With 'Genes' Like That, Who Needs an Environment? Postgenomics's Argument for the 'Ontogeny of Information'

Karola Stotz†‡

The linear sequence specification of a gene product is not provided by the target DNA sequence alone but by the mechanisms of gene expressions. The main actors of these mechanisms, proteins and functional RNAs, relay environmental information to the genome with important consequences to sequence selection and processing. This 'post-genomic' reality has implications for our understandings of development not as pre-determined by genes but as an epigenetic process. Critics of genetic determinism have long argued that the activity of 'genes' and hence their contribution to the phenotype depends on intra- and extraorganismal 'environmental' elements. As will be shown here, even the mere physical existence of a 'gene' is dependent on its phenotypic context.

1. Introduction: The Environment within the Gene. 'Genes' are not pre-determined entities lined up in the genome like beads on a string; rather, they are "things an organism can do with its genome" on the spot to create a template resource for a product a cell may need at any particular time (Stotz, Bostanci, and Griffiths 2006, 195). The 'same' DNA sequence potentially leads to countless different gene products, different sequences might code for identical products, and the need for a rare product asks for the assembly of a novel mRNA sequence. Hence the information for a product is not sufficiently encoded in the targeted DNA sequence but has to be supplemented through sequence information provided by elements outside the coding sequence, such as transcription, splicing, or editing factors. The

†To contact the author, please write to: Cognitive Science Program, Indiana University, 819 Eigenmann, Bloomington, IN 47408; e-mail: kstotz@indiana.edu.

‡I am grateful to the Biology Studies Reading Group at Indiana University, especially Lisa Lloyd and Colin Allen, for comments on an earlier draft. This research was supported by National Science Foundation grants 0217567 and 0323496 and supplemental funding by the University of Pittsburgh. Any opinions, findings, and conclusions or recommendations expressed in this material are mine and do not necessarily reflect the views of the National Science Foundation.

Philosophy of Science, 73 (December 2006) pp. 905–917. 0031-8248/2006/7305-0038$10.00
Copyright 2006 by the Philosophy of Science Association. All rights reserved.

'environment' for any gene is composed of (1) regulatory and intronic sequences that are targeted by transcription and splicing factors (proteins and noncoding RNAs) that bind to them and (2) the specific environmental signals that cue these factors or otherwise influence the gene's expression.

I understand 'genetic information' in its original meaning as it was spelled out by Crick as part of his formulation of the central dogma of molecular genetics, which still has considerable currency today: the coding sequence provides the *specification* of the linear sequence of amino acids in a polypeptide chain (Thieffry and Sarkar 1998). Against this background I restate: those important players that interactively regulate genomic expression are far from mere background condition or supportive environment; rather, they are on a par with genetic information since they *cospecify* the gene product together with the target DNA sequence.

1.1. From Molecular Preformationism to Molecular Epigenesis. The argument presented here is part of the historic debate between preformationist-reductionist and epigenetic-holist philosophies in the quest for understanding development, a debate that has resurfaced in the postgenomic era (Müller and Olsson 2003, 117).[1] Although twentieth-century molecular reductionism had many spectacular successes, it also made clear that a mere inventory of genes, proteins, and metabolites is not sufficient to understand the cell's complexity. There is remarkable integration of the various layers, both regulatory and structural, and most biological characteristics arise from interactions between numerous cellular constituents. Viewing the cell as a causal *network* of genes, RNAs, proteins, and metabolites with distributed agency offers a viable strategy for addressing the complexity of living systems. Therefore, a key challenge for postgenomic biology is to understand how interactions between these molecules determine the operation of a cell's enormously complex machinery, both in isolation and when surrounded by other cells.

