

Metabolic syndrome: Definition, pathophysiology, and mechanisms

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In 1988, Reaven¹ proposed that insulin resistance is central to the etiology of type 2 diabetes mellitus, hypertension, and coronary artery disease. The concept of insulin resistance and associated metabolic abnormalities leading to increased risk of cardiovascular disease became known by a variety of names, including metabolic syndrome, dysmetabolic syndrome, syndrome X, cardiometabolic syndrome, and insulin resistance syndrome. Today, metabolic syndrome is a matter of immense public concern for atherosclerosis prevention. The syndrome may affect more than 50% of the elderly in the United States and even higher percentages in various ethnic groups around the world.² Its prevalence continues to rise, probably as a result of increasing obesity in the United States and elsewhere.

In April 2002, a conference on "The Metabolic Syndrome of Cardiovascular Risk and Insulin Resistance" was held in Washington, DC. This is the first of 2 papers summarizing the discussion at that conference. This paper will address the definition of metabolic syndrome, the components of the syndrome, and the underlying pathophysiology. The second paper will address atherogenesis and therapeutic targets, as well as the design of future studies and clinical trials.

Defining metabolic syndrome

Clinical findings most commonly associated with metabolic syndrome include insulin resistance, dyslipidemia (specifically high triglycerides, low levels of high-density lipoprotein [HDL], and small dense low-density lipoprotein [LDL]), central obesity, hypertension, impaired glucose tolerance or diabetes mellitus, and high

rates of atherosclerotic disease. Recently, markers of inflammation and accelerated hemostasis/impaired fibrinolysis have been added to this list. Table I lists proposed components of metabolic syndrome and their associated findings.

Measurements of insulin resistance

Insulin resistance, implying depressed cellular sensitivity to insulin, is a central feature of metabolic syndrome. Insulin sensitivity varies by organ, cell type, and metabolic pathway examined. Clinical measurement of insulin resistance generally focuses on whole-body glucose uptake in response to circulating insulin. The resulting measurement is an aggregate response of a complex homeostatic system. Not surprisingly, a variety of techniques have been proposed and used to measure insulin sensitivity. The "gold standard" measurement of insulin resistance employs a euglycemic clamp technique in which insulin is infused intravenously at a constant rate and blood glucose is measured frequently so that glucose can be infused at a variable rate to maintain a constant glucose level. The plateau rate of glucose infusion is the critical measure of insulin sensitivity. In some studies, this technique has been modified by infusing somatostatin to inhibit endogenous insulin and glucagon release.³

Insulin sensitivity can also be estimated by analyzing glucose and insulin curves after bolus administration of a fixed glucose load. Mathematical modeling is required to determine parameters related to glucose distribution, pancreatic insulin response, insulin disposal, and insulin-sensitive and independent components of glucose uptake. Bergman's "minimal model" on the basis of a frequently sampled intravenous glucose tolerance test is the most widely used method of this kind.⁴

The simplest measures of insulin sensitivity are single determinations of plasma insulin in nondiabetic individuals, either in the fasting state or 1 to 2 h after administration of an oral glucose load. Plasma insulin alone, however, provides only a rough measure of insulin sensitivity or resistance. It may be adequate for large epidemiological studies, but it is not recommended for clinical assessment of individual patients.⁵ A refinement of this approach uses the product of fasting insulin and glucose, suggested by Matthews et al⁶ as part of

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Table I. Metabolic syndrome: proposed components and associated findings

1. Insulin resistance*
2. Hyperinsulinemia*
3. Obesity: visceral (central), but also generalized obesity*
4. Dyslipidemia: high triglycerides, low HDL, small dense LDL*
5. Adipocyte dysfunction
6. Impaired glucose tolerance or type 2 diabetes mellitus*
7. Fatty liver (nonalcoholic steatohepatitis, steatohepatitis)
8. Essential hypertension: increased systolic and diastolic blood pressure*
9. Endothelial dysfunction (discussed in next paper of this series)
10. Renal dysfunction: micro- or macroalbuminuria
11. Polycystic ovary syndrome
12. Inflammation: increased CRP and other inflammatory markers
13. Hypercoagulability: increased fibrinogen and PAI-1
14. Atherosclerosis leading to increased cardiovascular morbidity and mortality*

*Most widely incorporated into the definition of metabolic syndrome.

their homeostatic model assessment of insulin resistance (HOMA_{IR}). The exact formula is

$$\text{HOMA}_{\text{IR}} = \frac{\text{fasting insulin (mU/L)} \times \text{fasting glucose (mmol/L)}}{22.5}$$

where the constant divisor of 22.5 simply indexes HOMA_{IR} to 1.0 in normal individuals. Whereas HOMA_{IR} is easy to obtain and has been increasingly used in surveys and longitudinal studies, it has the drawback of relying upon fasting measurements of insulin and glucose to predict glucose metabolism in the insulin-stimulated state.

Epidemiological studies are beginning to ask whether insulin resistance as a single variable may help predict who is at higher risk for cardiovascular morbidity and mortality. A total of 147 healthy nonobese individuals were examined for insulin resistance and followed over 5 y to determine cardiovascular outcomes.⁷ Subjects were divided into tertiles of insulin resistance on the basis of steady-state plasma glucose measurements used to estimate insulin-mediated glucose disposal. Those individuals in the most insulin-sensitive tertile had no cardiovascular events, whereas 27% (13/49) of those in the most insulin-resistant tertile had a cardiovascular event.

Diagnostic criteria for metabolic syndrome

Several definitions and criteria have been proposed for the metabolic syndrome associated with insulin resistance. In 1998, the American Diabetes Association (ADA) issued a consensus statement identifying "glucose intolerance, central obesity, dyslipidemia (increased triglycerides, decreased HDL, increased small dense LDL), hypertension, increased prothrombotic and anti-fibrinolytic factors, and a predilection for atherosclerotic

vascular disease" as components of the metabolic syndrome associated with insulin resistance.⁸ The ADA statement stopped short of providing diagnostic cut-points or criteria for the syndrome. Other organizations stepped forward, however, and the 2 most widely used definitions come from the World Health Organization (WHO)⁹ and the US National Cholesterol Education Program (NCEP).¹⁰ The American College of Endocrinology/American Association of Clinical Endocrinologists (ACE/AACE) has also recently presented recommendations on key clinical signs and risk factors for the insulin resistance syndrome.

