Role of Early Hormonal and Nutritional Experiences in Shaping Feeding Behavior and Hypothalamic Development

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Abstract

Obesity in adults and children is increasingly becoming a major health problem worldwide. However, the precise biological mechanisms governing this disease have not been fully elucidated. Obesity involves the complex interaction of a wide range of environmental and genetic factors. Additionally, there is now a growing body of evidence suggesting that alterations in metabolic environment during important periods of organ development can predispose individuals to later development of obesity and diabetes. Maternal obesity or malnutrition during pregnancy increases the risk for metabolic disorders (including obesity) in the offspring. Similarly, early postnatal overnutrition also predisposes offspring to adult obesity. The hypothalamus appears to play an essential role in controlling appetite. It undergoes a tremendous growth during early stages of brain development, a period when it is particularly susceptible to environmental insults. In particular, there is growing appreciation that developmental programming of brain feeding pathways by the perinatal environment represents a possible cause for obesity in later life. Maternal obesity and diabetes during pregnancy increase the risk of obesity and type II diabetes in offspring. This observation is particularly troublesome, because it suggests that daughters of obese women may themselves be vulnerable to becoming obese and may be more likely to have offspring who share this vulnerability. Persistent maternal malnutrition or inadequate placental function during pregnancy also increases offspring’s susceptibility to develop obesity and associated disorders. Importantly, interference with 1 or more of these risk factors can lead to higher susceptibility to develop obesity. Maternal malnutrition and low birth weight followed by accelerated newborn-to-adolescent weight gain exacerbate the risk for obesity in adult life. The fact that similar metabolic outcomes are observed in the offspring of obese and malnourished mothers suggests that the programmed obesity observed in these individuals may be tied to the same physiological mechanisms.

Introduction

Over the past few decades, the prevalence of obesity has increased at an alarming rate in developing countries that have adopted a Western lifestyle involving decreased physical activity and overconsumption of energy-dense food. In addition to adversely affecting quality of life for affected patients, epidemiological studies have indicated that present rates of obesity will have a dramatic impact on life expectancy. Although there are some obvious sociological factors that contribute to the current rise in obesity, the precise biological mechanisms governing this disease have not been fully elucidated. In addition, it is not clear why different individuals exhibit different propensities for obesity. Numerous studies have demonstrated that developmental exposure of animals or humans to a variety of nutritional and metabolic insults (e.g., maternal obesity, diabetes, undernutrition, and early postnatal overnutrition) may result in predisposition to obesity in later life. Maternal obesity and diabetes during pregnancy increase the risk of obesity and type II diabetes in offspring. This observation is particularly troublesome, because it suggests that daughters of obese women may themselves be vulnerable to becoming obese and may be more likely to have offspring who share this vulnerability. Persistent maternal malnutrition or inadequate placental function during pregnancy also increases offspring’s susceptibility to develop obesity and associated disorders. Importantly, interference with 1 or more of these risk factors can lead to higher susceptibility to develop obesity. Maternal malnutrition and low birth weight followed by accelerated newborn-to-adolescent weight gain exacerbate the risk for obesity in adult life. The fact that similar metabolic outcomes are observed in the offspring of obese and malnourished mothers suggests that the programmed obesity observed in these individuals may be tied to the same physiological mechanisms.

There is general recognition that the developing brain is more susceptible to environmental insults than the adult brain. The hypothalamus is a region of the brain critical for regulation of homeostatic processes such as feeding and glucose homeostasis. To accomplish these complex physiological regulations, the hypothalamus...
been given to neurons of the arcuate nucleus of the hypothalamus (ARH). One subpopulation of arcuate neurons coexpresses neuropeptide Y (NPY) and agouti-related peptide (AgRP). Another subset of ARH neurons produces pro-opiomelanocortin (POMC). Both AgRP/NPY and POMC neurons are direct targets of leptin and insulin. Leptin and insulin activate POMC neurons and inhibit NPY neurons to reduce food intake and increase energy expenditure (19,22). These neurons send extensive projections to other parts of the hypothalamus, including the paraventricular nucleus (PVH), the dorsomedial nucleus (DMH), and the LHA, where they release their peptides to regulate energy balance. Therefore, leptin and insulin are particularly well suited to communicate nutrient availability in the environment to the “metabolic” hypothalamus during development.