The details of eukaryotic genetics show that eukaryotic DNA alone does not *specify* the primary sequence of amino acids of a protein, let alone the protein's tertiary structure or a complex phenotypic trait. In addition to the physical complexity and developmental contingency of gene expression involved in specifying a gene product, we learn that what constitutes a 'gene' in the first place—where it begins and ends, and which sequences it comprises—is determined by the genomic, cellular, and extracellular phenotype at each point in an organism's developmental trajectory. The whole determines what counts as a part. The main argument

1. Unless stated otherwise, 'epigenesis' and 'epigenetic' refer to the context dependence of developmental processes, here at the molecular level.

of this thesis derives from genomics itself by elucidating the gene regulatory mechanisms that cooperatively specify any product during the developmental process (Stotz, forthcoming). "Although the reduction of all biology to genes has occurred on an enormous scale, it is worth noting that new studies in molecular biology can be interpreted as demonstrating the epistemologic case for organicism. Indeed, we would argue that if there is a place to make the argument for organicism, it is at the level of the gene. . . . [We find] situations where *the information encoding a protein . . . is created rather than inherited*" (Gilbert and Sarkar 2000, 6; my emphasis). It is this "ontogeny of information" (Oyama [1985] 2000) that is being asserted here. The developmental process interactively constructs the informative-instructional content of genes. "Epigenesis is constitutive"; it "does not reduce to gene regulation, for genes themselves do not pre-exist developmental processes" (Robert 2004, 74). Any *program* notion has to be applied a posteriori to a self-organized network of genome expression with causally *distributed* agency.

As will be argued, the cellular context specifies a range of products from a gene through (1) the selective use of nucleotide information or (2) the creation of nucleotide information. The cellular context provides this specificity by means of (3) complex networks of genome regulation and (4) instructional environmental resources.

Sections 2 and 3 detail gene expression events that can be said to select, or even create novel, sequence information, while Section 4 describes some of the mechanisms responsible for these events. The molecules involved in these mechanisms react to environmental stimuli that are at the center of Section 5. Section 6 concludes with some reflections on the state of the central dogma of molecular genetics and the future of the field.

2. Ontogeny of Information I: Selection of Nucleotide Information. Genes are made of functional modules, each of which can be present in alternative copies that can be reassorted to form new genes in reaction to new types of regulation: upstream, intergenic, and downstream *cis*-regulatory modules; enhancers; promoters; transcription start sites (TSSs); 5′ untranslated regions (UTRs); noncoding introns; coding exons (including alternative splice sites, alternative reading frames [ARFs], and cryptic exons); 3′ UTRs; transcription termination sites (TTSs); and *trans*-regulatory modules. The context dependency of any possible gene starts with the selection of the sequences that will make up the gene in a particular case, with the rest of the genome functioning as part of the 'environmental' context of protein coding sequences. These are rendered transient through the necessity of transcription initiation and termination, the existence of alternative promoters, transcription start and end sites, and alternative splice sites (Communi et al. 2001). While alternative splicing of exons as

the simplest form of sequence selection results mostly in related protein isoforms, similar but more complicated expression patterns might be called 'overlapping genes' that produce unrelated functional products. Examples are cases where the intron of one splice variant forms the entire coding sequence for another splice form (Mottus et al. 1997), or where coding sequences are shared but read in different reading frames (Sharpless and DePinho 1999). In the yeast *Saccharomyces cerevisiae*, the open reading frame of TAR1 (Transcript Antisense to ribosomal RNA) is contained fully within the 25S rRNA sequence but is transcribed from the antisense strand (Coelho et al. 2002). While cases of alternative splicing and overlapping genes show the modularity of genetic components, examples of the cotranscription of two adjacent genes give evidence for the transient nature of the boundaries of 'classical' genes (Magrangeas et al. 1998). Another example for the role of 'frameshifting' in sequence selection is when noncoding exons of a pseudogene are reconverted into a coding sequence when cotranscribed with a preceding coding sequence and, consequently, read in an alternate reading frame (Finta and Zaphiropoulos 2000). Even in their noncoding state, pseudogenes, of which 20,000 are known in the human DNA and traditionally are perceived as nonfunctional, are shown to control the gene expression of its coding sister sequence (Gibbs 2003; Mattick 2004).

3. Ontogeny of Information II: Creation of Nucleotide Information. Another way that regulatory mechanisms of gene expression can increase the number of gene products is by reshuffling and modifying the original DNA sequence during the transcriptional or translational processes and thereby constituting new templates for protein *not mirrored* in any linear DNA sequence. Such cases might warrant speaking of the creation of nucleotide information either out of original DNA sequences or de novo.

