



Randy L. Jirtle, PhD

Epigenetics a Window on Gene Dysregulation, Disease

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SCIENTISTS STUDYING ONE FACET OF how gene activity is orchestrated may be starting to unravel the mystery of how genes interact with the environment to cause disease. They have found that many of these changes are epigenetic in nature. Epigenetic changes may be thought of as chemical switches that can turn on and off the expression of genes in response to environmental factors.

Randy L. Jirtle, PhD, of Duke University Medical Center in Durham, NC, explained that epigenetic changes may allow species to adapt rapidly in response to environmental signals early in life. But some epigenetic changes can increase risk of disease if they lead to dysregulation of genes or if there is a mismatch between the environment during development and the environment encountered in adulthood.

While the field of epigenetics is still in its infancy, scientists have already compiled compelling evidence for the role of epigenetics in cancer, and animal studies are providing provocative evidence that nutritional factors and other exposures at the earliest stages of development cause epigenetic changes that can increase the risk of disease later on (Jirtle RL and Skinner MK. *Nat Rev Genet.* 2007;8[4]:253-262).

JAMA: What is epigenetics?

Dr Jirtle: Epigenetics changes are heritable changes that alter the expression of the gene without changing the DNA sequence. Epigenetic programming involves chemical changes, such as the addition or removal of methyl groups from DNA or histones that alter whether certain genes are expressed. By epigenetically varying the repertoire of the genes expressed in cells, a single genome can result in the formation of liver cells, skin cells, neurons, and other cell types that have completely different functions.

JAMA: When are these changes likely to occur?

Dr Jirtle: Epigenetic changes occur most commonly during gestation, neonatal development, puberty, and old age. The epigenome is most vulnerable, however, to environmentally induced alterations during embryogenesis, when DNA synthesis is rapid and the DNA methylation patterning and chromatin structure required for normal development is established. Once established, these epigenetic alterations are faithfully passed on to the daughter cells during somatic cell division. If these epigenetic changes are not completely erased during gametogenesis, they can potentially affect health not only in the present, but also for future generations.

JAMA: What have animal studies been able to tell us about the role of epigenetics in disease?

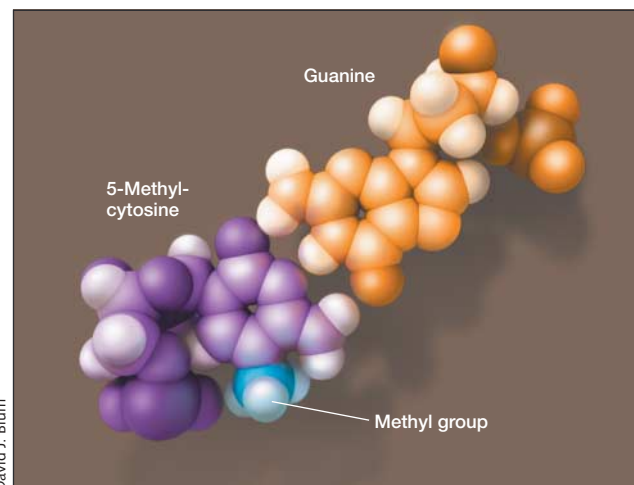
Dr Jirtle: Our studies using the yellow agouti strain of mice have shown that the mother's nutrition during pregnancy can permanently change the epigenetic programming of her offspring (Waterland RA and Jirtle RL. *Mol Cell Biol.* 2003;23[15]:5293-5300).

JAMA: Are there limits to what these types of animal studies can tell us about epigenetics?

Dr Jirtle: Epigenetic programming varies greatly between species. For example, we recently developed computer learning algorithms that predicted the presence of 600 imprinted genes in mice (Luedi PP et al. *Genome Res.* 2007;15[6]:875-884) and 156 imprinted genes in humans (Luedi PP et al. *Genome Res.* 2007;17[12]:1723-1730). Humans are predicted to have fewer imprinted genes than mice, and there is also only a 30% overlap between their imprinted gene repertoires. The divergence of imprinted genes between mice and humans indicates the need to identify the regulatory elements of imprinting in humans if we are to understand the role these genes play in the etiology of human diseases and neurological disorders.

JAMA: How has epigenetics been implicated in human disease?

Dr Jirtle: The role that epigenetics plays in cancer has been most clearly demonstrated. There are a number of tumor suppressor genes known to be inactivated epigenetically in human tumors (Baylin SB. *Nat Clin Pract Oncol.* 2005;2[suppl 1]:S4-S11). Imprinted oncogenes (eg, *IGF2*) are also frequently overexpressed in human cancers because of loss of



Chemical changes to DNA, such as the addition of methyl groups (pictured), may alter gene expression without altering the gene's DNA sequence.



imprinting—a condition that results in gene expression from two alleles of a gene rather than one (Feinberg AP and Tycko B. *Nat Rev Cancer*. 2004; 4[2]:143-153). Importantly, there are now cancer therapies (eg, 5-azacytidine and valproic acid) that aim to reverse inappropriate DNA methylation and histone modifications of such genes to allow them to function properly (Yoo CB and Jones PA. *Nat Rev Drug Discov*. 2006;5:37-50). These therapies can cause tumor regression, not by replacing a mutated gene with a good one, but rather by removing epigenetic marks so growth regulatory genes function properly.

