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Preface

The Emerging Epidemic of Obesity and Cardiometabolic Risk Factors

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The world is witnessing an obesity pandemic that threatens to pose serious challenge to every country's health system. The projected burden of an enormous continuing increase in the prevalence of atheroscelerotic cardiovascular disease, type 2 diabetes, hypertension, stroke and some forms of cancer will not only add to morbidity and mortality but would result in tremendous increase in health care expenditure. Obesity is also associated with an insidious, creeping increase in hitherto uncommon diseases such as non-alcoholic fatty liver disease and polycystic ovaries syndrome. A cluster of cardiometabolic risk factors has been described in association with obesity. These factors, both individually and collectively, enhance the risk of above mentioned diseases.

Amongst the most worrying problems is a steep rise in the prevalence of obesity among children which makes them much more prone to chronic diseases as they grow older, thereby curtailing both the longevity as well as quality of life(1). As Prof. Kate Steinbeck pointed out at the recently held 10th International Congress on Obesity in Sydney : ***'The children in this generation may be the first in history to die before their parents because of health problems related to weight'***. This phenomenon is already well recognized in developing countries with high infant mortality due to malnutrition, infections and infestations. Nevertheless, with epidimelogical and demographic transition, the world now has more fat people than hungry ones, with more than a billion overweight people compared to 800 million who are undernourished. Developing nations are facing a 'Double Trouble Syndrome'(2), with unhealthy coexistence of both communicable and non-communicable diseases , alongwith the spectre of HIV looming large on the horizon.

It is therefore timely to give serious thought to preventive aspects especially with regard to childhood obesity(3). It is being increasingly recognized that the environment of the baby and growing child plays a significant role in the development of obesity in childhood and adolescence.

The warning signs identified for enhancing obesity risk include watching more than eight hours of television a week; sleeping fewer than 10.5 hours each night; above average birth weight; obesity in both parents; excessive weight gain in the first year of life and rapid growth between birth and the age of two years.

Taking serious note of these trends in health transition, the CME Committee of NAMS in 2005 decided to organize an intra-mural national symposium devoted to Obesity and the related cardiovascular & metabolic risk factors. We were assigned the task of planning and organization of the symposium which was held at the PGIMER, Chandigarh on November 20, 2005. It required enormous preparatory efforts to ensure wider academic interaction and dissemination of information. Thus the deliberations of the Symposium were transmitted live to postgraduate residents and the Faculty at the Indira Gandhi Medical College, Shimla. This was done with the aid of PGIMER's Telemedicine Centre and the Centre for Development of Advance Computing, Mohali. The Real Time Video Conferencing was done successfully through the 3 BRI ISDN (384 Kbps Integrated Services Digital Network) lines with I.G. Medical College Shimla. The clarity and connectivity was excellent and the participants at Shimla Medical College were gratefully appreciative of such an academic interaction. The extensive coverage in the print and electronic media successfully disseminated health information about the subject content of Obesity to a wide readership and enlightened community leadership. The successful outcome and impact was therefore most gratifying.

In 2005, NAMS also established and constituted a standing Academic Committee with its objectives focused on enhancing quality of health care through continuing professional development, medical education and biomedical research. In its first meeting, the Academic Committee decided to develop National Guidelines in selected areas of health concerns. One of the topics selected was 'Metabolic Syndrome and Management of Risk Factors such as Dyslipidemia'. It has now been decided that the papers presented at the National Symposium held at Chandigarh should be published as a part of CME Monograph series, and that this publication may constitute an important background document, amongst others, to be deliberated upon by the Expert Group being convened in early 2007 for developing and recommending National guidelines regarding the Diagnosis and Management of Obesity and associated Cardiometabolic Risk Factors. Thus, the present monograph not only reflects the outcome of 2005 NAMS National Symposium, but also projects the futuristic development of background documentation for the 2007 NAMS Expert Group meeting.

We gratefully appreciate the cooperation extended by all those who were, and continue to be, deeply involved in these endeavours.

