Gene – Environment Contributions to the Development of Infant Vagal Reactivity: The Interaction of Dopamine and Maternal Sensitivity

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Abstract:
This study investigated dopamine receptor genes (DRD2 and DRD4) and maternal sensitivity as predictors of infant respiratory sinus arrhythmia (RSA) and RSA reactivity, purported indices of vagal tone and vagal regulation, in a challenge task at 3, 6, and 12 months in 173 infant – mother dyads. Hierarchical linear modeling (HLM) revealed that at 3 and 6 months, RSA withdrawal in response to maternal separation was greater (suggesting expected physiological regulation) in infants without the DRD2 risk allele than those with the risk allele. At 12 months, infants with the risk allele who were also exposed to maternal sensitivity showed levels of RSA withdrawal comparable to infants who were not at genetic risk. Findings demonstrate the importance of developmental analysis of gene – environment interaction.

Article:
Respiratory sinus arrhythmia (RSA), indexing the effect of the parasympathetic nervous system on the heart, is a psychophysiological marker for the regulation of arousal, state, and reactivity underlying individual differences in emotion, behavior, and personality in children (Fox & Stifter, 1989; Porges, 1991; Stifter, 1995; Stifter, Fox, & Porges, 1989). Although there are unresolved controversies in the field regarding the measurement of vagal tone (i.e., adjusting for respiration) and how measures of RSA should be interpreted (e.g., Denver, Reed, & Porges, 2007; Grossman & Taylor, 2007), the current study will use the term RSA as an index of cardiac vagal tone and RSA reactivity to challenge as an index of vagal regulation (see Porges, 2007), acknowledging the imperfect measures of vagal functioning that these indices provide.

Due to considerable individual variation in RSA functioning (RSA and RSA reactivity) and their links to behavioral and clinical outcomes, it is critical to understand better the factors that influence their development. Although several recent studies have examined RSA and RSA reactivity as the product of environmental factors (Burgess, Marshall, Rubin, & Fox, 2003; Calkins, Smith, Gill, & Johnson, 1998; Moore & Calkins, 2004; Porter, 2003; Porter, Wouden-Miller, Silva, & Porter, 2003), there has been almost no research on potential genetic correlates, although there is research suggesting some heritability for RSA reactivity (see below). Therefore, the primary goal of this study was to explore how specific genes may interact with parenting to influence development of infant RSA reactivity. This is consistent with the recent discussion of gene – environment interactions by Moffitt, Caspi, and Rutter (2006, p. 9) that suggests testing should be theoretically driven beginning with “plausible triads” (i.e., gene, environmental factor, behavioral phenotype). Although little is known about candidate genes that contribute to RSA, there is a small literature linking dopamine genes and general cardiovascular function that may be relevant, a larger body of research linking specific dopamine genes with certain behavioral phenotypes (e.g., Amenta, Ricci, Rossodivita, Avola, & Tayebati, 2001; Yeh et al., 2006) that are, in turn, related to infant RSA functioning, and research linking infant RSA functioning to parenting (reviewed below). Based on this indirect evidence, we tested a theoretical relation between infants’

RSA functioning (RSA and RSA reactivity) and candidate genes involved in regulating dopaminergic response (DRD2 and DRD4) and the moderating effect that parenting may have on this relation.

To support the proposed theoretical model of gene –environment interaction, first we review the literature that links RSA functioning, parenting, and infant regulation related to social behavior. Next, we discuss the literature on genetics of vagal tone and cardiovascular functioning. Third, we review research linking the dopamine receptor genes DRD2 and DRD4 to phenotypes related to behavioral regulation.

RSA and Vagal Tone

Parasympathetic control over cardiac functioning (i.e., vagal tone) is related to self-regulation, temperament, affect, and attention (Bornstein & Suess, 2000). The term vagal tone refers to control of the heart via the vagus nerve and is typically measured as the amplitude of RSA (i.e., beat-to-beat heart period [HP] associated with respiration), a parasympathetic index of heart rate variability (Porges, 1996; Porges & Byrne, 1992) and underlying regulatory abilities in mammals (Porges, 1996). Baseline vagal tone is considered to be a stable neurophysiological mechanism underlying autonomic and behavioral reactivity that provides a measure of resting state in the absence of environ-mental challenge. In the extant literature, RSA is commonly equated with vagal tone; however, it is important to remember that this is only one of its components and that there are many other influences on heart rate variability (Grossman & Taylor, 2007). Of these various components, however, RSA has been the most consistently examined in relation to dimensions of behavioral functioning in infants. For example, higher resting RSA during infancy has been associated with less temperamental difficulty (Stifter & Fox, 1990), greater sustained visual attention (Richards & Cronise, 2000), secure attachment (Izard et al., 1991), more sociable and explorative behavior (Fox, 1989; Stifter et al., 1989), and greater behavioral reactivity (Porges, Doussard-Roosevelt, Portales, & Suess, 1994).

Porges's (2007) polyvagal theory of social engagement asserts that the autonomic nervous system enhances restoration and growth by regulating the "vagal brake," which slows down the heart (i.e., activated vagal tone) during situations that do not present a challenge. However, during times of environmental stress, these internal processes are disrupted and the autonomic nervous system increases metabolic output to deal with external demands. A decrease in RSA (or RSA withdrawal) typically occurs when an individual is involved in an activity that requires active coping (Porges 1991, 1996), at which time the vagal "brake" is withdrawn (i.e., vagal tone is inhibited) to support an increase in heart rate. When environmental demands have ceased, the brake is reengaged (i.e., vagal tone is activated) to promote decreases in metabolic output and a return to a calm state. Thus, effective vagal functioning has been related to the ability to maintain homeostasis in the face of changing demands by allowing a shift from attention on internal demands to external ones that include the use of coping strategies to regulate affective or behavioral arousal.

Withdrawal of RSA during a challenging situation has been related to positive outcomes in infancy such as higher soothability (Huffman et al., 1998), more attentional control (Huffman et al., 1998; Suess, Porges, & Plude, 1994), and better emotion regulation (Calkins, 1997; Porges, Doussard-Roosevelt, Portales, & Greenspan, 1996). Furthermore, individual differences in RSA functioning have been examined as an index of characteristics such as aggression (Mezzacappa et al., 1997) and hostility (Sloan et al., 1994) in adolescents and adults. Although supporting data (Bazhenova, Plonskaia, & Porges, 2001; Moore & Calkins, 2004) are not conclusive, this research has led to the assumption that greater RSA withdrawal during a challenge context reflects more effective regulation.

Influence of early parenting on RSA. Due to the almost complete reliance on caregivers in assisting with both physiological and behavioral regulation during infancy, improvement in self-organization of these processes emerges with age (Spangler & Grossmann, 1993; Spangler, Schiechle, Ilg, Maier, & Ackerman, 1994). The way in which caregivers respond to the needs of their infants may have an impact on developing adaptive methods to modulate physiological stress (Derryberry & Rothbart, 1984) by infants' progressively internalizing the regulation strategies used within the dyad during the earlier months (Calkins et al., 1998; Thompson, 1994). RSA functioning, therefore, is a potential mediator of the relation between parenting and changes in infant emotional reactivity and regulation across development (see review by Propper & Moore, 2006). A number of studies have explored developing patterns of RSA functioning as the result of qualities of the infant – caregiver relationship and found that these qualities are, indeed, related to physiological functioning. Infants of dyads that spent more time in joint communicative sequences with continuous adjustment of behaviors in response to their partners, allowing for a range of emotional experience, had higher resting RSA than those in dyads that did not display this dyadic regulation (Porter, 2003). Moore and Calkins (2004) found that as early as 3 months of age, infants of dyads exhibiting lower synchrony showed higher physiological reactivity during a nor-mal play episode, less RSA withdrawal during a situation meant to elicit distress, and more difficulty returning to previous levels of reactivity and RSA following distress. Yet another study found that infants of more responsive parents displayed greater regulation of heart rate (RSA was not assessed) during a challenge task than infants of less responsive parents (Haley & Stansbury, 2003).

A recent study using the same sample as in the current report examined the link between infant– mother attachment relationships and infant change in RSA during the Strange Situation Procedure (SSP; Hill et al., 2008). Findings revealed that infants classified as insecure – avoidant displayed greater cardiac arousal and RSA withdrawal during separation and reunion episodes of the procedure than did infants classified as securely attached. This finding suggests that these infants may have been experiencing high levels of internal distress while minimizing behavioral displays of their distress. These infants appeared to recruit internal resources to actively cope with an external challenge and, therefore, may have relied on self-regulation to a greater degree than other infants, who may have relied more on their mothers. These findings are consistent with earlier discussions of the attachment relationship in terms of emotion regulation strategies (Cassidy, 1994). Cassidy suggested that the attachment relationship influences, over time, the behavioral strategies used by infants to successfully interact and respond to stress within that relationship. Infants classified as insecure – avoidant may show a behavioral pattern in response to a stressful situation (i.e., separation from mother) that minimizes their emotional expressions, perhaps because they have learned from experience to rely less on the attachment relationship as a source of comfort (Belsky, Rovine, & Taylor, 1984; Main & Solomon, 1986).

Genetics of RSA. Only a handful of infant studies have examined the associations of RSA functioning among family members. Healy (1992) studied mono-zygotic and dizygotic twins between the ages of 11 and 35 months and found no evidence of heritability of baseline RSA. Similarly, Bornstein and Suess (2000) looked at the relation between mother and child baseline RSA at 2 months and at 5 years of age and found no concordance within dyads. They did find, however, that RSA reactivity (measured as the base-line-to-challenge task difference) was concordant between mother and child at both time points, increasing from 2 months of age ($r = .23$) to 5 years of age ($r = .42$), suggesting an increase in some shared style of physiological response to environmental challenges within the dyad over time. Through repeated inter-actions, the mother's behavioral approach, or affective style, may transmit to her offspring a characteristic style of autonomic responsiveness. Additionally, genetic differences may contribute to observed changes in cardiovascular regulation and may be more pronounced during conditions of stress or challenge (Boomsma, Snieder, de Geus, & van Doornen, 1998; Kupper et al., 2005).

Support for this genetic hypothesis has been found in several behavioral genetics studies. Twin studies have reported RSA heritability estimates ranging from 13% to 55% (Boomsma, van Baal, & Orlebeke, 1990; Kupper et al., 2005; Singh et al., 1999; Sinnreich, Friedlander, Luria, Sapoznikov, & Kark, 1999; Voss et al., 1996). Of note, two twin studies found an increase in genetic variance associated with RSA during a challenging situation versus RSA at rest. The first one found that roughly 50% of the variance in RSA measured in adolescence was explained by genetic factors during two challenge tasks (i.e., reaction time and mental arithmetic), whereas only 25% of the variance was explained by genetic factors during a resting period (Boomsma et al., 1990). Similarly, a study of middle-aged twins found that heritability of RSA was 35% and 43% during two comparable challenge tasks (i.e., reaction time and mental arithmetic) and decreased to approximately 31% at rest (Snieder, Boomsma, Van Doornen, & De Geus, 1997). Results of these studies support the conclusion that there is genetic influence on RSA that may be more marked during a challenge.

