December 2008 Issue | Michael Skinner, PhD Center for Reproductive Biology

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Welcome to *Functional Medicine Update* for December 2008. Can you believe that we are ending another extraordinary year? The evolution of functional medicine over the last 26 years has been truly remarkable. December's issue is going to focus on epigenetics/epigenomics and how it relates to the future of medicine. We had the privilege of learning about this with Dr. Randy Jirtle, and I think we are now going to the next level by focusing on environmental epigenetics.

Environmental epigenetics (as described by our researcher of the month, Dr. Michael Skinner) is another component that ties environmental exposures to nutritional factors, lifestyle, and the expression of chronic disease entities, which then can change patterns of morbidity over time due to the heritable opportunities that these epigenetic marks pass down through the generations. I think there is a lot of alarm that can come from the observations you will learn about in this issue, but there is also a lot of hope for change in the future. As we move into 2009, an epic period of change in the world's history may be right ahead of us, certainly with the change in the United States government and all the things that are going on internationally. I think this topic we are discussing this month in *Functional Medicine Update*(a very small part of it-a microcosm) illustrates the remarkable nature of change that is occurring across all sectors--politics, economics, science, medicine, and so forth.

With that as a prelude, let me just quickly remind you what we mean by epigenetics. As you know, it is the post-mitotic modulation (or the change in the DNA marks) that then allow only certain portions of our book of life to be expressed; all regions of our genome are not equally readable after epigenetic modulation. This has to do with either putting methyl groups on certain regions of the genome, or putting acetyl groups on the histone proteins that encode the compaction of our DNA genetic information and open up regions of the DNA so that they can be read (so these would be "Read Here" notes versus the "Don't Read Here" notes that methyl groups impart).

Webinar Series on Nutritional Epigenomics
As some of you may know, I have just completed a two-part webinar series on basic nutritional epigenomics. This series will be available through Synthesis for those of you who are interested in doing more in-depth study on this emerging topic. I think this is a very, very important topic for reasons that we'll be discussing this month.

Emerging evidence obviously suggests a key role for epigenetics in a variety of human diseases, including inflammatory and even neoplastic disorders. The epigenome is modified by all sorts of

environmental factors throughout life, of which nutrition is one and that's why we called it "Nutritional Epigenomics." Environmental xenobiotics or foreign chemicals can have a role to play in modulating the epigenome, and that's what we will be discussing this month with Dr. Skinner: autoimmune and neoplastic consequences of cumulative epigenomic or epigenetic dysregulation that occurs throughout life.

We want to protect our book of life against putting marks that start reading the wrong chapters. There are individuals who live in cultures that have a history of long life (the so-called blue zones of our world, where centenarians are common). These are places where, maybe, the epigenetic marks that occur throughout life are not being placed on the regions of the book of life that would cause bad stories to be read and good stories, (like tumor suppressor genes) to be silenced.

Protecting the Epigenome from Genomic Instability

I think this model that is emerging gives a different view of the concept of diet, lifestyle, and environment related to health. It suggests (as we learned from Dr. Fenech earlier in 2008) that we want to protect our genome against genomic instability (agents that would cause it to be less able to keep the integrity of our message alive and well). Altered epigenomic marks or epigenetic marks would, in fact, influence the stability of the message and its integrity of being read in the right way.

We now recognize that there are a variety of dietary factors that influence this process of epigenetics and the "Stop" messages or the "Read Here" messages (the DNA methylation and acetylation messages). Those messages interrelate with diseases like autoimmune disease, cancer, diabetes, and heart disease. One condition that has been studied a great deal is colorectal cancer. Colorectal cancer is the most common cancer in nonsmokers and it poses a significant health burden. Observational studies have tended to support the impact of environmental factors, especially diet, on colorectal carcinogenesis. This is because the colon is like a conduit that is picking up the debris from our diet, and also the secondary byproducts from bacterial fermentation and the transference or the alteration of these materials into secondary chemicals that may cause injury at the cellular level.

We have started to look very seriously within the cells of the mucosal lining. We find that the cells exposed in the colon to these various chemicals in the gut can undergo altered genomic/epigenomic marks, the so-called DNA methylation. This is DNA methylation that is thought to occur at least as commonly as inactivation of tumor suppressor genes. In fact, compared with other human cancers, promoter gene methylation occurs most commonly within the gastrointestinal tract. This is very recent work that has been discovered, and an article on this was published in the journal *Epigenetics* (it has its own journal now) in 2008.[1] So emerging data suggest, then, that direct influence of micronutrients (for example, folic acid and selenium, as well as other methyl-donating nutrients) interact with toxins and can alter DNA methylation in the colon mucosal cells and alter, then, the potential for epigenetic silencing of tumor suppressing genes. This recent research suggests such interactions are likely to have a mechanistic impact on colorectal cancer carcinogenesis through these pathways and offer potential therapeutic benefit for dietary alteration and even selective nutrient supplementation through nutraceuticals.

You might say, "Are these epigenetic effects the same in all individuals?" Of course, the answer to that is "no" because we have to superimpose the concept of nutritional modulation of these epigenetic marks on the concept of genetic diversity (single nucleotide polymorphisms, the so-called SNP model). Although we only have about 25,000 genes in our human genome that code for proteins, we have several million

different variations on a theme called single nucleotide polymorphisms that are single letter alphabet changes in these stories that relate to those genes. These single letter alphabet changes sometimes may be benign, but other times they may produce a variant protein that has differing functional characteristics.

You could say there are different degrees of susceptibility to epigenomic changes based upon differing SNPs. This is making the field even more complicated. Epigenetics provides a potential explanation for how environmental factors modify the risk for common diseases. Individual variation in DNA methylation and epigenetic regulation has been reported at specific genomic regions, including transposable elements called transposons, genomically imprinted genes, and the inactive X chromosomes in females. Understanding of the factors that contribute to these inter-individual epigenetic variations is still emerging, but they do appear to relate, then, back to specific single nucleotide polymorphisms, or SNPs, that have unique diet and lifestyle sensitivity for their modulation.

As Dr. Jirtle told us in his interview earlier in 2008, of these 25,000 genes within our human genome, there are probably just a few-maybe a few hundred at most-that are very sensitive to epigenetic changes in the adult cells (meaning genetic messages that can be epigenetically modified in the adult). Those are the genes that we probably really want to spend our time learning more about because those are the epigenetic changes that can be both wiped clean and replaced with new marks, and we can do something to actually alter the marks themselves.