3.1. Trans-splicing. Sometimes *several* separately transcribed DNA sequences, either from the same sequence (homotypic) or separate sequences (heterotypic), are spliced together in *trans* to create *one* mature mRNA. In the case of homotypic *trans*-splicing, separately transcribed exons from one gene can be spliced together in a different order or appear in multiple copies within a transcript (exon scrambling or repetition; Flouriot et al. 2002; Takahara et al. 2002), or exons from separate genes—adjacent to each other, further apart, antisense at the same chromosome, or even from different chromosomes—can be spliced together to create a protein with an amino acid sequence that is not mirrored in the DNA (Blumenthal and Thomas 1988; Finta, Warner, and Zaphiropoulos 2002; Zhang et al. 2003). Sometimes a transcript that appears to be created by 'normal' *cis*-splicing is in fact produced through *trans*-splicing (Pirrotta 2002). The

autonomous transcripts need not be united into one final transcript but can be processed separately and only be connected at the translational or posttranslational level in a process called protein *trans*-splicing (Handa, Bonnard, and Grienenberger 1996).

3.2. RNA Editing. Another gene regulatory mechanism that can significantly diversify the proteome is RNA editing. Whereas most other forms of posttranscriptional modifications of mRNA (capping, polyadenylation, and *cis*-splicing) retain the *correspondence* of the primary structure of exon and gene product, RNA editing disturbs this correspondence by changing the primary sequence of mRNA after its transcription. The creation of 'cryptogenes' via RNA editing of the gene's pre-mRNA is therefore a very extreme mechanism of *genomic information modification*, which can be rather extensive with up to several hundred modified nucleotides. Editing events occur in such diverse organisms as viruses, slime molds, higher plants, and mammals and have, among other things, profound effects on the function of transmembrane receptors and ion channels in mammalian neural tissues, in erythropoiesis and inflammation, in cardiovascular disease, in cancer, and on the life cycle of viruses. Messenger, ribosomal, transfer, and viral RNAs all undergo editing in different systems through the site-specific *insertion* or *deletion* of one or several nucleotides, or nucleotide *substitution* (cytidine-to-uridine and adenosine-to-inosine deamination, uridine-to-cytidine transamination; Gray 2003). Most editing happens at the posttranscriptional stage at the pre mRNA transcript, but the family of mammalian APOBEC-1–related proteins also shows activity on DNA and is regulated by cells to enable diverse protein expression for the genome or to prevent protein expression from viruses (Samuel 2003). A-to-I editing of RNA transcripts with embedded Alu sequences has been shown to be a widespread phenomenon in the human transcriptome, especially in brain tissue. Such substitutions influence the receptor function and the channel's gating behavior of the mammalian glutamate receptors (GluRs), and the serotonin receptor subunit 2C (5-HT2CR) can modulate splice site selection in human brain cells and can sometimes mark nonstandard transcripts not destined for expression (Flomen et al. 2004; Kim et al. 2004).

3.3. Translational Recoding. A third process of modifying the original 'message' of a DNA sequence is through diverse mechanisms of translational recoding. During frameshifting the ribosome shifts the reading frame at a particular mRNA site to yield a protein encoded by two overlapping open reading frames. During 'programmed bypassing' (hopping) translation is suspended at a particular codon and is resumed at a nonoverlapping downstream codon. Finally 'codon redefinition' means the

localized alterations of codon meaning, for example, the redefinition of a stop codon to selenocysteine or to a standard amino acid (Baranov et al. 2003).

All of the above-mentioned expression patterns essentially increase the number of expressed products and therefore bridge the gap between the relatively small genome number in higher organisms and the complexity of their transcriptome. As an example, around 60% of human genes are alternatively spliced, with some of them having up to 100 different splice forms (Leipzig, Pevzner, and Heber 2004).

4. Ontogeny of Information III: A Gene Regulatory Network. Sections 2 and 3 deal with ways in which sequences directly involved in the coding process of proteins are manipulated. I now turn to those mechanisms that regulate, with the help of environmental cues that will be the topic of the following section, such gene expression patterns.