References :

1. *Rodriguez BL, Fujimoto WY, Mayer-Davis EJ, Imperatore G, Williams DE, Bell RA, Wadwa RP, Palla SL, Liu LL, Kershnar A, Daniels SR, Linder B, and for the SEARCH for Diabetes in Youth Study Group. Prevalence of Cardiovascular Disease Research Factors in U.S. Children and Adolescents With Diabetes: The SEARCH for Diabetes in Youth Study. Diabetes Care. 2006; **29**:1891-1896.*
2. *Bajaj JS. Diabetes care for the millions : A challenge in the next millennium. In : Diabetes 1994. Eds. S. Baba, T. Kaneko, Elsevier, Amsterdam, New York, Oxford, P. 95-102.*
3. *Lopez-Capape M, Alonso M, Colino E, Mustieles C, Corbaton J, Barrio J. Frequency of the metabolic syndrome in obese Spanish pediatric population. Eur. J. Endocrinol. 2006; **155**:313-319*

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Editorial

Metabolic Syndrome
From Inert Facts to Informed Action

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With due apologies to Charles Dickens, the recently generated intense debate on the Metabolic Syndrome (MS), may well be captioned as "A Tale of Two Worlds". The debate is essentially centered around *Egomantics*, a somewhat catastrophic combination of semantics *and* superego. While my distinguished colleagues at the International Diabetes Federation (IDF) concede that MS is 'perhaps an etiologic mystery but far from a myth'(1), the leadership of the American Diabetes Association (ADA) and European Association for the Study of Diabetes (EASD) insist that 'the MS has been imprecisely defined, there is a lack of certainty regarding its pathogenesis, and there is considerable doubt regarding its value as a cardiovascular disease risk marker' (2). Nevertheless, they also grudgingly admit that 'the term MS has now taken hold in the medical literature. It has been defined and institutionalized, principally by the World Health Organization (WHO) (3) and the Third Report of the National Cholesterol Education Program's Adult Treatment Panel (NCEP-ATP III) (4) albeit with different definitions. In addition, other organizations have developed similar, but again not identical, definitions (5,6). The fact that a version of the MS has its own ICD-9 code (277.7) also suggests that it is well thought out'.

Responding (or perhaps reacting) to what they call *'shot across the bow of the MS'*, the gallant knight(s), presumably representing the viewpoint of International Diabetes Federation, state (1) : 'we recognize the importance of debate : however, the appearance of this initiative on behalf of two of the world's leading regional diabetes organizations raises questions of motive and timing. Are the criticisms part of a "turf protection" scenario or do they have a valid scientific basis?'. This logic reflects the assumption

that the view of the whole i.e. IDF must have a greater credence and credibility than that of its individual constituents i.e. ADA and EASD. Almost like the logic behind MS and its constituents!

Essentially, the scientific argument of the protagonists is based on the facts that : (i) MS represents a risk-factor clustering that increases the "global risk" for atherosclerotic cardiovascular disease (ACVD) (7,8); (ii) the constituent risk factors appear to be of metabolic origin; (iii) the seemingly apparent contradictions between different definitions of MS result from the 'healthy process of evolution' (9); (iv) regardless of the precise metabolic pathways involved, both central obesity and insulin resistance are common risk conditions underlying the MS (7); (v) for a condition such as the MS in which there is a clustering of risk factors, precise clinical criteria are difficult to propose; (vi) proposed thresholds of the risk factors by necessity are arbitrary, but in the ATP III and IDF clinical definitions, their use has the virtue of simplicity; (vii) as current thresholds employed for clinical diagnosis are based on contemporary recommendations of accepted expert panels, Kahn and colleagues (i.e. opponents) are stretching credibility with the claim that the current clinical definition of the MS is imprecise; (viii) as a general rule, the risk from MS for major ACVD events is approximately twice as high as for those without the syndrome (10). For type 2 diabetes, MS confers an approximate 5-fold greater risk (10); (ix) it cannot be overemphasized that MS is not an *absolute risk predictor.*

Finally, the protagonists tend to close the argument for their forceful defense of MS by stating : 'the ADA/EASD attempt to disregard the MS will only confuse health professionals at all levels. The utility of the syndrome as a public health initiative has been put at risk by a statement that has "come out of the blue" and does not reflect the *past intellectual and constructive contributions of some of the individual authors!*' What a pity that an academic and scientific debate is closed by questioning the past intellectual and constructive contributions of some of the opponents!

