

April 2004 Issue | Elena Volpi, MD, PhD Associate Professor of Medicine

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Welcome to *Functional Medicine Update* for April 2004. The 11th International Symposium on Functional Medicine will take place next month, May 11-15, 2004, in Vancouver, British Columbia, where we will focus on “The Coming Storm: Reversing the Rising Pandemic of Diabetes & Metabolic Syndrome.” We are excited to have a Clinician of the Month in this issue of FMU who will discuss part of the story we will be talking much more about at the symposium.

In order to prepare for that discussion, I would like to talk about the protein connection to insulin, which has certainly received a lot of press recently. As I was driving down the freeway the other day, I noticed a big banner on a 7-11 store that read: “7-11 is Atkins Friendly.” It occurred to me that this issue has come full circle. When 7-11 becomes “Atkins Friendly,” I know the paradigm has shifted.

Let us turn to what is happening in the bioscience area related to insulin, dietary protein, and amino acids. I hope through this discussion you will get a somewhat different perspective as to what has been sensationalized, popularized, exaggerated, and marketed for promotional reasons in a way that subliminally seduces many consumers into believing something that may be quite different from what the facts are.

It is becoming common knowledge that body composition plays an important role in determining physiology. As body fat percentage increases at the expense of reducing body muscle percentage, there is an altered state of fatty acid metabolism, glucose metabolism, insulin sensitivity, and general mitochondrial oxidative efficiency, because muscle is rich in mitochondria and fat cells are poor in mitochondria. Therefore, oxidative chemistry and oxidative phosphorylation are shifted as body fat is increased, at the expense of body muscle.

Many years ago, Dr. Covert Bailey referred to the “thin/fat person” as one who, over the years, remains in the same dress or pants size, but whose internal body composition may have changed considerably due to lack of exercise and poor-quality diet. The “thin/fat person” tries to stay thin with calorie-restriction, but if CT whole-body scanning or examination of musculature is performed, it would likely reveal that his or her body is heavily marbled with fat, much like feedlot-fed beef.

There are some good examples in the literature that discuss the relationship of cross sectional CT scanning of various large muscles such as the gastrocnemius with percent body fat and body muscle in active versus sedentary individuals, and the finding that intracellular fat deposition begins to accumulate

in those with a sedentary lifestyle.

The type of fat of most concern, however, is not that which accumulates in the limbs, but that which accumulates around the abdomen, the so-called visceral adipose tissue, or VAT. We can say that health problems with fat are often present in people who are “over-VAT,” not “over-FAT.” The VAT (inter-abdominal body fat) is that which appears to present the greatest risk to age-related, chronic illness such as heart disease, diabetes, some forms of cancer, and perhaps also to cholelithiasis and gall bladder disease. It is these types of regional fat depositions that seem to be strongly associated with disease related to obesity or adiposity.

Determining Body Composition

One of the first things one needs to look for clinically is how much body muscle has been replaced by body fat, or the percentage of body fat, and the best way is to determine body composition. Traditionally, the simplest method is to measure height and weight and develop what is called the body mass index (BMI). Using a nomogram scale, the height can be determined in inches versus the weight in pounds to develop a BMI number. BMI>25 is overweight; BMI>30 is obese; BMI>40 is morbidly obese. (The only exception might be for individuals with heavy musculature, such as highly-trained athletes.) For the average individual, the BMI is a pretty good approximation of body composition, or percent body fat.

BMI is often coupled together with the waist-to-hip ratio. That ratio is achieved by measuring the circumference of the hips at the widest point and the waist circumference, about an inch or two above the umbilicus. Dividing the waist number by the hip number provides the ratio. If that number is greater than 0.8 for a woman, or greater than 1.0 for a man, it suggests increased incidence of abdominal obesity associated with increased BMI. Increased BMI and increased waist-to-hip ratio represent the VAT. Only a tape measure is needed to gather some inferential information about body composition.

CT Scanning and Bioimpedance (BIA) Measuring Techniques

There are more accurate methods of determining body composition, such as CT scanning neutron activation (not available to most individuals unless they are in a research situation), and bioelectrical impedance analysis (BIA), which uses resistance and conductivity measurements of the body. BIA is a pretty reasonable technology for evaluating body composition in a person who is properly hydrated and appropriately nourished relative to their normal diet. This would not include people who have consumed five cups of coffee and three drinks of Scotch before having a BIA, which would induce a diuretic effect. Subjects should be normally hydrated. Except for those who have various ponderous physiologies, the BIA regression equations built into the machines that give rise to the percent body fat, percent body muscle, and percent intracellular water calculations, are quite accurate and correlate nicely with other more sophisticated technologies. As a person reaches a very high BMI, this equation tends to break down. In these cases, the BIA calculation from normal bioimpedance analysis tends to be less accurate. For people in the normal or overweight range of body composition (19-30), the BIA machines provide accurate regression calculations compared to those determined by other methods. The simplest in-office technology for evaluating body composition that has the most qualitative inference is height-to-weight ratio and BMI. BIA renders a quantitative calculation that gives more compartmental and regional aspects of body composition and intracellular fluid.

A good paper on the comparison of BIA prediction equations for fat-free mass in a population-based sample of 75-year-olds was recently published in the journal *Nutrition*.¹¹ The investigators point out that

in cases where a person has a lot of sarcopenia and muscle wasting (such as an elderly person), the BIA is not very accurate, and it is not accurate for people who are morbidly obese. In the mid-range of normal body compositions, BIA has a fairly accurate correlation in terms of body fat, body muscle, and body water.

Once the body composition is determined, and let us say that person is a man with a percent body fat of 27 percent with increased extracellular fluid and compromised body muscle (low percent body muscle), what is a good clinical approach? Does he need to be on a low-calorie diet, exercise program, some form of anabolic steroid to increase muscle mass, or a nutritional supplementation program? If he is going to be placed on a lower-calorie diet, what type of diet? Should it be a diet high in complex carbohydrates, high in fiber, modest in protein and low in fat, or one that is high in protein, lower in carbohydrate, or any or none of the above? These are controversial and confusing questions as to how we personalize dietary approaches for individuals with altered body composition.

Similarly, the patient might be a woman with an initial percent body fat of 34 percent who has increased cellular water levels and who is at risk to obesity-related problems, with an increased waist-to-hip ratio. The same questions would prevail. What type of dietary and lifestyle intervention would be appropriate?

