The Journal of Nutrition
Supplement: Iron Works…The John Beard Memorial Symposium

Early Iron Deficiency Has Brain and Behavior Effects Consistent with Dopaminergic Dysfunction[1–3]

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Abstract

To honor the late John Beard's many contributions regarding iron and dopamine biology, this review focuses on recent human studies that test specific hypotheses about effects of early iron deficiency on dopamine system functioning. Short- and long-term alterations associated with iron deficiency in infancy can be related to major dopamine pathways (mesocortical, mesolimbic, nigrostriatal, tuberohypophyseal). Children and young adults who had iron deficiency anemia in infancy show poorer inhibitory control and executive functioning as assessed by neurocognitive tasks where pharmacologic and neuroimaging studies implicate frontal-striatal circuits and the mesocortical dopamine pathway. Alterations in the mesolimbic pathway, where dopamine plays a major role in behavioral activation and inhibition, positive affect, and inherent reward, may help explain altered social-emotional behavior in iron-deficient infants, specifically wariness and hesitance, lack of positive affect, diminished social engagement, etc. Poorer motor sequencing and bimanual coordination and lower spontaneous eye blink rate in iron-deficient anemic infants are consistent with impaired function in the nigrostriatal pathway. Short- and long-term changes in serum prolactin point to dopamine dysfunction in the tuberohypophyseal pathway. These hypothesis-driven findings support the adverse effects of early iron deficiency on dopamine biology. Iron deficiency also has other effects, specifically on other neurotransmitters, myelination, dendritogenesis, neurometabolism in hippocampus and striatum, gene and protein profiles, and associated behaviors. The persistence of poorer cognitive, motor, affective, and sensory system functioning highlights the need to prevent iron deficiency in infancy and to find interventions that lessen the long-term effects of this widespread nutrient disorder. J. Nutr. 141: 740S–746S, 2011.

Introduction

At the time of Oski and Honig's (1) seminal 1978 report of improved developmental test scores in iron-deficient anemic infants who received iron therapy, understanding of underlying brain mechanisms was limited. Little was known except that many enzymes in the central nervous system (CNS)[4] were iron dependent, and pioneering work by Dallman et al. (2,3) was showing that iron deficiency in rats lowered brain iron concentration and Youdim was documenting impaired dopamine function and related behaviors [see (4) for review]. Since then, much has been learned about neuroanatomical, neurochemical, neurometabolic, and genomic/proteomic effects of early iron deficiency (5–9), thanks in part to research in rodent models in the laboratories of John Beard (continued under the leadership of Erica Unger), James Connor, Barbara Felt, Michael Georgieff, Raghu Rao, and others. There have also been many more studies in human infants with iron deficiency anemia. These document poorer performance on global assessments of cognitive, motor, and social-emotional behavior (10–13) and alterations in such regulatory processes as the sleep-wake cycle (14).

[1] Published in a supplement to *The Journal of Nutrition*. Presented at the symposium, "Iron Works…The John Beard Memorial Symposium", held in State College, PA, November 2, 2009. The symposium was organized by the Department of Nutritional Sciences as a tribute to Dr. Beard's contribution to improving our understanding of iron metabolism. Its contents are solely the responsibility of the authors. The Supplement Coordinator for this supplement was Jere D. Haas, Cornell University. Supplement Coordinator disclosures: Jere D. Haas had no relationships to disclose. The supplement is the responsibility of the Guest Editor, to whom the Editor of *The Journal of Nutrition* has delegated supervision of both technical conformity to the published regulations of *The Journal of Nutrition* and general oversight of the scientific merit of each article. The Guest Editor for this supplement was Mary Cogswell, Centers for Disease Control. Guest Editor disclosure: Mary Cogswell had no relationships to disclose. Publication costs for this supplement were defrayed in part by the payment of page charges. This publication must therefore be hereby marked "advertisement" in accordance with 18 USC section 1734 solely to indicate this fact. The opinions expressed in this publication are those of the authors and are not attributable to the sponsors or the publisher, Editor, or Editorial Board of *The Journal of Nutrition*.

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[2] Supported by grant nos. P01 HD39386 and R01 HD33487 and a MERIT award (R23 HD31606) from the National Institute of Child Health and Human Development, B. Lozoff, Principal Investigator. The content is solely the responsibility of the author and does not necessarily represent the official views of the National Institute of Child Health and Human Development or the National Institutes of Health.

[3] Author disclosures: B. Lozoff, no conflicts of interest.

[4] Abbreviations used: CANTAB, Cambridge Automated Neuropsychological Test Assessment Battery; CNS, central nervous system; GABA, γ-aminobutyric acid.

First published online February 23, 2011; doi:10.3945/jn.110.131169.

Though consistently observed, such global outcomes give little indication of the CNS processes affected by iron deficiency in infancy. Guided by basic science and behavioral research in rodent models, some human infant studies have become more hypothesis driven and have tried to overcome the challenges in assessing specific CNS effects in infants. This review will focus on such studies. Results that are most related to Beard's many contributions regarding neurotransmitter function, especially dopamine, will be emphasized. Some of the more specific findings come from a cross-species NIH-supported program project grant entitled Brain and Behavior in Early Iron Deficiency (P01 HD39386), where Beard played key roles in the rodent project and analytical core, and from 2 NIH-supported longitudinal studies of long-term effects of iron deficiency anemia in human infants (R01 HD33487 and R23 HD31606). The infants in all studies were full term and healthy, and those with iron deficiency anemia received a full course of oral iron therapy. Specific criteria for anemia and iron deficiency varied from study to study but generally used cutoffs 2 SD below reference values for age and altitude (for hemoglobin). For instance, hemoglobin cutoffs might be ≤100 g/L at 6 mo or <110 g/L at 12 mo, depending on the study. Iron deficiency was defined as 2 or more abnormal iron measures, using study-dependent combinations of mean cell volume (<70 fL, <74 fL), transferrin saturation (≤10%, <12%), free erythrocyte protoporphyrin (≥100 μg/dL RBC), zinc protoporphyrin (>69 μmol/mol heme), ferritin (<12 μg/L), and red cell distribution width (>14%).

