

Metabolic syndrome: Evaluation of pathological and therapeutic outcomes

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In April 2002, a conference on “The Metabolic Syndrome of Cardiovascular Risk and Insulin Resistance” was held in Washington, DC. This is the second of 2 papers summarizing the discussion at that conference. The first paper addressed the definition of metabolic syndrome, the components of the syndrome, and the underlying pathophysiology. The objective of this paper is to address 3 key areas in the management of metabolic syndrome: (1) the etiology and detection of atherosclerosis, (2) current and future therapies for metabolic syndrome, and (3) health system issues, including regulatory considerations, clinical trial design, and cost effectiveness.

Atherosclerosis

Atherosclerosis is the primary pathological consequence of metabolic syndrome. Here, atherosclerosis will be characterized according to proposed etiologic mechanisms of several key risk factors, including lipoproteins, hypertension, diabetes mellitus, inflammation, and endothelial dysfunction.

Lipoproteins

Lipoproteins associated with major atherosclerotic risk (or protection) include low-density lipoprotein (LDL) cholesterol (LDL-C), high-density lipoprotein (HDL) cholesterol (HDL-C), and lipoprotein (a). Additional risk may accrue when levels of triglycerides and intermediate-density lipoproteins are high and when LDL particles are small and dense. The mechanisms by which lipoproteins influence atherogenesis are understood best for LDL, followed by HDL.

Measurements of apolipoprotein B in human arterial intima have led to the recognition of a distinctive pathophysiologic feature of this tissue: the estimated LDL concentration in arterial intima is approximately 10 times higher than that in other connective tissues in the body. The LDL concentration in human aortic intima is approximately equal to the LDL concentration in plasma.¹ In contrast, the LDL concentration in most connective tissues is assumed to be equal to the LDL concentration in lymph, which is one tenth of the LDL concentration in plasma.²

To understand the remarkably high levels of LDL in arterial intima, we must recognize that in other connective tissues, lymph vessels function as sumps to drain away excess macromolecular species. The arterial intima lacks lymph vessels, which allows LDL to accumulate to a very high concentration.

The pathogenic role of LDL is exacerbated by the fact that the intima is bounded by 2 permeability barriers—the arterial endothelium and the arterial tunica media. It is well known that endothelial tight junctions form a permeability barrier, but in the tunica media a tight proteoglycan meshwork also forms a permeability barrier that sterically excludes particles the size of LDL.³ The arterial intima is a loose structure without the same tight meshwork as the media. Because of the permeability barriers, LDL particles enter and exit the arterial intima at slow rates, and the half-life of LDL particles in the intima may be on the order of weeks or months. The long half-life and high concentration of LDL in the arterial intima afford ample opportunity for degradative, oxidative, and denaturing processes to disrupt the integrity of soluble LDL and render it aggregated, insoluble, and prone to cellular uptake.¹ The macrophage system that works well for tissue repair and cleanup in other connective tissues may be overwhelmed when presented with a high load of aggregated, oxidized, and degraded LDL.⁴

In addition to the pathophysiologic understanding described above, the role of LDL in promoting atherosclerotic events is supported by consistent epidemiologic data and clinical trial results. Low-density lipoprotein cholesterol has been used as a surrogate marker by clinicians and regulatory agencies in much the same way that blood glucose and blood pressure have been used. The issue of surrogate markers will be discussed in detail later.

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The protective effect of HDL against atherosclerosis has been ascribed to various functions: reverse cholesterol transport, antioxidant effects, anti-inflammatory effects, and others. Here, the focus will be on reverse cholesterol transport, which is the best demonstrated function of HDL.

Although a complete picture of lesion evolution in human atherogenesis is still not available, unesterified cholesterol likely plays a role in the pathogenesis of the lipid-rich, hypocellular core of atherogenic lesions. The transition from insignificant fatty streak lesion to clinically important fibrous plaque is marked by a distinctive shift from esterified to unesterified cholesterol in the deeper intimal layers. This is accompanied by the development of cholesterol crystals and the depletion of surrounding cells.⁵ Although both unesterified cholesterol and lipid peroxides may contribute to cell death in the atherosclerotic lipid-rich core, the circumstances of lesion progression favor unesterified cholesterol as the key responsible factor.

The chemical composition of cell membranes is highly regulated, and one consequence of excess cholesterol accumulation in the membrane is cell death. Table I shows some of the pathways provided by evolution for the maintenance of optimal cholesterol concentrations in the cell membrane.⁶⁻¹⁰ Of the 12 mechanisms listed in Table I, some are irrelevant to the arterial intima, and others may have limited efficacy when diffusion distances for tissue lipoproteins are large. When arterial cells reside near extracellular deposits of unesterified cholesterol, the aqueous-phase, physicochemical transfer of cholesterol from the deposits to the cells might override all other mechanisms by which cells defend their membrane cholesterol concentration. Nevertheless, HDL may help by removing unesterified cholesterol from intimal tissue (mechanisms 7-10 in Table I).