In multicellular organisms the proportion of non-protein-coding sequences increases as a function of complexity, as does the amount of regulation. New genes or splice variants need not only be specifically regulated and then integrated into the system, and regulators themselves need regulation. This accelerating control architecture imposes intrinsic functional complexity limits on systems. The received view of proteins as not only the primary functional and structural components of the cell but also the main regulatory agents does not sit squarely with the extrapolated regulatory overhead in bacterial genomes that seems to have imposed a ceiling of complexity in prokaryotes (Mattick 2004). In recent years the hypothesis that complex organisms have developed a digital regulatory system based on noncoding RNA signals able to bypass the intrinsic limits of protein-based regulation alone is gaining ground.

4.1. The Protein-Based Key-Lock System. There is a significant correlation between the size of intergenic DNA—upstream, downstream, and within intronic regions—of 'complex' genes and the diversity of functions in development and cell differentiations. Complex genes are also more often located in gene-poor regions with potentially more regulatory space available than through the flanking regions alone (Nelson, Hersh, and Carroll 2004). It is well known that a single site can be bound by different transcription factors, which often bind cooperatively, and also that multiple *cis*-regulatory modules involved in development often act independently of each other (Stern 2003, 146). The seeming lack of strong sequence constraints in many proposed eukaryotic transcription regulation sites, rather than indicating a lack of function, could be a natural consequence of the *flexibility* of the regulation machinery (Wray et al. 2003). A further role is played by *trans*-regulatory sites, for example, through alternative

splicing of transcription factor-encoding RNAs that affect the expression and activity of transcription factors (Davidson 2001). The number of proteins needed for transcription is staggering: the chromatin remodeling complex alone encompasses about a dozen proteins, and the RNA polymerase II holoenzyme complex is built out of about 15 proteins. Additional proteins necessary for transcription are one TATA-binding protein (TBP), approximately eight TBP-associated factors (TAFs or general transcription factors), several to many specific transcription factors (precise composition and number differ among loci and can vary in space and time and according to environmental conditions), and a diverse number of transcription cofactors (Lemon et al. 2001). Most of these factors react specifically to environmental stimuli.

4.2. Noncoding RNAs. Recently researchers have turned their attention to the up to 98% of 'junk' DNA in higher organisms that likely harbors novel genomic mechanisms for turning genes on and off during normal development and regulating mRNA processing. Such control mechanisms are non-protein-coding RNAs (ncRNAs) that function in basically two ways: (*a*) folded in two- and three-dimensional ways, they fulfill similar, analog functions as protein factors, such as catalyzing chemical reactions (ribozymes) or forming binding pockets for molecules (riboswitches); (*b*) they function as digital signals for DNA, RNA, and proteins through their complementary base pairing capacity (Mattick 2003, 2004).

A. Five of the nine known natural ribozymes catalyze self-cleavage using an internal phosphoester transfer reaction. Self-splicing introns assist in the processing of mature mRNA by enabling both *cis*- and *trans*-splicing in bacteria, viruses, chloroplasts in plants, and mitochondria in eukaryotes (Sturm and Campbell 1999). Riboswitches are long noncoding portions of various mRNAs that control gene expression by folding into receptors for specific environmental molecules. They are involved in such different regulatory mechanisms as inhibition of translation initiation and attenuation of both transcription and translation, leading to either activation or repression of gene expression. Known riboswitches regulate the metabolism of vitamins, amino acids, and purines. The combination of sequence conservation between large phylogenetic distances (all major branches of bacteria, archaea, and eukarya) and functional diversity suggests that riboswitches are possibly the oldest regulatory system (Mandal and Breaker 2004).

B. There is a large diversity of ncRNAs with digital functioning. The largest group is a diverse range of small RNAs that silence the expression of a variety of genes by either destroying the mRNA or interfering with its translation. RNA interference (RNAi) via double-stranded small interfering RNAs (siRNAs) has been implicated in several, different pro-

cesses including the temporal regulation of developmental gene expression, the prevention of transposon mobilization, and functioning as a resistance mechanism against virus infection (Novina and Sharp 2004). Thousands of microRNAs (miRNA) have been identified in both invertebrate and vertebrates that bind to specific transcription factor mRNAs to inhibit translation. They seem to regulate at least one-third of human genes involved in cell proliferation and death, developmental timing, or the patterning of the nervous system (Ambros 2004). Other forms of regulatory control, especially dosage compensation, are exerted by antisense RNAs, Xist RNAs, or roX RNAs (Gibbs 2003). Some sequences seem to be transcribed solely to block the transcription of the adjacent gene (Martens, Laprade, and Winston 2004). More well-known functional RNAs are small nuclear RNAs (snRNA) involved in assembling the spliceosome complex necessary for the splicing of nuclear genes (Mansfield et al. 2002), small nucleolar RNAs (snoRNA) that assist in RNA editing (among other functions), rRNAs of the ribosome, and transfer RNAs (tRNA) translating nucleic acid codons into amino acids.