Dopamine is important in regulating cognition and emotion, reward and pleasure, movement, and hormone release (15). Striatal networks with dopamine as the major neurotransmitter relate to higher order cognitive and emotional processes, motivated behavior, positive affect, and reward-related processing, as well as motor functioning (15,16). Thus, alterations in the striatum and basal ganglia more generally are likely to have many manifestations, given their role in widely distributed networks. Such effects have been observed in both animal models and humans with early iron deficiency.

Rodent models of diet-induced iron deficiency during development have helped generate specific hypothesis that we are testing in the human projects mentioned above. Rat studies show that brain and behavior effects and their reversibility with iron repletion vary depending on the timing and severity of iron deficiency and the timing of iron treatment (4,8,17–21). For instance, iron deficiency anemia reduces brain iron in rats, but the regional pattern and degree of reduction depends on timing and severity (17,20,22). Similarly, dopaminergic alterations vary, as shown in part by the extensive contributions of Beard and colleagues in the last 15 y or so. For example, reduced D1 and D2 receptor densities in the striatum, increased extracellular dopamine concentrations, and reduced densities of dopamine and other monoamine transporters vary with timing and severity (4–6,19,20,23–33).

Early rodent models resulted in severe iron deficiency anemia and poor growth. To be more relevant to iron deficiency anemia in human infants, Felt and Lozoff (34) developed a rodent model of iron deficiency during gestation and lactation with moderate iron restriction that avoids marked growth effects. This model, which is used in our program project, produces a more moderate level of brain iron deficiency than previous models (20,21). Even in this more moderate iron deficiency model, brain iron was reduced and dopamine and serotonin metabolism were altered while animals had iron deficiency anemia (20). The striatal metabolome was also affected (35). Some neurotransmitter alterations persisted in adulthood despite correction of anemia and brain iron content (except in the thalamus) (21). Behavioral alterations in iron-deficient rats are consistent with the CNS effects (18,20,21,36,37). During development, a number of sensory-motor reflexes are delayed (20). For instance, elicited forelimb placing emerged later during early development for rat pups with iron deficiency anemia (20), even though striatal metabolic alterations corrected with iron repletion (35). In young adulthood, rats with iron deficiency anemia during gestation and lactation had disrupted grooming sequences (21). These behavioral measures were chosen by Beard et al. and Felt et al. (20,21) specifically to assess striatal dopamine-dependent functional behaviors. Other persistent consequences related to the dopamine system include less exploration and more hesitancy in the face of novelty (18,21,34,36). Related CNS changes have been observed in monkeys. In the University of Wisconsin-Madison monkey project of our program project, Coe et al. (38) collaborated with Beard and colleagues to assess brain monoamines. Juvenile monkeys that had iron deficiency anemia as infants had lower dopamine levels in the cerebrospinal fluid (38) compared with monkeys without iron deficiency anemia.

Studies of iron deficiency in human infancy have now found differences that are consistent with altered dopaminergic function. The findings will be considered as they may relate to the 4 major dopamine pathways, i.e. mesocortical, mesolimbic, nigrostriatal, and tuberohypophyseal (15). However, there is overlap in these pathways (39) and we sometimes had to make educated guesses about which pathway is most involved in a particular functional outcome. Furthermore, we have oversimplified interpretation of results for heuristic purposes. Although the findings will be discussed in terms of specific hypotheses about the effects of early iron deficiency on dopamine pathways, we do not claim that dopaminergic dysfunction is the sole explanation. There is no doubt that iron deficiency affects other neurotransmitters and other processes, such as myelination, dendritogenesis, neurometabolism, and gene and protein profiles (4,6–9,40).

Mesocortical pathway

We have sought to include neurocognitive tasks where nonhuman primate studies or human neuroimaging studies implicate frontal-striatal circuits and dopaminergic function. The striatum sends dopaminergic projections to prefrontal cortex and is recruited in the control of executive functions (e.g. inhibitory control, planning, etc.), sustained attention, working memory, memory storage and retrieval, emotion regulation, and motivation (41). Cognitive control, which is essential for higher cognition, develops gradually throughout childhood and adolescence, probably due to prolonged maturation of the prefrontal cortex (42,43). Although several neurotransmitters are involved in inhibitory control, dopamine in its prefrontal-striatal circuits plays a key role (41). Relations between dopaminergic activity and performance on frontostriatal-dependent measures of executive functioning have been documented in human and nonhuman primates using dopamine agonists (drugs that stimulate dopamine release) and antagonists (drugs that block or inhibit dopamine) (44–48). In addition, functional MRI studies show that deliberately withholding a response requires integration of circuits in prefrontal cortex, basal ganglia, and the thalamus to modulate subcortical input to cortical motor areas (49–51).

Based on this research, we predicted that if early iron deficiency impairs dopamine function in prefrontal-striatal

circuits, it would be associated with poorer function on neurocognitive tasks that require inhibiting a familiar or prepotent response. Our first opportunity to test this hypothesis occurred in a long-term follow-up study in Costa Rica. We had previously reported that compared with children who were iron sufficient in infancy before and/or after iron therapy ("good iron status"), those who had had chronic, severe iron deficiency (with or without anemia) scored lower on global measures of cognitive, affective, and motor functioning in infancy (52,53) and at 5 (54,55) and 11–14 y (56) and overall cognitive functioning up to 19 y (57). At 19 y, we also assessed specific neurocognitive functions using the Trail Making Task (58) and the Cambridge Automated Neuropsychological Test Assessment Battery (version 3; CeNeS). Compared with young adults who had good iron status in infancy, participants who had chronic, severe iron deficiency as infants performed worse on tests involving inhibitory control, set-shifting, and planning, all of which are classified as executive functions and rely on the integrity of frontal-striatal circuits (59). In our other longitudinal study in Chile, results were similar, i.e. 10-y-old children who had iron deficiency anemia in infancy had poorer performance on inhibitory tasks compared with those who had been nonanemic (60).

Our first opportunity to test the hypothesis during a period of early iron deficiency anemia was in the human infant study of the program project grant's initial 5-y period. This infant study assessed 9- to 10-mo-old infants from inner-city Detroit (61–64). We used the A-not-B test, which is considered a precursor of executive function and requires inhibitory control (65,66). In this task, the infant is invited to retrieve an object that is hidden in location A for a few trials and then hidden in a new location B. Success in the task requires the infant to notice and remember when the toy is no longer hidden in the first location (A) and to inhibit the prepotent response of searching there. Object permanence is assessed before toy location changes in the A-not-B test to determine whether the infant can retrieve a hidden toy from a single location. There was a linear effect of iron status on object permanence; infants with iron deficiency anemia were least likely to exhibit object permanence, those who were iron-sufficient were most likely, and infants with iron deficiency without anemia were intermediate (64). Taken together, these results indicate short- and long-term effects of early iron deficiency on higher order cognitive processes (executive functions) and their precursors.

