Fetal Programming, Epigenetics, and Adult Onset Disease

Robert H. Lane, MD, MS

INTRODUCTION

Significant early life events program individuals toward adult health and disease. Epidemiologic evidence exists for this concept within communities across the globe as well as developed and developing countries. Biological programming is defined as the process in which cells develop, function, and adapt to the environment in response to an entrained set of executable commands, often emanating from the cell’s chromatin. Cells normally contain these programs. Significant early life events reformulate these programs, likely as adaptations to ensure early survival at the price of later disease.

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Department of Pediatrics, Children’s Hospital of Wisconsin, Medical College of Wisconsin, Suite 720, PO Box 1997, Milwaukee, WI 53201-1997, USA

E-mail address: rlane@mcw.edu

KEYWORDS

• DNA methylation • Developmental origins of disease • Epigenetics • Food desert • Histone covalent modifications • Insulin growth factor 1 • Insulin resistance • Obesity

KEY POINTS

• Early life events program the occurrence of significant adult diseases, including obesity and insulin resistance.
• Although our understanding started with issues that seem remote now, relevant current issues such as food deserts and prematurity continue to make programming a priority if adult disease is to be prevented, as opposed to treating it.
• Environmental epigenetics describes how our chromatin adapts to surroundings and is a likely mechanism central to programming.
• Environmental epigenetics involves changes to chromatin structure that changes the DNA accessibility as well as nontranslated RNAs.
• A key characteristic of environmental epigenetics is that it can be manipulated, so that it is a potential target for both personalized medicine and population health.

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EARLY LIFE PROGRAMMING PROVIDES A CONCEPTUAL INFRASTRUCTURE FOR HOW PEDIATRIC NEEDS TO CHANGE

The diseases associated with programming include those that cost the most in terms of human suffering and resources. The concept that early life events predict adult health and disease guides everything we do as a pediatric community. Moreover, this concept touches all 4 legs of the stool on which academic pediatrics stands.

- In term of education, young clinicians need to actively consider later life consequences of clinical interventions, and not just the immediate consequences. This statement is particularly true considering the paucity of prospective data informing us on how common interventions in the neonatal intensive care (NICU) affect long-term outcomes.
- In terms of research, investigators need to anticipate later life health and disease outcomes in the planning of their studies. Present funding fails to support this anticipation, but the presence of conceptual investigational infrastructure that accounts for later life health and disease outcomes keeps options open for key questions.
- In terms of community health, health care organizations need to think proactively on not only how to treat a community’s disease but also how to maintain and sustain a community’s health. This proactive thinking requires the removal or at least the moderation of environmental elements that prevent and threaten community health before disease becomes tenured.
- In terms of clinical care, clinicians need to act on the truism that preventing disease improves individual and community health more than treating disease (Box 1). A challenge to this truism resides in the reality that we do not know

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Box 1
A community pediatrician’s dilemma

Johnny is a reasonably happy 6 year old. His height is 44 inches (10th percentile), and his weight is 50 pounds (75th percentile). His past medical history is remarkable for being a 28-week former premature infant. His course in the NICU was relatively benign, with the most significant events including a few days of bubble CPAP, TPN, and antibiotics. He was discharged home to his single mother, who is working an hourly job. She cannot afford a car, and as such, she is limited to public transportation. He and his mother live in an economically depressed neighborhood, and they are the third generation of his family to live in this neighborhood. His mother is sincerely motivated to support positive changes for her family. She is involved in Johnny’s care, and she can verbalize the many risks associated with prematurity.

Running 15 minutes behind schedule, you enter the room to discover Johnny eating a bag of chips and drinking a large sugared soda. You are discouraged, but you try to understand why your efforts to encourage more healthy behaviors seem to be failing.