Under the WHO criteria, a disturbance of glucose/insulin metabolism must be present—type 2 diabetes mellitus, impaired glucose tolerance, or normal glucose tolerance with insulin resistance (defined as highest quartile of HOMA_{IR}). Diagnosis of metabolic syndrome, in addition, requires at least 2 of the following 4 components: (1) hypertension, either treated by medication or $\geq 160/90$ mm Hg untreated; (2) dyslipidemia with elevated plasma triglycerides (≥ 1.7 mmol/L or ≥ 150 mg/dL) and/or low HDL (< 0.9 mmol/L or < 35 mg/dL in men, < 1.0 mmol/L or < 39 mg/dL in women); (3) obesity with body mass index (BMI) > 30 kg/m² or central adiposity (waist-hip ratio > 0.90 in men or > 0.85 in women); and (4) microalbuminuria (urinary average excretion rate ≥ 20 $\mu\text{g}/\text{min}$ or albumin-creatinine ratio ≥ 20 mg/g).⁹

The NCEP criteria for metabolic syndrome require at least 3 of the following: waist circumference > 40 in men or > 35 in women, plasma triglycerides ≥ 150 mg/dL, HDL cholesterol < 40 mg/dL in men or < 50 mg/dL in women, blood pressure $\geq 130/85$ mm Hg, and fasting plasma glucose 110 to 125 mg/dL. Notably, the NCEP definition does not include a measure of insulin resistance, and it relies upon fasting glucose measurement rather than determination of glucose tolerance. According to the NCEP, metabolic syndrome is a precursor to, but does not include, type 2 diabetes mellitus, whereas the WHO criteria consider metabolic syndrome and diabetes to be intersecting diagnostic categories.¹⁰

ACE/AACE recognizes risk factors for metabolic syndrome that include the following key clinical signs: overweight or obesity emphasizing abdominal obesity, borderline-high or high triglyceride levels, low HDL cholesterol, moderately high or higher blood pressure, and either impaired glucose tolerance (2-h post 75 g oral glucose challenge) or impaired fasting glucose. Specific cutpoints are given in Table II. Other risk factors recognized by ACE/AACE include polycystic ovary syndrome, prior gestational diabetes, sedentary lifestyle, age, ethnicity, and family history of type 2 diabetes mellitus, hypertension, or cardiovascular disease. ACE/AACE did not specify a certain number of risk factors as defining metabolic syndrome, but stated that the

Table II. Metabolic syndrome: criteria and definitions

| ADA | NCEP* | WHO† | AACE‡ |
|------------------------------------------------------|----------------------------------------------------|------------------------------------------------------------------------------------------|------------------------------------------------------------------------------------|
| Glucose intolerance | Fasting plasma glucose 110-125 mg/dL | Type 2 diabetes, impaired glucose tolerance, or insulin resistance by HOMA _{IR} | Fasting plasma glucose 110-125 mg/dL or 2-h post 75 g glucose challenge >140 mg/dL |
| Central obesity | Waist circumference >40 in (men) or >35 in (women) | BMI >30 or waist-hip ratio >0.90 (men) or >0.85 (women) | BMI ≥25 or waist circumference >40 in (men) or >35 in (women) |
| Dyslipidemia: high TG, low HDL, small dense LDL | TG ≥150 mg/dL, HDL <40 (men), HDL <50 (women) | Dyslipidemia: TG ≥150 mg/dL, HDL <35 (men), HDL <39 (women) | TG ≥150 mg/dL, HDL <40 (men), HDL <50 (women) |
| Hypertension | Blood pressure ≥130/85 mm Hg | Hypertension: on medication or untreated blood pressure ≥160/90 mm Hg | High blood pressure ≥130/85 mm Hg |
| Increased prothrombotic and antifibrinolytic factors | | Microalbuminuria ≥20 µg/min | |
| Increased atherosclerotic vascular disease | | | |

TG, Triglycerides.

*NCEP: must meet 3 of 5 criteria (low HDL and high triglycerides are 2 criteria).

†WHO: must meet glucose/insulin criterion and 2 more.

‡AACE: these key clinical signs are considered risk factors. Other risk factors include polycystic ovary syndrome; sedentary lifestyle; age; ethnicity (certain groups); and family history of type 2 diabetes, hypertension, or cardiovascular disease.

syndrome becomes more likely as the number of risk factors increases.¹¹

Epidemiology and factor analysis

The prevalence of metabolic syndrome among US adults is approximately 22%, determined by applying NCEP criteria to the National Health and Nutrition Examination Survey III data set.² Prevalence increased with age from 6.7% in the 20 to 29 y of age range to 43.5% in the 60 to 69 y of age range. Hispanics were affected more than either blacks or whites. When divided by sex, black women had greater prevalence than white women, whereas white men had greater prevalence than black men.

Isomaa et al,¹² using WHO criteria, examined the prevalence and cardiovascular risk of metabolic syndrome in subjects participating in a large family study of type 2 diabetes mellitus in Finland and Sweden. Among men, 15% with normal glucose tolerance, 64% with impaired glucose tolerance, and 84% with type 2 diabetes mellitus had metabolic syndrome; among women, 10% with normal glucose tolerance, 42% with impaired glucose tolerance, and 78% with type 2 diabetes mellitus had metabolic syndrome. Over a 7-y period, cardiovascular mortality was 12% in those with metabolic syndrome and 2.2% in those without.

The concept of a disease affecting such a high proportion of the population, as indicated by these studies, is perplexing. Therefore, most investigators prefer the term *syndrome*, a condition aggravated by progressive weight gain, by sedentary lifestyle, and perhaps by aging per se. The question of whether to use liberal or stringent cutpoints for various components of

this syndrome leads to further questions: Are we recognizing or merely defining an epidemic condition in the modern postsubsistence world? How will event rates for study end points be affected by syndrome definitions? Synergistic interaction among multiple components of the syndrome might lead to event rates that are higher than otherwise expected.