Mesolimbic pathway

Through the mesolimbic pathway, dopamine plays a major role in systems of behavioral activation and inhibition, positive affect, and reward (67,68). Alterations in the mesolimbic pathway may help explain altered social-emotional behavior in early iron deficiency. Virtually every study that examined the social-emotional domain found differences comparing infants with iron deficiency anemia to those without (e.g. more wary, hesitant, solemn, unhappy, closer to their mothers, less social interaction, etc.) (53,61,69–74). Four of 6 randomized trials of supplemental iron that assessed this domain showed affective benefits of iron (e.g. more positive affect, social interaction, etc.) (12). The program project infant study adds to the mounting evidence by considering the severity of iron deficiency. We found dose-response relations between severity (iron deficiency anemia, iron deficiency without anemia, or iron sufficiency) and outcome. Linear effects showed that poorer iron status was associated with increased shyness, decreased orientation/engagement, and decreased soothability, and, when an examiner attempted to engage the infants in imitative play, decreased positive affect and engagement (61). The threshold for effects was iron deficiency with or without anemia. Social-emotional effects of iron deficiency even without anemia are supported by other studies as well. For instance, there was an early report of increased solemnity in nonanemic iron-deficient infants (74). In our preventive trial in Chile, infants who did not receive supplemental iron were less likely to show positive affect or interact socially (75). In addition, a study of human neonates reported a negative linear relation between cord-blood iron status across the full range and negative emotionality and a positive one for alertness and soothability (76). There is also evidence from nonhuman primate models. In the University of California-Davis monkey project of our program project (77), Golub et al. (78,79) observed increased boldness and impulsivity in infants of monkey mothers that did not receive prenatal iron supplements and increased tenseness and emotionality in monkey infants that were not postnatally supplemented with iron. None of the infants ever had iron deficiency anemia. Affective alterations were also observed in monkey infants with iron deficiency anemia (80). Taken together, these studies point to altered infant social-emotional behavior and affect with iron deficiency, regardless of whether the lack of iron is severe and chronic enough to cause anemia.

Notwithstanding the consistency of results, social-emotional effects have captured less attention than cognitive ones, but we previously speculated that they could equally result from direct effects of iron deficiency on associated brain systems (56,75). Findings of reduced positive affect are consistent with alterations in the mesolimbic dopamine pathway (67,68). We also have considered that behavioral alterations might be especially apparent in circumstances of novelty, unfamiliarity, or stress (53,81), because the dopamine system is involved with behavioral inhibition/activation. In the program project infant study, there was little difference in free play behavior, but several social-emotional differences became apparent when an examiner sought to engage the infant in elicited play (61). Further analyses showed that orientation and engagement with the examiner at least partially mediated the iron status effects on neurocognitive outcomes (64).

The program project's rodent study systematically investigated the behavioral domain in the moderate iron deficiency model. Behaviors that depend on striatal dopamine function were delayed or disrupted, with alterations into adulthood despite iron repletion and normalization of brain iron (20,21). Of particular relevance here are the observations of altered response to novelty, specifically, hesitancy, and reduced exploration (20,21). The results in human infants, monkey infants, and rodents in the short and long term contribute to our growing conviction that altered affect and response to novelty are among the core deficits in early iron deficiency.

Nigrostriatal pathway

The nigrostriatal system, which connects the substantia nigra and the striatum, is especially important for movement control and regulation (15). In the rodent project of the program project, Felt and Schallert included a naturalistic grooming sequence that had previously been shown to require intact dorsolateral striatal dopaminergic neurons (82). As adults, rats that experienced iron deficiency anemia during gestation and lactation had fewer complete grooming chains than control animals (21). In light of the motor sequence results in the rat model, we analyzed a particular motor task, toy retrieval from box, that required motor sequencing and bi-manual coordination in the human

infant study of the program project (63). In the box task, infants had to use their hands and arms in a coordinated sequential fashion to get a toy out of a transparent box while an examiner exerted light pressure on the box lid. There was a linear effect of iron status on the probability of retrieving the toy with good coordination: lowest in infants with iron deficiency anemia, intermediate in those with iron deficiency without anemia, and highest in iron-sufficient infants (63). This kind of task is thought to involve the motor loop of the basal ganglia. The basal ganglia play important roles in learning and execution of sequential movements (83) and also control of bi-manual coordination through motor inhibition (84). Furthermore, the basal ganglia have direct output to the supplementary motor area, which is known to be involved in the control of bi-manual coordination (85). Thus, the difficulty that iron-deficient anemic infants showed on the toy retrieval task is consistent with impaired striatal dopamine function.

In the human infant study of the program project, we also used the rate of spontaneous eye blink as a noninvasive way to assess dopaminergic function in the nigrostriatal pathway (86). Previous research in human and nonhuman primates showed that spontaneous eye blink rate can be increased by dopamine agonists and reduced by dopamine antagonists or specific lesions. The nigrostriatal system seems to be especially important (87,88) and dopamine appears to independently modulate spontaneous eye blink via D1 and D2 receptors (89). If early iron deficiency impairs dopamine functioning in this pathway, we hypothesized that the spontaneous eye blink rate would be lower in infants with iron deficiency anemia and would increase with iron therapy. In the Detroit study, iron-deficient anemic infants had a lower initial eye blink rate than nonanemic infants. After 3 mo, during which oral iron was provided to study infants, the eye blink rate increased significantly in the iron-deficient anemic group but was unchanged in the nonanemic group (86). These results provide perhaps the most direct evidence to date of reduced dopamine function in iron-deficient anemic infants. The clinical importance of a lower eye blink rate is unclear, but impaired dopamine functioning is likely to have broader impacts, given dopamine's many roles, as detailed above.

