40GENETIC SUCCEPTIBILITY TO METABOLIC SYNDROME

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1 . Introduction

The first recognition of clustering of hypertension, hyperglycemia and gout came already in twenties of 20th century . In 1988 Reaven identified Syndrome X originating from insulin resistance (IR). In 2004 The National Cholesterol Education Program's Adult Treatment Panel III (ATPIII) defined the metabolic syndrome with alternative names: IR syndrome, Reaven syndrome, characterized by following components:

- insulin resistance
- abdominal obesity
- atherogenic dyslipidemia and cardiovascular disease
- hypertension
- proinfammatory state
- prothrombotic state

Diagnostic criteria for MS were prepared by ATPIII by International Diabetes Federation (IDF). Upon ATPIII agreement, patients having at least 3 of 5 characteristics can be diagnosed as having the metabolic syndrome: abdominal obesity, elevated triglycerides, decreased HDL-cholesterol, increased blood pressure or increased fasting plasma glucose. IDF declares criteria fairly consistent with ATPIII. Slight differences include central obesity as the major feature and fulfilled two of four other manifestations. The borderline glucose concentration is lower in IDP criteria, with strong recommendation for OGTT when exceeded.

Three possible etiologies for the metabolic syndrome were postulated: Obesity was found to be responsible for excess release of free fatty acids, cytokines and other proinflammatory products which are implicated in the development of IR, hypertension and dyslipidemia.

IR as the second possible cause of metabolic syndrome rises a question whether it is possible to dissociate between obesity and IR. Indeed, IR exists to various degrees in all particular classes of body mass index, suggesting an independent inheritable contribution of it to at least some extent. Some populations (South Asians) with mild overweight display IR and this is said to be primary IR. From this point of view IR can be classified as a separate etiological factor for metabolic syndrome. Hyperinsulinemia as a consequence of IR is capable to increase VLDL secretion from the liver and cause hypertension. IR of muscle can cause hyperglycaemia, exaggerated with gluconeogenesis in insulin-resistant liver.

The third etiology is thought to include independent factors: immunologic, vascular, hepatic, which are influenced by specific genetic and environmental factors.

2.   Risk factors for metabolic syndrome

The following factors increase chances of having metabolic syndrome:

- *Age*. The prevalence of metabolic syndrome increases with age, affecting less than 10 percent of people in their 20s and 40 percent of people in their 60s. However, one study shows that about one in eight schoolchildren have three or more components of metabolic syndrome.
- *Race*. Hispanics and Asians seem to be at greater risk for metabolic syndrome than other races are.
- *Progressive weight gain*. Metabolic syndrome is present in about 5% of people with normal body weight, 22% of those who are overweight and 60% of those considered obese. Adults who continue to gain 5 or more pounds per year raise their risk of developing metabolic syndrome by up to 45%.
- *Obesity*. A body mass index (BMI) - a measure of your percentage of body fat based on height and weight - greater than 25 increases your risk of metabolic syndrome. So does abdominal obesity - having an apple shape rather than a pear shape.
- *History of diabetes*. You're more likely to have metabolic syndrome if you have a family history of type 2 diabetes or a history of diabetes during pregnancy (gestational diabetes).
- *Other diseases*. A diagnosis of high blood pressure, cardiovascular disease or polycystic ovary syndrome - a similar type of metabolic problem that affects a woman's hormones and reproductive system - also increases the risk of metabolic syndrome.
- *Low physical activity*. A sustainable exercise program, fore example 30 minutes 5 days a week is reasonable to start, providing there is no medical contraindication. (If you have any special concerns in this regard, check with your doctor first.) There is a beneficial effect of exercise on blood pressure, cholesterol levels, and insulin sensitivity, regardless of whether weight loss is achieved or not. Thus, exercise in itself is a helpful tool in treating metabolic syndrome.
- *Diet*. A detailed discussion of diet therapies, pros and cons of various diets etc. is beyond the scope of this article. However, there is now a trend toward the use of a Mediterranean diet -- one that is rich in "good" fats (olive oil) and contains a reasonable amount of carbohydrates and proteins (such as from fish and chicken). Mediterranean diet: A diet traditionally followed in Greece, Crete, southern France, and parts of Italy that emphasizes fruits and vegetables, nuts, grains, olive oil (as opposed to butter) and grilled or steamed chicken and seafood (as opposed to red meat). Plus a glass or two of red wine.
- *Lifestyle*: sedentary work, smoking, eating an excessively high carbohydrate diet, and consuming an alcohol-free diet.
- *Genetic factors*

Diet and exercise are still the preferred primary treatment of metabolic syndrome.