You learn that Johnny and his mother live in a neighborhood characterized by high rates of violent crime and school dropout. There are no supermarkets within walking distance; only stores that sell a few very expensive items of produce or calorie-dense low nutrition food are near their residence. She walks with him to school in the morning, but her job does not allow her to often be home after school. Fearing for his safety, she entices Johnny with video games to come home right away after school and to stay inside. This keeps him to some extent from the violence inherent to this neighborhood. For Johnny’s mother to be home, let alone make his appointments, she must take off work. Because she is an hourly worker, she loses pay and opportunities for advancement if she takes off. Johnny is presently eating chips because he was hungry because he missed lunch to catch the bus to clinic, and his mother obtained snacks from the vending machine in the lobby of the clinic. Moreover, the snacks serve as a reward for Johnny to be willing to come to clinic without a fuss (because Johnny really likes school).
how to do that yet secondary to the lack of clinically relevant prospective long-term studies, the broad continuum of the human experience, and the mathematical challenge of predicting multifactorial diseases. The mathematical challenge exists secondary to the matrixed set of confounding factors that impact the human experience, such as genetics, current environment, and family exposures across generations. Despite this challenge, a foundation exists that provides insight into the relevance and scope of the problems associated with fetal programming.

EARLY LIFE PROGRAMMING OCCURS SECONDARY TO SIGNIFICANT ENVIRONMENTAL EXPOSURES

Large epidemiologic studies and cohorts that involve multiple generations provide the foundation for understanding fetal programming. These initial studies focus on how maternal malnutrition, as an environmental exposure from a fetal stand point, program adult health and disease.6–11 These studies take advantage of spontaneous and sometimes tragic population-wide “experiments” induced by extreme environmental exposures (Table 1). Subsequently, these “malnutrition”-based studies provide utility by demonstrating common themes that build a conceptual foundation on which to understand fetal programming. These themes also become evident when looking at other environmental exposures, such as maternal stress, maternal smoking, pollution, and toxic exposures.

- Maternal malnutrition, usually defined by insufficient intake of macronutrients or micronutrients, affects adult health of the offspring regardless of the country of origin or ethnicity. Although the impact may differ based on the country, likely because of confounding environmental and genetics factors, an impact still occurs. Two concepts are basic to interpreting these studies as well as future studies in the field. The first concept is that most of these studies use birth weight as a surrogate measure of maternal malnutrition, as opposed to direct measure of maternal intact (the “Dutch Famine” studies are a population-wide exception). The second concept is that the definition of maternal malnutrition includes excess and not just deprivation.
- Maternal malnutrition’s timing during the gestation determines the impact on offspring adult health (Fig. 1). For example, in the Dutch Famine, offspring experiencing maternal malnutrition early in gestation suffer from an increased incidence of coronary artery disease, hypertension, and obesity as adults.12

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<th>Table 1</th>
<th>Tragic population—wide exposures</th>
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<tr>
<td>Name</td>
<td>Country</td>
</tr>
<tr>
<td>Dutch Famine</td>
<td>Netherlands</td>
</tr>
<tr>
<td>Leningrad Siege</td>
<td>Russia</td>
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<tr>
<td>Occupation of Guernsey</td>
<td>UK Channel Islands</td>
</tr>
<tr>
<td>Great Chinese Famine (Three Bitter Years)</td>
<td>China</td>
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Offspring experiencing maternal malnutrition during mid gestation suffer from an increased incidence of obstructive airway disease. Finally, offspring experiencing maternal malnutrition during late gestation suffer from insulin resistance. The absolute relationship between timing of the malnutrition during the gestation and outcome differs between studies, communities, and countries, but the theme remains true.

Maternal malnutrition affects the adult health of more than one organ system. Indeed, all of the morbidities noted in the previous statement require interactions of multiple organ systems. For example, in terms of coronary artery disease, this morbidity results from an interaction of the coronary arteries themselves, the immune system, and the liver (through its regulation of serum lipid homeostasis).

Maternal malnutrition affects adult morbidities, such as coronary artery disease, insulin resistance, and obesity, which are among the most costly on a population basis. Indeed, obesity owns the nickname of “public health enemy number one” in the United States by extracting greater than $147 billion a year in health care expenditures.