RNA-mediated regulations seem to be involved in such diverse processes as chromosome replication, transcriptional regulation, mRNA processing, splicing and modification, mRNA stability and transport, translation, protein degradation and translocation, genome immune system, chromatin remodeling, DNA methylation, dosage compensation, and transvection, which together seem to warrant talk of a "parallel digital regulatory system" (Mattick 2004, 319). The molecular mechanisms that control DNA synthesis and the dynamics of cell cycle regulation are so complex that their behavior cannot be understood by casual, hand-waving arguments à la the master control gene or a genetic program. Postgenomic systems biology signifies the move beyond the single gene description toward the understanding of the intricate molecular networks between protein, nucleic acid, and small molecules that mediate most cellular processes. The past years have witnessed immense progress in the understanding of complex network behavior, such as the interaction between transcription factors and regulatory modules, including the discovery of large changes in network architecture resulting from alteration of transcription factor interactions in response to diverse environmental stimuli (Luscombe et al. 2004).

5. Ontogeny of Information IV: Environmental Gene Regulation.

> Gene-control systems face an enormous challenge. They must coordinate numerous tasks that a typical cell carries out on an ever-changing cycle, and they must *interpret many different chemical and physical signals*. Even the simplest, single-celled organisms need to modulate the expression of

> hundreds of genes in *response to a myriad of cellular needs and environmental cues*. Gene-control systems, therefore, must have the ability to respond precisely to *specific signals*, rapidly bring about their intended genetic effect, and have sufficient *dynamic* character to fine-tune the level of expression for hundreds of different genes. (Gagen and Mattick 2004; my emphasis)

5.1. Cellular and Extracellular Regulation. One important regulation mechanism involves the coupling of transcription to the strength of intracellular signaling factors in order to continuously vary transcription rate (e.g., through mitogen-activated protein kinase–controlled transcription; Hazzalin and Mahadevan 2002). Many regulatory mechanisms of the cell react to extracellular signaling proteins that bind to the cell surface and thereby activate signal transducer and activator of transcription proteins latent in the cytoplasm (Levy and Darnell 2002). Environmental signals originating from different levels of organization alter the regulatory network dynamics and can have stable epigenetic effects at the genetic level. At the cellular level there are signals such as physiological and nutritional states of cells, at the extracellular level we find exogenous signals from other cells or the extracellular matrix such as hormones, and at the organism-external environmental level signals can be the ambient temperature, the circadian light cycle, and exogenous endocrine disrupters taken in by the mother. *Genes actually encode their own environmental sensors* (transcription factors and ncRNAs) to relay environmental information to the genome.

5.2. The Epigenetic Inheritance System. 'Epigenetic' regulation refers to mostly chromosomal mechanisms of gene regulation without changing the DNA sequence that are "non-DNA-based forms of mitotic and meiotic inheritance" (Müller and Olsson 2003, 117). With little exception, *different* cells that form organs as distinctive as brains or kidneys contain the *same* genetic material; however, they have inherited epigenetic information to express this genetic information differently. For example, while most maternal and paternal alleles turn on or off at the same time, imprinting can disrupt this balance and silence either the maternal or paternal allele. Chromatin (the protein packaging of DNA) controls access to DNA sequences by condensing and expanding sections dependently and effectively hiding whole swaths of the DNA from view while exposing other sections for transcription. Hence the position of a gene within the genome effects its regulation (Dillon 2003). Methyl-adding enzymes can lock genes in a silent—methylated—state that will be inherited by the daughter cells. Maternal care has been shown to affect the expression of

certain genes via methylation, which allows for the transmission of individual differences in stress reactivity across generations (Meaney 2001). Organizational structures such as membrane-based cellular and nuclear compartmentalization are part of the epigenetic system, which makes it possible that the position of a gene within the three-dimensional space of the nucleus could play an important role in the efficiency with which its transcripts are spliced or polyadenylated, or with which its mRNA is transported from the nucleus (Francastel et al. 2000). Steady-state dynamics of self-regulating systems of interacting enzymes are also epigenetically inherited (Moss 2003).