Extracellular Fluid and Diet Recommendations

In our experience, one of the first things that might be done in the case of a large amount of extracellular fluid, is to put a person on a diet as neutral and low-allergy as possible, without making sweeping changes in calorie intake. Some people call this an oligoantigenic diet; other people might refer to it as a detoxification program. Others might call it just a good, simple, clean diet. Whatever term might be used, the recommended diet is one that would employ foods grown in the ground with lots of color (fresh fruits and vegetables). It would avoid the color white in the diet—white sugar, white fat, white flour, and white alcohol—and include things as organic as possible, staying away from dairy and wheat products, or glutenous grains. We cannot be certain who might have some kind of sensitivity to the food families containing proteins to which many people are sensitive, but the most common allergenic proteins are found in dairy, soy, and wheat.

When putting a person on a fairly low-allergy diet, rice protein-based approaches are often used, because rice is well tolerated in western societies and has good-quality protein. Fiber would be increased, and vegetables and fruits would be increased. There are diet plans based on the Mediterranean Diet that avoid gluten grains. When an individual has been on this program for two to three weeks, immune system responses change. A lot of water weight that might have been retained as a consequence of immune system response to a purported offending agent, i.e., an inflammatory-promoting agent, may be lost. The person is now at a baseline physiology and it is easier to examine body composition. Extracellular fluid has decreased; some weight has been lost; the person is feeling better, and energy needs are in clearer focus. One can then move into the next phase, which is to tune up metabolism.

The Metabolic Tune-up

Dr. Bruce Ames discusses this in an article that appeared in the January 2004 issue of *Archives of Biochemistry and Biophysics*.^[2] He talks about the metabolic tune-up—supplementation and intervention with appropriate nutrients to improve muscle physiology and mitochondrial function.

That leads us to diet approaches. There are many different opinions on the ratio of protein, carbohydrate,

and fat amount in the diet, as well as type and percentage. We only have to look at the *New York Times* Best Seller List for the past three years to see the controversy that prevails about what the best diet is—the one with the appropriate ratio of protein, carbohydrate, and fat. My question is (we will be talking about this with our COM on Side 2), what is the best *kind* of protein, *kind* of fat, and *kind* of carbohydrate? That may be more important than the relative ratio. It seems as if we are ruled by numerology. We are so tied up with ratios, we get food-phobic about having exceeded a certain percentage of one nutrient versus another, rather than remembering that it is the *type* of carbohydrate. Is it complex? Is it whole-grain? Is it unrefined? Is it white starch? Is it sugar? What *type* of fat? Is it partially hydrogenated? Is it polyunsaturated? Is it monounsaturated? Is it saturated? Is it oxidized? What *type* of protein? Is it animal or vegetable protein?

These questions are exceedingly important in determining the outcome of the body's hormonal postprandial messaging response to the diet. It is the postprandial period, during which the orchestration of all sorts of hormones that sweep into the blood creates different tunes of genes downstream that ultimately regulates things like body composition. It is not as simple as just thinking about calories in and of themselves. It is the *type* of calories and *type* of nutrients that influence the messaging system of the body—such as the sex steroid hormones, glucoregulatory hormones, neurotransmitters, and the appetite-control hormones, including what are called adipocytokines, the fat-related neurotransmitters, neuroregulators, and immune-regulating substances. ?

Fat is now considered an endocrine organ. Isn't that remarkable? It produces a whole series of messenger molecules. In fact, in very under-lean people, the largest endocrine organ in their body is body fat; it can take over their physiology. It produces its own messenger molecules—TNF α , IL-1, IL-2, adiponectin, resistin, and leptin. These molecules—even sex steroid hormones like estrogen and C-reactive protein—can be manifestations of increased body fat percentage.

As body fat percentage increases, physiology changes. There is a functional change. The individual moves from one resting state of physiology to a new resting state, a new homeostasis. We often use the term “homeostasis” implying that it means healthy regulation. But one can be homeostatic with malignancy, cardiovascular dysfunction, diabetes, or inflammation. It is breaking the cycles that give rise to chronic illness that leads a person back to a resting physiology called a healthy homeostasis.

The focus of this issue of *Functional Medicine Update* is on the specific role that protein and possibly carbohydrate play in the regulation of the postprandial messages that alter body composition. Over weeks, months, and years of eating and living, these messages create a body that has altered percent body fat, altered percent body muscle, and altered percent extracellular fluid, as measured by BIA.

In order to understand the protein connection, I am going to trace back through some history.^[3] The first reported research I could find on this issue was that of Ignatowsky in 1908, when he examined the role of various macronutrients on atherosclerosis. You may recall that Atwater did his landmark work on the calorogenic content of food using human calorimeters at the turn of the 19th century. Ignatowsky's work follows Atwater's work on the calorie content of protein, carbohydrate, and fat. Ignatowsky believed there was a toxic metabolite in animal protein that led to atherosclerosis. When he fed high amounts of meat, as well as milk and egg yolk protein, to adult and weanling rabbits, he was able to cause atherosclerosis.

About the same time, the Russian physiologist Anitschkov was working with a similar strain of rabbits. He found that feeding the rabbits a soft diet rich in saturated fats and cholesterol could produce arterial sclerosis in the animals. It was a soft, fat, oily, gooey substance, and on necropsy, when he examined the animal's arteries, he found he could scrape it out on his fingers. It was oily, waxy, and gooey, and that led to the simple explanation that fat does not dissolve in water. If you eat a lot of fat, it clogs up your arteries and eventually results in atherosclerosis. That was the origin of the cholesterol hypothesis. I want to emphasize "hypothesis" as it pertains to the origin of heart disease.

About the same time, Ignatowsky was talking about dietary protein as a major contributor to atherosclerosis. There was an interesting battle being conducted between the two camps at that time. For the next two decades, experimental efforts from many laboratories were directed at determining which, if any, animal protein was the most atherogenic. Even in the face of the work by Anitchkov on cholesterol and fats producing heart disease, those in the protein camp continued, over the next 20 to 30 years, to follow-up on the protein hypothesis—that a toxic metabolite of protein caused atherosclerosis.

In 1926 Clarkson and Newburgh showed that the amount of cholesterol present in the animal protein they fed animals to make them atherosclerotic was insufficient to be atherogenic by itself, demonstrating that there was some factor other than dietary cholesterol (lipid) that determined atherogenicity of the diet, presumably protein. In 1940, Meeker and Kesten showed that animal protein (casein, the principal milk protein) was more atherogenic than plant protein (soy). The work of Carroll and his coworkers that followed the Meeker and Kesten work, showed that most proteins of animal origin were more cholesterolemic for rabbits than were proteins of vegetable origin, although there was some overlap.