There is now some preliminary clinical evidence showing that when individuals with specific types of cancer are given a drug that modifies epigenetic marks (e.g., by wiping the slate clean, so to speak) such as removing the silencing of tumor suppressor genes, there has been remission.. That certainly gets us to think very significantly about where the future might be in therapeutic medicine, but it also gets us to think about the role that everyday living plays in putting marks on and taking marks off of our genome to allow the good messages to be read and messages that we want to save for a rainy day (that hopefully never comes) to be silenced

The Important Role of Kinases in Intercellular Signal Transduction

In terms of dietary manipulation related to methylation patterns, you might jump too much on the bandwagon of folate, B6, B12, betaine as being the answer because we know there are many other variables and factors that influence these epigenetically modulated pathways. The signal that alters or stages the epigenetic marks is a signal that we call intercellular signal transduction. With this process, something that occurs outside the cell that triggers a series of events that transfer through the cytoskeleton of the cell, all the way into the genomic message locked deep within the nucleus of the cell in our book of life-this vault, this library that is more secure than Fort Knox by orders of magnitude-and allows, then, for the structure of our genome to be modified epigenetically by the placement of these marks (these methyl groups or these acetyl groups). The intercellular signal transduction process that communicates the outside world to the inside of the cell is regulated in part through a family of enzymes that I have discussed with you in the past called kinases. There are over 500 of these, and they participate in kind of a complex relay race, passing along the baton through the cytoplasm of the cell and ultimately through the nuclear envelope into specific regions (promoter regions) of genes to modulate their function.

Diet Plays a Role in Modulating Kinase Activity
You might ask, "Does diet play a role in modulating kinase activity?" The answer is yes. It has now being

discovered that there are many phytochemicals and nutrients that may play roles in augmenting or modulating kinase function within cells, which can then influence epigenomic remodeling of the genome. These are post-translational modifications that may influence what we call our nucleosome. The nucleosome is the complex unit structure that compacts and compresses the DNA message and locks it away safely within the cells so it is not easily accessible to injury from chemicals or radiation. The nucleosome is kind of a secure "vault." These post-translational modifications of the nucleosome occur through these processes that are kinase modulated and can also connect to things called NAD-dependent histone deacetylases or acetylases.

Resveratrol is an Example of a Phytochemical with Epigenetic Control
One of the phytochemicals that has a rich history and certainly has been in the news and speaks to the nucleosome in this capacity (to the NAD-dependent histone deacetylase) is resveratrol. This is related to the paradox of the French wine syndrome and this whole construct of phytochemicals playing a role in modulating the sirtuin genes, which are the so-called longevity genes. These genes are modulated through the epigenetic control that resveratrol has on this NAD-dependent histone deacetylase. When modified, this causes the compaction or the change in the reading structure of the genome. It allows different chapters to be read or not read, so this is an epigenetic modulation that is seen in the adult by the exposure to resveratrol. Resveratrol is just one of many phytochemicals that are being identified to have effects on epigenomic modulation.

I want to emphasize that the resveratrol work has principally been done, at this point, in very simple eukaryotic cell systems or in animals. We haven't yet seen the proof of concept in humans, but there is evidence, at least from the preliminary data, suggesting that this resveratrol or phytochemical connection to the NAD-dependent deacetylases that modulate histone protein epigenomics is real and can have influences over time on the functionality of gene expression.

I'm throwing a lot of words out there, aren't I? For those of you not molecular biologists or cell biologists this may appear very complicated, but I hope you are getting the drift of where the science is going. We are continuing to see how diet has an information signature that is much more complete, robust, and sophisticated than we previously thought. And this diet and environment connection to genetic expression is tied, in part, to cellular processes that translate these outside messages into epigenetic modulation of the genome that not only is going to influence that existing cell in its present state, but (if it occurs in a germ cell) is also transmissible to the next generation. That's the big "a-ha."

This concept may suggest that a changing environment can produce a transmissible factor very quickly, without having to rely on mutational injury to the genome as Darwinian natural selection might have suggested. This may occur very quickly by epigenetic modulation through these environmental factors, and we'll be hearing much more about this from Dr. Skinner. We've got both positive and negative influences of epigenetics on outcome, as it relates to health. Dietary manipulation can influence these post-mitotic, or let's call it epigenetic regulatory pathways (the histone deacetylation and acetylation pathways, and the methylation/demethylation and the phosphorylation pathways), all of which can play roles in modulating how our book of life is going to be read.

Maternal Nutrition Influences Disease Susceptibility Later in Life

Epidemiological and experimental studies have pointed toward maternal nutrition as a very major player

during prenatal development in influencing disease susceptibility later in life. The reason for this is clearly tied to these epigenomic or epigenetic changes. This was reviewed beautifully in the *Annual Review of Nutrition* in 2008.[2] This article discusses the influence of dietary sources on epigenetic programming during pregnancy. These messages can be passed on to the progeny as marks that alter the way their genes will be expressed throughout the course of their life unless they are able to wipe off those marks and replace them with new marks. Again, it has to do with sometimes aggressive lifestyle changes in behavior to reprogram and repattern some of these epigenetic systems. That is really where we are starting to see significant work going on now.

What does it take to modulate an epigenetic mark at one of these susceptible gene regulatory units? I think we can say we don't have the full answer to that question. The evidence would suggest that there are therapeutic opportunities for changing the course of expression. This is a very optimistic sense of the future. It doesn't lead to our belief of determinism; it leads to the view that aggressive intervention can remodel the ability of our book of life to be expressed in more favorable ways.

An article about this effect of in utero and early life conditions on adult health and disease was published in the *New England Journal of Medicine* in 2008.[3] Really it is about tying back to epigenetic programming or patterning what went on with the sperm and the egg prior to conception and then what happened in utero relative to the placement of epigenetic marks. Some questions that come out of this are: can this explain things like the rising prevalence of autistic spectrum disorder?; or can this explain the rising incidence of hyperactivity disorder?; or can it explain the rising incidence of things like asthma? All of these have been identified in animal models to have epigenetic relationships. Are we silencing certain genes and activating other genes to be expressed through these epigenetic environmental influences? These are very interesting questions. We are trying to learn not only where the effects occur, but how we can then replace those kind of marks with the marks in the right place on the genome to create the right (or more favorable) outcome.

Does Epigenetics Play a Role in Metabolic Syndrome?

People are talking now about epigenetic modulation in cancer metabolism. What about chronic diseases? Can you modulate a condition as broad based as metabolic syndrome, which we know has multiple genes that are influencing multiple functions? The *American Journal of Clinical Nutrition* featured a very interesting article in 2007 looking at epigenetic modulation and the effect of metabolic syndrome.[4] The article talked about how hyperinsulinemia and high insulin levels alter, post-mitotically, the epigenetic marks on the genome. In so doing, this causes regulation of expression in such a way as to induce higher inflammation, dyslipidemia, and vascular-related issues, which are things that we see clinically expressing themselves as type 2 diabetes, or cardiovascular disease, or even cancer, associated with hyperinsulinemia.