Epigenetic inheritance mechanisms "transmit *interpretations* of the information in DNA" and therefore phenotypes rather than genotypes (Jablonka and Lamb 2005, 119). Instead of just inheriting a developmental resource such as DNA sequences, organisms inherit a particular *relationship* to this resource; the phenotype, one might say, overrides the genotype.

6. Conclusion: The Challenged Dogma. What all the above examples of regulatory mechanisms of genome expression are able to show is that we have to revise most if not all our expectations of genes and their capacities. For the largest part of the past century we came to see genes as a material unit with structural stability and identity, with functional specificity by means of their template capacities that encode information, and with intergenerational memory; we came to see genes as the designator of life and the site of agency and even mentality (in containing a plan or program for and asserting control over developmental processes). In the postgenomic era, however, there is no DNA sequence that exhibits any or all of these traits without the help of an extensive and complex developmental machinery. The phenotype at the narrowest molecular level, under certain readings the genotype itself, and the information it contains, is constituted by epigenetic processes. Instead of a linear flow of information from the DNA sequence to its product, information is created by and distributed throughout the whole developmental system. The fact that even the structural identity of a gene is created by genome regulatory mechanisms and its environmental conditions makes it very difficult to draw a clear boundary between 'gene' and 'environment'. New knowledge of gene expression mechanisms should ultimately help to release the "tension between nature versus nurture that has been perpetuated in the popular concept of the gene" because it turns out that the gene is not "the ultimate entity of nature on which 'nurture can never stick'" (Falk 2000, 318). It seems to stick quite well.