From an epidemiologic viewpoint, compared with LDL concentrations, HDL concentrations are as predictive or slightly more predictive of atherosclerotic events.¹¹ Recent cardiovascular outcome studies support a role for increasing HDL in the amelioration of atherosclerotic risk.^{12,13}

Hypertension

Elevated blood pressure may contribute to atherogenesis by causing thickening of both the intimal and the medial layers of the arterial wall. Smooth muscle cells in these layers of the arterial wall respond to a rise in pressure and wall tension by exhibiting a growth response of hypertrophy or hyperplasia or by increasing their production of collagen and elastin. Whereas the molecular mechanisms continue to be explored, these physiologic responses appear to be intrinsic to smooth muscle tissue. As with smooth muscle in the walls of other hollow organs, the tension-bearing capacity and

Table I. Mechanisms by which cell membrane cholesterol-phospholipid composition is defended*

1. Removal of LDL from tissue spaces by lymph vessels
2. Down-regulation of cholesterol synthesis
3. Down-regulation of LDL receptors
4. Up-regulation of acyl-coenzyme A cholesterol acyltransferase
5. Decreased neutral cholesterol esterase activity
6. Stimulation of phosphatidylcholine and sphingomyelin synthesis
7. ABC-A1 transporter-mediated cholesterol transfer to apolipoprotein A-I
8. Aqueous phase transfer to HDL and other acceptors
9. Cholesterol esterification in HDL via lecithin-cholesterol acyltransferase
10. Cholesterol ester transfer from HDL to VLDL and LDL
11. Up-regulation of apolipoprotein E in peripheral cells
12. Activity of cholesterol 27-hydroxylase

ABC-A1, Adenosine triphosphate binding cassette A1.

*See text for references.

the wall thickness of the artery increase to match the imposed force.¹⁴

A reasonable hypothesis is that hypertension promotes atherogenesis mainly via thickening of the arterial intima. Pathological studies of human atherosclerosis have shown that lesion core development occurs only in arteries where a threshold of intimal wall thickness is exceeded.¹⁵ Thus, the mechanism of atherogenesis may involve excessive diffusion distances for intimal LDL and HDL in the arterial wall that has been thickened by hypertension.

Diabetes mellitus

Hyperglycemia is a primary determinant of the *microvascular* complications of diabetes, probably acting via the nonenzymatic formation of advanced glycosylation end products (AGEs) in target tissues. This theory is reinforced by the discovery that macrophages bear specific receptors for AGEs, which presumably represents an attempt by the body to limit the damaging impact of AGEs.¹⁶ Oxygen radical formation has been demonstrated to result from the reaction of molecular oxygen with AGEs.¹⁷

Atherosclerosis, the *macrovascular* complication of diabetes, may also be promoted by hyperglycemia via AGEs in the arterial wall, but the pathophysiologic and epidemiologic evidence is not nearly as well defined as it is for microvascular complications. In the United Kingdom Prospective Diabetes Study (UKPDS), rates of myocardial infarction showed a highly significant correlation with levels of hemoglobin A_{1c}.¹⁸ However, the slope of this effect was much less than it was for microvascular complications. Compared with untreated patients, patients randomly assigned to receive insulin or sulfonylurea to lower hemoglobin A_{1c} had only marginally reduced myocardial infarction rates (16% decrease, $P = .052$) and actually had a trend toward increased stroke rates (11% increase, $P = .15$).¹⁹

Hyperinsulinemia has been suggested as another determinant of accelerated atherosclerosis in diabetes, but the evidence is mixed.²⁰⁻²² The factors that augment atherogenesis in patients with diabetes mellitus thus remain to be determined; they may represent a complex interplay between glucose, insulin, dyslipidemia, insulin resistance, increased inflammatory cytokines, and endothelial dysfunction.

Inflammation

Over the past 2 decades, the idea that inflammation plays a significant role in the atherosclerotic lesion has gained acceptance.²³ More recently, a new hypothesis has been advanced, which states that atherogenesis may be influenced by the general state of inflammation in the body. This intriguing hypothesis has arisen largely through the recognition that the serum level of C-reactive protein (CRP), an acute-phase reactant, is a remarkably strong predictor of atherosclerotic events.²⁴ The epidemiologic relationship of CRP to atherosclerotic risk is now well established, but the exact causative link is less clear.²⁵ A direct role for CRP is unlikely. Instead, CRP is considered to be a marker for a systemic inflammatory response involving interleukin (IL)-6, IL-1, tumor necrosis factor α , and/or other cytokines and mediators. Some of these cytokines are known to be produced by adipocytes and are elevated in obese individuals.²⁶ Other etiologic possibilities include the following: (1) individuals differ in their genetic predisposition for a vigorous inflammatory response that promotes atherogenesis (the differences being reflected in CRP levels), and (2) inflammation elsewhere in the body (eg, chronic chlamydial infection of the lungs) may activate systemic inflammatory mediators, which raise CRP levels and also accelerate atherogenesis.

Other possible explanations for the strong association between CRP and atherosclerotic events must also be considered. Cytokines produced in atherosclerotic arteries might stimulate the liver to make CRP. Thus, atherosclerosis could stimulate the systemic inflammatory response (or at least CRP) rather than vice versa. In addition, CRP levels are elevated in people with obesity, low fitness levels, and/or metabolic syndrome.²⁷⁻²⁹ Moderate alcohol intake is associated with lower CRP levels compared to abstinence.³⁰ These variables may strongly influence atherothrombotic events and explain part of the statistical association with CRP. To contemplate the use of CRP as a treatment target, a more complete understanding of the systemic inflammatory response and its relationship to atherosclerosis will be needed.²⁵

Endothelial dysfunction

An understanding of endothelial function may help bridge the gap between clinical risk factors and the biochemical events that drive atherogenesis. The vaso-

dilatory response to acetylcholine or to increased blood flow is mediated largely by endothelial production of nitric oxide, which stimulates cyclic guanosine monophosphate-mediated relaxation of vascular smooth muscle. The arterial endothelium, in addition to governing smooth muscle tone and providing a permeability barrier for blood components, acts as a transducer for the inflammatory response that characterizes atherosclerosis.³¹ Abnormalities of endothelial-mediated vasodilation may correlate with overall endothelial dysfunction associated with atherogenesis. In support of this hypothesis, multiple atherogenic risk factors, including hypercholesterolemia, hypertension, smoking, ingestion of a high-fat meal, diabetes, and aging, are associated with impaired endothelial-mediated vasodilation.³² However, estrogen and antioxidants—2 factors cited to improve endothelial-mediated vasodilation—have failed to improve cardiovascular outcomes in randomized clinical trials.^{33,34}