The observations made over the last 100 years have been interesting. David Kritchevsky at the Wistar Institute in Philadelphia, has been discussing the dietary protein connection to atherosclerosis for many years. He was the first person I listened to 30 years ago who got me thinking about the subject. We often think that serum lipid abnormalities are a consequence of dietary lipid abnormalities because lipid makes lipid. But dietary protein, the carbon skeletons of the amino acids that make up protein, can also be converted by metabolic/anabolic function into triglycerides and serum lipids, and have effects on *de novo* cholesterol biosynthesis. It is not as simple as saying, "fat makes fat." Protein can make fat, as well, and it appears from the early work and on up through the work of Carroll et. al., that animal protein may be more atherogenic than vegetable protein. That is an interesting part of the story. Remember I said earlier, it is not just the ratio of protein to carbohydrate and fat; it is also the *type* of each of these.

In the 1970s and 1980s, David Kritchevsky led us to think that perhaps we should be concentrating on the difference between animal and vegetable protein. How do they vary when fat is taken out of the equation? To understand this, we need to go back to 1983 and look at papers like the one titled "Regression of casein and cholesterol-induced hypercholesterolaemia in rabbits."^[4] This is a fascinating study done in the Netherlands in which investigators showed they could develop arteriosclerosis in rabbits by feeding them a low-fat diet, but one that was enriched in casein animal protein. They could cause regression of hypercholesterolemia and arteriosclerosis by transitioning the animals onto an isocaloric amount of vegetable protein, in this case, soy protein. The concept that there is something toxic to the arteries in animal protein appeared to emerge from this research.

In 1985, a follow-on paper was done at the Department of Biochemistry, School of Medicine and Health Sciences, George Washington University; the Wistar Institute of Anatomy and Biology in Philadelphia,

and the Department of Pathology at the University of Pittsburgh, School of Medicine. The title of the paper was “Effects of Casein and Soy Protein on Hepatic and Serum Lipids and Lipoprotein Lipid Distributions in the Rat.”^[5] Again, a similar theme was found (moving from the rabbit to the rat), by feeding the animals an isocaloric diet reasonably low in fat, but containing either casein or soy protein in equal amounts, showing that the casein diet was much more atherogenic. Animal protein increased the *de novo* biosynthesis of cholesterol and serum lipids, whereas the soy protein diet did not. It also appeared that the animal protein diet had effects on hepatic 7 α -hydroxylase, the rate-limiting enzyme for the conversion of cholesterol to bile salts. Supplementation of the soy diet resulted in lowering of total lipids and increased cholesterol 7 α -hydroxylase. However (here is the important point), when they took the soy protein diet and supplemented it with the amino acid lysine to make it equivalent in its lysine content to that of beef or casein protein, the diet became atherogenic. It initiated increased concentrations of serum lipids and cholesterol, as well as atherogenicity. That opened the door in 1985 for examination of the fact that there was something about the arginine/lysine ratio in dietary proteins that could influence their atherogenicity and lipid effects.

What is that effect? This is where the story becomes quite fascinating. (I will be discussing the arginine/lysine ratio with our Clinician/Researcher of the Month on Side 2.) First of all, animal proteins are much higher in lysine and lower in arginine than vegetable proteins; conversely, vegetable proteins are higher in arginine and lower in lysine. Fortifying a vegetable protein with lysine to make the arginine/lysine ratio equivalent to that of beef protein, results in a similar cholesterol-increasing effect. There appears to be a thermostat of the cholesterol/lysine ratio on cholesterol and lipid biosynthesis, and therefore atherogenicity. What did I say earlier? It is the *type* of protein as well as the amount.

In 1986, work was done at Shizuoka University in Japan, looking at the relationship between amino acid composition of the diet and plasma cholesterol in growing rats fed a high-cholesterol diet.^[6] Feeding rats a cholesterol-laden diet, but with soy protein containing a low lysine/high arginine level, resulted in lowered risk to atherosclerosis and injury to the arteries. If the rats were fed a high cholesterol diet with a protein that was high in lysine/low in arginine, it increased atherogenicity. This produced a similar observation to that of Kritchevsky’s published in 1985.

In 1992, following the same theme, physiological research out of central Europe showed that casein and soy flour proteins and their amino acid content had a significant influence on liver *de novo* lipid biosynthesis, including cholesterol synthesis and triglyceride synthesis in experimental animals.^[7] Once again, investigators showed that the arginine/lysine ratio appeared to be one of the major determinants. The higher arginine/lower lysine foods (plant-based proteins like soy protein), had a lower cholesterol-stimulating effect and triglyceride-stimulating effect than did the higher lysine/lower arginine foods. This theme seems to have emerged over a period of 10 to 15 years, going all the way back to Ignatowsky’s observations about the vasculotoxic effects of animal protein.

In the *International Journal of Vitamin and Nutrition Research* in 1997, an interesting paper was published from a group out of the Beltsville, Maryland, U.S.D.A Research Labs in collaboration with the Western Region Research Branch of the Agriculture and the Agri-Food Canada research department.^[8] In this study, the investigators looked at the influence of casein, soy protein, the ratio of amino acids in those proteins, and the effects on cholesterol and lipoprotein fractions in guinea pigs. The effects of three dietary protein treatments on cholesterol content of plasma, lipoprotein fractions, and oxidation status of liver lipids were compared. This is the LDL oxidation model. All diets were adequate in soluble fiber and

well balanced in fatty acids, which provided 30 percent of the total energy. After seven weeks, dietary treatment with casein compared to soy protein increased cholesterol in a subfraction of LDL. Dense LDL, the most atherogenic particle, was increased with the isocaloric animal protein as compared to the soy protein. We are getting even more specific in the potential atherogenicity of animal protein when we begin to look at sub-fractionation.

The investigators in this study also found that there were adverse effects on HDL with the animal protein—a lowering of HDL. There were no effects of dietary treatment in the TVA, or the so-called lipid peroxide substances that were extracted from the liver; these diets appeared to principally alter cholesterol synthesis and dense LDL.

What does this lead us to? It leads to Ignarro and Murad and their Nobel Prize-winning discovery of the molecule nitric oxide (NO). Historically, pathfinders all the way back to Ignatowsky, and their hypotheses relating to the atherogenicity of animal protein in contrast to saturated fats in the animal protein-rich diet, suggest that the ratio imbalance of arginine to lysine (higher lysine and lower arginine) has something to do with the adverse effects of dietary protein on the arteries.