However, heritability estimates do not tell us any-thing about the specific genes that contribute to RSA functioning or in what way they do so. Only a small number of studies have examined specific genes as potential contributors to measures of cardiac functioning, most are animal models, and none of which we are aware has examined relations between specific genes and RSA. The results of the current study may provide a targeted direction for examining this relation. In addition, to date, research examining associations between specific genes and cardiovascular functioning has proceeded primarily from a health psychology approach, selecting candidate genes based on their association with disease conditions or other aspects of biological functioning. As outlined earlier, our approach was to select candidate genes based on association with behavioral phenotypes of interest. In addition, innovative prior research on the genetics of cardiovascular functioning has been conducted with samples ranging in age from adolescence to adulthood, without consideration of genetic contributions to the developing vagal system during infancy. Furthermore, in the extant literature, there is a lack of attention given to experiential factors, which may interact with genetic effects to better predict physiological outcomes (Gottlieb, 1998, 2003). The current study aimed to explore the dopamine receptor genes DRD2 and DRD4 that were selected on the basis of associations with behavioral phenotypes related to RSA functioning as potential predictors of RSA functioning in infancy and to examine possible interactions between these genes and infants' experience with parenting sensitivity across early infancy.

In addition to association with behavioral phenotypes, DRD2 and DRD4 are implicated in biological mechanisms of cardiovascular functioning. Dopamine regulates cardiovascular functioning by acting on the central and peripheral nervous systems, vascular smooth muscles, the heart, and the kidneys (Jose, Eisner, & Felder, 1999). Five distinct dopamine receptor genes have been identified and classified into two groups known as the "D1-like" super family (D1 and D5 receptors) and the "D2-like" super family (D2, D3, and D4 receptors; see Hyde, Knable, & Murray, 1996), each with its own molecular structure, mRNA coding and anatomical distribution, and chromosomal location (reviewed in Sibley & Monsma, 1992). The D2-like family of dopamine receptors has consistently been linked to the central nervous system regulation of blood pressure (Amenta et al., 2001; Bek, Eisner, Felder, & Jose, 2001; Jose, Eisner, & Felder, 1998; Yeh et al., 2006) and hypertension (Amenta, et al., 2001; Linthorst, van Giersbergen, Gras, Versteeg, & De Jong, 1994; Vaughan, van den Buuse, & Roland, 1999). There are a significant number of D2-like receptors found in the vagal complex (Hyde et al., 1996), suggesting that the DRD2 and DRD4 receptors are an important component of neural mechanisms regulating visceral function, including the cardiovascular system. Nevertheless, considerable research remains to be done to specify functional relations between these genes and RSA functioning in humans.

Dopamine Genes and Behavior

DRD2. Research on the associations of DRD2 and behavioral characteristics has been limited to adolescence and adulthood, with no studies done in infancy. Variations in DRD2 have been related to several dimensions of adult personality, including novelty seeking, sensation seeking, and aggressiveness (Cloninger, Adolfsson, & Svrakic, 1996; Noble et al., 1998). The risk allele of DRD2 (i.e., minor Taq1 A1 allele) has been related to high novelty seeking scores (Noble et al., 1998; Suhara et al., 2001). Possessing the Taq1 A1 allele of DRD2 has also been suggested as a risk factor for conduct disorder (Comings et al., 1996; Lu, Lee, Ko, & Lin, 2001), which is not surprising considering its association with impulsivity (Wiers, Sergeant, & Gunning, 1994) and disinhibition (McGue, Slutske, Taylor, & Iacono, 1997). Furthermore, several types of sub-stance abuse have been associated with the Taq1 A1 allele of DRD2, including alcoholism (Blum et al., 1991; Noble et al., 1994), nicotine (Comings et al., 1996; Noble et al., 1994), and opiate dependence (Lawford et al., 2000). Finally, this allele of DRD2 has also been associated with other addictive behaviors such as pathological gambling (e.g., Comings et al., 1996). These results are consistent with expectations, as the Taq1 A1 allele of DRD2 has been reported to be associated with a decrease in D2 dopamine receptor availability (Pohjalainen et al., 1998) and the number of receptor binding sites is lowest in A1 homozygotes. Therefore, given that D2 dopamine receptors have an inhibitory function in the dopaminergic system, a lower availability of these receptors may translate into less behavioral control.

DRD4. The DRD4 gene contains a repeated sequence polymorphism within its coding sequences that changes the length of the receptor protein that has been shown to have a moderate functional significance (Asghari et al., 1994). Individuals with longer versions of the polymorphism (L-DRD4; six to eight repeats) have significantly higher novelty seeking than those with the shorter version (S-DRD4; two to five repeats); the shorter the allele, the more efficient it is in binding dopamine (Plomin & Rutter, 1998), suggesting that the L-DRD4 allele is a risk allele. Research on the associations of DRD4 and behavioral characteristics has been conducted with adult and adolescent samples and more recently with infants and young children. Variations in DRD4 have been related to dimensions of adult personality such as novelty seeking, characterized by excitability, impulsiveness, and high exploratory behavior (Benjamin et al., 1996; Ebstein, Nemanov, Klotz, Gritsenko, & Belmaker, 1997; Noble et al., 1998), cigarette smoking (Shields et al., 1998), pathological gambling (Pérez de Castro, Ibàñez, Torres, Sáiz-Ruiz, & Fernández-Piqueras, 1997), and alcoholism (George, Cheng, Nguyen, Israel, & O'Dowd, 1993). L-DRD4 in 4- year-old children was associated with aggression as reported by mothers (Schmidt, Fox, Rubin, Hu, & Hamer, 2002). In 3-year-old children, L-DRD4 was associated with lower intensity of emotional reactions such as happiness, anger, and sadness than S-DRD4 (DeLuca et al., 2001). Infants at 12 months of age with the L-DRD4 allele showed less anger-related negative emotionality than did infants with the S-DRD4 allele in an anger-inducing task (Auerbach, Faroy, Ebstein, Kahana, & Levine, 2001). Earlier we discussed that individuals who fail to show expected RSA withdrawal in response to challenge situations may not be as reactive to those situations as most infants; infants with the L-DRD4 may be just such an example.

In summary, the DRD2 and DRD4 dopamine receptor genes are associated with personality characteristics and clinical disorders that are impulsive, reward seeking, and addictive in nature. These characteristics, overall, appear to be related to a lack of regulatory ability and behavioral control, suggesting an association between dopamine and RSA, which is also highly associated with regulatory ability and behavioral control. It is important to note, however, as with most direct gene –behavior associations, many of the earlier mentioned relationships have failed to replicate (e.g., Gebhardt et al., 2000; Gelernter et al., 1997; Jönsson et al., 1998). This failure to replicate may be due to differences in sample composition, such as ethnicity or sex, or to differences in phenotype measurements. However, of interest in the current study is the possibility that it is due to the lack of research attention to environmental factors.

Goals of the Current Study

Although studies have examined the influence of parenting on the development of RSA functioning (baseline RSA and RSA reactivity to challenge), thus far, little research has examined the genetic underpinnings of this process. Research has suggested a relationship between the DRD2 and DRD4 genes and characteristics that are related to impulsivity and behavioral dysregulation. Similarly, studies of RSA functioning have found associations between expected RSA withdrawal in response to challenge and better behavioral regulation. Thus, the current study used a theoretically driven approach to examine the association between these genes and RSA functioning. To explore possible gene –environment mechanisms that may influence the development of RSA functioning across early infancy, this study examined the interaction between these genes and early maternal parenting sensitivity as precursors of later RSA functioning.

Method

Participants

Participants were drawn from the Durham Child Health and Development Study (DCHDS), a longitudinal sample consisting of 206 healthy, full-term infants who were followed from 3 months to 3 years of age. Families were recruited from a largely urban community via fliers and postings at birth and parenting classes, as well as through phone contact via birth records. Approximately equal numbers of European American (EA) and African American (AA) families were sampled from low- and high-income groups. Infant's race was determined by the mother (or primary caregiver); income status was assessed based on the size of the family in relation to their household income in accordance with the 2002 Federal Poverty Guidelines. Demographic information was collected during the first visit at 3 months of infant age and was updated at each subsequent visit.

Infant –mother dyads from the DCHDS were seen within a week of infants' 3-, 6-, and 12-month birth-days. Dyads that completed the mother –infant free-play task at the 6-month visit (N = 173) made up the initial sample for this report. However, only those dyads with infants that had complete heart rate data at any of the three visits, as well as complete DRD2 genetic data, were included in analyses (N = 142). The final sample for analyses consisted of 67 males and 75 females. Seventy-two mothers reported their infants to be AA and 70 EA. At recruitment, the mean age of mothers was 27.8 (SD = 5.63, range = 18 – 40 years). Fifty-one percent of mothers reported having some college education, 49% had a high school education or some vocational training, and 44% of mothers were currently employed. Sixty-nine families reported in-comes that were classified as below poverty and 73 as above poverty. Of the 142 participants with complete DRD2 data, 11 fewer participants had complete DRD4 data. Those with incomplete DRD4 data did not differ by ethnicity or income.

Missing Data

Thirty-three of the dyads recruited at 3 months of infant age did not have complete data for the 6-month free-play task due to attrition (N = 27) or problems with coding due to video quality (N = 6). An additional 15 infants were missing genetic data due to difficulties with genotyping (N = 8) or refusal to provide a cheek cell sample (N = 7). Of the remaining infants, 18 were missing heart rate data at each of the three assessments. Eighty-three dyads had cardiac data at only one time point, 76 dyads had cardiac data at two time points, and 31 dyads had complete cardiac data across all three of the time points. The reasons for the missing data at each assessment, as well as the number of infants missing data per visit, are: heart rate monitor experienced problems or failure (3 months = 5,12 months = 2), collected data contained too many artifacts to use due to movement or removal of equipment by infant (3 months = 10, 6 months = 14, 12 months = 7), infant could not finish the task due to fussiness or extreme upset (3 months = 3, 12 months = 2), or the dyad did not complete the visit or task (6 months = 4, 12 months = 7). No systematic differences associated with missing data by ethnicity or income group were found.

Procedure

Visits were conducted both at the family's home and in the lab. At each visit, infants and their mothers participated in several joint and individual tasks followed by a standardized interview and completion of demographic questionnaires by mother. Although assessments were done every 3 – 6 months until the child reached 36 months of age, the present study examined data only from visits that occurred when infants were 3, 6, and 12 months old when observational tasks designed to assess infants' responses to maternal separation were conducted. These visits were targeted because although the mother –infant separation tasks differed between early infancy and age 12 months, they each provided an age-appropriate standardized assessment of infants' responses to withdrawal of maternal attention, thus allowing us to compare infants' physiological regulation in response to a typical, developmentally appropriate stressor. All tasks were videotaped for later coding.