An important question is whether endothelial dysfunction is predictive of cardiovascular events. The answer is probably yes, as several recent studies have shown a positive relation between endothelial dysfunction and subsequent cardiovascular events.³⁵⁻³⁷ However, the total number of patients studied thus far is limited, and one study used the simplest measure, flow-mediated dilation of the brachial artery.³⁶

Endothelial dysfunction in diabetes mellitus has been widely documented over the last 10 years.³⁸ In healthy, normal individuals, ingestion of a single high-fat meal was shown to induce transient endothelial dysfunction²⁹ and elevation of circulating free fatty acids by Intralipid-heparin infusion also caused endothelial dysfunction.³⁹ The relation between insulin resistance or fasting hypertriglyceridemia and endothelial dysfunction is controversial. Studies in young adults, who normally have robust dilatory responses, have shown impaired endothelial function in association with insulin resistance or fasting hypertriglyceridemia.^{40,41} However, these results have not been reproduced in studies of older individuals.⁴²

Soluble adhesion molecules comprise a set of biomarkers potentially related to endothelial dysfunction and to atherosclerotic risk. These molecules are soluble forms of the endothelial surface molecules that bind to ligands on circulating leukocytes and mediate the transmigration of inflammatory cells into the vessel wall. In an analysis of serum samples from almost 2000 men in the 16-year prospective British Regional Heart Study, soluble adhesion molecules were found to be predictive of coronary heart disease. The strongest effect was an odds ratio of 1.68 predicting coronary heart disease in the top compared with the bottom decile of levels of intercellular adhesion molecule-1. However, none of the associations between soluble adhesion molecules and coronary disease were independent of traditional coro-

nary factors and socioeconomic status. Based on their own and previous studies, the investigators believed that assays for soluble adhesion molecules add little to the current methods for predicting coronary risk.⁴³

In summary, endothelial function at the molecular level incorporates aspects of inflammation, thrombosis, and vasodilation.³¹ The response of endothelial function to treatment does not always predict cardiovascular benefit, as illustrated by estrogen and antioxidant administration. Nevertheless, clinical studies of endothelial function may help define mechanisms whereby new treatment approaches to metabolic syndrome will reduce atherosclerotic risk.

Noninvasive imaging technologies for atherosclerosis assessment

As the preceding discussion emphasizes, atherosclerosis and vascular dysfunction are central to the clinical consequences of metabolic syndrome. Clinical cardiovascular events are usually due to arterial occlusion related to preexisting atherosclerotic lesions. One might therefore hypothesize that clinical detection and quantitation of atherosclerosis could improve the prediction of cardiovascular events. In vivo imaging of arterial lesions remains suboptimal, but lesions imaged by angiography and ultrasound have significantly correlated with risk as have calcific deposits imaged by computed tomography (CT) scanning.⁴⁴ Here, we review some of the data relevant to potential use of carotid ultrasound and CT coronary calcium scoring in clinical studies of atherosclerotic risk in metabolic syndrome.

B-mode carotid ultrasound already has an established place in epidemiologic studies and clinical trials in atherosclerosis.^{45,46} Reflection of sound waves from tissues of differing densities allows one to define the combined intima-media thickness of the carotid arterial wall. The tissue characteristics—that is, fatty or fibrous—and therefore the histologic nature of arterial lesions usually cannot be defined by use of B-mode carotid ultrasound. This technique also has limited precision and reproducibility, requiring a skilled operator for accurate results.

An association between metabolic syndrome and increased carotid intima-media thickness has been shown with B-mode ultrasound, which may indicate increased atherosclerotic risk. Clinical trials, using trained operators and controlled conditions, have revealed that progression of intima-media thickness measured by B-mode carotid ultrasound predicts cardiovascular events.⁴⁷ Interventions that slowed progression or caused regression of intima-media thickness also decreased cardiovascular events, and the effect on progression-regression correlated with reduction of events.

Another noninvasive imaging technique focuses on detection of coronary calcium. In the 1970s, positive correlations were established between coronary calcium

visualized by fluoroscopy during coronary angiography and subsequent clinical events.⁴⁸ Electron beam (ultrafast) CT scanning has much greater sensitivity than fluoroscopy or conventional CT, allowing the detection and quantitation of coronary calcium in almost all patients with clinically significant coronary atherosclerosis. Coronary calcium scoring correlates well with the total burden of coronary atherosclerosis.⁴⁹ In addition, coronary calcium scoring predicts clinical events, as well as an estimate based on the number of stenotic coronary arteries determined by angiography⁵⁰ and almost as well as the Framingham risk calculation.⁵¹ A meta-analysis of 5 studies yielded a summary risk ratio of 4.2 for the combined outcome of death or nonfatal myocardial infarction in patients above or below the median coronary calcium score. However, only 2 of the studies in this meta-analysis adjusted results for patient age.⁵² Whether year-to-year changes in calcium score may be responsive to treatment and/or predictive of events is unclear at this time.⁵³ Recent innovations in gating conventional or helical CT scanners have led to accuracy in coronary calcium quantitation that rivals that of electron beam CT scanners.⁵⁴

As a potential clinical tool, coronary calcium quantitation by enhanced CT scanning has high diagnostic sensitivity but uncertain utility.⁵⁵ As a research tool, enhanced CT scanning may be used to provide entry criteria for studies, to stratify for treatment assignment, to provide intermediate or biomarker end points, or to determine a quantitative phenotype for risk factor or genetic studies. Coronary calcium scores are relatively reproducible and require less operator training than ultrasound techniques, and the coronary arterial bed is more relevant to cardiac events than the carotid arteries or the aorta.