The last statement was provocative enough to invite a befitting retort from Richard Kahn, the first author of the ADA/ EASD joint statement. In his article 'The Metabolic Syndrome (Emperor) wears no clothes (11)', Kahn laments the fact that despite over 10,000 articles on the subject, there is much missing information. He raises the critical question : 'whether we know enough about this constellation of risk factors to warrant adopting a unique clinical construct that has value to either physicians and / or patients?' He sums up the basic issues in support of his line of argument : (i) there is no biological basis for the diagnostic algorithm ; (ii) with many revisions of the diagnostic algorithm, there is a lack of specific biological evidence that warrants the change and / or clarifies as to

how the new/revised definition enhances the sensitivity, specificity, and positive predictive value of the diagnosis; (iv) MS, as defined, is a relatively insensitive indicator of insulin resistance (12,13); (v) if the syndrome includes other 'prothrombotic and proinflammatory states' or factors, why are there no such criteria in the definition?, and finally (vi) since all of the syndrome variables have no upper cut-off limits, many individuals will be so diagnosed because they have overt diabetes, hypertension, or severe lipid abnormalities.

The proponents, in a recent publication (14), draw support from the scientific statement published by American Heart Association and National Heart, Lung, and Blood Institute, endorsing essentially the concept and definition of MS (15,16). In a follow-up of this statement, Grundy reinforces the proponent's arguments and states (17) : 'Five risk factors of metabolic origin (atherogenic dyslipidemia, elevated blood pressure, elevated glucose, a prothrombotic state, and a proinflammatory state) commonly cluster together. This aggregation is frequently observed in clinical practice, and it has been convincingly documented in prospective studies by cluster analyses. Risk factor clustering cannot be explained by chance occurrence alone. Thus, if the metabolic syndrome is defined as multiple risk factors that are metabolically interrelated, then the syndrome certainly exists.' The discerning readers will readily notice the inclusion of 'a prothrombotic state, and a proinflammatory state' amongst the cluster of risk factors while the proponents donot include these in the 'international consensus'.

In this cacophony of arguments and counterarguments, what seems to have been forgotten (especially by those who live near Stratford-upon-Avon) is the axiom : *'you cannot stage Hamlet without the Prince of Denmark'*. Both the proponents and opponents cite the 'seminal' work of Gerald Reaven who, in his Banting Lecture in 1988, provided the conceptual framework of clustering of metabolic risk factors, suggesting the term 'Syndrome X' (18). His peers honoured him by naming the syndrome as Reaven's Syndrome. When the scientific level of current debate was reduced to questioning the *past intellectual and constructive contributions* of some of the individual authors of ADA/EASD statement, the Oracle finally spoke. In a thought provoking article : 'The Metabolic Syndrome : is this diagnosis necessary' (19), Gerald Reaven has imparted poise and equanimity to the raging debate. In his masterly review, Reaven concludes : '(i) providers should avoid labeling patients with the term metabolic syndrome ; (ii) adults with any major CVD risk factor should be evaluated for the presence of other CVD risk factors; and (iii) all CVD risk factors should be individually and aggressively treated. If these goals are achieved, there is no longer a need for a diagnosis of metabolic syndrome, a controversy about the best definition of the

metabolic syndrome, or any confusion as to the clinical approach to patients who, although they are at greater risk of CVD, do not qualify for a diagnosis of metabolic syndrome.'