**Periods of sensitivity for hypothalamic development**

Sensitive periods during the development of the brain are periods during which the brain is particularly sensitive to a variety of environmental exposures; these periods exist due to the temporal and regional emergence of critical developmental processes (i.e. cell proliferation, migration, differentiation, axon growth, synaptogenesis, and apoptosis). Evidence from numerous studies indicates that brain development extends from the embryonic period through adolescence. In general, the sequence of events is comparable among species, although the time scales vary considerably. The first important period for hypothalamic development is the period during which the hypothalamus begins to form, with neurogenesis occurring in the proliferative zone of neuroepithelial cells lining the 3rd cerebral ventricle. Birthdating studies employing administration of 5-bromo-2-deoxyuridine or [3H]thymidine at various stages of embryonic development indicate that hypothalamic neuronal proliferation occurs primarily during mid-gestation in rodents. In mice, the majority of hypothalamic neurons located in hypothalamic nuclei known to control energy homeostasis are born between embryonic d 12 (E12) and E14 (L. Ishii and S. Bouret, unpublished data). Rats exhibit a relatively longer neurogenic period, but most hypothalamic neurons are born between E12 and E17 in this species (23). There follows a sequence of developmental processes, including migration and differentiation, which appears to occur primarily during late embryonic development in rodents (23). Interruption of hypothalamic development during these important developmental periods can result in severe structural and functional abnormalities of the hypothalamus. For example, haploinsufficiency in Sim1, a transcription factor expressed in the PVH during embryonic development, causes structural and functional abnormalities of the hypothalamus associated with hyperphagia and obesity (24). Similarly, loss of the steroidogenic factor SF-1 results in a failure of ventromedial hypothalamic nuclei (VMH) neurons to differentiate and to project to other brain regions (25). The second important developmental period for hypothalamic development in rodents is during the first weeks of postnatal life, when neurons send their axonal projections to their target sites. In both mice and rats, projections from the ARH are immature at birth and develop mainly during wk 2 of life (26,27). Various parts of the hypothalamus, however, develop at different times. Efferent projections from the DMH and VMH develop prior to those from the ARH and are established by postnatal d (P) 6 (26). The normal ontogeny of hypothalamic development in rodents appears to differ from that in humans. In general, regional development of the rodent hypothalamus proceeds on a timeline of days compared with weeks to months in humans. In addition, although rodents display considerable postnatal hypothalamic development, hu-
mans undergo considerably more prenatal maturation of hypothalamic structures. Thus, whereas the hypothalamus appears largely immature until near the end of the first 3 wk of life in rodents, in primates, hypothalamic maturati-
on occurs primarily in utero. Hypothalamic cell proliferation is ini-
tiated during the first quarter of gestation in humans and develop-
ment of hypothalamic neural projections (precisely the projec-
tions from arcuate AgRP/NPY neurons) occurs as early as gestational d 100 (i.e., late second trimester of gestation) in nonhuman primates (28,29). Different species may therefore exhibit different periods of sensitivity based on the temporal and regional maturation patterns of the hypothalamus.

