The pathways to joint attention during free-flowing interactions change over developmental time.

This chapter is a pre-print article submitted to *Child Development* for publication. It investigates the processes through which infants and caregivers adapt to and influence the behaviour of each other to establish joint attention and examines how these dynamics change over developmental time. Subheadings and figure placement and style have been adapted to conform to the thesis format. The supplementary materials for this chapter are available in Appendix C.

5.1 Abstract

Joint attention (JA) has been found to correlate with many developmental outcomes. However, little is known on how naturalistic JA is established and develops during early infancy. We observed free-flowing tabletop toy play between infants at 5 and 15 months and their parents (N = 48 dyads) to (1) examine developmental changes in naturalistic JA, (2) investigate whether infants become better initiators or followers of JA, and (3) explore the role of intentionally mediated forms of communication. JA episodes increased in frequency and duration and initiations of JA became more evenly distributed between the members of the dyad. Older infants became better at leading as well as following their parents' attention behaviours and directed their attention toward their partner more often.

5.2 Introduction

From about 3 to 6 months old, infants begin to be able to coordinate their attention with a social partner in connection to a separate object or event (Butterworth, 2001; Carpenter et al., 1998; Corkum & Moore, 1998; Moore et al., 2014; Mundy & Newell, 2007; Mundy & Sigman, 2015). In the literature, this phenomenon is widely known as joint attention (JA). Overwhelming evidence correlates the ability to coordinate visual attention with others to an object or event with many developmental outcomes, including language learning (Mundy & Newell, 2007; Yu & Smith, 2013), social learning (Mundy & Newell, 2007), and other broader cognitive skills (Bornstein, 1985; Schroer & Yu, 2022). But how do infants become capable of coordinating their attention with others? Surprisingly, despite universal agreement about the importance of JA as the main source of learning opportunities in infancy, there is still little agreement on exactly how the *jointness* of JA is achieved and what its theoretical underpinnings are.

Some authors understand JA as "looking where someone else is looking" (Butterworth, 2001). Others, instead, emphasise the importance of internal models about the mental state of others. They highlight the importance of shared intentionality (Carpenter et al., 1998; Tomasello & Carpenter, 2007), and argue that to be in JA, both individuals must not only be experiencing the same thing at the same time, but "they must know together that they are attending to the same thing" (i.e. they must have common knowledge) (Tomasello & Carpenter, 2007). From this perspective, JA involves using communication cues to guide and share attention with the partner.

Current understanding shows that episodes of JA become more frequent as development progresses (e.g. Aureli et al., 2022; Bakeman & Adamson, 1984), but the mechanisms underlying these changes are still unclear. Response to and initiation of JA have been reported as the main components. Responding JA (RJA) refers to infants' ability to follow the direction of the gaze and gestures of others to share a common point of reference. Alternatively, initiating JA (IJA) involves infants' use of gestures and eye contact to direct others' attention to objects, to events, and to themselves (Mundy et al., 2007; Mundy & Newell, 2007). Research in the field has often examined how these skills develop through screen-based tasks involving eye-tracking or employing standardized tests such as the Early Social-Communication Scales (ESCS; Mundy et al., 2003). Generally, the development of RJA is thought to begin early (Scaife & Bruner, 1974) and is characterized by significant improvements in accuracy during the initial year of life (Jones et al., 2014; M. Morales et al., 2000; Mundy, 2018). For example, infants younger than 12 months can follow an adult's head turn correctly but are unable to accurately locate the target the adult focuses on when multiple targets are present (Butterworth & Jarrett, 1991) or when the object is out of sight (Delgado et al., 2002; Moll & Tomasello, 2004). Similarly, infants from around 10 to 11 months, but not younger, followed a head turn when the person's eyes were open but not when they were closed (Brooks & Meltzoff, 2005). IJA, instead, is believed to start developing later during the second half of the first year of life (Billeci et al., 2016; Mundy, 2018). From around 8 to 9 months, infants start to develop the ability to use pointing and gaze to initiate episodes of JA, alternating their direction of gaze between a person and an object to share engagement (Carpenter et al., 1998; Mundy, 2018).

Two competing theories focus on infants being either passive or active contributors to JA in social interactions. On the one hand, the theory of natural pedagogy suggests that human communication is specifically adapted to allow the transmission of generic knowledge between individuals and argues that human infants are prepared to be at the receptive side of such transmission (Csibra & Gergely, 2009, 2011). The most obvious ostensive signal in human communication is direct gaze towards the addressee (Csibra & Gergely, 2009). In line with this, studies have found that even newborns exhibit a preference for faces with direct gaze as opposed to averted gaze (Farroni et al., 2006). As discussed before, while the exact age remains a topic of discussion, it is generally accepted that the ability to follow a social partner's gaze develops significantly during the first year of life (e.g. Brooks & Meltzoff, 2005; Butterworth & Jarrett, 1991; Farroni et al., 2004; Flom & Johnson, 2011; Gredeback et al., 2008). Consequently, one possibility

is that infants' ability to detect and respond to ostensive signalling from adults improves with time, thereby improving their responsiveness to JA.

Alternatively, active learning theories view infants as proactive seekers of information. For example, infants, through social referencing, babbling, and pointing, selectively seek, elicit, and modulate the information they receive from informative social partners (e.g. Begus & Southgate, 2012, 2018; Gottlieb & Oudeyer, 2018). Guided by curiosity, infants use ostensive behaviours to actively direct their partners' attention to receive new information about their environment. For example, Liszkowski et al. (2004) showed that 12-month-olds point more when the adults actively share their attention and interest with them than when they do not (Liszkowski et al., 2004). In recent studies, it has been shown that 12-month-olds display increased pointing behaviours when provided with feedback that provides new information about an object, as opposed to situations where the experimenter merely shares attention and interest with the infant (Kovács et al., 2014). Additionally, 16-month-olds exhibit similar behaviours, pointing more towards adults perceived as competent in labelling objects compared to those perceived as ignorant (Begus & Southgate, 2012). Together, these results fit well with the idea that infants expect to learn something from the response they receive to their ostensive behaviours (Southgate et al., 2007). Importantly, these ostensive behaviours used by infants to initiate JA may also be used for less instrumental but more social purposes (Bates et al., 1976 in Mundy & Sigman, 2015; Mundy, 1995). Either way, the adult dynamically adapts to the child but not vice versa (Begus & Southgate, 2012, 2018; Wu & Gros-Louis, 2015). From these perspectives, as infants get better at signalling intention, their ability to initiate JA would also improve.

Most of this research, however, has used structured tasks (Northrup & Iverson, 2020). While useful, these contexts are quite distinct from unstructured social interactions in which JA "is embedded in a stream of free-flowing activity in which parents both react to and attempt to control toddlers' behaviours and toddlers react to, direct, and sometimes ignore parents as they pursue their own goals" (Northrup & Iverson, 2020; Yu & Smith, 2017). The repetitive nature of the tasks might prompt infants to exhibit behaviours that they would otherwise not typically display in naturalistic settings, where partner behaviours unfold within a continuum of multimodal and complex dynamics. This could potentially limit the generalizability of the findings (Tang et al., 2023). Over the past decade, significant efforts have been made to study the micro-dynamics of infant attention whilst they engage in more naturalistic, free-flowing interactions with their caregiver (e.g. Abney et al., 2020; Phillips et al., 2023; Yu & Smith, 2013). This has allowed researchers to both overcome important limitations from previous research, such as studying infants in isolation or within highly structured experimental paradigms, and significantly improve our understanding on the second-by-second dynamics and influences between infant-parent dyads (Phillips, 2023; Wass, Noreika, et al., 2018; Yu & Smith, 2013).

In these naturalistic contexts, socially coordinated shifts in attention are resolved in fractions of a second (Phillips et al., 2023; Yu & Smith, 2013). For example, several studies have now found that infants spend only a small proportion of time looking to their parent's face (Franchak et al., 2011; Phillips et al., 2023; Yu & Smith, 2013). Phillips et al. (2023) compared infant-led looks to JA and non-JA in 10-month-olds and found no differences in infants' use of behaviourally ostensive cues in the 5 seconds window before the initiation (i.e. looks to parents' face and vocalisations prior to leading a look), indicating that infants do not appear to direct the focus of their attention deliberately and actively when they guide the attention of their partners. Similarly, Yu and Smith (2017) studied JA in 12- and 18-month-old infants and found that when toddlers followed their parents' attention, they rarely did so by gaze following (<15%) but instead typically followed their parents' hands to the object (Yu & Smith, 2017): a spatially and attentionally simpler, and thus, faster, pathway to JA, compared to gaze following.

Together, these studies (and others, e.g. Yu & Smith, 2013) suggest that JA is a selforganizing outcome built upon the multimodal coupling of partners' individual sensorymotor behaviours (Yu & Smith, 2013). In other words, rather than controlled, intentionally mediated ostensive signalling, it is the fast-acting, sensory-motor coordination of both partners that largely drives and maintains episodes of JA. Based on this, one hypothesis is that, with time, infants become more adept at tracking low-level behaviours. This could result in an increase in infants following their partners into JA. Notably, rather than increased looks toward the social partner, improved JA abilities during early infancy could also involve a more sophisticated integration of cues like hand gestures, vocalisations, and body movements (Suarez-Rivera et al., 2019; Yu & Smith, 2013).

To our knowledge, our understanding of how different leader-follower processes function within the dyad and evolve over early developmental time to change JA remains limited. Our aims were to: (1) examine changes in naturalistic JA, (2) explore whether these are driven by infants becoming better initiators of new attention episodes (i.e. better leaders, IJA; Hypothesis 1) or by their improved coordination with play partners (i.e. better followers, RJA; Hypothesis 2), and (3) investigate whether these changes are driven by developments in intentionally mediated forms of communication (i.e. infants engaging in ostensive signals such as looks to partner) or not.

To do this, we manually coded the gaze of parents and their 5- and 15-month-old infants as they jointly played with toys (see 5.3.2 for more details). These ages were chosen because this is an age range where dramatic changes in both RJA and IJA occur (Mundy & Newell, 2007). During this time, infants acquire numerous new skills, encompassing cognitive, communicative, and motor abilities (Feldman, 2007; Yu & Smith, 2013). Additionally, according to Tomasello and colleagues (e.g. Tomasello, 2001) it is also the time in which the ability to share intentionally mediated JA starts to emerge. By studying these ages, we aim to understand how the mechanisms that drive JA change over this unexplored time period. Tracking the momentary visual fixations of each participant allowed us to determine how infants and their mothers enter moments of JA (i.e. by following their partner or leading them into JA) and how these dynamics changed over time. We also explored the sensitivity of one partner to changes in behaviours generated by the other partner and examined the contributions and influences of infant looks to partner in establishing and organising episodes of JA.

Hypothesis 1.1 predicted that JA is increasingly infant driven, leading to increased infantinitiated JA. Hypothesis 1.2 predicted that older infants would show more intention to involve others (Carpenter et al., 1998; Mundy et al., 2007; Tomasello & Carpenter, 2007). Accordingly, we hypothesised that intentional ostensive signals such as infant looks to partner *before* and *during* a leader look would increase with time not only to guide the partner towards the new attentional focus but also to ensure that partners are, indeed, directing their attention to the same object. Additionally, we also anticipated that changes in the infants' looking behaviour would have a greater influence on mother's attention as the infants got older. To test this, we looked at how mothers responded to changes in the infants' looking behaviour at the event level (see 5.3.3.6).

An alternative hypothesis is that developmental changes in JA are driven by infants becoming better at responding to their partner's social cues. Hypothesis 2.1 predicted that older infants become more like adults (i.e. more symmetrical, less child-led). That is, the increased ability of older infants to control their attention would make them more capable of adjusting their own looking behaviour in response to their partner's behaviour. Thus, we expected an increase in infant follower looks to JA (i.e. RJA) over time. We also hypothesized that this increase in follower looks would align with greater responsiveness to their mothers' looking behaviour. Consequently, changes in the mothers' looking behaviour were expected to have greater influences on infant attention as the infants got older (see 5.3.3.6). As a secondary hypothesis (Hypothesis 2.2), we expected a higher occurrence of infants looking towards their partners both *before* and *during* a follower look to verify both shared intention and attention to the same object.

In both scenarios, improved leadership or followership abilities were predicted to lead to a greater chance of infants coordinating their attention with their mothers. Nevertheless, understanding these differences in establishing JA and how these change over developmental time is an important goal, not only because differences in the frequency of use of JA behaviours are related to subsequent language, cognitive, and social development in typical samples, but also because it may offer insights into the underlying nature of various conditions, such as Autism and Attention-Deficit/ Hyperactivity Disorder, where impairments in establishing JA are frequently observed (Brooks & Meltzoff, 2005; Mundy et al., 2007).

5.3 Materials and methods

5.3.1 Participants

Participants were typically developing infants and their mothers. The catchment area for this study was East London, including boroughs such as Tower Hamlets, Hackney and Newham. Further demographic details on the sample are given in APPENDIX C, Table S1.

Participants were recruited postnatally through advertisements at local baby groups, local preschools/nurseries, community centres and targeted social media campaigns aimed at all parents in the area. We also operated a word-of-mouth approach. Ethical approval was obtained from the University of East London ethics committee (application ID: ETH2021-0076, see APPENDIX D).

Initial exclusion criteria include complex medical conditions, known developmental delays, prematurity, uncorrected vision difficulties and parents below 18 years of age. Further exclusion criteria as well as final numbers of data included in each of the analyses for both samples are summarised in APPENDIX C Table S2. The final samples included 24 5-months-old infants (11 females) and 24 15-month-old infants (11 infant females) and their mothers. Data was analysed in a cross-sectional manner. Average age for infants was 5.3 months (std= 0.55) and 15.77 months (std= 0.87) respectively. Average age for mothers was 35.24 years (std = 4.29, N= 23) at 5 months and 36.93 years (std= 4, N= 24) at 15 months. This is the first time that any of these data have been analysed and reported.

5.3.2 Experimental design

Mothers and infants were seated facing each other on opposite sides of a table. Infants were seated either in a highchair or on a researcher's lap, within easy reach of the toys (see APPENDIX C, Figure S1). At the beginning of the joint play session, a researcher placed the toys on the table and asked the mothers to "play with their infants just as they would at home". During the play session, researchers stayed behind a divider out of view of both the caregiver and the infant. The same three toys were used for each age group (see APPENDIX C, Figure S2). The average duration of the joint play interactions was 4.94 minutes (std= 1.36) at 5 months and 6.47 minutes (std= 1.44) at 15 months. Average duration differed significantly between 5- and 15-months (tt(46) =-3.778; p<0.001). Given that the analyses are conducted relative to the duration of each interaction (e.g. look counts per minute) or on specific events (e.g. the average durations of looks at objects), variations in interaction durations should not be an issue.

The interactions were filmed using three Canon LEGRIA HF R806 camcorders recording at 50 frames-per-second (fps). Two cameras were placed in front of the infant, one on each side of the mother, and another one was placed in front of the mother, just behind the right side of the infant. All cameras were positioned so that the infant's and the mother's gaze, as well as the three toys placed on the table, were always visible (see APPENDIX C, Figure S1).

5.3.3 Data processing and statistical analyses

5.3.3.1 Synchronisation of the video data.

The cameras pointing at the participants were synchronised via radio frequency (RF) receiver LED boxes attached to each camera. The RF boxes received trigger signals from a single source (computer running Matlab) at the beginning and end of the play session, and concurrently emitted light impulses, visible from each camera and an audible beep. The synchronisation of the video coding for maternal and infant behaviour was conducted offline by aligning the times of the LED lights of the three cameras and checking that the durations matched.

5.3.3.2 Gaze behaviour coding and processing.

The looking behaviour of the infants and their mothers was manually coded offline on a frame-by-frame basis, at 50fps (see Figure 5.1 for an example of raw data). The start of a look was the first frame in which the gaze was static after moving to a new location. The following categories of gaze were coded: looks to objects (focusing on one of the three objects), looks to partner (looking at their partner), inattentive (not looking to any of the objects nor the partner) and uncodable (see APPENDIX C, Figure S1). Uncodable moments included periods where: 1) their gaze was blocked or obscured by an object and/or their own hands, 2) their eyes were outside the camera frame, and/ or 3) a researcher was within the camera frame. To assess inter-rater reliability, ~15% of the data (13 datasets) were double coded by a second coder and Cohen's kappa was calculated.

There was substantial agreement (κ = 0.636, std= 0.143) (Landis & Koch, 1977). Looking behaviour data was then processed such that any look preceding and following an "uncodable" period was excluded from further analyses. Similarly, both the first and the last look of every interaction were also excluded from further analyses.

Next, similar to other studies (Phillips et al., 2023; Yu & Smith, 2013), we interpolated through infant and mother looks to their partner before calculating leader-follower dynamics around attention episodes to objects. This is because during periods of JA towards an object, caregivers, and, to a lesser extent, infants, alternated their attention frequently between the object and their partner. Without interpolation, each subsequent look back to the object would be classified as a separate follower look to the object. This procedure therefore allowed us to accurately identify moments where one partner was leading or following their partners' attention. Interpolation involved identifying moments where the caregiver or infant directed their gaze towards a specific object, subsequently shifted their attention to their partner for less than 2 seconds, and then reverted to focusing on the initial object. By interpolating through that partner look, the partner look became an extension of the proceeding and following object look.

After interpolation, the first and last frame of all looks to object were extracted and categorised into one of our look categories: leader or follower looks. Leader looks were defined when one of the partners shifted their gaze towards an object that the partner was not already looking at. These looks were further divided into two categories: leader looks that led to JA (these were the leader looks that were subsequently joined by the partner), and leader looks that did not lead to JA (these were leader looks that were not joined by the partner). Follower looks, on the other hand, were defined as those looks that followed the partners' attention (see APPENDIX C, Figure S1). JA was defined as the periods of time when both partners were looking at the same object at the same time.



