

# **Regular Article**

# Sympathetic nervous system functioning during the face-to-face still-face paradigm in the first year of life

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#### **Abstract**

Contemporary theories of early development and emerging child psychopathology all posit a major, if not central role for physiological responsiveness. To understand infants' potential risk for emergent psychopathology, consideration is needed to both autonomic reactivity and environmental contexts (e.g., parent–child interactions). The current study maps infants' arousal during the face-to-face still-face paradigm using skin conductance (n=255 ethnically-diverse mother–infant dyads; 52.5% girls, mean infant age = 7.4 months; SD=0.9 months). A novel statistical approach was designed to model the potential build-up of nonlinear counter electromotive force over the course of the task. Results showed a significant increase in infants' skin conductance between the Baseline Free-play and the Still-Face phase, and a significant decrease in skin conductance during the Reunion Play when compared to the Still-Face phase. Skin conductance during the Reunion Play phase remained significantly higher than during the Baseline Play phase; indicating that infants had not fully recovered from the mild social stressor. These results further our understanding of infant arousal during dyadic interactions, and the role of caregivers in the development of emotion regulation during infancy.

**Keywords:** arousal; electrodermal activity; face-to-face still-face; infants; psychophysiology; responsiveness; skin conductance; sympathetic nervous system

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Theories of early development and emerging child psychopathology suggest a central role for physiological responsiveness (e.g., Dadds & Frick, 2019). These theories posit that children's psychological outcomes are dependent upon individual factors, such as, propensity for autonomic nervous system (ANS) arousal, especially in response to their environment, including caregiver interactions (e.g., El-Sheikh & Erath, 2011). In this context, ANS arousal is considered a moderator of environmental influences (e.g., parenting conflict, maternal sensitivity, exposure to trauma) on child adjustment and the development of psychopathology (El-Sheikh & Erath, 2011). Across human and animal studies, the ANS has been shown to be particularly sensitive to early caregiving (Alkon et al., 2014; Hostinar et al., 2014; McLaughlin et al., 2015; Shonkoff et al., 2009; Sroufe, 2005), and has been implicated in the development of psychopathology (Anda et al., 2006; De Bellis & Zisk, 2014; Shonkoff et al., 2009). The individual differences observed in ANS responsiveness is thought to protect, or amplify, the effects of the environment in the development of psychopathology. Baseline levels of ANS arousal are thought to affect children's readiness to respond adaptively to

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environmental stimulus, and ANS activity is thought to indicate sensitivity to environmental changes (El-Sheikh & Erath, 2011). Moreover, the ANS comprises two neurological subsystems—the sympathetic nervous system (SNS) and parasympathetic nervous systems (PNS)—each of which likely plays a differential role in regulating responsivity (Berntson et al., 1993). To understand an infant's potential risk for emergent psychopathology, methods are needed to measure an infant's autonomic reactivity within important environmental contexts (e.g., parent–child interactions).

Laboratory-based stress tasks provide a means for researchers to study infant responsiveness within parent-child interactions in real time (Jones-Mason et al., 2018). Those paradigms that are most widely used to manipulate stress levels and examine sensitivity to environmental input in children and adults (such as the Trier Social Stress Test and Startle paradigm) are not appropriate for infants. Thus, the Face-to-Face Still-Face (FF-SF) paradigm is most commonly used to examine sensitivity to environmental input with infants (Adamson & Frick, 2003; Jones-Mason et al., 2018; Mesman et al., 2009). The FF-SF paradigm is considered a mild social stressor, which has been found to elicit an arousal response in infants (Adamson & Frick, 2003; Jones-Mason et al., 2018; Mesman et al., 2009). The FF-SF is thought to elicit infant distress because it involves a violation of social expectations within

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the caregiver-infant dyad (Tronick et al., 1978). Rather than providing the infant with the expected regulatory emotional scaffolding through movement and changes in facial expressions (Beeghly & Tronick, 1994; Stack & Muir, 1990; Weinberg et al., 2008; Weinberg & Tronick, 1996), mothers become nonresponsive to their child, interrupting the infant's goals for connectedness and social engagement which are typically realized with their mothers (Tronick et al., 1978). The FF-SF paradigm typically involves three phases: A Baseline Free-play where mothers are instructed to play with their infant as they would at home; the Still-Face segment where mothers are asked to become unresponsive to their infant; and a Reunion Play segment where mothers are instructed to resume playing with their infant (Tronick et al., 1978). To date, observational coding during the FF-SF has implied that when mothers become unresponsive to infant cues indicators of stress are evident in newborns (e.g., Bertin & Striano, 2006) through to children aged up to 2.5-years-old (e.g., Weinberg et al., 2008). For instance, during the Still-Face phase children tend to show increased gaze aversion, less smiling, more negative affect, increased motor activity, increased tactile object- and self-stimulation, and less positive affect compared to the baseline free-play phase (Adamson & Frick, 2003; Mesman et al., 2009).

A number of studies have examined whether infants also show psychophysiological indicators of arousal during the FF-SF (see Jones-Mason et al., 2018 for a review). These studies have mostly focused on specific PNS activity or nonspecific ANS activity (i.e., aggregating SNS and PNS activity, e.g., heart rate). This literature has reported that infants become more aroused moving from the Baseline Play to Still-Face periods, with evidence for significant increases in heart rate (e.g., Bazhenova et al., 2001; Conradt & Ablow, 2010; Moore & Calkins, 2004; Weinberg & Tronick, 1996) and decreases in respiratory sinus arrhythmia (e.g., Bazhenova et al., 2001; Conradt & Ablow, 2010; Ham & Tronick, 2006; Moore, 2009; Moore & Calkins, 2004; Weinberg & Tronick, 1996).