Health care expenditures toward diseases such as obesity affect the economies of all countries, diverting resources away from social improvement that may lead to prevention. As we grow into a “world” economy, an increase in adult morbidities due to fetal programming in one country distributes the impact across borders. A potential example of this exists in aftershock of the “Chinese Famine.” The Chinese Famine lasted 3 years from 1958 to 1961. Conservatively, 30 million deaths occurred during this time. The Chinese Famine differs from the Dutch Famine by spanning a longer period of time, affecting a population already struggling under the weight of chronic malnutrition, and varying across regions in terms of severity.

Subsequently, the impact of the Chinese Famine reaches extremes that the Dutch Famine fails to reach. On a population-wide basis, decreased adult height and neuro-developmental outcomes characterize first-generation adults who survived in utero during the Chinese Famine. Greater than normal birth weights characterized second-generation grandchildren. The impact of the Chinese Famine stretches across generations to increase the incidence of large-for-gestational age infants (LGA). LGA predisposes toward adult insulin resistance and obesity in affected individuals. Moreover, women suffering from insulin resistance and obesity are more likely to give birth to an LGA infant. Considering the millions of individuals impacted by the famine, the cost in terms of health and health care expenditures for this second generation and the subsequent third generation will be felt across the world economy, diverting essential resources from other priorities.

**Fig. 1.** Compilation of adult disease predisposition with trimester of affected pregnancy from the Dutch Famine. Although variation in the timing of maternal malnutrition and offspring predisposition’s exists, the important concept is that the timing of the insult is an important variable in offspring outcome.
Diversion of resources also needs to occur in the United States if we are to learn from the Chinese Famine. This diversion needs to occur in 2 arenas. The first arena exists with the concept of food deserts. The second arena exists in our growing population of former premature infants.

Food deserts play a role perpetrating the racial disparities in health that occur in the United States, such as with obesity (http://www.cdc.gov/obesity/data/adult.html) (Table 2). The United States Department of Agriculture defines a food desert as “a census tract with a substantial share of residents who live in low-income areas that have low levels of access to a grocery store or healthy affordable food retail outlet.” In New York City, the most severe food deserts exist within East and Central Harlem as well as North and Central Brooklyn. These communities contain a high proportion of low-income African Americans. In contrast, food oases within New York City exist in the Upper East Side, which contains middle and upper income families.

Children stranded within a food desert suffer from an increased risk of obesity. Children that live in communities enduring higher priced fruits and vegetables develop higher body mass indexes (BMIs) than children that live in communities with lower priced produce. A 10% increase in the price of produce associates with a 0.7% increase in childhood BMI. For the community pediatrician, the realities of the food desert go beyond the abstract nature of statistics because it predicts both present and future health. Childhood obesity predicts adult coronary artery disease, hypertension, and adult onset obesity. Maternal and paternal obesity predisposes toward obesity in their offspring, although the statistical link is stronger for maternal obesity. Maternal obesity increases the risk for preeclampsia and gestational diabetes. Moreover, maternal obesity increases the risk for spontaneous preterm birth, although the association is complex and involves ethnicity, parity, and environmental factors such as smoking.

Preterm births still account for approximately 12.5% of deliveries in the United States. Medical care services of premature infants cost the United States approximately $26.2 billion in 2005. Indirect costs involving early intervention services and lost household productivity additionally cost the United States another $7 billion. It is suspected that costs are not less in 2014. Moreover, evidence continues to slowly accumulate, suggesting that prematurity programs toward adult diseases. In referencing programming, pathophysiologies that involve direct cell damage are excluded, such as what happens with hypoxia-ischemia encephalopathy. Our insight into the relevant programming remains foggy secondary to the relative youth of neonatology as a field, the temporal and regional heterogeneity of neonatal practice, and the founders of environment and genetics. Nevertheless, a couple of themes pierce the fog, albeit bluntly.

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<th>Ethnic Group</th>
<th>Incidence of Obesity (Age-Adjusted Rates) (%)</th>
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<tr>
<td>Non-Hispanic Blacks</td>
<td>47.8</td>
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<tr>
<td>Hispanics</td>
<td>42.5</td>
</tr>
<tr>
<td>Non-Hispanic Whites</td>
<td>32.6</td>
</tr>
<tr>
<td>Non-Hispanic Asians</td>
<td>10.8</td>
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Prematurity predisposes toward childhood adiposity. Preterm children demonstrate increased waist circumferences despite decreased BMI relative to non-preterm children. These findings indicate a shift in adipose mass distribution toward visceral fat, which is significant considering the association between excess visceral fat and insulin resistance.