The Multi-Ethnic Study of Atherosclerosis, a National Institutes of Health-funded, multicenter trial,⁵⁶ is studying the progression of clinical and subclinical atherosclerosis examined by a variety of techniques, including ankle/brachial index, carotid B-mode ultrasound, CT scanning of coronary arteries, and magnetic resonance imaging of the heart, as well as endothelial function (brachial artery vasodilation). In other large studies, coronary calcium scores are being used as quantitative phenotypes for genomic investigation.⁵⁵ The use of these and other noninvasive imaging techniques requires further investigation and validation in predicting atherosclerotic risk in metabolic syndrome.

Therapeutic approaches to metabolic syndrome

Current and potential therapies for metabolic syndrome target obesity, insulin resistance, dyslipidemia, hypertension, inflammation, prothrombotic state, and the environmental conditions that promote obesity.

Given the diverse pathological mechanisms leading to the syndrome (see previous paper), a broad approach targeting multiple etiologic factors is reasonable.

Various components of metabolic syndrome may be addressed by dietary modification, exercise therapy, and pharmacologic agents. The pharmacologic agents most often proposed are those that target insulin resistance, such as metformin and thiazolidinediones (TZDs). There is little support for the use of insulin secretagogues or insulin therapy in nondiabetic patients with metabolic syndrome. Other pharmacologic agents such as statins, fibrates, niacin, angiotensin-converting enzyme (ACE) inhibitors, angiotensin II receptor blockers, and aspirin may be included in the treatment of metabolic syndrome to prevent or slow the progression of cardiovascular disease. A comprehensive management plan might include the following components: monthly nutrition and exercise classes, educational material, electronic records, phone follow-up, and paper and online log-books and forms for diet and exercise. This type of broad-based approach requires significant medical system infrastructure with associated cost.

Lifestyle modification

Lifestyle modification is the cornerstone of treatment for metabolic syndrome. Lifestyle modification, including both diet and exercise therapy, is aimed at achieving a net negative energy balance by decreasing energy intake and increasing energy expenditure, with the goal of decreasing both body weight and insulin resistance. Although no particular diet plan or dietary composition has been studied specifically in relation to metabolic syndrome, the current dietary recommendations include a balanced low-energy diet containing fruits, vegetables, whole grains, fish, and lean meats while minimizing fats, salt, simple sugars, and highly processed foods.

Similar to diet therapy, the use of exercise therapy in patients with metabolic syndrome without diabetes has not been extensively studied. A reasonable goal for most individuals is moderate exercise such as walking for 30 minutes per day at least 5 days per week.⁵⁷ Improvement in lipoprotein subfractions related to both the amount and the intensity of exercise was described recently by Kraus et al,⁵⁸ although not all subjects in the study had metabolic syndrome.

Both the Finnish Diabetes Prevention Study and the United States Diabetes Prevention Program (DPP) recently showed that diet and exercise had a significant effect on reducing the progression from impaired glucose tolerance to type 2 diabetes.^{59,60} The DPP also included a group assigned to metformin. The primary goal in the DPP was to prevent or delay the onset of type 2 diabetes; the secondary goals were to decrease cardiovascular disease risk factors, atherosclerosis, and cardiovascular events. These goals are similar to those of metabolic syndrome treatment. The DPP enrolled 3234

normotensive, mostly obese subjects including 68% women, 68% with family history of diabetes, and 50% minority. The average fasting plasma glucose of enrollees was 107 mg/dL, and average hemoglobin A_{1c} was 5.9%. Subjects were randomly assigned to intensive lifestyle modification (low-energy, low-fat diet to induce weight loss with 150 minutes of walking per week), 850 mg of metformin twice a day, 400 mg of troglitazone daily, or placebo. The troglitazone arm was dropped when rare cases of hepatic failure were reported from general use of the drug. The remaining treatment groups showed significant differences in the rate of development of new type 2 diabetes development, with 11% per year in the placebo group, 7.8% per year in the metformin group (a 31% decrease), and 4.8% per year in the intensive lifestyle group (a 58% decrease). In addition, significantly more patients were converted from impaired glucose tolerance to normal glucose tolerance by intensive lifestyle modification as compared to metformin, which in turn was significantly more effective than placebo. These improvements support the use of lifestyle recommendations for individuals with metabolic syndrome. However, the rates of cardiovascular events in the DPP were too low to be statistically significant, demonstrating the difficulty in obtaining hard cardiovascular end points in this population.

The currently active DPP Outcome Study has enrolled 2500 individuals with impaired glucose tolerance and 750 individuals with type 2 diabetes who were part of the original DPP cohort. All subjects in this follow-up study have received modified lifestyle intervention, and subjects on metformin in the original DPP study remain on metformin. Over 5 years, the DPP Outcome Study will examine (1) further prevention and/or delay of type 2 diabetes, (2) a composite microvascular end point, and (3) macrovascular end points, including carotid intima-media thickness, ankle-brachial index, and Framingham score.

Metformin and the UKPDS

Metformin is a biguanide that decreases hepatic glucose output and improves insulin sensitivity.^{61,62} Controlled studies of type 2 diabetes have shown that metformin reduces fasting plasma glucose by 22% to 26% and reduces hemoglobin A_{1c} by 1.2% to 1.7%, with minimal incidence of hypoglycemia.^{63,64} In short-term studies, metformin sometimes causes a small reduction in weight, an effect likely related to decreased food intake. Over a period of years, weight gain for diabetic patients on metformin was slower than that for patients on sulfonylureas or insulin. As discussed above, the DPP has demonstrated that metformin is effective at slowing metabolic deterioration in individuals with impaired glucose tolerance. Thus, metformin is a candidate drug for treatment of the glycemic components of metabolic syndrome.