Comparatively, few studies have examined SNS activity during the FF-SF (Bosquet Enlow et al., 2014; Busuito et al., 2019; Ham & Tronick, 2006, 2009; Suurland et al., 2017) with researchers commonly reporting methodological challenges (e.g., difficulty in obtaining resting state recordings due to excessive movement; likelihood of recording devices being removed during the experiment; see also, Mesman et al., 2009). Bosquet Enlow et al. (2014) found there were no groupwise variations in T-wave amplitude across the episodes of the FF-SF. Similarly, Suurland et al. (2017) found no groupwise variations across the episodes of the FF-SF in relation to infant preejection period. Ham and Tronick (2006) and Busuito et al. (2019) measured skin conductance and found differences across the FF-SF paradigm. In their pilot study (n = 12), Ham and Tronick (2006) found that skin conductance typically increased from the Baseline play to Still-face for 5-monthold infants. Similarly, Busuito et al. (2019) found that 6-month-old infants demonstrated a significant increase in skin conductance from the Baseline play to Still-face phase (n = 140).

Despite strong agreement on expected patterns of arousal across the phases of the FF-SF task in the behavioral literature (Mesman et al., 2009), patterns of arousal—as indexed by psychophysiological measures—have presented a more mixed picture from the still-face to the Reunion play periods across the literature. Findings are less clear from still-face to Reunion Play periods, with some studies examining PNS and nonspecific ANS arousal finding a reregulation effect with increases, on average, in respiratory sinus arrhythmia (e.g., Ham & Tronick, 2006; Moore & Calkins, 2004;

Weinberg & Tronick, 1996) and decreases, on average, in heart rate (e.g., Moore & Calkins, 2004; Weinberg & Tronick, 1996). In contrast, other studies examining PNS and nonspecific ANS arousal have found the opposite effects, on average, for respiratory sinus arrhythmia (e.g., Bush et al., 2017; Conradt & Ablow, 2010) and heart rate (e.g., Bosquet Enlow et al., 2014; Conradt & Ablow, 2010; Ham & Tronick, 2006; Moore et al., 2009). When considering studies examining SNS arousal, the two skin conductance studies found increases, on average, in tonic skin conductance activity between the still-face and Reunion play phases (Busuito et al., 2019; Ham & Tronick, 2006). It is possible that findings may differ according to which measure is being used, the sample employed (e.g., composition of infants with risk status, differing attachment, greater weight-for-length gain; Jones-Mason et al., 2018; Rudd et al., 2020; Suurland et al., 2017), and which part of the ANS is being measured. An examination of skin conductance may be preferable over PNS and nonspecific ANS functioning due to the specificity in measurement that skin conductance allows. In particular, skin conductance provides a direct measurement of SNS activity unlike measures of PNS functioning or measures of nonspecific ANS activity that aggregate SNS and PNS activity (i.e., where researchers cannot determine which activity is PNS or SNS functioning). SNS activity is also assumed to index infant arousal during the FF-SF task (Boucsein, 2012).

Although the two skin conductance studies (Busuito et al., 2019; Ham & Tronick, 2006) found an increase in arousal between the Still-face and reunion phases, on average, these results differ from the extensive behavioral coding of the FF-SF. It is possible that this consistency across the two skin conductance studies indicates that SNS arousal may be experienced even when there are fewer behavioral indications of arousal. However, the skin conductance findings reported by Busuito et al. (2019) from the Still-Face to the Reunion Play did not show the same pattern of results as their other infant psychophysiological measures that were taken at the same time. This suggests that infants were not at a higher arousal level (as indicated by the other measures). Although it is difficult to say with certainty why this has occurred (i.e., both studies have failed to mention the current circuits for the electrodes used), it is likely that build-up of counter electromotive force was not estimated in Busuito et al. (2019) or Ham and Tronick (2006). Counter electromotive force (e.m.f.) refers to the polarization of cellular membrane potentials in the skin that occurs with direct current electrodes over time (even when using nonpolarizing Ag/AgCl electrode gels; Boucsein, 2012). Such forces are typically regarded as difficult to estimate due to build-up being nonlinear and varying between individuals (Posada-Quintero & Chon, 2020). While this issue has not been explicitly investigated within the electrodermal activity (EDA) literature to-date (Boucsein, 2012; Grimnes et al., 2011), it is possible that—particularly in the FF-SF paradigm, which has such a long tradition of behavioral confirmation in the empirical literature—counter e.m.f. build-up contributes to differences between behavioral results and results from the relatively few attempts to apply skin conductance measures. If this hypothesis were true, then the confounding effects of counter e.m.f. would be maximized during the reunion phase of the FF-SF paradigm.

Collectively, research to-date using the FF-SF paradigm demonstrates four important findings: (1) It is possible to examine ANS activity in infants; (2) infants consistently demonstrate increased skin conductance arousal from Baseline play to Still-face phases of the protocol; (3) the pattern of arousal from Still-Face to Reunion phases is inconsistent across the literature; and (4) the

potential build-up of e.m.f. has not been appropriately modeled in previous FF-SF studies examining skin conductance. The FF-SF paradigm therefore provides an ideal task to be able to examine ANS arousal in the context of environmental influences (i.e., changes within mother-infant interactions) on child adjustment (i.e., including infants' degree of recovery during the Reunion phase). Further, infants' degree of recovery from the Still-face social stressor may be an important marker for the development of psychopathology as children age (Dadds & Frick, 2019; El-Sheikh & Erath, 2011).