REFERENCES

Ambros, Victor (2004), "The Function of Animal Micro RNAs," *Nature* 431: 350–355.
Baranov, Pavel V., Olga L. Gurvich, Andrew W. Hammer, Raymond F. Gesteland, and John F. Atkins (2003), "Recode 2003," *Nucleic Acids Research* 31 (1): 87–89.
Blumenthal, Thomas, and Jeffrey Thomas (1988), "Cis and Trans mRNA Splicing in C. elegans," *Trends in Genetics* 4 (11): 305–308.
Coelho, Paulo S. R., Anthony C. Bryan, Anuj Kumar, Gerald S. Shadel, and Michael Snyder (2002), "A Novel Mitochondrial Protein, TAR1p, Is Encoded on the Antisense Strand of the Nuclear 25S rDNA," *Genes and Development* 16: 2755–2760.
Communi, Didier, Nathalie Suarez-Huerta, Danielle Dussossoy, Pierre Savi, and Jean-Marie Boeynaems (2001), "Cotranscription and Intergenic Splicing of Human P2Y(11) SSF1 Genes," *Journal of Biological Chemistry* 276 (19): 16561–16566.
Davidson, Eric R. (2001), *Genomic Regulatory Systems: Development and Evolution*. San Diego: Academic Press.
Dillon, Niall (2003), "Positions, Please," *Nature* 425: 457.
Falk, Raphael (2000), "The Gene: A Concept in Tension," in Peter Beurton, Raphael Falk, and Hans-Jörg Rheinberger (eds.), *The Concept of the Gene in Development and Evolution*. Cambridge: Cambridge University Press, 317–348.
Finta, Csaba, S. C. Warner, and Peter G. Zaphiropoulos (2002), "Intergenic mRNAs: Minor Gene Products or Tools of Diversity?" *Histology and Histopathology* 17 (2): 677–682.
Finta, Csaba, and Peter G. Zaphiropoulos (2000), "The Human Cytochrome P450 3A Locus: Gene Evolution by Capture of Downstream Exons," *Gene* 260 (1–2): 13–23.
Flomen, R., J. Knight, P. Sham, R. Kerwin, and A. Makoff (2004), "Evidence That RNA Editing Modulates Splice Site Selection in the 5-HT2C Receptor Gene," *Nucleic Acids Research* 32 (7): 2113–2122.
Flouriot, G., H. Brand, B. Seraphin, and F. Gannon (2002), "Natural Trans-spliced mRNAs Are Generated from the Human Estrogen Receptor-Alpha (hER Alpha) Gene," *Journal of Biological Chemistry* 277 (29): 26244–26251.
Francastel, Claire, Dirk Schübeler, David I. K. Martin, and Mark Groudine (2000), "Nuclear Compartmentalization and Gene Activity," *Cell Biology* 1: 137.
Gagen, Michael J., and John S. Mattick (2004), "Imperatives and Inherent Limitations of Accelerating Networks in Biology, Engineering and Society," unpublished paper.
Gibbs, W. W. (2003), "The Unseen Genome: Gems among the Junk," *Scientific American* 289 (5): 46–53.
Gilbert, Scott F., and Sahotra Sarkar (2000), "Embracing Complexity: Organicism for the 21st Century," *Developmental Dynamics* 219: 1–9.
Gray, M. W. (2003), "Diversity and Evolution of Mitochondrial RNA Editing Systems," *IUBMB Life* 55 (4–5): 227–233.
Handa, H., G. Bonnard, and J. M. Grienenberger (1996), "The Rapeseed Mitochondrial Gene Encoding a Homologue of the Bacterial Protein Cell Is Divided into Two Independently Transcribed Reading Frames," *Molecular and General Genetics* 252 (3): 292–302.
Hazzalin, Catherine A., and Louis C. Mahadevan (2002), "MAPK-Regulated Transcription: A Continuously Variable Gene Switch?" *Nature* 3: 30–41.
Jablonka, Eva, and Marion J. Lamb (2005), *Evolution in Four Dimensions: Genetic, Epigenetic, Behavioral, and Symbolic Variation in the History of Life*. Cambridge, MA: MIT Press.
Kim, D. D. Y., T. T. Y. Kim, T. Walsh, Y. Kobayashi, T. C. Matise, S. Buyske, and A. Gabriel (2004), "Widespread RNA Editing of Embedded Alu Elements in the Human Transcriptome," *Genome Research* 14 (9): 1719–1725.
Leipzig, Jeremy, Pavel Pevzner, and Steffen Heber (2004), "The Alternative Splicing Gallery (ASG): Bridging the Gap between Genome and Transcriptome," *Nucleic Acids Research* 32 (13): 3977–3983.
Lemon, B., C. Inouye, D. S. King, and R. Tjian (2001), "Selectivity of Chromatin-Remodeling Cofactors for Ligand-Activated Transcription," *Nature* 414: 924–928.

Levy, David E., and J. E. Darnell Jr. (2002), "STATS: Transcriptional Control and Biological Impact," *Nature Reviews: Molecular Cell Biology* 3: 651–662.

Luscombe, Nicholas M., M. Madan Badu, Haiyuan Yu, Michael Snyder, Sarah A. Teichmann, and Mark Gerstein (2004), "Genomic Analysis of Regulatory Network Dynamics Reveals Large Topological Changes," *Nature* 431: 308–312.

Magrangeas, Florence, Gilles Pitiot, Sigrid Dubois, Elisabeth Bragado-Nilsson, Michel Cherel, Severin Jobert, Benoit Lebeau et al. (1998), "Cotranscription and Intergenic Splicing of Human Galactose-1 Phosphate Uridylyltransferase and Interleukin-11 Receptor Alpha-Chain Genes Generate a Fusion mRNA in Normal Cells," *Journal of Biological Chemistry* 273 (26): 16005–16010.

Mandal, Maumita, and Ronald R. Breaker (2004), "Gene Regulation by Riboswitches," *Nature Reviews: Molecular Cell Biology* 5: 451–463.

Mansfield, S. G., R. H. Clark, M. Puttaraju, and L. G. Mitchell (2002), "Spliceosome-Mediated RNA Trans-splicing (SMaRT): A Technique to Alter and Regulate Gene Expression," *Blood Cells, Molecules and Diseases* 28 (3): 338–348.

Martens, Joseph A., Lisa Laprade, and Fred Winston (2004), "Intergenic Transcription Is Required to Repress the *Saccharomyces cerevisiae SER3* Gene," *Nature* 429: 571–574.