Tuberohypophyseal pathway

Dopamine from the hypothalamus provides tonic inhibition of prolactin release from the anterior pituitary, primarily through D2 receptors (90,91). Serum prolactin has therefore been considered a peripheral indicator of central dopaminergic function. If dopamine function is impaired due to such factors as fewer D2 receptors, reduced reuptake, or decreased dopamine transporter, all of which have been observed in rodent models of early iron deficiency (4,5), there should be less inhibition of prolactin release and therefore higher prolactin levels. In keeping with this physiology, increased serum prolactin levels and liver prolactin-binding sites were reported years ago in iron-deficient rats (92,93).

We previously explored the question of dopaminergic alterations in human iron deficiency by assessing serum prolactin levels in the infant phase of the Costa Rica study (52,94). We did not find a significant relation between infant iron status and serum prolactin levels, perhaps due to the limited number of pre-iron treatment serum samples or the stress of venipuncture. However, a high serum prolactin level was associated with the behavioral profile of infants with iron deficiency anemia, i.e. wary and hesitant behavior during developmental testing (94).

We measured serum prolactin in the same cohort in early adolescence (95). Rather than the higher levels we predicted, the formerly iron-deficient children showed an earlier decline in serum prolactin concentration following venipuncture. For cortisol, another stress-responsive hormone, high levels in infancy are observed with stress, but lower or blunted response patterns can be observed later on (96–98). We speculated that the same might apply to early iron deficiency and prolactin (95). In the Chile preventive trial, we subsequently observed the expected higher prolactin levels in infants who did not receive supplemental iron and those with iron deficiency anemia (99). Combining the Chile infant findings and the Costa Rica long-term results, there appear to be higher serum prolactin levels with iron deficiency anemia in infancy, consistent with reduced dopamine functioning in the tuberohypophyseal pathway, and a long-lasting dysregulation of prolactin.

Other brain and behavior effects of early iron deficiency

To pay tribute to John Beard's many contributions regarding iron deficiency and dopamine biology, this review focused on results in animal models and human infants that are consistent with dopaminergic dysfunction. However, executive functions, positive affect and response to the unfamiliar, motor sequencing and coordination, spontaneous eye blink, prolactin release, and the related dopamine pathways are not the only brain and behavior systems affected by early iron deficiency. Studies in Connor's (5,9,100–102) laboratory (in collaboration with Beard in later years) have documented that early iron deficiency impairs myelination in rodent models and alters gene and protein profiling in rodent and monkey models, with both short- and long-term effects. In humans, short- and long-term latency delays in auditory and visual evoked potential studies are consistent with delayed myelination (103–107). Nor is dopamine the only neurotransmitter affected. In addition to earlier work in severe iron deficiency rodent models (4,8), later work by Beard and colleagues (21,35,38) in the milder iron deficiency rat model and the Madison monkey model found changes in other monoamine neurotransmitters, including serotonin and norepinephrine. Studies by Rao et al. (108) show changes in glutamate in both severe and moderate iron deficiency models and other research points to iron deficiency effects on y-aminobutyric acid (108–110). The opiate system and cholinergic neurotransmission appear to be affected as well (4,23,37).

There is also compelling evidence from rodent studies, especially by Georgieff, Rao, and colleagues, that early iron deficiency affects neurometabolism, dendritogenesis, and long-term potentiation in the developing hippocampus [reviewed in (7)]. Felt, Schallert, Georgieff and others have observed behavioral alterations consistent with these hippocampal effects, specifically poorer spatial learning performance (21,23,34,111) and altered trace conditioning (112,113). Among many important functions of the hippocampus, it is central to recognition memory processing (114), which can be assessed in infants and children. In the Detroit sample, we used event-related potentials and found electrophysiologic indications of delayed recognition memory (115). Evidence of poorer recognition memory in the long term has been electrophysiologically obtained in the Chile sample at 10 y (C. R. Algarin, E. L. Congdon, A. Westerlund, P. D. Peirano, M. Gregas, B. Lozoff, C. A. Nelson, unpublished data) and behaviorally in the Costa Rica sample at 19 y (59).

We want to emphasize again that the findings summarized in this review are unlikely to depend solely on a given CNS region or process. The correspondence between brain and behavior is not 1-to-1. Furthermore, the brain works as an integrated system, and disruption in one process, circuit, or region can affect other systems; age and experience also play a role. For instance, there

are functional interactions between prefrontal-striatal and hippocampal systems in humans, and dopamine seems to play a critical role in successful completion of hippocampus-based memory tasks (116). In rodents, the mesocortical dopamine system also modulates hippocampal-dependent long-term potentiation (117), thereby indicating that hippocampal and prefrontal neurons are connected at the level of cell (117) and system (118) and functionally integrated (117,119). Another example is prolactin release. The regulation of prolactin is complex and includes other neurotransmitter systems such as serotonin (91,120).

Iron is required for so many CNS processes that it is reasonable to expect a variety of subtle and diffuse effects. Interconnections between neurochemistry, neuroanatomy, neurometabolism, and genomics/proteomics may be particularly important during early development, when both vulnerability and plasticity often differ from what is observed later in life. The persistence of negative outcomes on measures of executive function and recognition memory and on other sensory, motor, affective, and neuroendocrine measures highlights the need to prevent iron deficiency in infancy and to find interventions that lessen the long-term effects of this widespread nutrient disorder.

Conclusion

Tremendous strides in understanding brain/behavior relations and the effects of early iron deficiency have been made in the last few decades, but there is still much uncertainty and much more to learn. In the second 5-y period of our program project grant, we are focusing on timing of iron deficiency and outcomes after early treatment. All projects (human infants, monkey infants, and developing rats) are investigating differential effects of pre- vs. postnatal iron deficiency and differences in reversibility, depending on timing of iron deficiency and its treatment. We are also considering the potential for adverse effects with excess iron or too-rapid iron repletion. John Beard played an important role in the conception and design of the relevant rodent experiments and the energetic discussions about the program project as a whole. He will be sorely missed by all members of our group and all those who seek to understand brain and behavior effects of early deficiency.