Factor analysis provides an objective look at metabolic syndrome in epidemiological studies. This statistical technique seeks to determine underlying linear combinations of multiple variables (planes in multidimensional space) that simplify the large set of variables into a smaller set of principal factors or components (defined as the planes that minimize residual variance). Factor analysis allows the assignment of some or all variables to groups on the basis of high correlations, termed *loadings*, with principal factors. This type of analysis was recently performed as part of the Insulin Resistance Atherosclerosis Study, which examined a multiethnic population in San Antonio.¹³ Patients with diabetes mellitus were excluded. The variables grouped by principal factors included a metabolic group, a hypertension group, and optionally, a proinflammatory factor group. In one model, the metabolic factor group (including BMI, waist circumference, fasting and 2-h glucose, insulin sensitivity, low HDL cholesterol, and high triglycerides) explained 28% of total variance, whereas the hypertension factor group (systolic and diastolic blood pressure) explained 9%. In a second model, the metabolic group (including BMI, waist circumference, fasting and 2-h glucose, insulin sensitivity, low HDL, high triglycerides, and plasminogen activator inhibitor-1 [PAI-1]) explained 23% of the variance; the proin-

flammatory group (BMI, insulin sensitivity, fibrinogen, and C-reactive protein [CRP]) explained 7%; and the hypertension group (systolic and diastolic blood pressure) explained 5%. These findings were consistent across ethnic groups.

The next generation: Metabolic syndrome in children

Similar to the trend in adults, there is currently an alarming increase in the rate of obesity and type 2 diabetes mellitus in children in the United States.^{14,15} Several studies have examined the interactions among fasting insulin, lipoproteins, blood pressure, and weight in children, pursuing the hypothesis that metabolic syndrome is a developmental disorder strongly influenced by risk factors in the first 2 decades of life.¹⁶⁻¹⁸

Sinaiko et al^{17,18} examined the relation among obesity, insulin resistance, and cardiovascular risk factors in children aged 11 to 14 y. A total of 357 children were studied by using both fasting insulin level and euglycemic insulin clamp technique to assess insulin resistance. The amount of intravenous glucose required to maintain euglycemia during constant insulin infusion defines the insulin sensitivity, termed M_{lbm} , after normalization to lean body mass. Girls had lower M_{lbm} than boys, and thus, more insulin resistance. Body mass index significantly correlated with fasting insulin and inversely correlated with M_{lbm} . Fasting insulin significantly correlated with systolic blood pressure and triglycerides, whereas M_{lbm} significantly correlated only with triglycerides (both sexes) and HDL (girls only). A clustering of metabolic syndrome risk factors was seen only in the children in the lowest quartile of M_{lbm} (the highest degree of insulin resistance). The study concluded that insulin resistance (as defined by M_{lbm}) was associated with risk factors for metabolic syndrome in 11- to 14-y olds, regardless of body fat. Ongoing research will repeat these studies at ages 18 to 19 and also examine parents and siblings. In the same study, review of diet showed increasing insulin sensitivity with increasing grain intake; this relationship was stronger in more obese children. Higher fiber intake was also associated with a lower fasting insulin level.

When dietary fat is converted to fatty acids and reesterified to triglyceride for storage in adipocytes, 3% of the energy in food is lost, but when dietary carbohydrates are converted and stored as triglyceride in adipocytes, 10% to 15% of the energy is lost. Therefore, eating the same number of calories as fat can result in more efficient energy storage and more weight gain. One should note, however, that the historical 38% fat content in the US diet has decreased to a current 32% fat content, although rates of obesity have increased. This finding is likely due to increased total caloric intake as well as reduced exercise.

Ethnic differences in the prevalence of metabolic syndrome in children have been described. Among 403 third-grade children, Batey et al¹⁹ found that a summary score of metabolic variables associated with insulin resistance was significantly higher in Mexican American children than in non-Hispanic white children. Among 144 mother-child pairs examined by Reaven et al,²⁰ Mexican American mothers and children had higher BMI, triglycerides, very low-density lipoprotein (VLDL), fasting insulin, and cholesterol/HDL ratio, and lower HDL levels than did non-Hispanic white mothers and children. Mother-child pairs also showed significant correlations of cardiovascular risk factors, owing to either genetic or environmental influences. These 2 studies show that metabolic syndrome begins early in life and is possibly related to early obesity and insulin resistance from genetic and/or environmental causes.

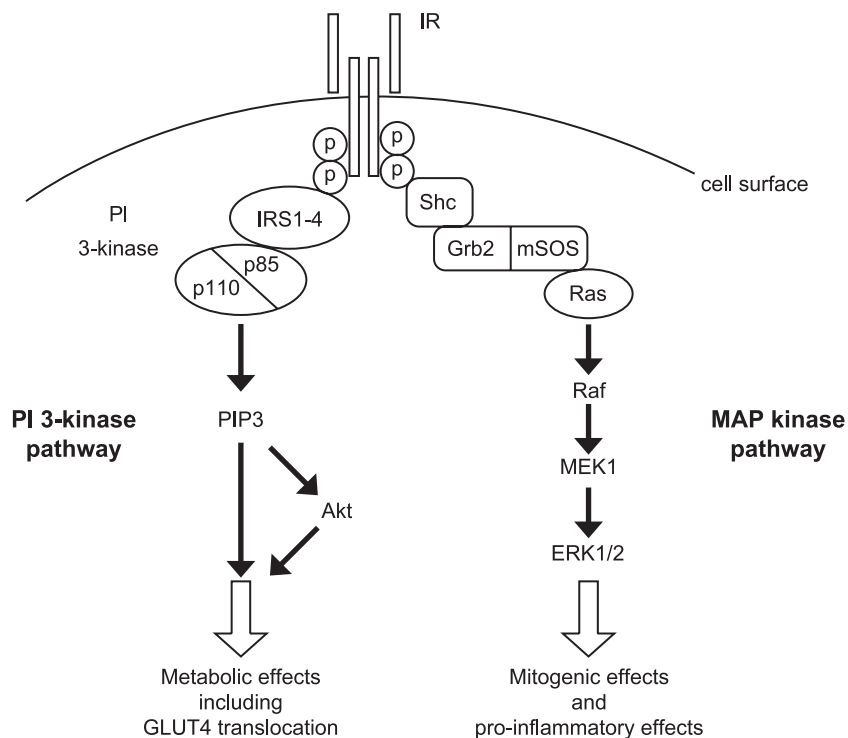
Pathophysiology: Molecular mechanisms of insulin action and insulin resistance

Metabolic syndrome is associated with insulin resistance, but it is not a consequence of insulin resistance alone, nor is it a direct consequence of the lack of insulin action. This is most evident in patients with insulin receptor mutations or autoimmune antibodies to the insulin receptor; they may have 100-fold or greater elevations of circulating insulin or require similarly high doses of exogenous insulin to control diabetes. These patients exhibit a distinct syndrome with acanthosis nigricans and a high risk of diabetes, but typically have no obesity, hypertension, or atherogenic dyslipidemia.²¹ Moreover, patients with type 1 diabetes mellitus, who lack insulin, do not exhibit the same atherogenic lipoprotein phenotype typical of patients with metabolic syndrome or type 2 diabetes mellitus. Lean type 1 diabetic mellitus patients do not characteristically have insulin resistance.