Murad and Ignarro were awarded the Nobel Prize in Physiology and Medicine for their discovery of the production of NO from arginine by an enzyme called nitric oxide synthase (NOS), which is mediated through the cyclic GMP pathway. NO is an essential biomolecule for endothelial health and therefore, it may be possible that the arginine/lysine ratio changes seen with different dietary protein sources have an effect on NO dynamics and the potential atherogenicity and immune effects associated with atherosclerotic disease or atherogenesis. More and more, this is now being seen as “the arteries on fire” related to autoimmunity and the concept that atherosclerosis has an immune origin. This may also tie back to modulation of factors associated with NO chemistry that appear in the artery wall or in the liver itself.

Arginine has a powerful immunomodulatory effect. This effect has been demonstrated in many studies, including work in post-surgical patients who have been infused with high doses of arginine to improve wound healing. It has also been demonstrated in lowered levels of arginine administered to animals, demonstrating effects upon immunochemical function of cell-mediated and non-cell-mediated immune defense, and looking at how certain T lymphocytes—natural killer cells—are influenced by arginine, as well. Arginine participates in a variety of immune functions that appear to be related to the various cell types associated with the atherogenic process. We are taking some fairly wide-sweeping brush strokes here. We are trying to tie together the arginine/lysine content at the whole organism level. Increased lipid levels are associated with a high lysine/low arginine diet, and that appears to be related to immune function, arterial health, and proper regulation of NO, which originates from arginine metabolism from NO synthase. There is a nice review on the immunomodulatory effects of arginine in the journal *Surgery*.^[9]

If we take that a step further, we might ask what influence the immunomodulatory effect, the NO modulating effect, the serum lipid-modulating effect, or arginine and lysine, have on insulin sensitivity. Insulin is tied to the web of interacting hormones that occur postprandially and regulate downstream function of the individual. Therefore, is there a relationship among atherogenicity, body composition, and adipocyte physiology? Is there something that relates to the type of dietary protein and its potential effect on insulin sensitivity?

There is a wonderful article by Sanchez and Hubbard from the Department of Nutrition, School of Public Health, and Department of Pathology, School of Medicine at Loma Linda University that appeared in *Medical Hypotheses*,^[10] titled “Plasma amino acids and the insulin/glucagon ratio as an explanation of the dietary protein modulation of atherosclerosis.” Does that title sound familiar relating to what I have been developing in this story? The authors state:

“The amino acid composition of the diet influences the postprandial levels of plasma amino acids along with the hormones insulin and glucagon in humans fed single test meals identical in composition except for protein source. Soy protein (hypocholesterolemic), vs. casein (hypercholesterolemic), contains a higher amount of arginine and glycine and induces an increase in postprandial arginine and glycine. Soy protein induces a low postprandial insulin/glucagon ratio in both hypercholesterolemic and normocholesterolemic subjects. Casein induces a high postprandial insulin/glucagon ratio among hypercholesterolemic subjects. Amino acids such as arginine and glycine are associated with a decrease, while lysine and branched-chain amino acids are associated with increased serum cholesterol levels. Our data are consistent with the hypothesis that the control of cholesterol by insulin and glucagon is regulated by dietary and plasma amino acids. From this hypothesis the insulin/glucagon ratio is proposed as an early metabolic index of the effect of dietary proteins on serum cholesterol levels, a risk factor and a common mechanism through which dietary and lifestyle factors influence cardiovascular disease.”

Improved insulin sensitivity is achieved with dietary proteins that are lower in lysine and higher in arginine.

Dietary glycemic load may play an important role in determining insulin sensitivity, but it may also be related to the source of dietary protein and amino acid composition. A high dietary glycemic load is associated with risk to colorectal cancer, as suggested by the Women’s Health Study. This information was recently published in the *Journal of the National Cancer Institute*,^[11] showing that high glycemic load diets, those that increase insulin and glucose levels, are associated with increasing incidence of colorectal cancer in women.

It is not just carbohydrate. It is not just the amount of protein, but it may be the *type* of protein. Glycemic load of the diet is not determined by just measuring carbohydrate alone. It is measuring the overall balance of these nutrients that influence postprandial gene response and messenger molecules that signal downstream how cells respond to their environment. As glycemic load comes of age, we should be looking at it not just as a measurement of available carbohydrate, but as one of all the signals coming from the diet that influence the regulatory hormones and alter adipocyte physiology, sarcomeric muscle cell physiology, mitochondria, and changing body composition. It is much more interesting and clinically important information that is emerging from this research.

An interesting article, titled “Glycemic load comes of age,” appeared in the *Journal of Nutrition* last year.^[12] Author David Ludwig says we should be looking at glycemic load as a total measurement of all the contributors to the various dietary components—protein, fat, carbohydrate, fiber, minerals, and vitamins—all the things that cause regulation of glucose. This might explain why type 2 diabetes and insulin resistance respond to a vegetarian diet, even though this diet is higher in carbohydrate. It is high in complex, unrefined carbohydrate, and higher in vegetable protein that is higher in arginine and lower in lysine. Type 2 diabetes can, in fact, be favorably influenced by a vegetarian diet. This is the topic of an article in the *American Journal of Clinical Nutrition*^[13] discussing how a diet richer in carbohydrate, but

unrefined, proper, and balanced in vegetable protein, will have beneficial effects on insulin sensitivity and help to manage the type 2 diabetic. David Jenkins is the principal author of this article and he will be a presenter at the 11th International Symposium on Functional Medicine.

In the Insulin Resistance Atherosclerosis Study, it was shown that high intake of whole grains was associated with increases in insulin sensitivity, not decreases.^[14] Carbohydrate, in and of itself, is not dangerous. It is the form in which it is administered to the body and whether it is balanced with the proper amount of vegetable protein.

Dietary protein in diabetes, as was described in the article in the *American Journal of Clinical Nutrition*, requires that we look at it in terms of whether it is properly balanced with regard to vegetable protein and the amino acid ratios that come from proteins. Amino acids play an important role in the regulation of lipid synthesis, insulin sensitivity, and gluconeogenesis. All these factors play a role in the glycemic load of the food and diet, and ultimately its effect on body composition. It is this discussion we will be sharing with our Clinician/Researcher of the Month on Side 2.