Such an approach finds a resounding echo in the recently published joint statement of American Diabetes Association and American Heart Association (20): 'Both the American Heart Association and the American Diabetes Association remain jointly committed to a reduction in heart disease, stroke, and new-onset diabetes. We strongly recommend that all providers assess patients for their global risk for CVD and diabetes. Despite many unresolved scientific issues, a number of *cardiometabolic risk factors* have been clearly shown to be closely related to diabetes and CVD : fasting / postprandial hyperglycemia, overweight/obesity, elevated systolic and diastolic blood pressure, and dyslipidemia. Although pharmacologic therapy is often indicated when overt disease is detected, in the early stages of these conditions, lifestyle modification with attention to weight loss and physical activity may well be sufficient.'

This clarion ***Call for Action*** is a pragmatic approach. While future research will continue to clarify MS-related issues, health care providers at all levels, from primary health care workers and primary care physicians to nurses, pharmacists, practitioners of Family Medicine, and finally upto and including specialists and tertiary care providers must join together to implement *'risk reduction'* approach. Health Policy Planners and Health Administrators must affect such changes not only in health policies but also in all related intersectoral policies such as agriculture, tobacco industry, information and broadcasting, and telecommunication to impart health-friendly orientation to all actions focused at 'risk reduction' and to educate communities regarding hazards of *coca-colonisation and McDonaldisation.*

If this debate results in the development and implementation of appropriate action plans as suggested above, and such plans *a priori* take into consideration the health needs and resources of each country, besides stimulating research efforts to provide the much needed information presently missing, it would have admirably served its purpose. It would certainly have the whole-hearted support of all of us, who have served the cause of primary diabetes care and of the delivery of preventive and promotive community health services to deprived population. Hopefully, such an appeal coming from one who has served and nurtured the International Diabetes Federation for nearly four decades, and has continuing admiration for the past and present leadership of both the IDF and ADA, may strike a sympathetic chord in those involved in this no holds-barred debate . Otherwise this appeal will remain, like that of exhortation by Reaven, a cry in the wilderness.

As Bertrand Russell in his ***Unpopular Essays*** states : *"for it is not enough to recognize that all our knowledge is, in a greater or lesser degree, uncertain and vague; it is necessary at the same time to learn to act upon the best hypothesis without dogmatically believing it".*

References :

1. Zimmet PZ, Alberti G (2005). The Metabolic Syndrome : Perhaps an Etiologic Mystery but Far From a Myth - Where Does the International Diabetes Federation Stand? *Medscape Diabetes & Endocrinology.* **7** (Posted 10/11/2005).
2. Kahn R, Buse J. Ferrannini E, Stern M (2005). The metabolic syndrome : time for a critical appraisal : joint statement from the American Diabetes Association and the European Association for the Study of Diabetes. *Diabetes Care.* **28** : 2289-2304.
3. World Health Organisation : Definition, Diagnosis, and Classification of Diabetes Mellitus and its Complications : Report of a WHO Consultation. Geneva, World Health Org., 1999.
4. Expert Panel on the Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (2001). Executive Summary of the Third Report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III) *JAMA.* **285**: 2486-2497.
5. Balkau B, Charles MA, Drivsholm T, Borch-Hohnsen K, Wareham N, Yudkin JS, Morris R, Zavaroni I, Van Dam R, Feskins E, Gabriel R, Diet M, Nilsson P, Hedblad B, European Group For The Study Of Insulin Resistance (EGIR) (2002). Frequency of the WHO metabolic syndrome in European cohorts, and an alternative definition of an insulin resistance syndrome. *Diabetes Metab.* **28** :364-376.
6. Einhorn D, Reaven GM, Cobin RH, Ford E, Ganda OP, Handelsman Y, Hellman R, Jellinger PS, Kendall D, Krauss RM, Neufeld ND, Petak SM, Rodbard HW, Seibel JA, Smith DA, Wilson PW (2003). American College of Endocrinology position statement on the insulin resistance syndrome. *Endocr Pract.* **9** :237-252.
7. Lemieux I, Pascot A, Couillard C et. al. (2000). Hypertriglyceridemic waist : a marker of the atherogenic metabolic triad (hyperinsulinemia ; hyperapolipoprotein B; small, dense LDL) in men ? *Circulation.* **102**: 179-184.
8. Grundy SM, Brewer HB Jr, Cleeman JI, Smith SC Jr, Lenfant C (2004). National Heart, Lung, and Blood Institute; American Heart Association. Definition of metabolic syndrome: report of the National Heart, Lung, and Blood Institute/American Heart Association conference on scientific issues related to definition. *Arterioscler Thromb Vasc Biol.* **24** : e13-e18.
9. Bloomgarden ZT (2003). The Endocrine Society meeting : topics in insulin sensitivity and hypertension. *Diabetes Care.* **26** : 2679-2688.
10. Eckel RH, Grundy SM, Zimmet PZ (2005). The metabolic syndrome. *Lancet.* **365** : 1415-1428.
11. Kahn R (2006). The Metabolic Syndrome (Emperor) Wears No Clothes. *Diabetes Care.* **29** : 1693-1696.
12. Cheal KL, Abasi F, Lamendola C, McLaughlin T, Reaven GM, Ford ES (2004).