Acknowledgments

All investigators in the first 5-year period of the Brain and Behavior in Early Iron Deficiency Program Project contributed to our thinking about dopamine-related effects in humans with early iron deficiency: Rosa Angulo-Barroso, Sandra Jacobson, Joseph Jacobson, and Charles Nelson with the human infant project; Mari Golub, Christopher Coe, and Gabriele Lubach with the monkey projects; and John Beard, James Connor, Barbara Felt, Michael Georgieff, Raghu Rao, and Timothy Schallert with the rodent project. I am also grateful for the dedicated efforts of 3 outstanding postdoctoral fellows with the Detroit infant project (Rinat Armony-Sivan, Matthew Burden, and Tal Shafir) and wonderful long-time colleagues in Costa Rica (Elias Jimenez) and Chile (Cecilia Algarin, Marcela Castillo, and Patricio Peirano). I wrote the manuscript based on the work of these and other investigators. The sole author had responsibility for all parts of the manuscript.

Literature Cited

1. Oski FA, Honig AS. The effects of therapy on the developmental scores of iron-deficient infants. J Pediatr. 1978;92:21–5.
2. Dallman PR, Siimes M, Manies EC. Brain iron: persistent deficiency following short-term iron deprivation in the young rat. Br J Haematol. 1975;31:209–15.
3. Dallman PR, Spirito RA. Brain iron in the rat: extremely slow turnover in normal rats may explain long-lasting effects of early iron deficiency. J Nutr. 1977;107:1075–81.
4. Youdim MBH. Brain iron: neurochemical and behavioural aspects. London: Taylor & Francis; 1988.
5. Beard JL, Connor JR. Iron status and neural functioning. Annu Rev Nutr. 2003;23:41–58.
6. Lozoff B, Beard J, Connor J, Felt B, Georgieff M, Schallert T. Long-lasting neural and behavioral effects of iron deficiency in infancy. Nutr Rev. 2006;64:S34–43.
7. Georgieff MK. The role of iron in neurodevelopment: fetal iron deficiency and the developing hippocampus. Biochem Soc Trans. 2008; 36:1267–71.
8. Beard J. Iron deficiency alters brain development and functioning. J Nutr. 2003;133:S1468–72.
9. Beard J. Recent evidence from human and animal studies regarding iron status and infant development. J Nutr. 2007;137:S524–30.
10. Grantham-McGregor S, Ani C. A review of studies on the effect of iron deficiency on cognitive development in children. J Nutr. 2001;131: S649–68.
11. McCann JC, Ames BN. An overview of evidence for a causal relation between iron deficiency during development and deficits in cognitive or behavioral function. Am J Clin Nutr. 2007;85:931–45.
12. Lozoff B. Iron deficiency and child development. Food Nutr Bull. 2007;28:S560–71.
13. Sachdev H, Gera T, Nestel P. Effect of iron supplementation on mental and motor development in children: systematic review of randomised controlled trials. Public Health Nutr. 2005;8:117–32.
14. Peirano PD, Algarin DR, Chamorro R, Reyes S, Garrido MI, Duran S, Lozoff B. Sleep and neurofunctions throughout child development: lasting effects of early iron deficiency. J Pediatr Gastroenterol Nutr. 2009;48:S8–15.
15. Dunnett SB, Bentivoglio M, Bjorklund A, Hokfelt Te. Dopamine. 21st vol. Amsterdam: Elsevier B.V.; 2005.
16. Dahl RE, Spears LP, editors. Adolescent brain development: vulnerabilities and opportunities. Ann N Y Acad Sci. 2004;1021:xi-458.
17. Pinero DJ, Li NQ, Connor JR, Beard JL. Variations in dietary iron alter brain iron metabolism in developing rats. J Nutr. 2000;130: 254–63.
18. Pinero D, Jones B, Beard JL. Variations in dietary iron alter behavior in developing rats. J Nutr. 2001;131:311–8.
19. Beard J, Erikson KM, Jones BC. Neonatal iron deficiency results in irreversible changes in dopamine function in rats. J Nutr. 2003;133: 1174–9.
20. Beard JL, Felt B, Schallert T, Burhans M, Connor JR, Georgieff MK. Moderate iron deficiency in infancy: biology and behavior in young rats. Behav Brain Res. 2006;170:224–32.
21. Felt BT, Beard JL, Schallert T, Shao J, Aldridge JW, Connor JR, Georgieff MK, Lozoff B. Persistent neurochemical and behavioral abnormalities in adulthood despite early iron supplementation for perinatal iron deficiency anemia in rats. Behav Brain Res. 2006;171:261–70.
22. Beard JL, Unger EL, Bianco LE, Paul T, Rundle SE, Jones BC. Early postnatal iron repletion overcomes lasting effects of gestational iron deficiency in rats. J Nutr. 2007;137:1176–82.
23. Youdim MB. Nutrient deprivation and brain function: iron. Nutrition. 2000;16:504–8.
24. Ashkenazi R, Ben Shachar D, Youdim MB. Nutritional iron and dopamine binding sites in the rat brain. Pharmacol Biochem Behav. 1982;17 Suppl 1:43–7.
25. Beard JL, Chen Q, Connor J, Jones BC. Altered monamine metabolism in caudate-putamen of iron-deficient rats. Pharmacol Biochem Behav. 1994;48:621–4.
26. Nelson C, Erikson K, Pinero DJ, Beard JL. In vivo dopamine metabolism is altered in iron-deficient anemic rats. J Nutr. 1997;127:2282–8.
27. Ben-Shachar D, Ashkenazi R, Youdim MBH. Long-term consequence of early iron-deficiency on dopaminergic neurotransmission in rats. Int J Dev Neurosci. 1986;4:81–8.
28. Erikson KM, Jones BC, Beard JL. Iron deficiency alters dopamine transporter functioning in rat striatum. J Nutr. 2000;130:2831–7.
29. Erikson KM, Jones BC, Hess EJ, Zhang Q, Beard JL. Iron deficiency decreases dopamine D1 and D2 receptors in rat brain. Pharmacol Biochem Behav. 2001;69:409–18.