Prematurity predisposes toward childhood and adult hypertension. The pathophysiology of the association likely lies in multiple mechanisms on the tissue level, including decreased nephron number, decreased vascular density, endothelial dysfunction, and increased arterial stiffness. The latter appears to affect smaller preresistance and resistance vessels relative to the large elastic arteries. An important observation in the relationship between prematurity and hypertension is that arterial stiffness proceeds the appearance of clinically relevant hypertension.

Prematurity possibly predisposes toward insulin resistance, although the findings are conflicting, likely confounded by the many factors ranging from genetics to variation in care practices, among others. The relationship between prematurity and insulin resistance appears to be regulated at least in part by postnatal growth.

Prematurity possibly predisposes to multiple other morbidities when rapid infancy weight gain exceeds relative gain in length. Evidence exists to support this paradigm for obesity, dyslipidemia, blood pressure, carotid intima-media thickness, and insulin resistance. If it is presumed that premature infants endure malnutrition early in their hospital course (although not for lack of trying, just that the isolette will never be better than mom), the observation that rapid infancy weight gain increases the risk for disease teleologically suggests the following.

Programming reformatted by the premature environment fails to accommodate for the surplus nutrition that is traditionally inflicted on NICU graduates. However, the conundrum exists that supplemented nutrition appears to improve neurodevelopmental outcomes, particularly when associated with good linear growth. Therefore, no clear answer exists in how much the premature infant should be fed in terms of quantity or quality that balances with certainty the risks for adult diseases versus the goal of optimizing neurodevelopmental outcomes. Eventually, this becomes a rich soil for the seed of personalized medicine.

The challenge in interpreting the field is that more reviews exist than hypothesis-driven double-blinded prospective data (the author recognizes the paradox of that statement). The challenge in treating infants is that little evidence-driven data exist to guide the clinician. As alluded to above, much of the data reflect confounding factors resulting in contradictory or ambiguous implications. Moreover, to design studies that aim to clarify the data, 2 truisms need to be acknowledged.

The first truism is that multiple mechanisms play a role in early life programming and subsequent adult disease. Evidence of varying degrees of confidence implicates mechanisms such as microbiome dyshomeostasis, mitochondrial dysfunction, apoptosis, genetic vulnerability, and epigenetics. Likely, all of these mechanisms play some role in early life programming toward adult disease. Subsequently, study design struggles to easily define a specific hypothesis based on mechanism, either directly or indirectly.

The second truism is that we need to pragmatically focus our hypotheses so that our studies generate reliable answers. This truism requires 2 steps: a commitment to a specific mechanism and a knowledge basis that informs us of the right questions. Epigenetics deserves this commitment.
EPIGENETICS PRIMER

Epigenetics regulates eukaryotic gene expression. Changes in epigenetic characteristics potentially change gene expression without affecting DNA sequence.

Epigenetics as a field divides conceptually into developmental epigenetics and environmental epigenetics. Developmental epigenetics studies the maintenance and execution of rigid patterns of gene expression involved in development, maturation, and tissue specificity. Environmental epigenetics studies how cells respond to the environment by changing chromatin structure or expressing small RNAs. This response changes gene expression by affecting the threshold toward how easy it is for a gene to be expressed (Fig. 2). A generally applicable difference between environmental epigenetics and developmental epigenetics is that changes in developmental epigenetics turn genes off and on. No one needs a neuron to turn into a hepatocyte. Changes attributed to environmental epigenetics modify gene expression in a more subtle and incremental manner. Responses to the environment that turn off an important gene are unlikely to benefit the cell or organism. The response is too drastic. In contrast, finely tuning expression in response to the environment does confer a survival advantage. In a broader sense, environmental epigenetics describes how the environment determines phenotype by modulating gene expression.