This raises a whole different view, doesn't it? It is a view about intervention that pertains to how to create the right marks in the right places (these epigenetic marks) to regulate the gene expression and to optimally produce outcome in the environment that is changing. This an environment in which we are being exposed to more and more things that are not native to the human species, but new within as little as a hundred years ago. We are having to develop new ways of modulating the effect of these exposures.

The Influence of B Vitamins on Epigenetics

What about B vitamins? Can we say that cultures that have low B vitamin intake, like concentration camp victims or prisoners of war, during starvation or famine, modulate the epigenome through lowered effect on genome methylation, acetylation, and phosphorylation? The answer appears to be yes. If you go back and look at individuals during time periods that were influenced by low nutrient intake, there is an influence on disease causality throughout the generations. This was discussed in an article in the *Proceedings of the Nutrition Society* in 2007.[5] This is the whole construct that temporal insufficiency of nutrients that modulate various critical epigenetic pathways can influence not only the immediacy of that person's health, but more importantly, the long-term outcome of their health, and maybe even passed on to progeny.

What I'm really talking about here is a structural approach for, kind of, functional modules: looking at the body as a biological network and how it interacts with its environment. This is a very different view than looking at single agents and single outcomes. This is the approach of looking at body function and health outcome as large biological networks in which cellular function interacts with the environment in such a way as to influence both immediate and long-term outcomes.

If we cast this approach against the past view of nutrition, nutritional supplementation, and nutritional pharmacology, it kind of re-contextualizes much of what we have been thinking for the last 30 years (actually it is probably more like the last 50 years). I recently authored a paper that appeared in *Alternative Therapies* in the September/October 2008 issue titled "The Future of Nutritional Pharmacology."[6] In that particular article I suggested that we go back and review the work of pioneers like Dr. Roger Williams, Dr. Linus Pauling, Dr.Abram Hoffer, and Dr. Kilmer McCully. So many discoveries, so many observations that were criticized heavily and were argued against by the "people in the know" at the time, are now being re-evaluated in light of nutritional epigenetics revolution. That is, how nutrients may not only influence the direct metabolism (i.e., metabolomics) through their effects as co-factors or antioxidants, but also their effect long-term on the signature of information that appears in the epigenome and how it regulates genetic expression and controls the proteosome, ultimately moving out into the metabolome, is now an area of interest.

I think we are really at a threshold-a very powerful threshold where we are taking this information and tying it together with our understanding of single nucleotide polymorphisms and diversity. We are going to be seeing many, many more genetic evaluations being done on people to try to personalize therapy, and coupling these two together to develop a construct that is the personalized medicine approach for that individual.

We now see that genetic tests are beginning to play a role in pharmacogenomics, in which people are being genetically typed for certain drug treatments. We are also seeing how the genotyping is being used to predict responsiveness of certain cancers to specific chemotherapy drugs. And now there are new organizations like 23andMe. Their website is 23andMe.com. They have a very robust website that is connecting a specific individual's genomic uniqueness to the world gene base to look at geneology. This is a very, very interesting trend that is occurring, and it is occurring, in this case, outside of traditional medicine. Molecular geneticists are behind this particular company, 23andMe. It is giving a whole different level of understanding of an individual's ancestry, legacy and past history. It is not just about advice to eat broccoli and exercise any more, but this whole connection between the specific strengths

and weaknesses within the genomic uniqueness of the person that tie together with their nutrition and lifestyle patterns to give rise to their outcome.

This is obviously a threshold. It is a revolution. When you hear Dr. Skinner you are going to learn more about how the environment is also a major component. And then various dietary substances can modulate this process hormetically. We had a wonderful conversation with Dr. Calabrese earlier this year in which he talked about nutritional hormesis and how small amounts of some substances working at regulatory nodes have larger effects than we expect on outcomes. Resveratrol is an example, and I have also talked about our own research that relates to the iso-alpha acids from *Humulus lupulus* (hops), which has a dramatic effect upon the regulatory networks that associate with inflammation and insulin signaling. There are also the effects you see hormetically from things like ginger (gingerols), or in licorice (some of the constituents in licorice have effects upon gene modulation through these regulatory nodes). Small amounts of intake can have much broader effects on physiology than we expect, through these hormetic mechanisms.

Hormetic Dietary Phytochemicals and their Role in Prevention of Neurodegenerative Diseases

There is a very nice paper that appeared in *Neuromolecular Medicine* just recently in 2008 that describes how hormetic dietary phytochemicals might play a role in prevention of neurodegenerative diseases through alteration of the regulatory pathways associated with apoptosis, neurofibrillary tangle formation, and beta amyloid accumulation with Alzheimer's disease, and how these phytochemicals can help reduce toxins that induce oxidative stress and free radical injury.[7] These phytochemicals include things like epigallocatechin gallate from green tea. We certainly see a tremendous amount of research coming out in that area. We also hear about various flavonoids that are associated with activation of the pathways associated with detoxification of foreign chemicals. I think this field is emerging to be more than just a cursory interest and is starting to translate down into clinical applications.

Let me close this discussion by focusing on things like the Mediterranean diet. We know the Mediterranean diet is very complex. It is rich in phytochemicals; it is nutrient dense. It is amazing the number of papers that are being published just within the past year or so on the Mediterranean diet composition on function and gene expression.

The rich array of phytochemicals in the Mediterranean diet play a significant role in imparting some of its clinical benefit, including improved insulin sensitivity, improved cardioprotection, and reduction of risk to inflammation. This may also translate into things like lowered risk to Alzheimer's through what is called type 3 diabetes, which is an insulin resistance situation that affects neurochemistry. We recognize that there are various dietary proteins within the Mediterranean diet that also play a role. This is a complex diet with many different factors, not one of which on its own has this influence; it is the full dietary array has impact on the cell signaling function.

Differing proteins have differing effects, not just as a consequence of their amino acid composition alone, but as a consequence of the fact that sequences of amino acids and proteins can impart different signals to the regulatory pathways. What did I just say? What I just said is that the sequence of amino acids in specific dietary proteins can modulate signaling pathways that are different than that of just individual amino acids in that protein by themselves. In the past we have always considered dietary protein was just delivery of amino acids because it is digested down to its requisite amino acids and absorbed as amino

acids; we didn't think there was any functional characteristics of the intact protein or the partially hydrolyzed protein.

This whole model is changing. It is being recognized that there are receptor sites on the surface of the GI system that can pick up information from small partial hydrolysate molecules in the digesta that can impart information to the immune or inflammatory system. This is new information suggesting the composition of proteins in the diet may play regulatory roles. This might explain things like how gluten from grains with alpha gliadin could have an inflammatory potential. They are made up of the same amino acids as other proteins, right? Why do they have a problem that other proteins don't? It is because of the structure/function relationships they have with specific genetic uniquenesses along receptor sites on the gut that activate transglutaminase or have different effects on the inflammatory pathway.