30. Chen Q, Beard JL, Jones BC. Abnormal rat brain monoamine metabolism in iron deficiency anemia. J Nutr Biochem. 1995;6:486–93.
31. Youdim MB, Yehuda S. The neurochemical basis of cognitive deficits induced by brain iron deficiency: involvement of dopamine-opiate system. Cell Mol Biol. 2000;46:491–500.
32. Burhans MS, Dailey C, Beard Z, Wiesinger J, Murray-Kolb L, Jones BC, Beard JL. Iron deficiency: differential effects on monoamine transporters. Nutr Neurosci. 2005;8:31–8.
33. Connor JR, Wang XS, Neely EB, Ponnuru P, Morita H, Beard J. Comparative study of the influence of Thy1 deficiency and dietary iron deficiency on dopaminergic profiles in the mouse striatum. J Neurosci Res. 2008;86:3194–202.
34. Felt BT, Lozoff B. Brain iron and behavior of rats are not normalized by treatment of iron deficiency anemia during early development. J Nutr. 1996;126:693–701.
35. Ward KL, Tkac I, Jing Y, Felt B, Beard J, Connor J, Schallert T, Georgieff MK, Rao R. Gestational and lactational iron deficiency alters the developing striatal metabolome and associated behaviors in young rats. J Nutr. 2007;137:1043–9.
36. Beard JL, Erikson KM, Jones BC. Neurobehavioral analysis of developmental iron deficiency in rats. Behav Brain Res. 2002;134:517–24.
37. Youdim MB. Brain iron deficiency and excess: cognitive impairment and neurodegeneration with involvement of striatum and hippocampus. Neurotox Res. 2008;14:45–56.
38. Coe CL, Lubach GR, Bianco LE, Beard JL. A history of iron deficiency anemia during infancy alters brain monoamine activity later in juvenile monkeys. Dev Psychobiol. 2009;51:301–9.
39. Bjorklund A, Dunnett SB. Dopamine neuron systems in the brain: an update. Trends Neurosci. 2007;30:194–202.
40. Lozoff B, Georgieff MK. Iron deficiency and brain development. Semin Pediatr Neurol. 2006;13:158–65.
41. Seamans JK, Yang CR. The principal features and mechanisms of dopamine modulation in the prefrontal cortex. Prog Neurobiol. 2004; 74:1–58.
42. Anderson VA. Development of executive functions through late childhood and adolescence in an Australian sample. Dev Neuropsychol. 2001;20:385–406.
43. Carlsona SM, Moses LJ, Casey BJ. How specific is the relation between executive function and theory of mind? Contributions of inhibitory control and working memory. Infant Child Dev. 2002;11:73–92.
44. Brozoski TJ, Brown RM, Rosvold HE, Goldman RS. Cognitive deficit caused by regional depletion of dopamine in prefrontal cortex of rhesus monkey. Science. 1979;205:929–32.
45. Sawaguchi T, Goldman-Rakic PS. D1 dopamine receptors in prefrontal cortex: involvement in working memory. Science. 1991;251:947–50.
46. Sawaguchi T, Matsumura M, Kubota K. Effects of dopamine antagonists on neuronal activity related to a delayed response task in monkey prefrontal cortex. J Neurophysiol. 1990;63:1401–12.
47. Mehta MA, Hinton EC, Montgomery AJ, Bantick RA, Grasby PM. Sulpiride and mnemonic function: effects of a dopamine D2 receptor antagonist on working memory, emotional memory and long-term memory in healthy volunteers. J Psychopharmacol. 2005;19: 29–38.
48. Mehta MA, Sahakian BJ, McKenna PJ, Robbins TW. Systemic sulpiride in young adult volunteers simulates the profile of cognitive deficits in Parkinson's disease. Psychopharmacology (Berl). 1999;146: 162–74.
49. Casey BJ, Thomas KM, Welsh TF, Badgaiyan RD, Eccard CHJ, Jennings R, Crone EA. Dissociation of response conflict, attentional selection, and expectancy with functional magnetic resonance imaging. Proc Natl Acad Sci USA. 2000;97:8728–33.
50. Fallgatter AJ, Ehlis AC, Seifert J, Konrad WS, Scheuerpflug P, Zillessen KE, Herrmann MJ, Warnke A. Altered response control and anterior cingulate function in attention-deficit/hyperactivity disorder boys. Clin Neurophysiol. 2004;115:973–81.
51. Giedd JN, Snell JW, Lange N, Rajapasake JC, Kaysen D, Vaituzis AC, Vauss YC, Hamburger SD, Kouch PL, et al. Quantitative magnetic resonance imaging of human brain development: ages 4–18. Cereb Cortex. 1996;6:551–60.
52. Lozoff B, Brittenham GM, Wolf AW, McClish DK, Kuhnert PM, Jimenez E, Jimenez R, Mora LA, Gomez I, et al. Iron deficiency anemia and iron therapy: effects on infant developmental test performance. Pediatrics. 1987;79:981–95.