#### The current study

The aim of the current study was to examine the SNS psychophysiological indicators of arousal during the FF-SF as detected by changes in skin conductance to achieve a better understanding of patterns of infant reactivity to parent-child interactions in early life. The present study will be the first FF-SF study to account for the potential build-up of nonlinear counter electromotive forces over the course of the task, which is critical to the appropriate investigation of SNS psychophysiological functioning over time when examining skin conductance with DC-based measurement.

This study investigated SNS psychophysiological functioning during the FF-SF with a sample of infants aged 5.4–10.7~(M=7.4; SD=0.9) months. Infants within the first year of life were of interest as emerging research has shown that SNS psychophysiological functioning may be shaped by, and regulated by, dyadic caregiver interactions (Busuito et al., 2019), which may result in cascading developmental psychopathology risk across infancy (Dadds & Frick, 2019; El-Sheikh & Erath, 2011). Further, the FF-SF paradigm is typically used within the first year of life, and there is limited movement using the soles of feet during this time, which allows for collection of skin conductance data with limited interfering noise.

In accordance with the extensive FF-SF literature using infant behavioral coding (Mesman et al., 2009) and two prior skin conductance studies (Busuito et al., 2019; Ham & Tronick, 2006), it was hypothesized that infants' skin conductance response will significantly increase between the Baseline play and Still-face phases. In accordance with our novel statistical design, it was hypothesized that infants would show a significant decrease in skin conductance during the Reunion play relative to the Still-face phase, where the impact of potential e.m.f. is expected to be maximized. Support for this second hypothesis would provide evidence that infants' skin conductance data is consistent with the extensive FF-SF behavioral coding literature. Moreover, such support would highlight the importance of designing and applying statistical models that can flexibly adjust for potential e.m.f. build-up as the only two FF-SF skin conductance studies previously conducted (Busuito et al., 2019; Ham & Tronick, 2006) both found a significant increase in skin conductance during the Reunion Play relative to the Still-Face phase, and, notably, do not employ statistical designs that adjust for potential e.m.f. build-up over the course of the task.

#### Method

# **Participants**

Mother-infant dyads were drawn from the "Watch Me Grow for REAL" longitudinal birth-cohort study (Doyle et al., 2020). The "Watch Me Grow for REAL" study follows 788 mother-infant dyads from a low socioeconomic urban area prospectively from mother's antenatal appointments (i.e., T0 assessment). This

sample was a normative community sample, but was recruited from this area as they were more likely to experience risk factors that might be associated with the development of child psychopathology (e.g., low socioeconomic status). Three hundred and ninety-six dyads attended the Time point 1 (T1) assessment (185 girls, 46.7%). Infants' age ranged from 5.4 to 10.7 (M = 7.4; SD = 0.9) months. All children were born in Australia. Infants were of European (n = 114, 28.8%), Middle-Eastern (n = 85, 21.5%), South Asian (n = 66, 16.7%), South-East Asian (n = 59, 14.9%), American (n = 22, 5.6%), Polynesian/ African/African Melanesian (n = 19, 4.8%), Hispanic/Latino (n = 15, 3.8%), East Asian (n = 10, 2.5%), and Aboriginal/Torres Strait Islander (n = 5,1.3%) descent, and one child had missing ethnicity data (0.3%). Mothers were aged between 19.0 and 55.5 years (M = 32.0; SD = 5.2). Three hundred and fifty-eight mothers (91.3%) were in a romantic relationship with the infant's father at T1, with 305 mothers (77.8%) reporting that they were married. Only 155 (39.1%) mothers were born in Australia. Eighty mothers (20.2%) lived in Australia for 0-5 years, 67 mothers (16.9%) lived in Australia for 6-10 years, 34 mothers (8.6%) lived in Australia for 11–15 years, 28 mothers (7.1%) lived in Australia for 16–20 years, and 182 mothers (46%) lived in Australia for 21 years or more (with 5 mothers, 1.3%, having missing data). Mothers' highest level of completed education (as self-reported at T1) was: Never attending school (n = 1, 0.3%), primary school (n = 6, 1.5%), Grade 10 or equivalent (n = 21, 5.3%), high school (i.e., Year 12 or equivalent; n = 61, 15.4%), technical or vocational qualification (n = 94, 23.7%), undergraduate university degree (n = 145, 36.6%), postgraduate university degree (n = 64, 16.2%), and 4 mothers (1.0%) had missing data. Prior to their child's birth, mothers reported their typical employment was full time work (n = 169, 42.7%), part time work (n = 47, 11.9%), casual work (n = 48, 12.1%), studying (n = 11, 2.8%), stay-at-home mother (n = 84, 21.2%), unemployed (n = 35, 8.8%), and 2 mothers had missing data (0.5%). According to the Australian Bureau of Statistics' socioeconomic indices for areas (Index of Relative Socio-Economic Disadvantage), which combines Australian census data on household income, education, and unemployment by postcode, it was found that 209 (52.8%) of the sample lived in an area in the lowest decile; indicating greater disadvantage. Mothers received a \$40 gift voucher to thank them for their time while infants were given a "Baby Scientist" certificate for their participation.