EPIGENETIC TOOLS

Cells use several epigenetic tools to regulate gene expression. These epigenetic tools can be divided into modifications of chromatin structure versus production of different species of small RNAs (Table 3). The scientific community’s understanding of epigenetics exists at an infancy stage, despite significant progress over the last 2 years. The scientific community’s understanding of how the different epigenetic tools influence and interact with each other is even more immature. Despite this immaturity, the efforts of many contribute to a foundation of understanding on which we continue to build; this is particularly true for the tools involving modifications of chromatin structure, DNA cytosine phosphate guanine (CpG) methylation, and histone covalent

Fig. 2. Conceptualization of “effort” necessary to transcribe a gene and the role environmental epigenetics plays. Transcription requires synchronized effort of multiple processes, including (1) intercellular signaling, (2) synthesis and accumulation of transcription factor complexes, and (3) synthesis and accumulation of chromatin-modifying enzymes that impart epigenetic changes. Conceptually, this can be seen in the difference between number 1 and number 2. The delta of number 2 is less than number 1 because of environmental epigenetics. Environmental epigenetics can also increase the delta.
These tools generally affect how accessible DNA is to transcription factor complexes or how efficiently transcription proceeds, including both initiation and elongation.

- DNA CpG methylation involves placing a methyl group on the cytosine of CpG dinucleotides. CpG dinucleotides occur disproportionately in CpG islands. Two-thirds of human promoters reside with CpG islands. Most promoters residing in these islands stay relatively unmethylated if the gene is expressed at any level within the tissue of question. DNA CpG methylation does characterize silenced genes, but initiation of silencing does not require methylation and most likely represents maintenance of transcriptional repression.

- Histone covalent modification involves modifying one N-terminal tail of a histone protein. Histone proteins form the nucleosome core around which DNA wraps. Nucleosomes and DNA form the backbone of eukaryotic chromatin. The pattern and combination of histone covalent modifications assimilate into the histone code. The histone code appears to participate directly in 3 basic functions.
  - Some combinations of histone codes modulate the charge of histone lysine residues and thereby regulate charge-dependent associations between nucleosomes and DNA. This association affects the affinity of chromatin-modifying and transcription-related protein to the chromatin’s DNA. A histone covalent modification that often associates with this function is histone acetylation of the N-terminal tail of histone 3.
  - Some combinations of histone code modulate the physical accessibility of chromatin-modifying and transcription-related proteins to the chromatin’s DNA. A histone covalent modification that often associates with this function is histone methylation of the N-terminal of histone 3.
  - Some combinations of histone code modulate the charge histone lysine residues and physical accessibility of chromatin-modifying and transcription-related proteins to the chromatin’s DNA. A histone covalent modification that often associates with this function is histone phosphorylation.

Interpreting and subsequently understanding the histone code remains a mystery overall, particularly when considering the challenges of integrating the possibility of a histone modification affecting transcription from a distance of hundreds of

<table>
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<tr>
<th>Chromatin-modifying Epigenetic</th>
<th>Non-coding RNAs</th>
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<tr>
<td>DNA CpG methylation</td>
<td>Micro-RNAs (17–25 nucleotides)</td>
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<tr>
<td>DNA CpG hydroxymethylation</td>
<td>Piwi-interacting RNA (26–31 nucleotides)</td>
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<tr>
<td>Histone covalent 3’ N-terminal tail modification: methylation</td>
<td>Promoter RNAs (90–200 nucleotides)</td>
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<tr>
<td>Histone covalent 3’ N-terminal tail modification: acetylation</td>
<td>Promoter RNAs (90–200 nucleotides)</td>
</tr>
<tr>
<td>Histone covalent 3’ N-terminal tail modification: phosphorylation</td>
<td>LncRNAs (&gt;200 nucleotides)</td>
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<tr>
<td>Histone Covalent 3’ N-terminal tail modification: ubiquitination</td>
<td>Telomeric repeat containing RNAs (100–9000 nucleotides)</td>
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<tr>
<td>Histone covalent 3’ N-terminal tail modification: citrullination</td>
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* Histone methylation can be mono-, di-, or tri-.
thousands of base-pairs. Further contributing to the mystery is that the histone code within a single cell contains more than $4 \times 10^{30}$ possible permutations. Based on varying combinations of histone covalent modifications, 51 distinct chromatin states exist. Further adding to the complexity, these states affect transcription and expression based on the individual complexes interacting with the chromatin and its DNA. Although daunting, this level of complexity and capacity offers opportunity as an information storage system that continually records in detail the impact of the environment on our chromatin.