If metabolic syndrome does not result purely from a lack of insulin effect, then how might insulin resistance generate other features of the syndrome? Proposed mechanisms center around 3 themes: effects of mild to moderate hyperglycemia, effects of compensatory hyperinsulinemia, and effects of unbalanced pathways of insulin action. Hyperglycemia, largely postprandial and below diabetic levels, may lead to a variety of effects usually associated with diabetes. For example, moderate hyperglycemia might be postulated to cause accelerated atherogenesis via advanced glycosylated end products or via enhanced collagen formation. The association between the level of glycemic control and atherosclerosis development, however, is not very strong (to be covered in the second paper of this meeting summary).²²

A more important mechanism may be compensatory hyperinsulinemia. The maintenance of normal post-

Figure 1



Two major pathways of insulin signaling. Binding of insulin to the insulin receptor causes autophosphorylation (P). The PI-3K pathway is initiated by tyrosine phosphorylation of insulin receptor substrate protein (IRS-1-4). Phosphorylated IRS-1-4 associates with p85, the regulatory subunit of PI-3K. Activated PI-3K, composed of p85 and p110 subunits, produces PIP₃. Elevated levels of the signaling molecule PIP₃ result in the activation of Akt and other downstream effector molecules that mediate metabolic effects of insulin signaling, including GLUT4 translocation to the cell surface. The upstream mediators of the MAP kinase pathway are Shc, Grb2, mSOS, and Ras. Activation of Ras triggers the cascade of Raf to MEK1 to ERK1 and ERK2. ERK1 and ERK2 are 2 MAP kinase subtypes that mediate the mitogenic and the proinflammatory effects of insulin signaling. IR indicates insulin receptor.

prandial glucose homeostasis requires that the pancreatic beta cells secrete a normal amount of insulin in response to the hyperglycemic challenge and that the resultant hyperinsulinemia (1) stimulates glucose uptake by muscle, the tissue responsible for the disposal of 80% to 90% of the ingested glucose load, and (2) suppresses endogenous glucose production, over 80% of which is derived from the liver. In insulin-resistant conditions, the ability of insulin to augment glucose uptake and inhibit hepatic glucose production is impaired. The resultant hyperglycemia presents a stimulus to the beta cells, which secrete large amounts of insulin after meals. Initially, attention was directed to the concept that certain organs and tissues can have lesser degrees of insulin resistance than skeletal muscle and liver. For example, the high insulin concentration required to produce normal glucose uptake in skeletal muscle may overstimulate cells of the arterial wall. In recent years, this concept has been expanded to include the idea that

not only different cell types, but also different metabolic pathways within the same cell, may differ in their responsiveness to insulin. The pathways of insulin action are thrown off balance.

Binding of insulin to the insulin receptor leads to activation of its tyrosine kinase activity and autophosphorylation of specific tyrosine residues of the receptor (Figure 1). In turn, the activated insulin receptor phosphorylates tyrosine residues on substrate proteins, initiating the intracellular signaling cascade. The 2 major pathways for insulin signaling are the phosphatidylinositol 3-kinase (PI-3K) and the mitogen-activated protein (MAP) kinase pathways.²⁵ The PI-3K pathway is initiated by tyrosine phosphorylation of a member of the insulin receptor substrate family (IRS-1/2/3/4), which associates with the p85 regulatory subunit of PI-3K, leading to activation of the enzyme. Phosphatidylinositol 3-kinase causes phosphatidylinositol 3,4,5-phosphate (PIP₃) to be produced, resulting in the activation of Akt and other

downstream effector molecules that mediate the metabolic response to insulin, which includes translocation of GLUT4 transporter to the membrane. The MAP kinase pathway begins with phosphorylation of SHC or insulin receptor substrate, which binds Grb2 and activates Ras via mammalian Son of Sevenless (mSOS). Ras then binds and disinhibits Raf, which activates another kinase, MEK1. MEK1 activates extracellular signal-regulated kinases ERK1 and ERK2. Activated ERKs, which are a type of MAP kinase, mediate the mitogenic and proinflammatory responses of insulin signaling. In metabolic syndrome and type 2 diabetes mellitus, the pathways leading to activation of PI-3K are blocked, possibly through serine phosphorylation of the insulin receptor and/or IRS proteins, whereas the MAP kinase pathway remains open and may even be hypersensitive.²⁴ Thus, antagonists of the appropriate serine kinase might reverse insulin resistance and restore the balance of MAP kinase and PI-3K signaling pathways.

To discern these effects *in vivo*, Hispanic offspring of 2 diabetic parents were studied by use of euglycemic clamps with muscle biopsy before the clamp and 30 min into clamp.²⁵ In these offspring, the chance of developing type 2 diabetes mellitus is 70% to 80%. Increases in fasting blood glucose correlated with hepatic insulin resistance, whereas 2-h postprandial blood glucose elevation correlated with muscular insulin resistance. The offspring had normal insulin receptor phosphorylation, but depressed tyrosine phosphorylation of IRS-1, as well as abnormal PI-3K and p85/IRS-1 signaling. ERK and MAP kinase pathways were intact. This experimental human model has been used to examine the effects of elevated free fatty acid levels on the development of muscular insulin resistance. Infusion of a lipid emulsion to elevate the plasma free fatty acid concentration reproduced the defects in IRS-1 and PI-3K demonstrated above. At the level of whole-body physiology, a key feature of metabolic syndrome is that free fatty acid production and release from adipocytes are not suppressed normally with the usual levels of insulin (see later section on free fatty acid metabolism). Furthermore, adipocyte resistance to the antilipolytic effect of insulin and the consequent elevated plasma free fatty acid levels may play an important role in the development of insulin resistance in muscle and other target tissues.