I want you to be thinking that these generic food families we talk about-carbohydrate, protein, and fat-are very limited in their usefulness when we are trying to design individual diets. The personalities and signatures of each of the components of protein, carbohydrate, and fat can have very dramatic, important roles to play in how they are witnessed, or seen, or read by the body. These bioactive components in food can modulate signaling in very, very important ways and can lead to altered bioenergetics and altered mitochondrial function. They can lead to different adipocyte function and engage in altered energy that leads to storage (storage we call obesity). So obesity may not be just solely a consequence of eating luxurious calorie diets. It may be a consequence of eating disinformation and inducing, in cellular physiology, altered epigenetic and proteomic/metabolomic outcomes.

As I said, the Mediterranean diet is a complex diet with many different dietary signatures. It is an orchestra with all the components: the strings, the woodwinds, the percussion, and the brass all playing together. It is the harmonious orchestration of that whole orchestra that gives rise to the effects on how genes are expressed across multiple tissues in men and women. There was a very nice paper about the rationale and evidence for the Mediterranean diet that was authored by Michel de Lorgeril and Patricia Salen; this paper talks about the extraordinary benefit that the Mediterranean diet has as a consequence of these dietary signatures.[8]

I think we are really starting to witness a changing paradigm-a paradigm that says lifestyle and diet intervention is a primary therapy. What we often call therapeutic lifestyle change, or TLC program, is a primary therapy for the management of complex metabolic disorders in which there is a distortion in the web of physiology rather than just a single point problem of necessity of blocking one enzyme, like using an ACE inhibitor, or using an H2 blocker, or using an HMG co-A-reductase inhibitor. We are talking about a much more complex modulation of the system (the network system), and the environment and the agents within the environment (beyond diet) play roles in sending signals as well. These are things like bisphenol A, which you probably know has been implicated recently in a whole range of metabolic, age-related disorders-a recent paper in the *Journal of the American Medical Association* brought that to our attention.[9,10] Bisphenol A is a plasticizer found in many different plastics. We might say, "A very small concentration isn't insignificant, is it?" But if you take the concept of hormesis (hormesis being small amounts having much bigger effects than expected), what has been found is that in animals, there are influences of this very small concentration on cellular signaling and cellular function, and when you look epidemiologically in humans, you find the same thing; it appears to correlate.

We have the good, bad, and the ugly: cellular signaling, depending upon what the environmental

exposures are and how they get translated through things like genomic stability or instability through epigenetic tagging, through the whole concept of transcription and ultimately into the formation of protein (the post-translational modification of protein), how that regulates metabolism and how metabolism eventually plays out to give rise to function. At every step along the way this system our environment is interfacing with those regulatory units to create our outcome. The way that we have basically managed this in medicine is to think, "Let's step in with a drug, which is a single molecule and block one function related to one end point so we'll create a good outcome." And then we wonder, "When we gave that single molecule, that person actually had what we call unexpected adverse symptoms. Where did they come from? I thought we were blocking only one function, one isoform of one enzyme or something of this nature." We find that our receptor sites for that specific functional characteristic on cells and tissues other than that which we wanted to treat, so we start getting action at a distance that gives rise to these other effects.

What we are really looking at now is a different philosophy--a different approach--and when people adhere to Mediterranean-type diets it leads to a reduction in the prevalence of clustered risk factors and clustered biomarkers, not just one biomarker: total cholesterol goes down, HDL goes up, LDL-C goes down, triglycerides go down, apo lipoprotein A goes up, apo lipoprotein B goes down. There are multiple things going on that indicate that we have changed the web of physiology. By the way, this is recently discussed in a very nice paper in the *European Society for Cardiology* on the role of the Mediterranean-type diet in reducing the multiple risk factors associated with cardiovascular disease.[11] It has also been found that the Mediterranean diet has a positive effect on bone mass in a sample of Mediterranean women, as well, as we might expect because we are modulating the web of physiology. This was published in 2008 in *Nutrition*.[12]

Hopefully we have set the stage for the wonderful interview we are going to have with Dr. Michael Skinner, who is really going to take us beyond the diet connection into the environment connection--to epigenetics and how that regulates function potentially throughout life and how those things ultimately translate into not only the immediacy of impact in the health of the individual, but also potential future generations. Obviously, the most sensitive period of life is during fetal development and infancy, and those times (or generations) may be the biomarkers of our society: how are we modulating the epigenetics of these individuals, either in utero or in infancy? How does that translate a legacy through the generations of altered functional status? And then that begs the question: what do we do to remove the marks that we don't want in certain places and put marks back where we need them? Stay tuned. You are in for a very exciting tour with Dr. Michael Skinner.

INTERVIEW TRANSCRIPT

Clinician/Researcher of the Month
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You all know how excited I am to this part of our monthly Functional Medicine Update, which is our clinician/researcher of the month section. It is here that we really learn what is happening at the forefront, at the cutting edge, at what I like to think is the leading-not the bleeding-edge, as it pertains to looking out the windshield rather than the rearview mirror of where medicine is going rather than where it has come from. As you know, in 2008, we have had the opportunity to talk with some people who are really creating a whole new vista, a whole new perspective. I would call it a post-Mendelian perspective of medicine-or a post-Darwinian perspective, even. We're not going to be disappointed this issue because we have another one of those seminal contributors to the evolution of this field, Dr. Michael Skinner.

Dr. Skinner and I share some degree lineage here, through our location. He is a professor at Washington State University in the School of Molecular Biosciences Center for Reproductive Biology. Also, I notice he is a graduate in chemistry from Reed College. I actually taught at Reed College for a year, so I have an Oregon affinity for him as well. That's probably where the similarity stops because his science has far surpassed that which I could have expected in my professional lifetime. He has done some extraordinary things. The first paper of his that I had the pleasure of reading, which really got me on to following his work more closely, was published in the June 2005 issue of Science magazine titled "Epigenetic Transgenerational Actions of Endocrine Disruptors and Male Fertility," which, as you'll hear through his voice, is the start of a whole interesting collaborative field.13 We heard a little bit about it from Randy Jirtle, but you are really going to get a sense from Dr. Skinner, I think, of how the environment influences not only the health of the F-zero generation (meaning us) but our progeny. Is this hormesis effect that we talked about with Dr. Calabrese really anything of clinical interest or is it just really an esoteric concept? If it is of clinical interest, how does it affect future generations?

With that as a context, Dr. Skinner I would like to welcome you to Functional Medicine Update. Thank you for your time. I guess my first question would be, how did you get into the area of environmental molecular toxicology?