Subsequently, reviewing how an early life event affects the epigenetics of a relevant gene generates value by providing a paradigm to build on when interpreting future investigations. A relevant gene to review is insulinlike growth factor 1 (IGF-1). IGF-1 is a polypeptide whose homology resembles pro-insulin. Hepatic production of IGF-1 determines serum levels, although local production of IGF-1 plays an important paracrine role for many tissues such as the lung. Several lines of evidence point to IGF-1 as a likely effector of early programming toward adult disease, particularly when considering the common theme morbidities of insulin resistance and coronary artery disease from above.

- In terms of insulin resistance, hepatic IGF-1 plays a significant role in modulating postnatal glucose homeostasis. In mice, elimination of hepatic IGF-1 production increases serum levels of insulin without affecting glucose.\textsuperscript{55} In humans, severe IGF-1 deficiency leads to clinically relevant insulin resistance, and the latter can be reversed with recombinant IGF-1.\textsuperscript{56} Finally, within the context of a more normal situation, euglycemic clamp studies in adolescents demonstrate that serum IGF-1 levels significantly correlated with insulin sensitivity.\textsuperscript{57}

- In terms of coronary artery disease, a recent nested case control study found that low IGF-1 levels predict increased risk for developing ischemic heart disease. Moreover, polymorphisms of the IGF-1 gene impact on coronary artery disease susceptibility.\textsuperscript{58} Evidence exists that suggests one mechanism through which IGF-1 reduces the risk of coronary artery disease occurs through opposing the effects of C-reactive protein on endothelial cell activation.\textsuperscript{59}

Epigenetic mechanisms play a role in regulating IGF-1 gene expression. A red flag for the role of epigenetic mechanisms in the regulating of IGF-1 gene expression is that the IGF-1 gene generates multiple transcribed products based on differential exon usage. Variation like this represents a key characteristic that differentiates mammalian gene expression from simpler organisms.

The hepatic IGF-1 gene generates multiple transcribed products that lead to a single protein product. IGF-1 promoter 1 (IGF-1 P1) initiates transcription from multiple start sites (Fig. 3). Hepatic IGF-1P1 usage predominates early in life. IGF-1 promoter 2 (IGF-1P2) also initiates transcription from multiple start sites as well as contains growth hormone response elements. IGF-1P2 becomes active in postnatal life and responds to both diet and growth hormone stimulation. The hepatic IGF-1 transcript may or may not contain exon 5. Transcripts that do not include exon 5 are designated IGF-1A. Transcripts that do include exon 5 are designated IGF-1B transcripts. The relevance of this alternative exon usage involves the production of 2 E-peptides, EA and EB, respectively. Although still open to debate, studies exist proposing that these peptides play roles in cell proliferation, migration, and survival.\textsuperscript{60}

To further build our paradigm of how an early life event affects IGF-1 epigenetic mechanisms, the extensive use of animal models needs to be acknowledged. Indeed, most our present insight on early life events and epigenetic mechanisms arises from animal models because of increased accessibility to tissue, briefer gestations, and
shorter life spans. A common animal model of early life events predisposing to adult insulin resistance involves the induction of uteroplacental insufficiency in the rat. Uteroplacental insufficiency in rats and humans leads to a fetal in utero environment characterized by malnutrition. Moreover, uteroplacental insufficiency in humans and rats associates with adult insulin resistance and obesity.

In humans, uteroplacental insufficiency decreases fetal and early postnatal serum IGF-1 levels. Later in life, there appears to be a dysregulation of IGF-1 homeostasis possibly varying with the extent of postnatal catch-up growth.