The PI-3K signaling pathway also increases nitric oxide, which is a potent inhibitor of vascular smooth muscle cell growth. Thus, an impairment in the PI-3K pathway could contribute to vascular endothelial dysfunction. Conversely, smooth muscle cell growth and proliferation are stimulated by activation of the ERK-MAPK pathway, which maintains normal sensitivity to insulin even in insulin-resistant conditions. The overall effect may be to enhance atherogenesis.^{24,26}

Pathophysiology: Dysfunctional energy storage and obesity

Some investigators regard insulin resistance as a mediating factor in metabolic syndrome, but not as the primary cause (Figure 2). These investigators consider dysfunctional energy storage to be the fundamental issue. In this theory, insulin resistance is thought to arise from abnormalities in the processing and storage of fatty acids and triglyceride, molecules that account for most of the body's energy utilization and storage. In most patients, the key abnormality is simply the presence of too much triglyceride, or body fat—that is, obesity. The purpose of adipose tissue throughout the body is energy storage: taking in food calories during and after meals, storing the calories as triglyceride, and then releasing calories in the form of fatty acids when energy is needed. It is safest for the body to store triglyceride in small peripheral adipocytes. If the capacity of these adipocytes to store triglyceride is exceeded, triglyceride accumulates in hepatocytes, skeletal myocytes, and visceral adipocytes. The abnormal triglyceride accumulation may lead to the development of hepatic and muscular resistance to insulin. This is referred to as the “overflow hypothesis.” Visceral adiposity can be measured by waist circumference, waist-hip ratio, or radiographic scans, and it correlates well with insulin resistance and other features of metabolic syndrome. Excess triglyceride in myocytes and in abnormally large peripheral adipocytes appears to engender insulin resistance in these cells.²⁷ Triglyceride in hepatocytes is recognized as fatty liver and may drive the formation and secretion of excessive VLDL.

This theory does not postulate an exclusive role for visceral adiposity, because abnormal peripheral fat cells and triglyceride-laden muscle cells participate in the dysfunctional state. Body mass index and waist circumference tend to load equally in factor analysis, if gender is taken into account. Body mass index may be a sufficient measure for many studies, particularly retrospective analyses of prior data sets.

Lipodystrophy syndromes offer striking examples of the effects of the inability to store triglyceride in the physiologically preferred small peripheral adipocytes.²⁸ The mutations underlying many cases of lipodystrophy are known. The disorders can present in childhood or in adult life with dramatic loss of subcutaneous fat below the shoulder girdle. These patients can develop severe hypertriglyceridemia, insulin resistance, fatty liver, and eventually diabetes mellitus. Patients infected with the human immunodeficiency virus, especially those treated with protease inhibitors, also develop partial lipodystrophy with similarly exaggerated features of metabolic syndrome.²⁹ Despite their loss of most subcutaneous fat, these patients develop abdominal obesity and fat pads around the base of the neck.

Figure 2

| Insulin Resistance | Dysfunctional Energy Balance |
|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| Insulin resistance ↓ Compensatory hyperinsulinemia to maintain euglycemia, especially postprandially ↓ Obesity (appetite stimulation?) Beta-cell exhaustion and diabetes Overstimulation of MAP kinase pathway Dyslipidemia due to high fatty acid flux ↓ Metabolic syndrome ↓ Increased atherosclerosis and cardiovascular disease | Triglycerides and fatty acids (molecules of energy storage and utilization) ↓ Triglyceride stored physiologically in small peripheral adipocytes ↓ Energy In > Energy Out ↓ With obesity, excess triglyceride goes to hepatocytes (fatty liver), skeletal myocytes, visceral adipocytes, abnormal large peripheral adipocytes ↓ Excess triglyceride leads to insulin resistance in these cells and metabolic syndrome with increased cardiovascular disease |

Complementary roles of insulin resistance and dysfunctional energy balance.

In addition to visceral fat, liver, and muscle, excess triglyceride can accumulate in abnormally large peripheral adipocytes. Cross-sectional studies indicate that large subcutaneous adipocyte size is associated with insulin resistance. In a study of Pima Indians, subcutaneous abdominal adipocyte size strongly correlated with risk for developing type 2 diabetes mellitus. This effect of adipocyte size on the risk of developing diabetes was independent of and additive to the effect of insulin resistance.³⁰ Danforth²⁸ has proposed that this correlation of adipocyte size and type 2 diabetes mellitus suggests a difficulty in differentiating new adipocytes. A failure of adipocyte differentiation limits the pool of adipocytes available for energy storage, and excess triglyceride overflows to other sites, leading to insulin resistance.²⁸

Studies of morbidly obese people who achieved a reduction in BMI from 45 to 35 with surgery showed normalization of insulin sensitivity. When the fat content of muscle was examined in these individuals, it had been reduced to zero, demonstrating that intramyocellular fat content is an important determinant of insulin sensitivity. Fat is found in muscle and liver when it overflows from overwhelmed adipocytes; once fat moves back from muscle and liver to adipocytes, it seems to be stored safely without causing metabolic derangement.^{28,31}

Adipocytes as an endocrine organ

The adipocyte is now recognized as the source of multiple bioactive peptides. Increased adipocyte mass

has been associated with increased expression of angiotensinogen, tumor necrosis factor- α (TNF- α), resistin, leptin, and PAI-1.³²⁻³⁵ Adiponectin is a protein expressed by adipocytes. Circulating adiponectin concentrations are actually decreased in obesity, as well as in the type 2 diabetes mellitus.³² Angiotensinogen is linked with the development of hypertension in obesity, and PAI-1 with antifibrinolysis, as discussed later. Overexpression of TNF- α has been demonstrated in obese rodents and human beings. By several approaches, TNF- α has been linked to insulin resistance in peripheral nonadipose tissues of rodents,^{34,35} although its role in the development of insulin resistance in human beings is less clear. Resistin was discovered by screening for genes that are induced during adipocyte differentiation and down-regulated in mature adipocytes treated with thiazolidinediones. In mouse studies, high levels of resistin correlated with insulin-resistant states, and resistin administration led to insulin resistance in vivo and in vitro.³³ Leptin is genetically deficient in the ob/ob mouse and is responsible for the striking development of obesity in this strain. There are leptin receptors in appetite centers in the brain, and their activation suppresses appetite. In rodents, leptin receptors in liver and muscle play an important role in the partitioning of substrate flux. The main physiological role of leptin may be to aid adaptation to low-energy intake rather than to resist high-energy intake. Recently, leptin administration was shown to partially reverse insulin

resistance and lipoprotein abnormalities in patients with lipodystrophy.³² As mentioned above, plasma levels of adiponectin are reduced in obesity and type 2 diabetes mellitus. Loss of adiponectin has been proposed to play a role in atherosclerosis, insulin resistance, and several components of metabolic syndrome, perhaps via effects on fatty acid metabolism in liver and muscle. Thiazolidinediones increase adiponectin gene expression and circulating plasma levels and enhance insulin sensitivity.³²