INTERVIEW TRANSCRIPT

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JB: Once again, it's time for our Clinician/Researcher of the Month. We are pleased to interview a world-renowned investigator, Dr. Elena Volpi, Associate Professor of Medicine at the University of Southern California, Division of Endocrinology and Diabetes. I think most of you are well aware of her work. It was highlighted for many of us in a landmark paper that appeared in the *Journal of the American Medical Association*, titled "Basal muscle amino acid kinetics and protein synthesis in healthy young and older men." [15] Dr. Volpi was a principal author of that paper. Some of us have presumed that basal muscle amino acid kinetics are slower in older men and faster in younger men, accounting for the difference in muscle mass. The work of Dr. Volpi and her colleagues dispelled that myth and raised new questions. It is with that in mind that we are going to talk with Dr. Volpi about the protein connection to body composition and how that relates to the insulin sensitivity story.

Dr. Volpi, thank you for being with us on FMU. My first question is, what led you to focus on protein-related issues in your research over the last 20 years?

Influence of Dietary Protein on Postprandial Insulin Mechanisms

EV: Thank you so much, Dr. Bland, for having me on FMU. I've always been fascinated with the mechanisms by which the proteins of the body work. Consider that proteins are the major translators of

the genetic information in the body. They can be enzymes; they can be structural proteins like muscle proteins or contractile proteins. I find it extremely interesting to look at how the genetic information is translated to the actual function of the body. I became interested because I'm an endocrinologist by trade and I was very curious about looking at metabolism in relation to hormones. My initial studies were done in healthy people in order to understand the physiology of hormone action, especially insulin, and also the physiology of nutrition in relation to the function of proteins.

From there, I moved into issues related to aging because it was quite an untouched field at the time. I started doing research on aging in 1996 when I moved to Texas. From that point on, I began studying the potential mechanisms by which muscle is lost with aging. That is my general orientation right now. From endocrinology, I moved into the field of gerontology, geriatrics, and functional medicine.

JB: I have been so impressed with your work because it embodies so many of the principles that we here at the Institute for Functional Medicine have been trying to get doctors to understand from a functional perspective. In 1996, you were a principal author of a paper indicating that dietary protein had an influence on postprandial insulin mechanisms.[16] Would you describe how you got into that study and what the results of that first investigation were?

EV: That study was done in healthy, young people. We were looking at the mechanisms, the relative contribution of hormonal stimuli and nutrients in the setting of postprandial protein anabolism. Gains in body proteins occur essentially in relationship to nutrition, because at the time of feeding, we are replacing the essential amino acids that have been destroyed by oxidation during the fasting period. Even if we exercise, there is a transient increase in protein anabolism, but overall, we are still missing the essential amino acids that have been lost during the fasting period. Nutrition is a pivotal point of anabolism for the body proteins. It is extremely important to look into that because that's how we gain and don't lose proteins during everyday life. The initial studies were on whole-body protein metabolism, the integrated sum of metabolism of all the body proteins—structural proteins, enzymes, plasma proteins, and others. We started with the problems of type 1 diabetes where there is insulin deficiency. The insulin-deficient subject loses protein. Insulin plays an important role in maintaining muscle proteins and whole-body proteins. Initially, we looked at insulin alone and saw that it decreases the breakdown of proteins at the whole-body level. That's probably one of the mechanisms by which it helps to maintain body proteins.

On the other hand, insulin is secreted in large amounts during a meal. We wanted to discern in healthy people how insulin interrelates to protein turnover and protein anabolism during the most anabolic moment for proteins, which is during meal absorption when dietary amino acids are being utilized. We administered a meal containing amino acids, fat, and glucose to one group of subjects. We normally stimulate insulin secretion via ingestion of glucose. Another group of subjects received a meal in which the amino acids were missing; it was a fat/glucose meal. Another group received only a placebo meal. We had a control group essentially drinking water and the other two groups on two meals different for amino acid content. We found that when amino acids are missing in a meal, a net whole-body protein anabolism does not take place. On the other hand, you slow down catabolism to a point where some significant effect is realized by reducing the whole-body protein breakdown.

Another interesting thing is that during the absorption of a meal, with or without amino acids, there is an increase in the synthesis rate of albumin, an important protein that can be considered as temporary storage

for dietary amino acids. There is no real storage area for amino acids in the body. Every protein in the body has a function. It's not like fat, where there are triglycerides sitting there waiting to be used if they're not needed right away. It's the same for glucose; there is glycogen.

What happens when amino acids are consumed is that they need to be used somehow. Albumin is an interesting protein because its synthesis can be doubled by insulin during meal absorption. The uptake of amino acids from the gut that are coming through the liver can be significantly increased. Albumin is synthesized in the liver. There is a kind of conjoined effect. The insulin is coming from the pancreas, the amino acids are coming from the gut, and they increase albumin synthesis. When amino acids are lacking, the albumin synthesis increases as well, meaning that insulin has a stimulatory effect on that. The overall albumin becomes kind of a temporary storage area and it is then taken and broken down everywhere in the body, including in muscle tissue so it can slowly release the amino acids during the fasting period. That's a very important mechanism because otherwise, amino acids coming in through the gut would be immediately oxidized. You cannot increase the free amino acid pool too much; otherwise, they can become toxic. In fact, this is one of the problems seen in liver disease, for example, when you have liver failure. These were the findings in the paper we published in *Diabetes* in 1996. It was a matter of physiology and the mechanisms that allow our body to increase the protein content.

JB: That's a wonderful explanation and a good segue into my next question. As far as I remember, albumin as a serum plasma protein is very high in percentage composition of branch-chain amino acids, those of the essential amino acid family—leucine, isoleucine, and valine. You have done some very interesting pioneering work looking at the difference in protein anabolic effects of branch-chain versus mixed amino acids. Would you tell us how the connection between type of amino acids within the protein connects to the story you are emerging?

Type of Amino Acids in Protein

EV: We don't really do those kinds of studies, but it is important to know that branch-chain amino acids, especially leucine, can directly stimulate muscle protein synthesis. Actually, they can activate initiation of translation of newly-synthesized proteins. Leucine for sure, but possibly other essential amino acids, may have some direct effect on the stimulation of protein synthesis. Branch chain amino acids alone, although able to stimulate protein synthesis, will probably be unable to sustain a prolonged increase in protein synthesis because to sustain the higher synthetic rate, all the amino acids are required to make those proteins. If the amino acids are not present in sufficient amounts, the synthesis rate slows down because you need to wait for the tRNA to be charged with the specific amino acid that is waiting to be used in protein synthesis. In this case, what I and also others believe is that you can induce stimulation with a single amino acid, but once you have started the increased synthetic process, then all of the amino acids are needed. The essential amino acids are the culprits at this point because you can actually make the non-essential amino acids, whereas the essential amino acids are coming either from protein breakdown or from the diet. Either way, if you have a higher synthetic rate and you don't have a flux of amino acids coming from the diet, then you must break down more protein.