Figure 5.1. Example of infant raw data at 5 (A) and 15 months (B).

5.3.3.3 Calculation of significance: Cluster-based permutation test

To estimate the significance of the time-series relationships in analyses 3 and 4.1, we chose a cluster-based test statistic and used the so-called Monte Carlo method to calculate significance. To do so, we used a function from FieldTrip (Maris & Oostenveld, 2007) called "ft_timelockstatistics". This nonparametric framework allowed us to both control for the multiple comparison problem that arises from the fact that the effect of interest is evaluated many times (e.g. adult attention around infant looks to the partner), and to reduce the potential for false negative effects (Meyer et al., 2021).

5.3.3.4 Analysis 1. Descriptive analyses of gaze behaviour.

We looked at how many times per minute infants and adults engaged in attention to play objects, partner or inattention and for how long these attentional episodes lasted on average. We used interpolated data to compute the descriptive for object looks, and noninterpolated data for looks to partner and inattention. To calculate the average frequency (count) of attention reorientations we calculated the occurrence of each of these types of looks and divided it by the length of the session. To calculate look durations, we subtracted the last from the first frame of each look and divided it by the sampling rate (50fps) to convert "duration in frames" to "duration in seconds". To test for significant differences across ages we employed unpaired two-sample t-tests. We present the same descriptive analyses using not interpolated data in the Supplementary Materials (APPENDIX C, Figure S3).

5.3.3.5 Analysis 2. Dynamics between leader and follower looks.

First, we calculated the proportion as well as average duration of our three different types of looks of interest: leader looks to objects that were not followed by the partner and did not lead to JA (referred to as looks to non-JA), leader looks to objects that were followed by the partner and led to JA (referred to as leader looks to JA), and looks that followed their partner's attention and thus, also led to JA (referred to as follower looks). We calculated these statistics at subject level and then averaged across subjects. To test for significant differences between age groups, we employed unpaired two-sample t-tests.

Next, we calculated two ratios: mother leader looks to JA relative to mother follower looks to JA, and infant leader looks to JA relative to infant leader looks to non-JA. We tested for significant age differences in these two ratios using unpaired two-sample t-tests.

We also looked at the duration and frequency (i.e. average count per minute) of JA episodes. This was conducted using interpolated data. We performed an unpaired two-sample t-test to test for significant differences.

Finally, we collapsed all looks that led to JA (leader looks to JA + follower looks to JA; referred to as joint looks) and all leader looks that did not necessarily lead to JA (leader looks to JA + leader looks to non-JA: referred to as leader looks). Additionally, we categorised all the JA looks based on which member of the dyad terminated the JA bout (referred to as breaker looks). We calculated the frequency (count) and the average

duration of all these three new look categories (i.e. joint, leader and breaker looks). To test for significant age differences, we employed unpaired two-sample t-tests.

5.3.3.6 Analysis 3. Responsivity to changes in partner gaze behaviour.

Here we looked at how each member of the dyad responded to changes in the partner looking behaviour. More specifically, we looked at responses to looks towards the partner (Analysis 3.1) and towards the objects (Analysis 3.2).

5.3.3.6.1 Analysis 3.1. Responsivity to partner looks.

To study infant responsivity to maternal looks to the partner (i.e. infant) we first selected all mother looks to the partner (i.e. infant) and took the onset of each of these looks. Second, we took the infant data and time locked it to each of the mothers' look-to-partner onsets. Third, we selected a 20-second segment of infant data before and after each onset. To study how infant *attention to objects* changed around moments when the mothers looked at their infants, we assigned '1' to each frame within the 20-second segment where the infant was looking to objects, and '0' to each frame where the infant was not (e.g. partner, inattention, etc.). Next, we calculated the proportion of looking at the objects by summing all the 1s across events (i.e. maternal looks to the infants) and subjects and dividing it by the total number of events. To study how *attention to partner* changed around moments when the mothers looked at the infant, we assigned '1' to each frame where the infant was looking to the partner, and '0' to any other infant look. We followed the same procedure to study maternal responsivity to infant looks to the partner.

5.3.3.6.2 Analysis 3.2. Responsivity to object looks.

To study infant and maternal responsivity to partner looks to object, we followed a similar procedure as the one described above.

We first looked at infant attention to objects around moments when the mothers look at the objects, and calculated the probabilities that infants would look at the same object at the time that mothers directed their attention to an object. To do so, we took the onsets of all the mother looks to Object 1. Second, we time-locked these maternal look onsets to Object 1 in the infant data and selected a 20-second segment of infant data before and after each onset. Next, we assigned '1' to each frame within the 20-second segment where the infant was looking to Object 1, and '0' to each frame where there was any other infant look. We repeated this procedure for Object 2 and Object 3. This allowed us to calculate the probability of the infants to respond to maternal looks to objects by looking at the same object as their mothers. We did the same thing to calculate maternal responsivity to infant looks to objects.

Finally, we studied how attention to the partner changed around moments when the mothers/ infants looked at the objects. To do so, we selected all mother/ infant looks to the objects (irrespective of what object) and followed the same procedure described in 2.3.6.1.

5.3.3.6.3 Control data and significance.

To compare whether the observed responses in Analysis 3.1 and 3.2 were significantly higher than chance, we generated control data. To do so, we followed the same steps as in 2.3.6.1 and 2.3.6.2 with one distinction: instead of using the original events (e.g. times when mothers looked at the infants), we selected 100 random times throughout each interaction and used these as events to create the control data. To compare the observed

responses with the control responses we employed a CBP test (see 5.3.3.3). Similarly, to compare the observed responses between the two timepoints (5 vs. 15 months) we also employed a CBP test. Prior to this, we applied a baseline correction to facilitate the comparison across the two timepoints. To do this, we averaged the data from the first 10 seconds of each 40-second segment and subtracted it from the rest of the 40-second segment.

5.3.3.7 Analysis 4. The role of partner looks in organising episodes of joint attention.

Here we looked at the probability of a partner look *during* as well as *before* leading an attentional look (Analysis 4.1) and the probability of mothers to follow an infant leader look (Analysis 4.2). Of note, this methodology (i.e. probabilities) has been employed before with comparable datasets (e.g. Yu and Smith 2013, 2017; Phillips et al., 2023) and it has been chosen as it appears to be the most suitable approach for investigating our question, enabling us to evaluate the probability of a particular event, such as looks to the partner, happening or not happening.

5.3.3.7.1 Analysis 4.1. Probabilities of a partner look *during* as well as *before* an attentional look.

First, we calculated the proportion of time 5 and 15-months-old infants looked at their mothers *during* an attentional episode to an object. To do this, we took the non-interpolated data, calculated how frequently the infant switched their gaze to their parents within a look to the same object and divided it by the total of times infants looked at an object. We did the same with the data from the mothers. We employed unpaired two-sample t-tests to calculate age differences.

Next, we calculated the likelihood of infants looking at their mother *before* initiating a look to the object. To do this, we took the interpolated data (see 5.3.3.2) and calculated the proportion of times any infant look to the object was preceded by a look to the partner. We did the same for leader (this included all looks in which the infant was the first to look to the object, irrespective of whether these led or not to JA) and follower (this included all looks when the parent first looked to the object and the infant followed) looks. We performed the same analyses using the data from the mothers.

Following this, we calculated the likelihood of infants looking at their parents before leading or following a look to JA. We first split all JA bouts into infant-led or infant-followed. Next, we selected a 20-second segment before and after the onset of each of these looks. We then assigned '1' to each time window in which the infant was looking to their partner and '0' to each time window when they looked elsewhere. This allowed us to calculate the proportion of parent looks around infant leader and follower looks to JA.

To compare whether the observed responses were significantly higher than chance, we generated control data. To do so, we followed the same steps as above with one distinction: instead of using the original events (e.g. onsets of infant leader looks to JA), we selected 100 random times throughout each interaction and used these as events to create the control data. Lastly, we compared the observed proportions of looking at their parents around leading or following a look against both the control data and across the two age groups (5 vs. 15 months) using CBP tests (described in 2.3.3). Similar to Analysis 3, before conducting age comparisons, we applied a baseline correction following the steps described in 2.3.6.3.

5.3.3.7.2 Analysis 4.2. Probability of mothers following an infant leader look.

Finally, to explore the effect of infant ostensive behaviours (i.e. looks to partner) on mothers, we compared the probability of mothers following an infant leader look that was preceded by a partner look versus an infant leader look that was not preceded by a partner look. To do so, we took all infant leader looks (i.e. leader looks to JA and leader looks to non-JA) and split them into two groups: the ones that followed from a partner look and the ones that did not. Next, we took these two groups and calculated the probabilities of mothers to follow them. To do so, we took each infant look, time locked it to the mother's data and checked whether in the 5 seconds following that look, the mother followed (i.e. the mother looked at the same thing as the infant) or not. We did this for each look of both age groups. Previous research has also used a 5 second window to check for follower looks (see Northrup & Iverson, 2020; Yu & Smith, 2013); however, we repeated these analyses using two different post-look onset time windows in the Supplementary Materials to ensure that this decision did not alter the outcomes.

These analyses enabled us to assess the probability of mothers following infant leader looks, distinguishing between those preceded by a partner look and those that were not. It also allowed us to compare these probabilities across age groups (5 months vs. 15 months). To perform these comparisons, we performed two linear mixed effects analyses using the MATLAB function "fitlme" (refer to the Supplementary Materials for more details on the specification of the different models).

5.4 Results

5.4.1 Analysis 1. Descriptive analyses on gaze behaviour.

This section presents descriptive statistics on infant and adult gaze behaviour. We investigated how many times per minute infants and adults engaged in attention to play

objects, partner or inattention (Figure 2A) and for how long these attentional episodes lasted on average (Figure 2B). The data on attention to play objects was interpolated (see Methods 5.3.3.2 for more details on the interpolation; refer to APPENDIX C, Figure S3 for descriptive statistics using non-interpolated data).

During early infancy, infants shifted their attention frequently between play objects (M= 8.94 (std= 3.6) object looks per minute) and rarely looked at their partners (M= 2.13 (std= 1.13) partner looks per minute) (see Figure 5.1 and 5.2A). As infants grew, these dynamics changed. Older infants looked less frequently to the play objects (M= 6.72 (std= 3.2) object looks per minute) (t(46)= 2.258, p= 0.028) and more frequently to their partner (M= 4.26 (std= 1.62) partner looks per minute) (t(46)= -5.295, p< 0.001) compared to younger infants (Figure 1 and 2A). Periods of inattention became non-significantly less common with age; but, when they did occur, they lasted significantly longer (t(46)= -2.973, p<0.01). Adult gaze behaviour, instead, remained stable between the two time points with some variation in the duration of these behaviours. Attention to objects (t(45)= -2.845, p<0.01) and inattention (t(44)= -2.568, p= 0.013) increased from 5 to 15 months, and attention duration to their partners (i.e. infants; t(45)= -2.14, p= 0.039) decreased. Additionally, we also examined the distribution of the proportion of all the looks that lasted less than or equal to 5 seconds and more than 5 seconds at both timepoints and for



Figure 5.2. Descriptive analyses on looking behaviour.

both members of the dyad (APPENDIX C, Figure S4). We observed that attention durations shorter than or equal to 5 seconds follow a positively skewed lognormal distribution at both time points and for both members of the dyad.

Figure showing average number of looks per minute (A) and duration (in seconds) (B). Looks to objects have been interpolated. Looks to the partner and inattention are not interpolated. Asterisks indicate significance (* = p>0.05, ** = p<0.01, *** = p<0.001).

5.4.2 Analysis 2. Dynamics between leader and follower looks.

Here we investigated the dynamics of three different types of looks: leader looks to objects that were not followed by the partner and did not lead to JA (object looks to non-JA), leader looks to objects that were followed by the partner and led to JA (leader looks to JA), and looks that followed their partner's attention and thus, also led to JA (follower looks).

We observed that, at 5 months, when infants do lead an attention shift this generally does not lead to JA with their partners (Figure 3A and 3D). The same was true for infants at 15 months. However, during later infancy, there was a significant increase in infant leader looks that did lead to JA relative to infant leader looks that did not lead to JA (i.e. proportion of infant leader looks to JA at 5 vs. 15m increased: t(44)=-2.199, p= 0.033; Figure 3D). Older infants also performed significantly fewer leader looks to non-JA (t(44)=3.494, p= 0.001; Figure 3A). Refer to APPENDIX C, Fig. S5 to see the average number of looks per minute.

At both ages, most of the JA moments were driven by the mother responding to the infant's initiations of attention (Figure 3A and 3C). However, with time, infants became more able to also follow their partners (i.e. proportion of infant follower looks at 5 vs. 15 months increased: t(44)= -4.899, p<0.001) (Figure 3A) and, as a result, the initiations of JA became more equally distributed within the members of the dyad (i.e. proportion of

mother leader looks at 5 vs. 15m increased: t(44)= 2.221, p= 0.031; Figure 3C). Importantly, both the duration (t(44)= -2.781, p= 0.007; Figure 3E) and the average frequency of JA episodes per minute (t(44)= -4.517, p< 0.001; Figure 3F) increased with developmental time.

Next, we collapsed all looks that led to JA (leader to JA + follower to JA; referred to as joint looks) and all leader looks (leader to JA + leader to non-JA: referred to as leader looks). We found that 15-month-old infants performed significantly more joint looks (t(44)= -3.494, p<0.001) and fewer leader looks (t(44)= 4.899, p<0.001) than 5-month-old infants (Figure 3G). These results can likely be attributed to the fact that older infants engage in more follower looks but fewer looks to non-JA (Figure 3A). On the other hand, the average durations of both adult joint (t(44)= -3.26, p= 0.002) and leader (t(44)= -2.694, p= 0.01) looks increased with time. This can likely be attributed to the fact that the duration of mother follower looks increased with older infants (t(44)= -3.347, p= 0.002). Together, these findings are consistent with the idea that, at 15 months, infants engage in more JA moments, which is driven both by an increased proportion of infant follower looks, and by an increased likelihood that infant leader looks will lead to JA.

Finally, we explored the dynamics of terminating JA. We observed that mothers with older infants terminate JA moments less than with younger infants (t(44)= -2.228, p= 0.031) (Figure 3G) and the average duration at which they terminate these moments increases with time (t(44)= -2.98, p= 0.005) (Figure 3H).

Overall, we observe that infant's initiations are more likely to lead to JA with their partner at 15 months compared to 5 months and that JA episodes get longer and more frequent with time.



Figure 5.3. Dynamics between leader and follower looks.

A-B: Figures showing the proportion (A) as well as averaged duration (B) of looks to non-JA (in red), leader looks to JA (in green) and follower looks to JA (in blue). Infant (left) and mother (right). C-D: Ratios for mother leader looks to JA relative to mother follower looks to JA (C); and infant leader looks to JA relative to infant leader looks to non-JA (D). (E) Shows the average duration of JA and (F) shows the average number of JA episodes per minute. G-H: Figure showing count (G) and averaged duration (H) of different types of looks. Joint looks (pink) are looks to JA). Leader looks (blue) are leader looks to JA and leader looks to non-JA. Breaker looks (grey) indicate who terminated the shared attentional moment (JA). Asterisks indicate significance (* = p>0.05, ** = p<0.01, *** = p< 0.001). Analyses are done using interpolated data.

5.4.3 Analysis 3. Responsivity to changes in partner gaze behaviour.

Next, we looked at responsivity to changes in the partner looking behaviour. More specifically, we looked at how an individual responded to different types of looks initiated by their partner. In Analysis 3.1, we examined how they responded to partner looks, while in Analysis 3.2, we examined how they responded to object looks. Studying behaviours at the event level is important as it allows us to gain a better understanding of proximate mechanisms and influences (Schroer & Yu, 2022).

5.4.3.1 Analysis 3.1. Responsivity to partner looks.

We found no age effects in the way infants or mothers responded to partner looks. However, the results showed some interesting infant/ mother asymmetries. For infants at both ages, the probability of looking at the object increased around (and after) moments where the adult was looking at the infant (p_{5M} < 0.001, p_{15M} < 0.001; Figure 4A). For adults, instead, the probability of looking at the object decreased around moments where the infant looked at the adult (p_{5M} < 0.001 , p_{15M} < 0.001; Figure 4B). We also observed that mothers with older, but not younger infants responded when infants looked at them by looking back at their infants (p_{15M} = 0.002; Figure 4D), but infants, even at 15 months, did not respond when mothers looked at them (p> 0.05; Figure 4C).

5.4.3.2 Analysis 3.2. Responsivity to object looks.

Here, we looked at how the probability of mothers and infants looking at the *same* object changed in response to their partners' attentional shifts towards the objects (more in Methods 2.3.6.2). For infants, attention to the same object increased around moments where the adult was looking at the objects (p_{5M} < 0.001 , p_{15M} < 0.001; Figure 4E). As a result, attention to their partner decreased around moments where the adult looked at an

object (p_{5M} = 0.007, p_{15M} < 0.001; Figure 4F). For mothers, the probability of looking at the same object as their infants increased around the time when infants looked at an object at both ages (p_{5M} = 0.002, p_{15M} < 0.001), but it was significantly higher at 15 months compared to 5 months ($p_{5Mvs15M}$ < 0.001; Figure 4G). Interestingly, the probability of mothers looking at their infant increased before infants directed their gaze towards an object and decreased after infant look-to-object onset (p_{5M} = 0.007, p_{15M} = 0.005; Figure 4H).









A-D responsivity to partner looks. Probability of infants (A) and mothers (C) to look at the object when their partner looks at them. Probability of infants (B) and mothers (D) to look at the partner when their partner looks at them. E-H responsivity to object looks. Probability of infants (E) and mothers (G) to look at the same object as their partners when their partner looks at the object. Probability of infants (F) and mothers (H) to look

at the partner when their partner looks at the objects. Green thick lines indicate infants (light green) or mothers (dark green) data at 5 months. Orange thick lines, instead, indicate infants (light orange) or mothers (dark orange) data at 15 months. Thin lines are control data at 5 months (green) or 15 months (orange). Red thick lines indicate significance from the CBP test (p<0.025, two-sided).