3.   Genetic risk factors for metabolic syndrome

Genetic factors could influence each individual component of the syndrome, and the syndrome itself. A family history that includes obesity, type 2 diabetes and/or insulin resistance greatly increases the chance that an individual will develop the metabolic syndrome. However there are some genetic loci, which are in linkage disequilibrium with metabolic syndrome.

1.   Genetics of metabolic syndrome

Kissebah et al. (2000) performed a genomewide scan by use of a 10-cM map in 2,209 individuals distributed over 507 nuclear Caucasian families and for the first identifying major genetic loci influencing the metabolic syndrome phenotypes. They showed a quantitative trait locus (QTL) on chromosome 3q27 strongly linked to 6 traits: weight, waist circumference, leptin, insulin, insulin/glucose ratio, and hip circumference (lod scores ranging from 2.4 to 3.5). A second QTL was found on chromosome 17p12 and was strongly linked to plasma leptin levels (lod = 5.0). Several candidate genes are located in both regions (Table 2.1.).

McCarthy and coworkers (2003) studied 207 SNPs in 110 candidate genes among coronary artery disease patients, a population enriched for metabolic abnormalities. The number of abnormalities (0 to 5) was determined in 214 male and 91 female patients, and the association with each polymorphism was evaluated. Polymorphisms in 8 genes were associated with metabolic syndrome in the whole population (P values ranging from 0.047 to 0.008): LDLR, GBE1, IL1R1, TGFB1, IL6, COL5A2, SELE) and LIPC. Variants in 7 additional genes showed significant gene interaction by gender. Separate analyses in men and women revealed a strong association with a silent polymorphism in the gene encoding low density lipoprotein receptor-related protein-associated protein-1 (LRPAP1) among females (P = 0.0003), but not males (P = 0.292). Several other genes showed association only in females; only 1 gene, PRCP, was significantly associated in men alone (P = 0.039).

Based on results of both genome wide studies, the genetic association studies with metabolic syndrome of genes listed in Table 2.1. are highly recommended.

Table 2.1. Candidate genes for genetic association studies with metabolic syndrome

LOW DENSITY LIPOPROTEIN RECEPTOR (LDLR) gene. The low density lipoprotein receptor is a cell surface receptor that plays an important role in cholesterol homeostasis. Mutations in this gene are associated with familiarly hypercholesterolemia.
GLYCOGEN BRANCHING ENZYME (GBE1) gene. The GBE1 gene encodes the glycogen branching enzyme (EC 2.4.1.18), which is involved in glycogen synthesis. Branching of the glycogen chains is essential to pack a very large number of glycosyl units into a relatively soluble spherical molecule of glycogen.
INTERLEUKIN 1 RECEPTOR, TYPE I (IL1R1) gene. Interleukin-1 consists of 2 separate but related proteins, IL1-alpha and IL1-beta. Both contain a single membrane-spanning segment, a large cytoplasmic region, and an extracellular domain. IL 1 is one of mediators in inflammation.
TRANSFORMING GROWTH FACTOR, BETA-1 (TGFB) gene encodes the multifunctional peptide that controls proliferation, differentiation, and other functions in many cell types. TGFB acts synergistically with TGFA in inducing transformation. It also acts as a negative autocrine growth factor. Dysregulation of TGFB activation and signaling may result in apoptosis.
INTERLEUKIN 6 (IL6) gene. IL6 is an immunoregulatory cytokine that activates a cell-surface signaling assembly composed of IL6, IL6RA, and the shared signaling receptor gp130. The aberrant production of IL6 by neoplastic cells has been implicated as a strong contributory factor to the growth of multiple myeloma and other B-cell dyscrasias, T-cell lymphoma, renal and ovarian cell carcinomas, and Kaposi sarcoma demonstrated repression of the IL6 gene promoter by p53. IL6 gene is one of the candidate genes for linkage studies of osteopenia and osteoporosis because the gene product stimulates osteoclasts through binding to its cell surface receptor (IL6R).
COLLAGEN, TYPE V, ALPHA-2 (COL5A2) gene.
SELECTIN E (SELE) gene. Endothelial leukocyte adhesion molecule-1 is expressed by cytokine-stimulated endothelial cells. It is thought to be responsible for the accumulation of blood leukocytes at sites of inflammation by mediating the adhesion of cells to the vascular lining.
HEPATIC LIPOPROTEIN LIPASE (LIPC) gene. Hepatic lipase, like lipoprotein lipase and

lecithin:cholesterol acyltransferase, plays a major role in the regulation of plasma lipids. Rare deficiencies of all of these enzymes have been identified in man, and all are associated with pathologic levels of circulating lipoprotein particles.

In addition, Robitaille study in 2004 found among 632 men increased frequency of the val162 allele of the leu162-to-val polymorphism in the PPARA gene, among those with abdominal obesity, hypertriglyceridemia, high plasma apoB and low HDL plasma levels, which are components of the metabolic syndrome. The frequency of the V162 allele was approximately 10% in their group.