In the UKPDS, 1704 overweight patients with newly diagnosed diabetes and no hyperglycemic symptoms were randomized to treatment with diet alone, metformin, sulfonylurea, or insulin.⁶⁴ Compared with patients taking placebo, patients receiving metformin (n = 342) over a median 10.7-year period had significant reductions in any diabetes-related end point, diabetes-related death, total mortality, and myocardial infarction, whereas no significant reductions were found in sulfonylurea- or insulin-treated patients for these end points. Myocardial infarction was reduced by 39% in patients taking metformin. Of note, the UKPDS data were used to create the UKPDS Risk Engine, which calculates the probability of cardiovascular events in diabetic patients and should be useful in planning clinical trials in diabetes.⁶⁵

PPAR- α and PPAR- γ agonists

Peroxisome proliferator-activated receptors (PPARs) are a family of nuclear transcription factors first cloned as mediators of the effects of drugs that cause proliferation of peroxisomes in rodents, although peroxisome function is only a minor component of the overall transcriptional activity of these agents.⁶⁶ Formation of a heterodimer between a PPAR and the receptor for 9-*cis*-retinoic acid (retinoid X receptor) is required for binding to specific response elements in the promoter region of target genes. Three PPAR isotypes have been described: α , β (also called δ or NUC1), and γ . Peroxisome proliferator-activated receptor α and PPAR- γ are the principal receptors mediating the effects of currently used drugs affecting lipid and glucose metabolism.

A summary of the characteristics and clinical correlates of PPAR- α is shown in Table II. The principal effects occur in the liver, where PPAR- α regulates fatty acid catabolism and apolipoprotein production.^{66,67} The clinically important hypotriglyceridemic effect of PPAR- α agonists appears to be due primarily to decreased transcription of the apolipoprotein C3 (apoC3) gene. Apolipoprotein C3 carried on triglyceride-rich very low-density lipoproteins (VLDL) inhibits the activity of lipoprotein lipase, the enzyme responsible for removing most of the triglyceride from VLDL. Down-regulation of apoC3 production thus activates lipase-VLDL interaction and lowers plasma triglycerides. Peroxisome proliferator-activated receptor α agonists also increase transcription of apolipoproteins A-I and A-II, thereby increasing HDL-C. Anti-inflammatory effects of PPAR- α agonists have been suggested from experiments with vascular cells in culture, but PPAR- α knockout mice exhibited more, not less, atherosclerosis when crossed with apolipoprotein E-deficient mice.⁶⁸

Early clinical cardiovascular outcome trials of PPAR- α agonists had mixed results, but more recent trials focusing on patients with low HDL-C or diabetes have had favorable results. In the World Health Organization

Table II. Characteristics and clinical correlates of PPAR- α

Sites of expression	Liver Brown adipose tissue Kidney Heart Skeletal muscle
Natural ligands	Unsaturated fatty acids (eg, linoleic) Branched, conjugated fatty acids Eicosanoids (8S-HETE, leukotriene B4)
Pharmacologic agonists	Clofibrate Gemfibrozil Fenofibrate
Target genes	Fatty acid uptake, binding, and oxidation Apolipoproteins
Clinical effects	↓ Triglycerides 40%-50% ↑ HDL-C 10%-15% ↔, ↓ LDL-C Anti-inflammatory

trial reported in 1978, clofibrate was not only associated with a trend toward decreased myocardial infarction, but also with a statistically significant 28% increase in total mortality related primarily to noncardiovascular mortality.⁶⁹ In the Helsinki Heart Study, gemfibrozil was associated with a 34% reduction in major coronary end points,⁷⁰ but nonsignificant trends toward increased noncardiovascular and total mortality were noted in follow-up.⁷¹ These primary prevention studies enrolled patients with hypercholesterolemia, although both drugs perform better for lowering triglycerides and raising HDL-C than for lowering LDL-C. More recently, in the Veterans Affairs HDL Intervention Trial (VA-HIT), which enrolled patients with low HDL-C levels and established coronary disease, gemfibrozil significantly reduced the combined end point of myocardial infarction and coronary death.¹² Noncardiovascular mortality was almost identical in the treatment and placebo groups in VA-HIT. Fenofibrate, a drug of considerable current interest, has not yet been studied in a trial large enough to examine clinical outcomes. Nevertheless, fenofibrate significantly inhibited progression of coronary stenosis in a large angiographic study of 418 diabetic subjects.⁷²

The characteristics and clinical correlates of PPAR- γ are summarized in Table III. The TZDs, including troglitazone, rosiglitazone, and pioglitazone, are PPAR- γ agonists used in clinical medicine, although troglitazone was discontinued because of hepatic toxicity. Thiazolidinediones improve insulin sensitivity and were approved for human use because they lower the glucose level in patients with type 2 diabetes mellitus. Thiazolidinediones appear to act through PPAR- γ to improve insulin sensitivity, but the exact mechanism remains unknown. Most of the TZD-mediated increase in glucose uptake occurs in skeletal muscle, which contains only trace levels of PPAR- γ . Thiazolidinediones reduce plasma free fatty acids via a PPAR- γ -mediated effect on adipose

Table III. Characteristics and clinical correlates of PPAR- γ

Sites of expression	Adipose tissue Colon Immune cells including monocytes/macrophages Retina Vascular endothelium
Natural ligands	Polyunsaturated fatty acids 15-deoxy-D ^{12,14} -PGJ ₂
Pharmacologic agonists	Troglitazone (withdrawn from market) Rosiglitazone Pioglitazone
Target genes	Adipocyte differentiation Fatty acid uptake Lipogenesis
Clinical effects	Insulin sensitizers Fat redistribution and weight gain \uparrow HDL-C, \downarrow triglycerides, \uparrow LDL-C Fluid retention and edema Decreased carotid intima-media thickness Anti-inflammatory

tissue, and the reduction in free fatty acids may secondarily improve insulin sensitivity in muscle. Peroxisome proliferator-activated receptor γ is a key activator of genes involved in adipocyte differentiation, an effect that may help explain weight gain in patients treated with TZDs.⁶⁶

No TZD trials with clinical cardiovascular outcomes have been completed. Because TZDs reduce insulin resistance, attention has been given to the possibility that they, like metformin, may prevent the onset of diabetes mellitus in patients with impaired glucose tolerance or metabolic syndrome. Multiple trials are currently studying this possibility.