Measures

Mother–infant free play (6-month home visit). To evaluate maternal behavior during interactions with infants, mothers were provided a standard set of toys and instructed to interact with their children as they normally would if playing during some free time on a typical day. The task lasted 10 min and was video-taped for later coding.

Free-play interactions were coded by two independent coders who were unaware of the study's hypotheses. From these observations, seven subscales of maternal behavior were coded (sensitive responsiveness, intrusiveness, detachment, positive regard, negative regard, stimulation of cognitive development, and animation). Coders were trained to reliability until interclass correlation coefficients of .80 or greater were established and maintained with criterion coders (and for each individual pair of coders). All interactions were double coded and final scores were agreed on by conferencing. An overall maternal sensitivity composite was created (guided by factor analyses) by aggregating the scores for five of the subscales, including sensitivity – responsiveness, positive regard stimulation of development, animation, and detachment – disengagement (reverse scored). Similar composite scores for maternal sensitivity have been used by the National Institute of Child Health and Human Development Study of Early Child Care (1997), the Family Life Project (Blair, Granger, Willoughby, Kivlighan, & the Family Life Project Key Investigators, 2006), and other reports based on the current DCHDS sample (Mills-Koonce et al., 2007).

Mother – infant still-face procedure (3-month home visit and 6-month laboratory visit). To assess infants' responses to maternal separation, the mother –child dyad participated in the still-face procedure (SFP; Tronick, Als, Adamson, Wise, & Brazelton, 1978). Mothers placed their children in an infant seat situated on a table and then sat on a chair directly in front of them. The normal play episode consisted of mothers playing with their babies as they normally would. After this 2-min episode, mothers were told to turn away from their infants for 15 s, after which time they turned back and began the still-face episode. During the still-face episode, mothers were asked to look at their children for 2 min without any facial movements or vocalizations. Mothers were then asked to turn away from their infants for another 15 s, which was followed by a 2-min reunion episode in which mothers were told to respond to their infants in any way that they felt was appropriate. Only HP and RSA data from the still-face episode of the SFP were used for the current analyses. During this time, infants experienced an episode of withdrawal of maternal attention, which requires them to regulate their own physiological arousal without the help of their mothers. We focused specifically on this episode of the procedure because it was individual, rather than dyadic, level of regulation that was of interest in the current study and because the episode was considered most analogous to the maternal separation task at 12 months.

Strange situation procedure (12-month laboratory visit). To assess infants' responses to maternal separation at 12 months, mothers and their infants were observed in the strange situation procedure (SSP; Ainsworth, Blehar, Waters, & Wall, 1978). Throughout the procedure, infants were exposed to eight brief episodes of increasing stress, including two mother –infant separations and reunions. The current study focuses on the first episode of mother –infant separation (fourth episode) at which time mothers leave the room and the infant is alone with a stranger for 3 min. Only HP and RSA data from this first separation episode of the SSP were used for the current analyses because the second episode of separation (sixth episode) elicited high levels of distress from almost all infants, creating a possible ceiling effect.

Cardiac monitoring. At the start of each visit, the experimenter placed two disposable pediatric electro-des on the infant's chest while he or she was seated in a baby seat or mother's lap. The electrodes were connected to a preamplifier, from which the output was transmitted to a heart interbeat interval (IBI) monitor (Mini Logger; Mini-Mitter/Respironics, Bend, OR) for R-wave detection. The infant wore a smock with a large pocket in which the monitor was placed. Once the monitor was securely in place and the infant was acclimated, an electronic signal was sent to the monitor by the experimenter (by manually pressing a button on the monitor) in order to mark the start of a 2- to 4-min baseline measure of IBI activity. During this period of time, mothers were asked not to interact with, or provide toys to, their infants so that stimulation was minimized and infants' IBI could be measured accurately during a neutral and calm state. At the completion of baseline, another electronic signal was used to mark the end of this episode. IBI data were continuously collected during the rest of the visit, with electronic signals to the monitor to mark the start and end of each task. During the SFP and the SSP, electronic signals marked the start and end of each episode.

Buccal cell collection (12-month laboratory visit). DNA was obtained through the collection of infant buccal cells (i.e., cheek cells). The experimenter put on latex gloves before handling any supplies and rubbed the inside of the infants' inner cheek and gums for 20 s with a Q-tip. The Q-tip was then immediately placed into a pint-sized zip-loc bag, sealed, and put into a storage freezer where it remained until sent to the laboratory for processing. Cheek cells for DNA isolation and analysis were sent to a genetics laboratory at North Carolina State University, Raleigh, NC. All the genotyping was done blind to the study's hypotheses and outcomes.

Data Reduction

Computation of RSA and HP. At the completion of each visit, a data file containing infant IBI data for the period of collection was transferred from the monitor to a computer in the laboratory. Such activities as bodily movements, infants tugging on electrodes, physical force to the monitor, and other such disruptions may affect IBI collection by recording artifactual points within the cardiac data. Thus, the IBI data files were edited and analyzed using Mxedit software (Delta Biometrics, Bethesda, MD) by two Mxedit reliable researchers. Editing the files consisted of scanning the data for outlier points relative to adjacent data and replacing those points by dividing them or summing them so that they would be consistent with the surrounding data. Due to difficulties in collecting cardiac data from infants of this age (i.e., pulling on electrodes, equipment failure), only participants who had full and sufficient data with less than 10% editing were used in the current analyses.

The present study used Porges' (U.S. Patent No. 4,510,944, 1985) method of calculating RSA and HP. The estimate of RSA is reported in units of ln(ms)2. RSA was calculated every 15 s for the baseline period and for each episode of the SFP and SSP. These epoch durations are typical for studies of short-duration tasks (Huffman et al., 1998). The mean RSA of the 15-s epochs within each episode was used in subsequent analyses. Larger values of HP indicate slower heart rate and larger values of RSA suggest greater vagal tone.

Computation of change in RSA and HP. To assess RSA reactivity, change in RSA (ΔRSA) was measured as the difference between baseline RSA and RSA during the still-face episode of the SFP at 3 and 6 months and the difference between baseline RSA and RSA during the fourth episode of the SSP at 12 months. Following previous research (Calkins, 1997; Moore & Calkins, 2004), difference scores were computed by subtracting episode RSA from baseline RSA, such that sign indicated direction of change, with positive values indicating greater RSA withdrawal (the expected response) and negative values indicating an increase in RSA during the challenge situation. Change in HP (ΔHP) was measured in the same way as ΔRSA (baseline HP minus episode HP). Positive values of HP indicated an increase in heart rate (the expected response) during the challenge situation and negative values indicated a decrease in heart rate. This method provided an index of change relative to each infant's baseline level.

Researchers have used both difference score and residual score methods to assess change in RSA. In the current data set, correlations between residual scores and difference scores for infants' RSA and HP ranged from.96 to.99. However, due to concerns that the use of difference scores might underestimate the effect between ΔRSA and infant genotype, we ran analyses using both difference scores and residualized scores and obtained the same results. Because the two scores have slightly different meanings, with residual scores indicating change if all participants started out equal in terms of baseline RSA and difference scores indicating raw change from baseline-to-challenge context, we chose the difference score method, as we believed that was most consistent with our research question.

Genotyping. Genomic DNA was extracted from each salivary sample using the Puregene DNA extraction kit by following the manufacturer's protocol for DNA isolation from 1 ml of body fluid. Saliva samples yielded DNA in adequate quantities for genotyping (approximately 200 μg/ml). Genotyping of the 48-bp repeat in Exon III of the DRD4 gene was performed as previously described (Propper, Willoughby, Halpern, Cox, & Carbone, 2007). The genotypes s/s, s/l, or l/l were assigned to each individual where s is the short allele and l is the long allele. Based on previous results (Anchordoquy, McGeary, Liu, Krauter, & Smolen, 2003; Benjamin et al., 1996; Schmidt et al., 2002), polymorphisms made up of homogeneous short alleles (s/s) were classified as Short (S-DRD4) and heterogeneous polymorphisms (s/l and l/l) were classified as Long (L-DRD4).

Genotyping of the DRD2 gene was performed by polymerase chain reaction amplification using the forward and reverse primers: 5'-ccgtcgacggctggc-caagttgtcta-3' (D2F1) and 5'-ccgtcgacccttcctgagtgt-catca-3' (D2R1; Miyake et al., 1999). The amplicon was subsequently digested with the restriction enzyme, *Taqα*1 (New England Biolabs, MA). This results in digestion products of the A1 allele (310 bp) and the A2 allele (180 + 130 bp). The allele status of DRD2 A_1^+ (A_1/A_1 and A_1/A_2 genotypes) and A_1^- (A_2/ A_2 genotype) were assigned to each individual based on previous studies (see Noble, 2003).

Results

Descriptive Statistics and Bivariate Relations Among Covariates

Forty-six percent of the sample carried the risk A_1^+ allele of DRD2, and 36% of the sample carried the risk L-DRD4 allele of DRD4. There was a significant difference in distributions of DRD2 polymorphisms as a function of ethnicity, with AA infants more likely to have the risk A_1^+ allele of DRD2 than EA infants, $\chi^2(1) = 3.99$, $p < .05$. This population stratification is consistent with existing research (Barr & Kidd, 1993). The impact of genetic differences by ethnicity, and the most appropriate way to address it in re-search designs, is currently the source of ongoing debate (Hoggart et al., 2003; Thomas & Witte, 2002; Wacholder, Rothman, & Caporaso, 2002). Because ethnicity in this study was measured by participants' self-report and individuals often are unaware of their full genetic heritage, making the measure of ethnicity subject to error, and because analyzing the groups separately would substantially attenuate power, the current analyses examined the two ethnic groups together but included ethnicity as a covariate in the model. There was no significant difference in the distribution of the DRD4 polymorphisms as a function of ethnicity.

Maternal sensitivity was significantly higher in EA families, $t(170) = 4.08, p < .001$ (see Table 1) and higher income families, $t(170) = 6.10$, $p < .001$. AA infants exhibited a lower mean value of ARSA, reflecting less RSA withdrawal at3 months, $t(145) = 2.23, p < .05$, and 6 months, $t(75) = 4.01$, $p < .05$, during the still-face episode of the SFP, and they had higher baseline RSA, on average, at 12 months, $t(99) = 3.00$, $p < .01$.