5.4.4 Analysis 4. The role of partner looks in organising episodes of joint attention.

Next, to understand the use of ostensive cues by the members of the dyad, we explored the probability of a mother/infant to look at their partner *during* as well as *before* an attentional look (Analysis 4.1). We also examined whether infant engagement affected the likelihood of mothers to follow an infant leader look (Analysis 4.2).

5.4.4.1 Analysis 4.1. Probabilities to look at the partner during as well as before an attentional look.

Looking at the partner during an attentional episode was infrequent for infants (Figure 5A). However, older infants were significantly more likely to look to their partners, at least once, during episodes of attention to objects compared to younger infants (t(44)= - 3.589, p<0.001; Figure 5A). There were no significant differences for mothers (t(44)= - 0.989, p=0.328).

Next, we explored the likelihood of looking at the partner before initiating a look to an object (Figure 5B and C). First, we looked at whether infants looked towards their mothers before *any* kind of attentional look. We found that older infants were more likely to look at their parents right before any type of attentional look (t(44)= -5.535, p<0.001; Figure 5B). There were no significant differences for mothers (t(44)= 1.686, p=0.099). Following this, we looked at whether the likelihood of looking towards the partner before an attentional look differed between leader and follower looks. We found that older

infants were also more likely to look at their parents before leading (t(44)= -4.081, p<0.001) as well as following (t(44)= -4.353, p<0.001) a look than younger infants (Figure 5C). There were no significant differences between the probabilities of looking at the partner before leading vs. following a look at any age ($t_{5M}(22)=1.878$, $p_{5M}=0.07$; $t_{10M}(22)=1.951$, $p_{10M}=0.064$; Figure 5C). We found no significant differences for mothers, within (leader vs. follower looks: $t_{5M}(22)=-1.141$, $p_{5M}=0.267$; $t_{10M}(22)=-0.269$, p=0.791) or between (5m vs. 15m: $t_{leader}(44)=1.207$, $p_{leader}=0.234$; $t_{follower}(44)=1.987$, $p_{follower}=0.0532$) age groups. Overall, the probability of older infants looking at the partner *during* (Figure 5A) but also *before* (Figure 5B and C) initiating an attentional look to an object were higher compared to younger infants.

Finally, we explored the probabilities of infants looking at their mothers before leading or following a look to JA. At 15 months, but not at 5 months, infants were significantly more likely to look at their partners before leading a look compared to control data (p_{15M} = 0.026, one-sided; Figure 5D and E). At 5 months, infants were significantly more likely to look at the partner *before* leading compared to following a look (p_{5M} = 0.002, two-sided; Figure 5E). At 15 months, instead, the probability of looking at the partner increased for follower compared to leader looks *after* look onset (p_{15M} = 0.014, two sided; Figure 5D). We found no significant increases in the probability of infants looking at their partners before a follower look at either age. There were no significant age differences (p>0.05, two-sided; see APPENDIX C, Figure S6). Overall, to a certain degree, infants at both ages seemed to be using their gaze to lead but not to follow their partners' gaze.



Figure 5.5. Probability of looking at the partner during and before initiating an attentional look.

Plot (A) Probability to look at the partner within an attentional look to an object. (B) Probability of infants (light colours) and mothers (dark colours) to look at their partners before any kind of attentional look to an object (i.e. irrespective of whether these looks led to JA and whether they were leader or follower looks). (C) Probability of infants (light colours) and mothers (dark colours) to look at the partner before leading or following a look. Asterisks indicate significance (* = p>0.05, ** = p<0.01, *** = p< 0.001). D-E: probability to look at the partner before a leader (in green) or follower (in blue) look to JA at 5 months (D) and 10 months (E). Thin grey lines represent the control data. Red lines indicate significance from the CBP test.

5.4.4.2 Analysis 4.2. Probability of mothers following an infant leader look.

Finally, we investigated the likelihood of parents following an infant leader look that followed from a partner look (i.e. instances where infants looked at the partner before leading a look) compared to infant leader looks that did not follow from a partner look.

At 5 months, there were no differences in the probability of mothers following an infant leader look that preceded a partner look compared to an infant leader look that did not precede a partner look (β = -0.03, p=0.12, refer to APPENDIX C, Table S3 for more

details on the model specification; Figure 6). However, at 15 months, mothers were more likely to follow an infant leader look if such look was preceded by a look to the partner (β = -0.09, p<0.001, refer to APPENDIX C, Table S3 for more details on the model specification; Figure 6). That is, the probability of following an infant look increased if, prior to this look, the infants gazed at their mothers.

Following this, we compared the likelihood of mothers to follow an infant look that was preceded by a partner look across the two ages. We found that mothers with older infants are more likely to respond to infant leader looks if these are preceded by a partner look compared to mothers with younger infants (β =0.13, p<0.001, refer to APPENDIX C, Table S4 for more details on the model specification; Figure 6). We repeated these same analyses using different time windows to calculate the probability of mothers following an infant leader look and results stayed the same (see APPENDIX C, Table S3 and S4 and Figure S7).



Figure 5.6. Probability of mothers following an infant leader look.

Probability of following an infant leader look that was preceded by a partner look (dark colours) versus a look that was not preceded by a partner look (light colours) at 5 months (green) and 15 months (orange). Asterisks indicate significance (* = p>0.05, ** = p<0.01, *** = p< 0.001).

5.5 Discussion

This study investigated the mechanisms that drive JA and how these change between 5and 15-month-old infants. First, we explored changes in real-world JA and asked whether these are driven by infants becoming better initiators or followers. We also examined whether these changes are driven by developments in intentionally mediated forms of communication (e.g. increased looks to partner) or not. To do so, we observed free-flowing tabletop toy play between N=48 infants at 5 and 15 months and their mothers, and micro-coded their gaze behaviours at 50fps (see Figure 1 as an example). By tracking the momentary visual fixations of each participant, we measured how often they attended to the same object at the same time and how they entered and organised these JA episodes.

During infancy, parents switched attention rapidly between play objects and the infant's face. This pattern did not change between 5 months and 15 months. In contrast, 5-monthold infants looked only occasionally toward the parent's face. As infants grew, these dynamics changed. Older infants shifted attention less frequently between the play objects and more often toward their partner (Figure 5.2A). Figure 5.2A is also important as it provides evidence that 5-months-old infants were as proficient as 15-month-olds in alternating gaze within this naturalistic setup, generating a comparable number of opportunities for JA as their older counterparts.

At both time points, most of the JA moments were driven by the mothers responding to the infants' initiations of attention rather than leading the infants' attention (74.5% of the time at 5M and 58.6% at 15M; Figure 5.3A and C). These findings are in line with previous research (Evans & Porter, 2009; Mendive et al., 2013; Phillips, 2023; Yu & Smith, 2013) and suggest that, during infancy, the adults' role is mostly in monitoring and contingently responding to re-orientations in their infants' gaze. In following the focus of their infants' attention at moments that they reorient towards a new object, "the caregiver 'catches' and extends infant attention with reactive and dynamic change in their salient ostensive behaviours, which infants are responsive to" (Figure 5.4) (Phillips, 2023; Suarez-Rivera et al., 2019; Wass, Clackson, et al., 2018). Over time, episodes of JA

increased in duration and frequency (Figure 5.3E and F). Next, we discuss potential factors that could have contributed to these changes in JA.

First, we observed that the probability of infants looking at the partner, at least once, during an attentional look to an object was higher for older infants compared to younger infants. This could indicate that older infants check more often whether their attention is being followed than younger infants. Similarly, older, but not younger, infants were more likely to look at their partners before leading a look (Figure 5.5E). Some researchers have interpreted these shifts in gaze between the partner and the goal (i.e. the toy) as evidence that infants' behaviours become increasingly intentional and communicative (Carpenter et al., 1998; Tomasello & Carpenter, 2007). In line with this, we also observed that mothers were, indeed, more likely to follow an infant leader look if such a look was preceded by a look to the partner at 15 but not at 5 months (Figure 5.6 and APPENDIX C, S7). Additionally, we also observed that mothers with older, but not younger infants, responded when infants looked at them by looking back at their infants (Figure 5.4D) and were also more likely to respond to infants' looks to objects by looking at the same object as their infants (Figure 5.4G). Taken together, these findings indicate that infants' behaviours at 15 months are causing changes in their partners' behaviour more than at younger ages, hinting at the possibility that older infants, through these increased looks to the parent, become better at signalling intention to share their attention with them. An explanation like this would also fit with the observed increase in infant leader looks that led to JA as opposed to leader looks that did not lead to JA over time (Figure 5.3D).

Some aspects of our findings, however, are harder to reconcile with this conclusion of infants becoming more efficient leaders (Hypothesis 1.1) through increased partner looks and increased awareness of shared intentionality (Hypothesis 1.2). That is, the proportion of infants' looks to the parent's face prior to leading a JA episode was relatively small (~ 0.2 at 5 months and ~ 0.4 at 15 months) and thus, these looks, at best, can explain only

a small proportion of the observed leading looks to JA episodes. Similarly, it is important to highlight that even at 15 months, infants did not perform many "checking partner looks" *after* leading a look to verify whether their partner followed their attention (Figure 5.5E). Consequently, these more frequent (though still relatively infrequent) "partner checking looks" appear to be a plausible factor, although not the only one, influencing infants' increased ability to lead their parents' attentional interest and possibly reflect a growing (but not fully developed) understanding of others as intentional beings. Other communicative behaviours coming from the infant such as vocalisations (Carpenter et al., 1998; Liszkowski & Tomasello, 2011), pointing (Begus & Southgate, 2012; Liszkowski & Tomasello, 2011) or other hand actions (e.g. object manipulation) (Yu & Smith, 2013), as well as other characteristics from the adult partners such as increased perceptivity (i.e. ability to understand their infants) from the mothers to older infants (Thorson et al., 2018) or reduced leadership (which would allow the infant to take more of a leading role in initiating JA; Evans & Porter, 2009; Kochanska & Aksan, 2004) might also play a role in establishing infant-led JA.

Interestingly, at 5 months, even though the likelihood of looking at their parents before leading a look did not differ from chance, we found that younger infants were still more likely to look at their parents before leading compared to following a look (Figure 5.5D). This suggests that processes important to triadic engagement such as looks to the parent's face may already be coming online at this age. This is striking as research using more structured paradigms has suggested that infants typically do not exhibit these behaviours until they reach 9 months of age (e.g. Carpenter et al., 1998). Together, these results indicate that infants become better leaders with developmental time, using their gaze (and possibly other signals) to actively lead and elicit certain responses from their partner.

Crucially, similar to other studies (Kochanska & Aksan, 2004; Scaife & Bruner, 1974), we also observed that older infants became better at following their partners' gaze. At both ages, infants were responsive to their partners' behaviours. For example, the probability of looking at the same object increased around moments where the adult was looking at the objects (Figure 5.4E). However, consistent with previous findings (Custode & Tamis-LeMonda, 2020; Yu & Smith, 2013), we found no evidence that looking at their partners' face played any role in guiding infants' follower looks to JA at either age (Figure 5.5D and E). One explanation for this might be that infants are more likely to follow the hand actions (e.g. object manipulation) of their partners rather than the direction of their gaze to coordinate visual attention with them (Yu & Smith, 2013). This provides not only a faster, but also a more spatially precise pathway into infant-followed JA (Yu & Smith, 2013, 2017). Alternatively, it is possible that caregivers, observing that older infants tend to be more inattentive (Figure 5.2B), chose to make themselves more salient (e.g. by increasing the rate of modulation of the voice or engaging in object-related talks) as a more efficient way to direct and maintain the infants' attention to certain objects (Phillips et al., 2023). At 15 months, however, infants were more likely to look at their parents before following a look compared to 5-month-old infants (Figure 5.5C). Additionally, older infants were more likely to shift their gaze towards their parents after following compared to leading looks (Figure 5.5E), perhaps to verify whether their parents were indeed focusing on the same object to which the infants had directed their gaze. However, it is important to note that the proportion of infants' looks to the parent's face at 15 months before and after following a JA episode was, again, relatively small (<0.5) so the increase in infant follower looks might only be partly explained by an increase in infant intentionally mediated behaviours. These results fit well with previous research from Yu and colleagues (e.g. Yu & Smith, 2013, 2017). Collectively, though, our findings might indicate not only the increased ability of older infants to control their attention and shift between person and object, but also the infant's growing appreciation that relationships are not always fully attuned to one's needs (Feldman, 2007; Gredeback et al., 2008).

Overall, the current study showed that, with time, the initiations of JA became more equally distributed within the members of the dyad. This shift evolved from the infants being passively engaged into JA by the mothers to the infants actively following and engaging the mothers some months later. As a result, older infants not only became more efficient leaders (i.e. performed more leader looks that led to JA, Hypothesis 1) but also followers (i.e. engaged in more follower looks to JA, Hypothesis 2). Older infants also appeared more intentional (i.e. performed more looks to the partner that resulted in mothers responding more); nevertheless, even at 15 months, JA was still predominantly achieved through mechanisms other than looking towards the partner.

5.5.1 Limitations

Our findings should be interpreted with consideration to a number of limitations of the study. First, we focused exclusively on overt (visual) attention and specifically examined looking behaviours of both infants and their mothers. However, we know that visual attention is not the only modality that can influence JA and that there are many other behaviours that can shape these dyadic processes through which JA is established such as vocalisations, gestures (e.g. pointing, object handling) and touch (amongst others) (Yu & Smith, 2017, Schroer & Yu, 2022, Suarez-Rivera et al., 2019, Deak 2000). Second, observing mother-infant dyads interact in a free play setting within a table-top interaction provided a naturalistic setting and increased generalizability of the study. However, using only this free play task may have limited the variability of interactional patterns that the dyads engaged in. It may have also put pressure on mothers to engage in the interaction more than they would have otherwise, since they were given directions to *"play with their children as they normally would"* while being video recorded (Abney et al., 2020). Third, our cross-sectional design limited our ability to make predictions regarding the stability and predictability of specific dyadic dynamics. It is likely that certain characteristics of

the dyad (e.g. mother or infant over- or under-responsiveness) may, to an extent, influence the way JA is organised and how it changes over time (Evans & Porter, 2009). Relatedly, we know that the history of interactions with others such as siblings, peers, and other caregivers, may also shape the way infants and mothers interact and establish JA. However, in this study we did not consider the potential varying levels of exposure to others nor its impact on the dynamics between infants and their mothers. Fourth, our sample is considerably homogeneous in terms of ethnicity, culture and socioeconomics and consists only of mothers (APPENDIX C, Table S1). It will be useful for future studies to include investigations from more heterogeneous groups (Feldman, 2007; Mundy et al., 2007) as well as to include fathers, given their increased involvement in their infants' lives (Aureli et al., 2022). Fifth, one of the criteria used to exclude participants from the current study was the presence of "known developmental delays." However, it is important to note that certain diagnoses, like Autism Spectrum Disorder (ASD) and Attention-Deficit/Hyperactivity Disorder (ADHD), may not be identifiable until later stages of development. Additionally, we know that ASD and ADHD often have a genetic component, however, the current study did not inquire about family history of these conditions. As a result, there is a possibility that some infants included in the study may have been at greater likelihood of developing these conditions than others. While there is a possibility that this factor may have influenced our results, it is also worth noting that our findings closely resemble those of other studies (e.g. Yu & Smith, 2013, 2017; Phillips et al., 2023).

Finally, throughout the discussion we considered this "checking behaviour" from the infant to the parent as reflecting some kind of monitoring of the partners' behaviours. Nonetheless, alternative interpretations have been proposed by other researchers. For example, it could be that that these behaviours from the infants might be merely to verify their mother's presence (e.g. for emotion regulation, see Carpenter et al., 1998) or just

displaying conditioned responses to their mothers' smiles, contingent vocalisations, and other expressions of pleasure and interest. Interpretations like these would indicate that infants do something different than monitoring the adults' intentional behaviour.

5.5.2 Overall implications

Joint attention is the main source of learning opportunities in infancy. We know more about JA than ever before but our understanding on how infants enter real-world JA remains limited because much of the previous work has used structured lab-based tasks, which are significantly distinct from the unstructured, fast-moving social interactions: do infants lead or follow their parents into JA?

We observed that both the duration and the average frequency of JA episodes per minute increased with developmental time and the initiations of JA became more equally distributed within the members of the dyad. Infants became better at leading as well as following their partners into JA with developmental time and seemed more intentional; nevertheless, even at 15 months, JA was still predominantly achieved through mechanisms other than looking towards the partner.

We know that deficits in joint attention are often early indicators of developmental disorders such as ASD and ADHD. Consequently, understanding the emergence of JA in real-world scenarios is not only important to understanding overall cognitive development and learning, but also crucial for identifying potential signs of atypical development at an early stage, allowing for timely intervention and support.

This work not only expands our understanding of the leader-follower dynamics of JA in naturalistic settings, and how these change over developmental time, but it also offers key insights into new methodological and analytical techniques to study these.
Chapter 6. General discussion

This final chapter summarises the three empirical studies conducted for this thesis. Next, it discusses the limitations of the work and presents recommendations for future research. Finally, it provides an interpretation and integration of the main findings in the context of the broader literature. Concluding remarks and the implications of the work are considered at the end of the chapter.