#### Procedure

Mother-infant dyads participated in the T1 laboratory visit for approximately 2 hours (for a full outline of measures at T1 see Doyle et al., 2020). During the T1 assessment, two gelled silver-silver chloride (Ag/AgCl) 'Skintact' electrodes were applied to both soles of each infant's foot. Infants' feet were selected for placement of electrodes because eccrine sweat glands are typically most dense on the hands and feet (Mendes, 2009), and placement on infants' hands would be more likely to interfere with the FF-SF task, and were not tolerated by infants during pilot testing. Electrodes were then connected to 'Consensys GSR' sensors (Shimmer, MA, USA), and a pair of socks was placed over the sensors and infant's feet to prevent the infants from detaching the electrodes either deliberately or due to artifact. Infants were seated in a raised bouncer chair with their mothers seated on a chair opposite their child. The FF-SF (Tronick et al., 1978) task was commenced to elicit infant's physiological responses to a mild social stressor involving the following sequence of events: The face-to-face Baseline play ("Baseline Play"

phase) was commenced for 240 s for which toys were provided; the researcher then returned to the room gave the instructions to commence the face-to-face "Still-Face" then left the room, and the Face-to-Face "Still-Face" phase was commenced for 90 s; finally, the researcher again returned to the room and a further face-to-face play ("Reunion Play") phase was commenced for 240 s.

#### Physiological assessments

Physiological EDA data were continuously collected throughout all phases of the experiment using a sampling frequency of 5 Hz. Phases corresponding with those of the FF-SF paradigm (described above) were time-stamped by researchers using the event marking feature of the ConsensysPRO software package (Shimmer Research Ltd., Dublin, Ireland). EDA data were preprocessed using the Frog (v.0.1.1) package in the R language developed specifically for the "Watch Me Grow for REAL" study. A range of psychophysiological features have been identified in the literature for EDA including skin conductance levels (SCL) and skin conductance peaks per epoch (PPE; see Boucsein, 2012 for a review). These features were extracted using an unsupervised process provided by the Affective Computing Lab at MIT (Taylor et al., 2015). The threshold for a skin conductance peak was set to 0.005 μSiemens. Artifacts were identified in 5-s epochs upon the basis of concomitant accelerometer data using a state-of-the-art multiclass classification algorithm from the same toolkit (Hemmelmann, 2018; Taylor et al., 2015).

#### Missing data

Of the 396 infant-mother dyads who attended the T1 assessment, 34 (8.6%) dyads did not attempt the FF-SF paradigm because the infant was too fussy, fell asleep before attempting the paradigm, or mothers ran out of time to complete all aspects of the broader T1 assessment. Of the 362 dyads who attempted the FF-SF paradigm, EDA data was not collected for 24 (6.1%) infants (e.g., computer program was not working; application of EDA sensors aborted due to infant distress/fussiness). From those who had data available, a further 83 (20.1%) infants were excluded: 34 infants (66 channels; [16.6% of the full sample]) for whom data were missing (e.g., event markers missing [14%] or erroneous [47%]; accelerometer data missing [39%]); 49 infants (49 channels; [12.3% of full sample]) with mean conductance  $< 5 \mu S$  or  $> 120 \mu S$  by device, indicating defective channels. In total, 255 infants (509 channels; [64.2% of the T1 sample]) were retained for statistical analysis, which is similar to, or better than, rates of data loss reported by past studies measuring skin conductance in infants (Busuito et al. (2019).

Sociodemographic differences between those who attended the T1 assessment (n=396) and those included in the final sample (n=255; 52.5% girls) were assessed. Infants in the final sample were of Caucasian (n=80, 31.4%), Middle-Eastern (n=56, 22.0%), South Asian (n=42, 16.5%), South-East Asian (n=34, 13.3%), African/African American (n=10, 3.9%), Polynesian/Melanesian (n=12, 4.7%), Hispanic/Latino (n=11, 4.3%), East Asian (n=6, 2.4%), and Aboriginal/Torres Strait Islander (n=3, 1.2%) descent, and one child had missing ethnicity data (0.4%). A generalized linear regression model (GLM) was fit to the data with birth weight, gestation duration, and the number of years the infant's mother has lived in Australia included as predictors. Although the GLM was found to fit the data well (Hosmer–Lemeshow goodness of fit test,  $\chi^2(8) = 13.45$ , p=.097), all predictors were found to be nonsignificant (all ps > .05). Similarly, no

differences were found between groups in child gender using Pearson's Chi-squared test with Yates' continuity correction,  $\chi^2(1)=2.42, p=.120$ , Cohen's  $\omega=.06$ . Finally, no differences were found between groups for mothers' age, Welch's two-sample t test, t(200.94)=.19, p=.850. These tests were repeated to examine whether there were differences between those initially recruited (N=788) and those included in the final sample in this study (n=255); however, again, nonsignificant differences were found (all ps>.05).

#### Data preparation

For each participant, data were extracted based upon the first event marker (i.e., transition in phase between Baseline Play and Still-Face); truncating recordings for Baseline Play and Reunion Play phases to uniform bin-widths. Data were resampled from 5 to 8 Hz in preparation for peak and artifact detection classification. Preparatory visualizations indicated the data did not conform to linear expectations at the extremes of the data set. To determine whether cohort effects or outlier behavior drove this tendency, individual plots were built for each participant leading to the identification and further exclusion of eight participants (3.1% of inclusions; 2.0% of full sample).