On average, infants with the DRD2 risk allele (A_1^+) had mothers who were less sensitive than infants without the risk allele (A_1^-), $t(154) = 2.10$, $p < .05$ (see Table 1). Infants with the A_1^+ allele exhibited lower values of ΔRSA at3 months, $t(128) = 2.92, p < .01$,and 6 months, $t(71) = 2.69$, $p < .01$, during the still-face episode than those without the risk allele (A_1^-). No mean differences were observed for maternal sensitivity or vagal functioning at any time point. As seen in Table 2, baseline RSA was stable over time, although ΔRSA was not. Baseline RSA was related concurrently to ΔRSA at 3 and 6 months but not at 12 months. Maternal sensitivity was uncorrelated with physiological variables.

Table 1

Means and Standard Deviations of Variables by Ethnicity and DRD2 *Polymorphisms*

	AA	EA		A_2/A_2 (A_1^-)	A_1/A_1 and A_1/A_2 (A_1^+)	
	M (*SD*)	*M* (*SD*)	Cohen's *d*	*M* (*SD*)	*M* (*SD*)	Cohen's *d*
Maternal sensitivity	9.38*** (4.05)	11.84*** (3.67)	.63	11.01*** (4.10)	9.65*** (4.05)	.34
Baseline RSA at 3 months	3.42 (0.95)	3.43 (1.03)		3.41 (0.92)	3.48 (1.07)	
Baseline RSA at 6 months	3.78 (0.95)	3.57 (0.80)		3.55 (0.70)	3.81 (1.08)	
Baseline RSA at 12 months	4.00** (1.00)	3.41** (0.88)	.60	3.68 (0.90)	3.65 (0.88)	
ΔRSA at 3 months	0.35* (0.90)	0.66* (0.75)	.37	0.68** (0.80)	0.26** (0.87)	.52
ΔRSA at 6 months	−0.19* (0.39)	0.39* (0.76)	.96	0.38** (0.70)	−0.08** (0.76)	.64
ΔRSA at 12 months	0.19 (0.60)	0.12 (0.70)		0.21 (0.65)	0.12 (0.64)	

Note. HP = heart period; RSA = respiratory sinus arrhythmia.
*$p < .05$. **$p < .01$. ***$p < .001$.

Model Specification

HLM were estimated using SAS proc mixed in order to examine changes in children's vagal functioning (baseline RSA, ΔRSA) over three time points from 3 to 12 months. This procedure allowed for the control of the nonindependence of observations due to the same individuals being repeatedly assessed over time. An HLM approach also accounts for missing-at-random outcome data, allowing the use of all available data for the outcome of interest (Little & Rubin, 1987; Raudenbush & Bryk, 2002). Restricted maximum likelihood was used in reporting model parameters, and degrees of freedom were estimated using the Satterthwaite method. Because of the correlations between demographic factors and model parameters, ethnicity and income were included in all HLM. Models were estimated for DRD2 and DRD4 separately. There were no significant findings for DRD4, so the following reports results from the DRD2 analyses only.

Preliminary Analyses: Vagal Functioning Over Time

Trajectories for levels of baseline RSA and levels of ΔRSA were first analyzed to assess normative change over time in these components of RSA functioning. Controlling for ethnicity and income, mean levels of baseline RSA increased from 3 to 12 months of child age by 0.25 SD (based on the distribution at 3 months of age), $F(2,173) = 4.50$, $p < .05$. Mean levels of ΔRSA during the challenge task decreased from 3 to 12 months of child age, $F(2,135) = 7.84$, $p < .001$. Because of the change in protocol to accommodate develop-mental changes in infants during the 1st year of life, it is not possible to determine whether changes in ΔRSA over time are due to child development or due to methodological artifact. Therefore, the following analyses of ΔRSA were conducted using z scores for ΔRSA rather than raw values. This approach is consistent with the main goal of the current research to analyze the effects of genetic and environmental factors on vagal functioning at different points in time as compared to analyzing their effects on trajectories of vagal functioning.

Predictors of Child Vagal Functioning Across 3 to 12 Months

Baseline RSA. There were no effects of DRD2, maternal sensitivity, or their interaction on baseline RSA. There was a main effect of ethnicity, $F(1, 242) = 9.46$, $p < .05$, indicating that AA infants exhibited higher levels of baseline RSA. Consistent with descriptive analyses presented earlier, this effect was moderated by time, $F(2,135) = 3.90$, $p < .05$, such that only AA infants displayed increases in baseline RSA over time, $F(2, 184) = 5.86$, $p < .01$.

Table 2

Correlations Among Physiological Variables

	1	2	3	4	5	6	7	8	9	10	11	12
1 Baseline HP at 3 months	—	.13	.31**	.59***	.19	.13	.61***	.26*	.13	.26**	.02	.12
2 Baseline HP at 6 months		—	.26†	−.05	−.23*	.20	.06	.50***	.27†	−.05	.12	.13
3 Baseline HP at 12 months			—	.08	−.15	.10	.22*	.30*	.73***	−.02	−.20	.20†
4 ΔHP at 3 months				—	.26*	−.03	.41***	.10	−.02	.54***	.16	.09
5 ΔHP at 6 months					—	−.18	.02	.01	−.20	.03	.50***	−.15
6 ΔHP at 12 months						—	−.03	−.03	.05	−.09	−.13	.69***
7 Baseline RSA at 3 months							—	.41***	.36***	.33***	−.11	.05
8 Baseline RSA at 6 months								—	.62***	.09	.28*	.01
9 Baseline RSA at 12 months									—	−.12	−.16	.18†
10 ΔRSA at 3 months										—	.20	.03
11 ΔRSA at 6 months											—	−.06
12 ΔRSA at 12 months												—

Note. HP = heart period; RSA = respiratory sinus arrhythmia.
†$p < .10$. *$p < .05$. **$p < .01$. ***$p < .001$.

Change in RSA. There was a significant effect of DRD2 on ΔRSA, $F(1, 242) = 9.46$, $p <.01$, such that the risk A_1^+ allele was associated with lower levels of DRSA (i.e., less RSA withdrawal). This effect was moderated by a significant three-way interaction between DRD2, maternal sensitivity, and time, $F(2, 242) = 4.26$, $p < .05$. Probing this interaction revealed that at early ages, there were significant main effects of child genotype on ΔRSA. Children with the A_1^+ allele were observed to have lower ΔRSA, indicating less RSA withdrawal, than children without the risk allele at 3 months of age, $F(1, 107) = 6.02$, $p < .05$, and at 6 months of child age, $F(1, 63) = 4.23$, $p < .05$ (see Table 1). At 12 months of child age, a significant interaction between maternal sensitivity and child genotype was observed, $F(1, 39) = 10.02$, $p < .05$, $\eta^2 = .13$. Probing this interaction revealed that high maternal sensitivity was associated with greater ΔRSA, indicating greater RSA withdrawal for infants with the risk A_1^+ allele, $F(1, 33) = 5.29$, $p < .05$, $R^2 = .15$. As seen in Figure 1, the A_1^+ risk allele was associated with lower levels of ΔRSA at 3 and 6 months, and by 12 months, infants with highly sensitive mothers were observed to have comparable levels of ΔRSA regard-less of their DRD2 polymorphism. Cells means in Figure 1 are adjusted for covariates.

Given the exploratory nature of the current research, post hoc analyses were conducted to assess a possible confound between DRD2 and ethnicity. First, in the model analyzed earlier, when controlling for child genotype and maternal sensitivity, the main effect of ethnicity on DRSA (reported in the Descriptive Statistics section) was no longer significant. Second, the interaction between sensitivity and ethnicity was examined and found to be nonsignificant, $F(1, 242) = 3.04$, $p < .1$, $\eta^2 = .06$. Figure 2 illustrates the effect of maternal sensitivity on ΔRSA for children of different genotypes, separately by ethnic groups.

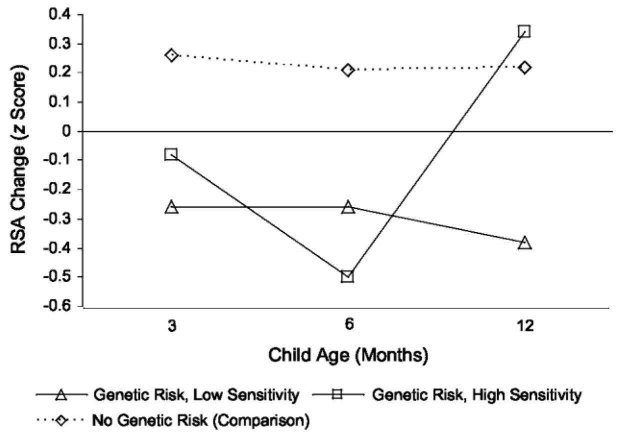

Figure 1. Maternal sensitivity moderates the relation between *DRD2* and infant change in respiratory sinus arrhythmia at 12 months.

Discussion

The primary goal of the present study was to explore genetic and environmental contributors to the development of RSA functioning related to social engagement across early infancy. The dopamine receptor genes DRD2 and DRD4 were selected on the basis of known associations with behavioral regulatory problems, which are, in turn, associated with infants' RSA functioning. The results of this study indicated that gene and environmental factors interacted in the development of RSA and RSA reactivity across 3– 12 months, highlighting the importance of develop-mental analyses of gene – environment interactions (Gottlieb & Halpern, 2002).

Infants carrying the A_1^+ allele of DRD2, which has been identified as a risk factor for problematic out-comes in adolescents and adults related to novelty seeking, aggressiveness, and impulse control disorders, showed significantly lower levels of RSA withdrawal in reaction to a task designed to be mildly distressing at 3 and 6 months of age. As RSA withdrawal in response to challenge is a purported indicator of effective physiological regulation, these findings suggest that the DRD2 genetic risk for behavioral disorders may be, in part, mediated by RSA functioning. Over time, however, maternal sensitivity moderated change in infant RSA reactivity to challenge. Although infants carrying the risk A_1^+ allele consistently exhibited less RSA withdrawal at 3 and 6 months, infants with the risk allele who were also exposed to sensitive maternal caregiving displayed an increase in RSA withdrawal in reaction to a distressing social situation by 12 months of age, reaching levels that were comparable to infants who were not at genetic risk.

RSA withdrawal in response to a distressing task is the typical response in infancy, and there is a body of research suggesting it indicates effective physiological regulation. Nevertheless, there are other interpretations of the current findings that cannot be ruled out. First is that infants who did not show RSA withdrawal were simply not distressed. In the current study, there is no way of accurately determining whether an infant did or did not become distressed by the procedures. Infants' behavioral and physiological indicators of distress are commonly uncorrelated in infancy (e.g., Gunnar, Mangelsdorf, Larson, & Hertsgaard, 1989; Weinberg & Tronick, 1996; cf. Bazhenova et al., 2001; Haley & Stansbury, 2003; Moore & Calkins, 2004). Measures of change in HP, which suggest arousal, were highly positively correlated with RSA reactivity, suggesting that infants who were more aroused showed greater RSA withdrawal. However, HP does not provide a measure of physiological arousal that is independent of parasympathetic influences. In future research, measures of sympathetic reactivity are needed, such as skin conductance, which has been used successfully to assess sympathetic reactivity in infants' responses (Ham & Tronick, 2006).