Neurotrophic signals orchestrating hypothalamic development

The array of molecules involved in modulating brain develop-
ment (neurogenesis, cell migration, axonal growth, and synapto-
genesis) is substantial. Hormones appear to exert a particularly important neurotrophic function in the developing hypothalamus. The involvement of steroid hormones in hypothalamic development has been known for decades [see (30) for review] and recent findings also show that metabolic hormones such as leptin may act as major neurotrophic signals during perinatal hypothalamic maturation. Leptin deficiency in mice results in abnormal development of the axonal projections from the ARH to other parts of the hypothalamus (Fig. 2) (31). Whereas numerous fibers from the ARH innervate the PVH on P12 in wild-type mice, only a few ARH axons are observed there in leptin-deficient (Lep<sup>ob/ob</sup>/Lep<sup>ob/ob</sup>) littermates. Furthermore, the dis-
ruption of ARH pathways in Lep<sup>ob/ob</sup>/Lep<sup>ob/ob</sup> mice appears to be permanent, because even at P60, a stage considered as mature in mice, there are fewer ARH fibers in each terminal field. The site of action for the developmental effects of leptin is unknown, but the effects appear to include direct action on ARH neurons. Application of leptin to isolated organotypic explants of the ARH can induce neurite extension. During neonatal life, leptin therefore appears to act as a trophic signal, directing key developmental events in the same hypothalamic pathways that will convey leptin signals in mature mice. However, not all regions that express leptin receptors respond to the trophic action of leptin. The DMH contains a substantial density of neurons that express leptin receptors, yet its projections to the PVH appear to be normal in Lep<sup>ob/ob</sup>/Lep<sup>ob/ob</sup> mice (31). Other metabolic signals besides leptin, such as insulin and ghrelin, may also exert profound neurotrophic effects on the developing hypothalamus. For example, postnatal injections of insulin have been associated with morphological changes in the VMH (32). Metabolic hormones may also act in concert to direct hypothalamic growth. This idea is supported by the pioneer work of Torran-Allerand et al. (33). More than 2 decades ago, these workers found that incubation of explants from fetal hypothalamus with insulin induces neuritic extension. Incubation with estradiol alone induces similar neurotrophic effects. However, sequential incubation with insulin and estrogen produces a synergetic enhancement of hypothalamic neurite outgrowth that is stronger than the additive impact of the 2 hormones acting independently (33). Because insulin and leptin share common intracellular signal transduction pathways (34), it is therefore possible that these 2 metabolic hormones may also act synergistically to mediate their neurotrophic effects on the hypothalamus.

Periods of maximal sensitivity to trophic factors

Many of the key events that occur during development of functional neural systems are particularly sensitive to developmental cues that arise during early developmental periods. Consistent with this idea, leptin acts primarily during a restricted neonatal critical period to promote formation of neural projections from the ARH to other hypothalamic nuclei; this developmental activity is greatly diminished in adult mice (31). However, leptin can still affect brain plasticity in adults by causing synaptic rearrangement of excitatory and inhibitory inputs on arcuate neurons (35), suggesting that these circuits remain relatively plastic throughout life. In addition, in terms of neurogenic responses, mature hypothalamic neurons still appear sensitive to external cues. For instance, injections of ciliary neurotrophic factor in adult obese mice induce marked neurogenesis in the hypothalamus (36). These findings support the idea that the hypothalamus remains relatively sensitive to neurotrophic cues during adult life. However, both the degree and the nature of hypothalamic neuroplastic responses may differ between adults and neonates.

![FIGURE 2 Neurotrophic action of leptin on hypothalamic feeding pathways. In wild-type mice, the ARH sends massive projections to discrete populations of neurons located in the DMH and PVH of the hypothalamus and in the LHA. Each of these cell groups plays a major role in the neural control of feeding. During early postnatal life, leptin appears to act as an important neurotrophic factor that directs the formation of hypothalamic feeding circuits. Mice lacking leptin display permanent disruption of projections from the ARH to each of its target sites. Leptin acts primarily during a restricted neonatal critical period to promote formation of hypothalamic neural projections; this developmental process appears greatly diminished in adult mice. The lower and upper panels show a schematic illustration of a coronal section at the level of the ARH and PVH, respectively. me, Median eminence; V3, 3rd ventricle. Schematic illustrations are based on Brain Maps: Structure of the Rat Brain (49).](https://academic.oup.com/jn/article-abstract/140/3/653/4600438)
Developmental influences of maternal and early postnatal nutrition on hypothalamic feeding pathways

Exposure to environmental metabolic insults is most likely to cause adverse effects when exposure occurs during development of the organ controlling the specific metabolic process that is affected. As described above, the developing hypothalamus is exposed to 2 major successive environments: one in utero, the other postnatally. These periods therefore represent important time windows during which alteration of the metabolic and hormonal environment may lead to abnormal hypothalamic development. Studies with animal models of maternal obesity have clearly demonstrated that prenatal nutrition influences the programming of hypothalamic appetite networks. High-fat feeding throughout pregnancy and lactation results in hyperphagic and obese offspring associated with dysregulation of hypothalamic gene expression (8,37). In addition to its adverse effects on expression of appetite-regulating genes, a maternal high-fat diet also affects central leptin sensitivity. Administration of leptin in rats born to obese dams results in reduced phosphorylated form of Signal Transducer and Activator of Transcription 3 (pSTAT3) staining, a marker of leptin receptor activation, in the ARH (37,38). As a result of this diminished leptin sensitivity, rats born to obese dams also show attenuated development of the neural projections from the ARH to the PVH (37).