6.1 Summary of findings

The present thesis was set to examine the developmental course of co- and self-regulation. More specifically, we looked at the development of physiological arousal and attention regulation. The development of these regulatory abilities during the first years of life is thought to be critical for the subsequent emergence of autonomy and the development of social, cognitive, and behavioural competencies (Bell & Calkins, 2000; Calkins et al., 2002; Cox et al., 2010; Fox & Calkins, 2003). The primary goal of this thesis was to contribute to the existing literature by addressing current gaps in the study of these early regulatory abilities, which included recognising a more active role for the infant, treating the dyad as a unit of analysis as opposed to studying infants in isolation (which seemed crucial to avoid overlooking the significance of co-regulation during infancy), shifting away from static methodologies, and enhancing the ecological validity of the research, amongst others (see chapter 2 for a detailed view of the gaps in the literature).

The first empirical study (chapter 3) examined developmental changes in naturalistic physiological co-regulatory processes during early life. This involved collecting day-long physiological arousal, proximity, and vocal data from 89 caregiver-infant dyads at 5 and 10 months, using wearable devices. More specifically, we measured the bi-directional dynamics between infants and their mothers around negative and neutral-to-positive

affect vocalisations from the infants. We investigated how these early inter-relationships within the dyad varied based on infant affect, evolved across developmental time, and associated with infant calming.

First, we found that infants' negative vocalisations were shorter at 10 months. Second, we examined whether these changes were due to improved self-regulation or more effective co-regulation. We were unable to replicate previous findings that concurrent infant-mother physiological synchrony increased around negative vocalisations. However, we found changes in sequential synchrony, such that infants' physiology forward-predicted changes in the mother's physiology around negative vocalisations at 5 but not at 10 months. In other words, at 5 months, mothers were more active partners in synchronizing to infants' state. Similarly, we observed greater and more long-lasting reductions in mother-child proximity following negative affect vocalisations at 5 months compared to 10 months. Similar to previous findings, we observed that the more mothers upregulated their physiological arousal around child negative affect vocalisations, the bigger the decrease in infant arousal was during the 3-5 minute window following the event at both time points. Interestingly, increased caregiver involvement did not associate with faster quieting at 5 months. Instead, caregiver responses became more predictive of infant quieting at 10 months. Based on this, we argued that dyadic strategies, rather than diminishing in importance, remain significant for infant physiological arousal regulation. In the following two chapters, we examined the development of attention regulation and co-regulation in solo- (chapter 4) and joint- (chapter 5) play settings, respectively. In chapter 4, we looked at the development of the capacity to regulate attention within complex, dynamic, real-world settings. To do this, we recorded brain activity, physiological arousal, and spontaneous attention patterns (i.e. looking behaviour) in 58 5- and 10-month-old infants during free solo play. We used time series analyses to

examine whether changes in physiological arousal and brain activity anticipate attention changes or follow on from them.

Early in infancy, slow-varying fluctuations in physiological arousal forward-predicted attentional behaviours, but cortical activity did not. By later infancy, fluctuations in fronto-central theta power – a cortical EEG rhythm commonly linked with attention in infancy (e.g. Jones et al., 2020; Meyer et al., 2019; Orekhova et al., 1999; Throm et al., 2023; Xie et al., 2019) – associated with changes in infant attentiveness and predicted the length of infant attention durations. But crucially, changes in cortical power followed, rather than preceded, infant attention shifts, suggesting that processes after (but not before) an attention shift determine how long that episode will last. We also found that changes in fronto-central theta power modulated changes in arousal at 10 but not at 5 months. Collectively, our results suggest that the regulation of real-world attention involves both arousal-based and cortical processes but point to an important developmental transition. As development progresses, attention control systems become dynamically integrated and cortical processes gain greater control over modulating both arousal and attention in naturalistic real-world settings.

In chapter 5, we looked at the dynamics and influences between infant-parent dyads in their capacity to share attention with a social partner. In essence, the study focused on the influences of the social environment on infants' capacity to regulate attention. First, we examined developmental changes in naturalistic joint attention (JA) and asked whether these are driven by infants becoming better initiators of new attention episodes (i.e. better leaders) or by their improved coordination with play partners (i.e. better followers). We also explored how the behaviours generated by one partner influenced the other partner. Second, we asked whether these changes in initiating and/or responding were driven by developments in intentionally mediated forms of communication or not. To test this, we observed free-flowing tabletop toy play between 48 infants at 5 and 15 months and their

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mothers. We micro-coded gaze behaviours at 50fps to determine whether infants and their mothers enter moments of JA by following their partner's gaze, or by leading them into JA, and to explore each partner's moment-by-moment sensitivity to their partner's gaze cues.

During early infancy, infants shifted their attention frequently between play objects and rarely looked to their parent. When younger infants did initiate an attention shift, this generally did not lead to a JA episode. Most moments of JA were driven by the mother responding to the infant's initiations of attention. The same was true for older infants; however, older infants directed their attention toward their partner *during* and *before* initiating looks more often. This, in turn, increased the likelihood of mothers following their gaze, resulting in a significant increase in infant leader looks that did lead to JA. Older infants also became better able to follow their partners into JA. Our findings suggest that, with time, initiations of JA shift from mostly being driven by the mother adapting to the infant to a more balanced pattern of initiations within the dyad. This transition involved older infants taking on a more active role in both initiating and responding appropriately to the social behaviours and demands of the social partner.

6.2 Limitations and future directions

Though specific study limitations are discussed at the end of each individual empirical chapter, there are further, more general, topics that require attention.

First, despite efforts to present ecologically valid research in this thesis, this was only partly successful. Even though no experimental manipulation was used, and the free-play interactions used in chapters 4 and 5 to measure infant and parental behaviours were designed to be as naturalistic as possible, data collection nevertheless took place in a relatively controlled and unfamiliar setting that may have influenced both infant and

parental behaviours. Similarly, in the home environment part of the study (chapter 3), caregivers, knowing they were being recorded, may have potentially altered their everyday interactions with their infants.

Second, we know that certain characteristics of the dyad (e.g. attachment style, Cox et al., 2010), the infant (e.g. temperament and reactivity, Braungart-Rieker & Stifter, 1996; Suata, 2023) and the caregiver (i.e. maternal mental health, Davis et al., 2018; de Barbaro et al., 2023; Smith et al., 2022) may all, to an extent, influence the co-regulatory dynamics within the dyad as well as the development of infants' appropriate regulation. Relatedly, we know that different family characteristics such as socio-economic background or number of siblings (Bridgett et al., 2013; Conejero & Rueda, 2018; Noble et al., 2007; Vernon-Feagans et al., 2016; Wass et al., 2019), may also shape the way infants and mothers interact with each other. Due to time limitations, we were unable to incorporate them into the current thesis. Future studies employing longitudinal approaches (instead of the current cross-sectional design) that consider the wider environment in which regulatory processes occur and develop as well as individual differences from the members of the dyad could unquestionably enhance and offer deeper insights into our findings. Additionally, a longitudinal approach like the suggested could also allow researchers to look at short- as well as long-term associations between the regulation of physiological arousal and attention. This aspect remained unexplored in the current thesis due to its cross-sectional design.

Third, our sample was considerably homogeneous in terms of ethnicity and culture, and consisted only of mothers. Consequently, our findings may not necessarily be representative of caregiver–child relations more generally. It will be useful for future studies to include investigations from more heterogeneous groups (Feldman, 2007; Mundy et al., 2007) as well as to include fathers, given their increased involvement in their infants' lives (Aureli et al., 2022). Relatedly, another pertinent issue to be considered

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among all the studies of the thesis is the self-selection bias, which refers to the systematic, non-random difference in characteristics between individuals who choose to participate in a study and those who do not. This may have impacted the likelihood of certain types of participants taking part in the research of this thesis.

6.3 Integration of main findings

At the beginning of the thesis (chapter 2) we reviewed the literature on regulation. More specifically, we discussed the development of two key regulatory abilities in infancy: regulation of physiological arousal and attention. Much of the infant/ child research on the development of these skills emphasizes the gradual increase in self-regulation. That is, with time, infants seem to be more able to regulate their arousal and attentional states on their own. This has led researchers to suggest a transition from the infant's initial reliance on the caregiver for direct regulatory assistance to the progressive internalization of regulation (Calkins et al., 1998; Ekas et al., 2013; Kopp, 1982, 1989; Thompson, 1994; Wass et al., 2023). Although the transition from dyadic to independent regulation strategies is considered a normative part of development, it is difficult to pinpoint exactly when this transition occurs (Atkinson et al., 2021). This led us to explore whether we could demonstrate that infants become progressively less contingent on others as time progresses.

In chapter 3 and 5, we found that dyadic strategies, rather than being phased out or replaced, seem to continue to play an important role at 10 (chapter 3) and 15 months (chapter 5) in both physiological and attentional regulatory processes. On the one hand, in chapter 3 we observed that mothers of 5-months-old infants, but not 10-months-old infants, were the more active partner in coordinating their physiological states with those of the infant. Mothers of younger infants also decreased the distance between them and their infants more and for longer than mothers of older infants. However, despite the fact

that mothers of 5-months-old infants appeared to be more responsive to their infants' signals, the relation with mother's responsiveness and infant's quieting strengthened with time. One possible explanation to this could be that only older, but not younger, infants might have been able to use their mothers' support and/or to adapt to them to co-regulate their physiological arousal more effectively.

In chapter 5, on the other hand, we found that mothers were responsible to create most of the shared attentional moments with their infants. However, older infants were better and more intentional at both initiating as well as responding to JA than younger infants. This resulted in a more balanced distribution of JA initiations between dyad members, less child led.

Collectively, these findings point at two potential developmental implications. First, it seems that infants, rather than becoming less dependent on others as we previously hypothesised, become progressively skilled in employing dyadic strategies. Some studies align with this idea. For example, research has found that regulatory strategies such as looking at or communicating to the mothers when infants faced emotion-eliciting situations (e.g. arm restrain) increased with time (Atkinson et al., 2021; Braungart-Rieker & Stifter, 1996; Rothbart et al., 1992; see 2.2.4 for more). Similarly, Bakeman & Adamson (1984) found that the time spent in coordinated attention with a social partner increased over development, but not the time spent in solitary object play. Collectively, these and our results challenge our initial hypothesis that infants would gradually become less dependent on others.

Second, our findings seem to also indicate that, early in development, the caregiver adapts to and coordinates with the infant more than the other way around. As development progresses, the infants seem to learn to also adapt to the caregiver's responses. Findings of bi-directional coordination are well-documented and extensively replicated in the literature. Generally, it is widely accepted that caregivers influence their infants and

infants, in turn, influence their caregivers (Bell et al., 1979; Cohn & Tronick, 1988; Jaffe et al., 2001). Nevertheless, the potential asymmetry within this bi-directional coordination, and how it changes over developmental time has been frequently overlooked (Beebe et al., 2016). Of the few studies that have examined co-regulation longitudinally, most have used Fogel's coding system where co-regulation is categorized into three patterns: symmetrical (when mother and child are engaged with each other and both contribute to the ongoing interactions), asymmetrical (when mother and child are engaged but only one contributes to the interaction) and unilateral (when one person ignores the other's bids for attention, engaging instead in their own activity) (Fogel et al., 2003). Results from these studies have consistently shown that unilateral coregulation, in which only the mother is actively involved, largely prevail at the beginning of the first year of life and then decreases linearly. Instead, time spent in symmetrical co-regulation follows a positive linear trend over time (Aureli et al., 2022; Aureli & Presaghi, 2010; Doiron et al., 2022; Evans & Porter, 2009). Using a different approach, Feldman et al., (1999) measured affect synchrony during the first year and found that significantly more dyads engaged in mutual synchrony at 9 months than at 3 months. Interestingly, they found that the lead-lag structure shifted from the parent's synchronizing with the infant state at 3 months to mutual adaptation at 9 months (Feldman et al., 1999). Together, summarising over different methodologies and modalities, these and our findings seem to indeed point to a shift from relying on one single partner (i.e. the mother) to the active involvement of both partners (i.e. mothers and infants) to maintain regulation over time (Aureli et al., 2022; Aureli & Presaghi, 2010; Beebe et al., 2016; Chow et al., 2010; Evans & Porter, 2009; Feldman et al., 1999). In other words, it seems that more symmetric dyadic regulation follows from caregiver-led regulation (Calkins, 2007; Sroufe, 2000; Taipale, 2016). This transition is important as dyadic regulation (i.e. co-regulation) has been suggested as a key contributor to the emergence of self-regulation (e.g. Butler & Randall, 2013; Feldman, 2007).

So far, we have argued for an increase in dyadic strategies and proposed that these changes are predominantly driven by the child taking on a more participative role in co-regulatory processes. But does this imply that self-regulation does not improve as infants mature? In the next final lines, we discuss the possibility that infants becoming a more active partner in co-regulatory processes might actually reflect (or be facilitated by) increased self-regulation.

In chapter 4 we observed that cortical processes – thought to relate with a more mature type of endogenous or internally directed attention – become progressively more important in the regulation of real-world attention than lower-order endogenous factors such as physiological arousal. Interestingly, we also found that cortical activity increased its control over the modulation of physiological arousal. One hypothesis is that, with time, the regulation of both physiological arousal and attentional episodes became more influenced by higher-order neural responses, resulting in more voluntarily regulated attentional, and potentially also physiological states. An explanation like this would not only align well with the current findings but also with idea that the maturation of cortical attentional systems (i.e. increased self-regulation of attentional states) might contribute to a better regulation and internalization of physiological arousal regulation (Rothbart et al., 1992, 2011; Wu et al., 2021; refer to 2.4 for more).

Generally, infants rely on their attentional skills to engage effectively with objects and caregivers. This, in turn, may create a calm and positive mental state, reducing the need for hypervigilance, which involves excessive scanning of the environment and is often accompanied by heightened arousal (Aston-Jones et al., 1999; Aston-Jones & Cohen, 2005; Wass, 2017; Wu et al., 2021). Simultaneously, active engagement with caregivers offers infants opportunities to seek assistance during distressing moments. Thus, one

hypothesis is that the maturation of attentional regulatory systems seen in chapter 4 could both contribute to the development of adaptive physiological regulation (see also Wu et al., 2021) and be the very factor that enabled infants to participate more actively in dyadic regulatory processes.

Of note, further research on short- and long-term interactions between attention and physiological regulation is required. Nevertheless, and even though infants may not have fully developed their capacity to effectively regulate their physiological and attentional states independently by the end of infancy, it is plausible that their progress in regulatory skills enabled them to increasingly participate and actively engage in dyadic regulation. An explanation like this would fit well with Calkins (2007), Sroufe (2000) or Taipale (2016), who, in separate reviews of the literature, suggested that "dyadic regulation follows upon the heels of caregiver-orchestrated regulation".

In sum, the less developed infant's self-regulatory capacities are, the more the caregivers not just *assist* but *manage* the infant's regulation. Over time, infants' capacity to effectively regulate physiological and attentional states independently develops, enabling them to take a more active role in dyadic regulatory processes.





The plot on the left shows the trajectories initially hypothesised: as self-regulation increases, co-regulation decreases. The plot on the right, instead, shows the new hypothesis: the increase in self-regulation enables an increase in co-regulatory processes.

6.4 Concluding remarks

One of the main contributions of this thesis is in taking a new, multi-method approach that combines neural, physiological, and behavioural techniques as well as home- and naturalistic laboratory-based research to study the development of regulatory processes in infancy. Methodological advances like these are important not only to identify potential new and more naturalistic ways to study infant development, but also to increase the generalisability of the results.

This thesis also aimed to stress the importance of both considering a more active role from the infants and moving beyond studying them in isolation. It also highlighted the importance of examining dynamic, two-way relationships that develop between infants and their immediate social and physical environment and explored new analytical approaches capable of capturing them. Additionally, it also aimed to emphasize the importance of understanding how these bidirectional relationships change over developmental time to shape (and adapt to) infant's development.

Finally, the present work also generated new perspectives on our current understanding of the development of early dyadic regulatory process by suggesting that infants, rather than becoming less dependent on others with developmental time as we previously hypothesised, become progressively skilled in employing dyadic strategies. From a developmental perspective, it is crucial to joint efforts into delineating these early regulatory trajectories and their developmental progression, as it could not only offer valuable insights into non-normative developments but also raise important new avenues for intervention research. For example, future studies focusing on the potential for parenting behaviour to support the development of infant regulatory behaviours may provide further evidence for parent-mediated interventions. Put differently, investigating how parental behaviours influence outcomes in infant regulatory behaviour could provide valuable insights for understanding parent-mediated risk for infant maladaptive self-

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regulation strategies. Parenting may be a suitable intervention target due to its influential role in shaping early experiences, fostering secure attachment relationships, modelling adaptive behaviours, and providing a supportive environment for growth and learning. By supporting parents in their caregiving roles, interventions can have an early, profound, and lasting impact on infant development.

On the other hand, extending this research to populations at higher risk of regulation difficulties, such as infants with a familial background of ASD or ADHD, would not only deepen our understanding of co- and self-regulation development in these groups in these populations, but also help us identify new targets for early intervention and possibly early identification as well.

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APPENNDIX A - Supplementary materials for: Do shorter infant distress episodes with age result from improvements in caregiver-infant coregulation or infant self-regulation?

- 1. Table S1. Demographic data split by age visits.
- 2. Table S2. Table summarising the final number of datasets and number of vocalisations for both samples as well as the reason for exclusion.
- 3. SM1. Voice Type Classifier (VTC) and Speech Emotion Recognizer (SER).
- 4. Figure S1. Percentage of vocalisations per hour split by affect.
- 5. Table S3. Model for Duration of Vocalisations.
- 6. Table S4. Average duration of vocalisations overall and split by affect.
- 7. Table S5. Number and duration of vocalisations per hour split by age.
- 8. Table S6. Number and duration of vocalisations per hour split by age and affect.
- 9. Table S7. Model for Infant Arousal at the time of a vocalisation.
- 10. Table S8. Model for Infant autonomic reactivity.
- 11. Table S9. Infant change in activity levels.
- 12. Table S10. Model for Caregiver Arousal at the time of a vocalisation.
- 13. Table S11. Model for Caregiver autonomic responsiveness.
- 14. Table S12. Caregivers change in activity levels.
- 15. Figure S2. Comparison of proximity levels based on age.
- 16. Table S13. Speed of recovery as a function of Age.
- 17. Figure S3. Association between caregiver's autonomic responsiveness and speed of infants' recovery.
- 18. Figure S4. Concurrent synchrony in infant-caregiver arousal using different window length and overlaps.
- 19. Figure S5. Results from the WCC and cluster-based permutation analysis using different time window widths.