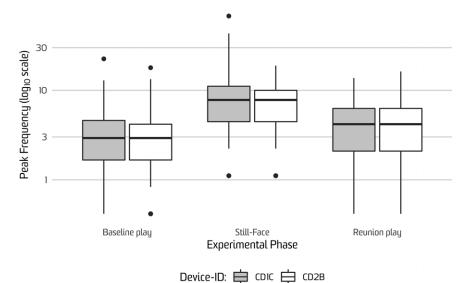
Devices (IDs CD1C and CD2B) were randomly placed on infants' feet and recorded EDA data throughout the FF-SF. At the time of data collection, it was not recorded which device ID was on each foot for each infant. Further, chirality data was not collected for each participant. Chirality data was not collected as handedness does not stabilize in infants until 18-24 months of age (Michel et al., 2016; Nelson et al., 2013) and was not expected to drive the target effects of this study. Thus, for each participant, we had two streams of EDA data which were associated a device ID (i.e. CD1C and CD2B). At the time of data analysis, to examine whether there would be a possibility of an interaction between handedness (unknown) and device ID (known), a simulated exploratory analysis was run. Naïve linear models were used to simulate (n = 10,000) the possible interaction between chirality and the specific instrument applied to the infant for the purposes of recording EDA (i.e. device IDs CD1C and CD2B). During each run of the simulation a single channel from one of the devices was randomly selected from each participant; thus, this approach counterbalances for device ID and controls for sampling bias. Results of this simulation indicated the presence of an instrumental difference of device ID. Device ID was therefore taken into account in subsequent analyses (see Supplementary File 1 for full details).

## Statistical Analyses

Statistical analyses were conducted using the R language v3.6.1 (R Core Team, 2021) within the RStudio IDE (RStudio Team, 2020). Full reproducible code and session information are available in Supplementary Files. As range of psychophysiological measures have been identified in the literature for EDA (Boucsein, 2012), the EDA data from this study were analyzed in two ways to examine whether the pattern of effects was consistent across these two measures (i.e. peak events by phase and tonic SCLs).

First, a one-way ANOVA was employed to analyze PPE data. To examine peak events by phase, post hoc comparisons using Tukey's HSD test were run.

Second, tonic SCLs were examined. As noted above, our design attempts to estimate the impact of e.m.f. as a potential confound within the tonic SCL data. By treating e.m.f. as an individual



**Figure 1.** Frequency of infant peak events detected during the Face-to-Face Still-Face paradigm factored by device ID. *y*-axis reflects the number of peak events detected by phase on log<sub>10</sub> scale. "Device ID" refers to the two Shimmer sensors that were each randomly assigned to infants' feet (CD1C, CD2B).

difference factor that increases over the duration of the experiment (i.e., as a random effect within a linear mixed effects model), it may be possible to partition variance associated with linear build-up of e.m.f. into a dummy variable whose impact is dynamically subtracted from fixed effects of interest. Accordingly, an adjustment variable was created such that the final sample per phase per person was centered upon zero with preceding samples approaching linearly from the negative direction. An interaction was specified between this adjustment variable and each phase, ranging over the full duration of the experiment. Moreover, by centering these data, intercepts were therefore fit to the end of each phase. This was done to ensure that differences between phase recordings would be maximized and also not be affected by potential SNS latency. Expectations of differences between devices were accounted for by introducing simple contrasts on device ID with the global intercept suppressed. Random intercepts were introduced for each participant with correlated random slopes expressing the same interaction between the adjustment variable and phase as defined in the fixed effects. This model was fit under restricted maximum likelihood (Harville, 1977) using the "nlme" package v3.1 (Pinheiro et al., 2007).

#### **Results**

### Analysis of peak events per epoch

Summary statistics for peak event data are presented in Table 1. Significant differences in mean peak event frequencies by phase were found using a 2 x (Device ID: CD1C, CD2B) between-subjects by 3 x (Phase: Baseline Play, Still-Face, Reunion Play) within-subjects mixed ANOVA with Hyunh–Feldt correction; F(2, 1185) = 220.16, p < .001, partial  $\eta^2 = .38$ . Post hoc comparisons using Tukey's HSD test indicated that the mean peak frequency for the Still-face phase was significantly higher than both the Baseline Play phase ( $M_{\rm d} = 4.96$ , p < .001; 95% CI [4.26, 5.66]) and the Reunion Play phase ( $M_{\rm d} = 3.81$ , p < .001; 95% CI [3.10, 4.52]). Likewise, the mean peak frequency for the Reunion Play phase was significantly higher than the Baseline Play phase ( $M_{\rm d} = 1.15$ , p < .001; 95% CI [0.45, 1.85]). These differences are visualized in Figure 1.