This is one of the potential mechanisms by which acute illness can actually induce severe muscle loss, especially in burn injuries. Bob Wolfe has done a lot of studies on that. In a burn injury, the patient has an extremely high breakdown rate of proteins in the muscle. At the same time, there is a very high synthesis rate of protein in the muscle, so there is a major increase in turnover of proteins in the muscle. The net protein balance in the muscle becomes negative because there is a huge outflow of amino acids from the

muscle into the liver. The liver needs these amino acids to build the acute-phase proteins such as fibrinogen and the complement, and all the acute-phase proteins necessary for a response to the acute injury.

If you look at the amino acid pattern in the acute-phase proteins on the muscle proteins, the amino acid pattern is different. You break down some muscle, and you need more muscle broken down to be able to make the same amount of acute-phase proteins because the amino acid pattern doesn't match. The theory is that if you don't provide large amounts of amino acids in the pattern required to make the acute-phase proteins, you're going to break down much more muscle and also oxidize the amino acids from the muscle that are not required to make the acute-phase proteins. That could sustain the muscle loss seen in this acute injury.

JB: When you, Bob Wolfe and your husband, Dr. Rasmussen, collaborated on the study that appeared in JAMA on basal muscle amino acid kinetics and protein synthesis in healthy young and older men,[17] were the results of your amino acid labeling study a surprise, or did you anticipate seeing those kind of results?

Basal Muscle Amino Acid Kinetics and Protein Synthesis in Healthy Young and Older Men

EV: Yes and no. That study followed a series of smaller studies we had done on the physiology of the response of muscle to nutrients in relation to age. Before the JAMA paper we did three studies in which we looked at whether muscle loss with aging was due to alteration in the anabolic response to increased nutrients. Nutrient intake is the most anabolic moment in our lives for muscle proteins and for proteins throughout our body. The major question was, does the muscle respond in older-age as it does at a younger age to the increased amino acid load during feeding? We published a paper in the Journal of Clinical Investigation in 1998 about infusing high doses of amino acids and looking at muscle protein synthesis breakdown in the baseline during the post-absorptive state.[18] We found that the response of muscle protein synthesis and anabolism to the amino acid infusion was normal in healthy older people.

There was some data from a French group who had seen in older people that the first-pass splanchnic uptake amino acids, those ingested and then taken up by the splanchnic tissues including the gut and liver, do not appear in the peripheral systemic circulation. They found that it (the uptake) was almost double in older people compared to younger people. The muscle, per se, is working with an IV infusion of amino acids. The question is, when you give amino acids orally, does their uptake decrease their appearance in the systemic circulation so that their actual availability for the muscle tissue is reduced?

We did another study in which we compared younger people and healthy older people. We looked at them in the baseline post-absorptive state and then during the ingestion of an amino acid mixture. We confirmed the data from the French researchers who found the increased splanchnic uptake at first-pass. We measured that using stabilized tracers (labeled) of amino acids. The interesting thing was that we didn't find any alteration in the response of the arterial concentration in the peripheral blood. Younger and older people had the same magnitude of increase in amino acid concentration in the arterial blood that was delivering the amino acids to the muscle tissue. Overall, the muscle response to that specific meal was identical in the young and in the older people. From this data, we concluded that there is an increase in the uptake from the splanchnic tissues, but there is also an increased release of unlabeled amino acids from the splanchnic tissues because otherwise, we couldn't have seen that same response in the blood amino acid concentrations. The conclusion is that in older people, there is both an increase in splanchnic

protein turnover, which is synthesis and breakdown of proteins within the splanchnic bed, but we don't know where. Is it the gut, the liver? We don't know. This does not impair their ability to respond to amino acids.

When we looked at the baseline data in the studies, we were surprised because we couldn't find any differences in the baseline muscle protein turnover rates—no difference in synthesis; no difference in breakdown; and no difference in the net protein balance in the baseline. That was quite surprising because other investigators, including Dr. Yarasheski from Washington University, St. Louis, Dr. Welle from Rochester University in NY and Dr. Nair at the Mayo Clinic, had reported in the past that there were significant differences in muscle protein synthesis rates between young and older people. Those differences were in the ballpark of a 30 percent reduction in the protein synthesis rates. We found no differences.

At that point, it became a puzzle because the other groups had measured protein synthesis rates using the same methodologies we used. There were no methodological differences that could have explained the differences in the findings. We thought that possibly the differences were due to the population studied, because the subjects were extremely healthy older people; they had no diseases, not even hypertension. They were very healthy. In a way, we had a potentially biased group of people because the subjects were so healthy. Another issue that may have been very important was the fact that in all the studies from the other investigators, there were no measures of muscle protein breakdown.

If you want to know exactly what the net balance of muscle proteins is, you must measure not only synthesis, but also breakdown. That gives you the net effect of what is going on in the muscle. When there is only synthesis, it might be that in the earlier studies they had a lower synthesis rate that was accompanied by a lower breakdown rate. That balance would have been normal so they were losing no muscle, but we didn't know that.

What was also done in those studies was to measure the synthesis rate of muscle proteins after the patients had been in a hospital bed for three days. That could have potentially created a difference. It is possible, but we don't know yet because we have no data from them. If you put an older person in the hospital for three days, their physical activity is significantly decreased. For example, if they had been gardening on a regular basis at home, obviously they couldn't do that in a hospital setting. They were sedentary for three days. Being sedentary, even for a very short time, can result in significant reduction in muscle protein turnover. There is no stimulation from even very mild exercise, such as walking or simple stair climbing in the home.

We decided we wanted to significantly increase the number of subjects because up until the study in the JAMA paper, our numbers were fairly small, usually token groups with less than 10 subjects per group. It is difficult to draw any conclusions from those kinds of numbers. We also decided not to have the subjects come in three days in advance because we wanted to study them under everyday life conditions. We told them to keep doing what they normally did in their homes, eat what they normally ate, and not change anything. We wanted them to come in the night before the study, sleep at the hospital, and we would do the study early in the morning. That way, we saw that there were no significant differences in protein synthesis and breakdown between the two age groups.