Mattick, John S. (2003), "Challenging the Dogma: The Hidden Layer of Non-protein-coding RNAs in Complex Organisms," *BioEssays* 25 (10): 930–939.

——— (2004), "RNA Regulation: A New Genetics?" *Nature Reviews Genetics* 5 (4): 316–323.

Meaney, Michael J. (2001), "Maternal Care, Gene Expression, and the Transmission of Individual Differences in Stress Reactivity across Generations," *Annual Review of Neuroscience* 24: 1161–1192.

Moss, Lenny (2003), *What Genes Can't Do*. Cambridge, MA: MIT Press.

Mottus, Randy C., Ian P. Whitehead, Michael O'Grady, Richard E. Sobel, Rod H. L. Burr, George B. Spiegelman, and Thomas A. Grigliatti (1997), "Unique Gene Organization: Alternative Splicing in Drosophila Produces Two Structurally Unrelated Proteins," *Gene* 198 (1–2): 229–236.

Müller, Gerd B., and Lennart Olsson (2003), "Epigenesis and Epigenetics," in Brian K. Hall and Wendy M. Olson (eds.), *Keywords and Concepts in Evolutionary Developmental Biology*. Cambridge, MA: Harvard University Press, 114–123.

Nelson, Craig E., Bradley M. Hersh, and Sean B. Carroll (2004), "The Regulatory Content of Intergenic DNA Shapes Genome Architecture," *Genome Biology* 5 (4): R25.

Novina, Carl D., and Phillip A. Sharp (2004), "The RNAi Revolution," *Nature* 430: 161–164.

Oyama, Susan ([1985] 2000), *The Ontogeny of Information: Developmental Systems and Evolution*. 2nd ed. Durham, NC: Duke University Press.

Pirrotta, V. (2002), "Trans-splicing in Drosophila," *BioEssays* 24 (11): 988–991.

Robert, Jason S. (2004), *Embryology, Epigenesis and Evolution: Taking Development Seriously*. Cambridge: Cambridge University Press.

Samuel, C. E. (2003), "RNA Editing Minireview Series," *Journal of Biological Chemistry* 278 (3): 1389–1390.

Sharpless, Norman E., and Ronald A. DePinho (1999), "The INK4A/ARF Locus and Its Two Gene Products," *Current Opinion in Genetics and Development* 9: 22–30.

Stern, David (2003), "Gene Regulation," in B. K. Hall and W. M. Olson (eds.), *Keywords and Concepts in Evolutionary Developmental Biology*. Cambridge, MA: Harvard University Press, 145–151.

Stotz, Karola (forthcoming), "Molecular Epigenesis: A Break in the Central Dogma," *History and Philosophy of the Life Sciences* 26 (4).

Stotz, Karola, Adam Bostanci, and Paul E. Griffiths (2006), "Tracking the Shift to 'Postgenomics'," *Community Genetics* 9 (3): 190–196.

Sturm, N. R., and D. K. Campbell (1999), "The Role of Intron Structures in Trans-splicing and Cap 4 Formation for the Leishmania Spliced Leader RNA," *Journal of Biological Chemistry* 274 (27): 19361–19367.

Takahara, T., D. Kasahara, D. Mori, S. Yanagisawa, and H. Akanuma (2002), "The Trans-

spliced Variants of Sp1 mRNA in Rats," *Biochemical and Biophysical Research Communications* 298 (1): 156–162.

Thieffry, Denis, and Sahotra Sarkar (1998), "Forty Years under the Central Dogma," *Trends in Biochemical Sciences* 23 (August): 312–316.

Wray, Gregory A., Matthew W. Hahn, Ehab Abouheif, James P. Balhoff, Margaret Pizer, Matthew V. Rockman, and Laura A. Romano (2003), "The Evolution of Transcriptional Regulation in Eukaryotes," *Molecular Biology and Evolution* 20 (9): 1377–1419.

Zhang, Cheng, Youmei M. Xie, John A. Martignetti, Tracy T. Yeo, Stephen M. Massa, and Frank M. Longo (2003), "A Candidate Chimeric Mammalian mRNA Transcript Is Derived from Distinct Chromosomes and Is Associated with Nonconsensus Splice Junction Motifs." *DNA and Cell Biology* 22 (5): 303–315.