Second, an increase in RSA in response to a challenge task may indicate increased attention and is related to higher levels of exploratory behavior (DiPietro, Porges, & Uhly, 1992). Given that the risk A_1^+ allele of DRD2 has been found to be associated with novelty seeking and risk-taking behavior in adults, it may be that infants with this genetic profile found the challenge tasks to be novel and interesting rather than distressing.

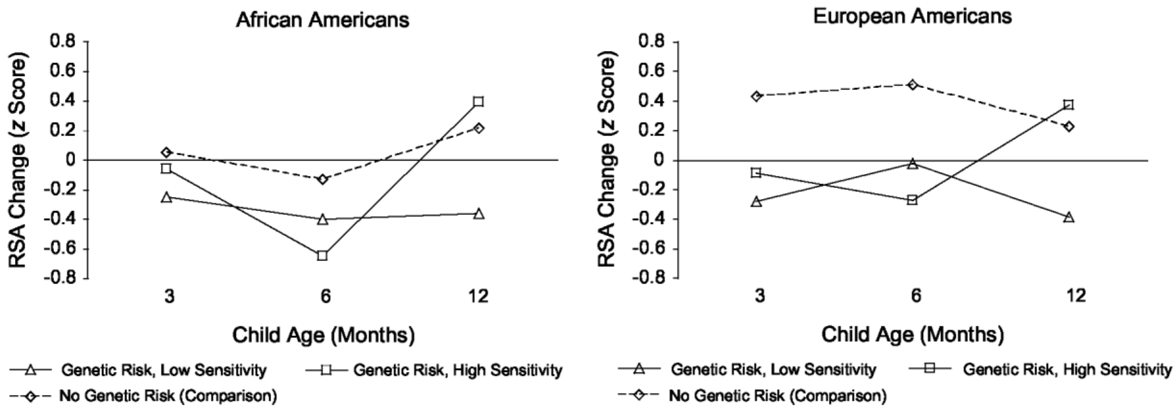

Figure 2. Interaction between maternal sensitivity and *DRD2* Risk Group by Ethnicity.

Third, in interpreting the interaction between genetic risk and maternal sensitivity over time, there are several possible explanations for the increase in RSA withdrawal seen for infants with the risk A_1^+ allele of DRD2 and highly sensitive mothers. One is that genetic risk carries an associated deficit in effective vagal functioning (indicated by a lack of or diminished RSA withdrawal to an expected stressor), but as a result of experience with a sensitive mother who presumably has provided support for the development of self-regulatory skills, these infants gradually acquired more effective physiological regulation by 12 months. Alternatively, these infants may already have had adequate physiological regulation but, until they accumulated sufficient experience with a highly sensitive mother, did not become distressed at separation from her. Analogously, their counter-parts in the genetic risk group who did not have highly sensitive mothers may either have had a deficit in effective vagal functioning or did not find the separation from mother distressing because of either an innate sense of novelty, as discussed earlier, or because it is not particularly distressing to be separated from a mother who is insensitive. This would be consistent with research finding that infants of depressed mothers were less likely to become distressed in response to mothers' still-face than other infants (Field, 1984).

It is also likely that, as with adults, there are individual differences in type of reactivity to stressors. Some individuals respond primarily with parasympathetic activation, some with primarily sympathetic activation, and some with activation of both systems. As stated earlier, additional measures of sympathetic reactivity are needed in future research to help to identify which of these many possible interpretations are most valid, and much larger sample sizes are needed to determine whether individual differences in patterns of autonomic reactivity can be identified in early infancy.

Although our theoretical model was based, in part, on associations between behavioral phenotypes and DRD2 and DRD4 genes, we found an association between infant RSA functioning and the DRD2 gene only. This could be a sample specific finding. Additional research needs to be done to replicate this finding as well as to understand better the biological basis of the functional relation between DRD2 and RSA functioning. Although a detailed discussion of the biological structure and function of dopamine receptors is beyond the scope of this article (see Missale, Nash, Robinson, Jaber, & Caron, 1998, for review), extant findings on the relationship between the dopaminergic system and cardiovascular functioning, mainly from animal models, may help explain the current findings. The "D2-like family" of dopamine receptors, which includes the DRD2 and DRD4 genes examined in the current study, has been found in the human vagal complex (Hyde et al., 1996), and dopamine receptors have been found in regions of the brain that are known to control cardiovascular function (van den Buuse, 1998), suggesting that they are an integral part of neural mechanisms that regulate cardiovascular function. However, the DRD2 gene specifically has been associated with hypertension in both humans (Thomas, Tomlinson, & Critchley, 2000) and rats (Linthorst, De Lang, De Jong, & Versteeg, 1991; Linthorst et al., 1994). Furthermore, Li et al. (1998) found hypertension in mice to be caused by increased activity of the adrenergic nervous system after disruption to the DRD2 gene (as cited in Bek et al., 2001), which suggests an indirect effect of dopamine on cardiac functioning. Injection of dopamine into the nucleus ambiguus of mice caused bradycardia (i.e., abnormally slow heart beat) due to the excitation of DRD2 receptors on vagal inhibitory neurons controlling heart rate (Chitravanshi & Calaresu, 1992). Taken together, this research suggests that the relation between DRD2 and RSA functioning found in the current study may reflect functional distinctions between the DRD2 and DRD4 receptor genes' effects on the vagal system. Our findings may also suggest that the relation between DRD2 and behavioral regulatory problems is mediated by RSA functioning, whereas the relation between DRD4 and behavioral regulatory problems is not or is related indirectly to RSA functioning.

Several methodological issues set an important context for interpreting the results. First, expected difficulties associated with obtaining valid cardiac data from young infants at three different time points resulted in a relatively small sample size. Second, because this was a longitudinal study, the challenge tasks in which we assessed infants' change in RSA needed to be developmentally appropriate. The challenge selected—maternal separation –is known to elicit stress responses from infants at various ages, thus remained consistent across age in the study. The context in which maternal separation occurred, how-ever, had to change to accommodate developmental changes in the degree of separation that infants would find stressful. Although the still-face paradigm and the SSP are different procedures, each presents the infant with an age-appropriate disruption in social interaction that involves a removal of maternal attention, and there is evidence to show that infants' responses to the still-face paradigm predict their responses to the SSP (e.g., Cohn, Campbell, & Ross, 1991), suggesting some functional equivalence between the two procedures. It is difficult to say with certainty that the task used at 12 months was eliciting the same emotional or physiological response from infants as the task used at 3 and 6 months, but this is a fundamental dilemma of developmental research.

Another methodological issue is the possibility that population stratification could have resulted in a confound, which can arise when the outcome of interest differs between ethnic populations for non-genetic reasons while the distribution of alleles also differs by ethnicity. In our sample, AA infants were significantly more likely to have the A_1^+ allele than EA infants, consistent with extant research on the frequency of the DRD2 A1 allele, where reported frequencies for EAs were found to be the lowest of 16 ethnic populations (Barr & Kidd, 1993). Although the issue of population stratification is an important one to consider, the degree to which this presents a problem remains controversial (Hoggart et al., 2003; Thomas & Witte, 2002), and reviews of the literature have found that the extent of population stratification bias has been exaggerated (Cardon & Palmer, 2003) and is likely to be small (Wacholder et al., 2002). Furthermore, in this study, ethnicity was assessed by mothers' self-report, and the sample was not likely to be accurately stratified in terms of genetic ancestry, especially in admixed populations where individuals may not be completely sure of their precise ancestry (Ziv & Burchard, 2003).

Exploring the possible confound suggested that, in terms of RSA reactivity, there was not a significant differential sensitivity to parenting for children of different ethnicities after controlling for genotype. Although not conclusive, this supports the interpretation of a gene – environment interaction across ethnic groups and suggests that ethnicity was not a confound. Ethnic differences found in other recent studies of genetic effects on physiological and behavioral outcomes (Propper et al., 2007; Williams et al., 2003), however, indicate the need to examine this issue in future research using larger and ethnically homogeneous samples or new methods that are being developed for detection and correction using within-family analyses or genome-wide covariates (Risch, Burchard, Ziv, & Tang, 2002; Rosenberg, Li, Ward, & Pritchard, 2003).

Finally, because there was a dearth of empirical research on which to base a search for specific candidate genes that affect the development of RSA and RSA reactivity, the justification for examining dopamine receptor genes in this study was indirect, theoretical, and based on known associations between dopamine and specific behavioral outcomes and between specific behavioral outcomes and infants' RSA reactivity. Although not ideal, this approach has been used to initiate or narrow the search for specific genes so that subsequent, molecular link-age studies may prioritize candidate genes in a hypothesis-driven search.

Keeping in mind the limitations and caveats noted earlier, the current findings provide an important first step in understanding possible genetic and environ-mental influences on the development of infants' RSA functioning. Although there are various interpretations of the current findings, one possibility is that these results show how infants' interactions with their caregivers over time could provide the necessary experience to effect change on genetic expression. The observed developmental change for one group of infants may reflect the impact of life experience; over time, reactions may be attributed not only to innate physiological mechanisms but also to experiential history (Sroufe, 1995). Between 3 and 12 months, infants undergo substantial developmental change, and recent developments in disparate areas of research are converging on the conclusion that inter-personal experiences strongly shape not just behavior but neural, physiological, and physical development (e.g., Siegel, 2001).

The 1st year of life is an important time for infants to develop regulatory abilities to adaptively cope with their environment. Parents may influence the way in which infants respond to situations, behaviorally and physiologically, through their help in alleviating negative emotions, reinforcing positive ones, and structuring the environment in which infants experience emotion. In the current study, at 3 and 6 months of age, infants may have exhibited RSA responses to their environments based on the influence of specific genetic polymorphisms of DRD2 rather than parenting. However, the contributions of DRD2 to the expression of physiological responses to the environment during the early weeks of life are likely to be, over time, only one of the many other factors that play a role in the development of these characteristics (Auerbach et al., 1999). By 12 months of age, specific experiential factors (i.e., parenting) may exert an increasingly potent influence on infants, as they become more likely to understand, interpret, remember, and apply past experiences to novel situations (Sroufe, 1990).

Thus, when examining gene – environment inter-actions, it is important to take into account the cumulative nature of environmental influences. Moffitt, Caspi, and Rutter (2005, p. 476) stated that "although the effects of a pathogen measured at a single point may be weak, the cumulative effects of extended or repeated exposure to that pathogen are often strong ... most risks derive from long-standing situations rather than acute events." The current study provides empirical support for this assertion. At 3 and 6 months of age, infants may not have had enough exposure to maternal behavior for it to exert an effect; by 12 months, however, the cumulative effect of maternal behavior may be more pronounced.