Relationship to insulin resistance of the Adult Treatment Panel III diagnostic criteria for identification of the metabolic syndrome. *Diabetes.* **53** :1195-1200.

13. McLaughlin T, Abbasi F, Cheal K, Chu J, Lamendola C, Reaven G (2003). Use of metabolic markers to identify overweight individuals who are insulin resistant. *Ann Intern Med.* **139** :802-809.
14. Zimmet P, Alberti G (2006). Diabetes and the metabolic syndrome : The IDF definition. *Diabetes Voice.* **51** : 11-14.
15. Grundy SM, Cleeman JI, Daniels SR, et. al. (2005) Diagnosis and management of the metabolic syndrome. An American Heart Association/National Heart, Lung, and Blood Institute Scientific Statement. Executive summary. *Circulation.* **112** : e285-e290.
16. Grundy SM, Cleeman JI, Daniels SR, et. al. (2005). Diagnosis and management of the metabolic syndrome. An American Heart Association/National Heart, Lung, and Blood Institute Scientific Statement. *Circulation.* **112** : 2735-2752.
17. Grundy SM (2006). Does the Metabolic Syndrome Exist? *Diabetes Care.* **29** :1689-1692.
18. Reaven GM (1988). Banting Lecture 1988. Role of insulin resistance in human disease. *Diabetes.* **37** : 1595-1607.
19. Reaven GM (2006). The metabolic syndrome : is this diagnosis necessary? *Am Jour of Clin Nutr.* **83** :1237-1247.
20. Eckel RH, Kahn R, Robertson RM, Rizza RA (2006). Preventing Cardiovascular Disease and Diabetes : A call to action from the American Diabetes Association and the American Heart Association. *Diabetes Care.* **29**:1697-1699.

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Demographic Determinants of the Epidemic of Obesity

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There is an alarming increase in the incidence and prevalence of obesity and its adverse health consequences worldwide justifying its recognition by WHO as a "Global Epidemic"(1). Especially the increase in less affluent developing countries and economies in transition poses additional burden on their already fragile infrastructure. The rise in childhood obesity with attendant increase in Type 2 Diabetes and other components of the Metabolic syndrome is a matter of grave concern.