JAMA: *Are there particular conditions that are likely to have an epigenetic component?*

Dr Jirtle: Disorders involving dysfunction of imprinted genes are likely to have an epigenetic component. Individuals have two copies, or alleles, of most genes. One is inherited from the mother and one from the father. When both copies of a gene work, it is like having a computer with a backup. But with imprinted genes, one copy is turned off epigenetically, and there is no back-up. These imprinted genes are susceptibility loci for disease since their normal function can be altered by a single genetic or epigenetic event.

JAMA: *What are some examples of human disorders related to deregulation of normally imprinted genes?*

Dr Jirtle: Disorders such as Beckwith-Wiedemann syndrome, Prader-Willi syndrome, and Angelman syndrome result not only from genomic mutations but also from epigenetic changes in imprinted genes (Murphy SK and Jirtle RL. *BioEssays*. 2003;25[6]:577-588).

Abnormal epigenetic programming of imprinted genes is associated, too, with an increased incidence of developmental disorders and failure to thrive conditions seen in humans born using assisted reproductive technologies (Arnaud P and Feil R. *Birth Defects Res C Embryo Today*. 2005; 75[2]:81-97).

JAMA: *Are there other clinical applications of epigenetics?*

Dr Jirtle: It may also be possible to prevent epigenetic changes that predispose individuals to diseases. For example, we showed that maternal supplementation of yellow agouti mice with compounds like folic acid or genistein during pregnancy blocked the negative effects (DNA hypomethylation) of bisphenol A on the epigenome of the offspring (Dolinoy DC et al. *Proc Natl Acad Sci U S A*. 2007;104[32]:13056-13061). We don't know if such preventive interventions would work if performed during adulthood rather than in early development. It is also unknown if such preventative strategies would be effective in humans. Currently, the molecular diagnosis of human disease susceptibility primarily relies on screening for gene mutations; however, this limited approach overlooks diseases that arise from changes in the epigenome. Thus, assessing for changes in DNA methylation and histone modifications may aid disease diagnosis.

JAMA: *What do you think the implications of epigenetics for toxicology testing might be?*

Dr Jirtle: Epigenetics needs to be integrated into toxicology testing programs. Also, the epigenome is most sensitive to perturbations in programming during the embryonic and the perinatal stages of development, while toxicology tests are primarily done in young adult animals—a time when the epigenome is resistant to deregulation.

JAMA: *What types of research are needed as we go forward?*

Dr Jirtle: The main thing we have to do now is define epigenetically labile genes. These are the genes that are most susceptible to being epigenetically deregulated and ultimately may give rise to increased susceptibility to diseases. Imprinted genes are particularly vulnerable to this type of deregulation.

JAMA: *Where would you start looking for epigenetically labile genes?*

Dr Jirtle: With the use of computer learning algorithms, my colleagues and I recently identified a subset of genes with high probability of being imprinted in humans (Luedi PP et al. *Genome Res*. 2007;17[12]:1723-1730). It is important to experimentally determine which of these candidate imprinted genes are indeed imprinted.

Once we've mapped these elements, we will have the ability to look at any condition, determine which genes have been deregulated, and, if so, at which locations. Eventually, you could make a chip and screen them all.

JAMA: *What about more traditional human studies?*

Dr Jirtle: Our mouse studies demonstrate that epigenetic changes that occur very early in development can result in enhanced disease susceptibility in adulthood. Human epidemiology data indicate that there may be similar epigenetic underpinnings for human chronic diseases. The National Children's Study will follow a large nationally representative sample of children from prior to conception until they are young adults. The samples collected during in this study should enable scientists to determine if early developmental changes in the epigenome are directly linked to the development of human disease later in life.

JAMA: *What are some other questions that need to be answered?*

Dr Jirtle: Imprinting is proposed to be evolutionarily advantageous because of its ability to potentially accelerate brain development (Badcock C and Crespi B. *J Evol Biol*. 2006; 19:1007-1032) and speciation (Vrana PB et al. *Nat Genet*. 1998;20:362-365). The presence of imprinted genes, however, can be disastrous to individual health because they lack a working backup copy, making them vulnerable to genetic and epigenetic change in function. Thus, a fundamental question that remains unanswered is, did imprinting help drive mammalian evolution? □