Glucose homeostasis

At a given level of obesity, an individual may have normal glucose tolerance, impaired glucose tolerance, or type 2 diabetes mellitus. The factors that determine the degree of impairment of glucose metabolism are insulin sensitivity and beta cell reserve capacity. Insulin sensitivity in normal individuals varies by approximately 3-fold.¹ In insulin-resistant individuals, the pancreatic beta cells must secrete more insulin to maintain euglycemia. However, prolonged hyperfunction can lead to "beta cell exhaustion." The biochemical/molecular basis of beta cell exhaustion has yet to be elucidated. Nevertheless, when the reserve capacity is exceeded by metabolic demands, insulin secretion is inadequate. Blood glucose levels then begin to rise, manifested first as impaired glucose tolerance and later as type 2 diabetes mellitus.

Free fatty acid metabolism

In addition to its role in stimulating glucose uptake in peripheral cells, insulin also inhibits free fatty acid release from adipocytes. An early manifestation of insulin resistance is the inability to suppress lipolysis in fat tissue. The excessive plasma free fatty acids are transported to the liver and muscle, where they stimulate hepatic glucose production and inhibit insulin-stimulated glucose uptake, worsening hyperglycemia.²⁸ Thiazolidinediones decrease free fatty acid concentrations and reduce free fatty acid turnover.

Although free fatty acids are important in the pathophysiology of metabolic syndrome, measurements are difficult to interpret, even in the clinical research setting, because of effects of diet and activity. The European Group on the Study of Insulin Resistance examined free fatty acids in 541 normoglycemic subjects by use of the euglycemic clamp technique.³⁶ Free fatty acid concentrations were positively correlated with fasting plasma glucose, fasting plasma insulin, and fasting triglycerides, but not with any other plasma lipoprotein concentrations. In other studies, the relation among free fatty acids, insulin resistance, and hypertension has been strong.¹ Unfortunately, concentrations of free fatty acids in the blood are highly variable over

the course of the day, precluding their practical use in clinical medicine.

Dyslipidemia

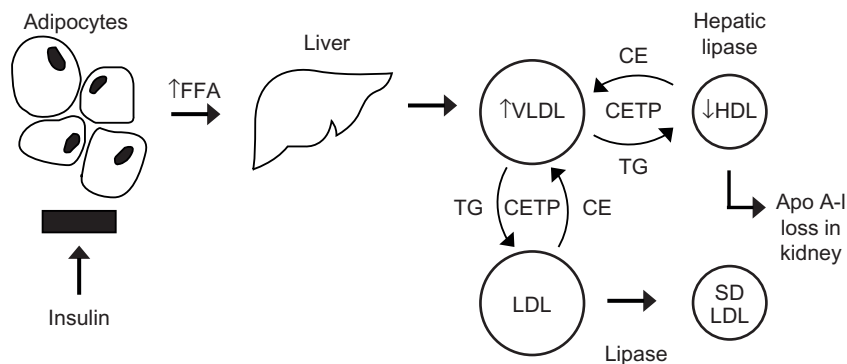
The dyslipidemia of metabolic syndrome is characterized by elevated triglycerides (VLDL), low HDL, and small dense LDL, a triad that has been termed the *atherogenic lipoprotein phenotype*.³⁷ In the Copenhagen Male Study, a prospective cardiovascular study of 5249 men, the combination of high triglyceride and low HDL concentrations was compared to high LDL concentrations as a risk factor for ischemic heart disease.³⁸ Subjects were divided into 4 groups on the basis of lipid levels, and ischemic heart disease events (ICD-8 codes 410-414) for each group were as follows: 6.6% in control group with normal triglyceride, HDL, and LDL levels; 11.4% in high triglyceride and low HDL group; 8.2% in high LDL group; and 17.5% in those with high triglycerides, low HDL, and high LDL. These results suggest that the dyslipidemia of metabolic syndrome is at least as powerful as high LDL in predicting cardiac risk.

Mechanisms linking insulin resistance and dyslipidemia are shown in Figure 3. Adipocytes release free fatty acids to circulating blood, which delivers them to the liver and muscle. In the liver, limited quantities of free fatty acids are oxidized, and most are reesterified to form triglyceride. Thus, fatty acids and triglyceride are constantly being transported between the liver and adipose tissue. If this process does not sufficiently favor transport toward adipose tissue, then the liver may accumulate triglyceride, a condition known as hepatic steatosis, or more commonly, fatty liver. Triglyceride can also accumulate in muscle cells because of inefficient oxidation or excess free fatty acid transport to muscle.

In the presence of insulin resistance, lipolysis in adipocytes is enhanced, plasma concentrations of free fatty acids increase, and more free fatty acids are transported to liver and muscle. At the same time, insulin drives lipogenesis in the liver directly. Elevated plasma glucose concentrations may also increase hepatic triglyceride synthesis by providing the carbon skeleton for glycerol. These changes enhance VLDL production by the liver. Excessive triglyceride and apolipoprotein B (apoB) enter the circulation as VLDL production is augmented.