Magnitude of the problem

According to WHO estimates around one billion people in the world are overweight and over 300 million of them are obese (BMI > 30) (2). In developed countries of Western Europe, USA and Australasia the epidemic started in 1980's whereas in developing countries and economies in transition like India the increase in obesity started more recently (3, 4). Across the world statistics have shown rising incidence in all sections of the population (3). Though comprehensive national statistics are not available for our country representative data from different regions shows a definite rising trend (5). The worrisome aspect is the rise in childhood obesity especially in urban areas brought to light by several representative surveys of school children. Our country has to grapple with the twin burdens of traditional problems of infectious diseases and malnutrition as well as the rise in non-communicable diseases like diabetes and cardiovascular disease engendered by the change in lifestyle. According to the recent mortality and morbidity statistics non communicable diseases contribute to 53 % of deaths in our country (6). Even at lower levels of BMI conventionally regarded as non obese, Indians have higher body fat especially visceral fat contributing to the higher risk of metabolic syndrome and cardiovascular disease. This has led to the redefinition of BMI cutoff point for Asians as <23 (7).

Etiology of the Obesity Epidemic-Genes vs. Environment

Arguments for and against Nature vs. Nurture in the causation of obesity abound but there is consensus that interplay of genetic and environmental factors is crucial (8). The importance of genetic factors is attested by concordance of body weights of children with biologic parents in adoption studies and concordance between monozygotic twins in over feeding studies (9). Still the rapidity of progression of the

epidemic across the world cannot be explained by genetic factors and must be largely due to changes in lifestyle brought about by demographic changes, urbanization and consequent socioeconomic and behavioral changes in different countries (10, 11).

An attractive Thrifty gene hypothesis has been put forward to explain the gene-environment interaction (12). Evolutionarily the hunter-gatherer lifestyle of early man with inevitable fasting and feasting cycle contingent on availability of food provided hypothetical 'thrifty genes' favoring storage of energy in the form of adipose tissue to be used up in times of starvation and insulin resistance at the muscle level to divert fuel to vital organs like brain. These protective genes became detrimental when food supply became uninterrupted and abundant with progress in civilization (12).

Simplistically obesity is a consequence of positive energy balance with energy intake exceeding expenditure. It is an ongoing debate whether increased energy intake or decreased energy expenditure due to physical inactivity plays the dominant role in the causation of obesity.

Obesogenic modern environment

Is it increased energy intake?

There is a worldwide nutrition transition in which traditional diets are giving way to energy dense, high fat, high refined carbohydrate 'western diets' (13). Compared to the Paleolithic diet of prehistoric man the contemporary Western diet has 3 fold increase in fat especially saturated fat, high refined carbohydrate and less protein (12). Even though population data on overall caloric intake and dietary fat consumption provide mixed results, there is no doubt that increase in consumption of energy dense foods, number of meals eaten outside home and consumption of sugary aerated drinks are major factors linked to obesity (13). The seeds for the epidemic are sown in early childhood by early weaning and introduction of formula foods followed later by affinity to energy dense, high fat, high glycemic index, low fiber junk foods (13). The relative low cost of these foods, attractive packaging, lure of increased portion sizes and aggressive marketing by fast food outlets fuel the epidemic. Social and environmental factors like conducive school milieu, peer pressure and busy working parents using food as reward or substitute for quality time with children compound the problem further.

Is it decreased physical activity?

Studies on energy balance in obesity have shown resting metabolic rate in obese subjects is unchanged or actually increased and likewise energy expenditure of physical activity may be actually increased whereas energy dissipation by thermogenesis is decreased (10, 13). Hence efficient energy storage mechanisms may be operative. Many researchers conclude after analysis of dietary patterns of

communities over time that decrease in physical activity is a more important cause of obesity than increased caloric intake (11, 13). Urbanization and a switch from agrarian to industrial economy have led to more sedentary physically less demanding work (14). Increased use of automated transport and energy saving technologies at home have led to reduction in energy expenditure of day to day activities. Passive leisure time pursuits have increased because of television and computers. Especially in children time spent on TV viewing and computer gaming is directly correlated to the risk of obesity in many studies (15). In a nutshell the creations of civilization and industrialization that contribute to physical inactivity are two types of machines -those that reduce the energy cost of work or transport (electric appliances and cars) and those that promote passive recreation (Television, videos) (16).