#### Analysis of tonic skin conductance

Linear mixed-effects modeling was used to examine tonic conductance by phase. Fixed effects for this model are reported in Table 2, with random effects and associated variance-covariance matrix reported in Table 3. A significant positive change in the gradient was observed between the Baseline free-play and still-face phases. Conversely, a significant negative change in the gradient was observed between the Still-Face and Reunion Play phases. The change observed from the Baseline Play to the Still-Face phase supports our experimental predictions that infants experience greater SNS arousal, as measured in skin conductance, with the still-face manipulation, in accordance with conventional results from the behavioral FF-SF paradigm. Concomitant decreases in SNS activity between still-face and Reunion Play phases suggest that mothers play an important role in externally regulating psychophysiological stress in infants. Predictions are further supported by significant differences observed between intercept values between the Reunion Play and Baseline Play phases. Fixed effects for device ID were small but nonetheless significant, vindicating the use of contrasts. Finally, significant interactions between the adjustment variable and both the still-face and Reunion Play phases—though, notably, not the Baseline Play phase—were identified, validating the inclusion of the adjustment variable for the purpose of adjusting for potential electromotive force build-up over the duration of the experiment. It is notable that SEs for the random effects of phase decrease for each consecutive phase while SEs for their interactions with the adjustment variable increase (see again, Table 2). Taken together, these features indicate a strong increase in SCL variability over the course of the experiment.

#### **Discussion**

We mapped the topography and temporal sequencing of SNS psychophysiological indicators of arousal during the FF-SF as detected by changes in skin conductance with a large and diverse sample of infants. In accordance with our first hypothesis, on average, infants showed a significant increase in SCL between the Baseline Free-Play phase and the Still-Face phase of the FF-SF paradigm. This result held across both the peak events and tonic skin conductance analyses. These findings align with the two

**Table 1.** Summary statistics for the time-adjusted frequency of peak events detected by phase and by device ID

Experimental phase	M (SD)	Device ID	n	M (SD)
Baseline play	3.37 (2.44)	CD1C	207	3.38 (2.53)
		CD2B	198	3.36 (2.35)
Still-face	8.33 (6.29)	CD1C	206	8.98 (7.61)
		CD2B	189	7.63 (4.34)
Reunion play	4.52 (2.79)	CD1C	197	4.51 (2.81)
		CD2B	191	4.53 (2.78)

Note. M = mean; SD = standard deviation.

**Table 2.** Estimated model fixed effects for infant skin conductance responses during the Face-to-Face Still-Face paradigm at 6 months

	Effect	Estimate	Std. error	t statistic	<i>p-</i> value	95% Confidence intervals	
						Lower	Upper
	Device <sup>a</sup>	-1.028	0.01	-84.20	<.001	-1.052	-1.004
	Baseline play	19.758	0.83	23.77	<.001	18.129	21.387
	Still-face	26.735	0.79	33.82	<.001	25.186	28.285
	Reunion play	23.128	0.83	27.99	<.001	21.508	24.748
Phase time:	Baseline play	0.005	0.00	1.87	0.062	0.000	0.009
Phase time:	Still-face	0.036	0.01	5.30	<.001	0.023	0.050
Phase time:	Reunion play	-0.036	0.01	-6.02	<.001	-0.048	-0.024

Note. <sup>a</sup>Reference category for device is CD1C. "Phase time" refers to the within-phase adjustment variable.

previously conducted studies of skin conductance arousal during FF-SF paradigm (Busuito et al., 2019; Ham & Tronick, 2006), as well as what tends to be seen behaviorally in the extensive FF-SF behavioral coding literature (Mesman et al., 2009). Thus, it would appear that there is robust evidence that during infancy the Still-Face manipulation acts as a universal mild social stressor by disrupting reciprocal engagement between infant and caregiver, as indicated by increased SNS activity. These findings also complement findings on PNS and nonspecific ANS psychophysiological arousal indicators (e.g., respiratory sinus arrhythmia, heart rate) showing increased arousal during the FF-SF (see Jones-Mason et al., 2018 for a review).

Supporting our second hypothesis, it was shown that, on average, there was a significant decrease in skin conductance during the Reunion Play when compared to the Still-Face phase. Although we did not examine coded infant behavior in this study, our skin conductance finding aligns with the extensive literature that has coded infants' behavior during the FF-SF in showing that mothers' reengagement in play has a buffering effect in reducing infants' arousal (Adamson & Frick, 2003; Mesman et al., 2009). In particular, behaviorally coded FF-SF data has shown that infants tend to display increased gaze toward their mother, less negative affect, and more positive affect during the reunion phase compared with the still-face phase (Adamson & Frick, 2003; Mesman et al., 2009).

**Table 3.** Estimated random effects accompanying with associated variance-covariance matrix

	Effect	Std. error	Correlations				
	Baseline play	13.235					
	Still-face	11.042	-69.483				
	Reunion play	10.274	-55.124	61.847			
Phase time:	Baseline play	0.038	0.262	-0.198	-0.159		
Phase time:	Still-face	0.108	-0.744	1.026	0.528	-0.002	
Phase time:	Reunion play	0.940	-0.180	-0.050	0.644	-0.001	0.000

Note. Random effects fitted with positive-definite structure using Log-Cholesky parametrization (Pinheiro & Bates, 1996). "Phase time" refers to the within-phase adjustment variable.

However, when using various psychophysiological measures (i.e., measures of SNS; PNS; and, nonspecific ANS, which include measures that do not differentiate between SNS and PNS functioning), there are more mixed findings in the literature for changes between the still-face to Reunion Play phases. Consistent with the present study, while some studies have found a re-regulation effect as evidenced by increases in respiratory sinus arrhythmia (e.g., Ham & Tronick, 2006; Moore & Calkins, 2004; Weinberg & Tronick, 1996) and decreases in heart rate (e.g., Moore & Calkins, 2004; Weinberg & Tronick, 1996), other studies have found opposite patterns for respiratory sinus arrhythmia (e.g., Bush et al., 2017; Conradt & Ablow, 2010), heart rate (e.g., Bosquet Enlow et al., 2014; Conradt & Ablow, 2010; Ham & Tronick, 2006; Moore et al., 2009), and skin conductance (Busuito et al., 2019; Ham & Tronick, 2006) response.