In contrast to the findings for RSA reactivity, there were no genetic effects of DRD2 on baseline RSA or its development. This is consistent with the findings of behavioral genetics studies discussed earlier in which the genetic variance associated with RSA reactivity during a challenge task was higher than that associated with baseline RSA (Boomsma et al., 1990; Kupper et al., 2005; Snieder et al., 1997). The current finding provides further evidence that conditions of challenge may elicit stronger individual differences in vagal regulation, leading genetic differences to become more pronounced (Boomsma et al., 1998).

The current findings may help to elucidate some of the nonreplications in the literature regarding the association of DRD2 and adult personality and psychological outcomes. Extant research has reported associations between the risk allele of DRD2 (A_1^+) and disorders related to the inability to regulate behavior (e.g., Comings et al., 1996; Noble et al., 1994), whereas other studies have failed to find these relationships (e.g., Edenberg et al., 1998). Our findings suggest that the risk allele may indeed be related to problems with physiological regulation during a stressor (i.e., lack of RSA withdrawal). However, when combined with a positive environment, the risks of this allele may diminish over time. Although the specific mechanisms that link DRD2 and RSA functioning cannot be determined by these exploratory analyses, we pro-vide initial data suggesting that this link may exist. Future studies should include analyses of genes and environment over time to further clarify their joint contribution to phenotypic outcomes (e.g., Caspi et al., 2002).

Another interesting finding of the current study was that infants with the risk allele (A_1^+) had mothers who were less sensitive than those without the risk allele (A_1^-). A strictly genetic explanation suggests that infants with the A_1^+ allele may have inherited this genotype from their mothers and that lack of sensitivity is associated with behavioral regulatory problems associated with this genetic risk. Another likely explanation is a "passive" gene – environment correlation, indicating that the influence of the mother's genotype on her own behavior influences her child's experiences and development (Rutter, 1997). From this perspective, our results are consistent with what we would expect if children possessed the same risk allele as their mothers; they may be at increased risk due to both genetic and environmental susceptibility for behavioral or physiological regulatory problems. Finally, children shape and select their own experiences by the way in which they act on their environment (Plomin, DeFries, & Fulker, 1988). An infant who is genetically predisposed to exhibit less RSA withdrawal in response to challenge may have more trouble regulating behavior. Infants with behavioral problems may be more difficult to care for, leading mothers to display less sensitive behavior in response.

Future Directions

This study is the first, to our knowledge, to address the interplay of genetic and environmental factors as they contribute to the development of early RSA functioning in humans. Findings have identified a number of important directions for future research, in addition to those already noted earlier. Research in molecular genetics is moving away from single gene association studies. Future research would benefit from examining multiple candidate genes in the same model (gene – gene interactions) as well as including haplotypes (i.e., sets of single alleles or closely linked genes that tend to be inherited together). Studies of this nature would require much larger sample sizes to detect multigene, multienvironment interactions.

Furthermore, differences found in maternal sensitivity as a function of ethnicity, with AA mothers being rated as less sensitive than EA mothers, are similar to reports in the literature that AA mothers show fewer displays of physical affection and warmth toward toddlers than EA mothers (Berlin, Brooks-Gunn, Spiker, & Zaslow, 1995; Bradley, Corwyn, McAdoo, & Garcia Coll, 2001). It may be the case that cultural norms dictate different parental behaviors, as well as the way in which that behavior is interpreted (Bradley et al., 2001; Deater-Deckard, Dodge, Bates, & Pettit, 1996; McLoyd & Smith, 2002). Coding systems may not be sensitive to these cultural differences. Future research should examine mechanisms, such as socioeconomic status or coding bias, which may underlie these differential effects.

In summary, the findings of the current study are the first to identify the effects of a gene – environment interaction between parenting in infancy and a specific gene that has been implicated in difficulties in behavioral self-regulation. Our findings suggest that maternal sensitivity may moderate potential negative effects of the risk allele of the dopamine gene DRD2 on infant RSA functioning as early as 12 months of age. Although additional research is needed, the current study provides a very exciting and innovative base for future research.

References

Ainsworth, M. D., Blehar, M. C., Waters, E., & Wall, S. (1978). Patterns of attachment: Assessed in the strange situation and at home. Hillsdale, NJ: Erlbaum.

Amenta, F., Ricci, A., Rossodivita, I., Avola, R., & Tayebati, S. K. (2001). The dopaminergic system in hypertension. Clinical and Experimental Hypertension, 23, 15 – 24.

Anchordoquy, H. C., McGeary, C., Liu, L., Krauter, K., & Smolen, A. (2003). Genotyping of three candidate genes after whole-genome preamplification of DNA collected from buccal cells. Behavior Genetics, 33, 73 – 78.

Asghari, V., Schoots, O., van Kats, S., Ohara, K., Jovanovic, V., & Guan, H. C. et al. (1994). Dopamine D4 receptor repeat: Analysis of different native and mutant forms. Molecular Pharmacology, 46, 364.

Auerbach, J. G., Faroy, M., Ebstein, R., Kahana, M., & Levine, J. (2001). The association of the dopamine D4 receptor gene (DRD4) and the serotonin transporter promotor gene (5-HTTLPR) with temperament in 12-month-old infants. Journal of Child Psychology and Psychiatry, 6, 777–783.

Auerbach, J. G., Geller, V., Lezer, S., Shinwell, E., Belmaker, R. H., & Levine, J., et al. (1999). Dopamine D4 receptor (D4DR) and serotonin transporter promoter (5-HTTLPR) polymorphisms in the determination of temperament in 2-month-old infants. Molecular Psychiatry, 4, 369–373.

Barr, C. L., & Kidd, K. K. (1993). Population frequencies of the A1 allele at the dopamine D2 receptor locus. Biological Psychiatry, 34, 204–209.

Bazhenova, O. V., Plonskaia, O., & Porges, S. W. (2001). Vagal reactivity and affective adjustment in infants during interaction challenges. Child Development, 72, 1314–1326.

Bek, M. J., Eisner, G. M., Felder, R. A., & Jose, P. A. (2001). Dopamine receptors in hypertension. Mount Sinai Journal of Medicine, 68, 362 – 369.
Belsky, J., Rovine, M., & Taylor, D. G. (1984). The Pennsylvania Infant and Family Development Project: 2. The origins of individual differences in infant-mother attachment: Maternal and infant contributions. Child Development, 59, 156 – 167.
Benjamin, J., Li, L., Patterson, C., Greenberg, B. D., Murphy, D. L., & Hamer, D. H. (1996). Population and familial association between the D4 dopamine receptor gene and measures of novelty seeking. Nature Genetics, 12,81–84.
Berlin, L., Brooks-Gunn, J., Spiker, D., & Zaslow, M. J. (1995). Examining observational measures of emotional support and cognitive stimulation in black and white mothers of preschoolers. Journal of Family Issues, 16, 664–686.
Blair, C., Granger, D., Willoughby, M., Kivlighan, K., & the Family Life Project Key Investigators. (2006). Maternal
sensitivity is related to hypothalamic-pituitary-adrenal axis stress reactivity and regulation in response to emotion challenge in 6-month-old infants. Annals of the New York Academy of Science, 1094, 263 – 267.
Blum, K., Noble, E. P., Sheridan, P. J., Finley, O., Montgomery, A., & Ritchie, T., et al. (1991). Association of the A1 allele of the D2 dopamine receptor gene with severe alcoholism. Alcohol, 8, 409–416.
Boomsma, D. I., Snieder, H., de Geus, E. J., & van Doornen, L. J. (1998). Heritability of blood pressure increases during mental stress. Twin Research, 1, 15 – 24.
Boomsma, D. I., van Baal, G. C., & Orlebeke, J. F. (1990). Genetic influences on respiratory sinus arrhythmia across different task conditions. Acta Geneticae Medicae et Emellologia: Twin Research (Roma), 39, 181 – 191.
Bornstein, M. H., & Suess, P. E. (2000). Physiological self-regulation and information processing in infancy: Cardiac vagal tone and habituation. Child Development, 2, 273–287.
Bradley, R. H., Corwyn, R. F., McAdoo, H. P., & Garcia Coll, C. (2001). The home environments of children in the United States part I: Variations by age, ethnicity, and poverty status. Child Development, 72, 1844–1867.
Burgess, K. B., Marshall, P. J., Rubin, K. H., & Fox, N. A. (2003). Infant attachment and temperament as predictors of subsequent externalizing problems and cardiac physiology. Journal of Child Psychology and Psychiatry and Allied Disciplines, 44, 819–831.
Calkins, S. D. (1997). Cardiac vagal tone indices of temperamental reactivity and behavioral regulation in young children. Developmental Psychobiology, 31, 125 – 135.
Calkins, S. D., Smith, C. L., Gill, K., & Johnson, M. C. (1998). Maternal interactive style across contexts: Relations to emotional, behavioral and physiological regulation during toddlerhood. Social Development, 7, 350 – 369.
Cardon, L. R., & Palmer, L. J. (2003). Population stratification and spurious allelic association. Lancet, 361, 598–604.
Caspi, A., McClay, J., Moffitt, T. E., Mill, J., Martin, J., & Craig, I. W. et al. (2002). Role of genotype in the cycle of violence in maltreated children. Science, 297, 851 – 854.
Cassidy, J. (1994). Emotion regulation: Influences of attachment relationships. In N. A. Fox (Ed.), The development of emotion regulation: Biological and behavioral considerations. Monographs of the Society for Research in Child Development, 59(2 – 3, Serial no. 240), 228 – 283.
Chitravanshi, V. C., & Calaresu, F. R. (1992). Additive effects of dopamine and 8-OH-DPAT microinjected into the nucleus ambiguus in eliciting vagal bradycardia in rats. Journal of the Autonomic Nervous System, 41, 121–127.
Cloninger, C. R., Adolfsson, R., & Svrakic, N. M. (1996). Mapping genes for human personality. Nature Genetics, 12,3–4.
Cohn, J. F., Campbell, S. B., & Ross, S. (1991). Infant response in the still-face paradigm at 6 months predicts avoidant and secure attachment at 12 months. Development and Psychopathology, 3, 367–376.
Comings, D. E., Rosenthal, R. J., Lesieur, H. R., Rugle, L. J., Muhleman, D., & Chiu, C. et al. (1996). A study of the dopamine D2 receptor gene in pathological gambling. Pharmacogenetics, 6, 223–234.