Thiazolidinediones have been shown to exert additional effects on components of metabolic syndrome.^{73,74} High-density lipoprotein cholesterol is increased by all of the TZDs. Rosiglitazone makes LDL subfractions larger and more buoyant, raises LDL-C, and has variable effects on triglyceride concentrations.⁷⁵ Pioglitazone, which may activate PPAR- α as well as PPAR- γ , usually reduces triglyceride concentrations and has no significant effect on LDL-C.⁷⁶ Plasminogen activator inhibitor-1 and fibrinogen levels are reduced by TZDs. Small decreases in blood pressure have also been shown. Both troglitazone and pioglitazone have been associated with decreased carotid intima-media thickness in human studies.⁷⁷

Other pharmacologic approaches

In addition to therapies that target glucose metabolism, a variety of other pharmacologic agents that target body weight, lipids, and blood pressure may improve outcomes in metabolic syndrome. Two agents, orlistat and sibutramine, are approved for weight reduction in the United States. Orlistat inhibits pancreatic lipase in the intestinal tract, thereby reducing fat absorption by

approximately 30%. Sibutramine appears to reduce appetite by inhibiting the reuptake of serotonin and norepinephrine in hypothalamic neurons regulating hunger. Both agents showed efficacy in reducing body weight by an average of 8 to 12 lb more than placebo in randomized trials.^{78,79} Both agents were also shown to improve glucose control in diabetic patients and to improve lipoprotein fractions in obese subjects.⁸⁰⁻⁸² The cardiovascular risk profile of sibutramine is mixed because small increases in both average pulse rate and average blood pressure also occur. Further development and testing of these or similar weight-loss agents specifically for the treatment of metabolic syndrome are inhibited because healthcare payors have deemed them to serve cosmetic purposes (ie, weight reduction) rather than amelioration of health risk.

Other drugs that specifically target lipids and blood pressure are also useful in the management of metabolic syndrome. Review of these medications was beyond the scope of the meeting, but intriguing observations on diabetes prevention from 3 clinical trials deserve mention. In the Heart Outcomes Prevention Evaluation (HOPE) trial, ramipril, an ACE inhibitor, was associated with a 34% decrease in the rate of self-reported, new-onset diabetes mellitus ($P < .001$).⁸³ No similar observations with other ACE inhibitors are available. In the Losartan Intervention for Endpoint Reduction in Hypertension (LIFE) trial comparing the effects of losartan, an angiotensin II receptor blocker, versus atenolol on blood pressure and cardiovascular outcomes, the losartan group experienced a 25% lower risk of new-onset diabetes mellitus ($P = .001$). This result might be due to either an increase in diabetes incidence caused by atenolol or a decrease in diabetes incidence caused by losartan.⁸⁴ Finally, in the West of Scotland Coronary Prevention Study (WOSCOPS), pravastatin given for primary prevention of coronary disease was associated with a significant 30% decrease in cases of new-onset diabetes ($P = .04$).⁸⁵ However, the Anglo-Scandinavian Cardiac Outcomes Trial (ASCOT), which assessed 10 mg of atorvastatin versus placebo in almost 20 000 subjects, did not reproduce this result.⁸⁶

Future therapies

From a national perspective, both individual- and population-based approaches to metabolic syndrome are necessary. The prevention or reversal of obesity, for example, may be achieved by behavioral, pharmacologic, or even surgical modalities. Insulin sensitization may be achieved by behavioral (exercise and weight reduction) or pharmacologic means. Behavioral change might be facilitated by medications targeted at the hypothalamic centers for appetite. At the other end of the spectrum, the building of bicycle paths and the maintenance of safe parks for outdoor recreation can provide metabolic benefits for a population. Statistically

significant benefit and cost effectiveness must be shown by clinical studies with rigorous outcomes. The DPP, described earlier, provides benchmarks against which both lifestyle and pharmacologic approaches to diabetes prevention may be measured.

The pharmaceutical search for insulin sensitizers and other agents to treat metabolic syndrome, reviewed recently by Moller,⁸⁷ is broad-based, encompassing agents that affect transcription factors, intracellular and extracellular signaling, and basic glucose biochemistry; the latter includes the glycolytic pathway, citric acid cycle, and glycogen formation and breakdown pathways. Much attention is currently given to PPAR- γ agonists and PPAR- γ/α combined agonists.

Drug development will benefit from powerful molecular techniques applied *in vitro* and *in vivo*. The National Institute of Diabetes and Digestive and Kidney Diseases has initiated a program to develop a functional atlas of nuclear receptors.⁸⁸ These receptors exert their effects through aggregations of signaling molecules. Various coactivators and corepressors interact with the receptors, often resulting in tissue-specific effects subject to multiple influences beyond mere ligand binding.⁸⁹ The example of selective estrogen receptor modulators, such as raloxifene and tamoxifen, shows how differing specificities can be achieved through knowledgeable drug design.⁹⁰ Similar specific design characteristics might be possible within classes of PPAR agonists.

New molecular targets in metabolic syndrome will likely be discovered because of powerful genetic techniques currently in use. Mapping of haplotypes by single nucleotide polymorphisms may identify additional genes involved in obesity and insulin resistance. Proteomics facilitated by new methods of mass spectroscopy can be combined with genome sequence data to provide crucial information on gene expression.