In regards to the specific skin conductance findings, our Stillface to Reunion Play results differ to findings from Busuito et al. (2019) and Ham and Tronick (2006), which found that skin conductance increased, on average, across the FF-SF task. These prior findings, however, may be explained by the lack of explicit statistical control for e.m.f. build-up when using DC electrodes, which this study aimed to overcome (for comparison between models with and without our adjustment variable, see Supplementary File 2). While the overall variance accounted for by our solution was modest (see Table 2), it was nonetheless significant and aligns with the results of the simulation analysis (see Supplementary File 1) indicating a pattern of increased variance across the FF-SF task. Furthermore, the absence of explicit controls for potential e.m.f. build-up in Busuito et al. (2019) may explain why their skin conductance findings between Still-face and Reunion play did not show the same pattern of results with their other infant psychophysiological measures taken at the same time. It is likely, but less clear, that potential effects of counter e.m.f. explain the pattern of results in Ham and Tronick's (2006) pilot work. Due to the small sample size, it is more challenging to deduce whether sample composition (e.g., composition of infants with risk status, differing attachment, greater weight-for-length gain; Jones-Mason et al., 2018; Rudd et al., 2020; Suurland et al., 2017) and/or measurement problems (e.g., counter e.m.f. build-up) are influencing the groupwise and sub-group findings. Our present findings highlight the importance of adjusting for potential e.m.f. build-up across time

when employing skin conductance measures with direct current circuits in future studies.

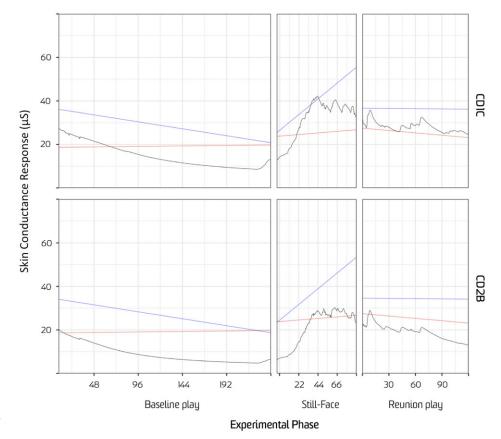
Although infants' arousal reduced from the Reunion Play when compared to the Still-Face phase, arousal remained significantly higher than during the Baseline Play phase. These findings indicate that infants recover once the stressor (i.e., Still-Face) has been removed, and highlights the likely role of self-regulation and maternal co-regulation behaviors (Adamson & Frick, 2003; Mesman et al., 2009). It is possible that increasing missingness in the data may have artificially deflated Reunion Play estimates as infants at times fidgeted or "kicked off" electrodes. That is, each successive phase over time had fewer electrodes with active recordings (i.e., infants were more likely to fidget/"kick off" the electrodes over time simply as a matter of opportunity) and therefore there were a greater proportion of channels with missing active recordings and/or poor skin contact during the Still-Face and Reunion Play phases of the experiment, which could have negatively impacted fixed effects estimated for these phases. However, we believe it is unlikely that this issue wholly accounted for our results for two reasons: First, the random effects structure applied in the analysis of SCLs adjusts for the contribution of high variance channels via partial pooling within the random effects structure of our analytic models; second, the tonic skin conductance dynamics match those identified in the analysis of PPE for which no partial pooling was applied by participant or by phase. Nonetheless, it is notable that, to our knowledge, the confound associated with increased data loss over time has not been considered within the EDA literature, particularly in relation to infants for whom the collection of psychophysiological data is especially challenging and artifact and data loss are ineluctable. Indeed, when examining missing data trends across this literature, Busuito et al. (2019) reported 40%-58% of infants in their sample had missing skin conductance data during each phase of the FF-SF; which was the highest level of missingness across all of the infant psychophysiological measures examined. Similar to the present study, Busuito et al. (2019) also collected skin conductance data from infants' feet. Missingness for SCLs in the present study was markedly lower when compared to Busuito et al. (2019) across all phases of the FF-SF paradigm: Baseline Play: 5 (2.0%); Still-Face: 6 (2.4%); Reunion Play: 12 (4.7%). Future research should take care to ensure that infants are fitted with EDA sensors in locations that are least likely to result in disconnection due to fidgeting, infant interference, and/or removal due to expected movement during the FF-SF paradigm (e.g., increased motor activity or increased tactile object- and self-stimulation; Adamson & Frick, 2003; Mesman et al., 2009). Future research might also seek to identify and include data for missing samples due to such disconnection issues to better allow statistical models to adjust in relation to other parameters (e.g., participant; phase; device ID).