53. Lozoff B, Klein NK, Nelson EC, McClish DK, Manuel M, Chacon ME. Behavior of infants with iron deficiency anemia. Child Dev. 1998;69: 24–36.
54. Lozoff B, Jimenez E, Wolf AW. Long-term developmental outcome of infants with iron deficiency. N Engl J Med. 1991;325:687–94.
55. Corapci F, Radan AE, Lozoff B. Iron deficiency in infancy and mother-child interaction at 5 years. J Dev Behav Pediatr. 2006;27:371–8.
56. Lozoff B, Jimenez E, Hagen J, Mollen E, Wolf AW. Poorer behavioral and developmental outcome more than 10 years after treatment for iron deficiency in infancy. Pediatrics. 2000;105:E51.
57. Lozoff B, Jimenez E, Smith JB. Double burden of iron deficiency and low socio-economic status: a longitudinal analysis of cognitive test scores to 19 years. Arch Pediatr Adolesc Med. 2006;160:1108–13.
58. Army Individual Test Battery. Manual of directions and scoring. Washington, DC: Adjutant General's Office; 1944.
59. Lukowski AF, Koss M, Burden MJ, Jonides J, Nelson CA, Kaciroti N, Jimenez E, Lozoff B. Iron deficiency in infancy and neurocognitive functioning at 19 years: evidence of long-term deficits in executive function and recognition memory. Nutr Neurosci. 2010;13:54–70.
60. Algarin CR, Peirano PD, Just E, Garrido MI, Nelson CA, Lozoff B. Executive cerebral functions in former iron deficient anemic preadolescents. Pediatr Res. 2004;EPAS2004:1589.
61. Lozoff B, Clark KM, Jing Y, Armony-Sivan R, Angelilli ML, Jacobson SW. Dose-response relationships between iron deficiency with or without anemia and infant social-emotional behavior. J Pediatr. 2008; 152:696–702.
62. Shafir T, Angulo-Barroso R, Jing Y, Jacobson S, Lozoff B. Iron deficiency and infant motor development. Early Hum Dev. 2008;84:479–85.
63. Shafir T, Angulo-Barroso R, Su J, Jacobson SW, Lozoff B. Iron deficiency anemia in infancy and reach and grasp development. Infant Behav Dev. 2009;32:366–75.
64. Carter RC, Jacobson JL, Burden MJ, Armony-Sivan R, Dodge NC, Angelilli ML, Lozoff B, Jacobson SW. Iron deficiency anemia and cognitive function in infancy. Pediatrics. 2010;126:e427–34.
65. Diamond A. Development of the ability to use recall to guide action, as indicated by infants' performance on AB. Child Dev. 1985;56: 868–83.
66. Diedrich FJ, Thelen E, Smith LB, Corbetta D. Motor memory is a factor in infant perseverative errors. Dev Sci. 2000;3:479–94.
67. Bressan RA, Crippa JA. The role of dopamine in reward and pleasure behaviour: review of data from preclinical research. Acta Psychiatr Scand Suppl. 2005;14–21.
68. Wild B, Rodden FA, Grodd W, Ruch W. Neural correlates of laughter and humour. Brain. 2003;126:2121–38.
69. Oski FA, Honig AS, Helu B, Howanitz P. Effect of iron therapy on behavior performance in nonanemic, iron-deficient infants. Pediatrics. 1983;71:877–80.
70. Lozoff B, Klein NK, Prabucki KM. Iron-deficient anemic infants at play. J Dev Behav Pediatr. 1986;7:152–8.
71. Walter T, Kovalskys J, Stekel A. Effect of mild iron deficiency on infant mental development scores. J Pediatr. 1983;102:519–22.
72. Deinard AS, List A, Lindgren B, Hunt JV, Chang P-N. Cognitive deficits in iron-deficient and iron-deficient anemic children. J Pediatr. 1986;108:681–9.
73. Lozoff B, Wolf AW, Urrutia JJ, Viteri FE. Abnormal behavior and low developmental test scores in iron-deficient anemic infants. J Dev Behav Pediatr. 1985;6:69–75.
74. Honig AS, Oski FA. Solemnity: a clinical risk index for iron deficient infants. Early Child Dev Care. 1984;16:69–84.
75. Lozoff B, De Andraca I, Castillo M, Smith J, Walter T, Pino P. Behavioral and developmental effects of preventing iron-deficiency anemia in healthy full-term infants. Pediatrics. 2003;112:846–54.
76. Wachs TD, Pollitt E, Cuerto S, Jacoby E, Creed-Kanishiro H. Relation of neonatal iron status to individual variability in neonatal temperament. Dev Psychobiol. 2005;46:141–53.
77. Golub MS, Hogrefe CE, Tarantal AF, Germann SL, Beard JL, Georgieff MK, Calatroni A, Lozoff B. Diet-induced iron deficiency anemia and pregnancy outcome in the rhesus monkey. Am J Clin Nutr. 2006;83: 647–56.
78. Golub MS, Hogrefe CE, Germann SL. Iron deprivation during fetal development changes the behavior of juvenile rhesus monkeys. J Nutr. 2007;137:979–84.