MS: Thanks for the opportunity to participate. It's a pleasure. I would say I'm not an environmental toxicologist by training, for sure. We stumbled into this. Many major studies or observations in science you would call serendipitous. I will kind of go through it quickly, but yes, this was a serendipitous observation that led us down this path-something we would not have predicted.

I am actually a reproductive biologist. I have studied the development of the testis and the ovary for many years, in terms of the basic molecular and cellular control of their function. We were studying sex determination during embryonic development and looking at testis development. To investigate that further I decided to try some environmental compounds (some endocrine disruptors) to treat a pregnant mother to see if we could interfere with or alter sex determination. We put forward a series of experiments in vitro and in test tubes and everything appeared to work really well (it looked like we were getting effects), but then when we did in vivo where we simply injected a pregnant mother (we used a rat as a model), we basically didn't see any change in the sex determination. We didn't see any alteration in testis development. And, in fact, we looked out through puberty development up to the adult and we really didn't get an effect. Most people would say that was a negative experiment (essentially the experiment didn't work).

But then there was a serendipitous observation: the adults, after they matured, developed a testis defect on most of the spermatogenic cells undergoing spermatogenesis that are eventually going to turn into sperm;

they started dying in the testis. They would undergo apoptosis and die. It was a sub-fertile sort of condition. We actually published that article and put it out and we thought that was pretty much it. There was a postdoctoral fellow in my lab-her name is Andrea Cupp-and she came in the office one day upset because she accidentally had bred the F1 generation animal to make an F2. We didn't really plan that experiment, so we hadn't planned on using the animals that way. I said, "Don't worry about it. Just go ahead and look at the phenotype (what the testis looked like in that F2)." Lo and behold, it had the same phenotype. Greater than 90{56bf393340a09bbcd8c5d79756c8cbc94d8742c1127c19152f4230341a67fc36} of the males had this hermatogenic cell defect. She came back and said that was going on. I didn't quite understand because the only animal exposed to the toxin was a very transient exposure during sex determination of the mother. She went back and repeated it a number of times. Basically, we took it out to the fourth generation and we found that this became a transgenerational male defect in spermatogenesis that went out four generations and greater than 90{56bf393340a09bbcd8c5d79756c8cbc94d8742c1127c19152f4230341a67fc36} of animals in each generation were affected. So this would not follow a normal Mendelian genetic sort of trend, and having 90{56bf393340a09bbcd8c5d79756c8cbc94d8742c1127c19152f4230341a67fc36} of the males of all those different generations also would not follow normal DNA sequence mutation events either. So this turned out to be a transgenerational disease state.

As those animals age it turns out that they develop a whole series of diseases-prostate disease, kidney disease, immune abnormalities, and a whole series of tumors, including breast tumors, as well. About 85-90{56bf393340a09bbcd8c5d79756c8cbc94d8742c1127c19152f4230341a67fc36} of the animals would develop one disease or more, so this was an extremely high level of disease. The only animal exposed to the environmental factor was a pregnant mother, four generations later causing a disease state, so this is a transgenerational disease state. The only real way to do this, mechanistically, is epigenetics, which I can describe for you. It turned out to be a non-Mendelian sort of approach. It turns out what we were doing was reprogramming the germ line during that embryonic development during sex determination and permanently imprinting DNA methylation marks on the DNA, which then became permanently programmed such that that sperm would pass forward to each generation this disease phenotype. It turned out to be an epigenetic transgenerational disease phenomenon.

JB: You know, when I hear people like yourself--primary discoverers of major new paradigms--it often just flows off their lips so eloquently and so smoothly, but behind the scenes are all sorts of extraordinary seismic events in the body of knowledge, which often doesn't undergo transition easily. We hold onto our beliefs (even scientists) very, very strongly. When we talk about non-Mendelian and alterations of phenotypic expression from non-mutational changes in DNA, how did that get responded to by your colleagues?

MS: That was the hypothesis I put forward and then we actually had to do experimental studies to show that we were not causing a DNA sequence mutation. We did some experiments recently using some genomic approaches to show that indeed there are no increases in point mutations. We had to also show that there was a DNA methylation change in the germ line in the sperm, itself. Originally, four years ago, we did much more cruder approaches than what are available today. Today we take much more elegant molecular approaches. Basically we had to show that the DNA was getting methylated differently, and that was carried forward for four generations. So we had to have that experiment in hand before anyone would let us state that this could be an epigenetic phenomenon. It took us several years to actually get that done.

JB: After having completed that work-by the way, as I was mentioning to you, I have had the privilege of reading 17 of your papers, which are just really excellent science, just doing the heavy lifting of this work-after you completed that and published it, was there a general sense of "a-ha," or did you find there was still a push-back and people trying to find fault with the work? Or did they look at other investigators (we've had a chance to talk with Randy Jirtle and the Agouti mouse model) and maybe they started to see that there was collaborative discovery... how did that actually transition for you?

MS: The same year that we published this article in Science there was another article published on twins (identical twins) basically having different disease frequencies even though the same genetics were there.14 As those twins age in different regions from where they were raised they would develop different disease frequencies, suggesting an environmental impact on disease development. So that suggested, again, an environmental impact, and one of those investigators actually looked at the epigenome of a cell type showing that it was different between those twins, even though the genetics was the same. That came out at the same time, so that was supportive evidence that indeed epigenetics may be the mode of action for environmental factors on genome activity. Randy Jirtle has been studying for a long time the Agouti mouse locus and he had also found environmental impacts on that locus, so he had done that previously, and more recently he's done the same thing with some environmental toxicant-type substances.

Approximately eight months or so later, there was a fellow named Marcus Pembrey in England who did a study (he was more of an epidemiologist).15 They looked at a famine case where several generations after the famine they had increased disease frequency versus controls in a European population (sort of archival epidemiology). That supported the concept that an environment would affect an epigenetic phenomenon that may go forward in terms of disease. So all of those things sort of broke within about a year, and when all the multiple things started breaking more of the scientific community started to accept that something was probably going on. But I must say, from the beginning even up to now, we still get a push back by the genetic community because it is a fairly built in dogma to our current scientific paradigm that genetics really is the principal cause for disease. What our data would suggest is it is a factor, but epigenetics probably has an equally important role.

JB: With that in mind, now you start birthing a new field (or new discipline) within science. It is always interesting when you start seeing that happen how many people attend the first meeting. You start forming a foundation or an institute or a society and you have a group of epigeneticists that have maybe come from different disciplines but you are similarly bonded together by a similar observation or a shared observation. Tell us a little bit about the birthing of the field of epigenetics, because clearly you were one of the first guys as a club member.

MS: Epigenetics has been around for awhile. Just to give you a little history, there was a fellow named Conrad Waddington in the 1940s that came up with the term "epigenetics." He came up with regards to environment-gene interactions that generated phenotype, and that was done back in the 40s. It was mostly theoretical, and he had some very nice papers around that.