The current findings should be considered in light of some methodological limitations. First, because EDA provides a measure of emotion-related arousal—but not discrete emotional states or associated valence—we are unable to differentiate which discrete emotional states or associated valence were captured with EDA (Collet et al., 1997). This is because EDA provides a measure of emotion-related arousal, but not discrete emotional states or associated valence. Second, resting state EDA recordings could not be taken with this infant sample. The primary reason for this was that infants appeared to be mildly uncomfortable following the application of the electrodes associated with the Shimmer sensors (see Figure 2). This meant that any resting state recordings might have been confounded due to infants' differing capacity to tolerate the

application of the electrodes; a limitation shared by most infant studies. Although EDA measures with adult participants typically uses differences between resting state EDA recordings and experimental EDA recordings as a within-subjects dimension of genuine skin conductance responding, in this study differences between the three phases of the FF-SF paradigm were examined. Third, as outlined earlier, a measurement of chirality was not collected because handedness does not stabilize in infants until 18-24 months of age (Michel et al., 2016; Nelson et al., 2013). Nonetheless, future research may wish to empirically test the possibility of an interaction between infants' handedness and the devices used or ensure that devices are counterbalanced across recording site. Finally, this study did not include a measure of infants' behavior. Extensive previous research, however, has been conducted on infants' behavioral responding during the FF-SF (Adamson & Frick, 2003; Mesman et al., 2009), with which our psychological findings aligned with typical behavioral patterns. As some studies (e.g., Busuito et al., 2019) have not shown concordance between psycho-physiological measures and coded behavior, it is important to note that it is possible that behavior in the present sample may not align with our skin conductance arousal findings nor expected behavioral patterns; for example, some infants in the current sample may have discordant behavioral responses. Future research is needed to clarify if the current sample's skin conductance aligns with coded behavioral measures within the present sample.

Despite these limitations, this study features some noteworthy strengths. This study was the first study to examine SNS functioning, and specifically SCR, during the FF-SF paradigm with a heterogeneous sample (i.e., low SES; varied ethnicity; larger infant age range; families from culturally and linguistically diverse backgrounds) of mother-infant dyads constituting the largest sample of infants to date (N = 255). This is a notable study strength as the psychological literature is typically dominated by studies of predominantly Caucasian samples, as is previous studies investigating skin conductance during the FF-SF (Busuito et al., 2019; Ham & Tronick, 2006). Consideration of both autonomic reactivity and environmental context is needed to understand infants' potential risk for emergent psychopathology. Thus, in order to ensure that SNS responses to the FF-SF paradigm are robust, it is essential that researchers examine infants with a range of risk profiles. A second study strength unique to the present study was that statistical analyses were designed to adjust for the potential e.m.f. build-up across the duration of the paradigm. This statistical approach provides confidence that instrumental factors with respect to the choice of skin conductance electrodes do not account for our findings. Moreover, our results also appear internally reliable as they were consistent across multiple psychophysiological measures of EDA (i.e., PPE and SCLs), and align with what tends to be seen behaviorally in the large body of literature that has examined the FF-SF using behavioral coding (Adamson & Frick, 2003; Mesman et al., 2009).

Our results stand to inform conceptualizations of the role that the ANS, particularly its sub-component of the SNS, plays in early life. Across human and animal studies, the ANS has been shown to be particularly sensitive to early caregiving (Alkon et al., 2014; Hostinar et al., 2014; McLaughlin et al., 2015; Shonkoff et al., 2009; Sroufe, 2005), and has been implicated in the development of psychopathology (Anda et al., 2006; De Bellis & Zisk, 2014; Shonkoff et al., 2009). In this study, the FF-SF paradigm provided the ideal context in which to examine infants' SNS functioning during maternal interactions with a diverse community birth-cohort sample of infants. It was shown that for all infants, on



**Figure 2.** Exemplar EDA recording by experimental phase and device ID, with fixed and random effects overlaid.  $\mu S = micro$  Siemens. Experimental phase axes are measured in seconds. Fixed effects are represented by the red lines; random effects by the blue lines. "Device ID" refers to the two Shimmer sensors that were each randomly assigned to infants' feet (CD1C, CD2B).

average, the Still-face manipulation was a mild social stressor; yet infants also differed in their relative Baseline free-play skin conductance, as well as the extent to which they experienced SNS arousal to the social stressor. There was strong variability over the duration of the paradigm that might be explained by individual difference factors, for which future research attention is needed. By establishing theoretically important and generalized responses to social interactions, it allows the paradigm to measure individual differences in arousal that may provide insight into which infants are predisposed to mental health problems later in life (Dadds & Frick, 2019). The strong variability observed over the duration of the paradigm may well be explained by individual difference factors (such as infants' patterns of reactivity and recovery time), and therefore warrants future research attention. By further examining infants' physiological responsiveness within dyadic caregiver interactions, researchers may better understand the extent to which infants' responsiveness protects, or amplifies, the effects of the environment on the development of child psychopathology (Dadds & Frick, 2019).

In conclusion, an infant's potential risk for emergent psychopathology, is linked to both autonomic reactivity (e.g., skin conductance) and environmental contexts (e.g., parent-child interactions). EDA data from 7-month-old infants showed that tonic SCLs —as a measure of SNS functioning—significantly increased, on average, during the Still-face and then significantly reduced, on average, during the Reunion Play phase. Infants' skin conductance during the Reunion Play phase still remained significantly higher than during the Baseline Play phase; indicating that they had not fully recovered from the mild social stressor. Moreover, an identical pattern in SNS responding was found when separately analyzing peak events data. Unlike previous examinations of infants' skin

conductance during the FF-SF paradigm, these findings align with what tends to be seen the extensive behavioral coding literature on the Face-to-Face Still-Face paradigm.

**Supplementary material.** The supplementary material for this article can be found at https://doi.org/10.1017/S0954579421001553

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