Animal models for metabolic syndrome studies

For studies of metabolic syndrome two of animal models were developed: a transgenic mice with 11-beta-hydroxysteroid dehydrogenase type 1 overexpressing and the Neil1 knockout mice.

The transgenic mice overexpressing 11-beta-hydroxysteroid dehydrogenase type 1 selectively in adipose tissue was to an extent similar to that found in adipose tissue from obese humans. These mice had increased adipose levels of corticosterone and developed visceral obesity that was exaggerated by a high-fat diet. The mice also exhibited pronounced insulin-resistant diabetes, hyperlipidemia, and, surprisingly, hyperphagia despite hyperleptinemia. Increased adipocyte 11-beta-hydroxysteroid type 1 activity may be a common molecular etiology for visceral obesity and the metabolic syndrome.

Vartanian et al. (2006) found that Neil1 knockout mice were born at expected mendelian ratios and the phenotype of Neil1 -/- pups was normal through the first 4 to 6 months of life. At about 7 months, however, male Neil1 -/- mice developed severe obesity, and female Neil1 -/- mice were modestly overweight. Mutant mice also showed dyslipidemia, fatty liver disease, and a tendency to develop hyperinsulinemia, similar to metabolic syndrome in humans. Histologic studies showed significant kidney vacuolization, and mitochondrial DNA from Neil1 -/- mice showed increased levels of steady-state DNA damage and deletions, compared to wildtype controls.

2. Genetics of individual components of metabolic syndrome

Genetic factors could influence each of components in metabolic syndrome individualy. In following chapters the genetic factors in obesity and insulin resistance as the main causes of metabolic syndrome, will be rewieved in short.

1. *Genetics of obesity*

The state of nutrition is best described by body mass index (BMI) which is, with some exceptions, in good correlation with the amount of total body fat. According to the BMI, the following categories of excessive body mass or nutrition are postulated:

BMI 25-30 kg/m2   overweight
BMI 30-40 kg/m2   obesity
BMI 40-50 kg/m2   morbid obesity
BMI >50 kg/m2   extreme obesity

Pathophysiology of obesity

Each individual has genetically determined the weight set-point and hence body weight is tightly regulated by an energetic homeostatic mechanism. Adipocytes secrete leptin and β-cells secrete insulin, both in proportion to body-fat content. The two hormones enter the brain. They bind to their central receptors on the hypothalamic neurons exerting effects to reduce body weight. Hypothalamic nevrons express peptides and their receptors which could be categorized as orexigenic: neuropeptide Y, agouti-related protein (AgRP), melanin-concentrating hormone (MCH), orexins A and B; or anorexigenic: melanocortins (i.e. melanocyte-stimulating hormone, α-MSH) and cocaine and amphetamine related transcript (CART). In leptin or insulin abundance anorexigenic pathways prevail: increase of energy expenditure, increase of thermogenesis, diminished food intake. Particularly leptin-melanocortin anorexigenic signalling pathway appears to be very conserved among species and mutations in genes encoding for components of this pathway: leptin, leptin receptor, pro-opiomelanocortin (POMC), prohormone-convertase 1 (PC1), and melanocortin 4 receptor (Mc4R), cause rare forms of morbid monogenic obesity and lead to some naturally occuring murine models of obesity (ob, db, Ay and mg;). On the contrary, knockouts in genes for orexigenic pathways in mice fail to produce lean fenotypes, which demonstrates extremely powerful mutual effects of anabolism and weight gain system components.

Leptin and insulin mediate long-term body-mass regulation. They are active also in short-term signals that effect single meal to be initiated and terminated. In addition, there are some other shortly acting hormones/factors which accompany food intake: ghrelin, motilin, neuromedin U, neurotensin, cholecystokinin, peptide YY_{3-36} (PYY; 72) and glucagon-like peptide-1, all secreted by the gastrointestinal tract, and vagal afferent signalling .

Genetics of obesity

Today's high incidence of obesity could be explained by a »thrifty genotype« hypothesis: over periods of time the alleles were selected which favored weight gain and fat storage in order to provide enough nutrients for times of food deprivation. In today's times of food availability and decreased physical activity such genotypes cause obesity.

Besides monogenic forms of obesity, there are at least 20 rare syndromes with obvious genetic basis, which appears to be more complex as it predisposes more dysfunctions (mental retardation, multiple signs of hypothalamic disorder).

The common human obesity is thought to be oligogenic state and its expression is modulated by multiple modifier genes and by environmental factors: food intake, physical activity, and smoking. Genetic basis in the pathophysiology of obesity is estimated to be 40-80%. At least 204 putative gene loci associated with obesity have been identified, and those, which have been confirmed by multiple studies, are presented in Table 2.2.