Advances in mouse genetics are providing tissue-specific knockout or overexpression models. For example, knockout of insulin receptors in the liver, as opposed to in muscle and fat, showed much larger effects on systemic glucose homeostasis.⁹¹ Knockout of insulin receptors in pancreatic beta cells has implicated beta cells as an important target of insulin, not merely the source of insulin.⁹²

Research is also focused on adipose tissue, which is now recognized as an endocrine organ. Leptin, tumor necrosis factor α , angiotensinogen, resistin, adiponectin, IL-6, and other polypeptide mediators produced in adipocytes appear to exert distant or systemic effects. These adipocytokines were reviewed in the first part of the meeting summary. Subcutaneous administration of leptin has ameliorated hyperglycemia, hypertriglyceridemia, and fatty liver in patients with severe lipodystrophy. This disorder is characterized by deficiency or destruction of adipose tissue cells, leading to an exaggerated manifestation of metabolic syndrome.⁹³

Research is leading to a better understanding of the signaling pathways for insulin, leptin, and other hormones, as well as of modulators such as I κ B, which attenuates insulin signaling. Salicylate was found to weakly inhibit the I κ B pathway and, thus, in high doses to enhance insulin sensitivity.⁹⁴ This pathway provides a possible link between inflammation and insulin resistance.

As we understand more about the causation of metabolic syndrome, its heterogeneity becomes increasingly apparent. Phenotyping may allow identification of subsets of patients for whom one class of drug is superior to another. At the same time, we must be cautious and avoid overinterpreting the capabilities of new technologies. If blood or tissues are submitted to gene expression arrays, is it best to look for patterns or to try to interpret individual data points? Suppose that the *ras* oncogene exhibits a 2-fold increase in expression with some treatment. Should the treatment be considered “tumorigenic” and discarded even if another 50 oncogenes are unchanged? We need to understand how the powerful new techniques of gene analysis will translate to clinical outcomes.

Health system issues

Regulatory issues

Regulatory issues intersect with clinical needs, research discoveries, safety concerns, and cost-effectiveness estimates. Here, we consider regulation principally with regard to new drug development. Drugs are approved and labeled based on *expected benefits and risks* of therapy for the target disease in a particular population. To predict both efficacy and safety, clinicians, researchers, and regulators sometimes rely on “markers” as substitutes or stand-ins for the clinical end points of mortality and morbidity.

The use of such markers is well established in medicine and especially in metabolic disorders. Examples include blood pressure in hypertension, glucose and hemoglobin A_{1c} in diabetes mellitus, and LDL-C in atherosclerosis prevention and treatment. Hepatic transaminases are important safety markers. Historically, drugs have been approved and used largely on the basis of their effects on such markers, but today much more emphasis is placed upon the ability of drugs to improve mortality and morbidity.

A “surrogate” is a validated marker. In 1999, a National Institutes of Health conference defined a surrogate as a substitute for clinical benefit that correlates with outcome, explains most of the treatment benefit, and requires vigorous validation.⁹⁵ From a regulatory viewpoint, a change in a surrogate marker must be shown to be a reliable predictor of benefit and/or risk for a specific molecule or class molecules used to treat a

particular disease. Trials using the surrogate must show favorable effects, display no troubling safety signals, study large samples, and have long durations. Even when these criteria are met, the acceptance of a surrogate remains a leap of faith.⁹⁶

However, the suggestion that clinical evaluation should end with a surrogate is hazardous. From the point of view that regards them as final end points, almost no surrogates are validated. In lipidology, LDL-C is considered to be a reliable surrogate, and most methods and drugs for lowering LDL reduce cardiovascular risk. However, oral estrogen and doxazosin received favorable attention for their ability to lower LDL, but were subsequently shown to have unfavorable clinical outcomes.^{33,97}

Many metabolic end points commonly considered to be surrogates are in fact biomarkers—educated guesses about what will work that have not been validated. In many situations, single biomarkers are not adequate for outcome prediction; thus, biomarker arrays may be a solution. Metabolic syndrome itself may be considered a biomarker array, and in treating metabolic syndrome, a variety of end points and outcomes can be evaluated. For example, we might seek to (1) decrease insulin resistance, (2) decrease insulin levels, (3) improve some aspect of dyslipidemia, (4) lower blood pressure, (5) reduce an aggregate estimate of risk, (6) prevent diabetes mellitus, (7) improve 1 or more markers of atherosclerosis, (8) prevent atherosclerotic cardiovascular events, and/or (9) decrease overall mortality. The attainment of metabolic goals is expected to decrease clinical end points, but the expectation may not be fulfilled for some treatment modalities. To choose the best measures of efficacy, we must determine what primary and secondary metabolic abnormalities contribute to morbidity and mortality. Furthermore, how do effects on these markers combine to influence clinical events? If we choose 4 components of the syndrome—say, blood pressure, weight, glucose, and insulin resistance—and if 3 improve and 1 worsens with a particular treatment, what should be done with the data? If there is a central mechanism underlying the syndrome, we would expect all components to go in the same direction. The presence of a consistent pattern involving both metabolic markers and intermediate markers of vascular risk may help establish clinical benefit.

In considering potential drugs to treat metabolic syndrome, regulators are likely to be more comfortable with drugs that have long, safe usage histories in other conditions, such as metformin or statins, than with novel drugs. A novel treatment or drug will require more testing, especially because the goal is prevention rather than treatment of morbidity. The regulatory balance involves decisions about the degree to which efficacy and safety are ensured on the one hand, and the

adequacy of financial incentives for drug development on the other. It may be possible to introduce a drug on the basis of metabolic data, but clinical outcome trials will be needed before widespread marketing. Phased adoption of treatment strategies may be used. After a drug is on the market, adequate follow-up is required as the product is used in large numbers of people. Because of the need for hard clinical end points to establish benefit and risk, the Food and Drug Administration will ask, at minimum, for commitment to perform phase IV clinical outcome trials if therapies are to be approved for metabolic syndrome indications.