A number of reports have also focused on the effects of maternal undernutrition on the growth of the offspring. Animals born to undernourished dams show a strikingly similar phenotype to that of pups born to obese mothers, i.e. an increased predisposition to develop obesity [see (5,39) for review]. However, in contrast to maternal obesity, which causes hyperleptinemia throughout the postnatal and adult life (37), prenatal underfeeding resulted in reduced leptin levels during the postnatal period (40). Nevertheless, similar to observations made in offspring of obese dams, prenatal underfeeding was also associated with a reduced anorectic effect of leptin and an altered hypothalamic response to leptin, as evidenced by decreased leptin-induced cFos immunoreactivity (a marker of neuronal activation) in the PVH (40). These data suggest that correct levels of leptin during neonatal life are required for normal energy balance regulation and hypothalamic function. Remarkably, daily leptin treatment from P3 to P13 in prenatally energy-restricted rats normalized their energy intake, body weight, adiposity, and plasma glucose (41). This observation implies that metabolic (mal)-programming is potentially reversible upon injection of leptin during specific phases of developmental plasticity. Whether the reprogramming effects of leptin are caused by rewiring of the hypothalamus or by effects on the plasticity of other organs remains to be determined.

In addition to environmental factors, genetic predispositions also appear to be important contributors to obesity risk. A recent twin study suggested that up to 70% of the differences between individuals with respect to body weight can be attributed to genetics (42). Obviously, genetics alone cannot explain why obesity has increased so rapidly in recent decades. Genetic predispositions may, rather, synergistically interact with a Western dietary pattern (the wide availability of energy-dense food) in determining obesity risk. Rodent models of obesity can provide valuable insights into the biological processes underlying the development of obesity in humans. Rats genetically prone to develop diet-induced obesity are particularly suited to the task, because their tendency to become overweight shares several features with human obesity, including contributions from multiple genes (43). Rats with genetic susceptibility to development of diet-induced obesity display reduced leptin sensitivity prior to development of the obese phenotype (44–46). In these rats, diminished leptin action is characterized by decreased expression of mRNA for the long form of leptin receptor and is associated with reduced 125I-leptin binding and leptin receptor signaling in hypothalamic nuclei known to mediate the anorectic actions of leptin, including the ARH (47,48). Importantly, diminished central leptin sensitivity in these rats occurs as early as P10, i.e. the period when leptin exerts its maximal neurotrophic effects on ARH neural projections. Consistent with these observations, diet-induced obesity rats showed attenuated development of neural projections from the ARH to the PVH (48). These findings suggest that in addition to environmental factors, genetic predispositions to obesity may also cause developmental malprogramming of neural pathways involved in feeding and energy balance.

In conclusion, very clear evidence exists that nutritional changes during important periods of development can have lasting and potentially irreversible effects on feeding and metabolism. The hypothalamus is capable of sensing subtle changes in the hormonal milieu and develops over a relatively long period of time extending from the embryonic period throughout postnatal life. This extended developmental period tends to make the hypothalamus extremely sensitive to environmental metabolic cues. The precise mechanisms by which environmental factors influence hypothalamic development and subsequent function remain largely unknown. However, identification of perturbations of normal developmental processes by environmental factors has led us to the recognition that a developmental insult can initiate a cascade of alterations that may not be detected structurally or functionally until much later in life. These effects may be manifested at a time far removed from the critical time window during which exposure occurred. A better understanding of the mechanisms mediating hypothalamic functional misorganization, obtained through study of experimental models of programmed obesity, will further our understanding of the developmental origin of adult metabolic diseases, improve our ability to predict adverse outcomes in animals, and assist in the extrapolation of these adverse outcomes to risks to human health.

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Literature Cited