That sort of concept, however, was set aside for several decades, and in the mid-70s, the first epigenetic phenomena called DNA methylation, where cytosine residues and DNA get methylated was identified by Holliday in the mid-70s. Then in the late 80s/early 90s, imprinted genes came around and also histone modifications, and so for about a decade and a half (almost two decades) the concept of having things around the DNA that regulate genome activity independent of sequence has been known.

The new phenomena that came out around the time we published was that environmental factors that could actually affect disease has been known for quite awhile by epidemiologists that have been telling us that for decades, but we really didn't have a mechanism to pinpoint how the environment could regulate the DNA since most things don't change DNA sequence. So our study and subsequent studies have started to show that epigenetics probably is the main mechanism by which the environment can influence genome activity. That's really what's pushed. So the field of environmental toxicology is fairly mature; there is a big population of people in it. And the field of epigenetics is reasonably mature as well. What we did is sort of brought those two together, and so now most environmental people are moving towards doing epigenetic-type studies. In addition, people in the field of disease, or epidemiology, or just interested in disease etiology, started moving that way (whether it be cancer researchers or whatever disease they have an interest in) because they've started to get the concept that epigenetics may be a factor in those diseases. I think it was multiple fields sort of merging on a central theme where epigenetics clearly was the bridge between them.

JB: You know, the tradition of toxicology is built around Tolman's laws and this kind of linear dose-response model of extrapolating back to the origins of zero-zero effects, so you have some kind of straight line or curvilinear response. This epigenetic-environment connection suggests (at least, to me, as I read the papers) that this may be a nonlinear extrapolation back to the origin and there may be some profound unexpected effects at low concentrations, which sounds a little like this hormesis concept. Am I at all moving down the right path in this kind of connection?

MS: In part. Clearly the newer concepts over the past few years (for example, in the area of using bisphenol A and so forth) have shown that extremely low doses below what you would anticipate for receptor binding, that those very low doses have physiological effects that are actually either lost or are different at higher doses that you'd expect would be more appropriate to binder receptors. Fred von Saal (with bisphenol A) has been talking about that for a number of years. So that type of activity has been going on, and that is a factor in terms of how much it would take to regulate an epigenetic effect versus how much it would take to regulate a physiological response. Those two things can be quite different. It may take a very little amount to affect epigenetics, which indirectly, then, would affect the physiology. Unless you know the direct target for the substance you are interested in, it's a little bit difficult to go after dose. But probably more important for epigenetics is this is really getting towards the heart of developmental biology, so the fetal basis of adult-onset disease. What it looks like is early life exposures, when an organ system is undergoing a rapid development, is susceptible to environmental factors, changing its epigenome. It then turns around and changes the transcriptome, and eventually that could potentially lead to a disease later in life. So it is much more of a developmental phenomenon, more so than we previously appreciated in disease etiology.

JB: People undoubtedly ask you the question-I mean, there are many questions I've asked you, but one that come to mind from what you just said is: how about cells that have differentiated and are beyond the state of a germ cell, so they are in some form of mitotic division by cell repair and turnover, but they are not in the germ cell state? Is the organism, at that point in the lifecycle, susceptible to epigenetic effects, or is it only that where you have a rapidly dividing fertilized ovum?

MS: Certainly the most sensitive period for exposures is in utero, in the fetus. The next density of area would be early postnatally (when you are a very young individual). The next step would be pubertal development. And then, as an adult, you are relatively insensitive to most things (this is just general

toxicology). And then as an aged adult, that's when a lot of those adult-onset diseases develop. So generally, as adults, we are not as sensitive to environmental effects because of that very process. Those cells are differentiated. Even though we might cause some damage to a tissue, they can repair themselves and so forth, but it's not a developmental process. Therefore, the reason those early ages are more susceptible is organ systems are rapidly developing, so it doesn't take much to shift that development a little bit to then eventually cause a disease as an adult.

JB: In the compounds that you have seen that cause these effects, are they trans-placentally transferred and can it have extrapolation to human, or is that still an area for controversy?
MS: Depending on what time during development, many compounds can actually go across and reach the fetus. There is not a barrier, per se. As the fetus gets older, there becomes more of a barrier. Yes, the compounds we are using can get across that. Bisphenol A, the phthalates, lots of the environmental compounds actually can get across that. The question is whether the amount exposed to the mother, and the amount that gets across, has a physiological effect on the fetus. Those are questions that have not been rigorously looked at in a toxicology manner in terms of levels and metabolism and so forth. For our purposes, we're using (and this is important) the vinclozolin, which is the principal compound we use, and which is the most commonly used fungicide worldwide in the fruit industry, including the wine and apple industries. It is a compound that basically, in occupational exposures and potentially the water supply around these sources, could be available. It breaks down relatively rapidly. There have been a few toxicology-type studies done to show that it takes reasonably high levels to cause an effect. The level that we use exceeds what you would expect in the environment. We also use injections (interperitoneal injections) so we can control the dose, which is also not your typical exposure route. We are strictly using this as a pharmacologic agent to induce an epigenetic transgenerational disease so we can study that basic molecular process of how a compound may influence epigenetics, which then can promote disease onset. With basic information on a molecular level, then you can go back and do much more sophisticated toxicology than doing classic toxicology, in terms of looking at a dose and a phenotype. So that's the approach I think that we are going to end up taking in the future: taking more molecular approaches to study environmental toxicology.

JB: With that in mind, and the fact that methylation patterns are changing upon exposure to these compounds. It raises the question, obviously, as to whether there are other covariables that relate to the regulation of these methylation patterns, and the one that is obvious that people think about is the folate cycle and 5-methyltetrathydrofolate through S-adenosylmethionine. Are there any ways, with nutrients, to modulate the relative sensitivity of these agents on methylation patterns?

MS: Sure. Clearly folate, which is the methyl donor (rides the methyl donor, eventually) for DNA methylation is (and there are a couple of things that will do that)... basically, if there were sufficient amounts around, then you'd get a normal sort of methylation. If there were insufficient amounts around, then you actually may get an effect on methylation that could be abnormal. However, if there was too much around, you could also get an effect that would be detrimental. So putting in additives like folate, you potentially might protect for some things, but you also could turn around and induce others. For example, it has recently been shown that excess folate can actually induce asthma and immune abnormalities in a rodent model, and potentially there is a correlation. Having too little is bad and having too much is bad, so you have to be careful, when considering those additives, of exactly what you are doing. I think that the field is a little bit premature to actually start using that information to suggest how humans supplement their diets. I suspect in the future, however, that type of thing can be used potentially

as a protective-type agent to environmental things, but we just simply need more data before we know exactly how to use them. In other words, what I like to stress is there could be equally as much harm done from putting in too much than having not enough.