79. Golub MS, Hogrefe CE, Germann SL, Capitano JL, Lozoff B. Behavioral consequences of developmental iron deficiency in infant rhesus monkeys. Neurotoxicol Teratol. 2006;28:3–17.
80. Golub MS, Hogrefe CE, Widaman KF, Capitanio JP. Iron deficiency anemia and affective response in rhesus monkey infants. Dev Psychobiol. 2009;51:47–59.
81. Angulo-Kinzler RM, Peirano P, Lin E, Algarin C, Garrido M, Lozoff B. Twenty-four-hour motor activity in human infants with and without iron deficiency anemia. Early Hum Dev. 2002;70:85–101.
82. Aldridge JW, Berridge KC. Coding of serial order by neostriatal neurons: a "natural action" approach to movement sequence. J Neurosci. 1998;18:2777–87.
83. Lehericy S, Benali H, Van de Moortele PF, Pelegrini-Issac M, Waechter T, Ugurbil K, Doyon J. Distinct basal ganglia territories are engaged in early and advanced motor sequence learning. Proc Natl Acad Sci USA. 2005;102:12566–71.
84. Scholz VH, Flaherty AW, Kraft E, Keltner JR, Kwong KK, Chen YI, Rosen BR, Jenkins BG. Laterality, somatotopy and reproducibility of the basal ganglia and motor cortex during motor tasks. Brain Res. 2000;879:204–15.
85. Vink M, Kahn RS, Raemaekers M, van den Heuvel M, Boersma M, Ramsey NF. Function of striatum beyond inhibition and execution of motor responses. Hum Brain Mapp. 2005;25:336–44.
86. Lozoff B, Armony-Sivan R, Kaciroti N, Jing Y, Golub M, Jacobson SW. Eye-blinking rates are slower in infants with iron-deficiency anemia than in non-anemic iron-deficient or iron-sufficient infants. J Nutr. 2010;140:1057–61.
87. Lawrence MS, Redmond DE Jr. MPTP lesions and dopaminergic drugs alter eye blink rate in African green monkey. Pharmacol Biochem Behav. 1991;38:869–74.
88. Karson CN. Physiology of normal and abnormal blinking. Adv Neurol. 1988;49:25–37.
89. Elsworth JD, Lawrence MS, Roth RH, Taylor JR, Mailman RB, Nichols DE, Lewis MH, Redmond DE Jr. D1 and D2 dopamine receptors independently regulate spontaneous blink rate in the vervet monkey. J Pharmacol Exp Ther. 1991;259:595–600.
90. Benker G, Jaspers C, Hausler G, Reinwein D. Control of prolactin secretion. Klin Wochenschr. 1990;68:1157–67.
91. Ben-Jonathan N, Hnasko R. Dopamine as a prolactin (PRL) inhibitor. Endocr Rev. 2001;22:724–63.
92. Barkey RJ, Ben-Shachar D, Amit T, Youdim MBH. Increased hepatic and reduced prostatic prolactin (PRL) binding in iron deficiency and during neuroleptic treatment: correlation with changes in serum PRL and testosterone. Eur J Pharmacol. 1985;109:193–200.
93. Barkey RJ, Amit T, Ben-Shachar D, Youdim MBH. Characterization of the hepatic prolactin receptors induced by chronic iron deficiency and neuroleptics. Eur J Pharmacol. 1986;122:259–67.
94. Lozoff B, Felt BT, Nelson EC, Wolf AW, Meltzer HW, Jimenez E. Serum prolactin levels and behavior in infants. Biol Psychiatry. 1995; 37:4–12.
95. Felt B, Jimenez E, Smith J, Calatroni A, Kaciroti N, Wheatcroft G, Lozoff B. Iron deficiency in infancy predicts altered serum prolactin response 10 years later. Pediatr Res. 2006;60:513–7.
96. Gunnar M, Quevedo K. The neurobiology of stress and development. Annu Rev Psychol. 2007;58:145–73.
97. Gunnar MR, Vazquez DM. Low cortisol and a flattening of expected daytime rhythm: potential indices of risk in human development. Dev Psychopathol. 2001;13:515–38.
98. Gunnar MR, Frenn K, Wewerka SS, Van Ryzin MJ. Moderate versus severe early life stress: associations with stress reactivity and regulation in 10–12-year-old children. Psychoneuroendocrinology. 2009;34:62–75.
99. Dimitrijevic M, Tao M, Lozoff B, Felt BT. Iron deficiency in infancy lowers serum cortisol levels. Pediatr Res. E-PAS2008:4310.8.
100. Siddappa AJ, Rao R, Wobken JD, Casperson K, Liebold EA, Connor J, Georgieff M. Iron deficiency alters iron regulatory protein and iron transport protein expression in the perinatal rat brain. Pediatr Res. 2003;53:800–7.
101. Clardy SL, Wang X, Zhao W, Liu W, Chase GA, Beard JL, Felt BT, Connor JR. Acute and chronic effects of developmental iron deficiency on mRNA expression patterns in the brain. J Neural Transm Suppl. 2006;173–96.
102. Wang X, Wiesinger J, Beard J, Felt B, Menzies S, Earley C, Allen R, Connor J. Thy1 expression in the brain is affected by iron and is decreased in Restless Legs Syndrome. J Neurol Sci. 2004;220:59–66.
103. Li YY, Wang HM, Wang WG. [The effect of iron deficiency anemia on the auditory brainstem response in infant]. [Chinese]. Zhonghua Yi Xue Za Zhi. 1994;74:367–9.
104. Roncagliolo M, Garrido M, Walter T, Peirano P, Lozoff B. Evidence of altered central nervous system development in infants with iron deficiency anemia at 6 mo: delayed maturation of auditory brain stem responses. Am J Clin Nutr. 1998;68:683–90.
105. Cankaya H, Oner AF, Egeli E, Caksen H, Uner A, Akcay G. Auditory brainstem response in children with iron deficiency anemia. Acta Paediatr Taiwan. 2003;44:21–4.
106. Amin SB, Orlando M, Eddins A, MacDonald M, Monczynski C, Wang H. In utero iron status and auditory neural maturation in premature infants as evaluated by auditory brainstem response. J Pediatr. 2010; 156:377–81.
107. Algarin C, Peirano P, Garrido M, Pizarro F, Lozoff B. Iron deficiency anemia in infancy: Long-lasting effects on auditory and visual systems functioning. Pediatr Res. 2003;53:217–23.
108. Rao R, Tkac I, Townsend EL, Gruetter R, Georgieff MK. Perinatal iron deficiency alters the neurochemical profile of the developing rat hippocampus. J Nutr. 2003;133:3215–21.
109. Hill JM. The distribution of iron in the brain. In: Youdim MBH, editor. Brain iron: neurochemical and behavioural aspects. London: Taylor and Francis; 1988.
110. Li D. Effects of iron deficiency on iron distribution and y-aminobutyric acid (GABA) metabolism in young rat brain tissues. Hokkaido Igaku Zasshi. 1998;73:215–25.
111. Carlson ES, Tkac I, Magid R, O'Connor MB, Andrews NC, Schallert T, Gunshin H, Georgieff MK, Petryk A. Iron is essential for neuron development and memory function in mouse hippocampus. J Nutr. 2009;139:672–9.
112. McEchron MD, Cheng AY, Liu H, Connor JR, Gilmartin MR. Perinatal nutritional iron deficiency permanently impairs hippocampus-dependent trace fear conditioning in rats. Nutr Neurosci. 2005; 8:195–206.
113. Gewirtz JC, Hamilton KL, Babu MA, Wobken JD, Georgieff MK. Effects of gestational iron deficiency on fear conditioning in juvenile and adult rats. Brain Res. 2008;1237:195–203.
114. Nelson CA. The ontogeny of human memory: a cognitive neuroscience perspective. Dev Psychol. 1995;31:723–38.
115. Burden MJ, Westerlund AJ, Armony-Sivan R, Nelson CA, Jacobson SW, Lozoff B, Angelilli ML, Jacobson JL. An event-related potential study of attention and recognition memory in infants with iron-deficiency anemia. Pediatrics. 2007;120:e336–45.
116. Takahashi H, Kato M, Hayashi M, Okuba Y, Takano A, Ito H, Subara T. Memory and frontal lobe functions; possible relations with dopamine D^2 receptors in the hippocampus. Neuroimage. 2007;34:1643–9.
117. Gurden H, Tassin JP, Jay TM. Integrity of the mesocortical dopaminergic system is necessary for complete expression of in vivo hippocampal-prefrontal cortex long-term potentiation. Neuroscience. 1999;94:1019–27.
118. Thierry AM, Gioanni Y, Degenetais E, Glowinski J. Hippocampo-prefrontal cortex pathway: anatomical and electrophysiological characteristics. Hippocampus. 2000;10:411–9.
119. Gurden H, Takita M, Jay TM. Essential role of D1 but not D2 receptors in the NMDA receptor-dependent long-term potentiation at hippocampal-prefrontal cortex synapses in vivo. J Neurosci. 2000;20: RC106.
120. Albinsson A, Palazidou E, Stephenson J, Andersson G. Involvement of the 5–HT2 receptor in the 5-HT receptor-mediated stimulation of prolactin release. Eur J Pharmacol. 1994;251:157–61.