Very low-density lipoprotein triglyceride is exchanged for cholesteryl ester in HDL and LDL by the action of cholesteryl ester transfer protein (CETP). The consequences of this exchange for atherosclerosis are complex. Most of the cholesteryl ester delivered to VLDL in this manner ultimately returns to the liver in remnant particles after VLDL triglyceride has been hydrolyzed by lipoprotein lipase. This is part of the reverse cholesterol transport pathway, and thus, is regarded as a favorable

Figure 3



Insulin resistance and dyslipidemia. When insulin resistance (bar) blocks the inhibitory effect of insulin on adipocyte lipolysis, adipocytes increase the release of FFAs, which are transported to the liver. The liver incorporates FFAs into TG, which is assembled into VLDL. Via the action of CETP, VLDL exchanges TG for cholesteryl ester from low-density lipoproteins and high-density lipoproteins. Lipases then act on TG-rich LDL to create SD LDL. Apolipoprotein A-1 is lost from TG-rich HDL in the kidney and at other sites, thus, reducing available HDL. This series of events produces the atherogenic lipoprotein phenotype of high TGs, low HDL, and small dense LDL. FFA indicates free fatty acid; SD LDL, small dense LDL; and TG, triglyceride. Adapted from *J Clin Invest.* 2000;106:453.

effect of CETP. However, some portion of cholesteryl ester in remnant particles may end up in the arterial wall—a proatherogenic effect.

Other major consequences of CETP activity in metabolic syndrome relate to triglyceride enrichment of HDL and LDL. When enriched in triglyceride, both of these lipoproteins are subject to lipolysis by hepatic lipase. Upon lipolysis, the HDL and LDL become smaller. The lipolyzed HDL are cleared more rapidly from the circulation, resulting in reduced HDL cholesterol and apolipoprotein A-I (apoA-I) concentrations. Apolipoprotein A-I and HDL (containing apoA-I) are antiatherogenic by participating in reverse cholesterol transport and probably by antioxidant mechanisms. Therefore, a metabolic disturbance that begins with increased VLDL triglyceride leads to an atherogenic reduction of HDL.

Triglyceride-enriched LDL is also lipolyzed and becomes smaller. Although all LDLs are atherogenic, small dense LDL are more atherogenic, perhaps due to increased penetration of the arterial intima, decreased antioxidant capacity, or other properties. For any given LDL cholesterol level, small dense LDL will include a greater number of particles, and therefore, a higher apoB concentration. The question arises as to whether an increase in LDL cholesterol is benign, as long as apoB and LDL particle numbers remain the same. Although results do not allow for a definitive answer at this time, most investigators in the lipid field would suggest that cholesterol matters and that such an increase in LDL cholesterol should not be regarded as benign.

Increased free fatty acid release from adipocytes and enhanced triglyceride synthesis in the liver are highlighted here as key steps in the genesis of lipid

abnormalities in metabolic syndrome. An important therapeutic goal is to sensitize adipocytes to insulin, thereby improving their ability to retain triglyceride. This shifts triglyceride and atherogenic lipoproteins from pathologic sites—that is, muscle, liver, arterial wall—to “safer” storage depots.

Many current studies are focusing on HDL and reverse cholesterol transport. “Nascent HDL” or pre- β -HDL is a lipid-poor apoA-I/phospholipid assembly secreted by the liver, but may also be formed in peripheral tissues from spherical HDL. Peripheral tissue cells deliver cholesterol to nascent HDL via the ATP-binding cassette-A1 (ABC-A1) membrane transporter. The cholesterol is subsequently esterified by lecithin-cholesterol acyltransferase, which aids in the formation of mature spherical HDL particles. Cholesteryl ester transfer protein then functions to transfer cholesteryl ester from HDL into VLDL and LDL, as described earlier, facilitating transport back to the liver. ABC-A1 is mutated in Tangier disease.³⁹ Both Tangier disease and recessive loss-of-function mutations of lecithin-cholesterol acyltransferase result in decreased reverse cholesterol transport and decreased ability to form stable, mature, and spherical HDL particles. Heterozygous ABC-A1 deficiency leads to low HDL cholesterol concentrations, but is not responsible for most cases of low HDL. The implications of metabolic syndrome for this system of reverse cholesterol transport have yet to be fully investigated.

Hypertension

Elevated blood pressure is included in most definitions of metabolic syndrome, but its relation to the syndrome

is complex. Multivariate techniques such as factor analysis, reviewed earlier, show that hypertension tends to segregate independently of other variables in metabolic syndrome. Nevertheless, most experts agree that hypertension should remain one of the options in diagnostic criteria. Key points of the argument include the clear correlation between hypertension and body weight, the increased incidence of hypertension in diabetic patients,⁴⁰ the negative correlation between insulin sensitivity and hypertension (although at relatively low loading in multivariate analysis),⁴¹ and the interactive role of hypertension and other factors in atherosclerotic risk.⁴² The last point—the interactive role in atherogenesis—would not be sufficient on its own, because LDL cholesterol also synergistically increases atherosclerotic risk but largely lacks the other correlations and is not considered part of metabolic syndrome.

The kidney plays the key role in controlling blood pressure, notably through pressure natriuresis.⁴³ Angiotensin and other neurohormonal mediators of hypertension have effects on intrarenal hemodynamics and renal electrolyte handling, and these factors determine long-term blood pressure control. Increased adipocyte mass leads to increased angiotensinogen production in adipose tissue, providing a potential mechanism for increased blood pressure.⁴⁴ Adipocytes also make angiotensin-converting enzyme and cathepsins, which effect local angiotensin conversion and catabolism, respectively. Free fatty acids were shown to increase angiotensinogen production by arterial wall cells in culture. Fatty acids may promote oxidative stress in endothelial cells, an effect that is amplified by angiotensin.⁴⁴

In a study of the relation among obesity, hypertension, and free fatty acids, 17 abdominally obese individuals with and without hypertension were evaluated by use of the euglycemic clamp technique. Fatty acid levels and turnover were much more resistant to suppression by insulin in obese hypertensive subjects vs lean or obese normotensive subjects. Correlations between fatty acids and blood pressure were also evident in the course of an insulin tolerance test in 30 additional subjects with a wide range of cardiovascular risk factors.⁴⁵ In hypertensive subjects with abdominal obesity, administration of an angiotensin-converting enzyme inhibitor not only lowered blood pressure, but also improved the ability of insulin to reduce plasma free fatty acid levels. Thus, hypertension appears more closely related to impairment of insulin action on free fatty acids vs on glucose.⁴⁴

Among patients with hypertension, approximately half are insulin resistant. Insulin resistance might also increase blood pressure via reduced nitric oxide-mediated vasodilation, increased salt sensitivity, or plasma volume expansion. These mechanisms might also underlie the decreased response to antihypertensive therapy seen in insulin-resistant individuals.⁴⁶⁻⁴⁸