JB: So when we take this extraordinary discovery that you and your colleagues have made and we map that against (or superimpose it, or intermix it) with the developing field of nutrigenomics and the recognition of how many single nucleotide polymorphisms exist, it both raises the complexity and it also gives us understanding of points of differentiation moving away from the Gaussian representation of the human. How do you see nutrigenomics interfacing with this concept of epigenomics?

MS: I have been using endocrine disrupters to induce these types of phenomena, and so those are environmental compounds that are sort of readily in our environment. But another major environmental factor that will induce this type of phenomena is nutrition. Caloric restriction in the embryo has been shown in both animal and now human models to actually influence disease onset later in life-things like diabetes, obesity, and so forth. Nutrition is clearly, during pregnancy and early postnatally, a significant factor in terms of environmental factors that will affect your eventual adult onset disease. Understanding that more-understanding what is bad and what is good-will be a significant impact of the nutritional field. I think that is one of the major areas that we are going to expand, and now just in the last year or so there has been some epigenetics linked to those nutritional effects that they are looking at. I think it will be very important, and we just need to get more molecular information on how the nutrient supply is affecting sort of the programming of the epigenome.

JB: In previous discussions we have had the privilege of talking to Dr. Bruce Ames and Michael Fenech and different conversations about factors related to genomic stability which then translates into phenotypic changes. Do these differing methylation patterns then alter the degree of genomic stability in such a way as to have effects that I guess you would call global effects on metabolic disorders?

MS: It has actually been known for a while. Particularly, the cancer field is one of the first ones to come out with the concept that the epigenome can influence genome stability, and so the transformation process in a cancer can actually be influenced initially by whether you have a normal or abnormal epigenome. There are a number of laboratories studying that in the realm of cancers, on several different types of cancers. And so, yes. This is where I would sort of step out and make the speculation that I don't see very many diseases in the human condition that would not have an early-life effect, (having a reprogramming of the epigenome) that then has some sort of susceptibility to an eventual sort of disease onset, and that includes cancer. Now which one comes first, whether it is a genetic susceptibility or whether it is an epigenetic-induced susceptibility, I don't know. It is probably some combination of the two. I guess what I am saying is, I don't believe it is now possible to think about genetics as the only source for disease or susceptibility. It is going to be a combination of the epigenome and genetics, and so it will be a combination of epigenetics and genetics that is going to fully explain disease etiology susceptibility and what diseases you are going to get.

JB: I may be asking a question here that is inappropriate and please tell me if it is because I'm not asking you to speculate beyond that with which you are comfortable. When we take what you just said and we kind of weave it into society today, we can see there are some conditions within our population in children that are increasing quite dramatically relative to what they were even 40 years ago, which are hard to explain on the basis of just maybe Mendelian types of traditional views. Two I am thinking about

are ASD (autistic spectrum disorder) and the other is NALFD (nonalcoholic fatty liver disease) which, when I was in medical school in the 60s we were told that wouldn't probably ever be seen very frequently, and now we are seeing it quite frequently. Both of those kind of speak to the liver and the brain, something of lipophillic nature that could modulate things like epigenomics. I'm using wide brush strokes here so you can certainly censor me. Do you think any of this that we are seeing in society, beyond that of just better diagnosis, relates to some of these potential things that you've discovered?

MS: Yes. I think there are a number of aspects of disease which suggest a significant environmental impact. The first is if you go anywhere in the world, every region has a different disease frequency, even if you have similar genetics. Sometimes if you take a person early in life and put them in another area they actually develop the disease frequency where they move to, suggesting environmental impact. The second most compelling sort of argument is the increase in the disease frequency for nearly every disease over the past two or three decades, sometimes going from 1-2{56bf393340a09bbcd8c5d79756c8cbc94d8742c1127c19152f4230341a67fc36} up to 15-20{56bf393340a09bbcd8c5d79756c8cbc94d8742c1127c19152f4230341a67fc36} of the population. That kind of an increase within that short of a period has to be an environmentally induced phenomena; it cannot be a simple Mendelian genetic sort of issue. So, yes, I think when you see a dramatic increase in something like autism (the rate it has gone up), there is sort of an argument that clearly diagnosis is an issue (the efficiency of it), but if you can count that, you still have an increase in the disease that has to be an environmentally induced. Now one area that neuroscientists have been talking about for over a decade is they have not been able to explain neurodegenerative diseases very well using genetics. Actually there are very few genetically-based neurodegenerative diseases, but epigenetics could explain a great deal of that and they have been speculating that for awhile. I think the role that epigenetics plays in the brain may be very important.

JB: With that in mind, let me go to an example that you probably are familiar with because it seems like it is almost directly mapped to your discoveries and work and, again, I may be extrapolating ad absurdum here. I'm talking about David Jacobs/Duk Lee's work, which you probably are familiar with, looking at an association between serum concentrations of persistent organic pollutants and insulin resistance, metabolic syndrome, and type 2 diabetes16 in which they find, by looking at NHANES 3 data (the National Health and Nutrition Examination 3 data for the US) that there actually is no strong statistical correlation between obesity or elevated BMI and diabetes in the absence of people having high normal or elevated GGTP in their serology (gamma glutamyl transpeptidase), which is a surrogate marker (it appears, from work that has been published over the last few years) for exposure to environmental toxicants or xenobiotics that upregulate the use of glutathione and cause the body to compensate by enhancing glutathione synthesis. It suggests from this-and again I want to emphasize "suggests"-that there is a connection between persistent organic pollutants (POPs) and disease, which then begs the question: how do these POPs influence disease? To me, the explanation is tied to your discoveries. And then we take that one step farther and look at the Journal of the American Medical Association, in the September 17 issue this year, at the paper that Ian Lang and his colleagues published on the association between urinary bisphenol A concentrations with a complex range of medical disorders and laboratory abnormalities, which again seems to suggest that we're seeing these chronic metabolic diseases. In fact, the editorial that follows it even emphasizes that-that from nontraditional dose-response type relationships of exposure that are influencing a wide panorama of diseases, and they seem very prevalent in people with high normal GGTP levels in their serology. To me, this sounds like a clinical indicator for what you have observed in your studies. Am I at all making an association that has any reasonable connection?

MS: I think you are on the nose. For example, diabetes and obesity is clearly a metabolic syndrome-it's a metabolic disease-and it is fairly well established as to the etiology of the disease. The brain has a significant role in the regulation of metabolic disease as well. I think you are, overall, influencing the system (the entire system of metabolism), and some of the outcomes of that are various diseases like diabetes or obesity. Clearly, the data to suggest nutrient support, environmental factors, and environmental compounds can influence those types of diseases is there. I think, yes, it could be that metabolic disease is clearly more of an environmentally influenced phenomenon. If that's the case, then I would suggest that it is going to be an earlier life exposure causing a sort of programming event that later in life causes an abnormal metabolism, which then subsequently leads to the adult onset disease. I think that that overall trend is there. The proof now will be to actually go in and look at what is the exposure, showing the epigenetic changes, how that influences the metabolism, which then correlates to the disease. So I think you are definitely correct in your hypothesis, and now we just need more sort of a systems approach to actually approach it.