	5 MONTHS	10 MONTHS
Maternal education		
Post-graduate degree	45.45	43.48
Degree	45.45	43.48
F.E. Qualification	0	0
A Level	6.82	8.70
GCSE's	2.27	4.35
No formal qualification	0	0
Other	0	0
Prefer not to answer	0	0
Household income		
Under 16k	0	0
16k - 25k	0	0
26k - 35k	0	0
36k - 50k	6.82	4.35
51k - 80k	25.00	26.09
More than 80k	54.55	60.87
Prefer not to answer / No reply	13.64	8.70
Maternal ethnicity		
White British	45.45	69.57
Other White	22.73	8.70
Asian (Chinese, Indian)	13.64	4.35
Black	6.82	8.70
Mixed – White and Black Caribbean	4.55	4.35
Prefer not to answer / No reply	6.82	4.35

APPENDIX A - Table S.1. Demographic data split by age visits.

APPENDIX A - **Table S.2.** Table summarising the final number of datasets and number of vocalisations for both samples as well as the reason for exclusion.

	Negative vocalisations		Neutral vocalisations			
5 MONTHS	Vocalisation count	%	Number of participants remaining	Vocalisation count	%	Number of participants remaining
Initial numbers	26770	100	60	15546	100	60
Number of vocalisations with full datasets *	25260	100	57	14931	100	57
Discarded, proximity >10m	-2708	10.72	57	-1380	9.24	57
Discarded, vocalisation within first/ last 10min	-426	1.89	57	-346	2.55	57
Leads 80% **	-5644	25.51	51	-3338	25.28	51
Leads 60% **	-708	4.3	51	-375	3.8	51
Less than 10 vocalisations ***	-11	0.07	49	-7	0.07	49
Final numbers	15763	62.4	49	9485	63.53	49

10 MONTHS	Vocalisation count	%	Number of participants remaining	Vocalisation count	%	Number of participants remaining
Initial numbers	16080	100	33	9009	100	33
Number of vocalisations with full datasets *	15990	100	32	8923	100	32
Discarded, proximity >10m	-2266	14.17	31	-928	10.4	31
Discarded, vocalisation within first/ last 10min	-298	2.17	31	-249	3.11	31
Leads 80% **	-4663	34.73	27	-2867	37.01	27
Leads 60% **	-1088	12.42	26	-570	11.68	26
Less than 10 vocalisations ***	0	0	26	0	0	26
Final numbers	7675	48	26	4309	48.29	26

* Some datasets were not completed (i.e., the data for caregiver or infant was missing, mostly due to recoding error).

** ECG leads were not detected for more than $80\%\,/\,60\%$

*** After all the pre-processing steps, participants that had less than 10 vocalisations remaining were excluded from further analyses.

APPENDIX A - SM1. Voice Type Classifier (VTC) and Speech Emotion Recognizer (SER).

To ascertain the precision/recall we could expect from each choice of VTC confidence estimate threshold we compared the estimates from the VTC against the hand coding. For this paper, we chose a relatively high confidence estimate threshold, which yield a precision of 0.8 and a concomitant recall of 0.3.



Below is shown the confusion matrix used for evaluating the performance of the SER classifier.



INFANT ACOUSTIC VALENCE CLASSIFIER



and darker colours are for data at 10 months (V2).

APPENDIX A - **Figure S.1.** Percentage of vocalisations per hour split by affect. Bar plot showing the percentage of negative (red) and neutral (green) vocalisations per hour at both time points (5 and 10 months). Lighter colours are for data at 5 months (V1)

APPENDIX A - Table S.3. Models to test effect of Affect, Age, and Affect * Age

interaction on duration of infant vocalisations.

Model for Duration of Vo	calisations.							
Dependent/ Response varia	ble = vocalisa	ation durations.						
Fixed effects (predictor var	iables) = Affe	ect, Age and inte	eraction betw	een Affect *	Age.			
Random effect for subject.								
	BValue	Std.Error	DF *	t-value	p-value			
Affect	-0.217	0.044	65013	-4.931	0.000			
Age_group	-0.183	0.078	87	-2.340	0.022			
Affect:Age_group	0.106	0.030	65013	3.534	0.000			
Duration of Vocalisations	as a function	n of Affect						
Dependent/ Response varia	ble = vocalisa	ation durations a	tt 5M (1) or a	at 10M (2).				
Fixed effects (predictor var	iables) = Affe	ect (neutral/ neg	ative).					
Random effect for subject.								
	BValue	Std.Error	DF *	t-value	p-value			
1. 5M - Affect	-0.111	0.019	40133	-5.727	0.000			
2. 10M - Affect	-0.005	0.022	24880	-0.209	0.835			
Duration of Vocalisations	as a function	n of Age.						
Dependent/ Response varia	ble = vocalisa	ation durations (irrespective	of the Affect)	(1) and			
vocalisation durations for n	eutral (2) or 1	negative Affect	(3).	,				
Fixed effects (predictor var	iables) = Age	e (5M/ 10M).						
Random effect for subject.	, C	. ,						
	BValue	Std.Error	DF *	t-value	p-value			
1. Duration of <i>a</i>	0.142	0.078	87	1 817	0.073			
vocalisation	-0.142	0.070	07	-1.01/	0.075			
2. Neutral - Age_group	-0.036	0.079	87	-0.452	0.652			
3. Negative - Age_group	-0.193	0.084	87	-2.305	0.024			

* Analyses include all subjects with full datasets ($N_{5M} = 57$, $N_{10M} = 32$).

	Average duration (s)	std
VISIT 1 (5 months)	1.56	1.85
Neutral	1.45	1.74
Negative	1.63	1.91
VISIT 2 (10 months)	1.44	1.65
Neutral	1.41	1.76
Negative	1.46	1.58

APPENDIX A - **Table S.4.** Average duration in seconds (with standard deviation) of vocalisations overall and split by affect (neutral vs. negative) at 5 and 10 months.

	5 months			10 months			
	Number	Duration (s)	std	Number	Duration (s)	std	
1st hour	6412	1.62	1.81	3754	1.70	1.96	
2nd hour	4896	1.56	1.84	2620	1.56	1.70	
3rd hour	5300	1.48	1.68	3583	1.44	1.47	
4th hour	6058	1.65	2.03	3577	1.40	1.86	
5th hour	4919	1.55	1.75	3169	1.32	1.68	
6th hour	4857	1.48	1.70	2892	1.34	1.47	
>7th hour	7749	1.45	1.89	5318	1.26	1.02	

APPENDIX A - **Table S.5.** Number (count) and average duration in seconds (with standard deviation) of vocalisations per hour split by age.

	Negative vocalisations					Neutral vocalisations				
	5 months		10 months		5 months		10 months			
	Number	Duration (std)	Number	Duration (std)	Number Duration (std)		Number	Duration (std)		
1st hour	4257	1.53 (1.78)	2656	1.38 (1.41)	2155	1.59 (1.96)	1098	1.40 (1.38)		
2nd hour	3145	1.60 (1.78)	1712	1.29 (1.25)	1751	1.59 (1.97)	908	1.41 (1.45)		
3rd hour	3126	1.61 (1.89)	2490	1.32 (1.32)	2174	1.62 (1.88)	1093	1.42 (1.37)		
4th hour	3994	1.58 (1.93)	2194	1.32 (1.30)	2064	1.55 (1.75)	1383	1.38 (1.40)		
5th hour	3181	1.56 (1.79)	1931	1.39 (1.39)	1738	1.65 (1.95)	1238	1.26 (1.24)		
6th hour	2878	1.65 (1.98)	1856	1.33 (1.30)	1979	1.51 (1.63)	1036	1.31 (1.34)		
>7th hour	4679	1.50 (1.58)	3151	1.33 (1.26)	3070	1.74 (2.13)	2167	1.23 (1.46)		

APPENDIX A - **Table S.6.** Number (count) and average duration in seconds (with standard deviation) of vocalisations per hour split by age and affect.

APPENDIX A - Table S.7. Models to test effect of Affect, Age, and Affect * Age

interaction on infant arousal at the time of a vocalisation.

Model for Infant Arousal	at the time of	² a vocalisation	1		
Dependent/ Response verial	$a_1 = infort or$	a vocalisation	a of a vocali	action	
Dependent/ Response varial	ble = lint ar	ousai at the tim	le of a vocali	sation.	
Fixed effects (predictor vari	(ables) = Affective	ct, Age, and int	eraction betw	ween Affect*	Age.
Random effect for subject.			1	1	1
	BValue	Std.Error	DF	t-value	p-value
Affect	-0.068	0.034	36416	-2.016	0.044
Age_group	-0.153	0.052	73	-2.941	0.004
Affect:Age_group	0.009	0.024	36416	0.387	0.699
		1	1		1
Infant arousal as a function	on of Affect				
Dependent/ Response varial	ble = infant ar	ousal at the tim	e of a vocali	sation at 5M	(1) or at
10M (2).					
Fixed effects (predictor vari	ables) = Affec	ct (neutral/ nega	ative).		
Random effect for subject.	,				
	BValue	Std.Error	DF	t-value	p-value
1. 5M - Affect	-0.058	0.013	24873	-4.329	0.000
2. 10M - Affect	-0.049	0.020	11543	-2.476	0.013
		·			
Infant arousal as a function	on of Age.				
Dependent/ Response varial	ble = infant ar	ousal for neutra	al (1) or nega	ative (2) Affe	ct
vocalisations.					
Fixed effects (predictor vari	ables) = Age	(5M/ 10M).			
Random effect for subject.		. ,			
	BValue	Std.Error	DF	t-value	p-value
1. Neutral - Age_group	-0.132	0.051	73	-2.590	0.012
2. Negative - Age_group	-0.160	0.055	73	-2.886	0.005
		1	I		1

APPENDIX A - Table S.8. Models to test effect of Affect, Age, and Affect * Age

interaction on infant autonomic reactivity.

Model for Infant autonomic reactivity.										
Dependent/ Response variable = infant autonomic reactivity.										
Fixed effects (predictor variables) = Affect, Age, and interaction between Affect*Age.										
Random effect for subject.										
	BValue	Std.Error	DF	t-value	p-value					
Affect	0.047	0.027	36696	1.721	0.085					
Age_group	-0.079	0.046	73	-1.708	0.092					
Affect:Age_group	-0.050	0.019	36696	-2.595	0.010					
Infant autonomic reactiv	vity as a functi	ion of Affect								
Dependent/ Response vari	able = infant a	utonomic rea	ctivity at 5M	(1) or at 10M	(2).					
Fixed effects (predictor va	riables) = Affe	ect (neutral/ n	egative).							
Random effect for subject	•									
	BValue	Std.Error	DF	t-value	p-value					
1. 5M - Affect	-0.004	0.011	24987	-0.343	0.732					
2. 10M - Affect	-0.055	0.016	11709	-3.479	0.001					
Infant autonomic reactiv	vity as a functi	ion of Age.								
Dependent/ Response vari	able = infant a	utonomic rea	ctivity to neu	utral (1) or neg	ative (2)					
Affect vocalisations.										
Fixed effects (predictor va	riables) = Age	e (5M/ 10M).								
Random effect for subject	•									
	BValue	Std.Error	DF	t-value	p-value					
1. Neutral - Age_group	-0.128	0.043	73	-2.997	0.004					
2. Negative - Age_group	-0.090	0.052	73	-1.746	0.085					

Here we explored whether changes in infant autonomic reactivity were parallel to changes in activity levels. To explore this, we calculated the changes in activity in infants relative to infant vocalisations. Changes in activity were defined by taking the average activity levels during the 10 minutes before the vocalisation onset and subtracting it from the average activity levels during a 1-minute period right after the vocalisation (post – pre). We explored whether there were significant differences in activity around different types of vocalisations (i.e. changes in activity around negative vs. neutral vocalisations) as well as across the different ages (i.e. changes in activity around negative vocalisations at 5 months vs. at 10 months). There were no significant age differences in the way activity changed around negative and neutral vocalisations.

APPENDIX A - **Table S.9.** Models to test effect of Affect, Age, and Affect * Age interaction on infant change in activity levels.

Infant change in activity levels.									
Dependent/ Response variable = Infant change in activity levels relative to infant									
vocalisations at 5 and 10 months.									
Fixed effects (predictor va	ariables) = Af	ffect, Age, and i	nteraction be	tween Affect*.	Age.				
Random effect for subject	-	_			_				
	BValue Std.Error DF t-value p-valu								
Affect	0.001	0.021	37155	0.054	0.957				
Age_group	-0.033	0.029	73	-1.157	0.251				
Affect:Age_group	-0.018	0.015	37155	-1.222	0.222				

APPENDIX A - Table S.10. Models to test effect of Affect, Age, and Affect * Age

Model for Caregiver Arousal at the time of a vocalisation.								
Dependent/ Response variable = caregiver arousal at the time of a vocalisation.								
Fixed effects (predictor variables) = Affect, Age, and interaction between Affect*Age.								
Random effect for subject.								
	BValue	Std.Error	DF	t-value	p-value			
Affect	-0.032	0.032	35570	-1.003	0.316			
Age_group	-0.012	0.028	73	-0.434	0.666			
Affect:Age_group	-0.007	0.023	35570	0.307	0.759			

APPENDIX A - Table S.11. Models to test effect of Affect, Age, and Affect * Age

interaction on caregiver autonomic responsiveness.

Model for Caregiver autonomic responsiveness.									
Dependent/ Response variable = caregiver autonomic responsiveness.									
Fixed effects (predictor variables) = \overline{A} ffect. Age. and interaction between Affect*Age.									
Random effect for subject.									
	BValue	Std.Error	DF	t-value	p-value				
Affect	-0.019	0.027	36221	-0.722	0.470				
Age_group	-0.035	0.033	73	-1.072	0.287				
Affect:Age_group	-0.020	0.019	36221	-1.037	0.300				

Here we explored whether changes in caregiver autonomic responsiveness were parallel to changes in activity levels. To explore this, we calculated the changes in caregivers' activity relative to infant vocalisations. Changes in activity were defined by taking the average activity levels during the 10 minutes before the vocalisation onset and subtracting it from the average activity levels during a 1-minute period right after the vocalisation (post – pre). We explored whether there were significant differences in activity around different types of vocalisations (i.e. changes in activity around negative vs. neutral vocalisations) as well as across the different ages (i.e. changes in activity around negative vocalisations at 5 months vs. at 10 months). We did not find significant differences in the way caregiver activity changed relative to different type of vocalisations (negative vs. neutral) at either time point.

APPENDIX A - **Table S.12.** Models to test effect of Affect, Age, and Affect * Age interaction on caregiver change in activity levels.

Caregivers change in activity levels.									
Dependent/ Response variable = Caregiver change in activity levels relative to infant									
vocalisations at 5 and 1	10 months.								
Fixed effects (predicto	r variables) = A	Affect, Age, and	interaction b	etween Affect	*Age.				
Random effect for subj	Random effect for subject.								
	BValue	BValue Std.Error DF t-value p-value							
Affect	-0.009	0.021	37155	-0.413	0.680				
Age_group	-0.025	0.023	73	-1.105	0.273				
Affect:Age_group	-0.016	0.015	37155	-1.062	0.289				



APPENDIX A - Figure S.2. Comparison of proximity levels based on age.

Proximity levels around negative (A) and neutral (B) vocalisations at 5 months (lighter colours) and 10 months (darker colours). Units are in metres. Shaded areas show standard error. Straight red lines indicate significant clusters (p<0.025, two sided) identified using a cluster-based permutation test.

APPENDIX A - Table S.13. Models to test the speed (i.e. slope of recovery) at which the

infants recovered from negative affect vocalisations.

Speed of recovery as a function of Age. Dependent/ Response variable = slope of the recovery line at 5 and 10 months. Fixed effects (predictor variables) = Age. Random effect for subject.							
	BValue	Std.Error	DF	t-value	p-value		
Slope from min1 to min4 (5m) and min1 to min5 (10m)	0.075	0.051	22681	1.457	0.145		
Slope from min1 to min5 (5m) and min1 to min5 (10m)	0.089	0.054	22609	1.629	0.103		



APPENDIX A - Figure S.3. Association between caregiver's autonomic responsiveness and speed of infants' recovery.

Here we repeated Analysis 3.2 (Chapter 3, Figure 3.7D) using a recovery line from the 1^{st} to the 5th (instead of 4th) minute at 5 months to make sure our decision to use different times to calculate the recovery lines was not changing the results. The figure shows the results at 5 months, R_{5M} = -0.129, p_{5M} = 0.378.

MATCHING FINAL Ns

B)



APPENDIX A - **Figure S. 4**. Control analysis, retrieving random sample of 26 infants (matched gender) from the 5-month group.

Here we randomly selected a sample of 26 dyads at 5 months (matching the gender with the sample at 10 months) and repeated analyses 3.2 following the steps described in 2.3.7.2. (A) Average arousal across the 5-minute recovery window. Light red shows data at 5 months and dark red shows data at 10 months. Asterisks indicate significance (* BH adjusted p< 0.05, ** BH adjusted p<0.01, *** BH adjusted p<0.001). (B) Slopes of the recovery lines at 5 (left) and 10 months (right). No significant differences were found between age groups (t(50) = -1.2797, p= 0.2066). (C) Results showing the association between caregivers' autonomic responsiveness relative to the onset of a negative infant vocalisation and infants' recovery indexed as the slope of the recovery line at 5 months (left), and at 10 months (right). There was a significant relationship between caregiver heart rate change and infant recovery at 10 months (R= -0.5447, p= 0.0040) but not at 5 months (R= -0.103, p= 0.6165).