Deater-Deckard, K., Dodge, K. A., Bates, J. E., & Pettit, G. S. (1996). Discipline among African American and European American mothers: Links to children's externalizing behaviors. Developmental Psychology, 32, 1065 – 1072.
DeLuca, A., Rizzardi, M., Torrente, I., Alessandroni, R., Salvioli, G. P., & Filograsso, N. et al. (2001). Dopamine D4 receptor (DRD4) polymorphism and adaptability trait during infancy: A longitudinal study in 1- to 5- month-old neonates. Neurogenetics, 3, 79 – 82.
Denver, J. W., Reed, S. F., & Porges, S. W. (2007). Methodological issues in the quantification of respiratory sinus arrhythmia. Biological Psychology, 74, 286 – 294.
Derryberry, D., & Rothbart, M. K. (1984). Emotion, attention, and temperament. In C. E. Izard, J. Kagan, & R. B. Zajonc (Eds.), Emotions, cognition, and behavior (pp. 132 – 166). Cambridge, UK: Cambridge University Press.
DiPietro, J. A., Porges, S. W., & Uhly, B. (1992). Reactivity and developmental competence in pre-term and full-term infants. Developmental Psychology, 28, 831 – 841.
Ebstein, R. P., Nemanov, L., Klotz, I., Gritsenko, I., & Belmaker, R. H. (1997). Additional evidence for an association between the dopamine D4 receptor (D4DR) exon III polymorphism and the human personality trait of novelty seeking. Molecular Psychiatry, 2, 472 – 477.
Edenberg, H. J., Foroud, T., Koller, D. L., Goate, A., Rice, J., & Van Eerdewegh, P. et al. (1998). A family-based analysis of the association of the dopamine D2 Receptor (DRD2) with alcoholism. Alcoholism: Clinical and Experimental Research, 22, 505–512.
Field, T. (1984). Early interactions between infants and their postpartum depressed mothers. Infant Behavior and Development, 7, 517–522.
Fox, N. A. (1989). Psychophysiological correlates of emotional reactivity during the first year of life. Developmental Psychology, 25, 364 – 372.
Fox, N. A., & Stifter, C. A. (1989). Biological and behavioral differences in infant reactivity and regulations. In G. Kohnstamm, J. Bates, & M. Rothbart (Eds.), Handbook of temperament in childhood. Chichester, UK: John Wiley.
Gebhardt, C., Leisch, F., Schussler, P., Fuchs, K., Stompe, T., & Sieghart, W., et al. (2000). Non-association of dopamine D4 and D2 receptor genes with personality in healthy individuals. Psychiatric Genetics, 10, 131 – 137.
Gelernter, J., Kranzler, H., Coccaro, E., Siever, L., New, A., & Mulgrew, C. L. (1997). D4 dopamine-receptor (DRD4) alleles and novelty seeking in substance-dependent, personality-disorder, and control subjects. American Journal of Human Genetics, 61, 1144 – 1152.
George, S. R., Cheng, R., Nguyen, T., Israel, Y., & O'Dowd, B. F. (1993). Polymorphisms of the D4 dopamine receptor alleles in chronic alcoholism. Biochemical and Bio-physical Research Communications, 196, 107 – 114.
Gottlieb, G. (1998). Normally occurring environmental and behavioral influences on gene activity: From central dogma to probabilistic epigenesis. Psychological Review, 105, 792–802.
Gottlieb, G. (2003). On making behavioral genetics truly developmental. Human Development, 46, 337–355.
Gottlieb, G., & Halpern, C. T. (2002). A relational view of causality in normal and abnormal development. Development and Psychopathology, 14, 421 – 435.
Grossman, P., & Taylor, E. (2007). Toward understanding sinus arrhythmia relators to cardiac vagal tune, evolution, and biobehavioral functions. Biological Psychology, 74, 263–285.
Gunnar, M. R., Mangelsdorf, S., Larson, M., & Hertsgaard, L. (1989). Attachment, temperament, and adrenocortical activity in infancy: A study of psychoendocrine regulation. Developmental Psychology, 25, 355–363.
Haley, D. W., & Stansbury, K. (2003). Infant stress and parent responsiveness: Regulation of physiology and behavior during still-face and reunion. Child Development, 74, 1534 – 1546.
Ham, J., & Tronick, E. (2006). Infant resilience to the stress of the still-face: Infant and maternal psychophysiology are related. Annals of the New York Academy of Sciences, 1094, 297–302.
Healy, B. T. (1992). The heritability of autonomic nervous system processes. In T. M. Field, P. M. McCabe, & N. Schneiderman (Eds.), Stress and coping in infancy and childhood (pp. 69–82). Hillsdale, NJ: Erlbaum.

Hill, A., Mills-Koonce, R., Propper, C., Calkins, S., Granger, D., Moore, G., et al. (2008). Physiological responses to the strange situation: Vagal withdrawal, salivary *a*-amylase and salivary cortisol in infants and mothers as a function of attachment status. Manuscript under review.
Hoggart, C. J., Parra, E. J., Shriver, M. D., Bonilla, C., Kittles, R. A., Clayton, D. G., et al. (2003). Control of confounding of genetic associations in stratified populations. American Journal of Human Genetics, 72, 1492–1504.
Huffman, L. C., Bryan, Y., Del Carmen, R., Pederson, F., Doussard-Roosevelt, J., & Porges, S. W. (1998). Infant temperament and cardiac vagal tone: Assessments at twelve weeks of age. Child Development, 69, 624 – 635.
Hyde, T. M., Knable, M. B., & Murray, A. M. (1996). Distribution of dopamine D1-D4 receptor subypes in human dorsal vagal complex. Synapse, 24, 224 – 232.
Izard, C. E., Porges, S. W., Simons, R. F., Haynes, O. M., Hyde, C., & Parisi, M. et al. (1991). Infant cardiac activity: Developmental changes and relations with attachment. Developmental Psychology, 27, 432–439.
Jönsson, E. G., Nöthen, M. M., Gustavsson, J. P., Neidt, H., Forslund, K., Mattila-Evenden, N., et al. (1998). Lack of association between dopamine D4 receptor gene and personality traits. Psychological Medicine, 28, 985 – 989.
Jose, P. A., Eisner, G. M., & Felder, R. A. (1998). The renal dopamine receptors in health and hypertension. Pharmacological Therapy, 80, 149 – 182.
Jose, P. A., Eisner, G. M., & Felder, R. A. (1999). Dopaminergic mechanisms in the development of hypertension. In R. McCarty, D. A. Blizard, & R. I. Chevalier (Eds.), Handbook of hypertension (pp. 1 – 44). Amsterdam: Elsevier Science.
Kupper, N., Gonneke, W., Posthuma, D., de Boer, D., Boomsma, D. I., & de Geus, E. J. (2005). A genetic analysis of ambulatory cardiorespiratory coupling. Psychophysiology, 42, 202–212.
Lawford, B. R., Young, R. M., Noble, E. P., Sargent, J., Rowell, J., & Shadforth, S., et al. (2000). The D2 dopamine receptor A(1) allele and opioid dependence: Association with heroin use and response to methadone treatment. American Journal of Medical Genetics, 96, 592 – 598.
Li, X. X., Asico, L. D., Low, M., et al. (1998). Disruption of the dopamine D2 receptor produces non-renal dependent hypertension. Journal of the American Society of Nephrology, 9, 311.
Linthorst, A. C. E., De Lang, H., De Jong, W. T., & Versteeg, D. H. G. (1991). Effect of the dopamine D2 receptor agonist quinpirole on the in vivo release of dopamine in the caudate nucleus of hypertensive rats. European Journal of Pharmacology, 201, 125 – 133.
Linthorst, A. C. E., van Giersbergen, P. L., Gras, M., Versteeg, D. H. G., & De Jong, W. (1994). The nigrostriatal dopamine system: Role in the development of hypertension in spontaneously hypertensive rats. Brain Research, 639, 261 – 268.
Little, R. A. A., & Rubin, D. B. (1987). Statistical analysis with missing data. New York: Wiley.
Lu, R. B., Lee, J. F., Ko, H. C., & Lin, W. W. (2001). Dopamine D2 receptor gene (DRD2) is associated with alcoholism with conduct disorder. Alcoholism: Clinical and Experimental Research, 25, 177 – 184.
Main, M., & Solomon, J. (1986). Discovery of a new, insecure-disorganized/disoriented attachment pattern. In T. B. Brazelton & M. Yogman (Eds.), Affective development in infancy (pp. 95 – 124). Norwood, NJ: Ablex.
McGue, M., Slutske, W., Taylor, J., & Iacono, W. G. (1997). Personality and substance use disorders: I. Effects of gender and alcoholism subtype. Alcoholism: Clinical and Experimental Research, 21, 513 – 520.
McLoyd, V. C., & Smith, J. (2002). Physical discipline and behavior problems in African American, European American, and Hispanic children: Emotional support as a moderator. Journal of Marriage and Family, 64, 40 – 53.
Mezzacappa, E., Tremblay, R. E., Kindlon, D., Saul, J. P., Arseneault, L., & Seguin, J. et al. (1997). Anxiety, antisocial behavior, and heart rate regulation in adolescent males. Journal of Child Psychology and Psychiatry, 38, 457 – 469.
Mills-Koonce, W., Gariépy, J., Propper, C., Sutton, K., Calkins, S., Moore, G., et al. (2007). Infant and parent factors associated with early maternal sensitivity: A caregiver-attachment systems approach. Infant Behavior and Development, 30, 114 – 126.
Missale, C., Nash, S. R., Robinson, S. W., Jaber, M., & Caron, M. G. (1998). Dopamine receptors: From structure to function. Physiological Review, 78, 189 – 225.