JB: It's likely that many of our listeners have had the opportunity to review your JAMA paper and have

probably seen your 2003 paper in the American Journal of Clinical Nutrition. They haven't yet seen the 2004 paper in the American Journal of Physiology Endocrinology Metabolism (due to be published this month), titled "Amino acid ingestion improving muscle protein synthesis in the young and elderly." Our clinicians might ask, what role does dietary protein play? Should they take a different approach to counseling their patients? There is the current concept of increasing dietary protein, and even popularization of the so-called Atkins Diet. How do you see your work translating into the management of patients who may be at risk to sarcopenia?

EV: Referring back to the previous question and the previous explanation of the JAMA paper, we saw no difference in the baseline muscle protein turnover rates in the younger or older people. That might have been biased because those people were extremely healthy. On the other hand, that presents the idea that age, per se, with no complications from disease, does not actually affect muscle protein turnover. Yet, those individuals we studied for the JAMA paper had decreased muscle mass because they had smaller muscles as compared to their younger counterparts. Although their basal protein turnover rates were normal, their muscle mass was smaller.

Our general conclusion from that paper was that we were very happy to see no fundamental impairment in the muscle protein synthesis rate in older men. Had it been there, it would have been a serious problem to counteract. We would have to resort to treatments that would actually increase baseline muscle protein turnover, and we don't even know how to do that—probably with androgens, but that's for males only and wouldn't apply to women. When we saw that the baseline was not a problem, we were reassured that it is probably more of a compounded problem of smaller deficits in the anabolic increases in muscle protein synthesis, possibly during the anabolic stimuli that do not adapt over the years and decades, eventually leading to a measurable muscle loss.

After having demonstrated that amino acids alone, given orally or by IV were working, we went on to give a more physiological type of chemically-defined meal. We gave amino acids with glucose. We did the same with a large amount of amino acids—40 grams, which is a very large amount. Then we added another 40 grams of glucose to that. We found the gains in muscle protein synthesis that we would have expected due to the presence of amino acids in this meal, were totally lost. Protein synthesis in older people did not increase at all with this kind of meal, whereas in the younger controls, we had a huge increase in protein synthesis, double what we would have seen with amino acids alone. There was a completely different effect of this mixed meal on muscle proteins in young and older people. In both groups, we also saw a decrease in muscle protein breakdown. Overall, there was an increase in net muscle protein balance, but this increase was blunted in the older people because obviously, there was the component that the synthesis rate was missing. The total increase in net balance was entirely due to the decrease in muscle protein breakdown.

You need to remember that the net balance of muscle proteins is due to the synthesis rate minus the breakdown rate. If the breakdown rate decreases significantly, the net balance can go up, but it is due to a total suppression, or decrease, in the muscle protein turnover rate. We concluded from that study that in young people when you add carbohydrate (an insulin stimulus which comes with additional carbohydrate in the diet), you see a significant increase of muscle protein anabolism and synthesis above and beyond what you see with amino acids alone.

In elderly people, there is no net benefit with additional carbohydrate, but on the other hand, this addition

can be detrimental for muscle remodeling because there is a decrease in the total turnover rate of muscle proteins as the breakdown decreases and the synthesis doesn't increase. There is a reduction in the ability of muscle to exchange the older proteins. That could be one of the reasons why, over time, we see a slow, creeping decrease in muscle mass that could be due to the collection of smaller increases in net balance with mixed meals.

I cannot say that the Atkins Diet or the carbohydrate-free diet would work in older people, because there's no long-term data. We are still in the hypothesis stage on that issue. We don't know what happens over the long term when you change the diet or add nutritional supplements that do not include carbohydrate and do not stimulate insulin secretion. We don't know what's going to happen with that over the long term. We need to do the studies. I turned in a grant proposal just recently so we can see if, with six months of nutritional supplementation with amino acids (essential amino acids in particular, which apparently are the ones most active anabolically for muscle in older people), we can make a difference and regrow muscle mass.

Loss of response to the mixture of amino acids and carbohydrate on the other hand indicates a need to focus on the effects of insulin. In fact, when we were writing the paper about the combination of amino acids and glucose, we wondered what would cause this. Obviously, we could not dissect it from the data because we gave one group amino acids and one group amino acids with glucose. We thought that if the glucose we were giving was the culprit, how did it work? We didn't know that because, if anything, it would have added some calories, some energy, to the meal. Protein synthesis is an energy-consuming process so it should have had a positive effect.

The other major variable that changed during the carbohydrate meal with amino acids was insulin. I currently have a grant funded by the National Institutes of Health, to look at the potential for some kind of insulin resistance of muscle proteins which results with age. That might explain the impairments in muscle protein turnover during mixed feeding. I have some preliminary data. We are writing the paper right now, and hope to publish it over the next several months. There is definitely impairment in the response of muscle protein synthesis, once you expose the muscle to postprandial concentrations of insulin in older people. In younger people, there is an increase in muscle protein synthesis at these insulin concentrations, whereas in older people, we see no change in muscle protein synthesis and a decrease in breakdown. That's one of the potential culprits (I wouldn't say it's the only one), that might explain muscle loss with age.

JB: That is fascinating work and obviously we are going to be patiently waiting for continued results from your experiments and research. It seems to me that there is some kind of an age-related change that may be related to different signaling molecules focused on muscle physiology in older-age individuals versus younger individuals, and that insulin somehow plays a role. Perhaps diets should change as we grow older. That's where your research may be taking us.

Aging and Diet Changes

EV: That's possible. I can't give any nutritional advice right now, because we don't have the long-term data yet. We still need to do the studies, and it can be dangerous if you go on a whole-protein diet, carbohydrate-free diet. Perhaps split diets would be feasible where you eat carbohydrate separately from the meat (protein) . Again, this needs to be confirmed by long-term studies.

On the other hand, we do know that if you want to increase muscle mass in older people, they must exercise. That's the other part of the equation that needs to be stressed. Nutrition is one factor, but exercise is the other. Many people lose muscle just because they cut down on physical activity. Any kind of physical activity is important. Weight-lifting has been repeatedly shown by Drs. Evans, Yarasheski, and Welleto be extremely helpful in stimulating muscle protein synthesis in adults, and also muscle growth over the long term. I understand that weight lifting is not very appealing, so we need to consider aerobic exercise to help the regrowth of muscle—walking, swimming, or cycling. We are only beginning to examine this.