Clinical trials for metabolic syndrome

When planning clinical trials, the following general therapeutic principles should be kept in mind: (1) therapeutic effects are usually modest; (2) unintended targets are common; (3) long-term versus short-term effects differ; (4) combinations are unpredictable; (5) class effect may not be valid; and (6) most treatments have a combination of benefits and risks. Examples illustrating these principles abound in cardiovascular trials from the past 20 years. The Cardiac Arrhythmia Suppression Trial (CAST), in which antiarrhythmic drugs given for premature ventricular contractions increased risk of sudden death, taught us that therapies do not always work as expected.⁹⁸ The problem of unintended targets has been demonstrated by congestive heart failure trials of inotropic agents, which ultimately were proven to have negative effects. The use of combination medications is becoming more common; however, 2 or 3 good agents may be detrimental when given together. For example, ACE inhibitors, angiotensin II receptor blockers, and β -adrenergic blockers are each beneficial when given as monotherapy, but the effect of triple therapy remains unknown. The issue of class effects remains unresolved for most types of drugs.

As long-term clinical trials of pharmacologic treatment of metabolic syndrome are planned, numerous trial design issues must be considered. Appropriate subject selection is vital to ensure that results will be generalizable and that sufficient end points will occur to determine statistically significant benefit or harm. Background therapy should include diet, exercise, weight loss, and weight maintenance. In addition, one must consider background treatment of lipids and hypertension according to established guidelines.

The most readily attainable clinical goal in metabolic syndrome therapy may be the prevention of diabetes mellitus. The DPP has set a standard against which future therapies and trials must be measured. The microvascular problems associated with diabetes and hyperglycemia are known to decrease quality of life and increase healthcare costs. However, the prevention of clinical diabetes might not translate into effects on macrovascular disease, or cardiovascular benefit, as

atherosclerotic risk is already very high when type 2 diabetes mellitus is first diagnosed. In addition, even if lower blood glucose is better, the prevention of diabetes by lifetime treatment with a drug could lead to unanticipated chronic complications and toxicities associated with the drug. Postmarketing, phase IV trials will be needed to assess these possibilities.

NAVIGATOR: an example of trial design

One model for trial design in patients with impaired glucose tolerance is the Nateglinide and Valsartan in Impaired Glucose Tolerance Outcomes Research (NAVIGATOR) study.⁹⁹ This study, launched in November 2001, is the largest to date for diabetes prevention and will involve sites in 40 countries. NAVIGATOR has a short-term end point of diabetes prevention and long-term end points of safety and cardiovascular outcomes. Nateglinide might prevent new onset of diabetes, as this drug restores early phase insulin secretion, an early defect in the pathogenesis of type 2 diabetes. Valsartan is an angiotensin II receptor blocker that is similar to losartan, which was superior to atenolol in diabetes avoidance in the LIFE trial.⁸⁴ Because individuals with impaired glucose tolerance are at high risk for cardiovascular disease, the study is powered to look beyond diabetes prevention and to examine a potential decrease in cardiovascular events. Nateglinide and valsartan are each compared to placebo in a 2×2 factorial design. All subjects will receive lifestyle intervention with monthly diet and exercise counseling. Among subjects enrolled to date, one third have a previous cardiovascular event. Monitoring for new-onset diabetes will occur with evaluation of fasting plasma glucose every 6 months and annual oral glucose tolerance testing, to be repeated if fasting or postprandial glucose is elevated. Cardiovascular outcomes will be evaluated over 6 years with a composite outcome measure. Given the use of 2 drugs and 2 sets of end points, statistical issues that have to be addressed include splitting α . In this case, study designers have agreed to do hierarchical testing.

For subjects in NAVIGATOR who develop diabetes or hypertension during the trial, the addition of nonstudy medications may cloud interpretation. Intent-to-treat analysis, regardless of other medications, is planned. As a general principle, "local/best care if treated" will be provided. Thus, most subjects with new diabetes will get metformin, and new hypertensives will get an ACE inhibitor. Although for research purposes this is not the "cleanest" study design, it is the most reasonable ethical approach.

Cost effectiveness

Issues of cost and cost effectiveness for both the health system and society have become increasingly important as healthcare costs have continued to rise. A cost analysis based on the DPP estimated that health

system incremental costs, relative to the minimal costs of placebo treatment, were \$2191 per participant over 3 years for the metformin intervention and \$2269 for the lifestyle intervention. These incremental costs were considered modest. The balance of costs versus health benefits conferred by prevention of diabetes remains to be determined.¹⁰⁰ Computer modeling of direct and indirect costs will be used to estimate the quality-of-life-adjusted years gained because of decreased rates of new-onset of diabetes compared to costs of the program.

The example of the DPP shows that behavioral strategies aimed at individual patients can entail substantial costs, given the very high prevalence of metabolic syndrome in the population. Drug-based prevention might be more or less costly depending on effectiveness, side effects, and compliance. Regardless of regulatory approval of allowable treatment strategies, insurance payors must be persuaded to reimburse preventive medical treatment. Alternative behavioral interventions can be aimed for the society at less cost—for example, dietary recommendations, bicycle trails—but the medical community may have to tackle the food and entertainment industries as well as political interests.

Summary

Despite its etiologic complexities, regulatory hurdles, and clinical uncertainties, metabolic syndrome must be recognized as a looming danger to public health in the United States. It demands the best efforts of clinicians, bench scientists, regulators, advocates, and clinical trialists to find viable solutions in the near future.

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