In rats, uteroplacental insufficiency decreases serum IGF-1 levels and hepatic IGF-1 transcripts, although the effect is transcript-specific, further pointing toward epigenetic mechanisms. For example, uteroplacental insufficiency decreases IGF-1P1 and EA transcripts at day 0 of life. In contrast, uteroplacental insufficiency decreases IGF-1P2 and IGF-1B transcripts at both day of life 0 and day of life 21 (Fig. 4). The latter represents the time when rat pups wean from their mothers.

The impact of uteroplacental insufficiency on hepatic IGF-1 expression corresponds with changes in the epigenetic characteristics of the whole rat hepatic IGF-1 gene. For example, uteroplacental insufficiency enriches DNA CpG methylation at IGF-1P2 at day of life 21. Uteroplacental insufficiency also decreased lysine 36 trimethylation on histone 3 across the whole gene, with the greatest impact toward the 3′ region of the gene (Fig. 5). In vitro studies postulate that lysine 36 trimethylation on histone 3 facilitates RNA polymerase elongation of transcripts. Unfortunately, technology does not exist that presently allows for the generation of a transgenic animal that specifically decreases hepatic IGF-1 3′ lysine 36 trimethylation. However, another rat model that uses maternal hyperglycemia as an early life event to induce adult cardiovascular disease similarly finds decreased lysine 36 trimethylation of the hepatic IGF-1 gene.

The paradigm that these and other studies (both in vivo and in vitro) generate includes the following principles, which are helpful when interpreting epigenetic studies:

- Environmental challenges often initiate different responses between the genders.
Environmental challenges rarely affect only strong promoters or other key regulators of gene expression. The impact of environmental challenges occurs along the length of the gene. Looking at only a single site within an epigenetic study limits the informative nature of the study.

Environmental challenges and their impact on chromatin cannot be interpreted via a single site of change. Changes in DNA methylation or the histone covalent modifications affect transcription based on the (1) location within a gene’s organization; (2) context of other surrounding epigenetic modifications; and (3) the nuclear milieu of chromatin-modifying enzymes and transcription factor complexes.

Environmental challenges impact epigenetic characteristics early in life and later in life relative to a control or nonexposed group. However, the differences between the groups may be different early in life versus the differences later in life, and likely due to the reality that epigenetic characteristics change as mammals mature. An early change therefore leads to a different progression of epigenetic maturation.

Environmental challenges impact epigenetic characteristics early in life, but the impact in terms of gene expression or phenotype does not become evident until much later. A relevant example of this involves epigenetic changes involving sex

Fig. 4. Hypothetical impact of uteroplacental insufficiency on the “effort” necessary to transcribe the hepatic IGF-1 gene. Similar to Fig. 2, delta 1 represents the normal effort to transcribe hepatic IGF-1, the gene presented as a paradigm within this review. Delta 2 represents the similar effort, but after uteroplacental insufficiency-induced chromatin changes increase the effort necessary for successful transcription of a functional transcript.

Fig. 5. Effect of uteroplacental insufficiency on IGF-1 gene histone covalent modifications. Location of the star designates where intrauterine growth retardation decreased hepatic IGF-1 lysine 36 trimethylation on histone 3 enrichment. Location of the diamond designates where maternal hyperglycemia decreased offspring postnatal hepatic IGF-1 lysine 36 trimethylation on histone 3 enrichment.
steroid response elements, likely due to transcription machinery that interacts with epigenetic mechanisms changes as the animal matures. An easy example of this resides within sex steroid biology. A corollary of this tenant is that the impact of a specific set of epigenetic changes on gene expression and phenotype may be different in later life relative to the impact early in life.

Other epigenetic mechanisms exist through the expression of different species of noncoding RNAs. Our present understanding of the relevance of these noncoding RNAs in vivo is descriptive, although in vitro studies suggest that these noncoding RNAs provide another layer of epigenetic regulation. Considering where the epigenetic community is in terms of translating the significance of these molecules toward human disease, only 2 of these noncoding RNAs are briefly described in later discussion. These noncoding RNAs arise from the 98% of our genome that does not encode proteins. The biological significance of these noncoding RNAs becomes apparent when acknowledging that the ratio of non-protein-encoding DNA to protein-encoding DNA correlates with biological complexity. Most of these noncoding RNA appear to mediate posttranscriptional regulation of gene expression. For example, micro-RNAs contain approximately 22 nucleotides, and they affect expression of up to 60% of our genome. They silence expression by binding to complementary base-pairs of mRNA, thereby preventing translation.