Miyake, H., Nagashima, K., Onigata, K., Nagashima, T., Takano, Y., & Morikawa, A. (1999). Allelic variations of the D2 dopamine receptor gene in children with idiopathic short stature. Journal of Human Genetics, 44, 26–29.
Moffitt, T. E., Caspi, A., & Rutter, M. (2005). Strategy for investigating interactions between measured genes and measured environments. Archives of General Psychiatry, 62, 473–481.
Moffitt, T. E., Caspi, A., & Rutter, M. (2006). Measured gene-environment interactions in psychopathology: Concepts, research strategies, and implications for research, intervention, and public understanding of genetics. Perspectives on Psychological Science, 1, 5–27.
Moore, G. A., & Calkins, S. D. (2004). Infants' vagal regulation in the still-face paradigm is related to dyadic coordination of mother-infant interaction. Developmental Psychology, 40, 1068 – 1080.
National Institute of Child Health and Human Development Study of Early Child Care. (1997). Effects of infant child care on infant-mother attachment security: Results of the NICHD study of early child care. Child Development, 68, 860–879.
Noble, E. P. (2003). D2 dopamine receptor gene in psychiatric and neurologic disorders and its phenotypes. American Journal of Medical Genetics Part B: Neuropsychiatric Genetics, 116B, 103–125.
Noble, E. P., Ozkaragoz, T. Z., Ritchie, T. L., Zhang, X., Belin, T. R., & Sparkes, R. S. (1998). D2 and D4 dopamine receptor polymorphisms and personality. American Journal of Medical Genetics: Neuropsychiatric Genetics, 81, 257–267.
Noble, E. P., Syndulko, K., Fitch, R. J., Ritchie, T., Bohlman, M. C., Guth, P. et al. (1994). D2 dopamine receptor Taq1 A alleles in medically ill alcoholic and nonalcoholic patients. Alcohol, 29, 729 – 744.
Pérez de Castro, I., Ibáñez, A., Torres, P., Sáiz-Ruiz, J., & Fernández-Piqueras, J. (1997). Genetic association study between pathological gambling and a functional DNA polymorphism at the D4 receptor gene. Pharmacogenetics, 7,345–348.
Plomin, R., DeFries, J. C., & Fulker, D. W. (1988). Nature and nurture during infancy and early childhood. New York: Cambridge University Press.
Plomin, R., & Rutter, M. (1998). Child development, molecular genetics, and what to do with genes once they are found. Child Development, 69, 1223 – 1242.
Pohjalainen, T., Rinne, J. O., Nagren, K., Lehikoinen, P., Anttila, K., Syvalahti, E. K. G., et al. (1998). The A1 allele of the human D2 dopamine receptor gene predicts low D2 receptor availability in healthy volunteers. Molecular Psychiatry, 3, 256 – 260.
Porges, S. W. (1985). Method and apparatus for evaluating rhythmic oscillations in a periodic physiological response system (U.S. Patent No. 4,510,944). Washington, DC: U.S. Patent Office.
Porges, S. W. (1991). Vagal tone: An autonomic mediatory of affect. In J. A. Garber & K. A. Dodge (Eds.), The development of affect regulation and dsyregulation (pp. 11 – 128). New York: Cambridge University Press.
Porges, S. W. (1996). Physiological regulation in high-risk infants: A model for assessment and potential intervention. Development and Psychopathology, 8, 43 – 58.
Porges, S. W. (2007). The polyvagal perspective. Biological Psychology, 74, 116 – 143.
Porges, S. W., & Byrne, E. A. (1992). Research methods for measurement of heart rate and respiration. Biological Psychology, 34, 93–130.
Porges, S. W., Doussard-Roosevelt, J. A., Portales, A. L., & Greenspan, S. I. (1996). Infant regulation of the vagal "brake" predicts child behavior problems: A psychobiological model of social behavior. Developmental Psycho-biology, 29, 697–712.
Porges, S. W., Doussard-Roosevelt, J. A., Portales, A. L., & Suess, P. E. (1994). Cardiac vagal tone: Stability and relation to difficultness in infants and 3-year-olds. Developmental Psychobiology, 27, 289–300.
Porter, C. L. (2003). Coregulation in mother-infant dyads: Links to infants' cardiac vagal tone. Psychological Reports, 92, 307–319.
Porter, C. L., Wouden-Miller, M., Silva, S. S., & Porter, A. E. (2003). Marital harmony and conflict: Links to infants' emotional regulation and cardiac vagal tone. Infancy, 4, 297–307.
Propper, C., & Moore, G. A. (2006). The influence of parenting on infant emotionality: A multi-level psychobiological perspective. Developmental Review, 26, 427 – 460.

Propper, C., Willoughby, M., Halpern, C. T., Cox, M., & Carbone, M. A. (2007). Parenting quality, DRD4, and the prediction of externalizing and internalizing behaviors in early childhood. Developmental Psychobiology, 49, 619–632.
Raudenbush, S. W., & Bryk, A. S. (2002). Hierarchical linear models. Thousand Oaks, CA: Sage.
Richards, J. E., & Cronise, K. (2000). Extended visual fixation in the early preschool years: Look duration, heart rate changes, and attentional inertia. Child Development, 71, 602 – 620.
Risch, N., Burchard, E., Ziv, E., & Tang, H. (2002). Categorization of humans in biomedical research: Genes, race and disease. Genome Biology, 3, 1 – 12.
Rosenberg, N. A., Li, L. M., Ward, R., & Pritchard, J. K. (2003). Informativeness of genetic markers for inference of ancestry. American Journal of Human Genetics, 73, 1402–1422.
Rutter, M. L. (1997). Nature-nurture integration: The example of antisocial behavior. American Psychologist, 52,390–98.
Schmidt, L. A., Fox, N. A., Rubin, K. H., Hu, S., & Hamer, D. H. (2002). Molecular genetics of shyness and aggression in preschoolers. Personality and Individual Differences, 33, 227–238.
Shields, P., Lerman, C., Audrain, J., Main, D., Boyd, N., & Caporaso, N. (1998). Dopamine D4 receptors and the risk of cigarette smoking in African-Americans and Caucasians. Cancer Epidemiology, Biomarkers, and Prevention, 7, 453 – 458.
Sibley, D., & Monsma, F. (1992). Molecular biology and dopamine receptors. Trends in Pharmacological Science, 13, 61–69.
Siegel, D. J. (2001). Toward an interpersonal neurobiology of the developing mind: Attachment relationships, “mindsight,” and neural integration. Infant Mental Health Journal, 22, 67–94.
Singh, J. P., Larson, M. G., O’Donnell, C. J., Tsuji, H., Evans, J. C., & Levy, D. (1999). Heritability of heart rate variability: The Framingham Heart Study. Circulation, 99, 2251–2254.
Sinnreich, R., Friedlander, Y., Luria, M. H., Sapoznikov, D., & Kark, J. D. (1999). Inheritance of heart rate variability: The kibbutzim family study. Human Genetics, 105, 654–661.
Sloan, R. P., Shapiro, P. A., Bigger, J. T., Bagiella, M., Steinman, R. C., & Gorman, J. M. (1994). Cardiac autonomic control and hostility in healthy subjects. American Journal of Cardiology, 74, 298 – 300.
Snieder, H., Boomsma, D. I., Van Doornen, L. J., & De Geus, E. J. (1997). Heritability of respiratory sinus arrhythmia: Dependency on task and respiration rate. Psychophysiology, 34, 317–328.
Spangler, G., & Grossmann, K. E. (1993). Biobehavioral organization in securely and insecurely attached infants. Child Development, 64, 1439 – 1450.
Spangler, G., Schiechle, M., Ilg, U., Maier, U., & Ackerman, C. (1994). Maternal sensitivity as an external organizer for biobehavioral regulation in infancy. Developmental Psychobiology, 27, 425–437.
Sroufe, L. A. (1990). An organizational perspective on the self. In D. Cicchetti & M. Beeghly (Eds.), The self in transition: Infancy to childhood (pp. 281–307). Chicago: University of Chicago Press.
Sroufe, L. A. (1995). Emotional development: The organization of emotional life in the early years. Cambridge, UK: Cam-bridge University Press.
Stifter, C. A. (1995). Approach/withdrawal processes in infancy: The relationship between parasympathetic tone and infant temperament. In K. Hood, E. Tobach, & G. Greenberg (Eds.), Approach/withdrawal theory and behavioral development (pp. 371 – 399). Hillsdale, NJ: Erlbaum.
Stifter, C. A., & Fox, N. A. (1990). Infant reactivity: Physiological correlates of newborn and 5-month temperament. Developmental Psychology, 26, 582 – 588.
Stifter, C. A., Fox, N. A., & Porges, S. W. (1989). Facial expressivity and vagal tone in 5- and 10-month-old infants. Infant Behavior and Development, 12, 127 – 137.
Suess, P. E., Porges, S. W., & Plude, D. J. (1994). Cardiac vagal tone and sustained attention in school-age children. Psychophysiology, 31, 17–22.
Suhara, T., Yasuno, F., Sudo, Y., Yamamoto, M., Inoue, M., Okubo, Y., et al. (2001). Dopamine D2 receptors in the insular cortex and the personality trait of novelty seeking. Neuroimage, 13, 891–895.
Thomas, G. N., Tomlinson, B., & Critchley, J. A. J. H. (2000). Modulation of blood pressure and obesity with the dopamine D2 receptor gene Taq1 polymorphism. Hyper-tension, 36, 177 – 182.
Thomas, D. C., & Witte, J. S. (2002) Point: Population stratification: A problem for case-control studies of candidate-gene associations? Cancer Epidemiology, Bio-markers and Prevention, 11, 505 – 512.

Thompson, R. A. (1994). Emotion regulation: A theme in search of a definition. In N. A. Fox (Ed.), Emotion regulation: Behavioral and biological considerations. Monographs of the Society for Research in Child Development (2 – 3, Serial no. 240), 25 – 52.
Tronick, E. Z., Als, H., Adamson, L., Wise, S., & Brazelton, B. (1978). The infant's response to entrapment between contradictory messages in face-to-face interaction. Journal of the American Academy of Child Psychiatry, 1, 1 – 13.
van den Buuse, M. (1998). Role of the mesolimbic dopamine system in cardiovascular homeostasis. Stimulation of the ventral tegmental area modulates the effect of vasopressin on blood pressure in conscious rats. Clinical and Experimental Pharmacology and Physiology, 25, 661–668.
Vaughan, C. E., van den Buuse, M., & Roland, B. L. (1999). Brain dopamine D2 receptor mRNA levels are elevated in young spontaneously hypertensive rats. Neuroscience Research, 34, 199 – 205.
Voss, A., Busjahn, A., Wessel, N., Schurath, R., Faulhaber, H. D., Luft, F. C., et al. (1996). Familial and genetic influences on heart rate variability. Journal of Electro-cardiology, 29, 154 – 160.
Wacholder, S., Rothman, N. & Caporaso, N. (2002). Counterpoint: Bias from population stratification is not a major threat to the validity of conclusions from epidemiological studies of common polymorphisms and cancer. Cancer Epidemiology, Biomarkers and Prevention, 11,513–520.
Weinberg, M. K., & Tronick, E. Z. (1996). Infant affective reactions to the resumption of maternal interaction after the still-face. Child Development, 67, 905 – 914.
Wiers, R. W., Sergeant, J. A., & Gunning, W. B. (1994) Psychological mechanisms of enhanced risk of addiction in children of alcoholics: A dual pathway? Acta Paediatrica Supplement, 404, 9 – 13.
Williams, R. B., Marchuk, D. A., Gadde, K. M., Barefoot, J. C., Crichnik, K., Helms, M. J., et al. (2003). Serotonin-related gene polymorphisms and central nervous system 2serotonin function. Neuropsychopharmacology, 28, 533 – 541.
Yeh, T., Yang, Y., Chiu, N., Yao, W., Yeh, S., Wu, J., et al. (2006). Correlation between striatal dopamine D2/D3 receptor binding and cardiovascular activity in healthy subjects. American Journal of Hypertension, 19, 964 – 969.
Ziv, E., & Burchard, E. G. (2003). Human population structure and genetic association studies. Pharmacogenomics, 4, 431–441.