APPENDIX A - Figure S.5. Concurrent synchrony in infant-caregiver arousal using different window length and overlaps.

Results from the SWC analysis using a window length of 60s, 180s and 300s with an overlap of 30s, 90s and 150s respectively at 5 months (top row) and at 10 months (bottom row). Shaded areas show standard error. Straight lines are significant clusters identified using a cluster-based permutation test.



APPENDIX A - Figure S.6. Results from the WCC and cluster-based permutation analysis using different time window widths.

Windowed cross-correlation analysis using a window width of 60s (A and G), 180s (C and I) and 300s (E and K) at 5 and 10 months respectively. Negative lags indicate infant arousal forward-predicting adult arousal. Positive lags indicate caregivers' arousal forward-predicting infants' arousal. Results from the cluster-based permutation test using a window width of 60s (B and H), 180s (D and J) and 300s (F and L) at 5 and 10 months respectively. No significant clusters were identified at 10 months for any time window.

APPENDIX B - Supplementary Materials for: The neural and physiological substrates of real-world attention change across development.

- 1. SM1. Changes in the number and duration of attentional episodes throughout the solo play interaction
- Figure S1. Changes in the number and duration of attentional episodes throughout the solo play interaction
- 3. Figure S2. Calculation of theta power changes around an attention episode at 5 months after matching the final number of accepted trials to those at 10 months.
- 4. Table S1. Results from the 3-way ANOVA performed in Analysis 4 for theta PSD.
- 5. Table S2. Multiple comparison test between time within an attentional episode and age (in theta PSD activity).
- 6. Table S3. Multiple comparison test between channel cluster and age (in theta PSD activity).
- 7. Figure S3. Calculation of alpha power changes within an attention episode.
- 8. Table S4. Results from the 3-way ANOVA performed in Analysis 4 for alpha PSD.
- 9. SM 2. Analyses on EEG data quality
- 10. Table S5. Table showing the average percentage (and standard deviation) of channel interpolation, segments removed, and ICA components rejected at 5 and 10 months.
- 11. Figure S4. Bar plots showing the average of interpolated channels per cluster of channels of interest.
- 12. Table S6. Table showing the average percentage (and standard deviation) of interpolated channels for each cluster of channels of interest.
- 13. Figure S5. Photos of the toys.
- 14. SM 3. Analyses on the influence of the mothers on infants' behaviours.
- 15. Table S7. Table summarising the numbers of datasets included in each of the analyses for both samples as well as reason for exclusion.
- 16. Figure S6. Topoplot showing our channel clusters in the 32-channel (A, 10 months) and the 64-channel (B, 5 months) BioSemi gel-based ActiveTwo montage.

SM 1. Changes in the number and duration of attentional episodes throughout the solo play interaction

These analyses are aimed to explore whether the average number of attentional episodes and duration changed throughout the play session and are complementary to analysis 1 (Figure 4.2B and D). To do this, we calculated the best fit lines for number (Figure S1A and B) and duration (Figure S1C and D) of attentional episodes and compared their gradients between ages (to check for developmental differences) and against zero (to see whether there were increases or decreases in these variables throughout the interaction at both ages).

Average number (count) of attentional reorientations per minute (Figure 4.2A and B)

- 5-months-old infants, mean gradient = -2.33179.
- 10-months-old infants, mean gradient = -1.20763.

We observed a significant decrease in attentional reorientations over the course of the interaction at both time points (at 5 months: t(30)=-3.483, p=0.0015; at 10 months: t(28)=-3.331, p=0.0024). We found no significant differences between the two groups. Average duration (sec) of attentional episodes per minute (Figure 4.2C and D)

- 5-months-old infants, mean gradient = 0.2528.
- 10-months-old infants, mean gradient = 0.689 (0.1043 without outlier in Figure S1D).

No significant differences were found in the way attentional episodes durations changed throughout the interaction, neither within nor between groups. Results stayed the same after removing outlier in Figure S1D.





Figure showing the best fit lines for individual number of attentional episodes per minute at 5 months (A) and at 10 months (B); and best fit lines for average duration of attentional episodes at 5 months (C) and at 10 months (D).



APPENDIX B - Figure S.2. Calculation of theta power changes around an attention episode at 5 months after matching the final number of accepted trials to those at 10 months.

Results of the linear mixed effects models conducted to examine whether individual looks accompanied by higher theta power are longer lasting. For each look, we calculated the association between the total duration of the look and relative theta power during five time-windows (-2000msec to -1000msec and -1000msec to 0 prior to the look, and 0 to 1000msec, 1000 to 2000msec and 2000 to 3000msec before the look), using a series of separate linear mixed effects models. Y-axis is the t value. Asterisks (*) indicate p values < .05. Central channels include: 'FC1', 'FC2', 'C3', 'C2', 'C4', 'CP1' and 'CP2'.

APPENDIX B - **Table S.1.** Results from the 3-way ANOVA performed in Analysis 4 for theta PSD.

Analysis of Variance								
Source	Sum Sq.	d.f.	Mean Sq.	F	Prob>F			
attentional phase	0.174	2	0.08719	4.56	0.0105			
channel cluster age	0.045 0.022	2	0.02255 0.02245	1.18	0.30// 0.2788			
attentional phase*channel cluster	0.019 0.213	4	0.00478 0.10668	0.25 5.58	0.9098 0.0038			
channel cluster*age	0.458	2	0.22918	11.98	0			
attentional phase*channel cluster*age Error	0.026 136.545	4 7137	0.00644 0.01913	0.34	0.8535			
Total	137.741	7154						

Constrained (Type III) sums of squares.

"Attentional phase refers" to time within an attentional episode (i.e., start, middle, termination); "Channel cluster" includes Central, Frontal and Occipital channels; and "age" includes 5- and 10-months-old infants.

time within an attentional episode (AE)	age	Lower Limit	A-B	Upper Limit	P-value
AE=1,age=1	AE=2,age=1	-0.021	-0.002	0.017	1.000
AE=1,age=1	AE=3,age=1	-0.004	0.009	0.022	0.365
AE=1,age=1	AE=1,age=2	0.006	0.019	0.033	0.001
AE=1,age=1	AE=2,age=2	-0.026	-0.007	0.011	0.860
AE=1,age=1	AE=3,age=2	-0.007	0.007	0.022	0.719
AE=2,age=1	AE=3,age=1	-0.009	0.011	0.031	0.643
AE=2,age=1	AE=1,age=2	0.001	0.021	0.042	0.039
AE=2,age=1	AE=2,age=2	-0.029	-0.006	0.018	0.986
AE=2,age=1	AE=3,age=2	-0.012	0.009	0.030	0.826
AE=3,age=1	AE=1,age=2	-0.005	0.010	0.026	0.382
AE=3,age=1	AE=2,age=2	-0.036	-0.016	0.003	0.150
AE=3,age=1	AE=3,age=2	-0.018	-0.002	0.014	1.000
AE=1,age=2	AE=2,age=2	-0.047	-0.027	-0.007	0.002
AE=1,age=2	AE=3,age=2	-0.029	-0.012	0.005	0.305
AE=2,age=2	AE=3,age=2	-0.006	0.015	0.035	0.318

APPENDIX B - **Table S.2.** Multiple comparison test between time within an attentional episode and age (in theta PSD activity).

AE indicates "time within an attentional episode" where AE1= first second into the look (start), AE2 = third to fourth second into the look (middle), and AE3 = last second into the look (termination). Age1 = 5 months, age2 = 10 months.

channel cluster (ch)	age	Lower Limit	A-B	Upper Limit	P-value
Ch=1,age=1	Ch=2,age=1	-0.028	-0.010	0.008	0.571
Ch=1,age=1	Ch=3,age=1	-0.004	0.014	0.031	0.252
Ch=1,age=1	Ch=1,age=2	-0.005	0.014	0.032	0.287
Ch=1,age=1	Ch=2,age=2	-0.009	0.010	0.028	0.651
Ch=1,age=1	Ch=3,age=2	-0.026	-0.008	0.010	0.820
Ch=2,age=1	Ch=3,age=1	0.006	0.024	0.042	0.002
Ch=2,age=1	Ch=1,age=2	0.005	0.024	0.042	0.003
Ch=2,age=1	Ch=2,age=2	0.002	0.020	0.039	0.023
Ch=2,age=1	Ch=3,age=2	-0.016	0.002	0.021	0.999
Ch=3,age=1	Ch=1,age=2	-0.018	0.000	0.018	1.000
Ch=3,age=1	Ch=2,age=2	-0.022	-0.004	0.015	0.993
Ch=3,age=1	Ch=3,age=2	-0.040	-0.022	-0.003	0.011
Ch=1,age=2	Ch=2,age=2	-0.023	-0.004	0.015	0.993
Ch=1,age=2	Ch=3,age=2	-0.041	-0.022	-0.003	0.016
Ch=2,age=2	Ch=3,age=2	-0.037	-0.018	0.001	0.082

APPENDIX B - **Table S.3.** Multiple comparison test between channel cluster and age (in theta PSD activity).

Ch1 = central channels, Ch2 = occipital channels, and Ch3 = frontal channels. Age1 = 5 months and Age2 = 10 months.



APPENDIX B - Figure S.3. Calculation of alpha power changes within an attention episode.

Bar plots for the average relative alpha power throughout a look, at both time points (5 and 10 months) and at different groups of electrodes (central, occipital, and frontal). Error bars represent SEMs.

APPENDIX B - **Table S.4.** Results from the 3-way ANOVA performed in Analysis 4 for alpha PSD.

Analysis of Variance							
Source	Sum Sq.	d.f.	Mean Sq.	F	Prob>F		
attentional phase	0.0027	2	0.00136	0.39	0.6757		
channel cluster	0.0597	2	0.02983	8.58	0.0002		
age	0.6072	1	0.60725	174.6	0		
attentional phase∗channel cluster	0.0002	4	0.00006	0.02	0.9994		
attentional phase∗age	0.0117	2	0.00585	1.68	0.186		
channel cluster∗age	0.0051	2	0.00256	0.74	0.4787		
attentional phase*channel cluster*age	0.0025	4	0.00062	0.18	0.949		
Error	24.8216	7137	0.00348				
Total	25.5905	7154					

Constrained (Type III) sums of squares.

"Attentional phase refers" to time within an attentional episode (i.e., start, middle, termination); "Channel cluster" includes Central, Frontal and Occipital channels; and "age" includes 5- and 10-months-old infants.

SM 2. Analyses on EEG data quality

To compare the quality of the EEG data at 5 and 10 months we performed a series of analyses on percentage of channels interpolated, percentage of segments removed (i.e. zeroed out) and percentage of ICA components rejected. None of these variables differed significantly between age groups.

APPENDIX B - **Table S.5.** Table showing the average percentage (and standard deviation) of channel interpolation, segments removed, and ICA components rejected at 5 and 10 months.

	Channels interpolated (%)		Segments r (%)	removed	ICA components rejected (%)	
	Mean	Std	Mean	Std	Mean	Std
5 months	14.130	8.782	8.066	9.617	51.630	15.479
10 months	9.783	10.292	8.360	6.216	48.153	16.243
Statistics	t(44)= 1.541,		t(44) = -0.124,		t(43) = -0.735,	
	p=0.1	304 *	p = 0.902 *		p = 0.466 [†]	

There were two (*) or three $(^{\dagger})$ datasets for which this data was missing at 10 months.

We also calculated the percentage of channel interpolation within our clusters of channels of interest (i.e. frontal, central, occipital vs. other channels). The percentage of interpolated channels within our cluster of channels of interest did not statistically differ between our two groups. Instead, the percentage of other channels removed (i.e. channels not included in any of our analyses) did differ significantly.



APPENDIX B - Figure S.4. Bar plots showing the average of interpolated channels per cluster of channels of interest.

Red lines indicate the equivalent percentage of 1.5 channel interpolated. Significance is indicated with asterisks where * = p < 0.05.

	P							
	Frontal		Central		Occipital		Others	
	Mean	Std	Mean	Std	Mean	Std	Mean	Std
5 months	2.378	2.575	1.902	2.400	0.340	0.659	9.511	6.458
10 months	1.630	2.960	1.359	2.799	1.223	2.621	5.571	6.792

t(44) = -1.567,

p=0.124

t(44)=2.0163,

p= 0.050

t(44)=0.710,

p=0.483

Stats

(t-test)

t(44)=0.914,

p=0.366

APPENDIX B - **Table S.6.** Table showing the average percentage (and standard deviation) of interpolated channels for each cluster of channels of interest.



APPENDIX B - Figure S.5. Photos of the toys.

Toys employed during the solo play at 5 months (A to C) and at 10 months (D to F).

SM 3. Analyses on the influence of the mothers on infants' behaviours.

To examine the possibility that mothers might have influenced infants' behaviours, we performed a set of analyses in which we coded maternal looking behaviour in a subsample of our participants across four different interactions at both time points. Mean average duration was 3.475 minutes (std= 0.624) at 5 months and 3.704 minutes (std= 0.803) at 10 months. There were no significant differences between groups in the duration of maternal interactions that were coded (t(6)=-0.448, p=0.669).

First, we calculated the percentage of time mothers and infants spent looking at the same object at the same time. At 5 months, the percentage of time spent looking at the same object was 4.455% (std= 4.378) and, at 10 months, it was 10.902% (std= 8.936). There were no significant differences between age groups (t(6)= -1.296, p= 0.246). Second, we coded Cohen's Kappa to index the agreement in looking behaviour between our infant-mother pairs. At 5 months, Cohen's Kappa was 0.003 (std= 0.025) and 0.047 (std= 0.026) at 10 months. Suggesting that, at both time points, there was virtually none to slight alignment between the looking behaviour time series of infants and their mothers. Finally, we calculated the correlation between infant and mother looking behaviours. Again, there were no significant differences between the two time points (R_{5M} = -0.0313, R_{10M} = 0.1001; t(6)= -1.159, p= 0.29).

These results preclude the potential for maternal influence on infants' behaviour, and they also demonstrate that the impact of the mothers on the infants' behaviour did not differ between age groups. This is as expected because, as we describe in the methods section, mothers were instructed "not to interact with the infant" during the play sessions and were given explicit instructions not to label the toys they were playing with, to refrain from addressing their infants directly, and to avoid making direct eye contact with them.

APPENDIX B - **Table S.7.** Table summarising the numbers of datasets included in each of the analyses for both samples as well as reason for exclusion.

	at 5 months	%	at 10 months	%
total SP sessions	60	100	81	100
EEG datasets	60	100	81	100
bad EEG data *	33	55.0	21	18.5
technical issues with data recording (e.g., video missing)	1	1.7	8	8.6
further rejection - sync problems	-	-	19	12.3
further rejection - behaviour not coded	-	-	26	27.2
further rejection - too few looks	3	5.0	3	2.5
Final EEG datasets	23	38.3	25	30.9
ECG [†]	31	51.7	29	35.8
noisy/ bad data	2	3.3	2	6.9
sync problems	1	1.7	3	10.3
file corrupted/ recording error/ missing data	-	-	10	34.5
Final ECG datasets	28	46.7	14	17.3
Final datasets (N)				
Looking behaviour coded	31		29	
Behaviour + EEG	23		25	
Behaviour + ECG	28		14	
ECG + EEG	22		10	

* Bad EEG data also includes these datasets where the infant was fussing out, pulling out the EEG wires, falling asleep, etc.

[†] ECG data is explained based on the datasets that have gaze behaviour coded


APPENDIX B - Figure S.6. Topoplot showing our channel clusters in the 32-channel and the 64-channel BioSemi gel-based ActiveTwo montage.

Topoplot showing our channel clusters in the 32-channel (A, 10 months) and the 64channel (B, 5 months) BioSemi gel-based ActiveTwo montage. In blue, Frontal channels ('Fp1', 'Fp2', 'AF3', 'AF4', 'Fz'); in orange, Central channels ('FC1', 'FC2', 'C3', 'Cz', 'C4', 'CP1', 'CP2'); and in green, Occipital channels ('PO3', PO4', O1', 'Oz', 'O1). Channel names are organised from top to bottom and from left to right. APPENDIX C - Supplementary materials for: Who leads and who follows? The pathways to joint attention during free-flowing interactions change over developmental time.

- 1. Table S1. Demographic data at 5 and 15 months.
- 2. Table S2. Table summarising the numbers of datasets included in the analyses for both samples as well as reason for exclusion.
- 3. Figure S1. Experimental setup.
- 4. Figure S2. Toys employed at both time points.
- 5. Figure S3. Descriptive analyses using not interpolated data.
- 6. Figure S4. Histograms with look durations at 5M and 15M.
- 7. Figure S5. Average number of looks per minute.
- Figure S6. Analysis comparing the probability to look at the partner before leading/ following a look to JA at 5M vs. 15M.
- 9. Table S3 and Table S4. Results and specific models employed for the LME analyses on probability of mothers to follow infant leader looks.
- 10. Figure S7. Probability of mothers to follow an infant leader look using different time windows.

	5 months	15 months
Maternal education	%	%
Post-graduate degree	53.33	47.06
Degree	40	35.29
F.E. Qualification	-	-
A Level	6.67	11.76
GCSE's	-	5.88
No formal qualification	-	-
Other	-	-
Prefer not to answer	-	-
Household income	%	%
Under 16k	-	-
16k - 25k	-	-
26k - 35k	-	-
36k - 50k	6.67	5.88
51k - 80k	20	17.65
More than 80k	60	58.82
Prefer not to answer	13.33	17.65
Maternal ethnicity	%	%
White British	60	64.71
Other White	13.33	5.88
Asian, Indian	6.67	11.76
Black	13.33	5.88
Mixed – White/Afro-	-	-
Caribbean		
Not answered	6.67	11.76

APPENDIX C - Table S. 1. Demographic data at 5 and 15 months.