Another small RNA epigenetic mechanism includes long noncoding RNAs (lncRNAs), which is a diverse class of transcripts that lack an open reading frame. LncRNAs are typically greater than 200 nucleotides, and cells express tens of thousands of these transcripts. LncRNAs regulate several of the serial steps involved in transcriptional regulation, including modifying transcription factor activity and regulating the association with chromatin. LncRNAs may also function as an interface between transcriptome and proteosome.

ENVIRONMENTAL EPIGENETICS AS FIT FOR AND PERSONALIZED MEDICINE AND FITNESS

The future of epigenetics lies in its potential as a tool to apply toward personalized medicine. Personalized medicine proposes the customization of health care, with decisions, practices, and products being tailored to individual patients. The future of epigenetics lies in this field because environmental epigenetics represents the recording of the interactions between our genome and our environment. Environmental epigenetics contains the capacity and specificity to do the following:

- Allows for tissue-specific and gender-specific responses to early life environmental events.
- Allows for the variability and complexity of the human experience (the capacity of the histone code is particularly exciting for this reason).
- Incorporates and acknowledges the pivotal role non-protein-encoding DNA plays in determining gene expression and subsequent phenotype.

Environmental epigenetics also allows for the concept of Lamarckian inheritance. Lamarckian inheritance exists as the idea that mammals pass on adaptations acquired during a lifetime of environmental exposures to their offspring with the teleologic goal of increasing the survival odds. Describing research as Lamarckian used to be considered diminutive, but evidence of cross-generational impact of early life events such as with the Chinese Famine suggests important applicability to the concept. Intuitively, several keen observers of the human condition, including the author’s children, note that individuals cannot change their genetics, but they can change their epigenetics.
EPIGENETICS AS A BIOMARKER AND AN INTERVENTION TARGET

The concept that epigenetic characteristics can be purposefully and strategically modified belies a key characteristic of environmental epigenetics as a future basis for personalized medicine. Our present studies focus on the use of epigenetics as a biomarker, as either a pattern via high through-put studies or detailed characteristics of an individual gene. For example, life variables, such as maternal macronutrient and micronutrient consumption, multiple gestations, mode of conception, mode of delivery, and maternal smoking, associate with various differences in DNA CpG methylation. With rare exception, the present understanding of epigenetics and technology, in terms of both the biology and mathematical computation, suffer from naïve but well-intended reductionism in terms of many of these studies, providing actionable data. However, this will change.

The field will progress through the fruitful efforts of many. Future studies will allow us to take the next steps to specifically intervene and thereby soften the adult impact of an early life event. Because all interventions carry some aspect of risk, identifying the right intervention for the right person stands as the end prize to make personalized medicine real on a population-wide basis. Caution must be used in terms of intervening until our knowledge base increases. Interventions that aim at changing epigenetic characteristics of a specific gene or subset of genes in a specific tissue for a specific disease may result in either immediate or long-term unintended consequences. Presently, few studies account for this contingency.

Despite this caution, environmental epigenetics represents hope for biomarkers and intervention in diseases acquired by programming. This hope resides in the primary difference between genetics and epigenetics, which has been paraphrased by several pundits in the following observation, “you can’t change your parents, but you can change your epigenetics.”

How early life events program adult disease is undergoing a transition from the broad field of maternal malnutrition to the currently relevant issues of food deserts and prematurity. Although many adult diseases and morbidities associate with various early life events and programming, the morbidities of insulin resistance, cardiovascular disease, and obesity appear to be common end points of many early life events despite potential confounders. Environmental epigenetics as a mechanism becomes particularly relevant because it contains the capacity to account for the complexity intrinsic to mammalian biology and environmental factors, while allowing for adaptation and change.

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REFERENCES