Table 2.2. A list of genes associated with obesity confirmed by 5 or more studies:

Gene name (accord. to HUGO nomenclature committee)	Protein name
ADIPOQ	adiponectin
ADRA2A	adrenergic receptor α-2A
ADRA2B	adrenergic receptor α-2B
ADRB1	adrenergic receptor β-1
ADRB2	adrenergic receptor β-2
ADRB3	adrenergic receptor β-3
DRD2	dopamine receptor D2
LEP	leptin
LEPR	leptin receptor
NR3C1	nuclear receptor subfamily 3, group C, member 1
PPARG	PPAR-γ
UCP1	uncoupling protein 1
UCP2	uncoupling protein 2
UCP3	uncoupling protein 3
TNF	TNF-α
LIPE	hormone sensitive lipase

2. Genetics of insulin resistance and diabetes mellitus type 2

Insulin resistance (IR) is defined as less than normal response to insulin, which leads to hyperinsulinemia for euglycaema to be maintained. The hyperinsulinemia causes disinhibition of gluconeogenesis, impaired uptake of glucose by muscle and disinhibition of lipolysis in adipose tissue. Clinical markers of IR are visceral obesity, acantosis nigricans, acne, hirsutism, hepatic steatosis.

Etiology of IR includes genetic factors resulting in syndromic forms of IR, and environmental factors: food intake, poor physical activity, aging, smoking or administration of drugs – thiazide diuretics, beta-adrenergic antagonists, glucocorticoids, which can cause or contribute to IR. The most important factor is obesity, which is usually of combined polygenetic and environmental origin. Abdominal adipose tissue is a source of free fatty acids and various hormones (adipokines) being implicated in the development of IR.

Genetic defects in insulin signalling
A minority of insulin resistant cases is characterized by a single genetic or acquired trait. Anti-insulin autoantibodies have been found in diabetes mellitus type 1. On the other hand, more than 60 mutations have been identified in the insulin receptor gene. Among them type A IR is associated with heterozygous mutation state which underlies decreased Tyr phosphorylation of the β-subunit after insulin binding. In Rabson-Mendenhall syndrome and leprechaunism, (Donohue syndrome) insulin receptor gene mutations are presumed to impair

insulin binding to the receptor. IR may be due to abnormal production of anti-insulin-receptor antibodies (type B IR). PPAR-γ mutations, which are not associated with lipodystrophy, are also reported to cause IR.

Genetics of DM2

Affected genes in monogenic forms of diabetes, insulin resistance and lipodystrophy represent an excellent base for the search of susceptibility genes for polygenic multifactorial DM2, although the latter can be distinguished from monogenic Mendelian diseases. Namely, on certain genetic backgrounds, with particular gene interaction – epistasis and with certain envinronment influence the same genes could contribute to DM2.

Maturity–onset diabetes of the young (MODY) is a monogenic from of diabetes and exists in 6 forms due to 6 affected MODY genes. From them, *HNF4A*, *TCF1* (or *HNF1A*) and *GCK* genes which encode for two transcriptional factors and glucokinase in the β-cells, respectively, were reliably proved to be involved in DM2.

From the genes responsible for monogenic form of insulin resistance, the gene for insulin with class III variable number tandem repeat alleles and the *PPARG* gene have been associated with multifactorial DM2 as well. Additionally, the *AGPAT2* (acylglycerolphosphate-acyltransferase) gene underlying Berardinelli-Seip congenital lipodystrophy and *Mt-tRNA Leu(UUR)* mitochondrial gene otherwise associated with maternally inherited diabetes and deafness (MIDD) syndrome have been shown to be implicated in DM2 .

Search for susceptibility genes among the genes dealing with insulin secretion and action gave positive results for already mentioned *PPARG* gene with another polymorphism P12A (different than the one implicated in monogenic IR) and *KCNJ11* gene, encoding for protein contained in K^+_{ATP} channels. Arising but not yet definite evidence for implication in DM2 concerns *IRS-1*, *GLUT2* and *PGC1A* (co-activator of PPAR-γ) genes.

The only proved DM2 susceptibility gene found through genome wide-scan methods is *CPN10* which encodes for calpain-10 from the familiy of calcium-activated neutral proteases, expressed in β-cells. This gene was identified after strong linkage disequilibrium found with DM2, which was followed by positional cloning.

When studying the importance of particular susceptibility genes one must bear in mind that their effect is modest. The listed susceptibility genes does contribute to the basis for future diagnosis, prognosis and therapy, but for now, the mutual influences of various gene loci and interactions of genes with dietary and other lifestyle factors remain to be exactly determined and quantitated.

Recommended literature:

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