APPENDIX C - **Table S.2.** Table summarising the numbers of datasets included in the analyses for both samples as well as reason for exclusion.

Final datasets	5 months	15 months
Gaze coded (N)	47	48
Infants	24	24
Mothers	23	24
Dyads incomplete *	1	2
Dyads completed	23	23

* The data for one of the members of the dyad (infant or mother) could not be recovered (i.e. datasets were either corrupted or missing).

Experimental set-up



APPENDIX C - **Figure S.1.** Experimental set up and an example of looking behaviour. Top figure shows the experimental set up for the joint play condition. Two cameras pointed at the infant (view in photos 1 and 2) and one camera pointing at the mother (view in photo number 3). Looking behaviour was coded manually for object and partner looks from both the mother and the infant. Photos in the middle row show the different categories of interest. Bottom figure shows the looks of interest and how are these defined. JA = Joint attention, MG = Mutual gaze.



APPENDIX C - Figure S.2. Photos of the toys employed at both time points.(A) panda, (B) a book, and (C) rattle.



APPENDIX C - Figure S.3. Descriptive analyses using not-interpolated data. Figure showing average number of looks (top) and duration (bottom) per minute. Data is not interpolated. Asterisks indicate significance (* = p>0.05, ** = p<0.01, *** = p<0.001).



APPENDIX C - Figure S.4. Histograms showing the distribution of look durations during the whole session.

A-B: show the distributions of look durations at 5 months, for infants (A) and mothers (B). C-D: show the distributions of look durations at 15 months, for infants (C) and mothers (D).



Average num	ber of lool	ks per min	ute			
	Leader looks to JA		Follower looks		Leader looks to non-JA	
	Mean	(std)	Mean	(std)	Mean	(std)
Infants 5M	2.52	1.08	1.32	0.83	4.88	2.53
Infants 15M	2.32	1.09	1.79	1.28	2.68	1.47
Mothers 5M	1.47	0.72	3.31	1.40	2.51	1.12
Mothers 15M	1.93	0.88	3.12	1.37	2.81	1.01

APPENDIX C - Figure S.5. Average number of looks per minute.

Average number of looks per minute of leader looks to non-JA (in red), leader looks to

JA (in green) and follower looks to JA (in blue). Infant (left) and mother (right).



APPENDIX C - **Figure S.6.** Probability of infants to look at the partner before a look to JA.

Analysis comparing the probability to look at the partner before leading/ following a look to JA at 5M vs. 15M. Probability of infants to look at the partner around a leader (A) or a follower (B) look to JA at 5 months (in light green) and 15 months (in light orange). Red thick lines indicate significance from the CBP test.

APPENDIX C - **Table S.3.** Model for probability of mothers following an infant leader look as a function of a leader look following a look to partner or not.

Dependent/ Response variable = probability of following an infant leader look at 5M (Model 1) or 15M (Model 2) Fixed effects (predictor variable) = Leader look that follows a partner look vs leader look that did not Random effect for subject Model 1 - 5 months Time window ß value Std.Error DF * t-value p-value 2 min -0.03 0.02 40 -1.37 0.18 5 min -0.03 40 0.02 -1.58 0.12 10 min -0.02 0.02 40 -1.43 0.16 Model 2 - 15 months Time window ß value Std.Error DF t-value p-value 2 min -0.10 0.02 44 -3.86 0.00 5 min -0.09 0.02 44 -4.71 0.00 10 min -0.08 0.01 44 -6.25 0.00

* Subjects with only one look were excluded from the analyses

APPENDIX C - **Table S.4.** Model for probability of mothers following an infant leader look as a function of age.

Dependent/ Response variable = probability of following a leader look that is preceded by a partner look

Fixed effects (predictor variable) = Age group

Random effect for subject

Time window post look = 2sec

l				
ß value	Std.Error	DF	t-value	p-value
0.14	0.03	40	4.17	0.00
Time window post lo	ook = 5sec			1
ß value	Std.Error	DF	t-value	p-value
0.13	0.03	40	4.81	0.00
Time window post lo	ook = 10sec			1
ß value	Std.Error	DF	t-value	p-value
0.12	0.03	40	4.64	0.00



APPENDIX C - **Figure S.7.** Probability of mothers to follow an infant leader look using different time windows.

Probability of mothers to follow an infant leader look that was preceded by a partner look (dark colours) versus a look that was not preceded by a partner look (light colours) at 5 months (green) and 15 months (orange). A) Figure showing the comparison between infant leader looks that were preceded by a partner looks and infant leader looks that were not. B) Comparison between probability of mothers to follow an infant leader look that was preceded by a partner look at 5 months.

APPENDIX D – Ethics

Ethics approval for the project was submitted and obtained by James White on behalf of the PhD students working on the project; Myself and James White. Below are the ethical approval letter and the approved ethics application.

Ethical approval letter



Pioneering Futures Since 1898

Dear James

Application ID: ETH2021-0076

Project title: Doctoral Research Project

Lead researcher: Mr James White

Your application to University Research Ethics Sub-Committee was considered on the 24th of February 2021.

The decision is: Approved

The Committee's response is based on the protocol described in the application form and supporting documentation.

Your project has received ethical approval for 4 years from the approval date.

If you have any questions regarding this application please contact your supervisor or the secretary for the University Research Ethics Sub-Committee.

Approval has been given for the submitted application only and the research must be conducted accordingly.

Should you wish to make any changes in connection with this research project you must complete <u>'An application for</u> approval of an amendment to an existing application'.

The approval of the proposed research applies to the following research site.

Research site: University of East London Stratford Campus and in home visits to UK residents.

Principal Investigator / Local Collaborator: Mr James White

Approval is given on the understanding that the <u>UEL Code of Practice for Research and the Code of Practice for</u> <u>Research Ethics</u> is adhered to.

Any adverse events or reactions that occur in connection with this research project should be reported using the University's form for <u>Reporting an Adverse/Serious Adverse Event/Reaction</u>.

The University will periodically audit a random sample of approved applications for ethical approval, to ensure that the research projects are conducted in compliance with the consent given by the Research Ethics Committee and to the highest standards of rigour and integrity.

Please note, it is your responsibility to retain this letter for your records.

With the Committee's best wishes for the success of the project

Yours sincerely

Fernanda Silva

Administrative Officer for Research Governance

Docklands Campus University Way London E16 2RD **Stratford Campus** Water Lane London E15 4LZ **University Square Stratford** Salway Road London E15 1NF +44 (0)20 8223 3000 srm@uel.ac.uk uel.ac.uk



Ethics application

Ethics ETH2021-0076: Mr James White (High risk)

Date	06 Jan 2021
Researcher	Mr James White
Student ID	2067263
Project	Mechanisms, capabilities and applications of infant neural and
	physiological oscillatory entrainment
School	Psychology

Ethics application

Checklist for research projects conducted during a pandemic

Project details

1.1 Is your research project taking place during a pandemic? Yes

1.2 Where will the research take place?

1. Lab visits - University of East London - Baby Development Lab - Stratford Campus - Arthur Edwards Building

2. Home visits - In participants' homes

1.3 How will the research methodology be amended in response to the pandemic, e.g. change from face-to-face to remote working? How will you address any changing circumstances that may affect the research project and what measures will be put in place?

Ensuring that our study is COVID secure is a top priority and has been a concern that has been well thought through with multiple risk assessments and group discussions.

General:

Our research will be conducted online or remotely as much as possible, including: communication between researchers, coding of collected data, data analysis, write up of experimental results and any other work that could be completed online. Where possible, any questionnaires for participants will be completed online to minimise time spent in the lab and contact with additional resources. While completing online and remote working, researchers will have no physical contact with each other.

Lab visits:

A specific COVID risk assessment for the lab visits has been attached to this ethics application and details 27 points where changes have been made, reasons for these changes and the risks involved have been scored.

Home visits:

A specific COVID risk assessment for the home visits has been attached to this ethics application, detailing the procedure that we wish to use, the 24 points where changes have been made to ensure that risks have been mitigated and a score for risks involved.

1.4. Is there a plan in place as to how the research project will be conducted in the event of a significant change to the timescale of the research project, changes to the research team, limited access to resources or laboratory space, or restrictions on travel or entering premises?

Yes

1.5. What are the current guidelines/requirements for the location of the research, locally, nationally or/and internationally?

Current Government guidance specifies that labs and research facilities are able to remain open, providing they are COVID secure (Guidance of 6 January 2021). The BabyDev Lab has been cleared by the UEL Gold Committee to continue testing, providing stringent COVID measures are followed; and currently a full research programme is underway.

The UK government has published and been updating this guide for working safely during the coronavirus. https://www.gov.uk/guidance/working-safely-during-coronavirus-covid-19/labs-and-research-facilities This includes guides on lockdown and tier measures, how often to clean and other practices.

International health and safety requirements are not necessary in this scenario as all researchers, participants and research proceedings will be UK based.

1.6. Does your research project comply with all legal and ethical requirements and other applicable guidelines, including those from other organisations and/or countries? Yes

1.6.1 If yes, please provide details.

As per the government guidance all research facilities can remain open following a COVID-19 risk assessment, and when following COVID safe rules when testing participants. The Baby Development Lab has completed and had a COVID-19 risk assessment approved by a gold level committee. Participants are required to follow the COVID safe rules during testing.

No other organisations will be involved in this project.

Research Team

2.1. Who will be responsible for ensuring that the research project adheres to current UK Government guidelines?

2.2. Has consideration been made for supervision and management of the research team members considering remote communication and working remotely/offsite?

If yes, please provide details.

2.3. Are systems in place to safely share research data with members of the research team that are working remotely?

Participants

3.1. Are mechanisms in place to ensure that the informed consent process is clear allowing an appropriate forum for questions and right to withdraw, despite resource limitations and in the context of online or remote communication?

3.1.1 If yes, please provide details.

3.2. How will you manage any necessary changes to the participant consenting process should you be unable to recruit participants using your preferred method of data collection?

3.3. Is the research proposal mindful of factors which may impact negatively on equality of participation?

If yes, please provide details.

Risk Assessment

4.1. Have the potential risks to the research or the health, safety and well-being of researchers, research participants and others been fully considered?

4.1.1 If yes, please provide details.

4.2. How will you conduct the research ensuring that you adhere to Government regulations and taking in to account any changes to local and national requirements, e.g. compliance with any measures that adhere to current infection control guidelines, protective equipment, social distancing?

4.3. Have health and safety requirements been evaluated for specific research environments inherent to the project design e.g. laboratory work, access to appropriate equipment, interviews at external organisations or schools?

4.4. If exposure to the pandemic during the research compromises health and safety, how will you ensure that local, national and where required, international, health and safety requirements are met?

4.5. How will you monitor any changes in national/international Government or equivalent, rules or restrictions and any other health and safety guidance which may affect the research project?

4.6. Do you have a process in place to maintain compliance with legal and ethical requirements in emergent circumstances, including those from other countries, and staying current as they may change, including consideration of an agreed upon standard if national/regional standards differ?

4.6.1 If yes, please provide details.

Data collection and storage

5.1. Have you included sufficient lead time to complete data collection should your research project be suspended or significantly delayed due to unforeseen circumstances?

Collaborators

6.1. Have agreements been reached or formalised relating to collaborative working, particularly where the collaborations cross institutional, national, disciplinary or sector boundaries?

If yes, please provide details.

6.2. Is there a plan in place for ensuring that all contributors are given sufficient opportunity to participate despite limited resources or difficulties with working during emergent circumstances?

If yes, please provide details.

Contracts

7.1. Has consideration been made for possible modifications to contracts and financial agreements relating to the research, including possible delays in meeting deliverables or changes in research design?

If yes, please provide details.

Intellectual Property and Authorship

8.1. Have agreements related to intellectual property, publication and authorship, or collaboration, including the roles and responsibilities of research team members, been agreed and formalised? Have any of the agreements been affected by the pandemic?

If yes, please provide details.

Conflict of Interest

9.1. Have any conflicts of interest relating to the research been identified, declared and addressed in the context of the pandemic, including consideration of pressure to accelerate research projects?

If yes, please provide details.

Monitoring and Audit

10.1. Have monitoring and audit requirements been considered in the context of a pandemic, e.g. secure record keeping, data exchange?

If yes, please provide details

10.2. Are systems in place to safely share data with those conducting monitoring and auditing?

Dissemination

11.1. Will the research and its findings be reported accurately, honestly and within a reasonable time frame? Has consideration been given to the best ways to communicate research findings considering any restrictions on certain methods of dissemination due to infection control measures?

1. Project details

1.1. Title of proposed research or consultancy project

Oscillatory neural and autonomic correlates of social attunedness during early life: new mechanistic insights into how we learn to learn from one another

1.2. UEL Researchers or Consultancy lead'

Mr James White

Miss Marta Perapoch Amado

Ms Louise Goupil

Prof Samuel Wass

1.3. Start date of project for which ethical approval is being sought 01 Mar 2021

1.4. Anticipated end date of project for which ethical approval is being sought 31 Aug 2025

1.5. If this project is part of wider research or consultancy work, please provide the UREC, EISC, URES, RRDE, SREC, CREB or NHS research ethics approval number

1.6. If this project is part of a wider research study or consultancy work please state the start and end dates

01/09/2020 - 31/08/2025

1.7. Specify where the research or consultancy project will take place

University of East London Stratford Campus and in home visits to UK residents.

2. Aims and methodology

2.1. Aims and objectives of the project

We are a social species. Most infants, and young children, spend the majority of their early waking lives in the company of others. But, for practical reasons, almost everything that we know about how the brain subserves early attention and learning comes from studies that examined brain function in

one individual at a time. This means that we understand lots about how children attend and learn from information presented while they are alone, viewing a computer screen - but little about how attention is shared between people during social interaction. ONACSA will develop new techniques to look, for the first time, at how two brains dynamically interact with one another during early learning exchanges. The project will determine how children's active, participatory bids during learning lead to reactive changes in both members of the dyad – and how these changes, in turn, influence both partners' subsequent attention, and learning. It will also determine how, and why, some infants, and some parents, show greater sensitivity during social exchanges than others. And, using targeted interventions, it will investigate whether social sensitivity can be improved. The question of how two brains dynamically influence one another during learning exchanges has been described as the 'dark matter' of social neuroscience. Yet nobody has looked at these questions before from the perspective of early learning. Our results may help us to move beyond viewing children primarily as passive recipients of information during learning exchanges, to a perspective that better appreciates children's role as active participants in learning. Our findings may also have practical implications for educationalists, and clinicians.

2.2. Methodology, data analysis and recruitment for the project

We aim to collect neuroimaging and physiological data from 150 infants and their parents. The infants will be ages 2-36 months and will have multiple lab and home sessions as they develop at with one of each session at 2, 6, 12, 18 and 36 months. This data will be collected from participants while they engage in screen-based tasks, tabletop free play sessions or naturalistic at home data. I have attached lab and at home COVID risk assessments in the attachments section to demonstrate how all sessions will be kept COVID safe.

Questionnaires and interview

Once enrolled in the study, participants will be asked to fill in the family background information questionnaire. At each of the testing sessions participants will be asked to complete the following questionnaires in each of the visits: Infant Behaviour Questionnaire, Communicative Development Index, GAD-7, PHQ-9, PSWQ, CHAOS, perceived stress scale. These are all attached in the attachments section

A subset of participants (~30) will be asked to opt-in to the Adult Attachment Interview, a one-hourlong, semi-structured interview. It will be used with a sub-sample of 30 adult participants at the 2month check. The interview will be conducted once, by one researcher and the adult (parent) participant; either during the lab visit or at home, while the baby is sleeping or otherwise quiet/not needing much adult interactions (so for instance, while feeding).

Lab Sessions

Parent will be seated in a chair across the table from the infant who will be seated in an age appropriate highchair. Both participants will be required to wear specialised head caps which contain 64 electrodes or sensors to record brain activity from the scalp. Additional electrodes will be attached to the participants' face to collect muscle movement data and on the right clavicle and left rib area to record physiological activity, these electrodes will be stuck onto the skin using a small 13 x 8 mm paper washers. Additionally, in accordance with the manufacturing guidelines a small amount of nontoxic water-saline-based gel will be applied to each sensor in order to increase signal quality. Each participant's sensors will then be plugged into individual A/D amplifiers. These amplifiers are electrically isolated units meaning that they only record electrical activity. Both amplifiers will then be

connected via a fibre optic cable, and the summed data will be transmitted into an acquisition computer. All cables will be out of reach of infants and where possible held within plastic cable housing. The length of recording time will vary with different ages and will be maintained at an age appropriate level with regular breaks if needed. Participants will also be video recorded, as to allow for additional behavioural analysis post hoc. All video data recorded will be stored on a UEL computer which is not connected to the internet. Additionally, each member of the research team will have an additional copy of the data on an encrypted hard drive. All lab session techniques have been used before in previous studies in labs globally without problems.

During initial preparation for the session, infants and parents will be invited into the BabyDevLab, set up in a relaxed format with comfortable chairs and toys. All EEG and physiological apparatus will be applied (as above) to both participants in this room before moving to the testing room. If the participants become distressed, soothing techniques will be encouraged from the parent. If either of the participants becomes too distressed the session will be ended.

During the screen-based task, participants will be asked to watch a screen-based display lasting less than 10 minutes, which is designed to show visual and auditory stimuli that cause neural entrainment. Stimuli include a continuously playing cartoon and audio stimuli with changing visual patterns surrounding the cartoon display and a chirping sound as part of the auditory sound. Participants will be seated in an age appropriate chair, and younger/fussy infants will be asked to sit on their parent's lap in front of the screen. No other objects will be within reach of the participants. Participant response will be passively measured using EEG caps. A researcher will be in the room at all times to monitor the participants for signs of distress.