Hypertension is a therapeutic target for angiotensin-converting enzyme inhibitors and angiotensin receptor blockers. The role of these agents may extend to metabolic syndrome. The Heart Outcomes Prevention Evaluation showed a decrease in new cases of self-reported diabetes in patients assigned to ramipril.⁴⁹ In the Losartan Intervention for End Point Reduction in Hypertension trial, prevention of diabetes was confirmed in patients assigned to losartan. Investigators hypothesized that effects of losartan on free fatty acids might account for this beneficial effect.^{50,51} Another theory is that modulation of the renin-angiotensin system may dampen oxidative stress, resulting in either improved insulin resistance or better maintenance of beta cell reserve.^{44,52}

Microalbuminuria

Microalbuminuria is defined as urinary albumin above the normal excretion rate and in the range of 30 to 300 mg per 24 h. Because the renal glomeruli contain vascular endothelial cells, as well as modified vascular smooth muscle cells known as mesangial cells (similar to cells in the vascular wall), the hypothesis has arisen that glomerular function (in particular, the function of excluding albumin rigorously from excreted urine) might mirror endothelial function and predict vascular disease. Microalbuminuria is a risk factor for coronary artery disease and progressive renal disease in patients with diabetes mellitus.⁵³ Some correlations have been shown between microalbuminuria and indicators of cardiovascular risk in nondiabetic patients as well,⁵⁴⁻⁵⁶ but conflicting data have also been reported.⁵⁷ These studies may be summarized by saying that insulin resistance is common among people with microalbuminuria, but that microalbuminuria is rare among all subjects with insulin resistance. Available data for nondiabetic normotensive patients do not support microalbuminuria as a risk factor for coronary artery disease or as a significant component of metabolic syndrome.⁵⁷

Inflammation and coagulation

In recent years, markers of systemic inflammation and certain components of the hemostatic system have been found to predict atherosclerotic risk. Some of these factors are associated with insulin resistance or other components of metabolic syndrome. C-reactive protein, when measured by highly sensitive immunoassay, has emerged as a very strong atherosclerotic risk factor comparable in predictive power to the total/HDL cholesterol ratio.⁵⁸ C-reactive protein was found to be independently associated with body weight (BMI), insulin resistance, and systolic blood pressure in the multiethnic Insulin Resistance Atherosclerosis Study.⁵⁹ White blood cell count and fibrinogen were also

associated with fatness and insulin resistance, though not as strongly.

The hemostatic components most prominently associated with atherosclerotic risk are fibrinogen and PAI-1. These were among 11 hemostatic and 10 metabolic variables included in a factor analysis performed with data from 322 nondiabetic elderly subjects in the Cardiovascular Health Study.⁶⁰ The analysis identified 7 uncorrelated factors, characterized as body mass, blood pressure, insulin/glucose, lipids, procoagulation, inflammation, and vitamin K-dependent protein factors. Interestingly, PAI-1 loaded with the body mass factor and partially with the insulin/glucose factor, but minimally with other factors. Fibrinogen, CRP, coagulation factors VIII and IX, plasma- α 2-antiplasmin, and fibrin D-dimer fragment contributed to a highly intercorrelated “inflammation” factor in this study.

Among 1047 nondiabetic subjects in the Insulin Resistance Atherosclerosis Study, 144 had new-onset diabetes over a 5-y period. Baseline CRP, fibrinogen, and PAI-1 were all significantly associated with risk of converting to diabetes. The effect of PAI-1 was independent of BMI, insulin resistance, and other known risk factors for diabetes.⁶¹ Thus, chronic inflammation, especially as assessed by PAI-1, may be a risk factor for the development of type 2 diabetes mellitus.

Labels such as “inflammatory” or “hemostatic” tend to be applied indiscriminately to various bioactive molecules. In some ways, this is appropriate. Just as cell biologists have learned that the name initially given to a cytokine may vastly understate its biological activity (TNF is an example), we should be careful about sorting molecules into predefined categories. What actually exists is a complex interactive system of biochemical pathways, biophysical forces, and patterns of gene expression, and ultimately, it will be necessary to learn both the molecular details and the whole system responses. Using names such as inflammation, hemostasis, insulin resistance, and metabolic syndrome is an attempt to reduce the entire system to humanly understandable subsystems, but the representation is necessarily imperfect.

Summary and conclusions

Metabolic syndrome is a highly prevalent but under-recognized and undertreated condition. The concurrence of abdominal obesity, borderline hyperglycemia, atherogenic lipoprotein phenotype, and/or hypertension in a patient constitutes a system of linked pathogenesis and high atherogenicity. The complex pathogenetic links involve glucose and fat metabolism, insulin, and a variety of adipocyte-derived hormones and cytokines.

Current understanding can be summarized as follows: insulin resistance, a central feature of metabolic syn-

drome, is specific for the PI-3K pathway of insulin signaling, whereas the MAP kinase pathway of insulin signaling may be overstimulated. Dysfunctional energy balance, characterized by accumulating triglyceride from food energy intake exceeding energy output, overwhelms appropriate storage available in small peripheral adipocytes, resulting in increased concentrations of free fatty acid in plasma as well as dysregulation of adipocyte endocrine and paracrine function. Plasma free fatty acids and cytoplasmic triglyceride in muscle suppress insulin-stimulated glucose uptake, whereas insulin resistance in adipocytes leads to insuppressible fatty acid release. The atherogenic lipoprotein phenotype of high triglycerides, low HDL, and small dense LDL results primarily from increased flux of free fatty acids from adipose tissue to liver. Hypertension is linked to this disordered system, in part, via angiotensinogen synthesized by adipocytes and via effects of free fatty acids on central organs and blood vessels.

Energy metabolism in human beings is adapted by evolution for efficient conversion of food energy to muscular effort and for survival in times of food energy deprivation. Metabolic syndrome embraces multiple interactive systems that are maladaptive in the face of food energy abundance and low levels of muscular activity. In this first part of the meeting summary, we have reviewed the current understanding of the pathogenesis of metabolic syndrome. In the second part, we will summarize the convergence of various components that promote the atherogenesis, the current status of therapeutic and pharmaceutical endeavors, and the emerging clinical research goals that will define success in treating metabolic syndrome.

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