JB: I know that you are actively involved in some of that systems biology-type research. Could you kind of give us a glimpse into the future as where you see your (and your colleagues') work heading, knowing that this is very complex? This is like trying to eat the elephant in a single bite.

MS: I can give you two examples of two ongoing studies right now, and then I can speculate sort of what this might be leading to in the future. The first: we have done a series of experiments where we did this exposure in utero to this endocrine disruptor. And this exposure occurred during sex determination, so we affected gonadal development and the germ line development. And it also occurs during brain development, when the brain is actually developing, as well. So we speculated that we'd have some significant brain effects. So we have taken three generations removed from the exposure (brains) and found that indeed the brains, in several different regions, have different transcriptomes (what sets of genes are expressed). Hundreds of genes are actually changed, and it is a transgenerational thing because it goes between generations, the same set of genes being affected. This transgenerational transcriptome would then likely lead to some sort of neural abnormality. When we have done behavioral studies on these animals, indeed what we found was that the females have a statistically significant higher level of anxiety, and the males have a statistically lower level of anxiety and are higher risk takers. So there is a behavioral effect on those animals, and it is opposite between male and female, and it is linked to the brain transcriptome and this early life, transgenerational exposure. That is a study that we are putting out, sort of showing that through an epigenetic change in the germ line we can actually alter brain development and subsequent behavior, so this is sort of providing that systems link from neuroscience to sort of this environmental exposure. That is a current study that we are sort of in the midst of putting out. The other corresponding study was in 2007. We published an article on a similar type of phenomenon showing sex preference (or mate preference) in sexual selection was actually altered in the males and females as well-again, a brain-initiated function.17 I think that whole issue that you brought up earlier on how the brain may be involved in the diseases associated with abnormalities in the brain could be epigenetic... we're moving in that direction.

In a second series of studies we are doing a much more genome-wide approach and moving towards the human to actually look at the epigenome in the human and mapping some of these epigenetic changes, both in the germ line and in somatic tissues to see if we can identify epigenetic biomarkers. This is where you'd have a DNA methylation change in the DNA, or a group of them, that may correlate to a specific disease or another. The potential to have these epigenetic biomarkers as diagnostics for the disease to then

go in and potentially (early in life) identify that you have a 95{56bf393340a09bbcd8c5d79756c8cbc94d8742c1127c19152f4230341a67fc36} chance that 20 years from now you are going to develop this disease (that type of a sort of epigenetic biomarker). This is a much more of a developmental effect. In the past, we have never had the ability to have those early-stage diagnostics.

So the future could be that when you are in your 20s you would have your epigenome mapped, and because of certain biomarkers you might have, you could predict what diseases you may get later in life. Instead of waiting for the disease to develop to treat it, when you are in your 30s or 40s, prior to the disease onset, you could potentially map what changes are happening in that tissue, and therapeutically treat them to prevent the disease from developing. This would lead us down the path to preventive medicine, and the ability to do that would be having these early-stage diagnostic markers that could be epigenetic biomarkers that would be linked to those diseases. I think that potentially will be the future of medicine: going from a reactionary medicine situation to a more preventative medicine situation. The key, really, is a better understanding of the etiology of the disease, including the epigenome.

JB: That's a really fascinating vision for the future. I have to say, just parenthetically, that is certainly consistent with what we have tried to structure as out functional medicine model as it pertains to the connection between genes and environment and outcome. I'd like to close with just one question and I don't want this to sound alarmist because I think there is always a tendency, probably, when people hear this work that it sounds a little doom and gloomish... I'd like to phrase it in a slightly different way. We all know that this article that appeared in the New England Journal of Medicine in 2005 that was a consortium group of high profile individuals that wrote about future life expectancy and suggested that the lifespan of our children born today maybe shorter than that of their parents, on a statistical average, which would be the first time in the history of the United States.18 This is at a time when we are spending twice per capita on health care than any country in the developed world (we are 30-something in health outcomes according to the World Health Organization). That then brings the question up: if you get the diminishing returns of a model indicating that maybe your model is broken or needs modification, and if transitions occur with the epigenome and we start to see it mapping against certain diseases, what do we do in our curriculum? What do we do in our society to educate people about how the decisions that are made everyday influence not only that which we are living, but that which subsequent generations will live? Is it through the science? Is it through public activism? How would you see this transmitting into a cultural change?
MS: Well, it's a combination. Ten years ago, if you basically asked a scientist that question they would tell you that you will have to focus on the DNA sequence and the genetics and that is pretty much going to be your answer. I think clearly that that is part of the answer, but we had this huge black box, which now epigenetic may help fill, that just develops and stands in terms of its application. So I don't think you can ever say that science is fully aware of everything we need to know about disease or health. I think you need to move the science forward to develop those new insights--things we didn't expect-to help answer some of those paradigms or conundrums that we couldn't explain. Part of our movement forward, in terms of improving our health, will be to advance the science, and so as a society we are going to have to support basic research and moving that science forward, otherwise we will simply get into a situation where we think we know how everything works, but probably we don't. That will be the first.

The other thing that scientists need to do, which your program does for physicians but we probably need to take it to the more public level, is the scientists need to educate the public about some of these fast-

paced, moving scientific advancements, so that they are aware of the situation. I think if the general community realized that things like caloric restriction during pregnancy may actually induce obesity or a diabetes situation in their children there would be a consideration or a request from the health community on what they should or shouldn't do to avoid that. So just educating the public about these scientific advances is probably something we should do in the sciences as well. So those are the two things I would suggest, on a societal level. We do definitely need to move the science forward, but we also need to educate the public more.

JB: I can't tell you how much I appreciate you spending the time with us, and also the diligence and the difficult that you have done in bringing this whole field up with your colleagues to the level of understanding that you have. It's clearly obvious that we have a lot of work ahead of us. I guess the good news is if you can put these imprints on and you can take them off and this leads to some degree of plasticity versus the deterministic view of Mendelian genetics, so I guess that's the nice part of the story: if we can clean up our mess, we can also clean up our epigenome.

MS: Generally, knowing more of the basic reasons why things happen helps us actually fix the problems, correct?

JB: Yes. Dr. Skinner, thank you very, very much. We wish you the best in your continued work. We will be following it very, very closely.

MS: Thanks very much and I appreciate the opportunity.

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