During the screen-based interaction task, participants will be asked to interact with each other via a video link. Participants will be placed in separate rooms so that live audio and visual communication cannot occur without the video link. Infants will be placed in an age appropriate chair positioned in front of a table, on the table may be toys or other age appropriate objects that the infant can play with as well as a screen displaying the parent. The parent will be placed in a chair in front of a table, on the table may be a set of toys or other infant age appropriate objects that the adult can play with as well as a screen displaying the infant. The video screen will be used to manipulate the course of the social interaction.

During the tabletop, free play task, participants will be placed on opposite sides of a table in age appropriate chairs. Between the participants may be age appropriate toys or other objects that the pair can play with.

Home Sessions

These sessions take place in the family's home. It involves parent and baby wearing a small wearable device during a day and a night session. This device has been designed to record continuous data for ~10 hours in a variety of parameters such as: heart rate, visual and auditory data, proximity between paired devices and location. All home session techniques have been used before in previous studies without problems.

For the set-up, a researcher will travel to the participants' homes at an agreed time and day and fit both participants with our devices. After that, he/she will leave the home. At the end of the session (that is, evening for day-time sessions or the following morning for night-time sessions) the researcher will travel back to their homes to collect the device.

The monitor will be worn for periods of the day/night but never during washing. The device is not waterproof, so we ask participants not to go swimming or have wet play whilst wearing it. Other than that, participants can nap/sleep, have her nappy changed, feed and generally do anything they like. During the periods when the monitor is being worn, the parent will be asked to fill in a logbook to record the infant's sleep patterns.

The devices will all be encrypted and the raw data will only be accessible to the research team. All adults engaging with infants will have acquired full DBS safety checks prior to any contact with infant.

2.3. Is the data accessed, collected or generated of a sensitive nature?

Yes

2.3.1. If yes, please provide details. Please ensure that all data of a sensitive nature is handled carefully and stored appropriately.

Video and audio recording data - will be personally identifiable, including information of vulnerable (infant) participants.

All video data will be stored on UEL's OneDrive for Business. Only one file will be kept in which participant numbers are linked to personally identifiably information (names and addresses). This will be stored on a separate computer and kept in a locked office, on a password-protected computer, in an encrypted file. Data will be backed-up onto OneDrive for Business.

Immediately after recording, all video data will be transferred to dedicated, encrypted hard disks that have been purchased specially for this purpose. Recordings will be securely deleted from the device that was used to make the recording. All data collected will only be accessible to members of the research team. It will be stored only on encrypted hard disks, in password-protected format. Between recording sessions, recording devices will be stored in a locked secure location.

Questionnaire and interview data - will include questions of a sensitive nature related to demographics, socio-economic status and mental health.

All participants will be assigned unique ID numbers. Participants' ID number will be used at all times when managing the research data. All questionnaire data will only be accessible to the research team and will be kept on password protected, encrypted computers and backed up on password protected, encrypted hard drive.

3. About your project

3.1. Is the research/consultancy project funded? Yes

3.2. Does the project involve external collaborators? No

3.3. Does the project involve human participants? Yes

3.4. Does the project involve non-human animals? No 3.4.1. If yes, where is the research project taking place?

3.5. Does your project involve access to, or use of, material (including internet use) covered by the Terrorism Act (2006) and / or Counter-Terrorism and Border Security Act (2019) or which could be classified as security sensitive? No

3.6. Does the project involve secondary research, secondary data or analysing an existing data set?

No

3.7. Does the project raise ethical issues that may impact on the natural environment over and above that of normal daily activity? No

3.8 Does the research/consultancy project involve data collected online via social media, advertising the project online or via social media or include a questionnaire/survey? No

If yes, please provide details.

3.9. Will the research/consultancy project take place overseas? No

3.10. Will the researcher or research team be responsible for the security of all data collected in connection with the research/consultancy project? Yes

3.11. Does your research/consultancy project require third-party permission? No

If yes, please provide details.

3.12. Does your research/consultancy project involve any circumstances where the professional judgement of you and/or the team is likely to be influenced by personal, institutional, financial or commercial interests?

If yes, please provide details.

3.13. Does the project involve consultancy or contract research?

If yes, please provide details.

4. Funding

4.1. Funder(s) European Research Council (ERC)

4.2. Grant type Research Council European Union

If you selected other, please provide further details.

4.3. Value of grant £ 1355696

4.4. Please upload a letter advising of the award of the grant.

6. Recruitment

6.1. Are the research participants able to give informed consent (in written or verbal form)? No

6.1.1. If no, is this because they are perceived to lack mental capacity or because they are vulnerable? Vulnerable

6.1.2. If the participants are perceived to lack mental capacity, please provide the reason(s).

6.1.3. Further details

6.1.4. If the participants are perceived to be vulnerable, please provide details of the vulnerability.

Research will involve children aged 2-36 months at time of testing. In line with previous studies of this type undertaken by the university, parents will be asked to consent to each test proposed in this study. Written information about each test will be emailed to parents at least a week before their visit to the lab, and they will be encouraged to ask for any clarification they feel they need. All participants are invited to ask questions and voice concerns about our consent and information documents, so that we can respond to or expand on any part of the process that is not clear.

6.1.5. Does the research/consultancy project involve children or young people under the age of 16?

Yes

6.1.6 If yes, are the children or young people able to give informed assent? No

6.1.7. If no, is this because they are perceived to lack mental capacity or because they are vulnerable? Vulnerable 6.1.8. If the participants are perceived to lack mental capacity, please provide the reason(s).

6.1.9. Further details

6.1.10. If the participants are perceived to be vulnerable, please provide details of the vulnerability.

The infant participants will be aged between 2 and 36 months at the time of testing. At these ages, the babies will be too young to give informed consent before starting, therefore parents will consent on behalf of their infants/children, as standard in this age cohort. In addition, they will be constantly monitored for signs of distress, and any procedure that is deemed upsetting to either parent, researcher or the baby will be halted. All participants will be informed that they have the right to halt proceedings at any stage. Babies and children will be constantly monitored, and any test will be discontinued should they become distressed or show signs of discomfort.

6.2. How will participants be recruited?

Infants will be recruited in four ways: 1) Infants will be recruited from the existing participant database in the Baby Development Lab at UEL; 2) infants will be recruited from third party infant groups such as local prenatal groups, baby-groups and children's centres. Third party infant groups will be found through social media and search engines and the leadership of these groups will be approached to gain permission to share information about infant research at UEL. No information will be shared with any participants of any infant groups without the permission of the gatekeepers. None of these groups will be NHS run groups (so no NHS ethics application will be required). For this project a live link to recruitment places will be sent to research ethics through Emily Greenwood. In person talks will only be conducted providing that this is legally permitted under COVID-19 regulations and we have gained specific permission for this from the leadership of these groups. 3) during lockdown, social media and word of mouth have proved to be the most fruitful participant recruitment avenues. Infants will be recruited through targeted posts on local Facebook parenting groups, and parents invited to get in contact with the lab. Should the group be private, permission will be sought to enter the group where we will disclose our intentions to highlight the research at the Baby Development Lab at UEL. Parents who contact the lab or who take part in existing studies will be asked to disseminate our information to any other parents/expectant mums they know. 4) infants will be recruited through 'flyering', where leaflets inviting parents to learn more information about this project and other research at the UEL baby development lab will be offered to members of the public in public spaces or by delivering leaflets to houses in the local area. When in public spaces, if relevant, permissions will have been sought from the management of the space. Members of the public will have no obligation to take or act on a leaflet. We will not employ flyering during lockdown due to COVID restrictions. For each of these methods, we will strictly comply with the relevant COVID-19 procedures enforced at the time. Fully informed consent will be obtained before testing commences. All of the parents of babies in this study will have received an information sheet and been given an

explanation of the aims of this study before providing their contact details.

When parents are initially contacted, they will be given more detailed information about what participation in the study involves via an information sheet, which will be emailed to them. At each interaction, an opportunity will be given to ask any questions or gain clarification. Before any data is collected, the parent/carer will be asked to sign a consent form. All participants are invited to ask

questions and voice concerns about our consent and information documents, so that we can respond to or expand on any part of the process that is not clear.

6.4. How many participants are being sought for the project?

150 pairs (one adult, one infant)

6.5. How long will participants be required for the project?

Prior to the testing sessions, parents will be asked to complete a series of questionnaires that should take no more than an hour. A small subset of adult participants (~30) will be asked to complete the Adult Attachment Interview, which should take an hour. Participation in the AAI will be voluntary.

Ten testing sessions are planned at infant ages 2, 6, 12, 18 and 36 months.

At each age range participants will be invited to the lab for a lab session lasting no longer than a few hours. Both adult and infant participants should be able to complete the entire lab session without needing breaks, however if infants are fussy and need a break then these will be taken flexibly during the session until the infant has calmed down. If the infant remains fussy throughout the break then the testing sessions may be ended early and rescheduled or cancelled. Infants are also welcome to take naps and sessions will be longer to accommodate for this.

Participants will also be invited to participate in an at home session. A researcher will travel to the participant's home to provide wearable equipment for the parent and infant, along with instructions on how to put on and remove the wearables. The researcher will also be available to answer any questions that the parents may have, this should take no longer than 45 minutes. The participants will be asked to keep the wearable devices on all day (~8-10 hours) until the researcher returns in the evening to remove and collect the wearable devices. This should take no longer than 20 minutes. Participants will be informed that they are able to stop or suspend recording on either the adult or infant devices at any time, for any reason. However, the devices will be comfortable so that wearing the device for the duration of the day is not an issue. Breaks are not required as part of the at home testing session.

The night-time wearable device will also be installed at the same time and any questions may be asked. This will add an extra 20 minutes to the evening visit. The night-time wearable device will be collected in the morning in a separate visit which should take no longer than 20 minutes. As with the daytime at home wearable device, the night-time wearable device has been designed to be comfortable so that it can be worn throughout the night without requiring any breaks. Participants will be informed that they are able to stop or suspend recording at any time for any reason.

6.6. Will the participants be remunerated for their contribution?

Yes

6.6.1. If yes, please specify monetary value of cash or giftcard / vouchers.

Participants will be offered the cost of travel costs to and from the lab for lab sessions.

Participants will also be offered a £10 Love To Shop voucher for each session that is completed.

7. DBS

7.1. Do you require Disclosure Barring Service clearance (DBS) to conduct the research/consultancy project?? Yes

7.2. Is your DBS clearance valid for the duration of the research/consultancy project? Yes

7.2.1. If you have current DBS clearance, please provide your DBS certificate number. 001717370584

8. Medical

8.1. Is your project a clinical trial and / or involves the administration of drugs, substances or agents, placebos or medical devices? No

8.1.1. If you answered yes, please explain why you have chosen to use this application form instead of the NHS/HRA ethics application form. If you have selected yes, your project requires approval by the NHS/HRA, as it is falls under the classification of Medicines for Human Use (Clinical Trials) Regulations (2004) or Medical Devices Regulations (2002) or any subsequent amendments to the regulations.

8.2. Does your project involve the collecting, testing or storing of human tissue / DNA including organs, plasma, serum, saliva, urine, hair, nails or any other associated material? No

8.2.1. If you answered yes, please explain why you have chosen to use this application form instead of the NHS/HRA ethics application form. If you have selected yes your project requires approval by the NHS/HRA, as it is falls under the classification of the Human Tissue Act (2004).

9. Risk

9.1. Does the project have the potential to cause physical or psychological harm or offence to participants and / or researchers?

9.1.1. If yes, please provide details of the risk or harm explaining how this will be minimised.

9.1.2. Please complete and upload a research risk assessment form9.2. Does the project involve potential hazards and/or emotional distress? Yes

9.2.1. If yes, provide an outline of support, feedback or debriefing protocol. Infants sometime express mild distress on the application of the EEG equipment. However, we have received extensive training in how to minimise this. We do not obtain any usable EEG recordings if infants are at all distressed, so we take every possible step to minimise this when it occurs. The process of recording EEG data from infants is in place in numerous other research labs across the world, and the process we follow will be identical to those used in other labs.

Some questions within the questionnaires (all attached in the attachments section) or the adult attachment interview (interview questions also attached in the attachments section) may be considered to involve emotional discomfort or distress to some participants. All participants will be informed that they do not need to complete any question or questionnaire that they don't want to. The adult attachment interview will be opt in for a small (~30) subset of participants and all participants will be informed that they can skip any question or stop the interview at any time.

After the participant has finished the session they will be given a debriefing form (see attached), in the unlikely event that the infant or adult participant has come to any distress they will be able to contact the principal investigator (Dr. Sam Wass) or the ethics subcommittee, both of which are listed in the debriefing sheet.

9.3. Provide an outline of any measures you have in place in the event of an adverse event or reaction or unexpected outcome, the potential impact on the project and, if applicable, the participants.

We do not expect adverse events arising from the study procedures. Under circumstances of an unexpected adverse event, the participants' health and safety will be the highest priority. If health and safety is in any way compromised, the participant will be withdrawn from the study, with clear reasoning given.

10. Anonymisation

10.1. Will the participants be anonymised at source? No

10.1.1. If yes, please provide details of how the data will be anonymised.

10.2. Are participants' responses anonymised or are an anonymised sample? No

10.2.1. If yes, please provide details of how the data will be anonymised.

10.3. Are the samples and data de-identified? Yes

10.3.1. If yes, please provide details of how the data will be anonymised/pseudonymised.

Direct and indirect identifiers will be removed from data and participants will be assigned a participant code. This will be entered along with the date and time of testing on the files containing raw data, and will be used to record all other data collected during pre-post assessments. Participant names and ID numbers will be stored in a separate password protected database. This is so that participants can withdraw their data up to the point at which it is included in the final analysis. Consent forms will be kept securely and separately from the raw data. Only members of the research team will have access to both the raw data and consent forms. Data will be retained in a secure place at the end of the

project as, should funding allow, we might wish to follow up the sample over a longer time period. Video recording of the behavioural paradigms will only be viewed by members of the research team.

10.4. Please provide details of data transcription.

All data will be analysed using MATLAB software. Behavioural paradigms will be coded according to their manuals. UEL students may be recruited as research assistants in order to code videos; assistants will be fully briefed on ethics related to the study and will not be allowed access to consent forms, code keys, or any other data that would allow them to identify the participant. Research assistants will only be permitted to code data on university computers (that are disconnected from the internet) and will not be allowed to copy or move video files from the university. Research assistants will be asked to complete a confidentiality agreement before undertaking any work on the project. An ethics amendment form will be completed and submitted to add research assistants to this ethics application before data transcription and coding starts.

10.4.1 Will the data be transcribed by person(s) outside of the project team?

If yes, please upload a blank copy of the confidentiality agreement.

10.5. If applicable, will all members of the project team know how the code links the data to the individual participant? Yes

10.5.1. If no, in the event of a researcher's absence please specify the process should access to the research data be required.

10.6. Will participants be anonymised/pseudonymised in publications that arise from the research/consultancy project? Yes

10.6.1. If no, please provide details.

10.7. Will participants have the option of being identified in the study and dissemination of research findings and / or publication? No

10.7.1. If yes, please provide details.

16. Data security

16.1. Will the researcher or the project team be responsible for the security of all data collected in connection with the proposed research/ consultancy project? Yes

16.1.1. If no, please provide details.

16.2. Will the research/consultancy data be stored safely on a password protected computer?

Yes

16.2.1. If no, please provide details.

16.3. Will the research/consultancy project data be stored on a UEL data managed device? Yes

16.3.1. If no, please specify where the electronic data will be stored and how the data will be kept secure.

16.4. Will you keep research/consultancy project data, codes and identifying information in a separate location? No

16.4.1. If yes, please explain how you will store the research data.

16.5. Will the raw data be shared with individuals outside of the project team? No

16.5.1. If yes, please specify the names, positions and their relationship to the research/ consultancy project

Name

Position

Relationship to research

16.6. Will participants be audio and/or video recorded? Yes

16.6.1. If yes, please explain how you will transfer, store and, where relevant, dispose of audio and/or video recordings.

Digital audio-video recordings will be transferred onto the UEL secure computer network that only the research team will have access. Video files may need to be stored externally; in this case, recordings will be stored and transferred on a password-encrypted hard drive with access limited to members of the research team.

16.7. If audio and/or video recordings will be retained, please provide details and state how long the recordings will be kept.

Recording will be kept for up to a year after the completion of the study and data analysis unless further funding can be obtained and the data is used again in further analyses, for which ethics approval of analyses of secondary data will be sought.

16.8. Will you retain hard copies of the data?

16.8.1. If yes, please provide details of how the data will be transported safely and, where relevant, undergo secure disposal.

16.9. Will the research/consultancy project data be encrypted and transferred inside of the UK? Yes

16.9.1. If no, provide details of where the research data will be stored and measures in place to keep the data secure.

Research data will be kept in the UK only. Research data will be stored on password protected, encrypted researcher laptops, dedicated password protected hard disks and will be backed up to UEL's OneDrive for Business. Only research team members will have access to this data.

16.10. How long will the research data that details personal identifiers be stored?

Any personal identifiers will only be retained until the research has been published and it is no longer possible for participants to request to withdraw their data.

16.11. Please upload a copy of your Data Management Plan.

18. Dissemination

18.1. Will the results be disseminated? Yes

18.1.1. If yes, how will the results of the research/consultancy project be reported and disseminated? Dissertation / Thesis

Peer reviewed journal Conference presentation Written feedback to research participants Books or chapters Blogs

18.1.2. If you selected other, please provide further details.

18.1.3. If the results of the research/consultancy /project will not be reported and disseminated, please provide a reason

20. Attachments

You can generate a Participant Information Sheet and Consent Form using the answers provided in your ethics application form. The Word files generated can be edited. You should upload the final version(s) before submitting your application form.

20.1. Upload any additional files to support your application which have not already been uploaded within your application.