There are definitely some dietary components that can be helpful. I do not subscribe to the Atkins Diet from an endocrinologist's or physician's perspective because it makes people ketotic and it may actually be dangerous in certain circumstances. A balanced diet is always a good idea to start with, and we can look into whether it needs to be supplemented. It is also important to emphasize, especially in the oldest old—people over 70-75—that they should not strive to lose weight. Weight loss in the elderly is strongly correlated with increased mortality.

There are lots of questions still to be answered. The older we get, the higher the risk to become un nourished. We need energy and carbohydrate and fat can help with that. In prescribing diets for older people, one needs to look at body composition, and whether weight has been gained, lost, or is stable. If a patient is losing weight, packing calories is needed. Any cheesecake is good. My patients have always been very happy when I prescribe cheesecake because they feel reborn after years and years of fat-free diets for their heart disease. All the potential problems need to be factored into the equation, including the fact that weight loss is associated with higher mortality in older people.

JB: Thank you, Dr. Volpi. You've done an eloquent job of covering a very sophisticated topic. It seems so obvious that we should know more about carbohydrate, protein, and fat, and their relationship to endocrine function in aging individuals. As you've outlined it, it sounds like a lot of the important, first-level studies are presently being conducted by you and your colleagues. It's interesting what we've taken for granted that we really didn't know.

EV: I agree. It's a wide-open field right now.

JB: Thanks again for being with us today. We look forward to visiting with you at the 11th International Symposium on Functional Medicine.

Dr. Volpi's presentation raised many questions for us and also provided some insight relating to the diet controversy about protein, carbohydrate, and fat. One of the takeaways is that the total environment of the patient determines the way they respond to dietary variables, and that these are messenger molecules, dietary signals, that create the outcome. The outcome is different in different environments. If the patient is in an inflammatory state, is insulin resistant or diabetic, the response to meals may be very different from individuals who do not carry those considerations.

Increased Dietary Protein and Blood Glucose Response in Type 2 Diabetics

This information helps us to better understand results that were published in the *American Journal of Clinical Nutrition*, titled "An increase in dietary protein improves the blood glucose response in persons

with type 2 diabetes.”^[19] In this paper, the investigators used a high-protein diet that was shown to lower blood glucose postprandially in patients with type 2 diabetes, and improve overall glucose control. The authors state that longer-term studies are necessary to determine the total magnitude of response, possible adverse effects, and the long-term acceptability of the diet.

A New Look at Dietary Protein in Diabetes

The editorial that followed this paper also confirms that there seem to be some interesting results of increasing dietary protein as a consequence of trying to monitor, measure, or modulate type 2 diabetes, insulin resistance, and hyperinsulinemia.^[20] Could the state a person is in, with increased inflammatory mediators associated with type 2 diabetes/insulin resistance, affect which protein at an enhanced level has a beneficial effect? As Dr. Volpi said, perhaps uptake, muscle resynthesis, reformation, and remodeling are modified to a greater extent by increased dietary protein in the person with insulin resistance/hyperinsulinemia than in the person in whom inflammatory mediators are absent.

That would tend to be consistent with the Volpi et al. paper that appeared in *JAMA* in 2001 that looked at basal muscle amino acid kinetics and protein synthesis in young and older men, suggesting that the postprandial environment, inflammatory mediators, and the messenger molecules that differ at young age versus older age, might have something to do with muscle protein synthesis, recalling that, in general, older men do not have the same muscle mass as younger men. It may be the total environment. We need to personalize the diet to the environment.

As I mentioned earlier about animal versus vegetable protein, the Volpi et al. work indicates that essential amino-acid rich proteins are very desirable for establishing the right postprandial environment for muscle formation integrity. Essential amino acid-rich diets can be delivered with vegetable proteins fortified with specific essential amino acids, as well as with animal protein diets. Although, with soy protein fortified with sulfur amino acids, i.e., methionine, one might worry about soy protein because it contains phytoestrogens, isoflavones that can modify estrogen receptivity.

In a recent paper in the *American Journal of Clinical Nutrition*, titled “Dietary phytoestrogen and breast cancer risk,”^[21] the investigators looked at the dietary intake in 15,550 women, age 49 to 70, and constituted a cohort dietary intake looking at isoflavones and breast cancer incidence. They found that a high intake of isoflavones or lignans was not significantly related to breast cancer risk, again dispelling some of the unfounded risk concerns that certain people have about isoflavones.

Flaxseed versus Soy Supplementation in Estrogen Metabolism

Flaxseed lignans also play a role in estrogen metabolism in postmenopausal women, and may have a more salutary effect than soy itself.^[22] Lignans from flax and soy isoflavones appear to be safe in moderate dietary intake. Soy protein has a good arginine/lysine ratio balance to achieve the effects we were talking about earlier.

The whole story of protein, carbohydrate, and fat is more complex than it appears on the surface. We should be looking at personalizing individual dietary needs because diet signals create an outcome that leads to muscle preservation.

Lignans and Estrogen Modulation

When we talk about increasing vegetable protein, most individuals recommend soy-based foods, beans, or legumes, which contain various types of proteins. These are food products that also contain other phytonutrients, other plant-derived chemicals—a whole series of agents in different plant foods that modulate gene expression and function. One of these groups is lignans. Lignans are interesting because they are metabolized by enteric flora in the secondary compounds that have hormone-modulating effects. These secondary compounds from lignans can modulate estrogen metabolism.

In a recent paper in the *American Journal of Clinical Nutrition*, investigators found that the urinary concentrations of the 2-hydroxyestrogens were increased significantly when postmenopausal women consumed a diet higher in flax lignans. The 16-hydroxyestrogens, the more mitogenic estrogens, were lower. Therefore the 2-16 hydroxyestrogen ratio was favorably improved with improved 2-hydroxylation and lowered 16-hydroxylation. This is as a result of consumption of lignan-rich diets, such as those enriched in flax seeds.

These plant-based foods may have a whole series of salutary benefits on cell signaling and hormonal messaging beyond that of the protein/carbohydrate ratio or the amino acid ratio. We need to keep a broad perspective, open to all the variables in the diet that influence the complex processes in endocrinology. It is in the unrefined or whole foods diet where one is getting the complex signaling molecules in their natural forms, versus a highly purified diet (giving amino acids and glucose), where one is getting a whole different orchestration of function.

As we simplify the diet and refine it more and more, it tends to address only one player in the orchestra, which creates a different symphonic sound than when we have all the players working together. Diets containing natural foods that are minimally processed, rich in color, with proper levels of vegetable protein and complex unrefined carbohydrates rich in phytochemicals, appear to be what would be recommended from much of the research.

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