Nutrition Transition in the developing world

Evolution of the human race and Civilization have brought in their wake several transitions from time to time (17). These can be classified into (i) Demographic transition (From high fertility and high mortality to low fertility and low mortality) (ii) Epidemiologic transition (From Infectious diseases and malnutrition to chronic non-communicable diseases) and (iii) Nutrition Transition (from Traditional diet to "Western diet") (17). The pace of change in these areas has accelerated especially in the last 3 centuries.

In evolution Man has progressed from the age of hunting and collecting food to the age of continuous food supply and concomitant chronic degenerative diseases and this has been brought about by agricultural and industrial revolutions (17).

Determinants of Nutrition Transition

Nutrition transition is attributable to a complex interplay of changes in several sectors namely agriculture, demography, socio-economic and health. Of these socioeconomic changes are key contributors. Changes in role of women especially with respect to time allocation to household chores because of increase in the number of working women is found to be one of the major determinants by demographic researchers (14, 17). Changes in income patterns, household food preparation technology, food production and processing technology and alterations in family and household composition are the other contributors. This has led to increase consumption of more processed instant foods with less fiber and more refined carbohydrates and fats which are obesogenic (14, 17).

Urbanization is the culprit!

Progressive increase in the urban population through migration from rural areas is witnessed in the developing world. The urban population in India had increased from 10.84% in 1901 to 25.72 %

in 1991 (18). The number of mega cities (cities with more than 8 million population) is showing an exponential increase in developing countries, while it remains almost constant in developed countries (19). This is largely because of internal migration of people from rural to urban areas. This brings about a switch of work force from agriculture to industrial and service sectors with more sedentary occupations contributing to weight gain.

Urbanization leads to increase in continuous availability of food with a wide variety, a significant percentage of it from processed commercial food sector. Shift in occupation patterns leads to reduced compatibility of jobs with home food preparation and necessitates eating out of home (14, 17). Socioeconomic factors also contribute equally. Direct correlation between annual per capita income and consumption of fats and refined carbohydrates is noted in several countries of the developing world (17). National nutrition surveys in our country have shown almost 2 fold difference in fat intake between urban and rural areas of the country (4).

The urban structure necessitates automated transport and constraints of time, space and safety constraints discourage simple physical activities like walking and cycling (13). High rise apartments and luxury hotels promote use of elevators and discourage or actually do not have free access to stairs (16). In children these problems of urbanization are compounded by long hours spent in school and coaching classes necessitated by the academic rat race.

Prevention /reversal of the obesity epidemic- The population approach

The salutary effects of lifestyle modification on reversing obesity and metabolic syndrome have been shown already in select populations and interventional studies for prevention of Type 2 Diabetes (20). For this we need to alter the obesogenic milieu through measures starting from early childhood. Education through mass media regarding adverse health consequences of junk food and sedentary habit, promotion of optimum maternal nutrition and breast feeding, inculcation of healthy feeding practices from infancy, incorporation of healthy life style advice in school curriculum and provision of opportunities for increasing physical activity in the urban milieu are some of the measures in this direction (15, 16, 21). This will require conviction, commitment and coordinated efforts from scientists, health care providers, school authorities, food industry and policy makers. In a nutshell large scale lifestyle changes brought about by demographic and socioeconomic changes and the consequent nutrition transition are fueling the obesity epidemic. Addressing lifestyle issues is the only way to stall the epidemic.

References :

(1) World Health organization. Obesity: preventing and managing the global epidemic. Geneva WHO 2000

(2) World Health organization. Global strategy on diet, physical activity and health. Geneva WHO 2003

(3) Stein CJ and Colditz GA (2004). The Epidemic of obesity *J Clin Endocrinol Metab* **89:** 2522-2525

(4) Settee PS (2002). Nutrition transition in India. *Public Health Nutrition* **5(1A):** 175-182

(5) Gopalan C (1998). Obesity in the urban middle class *NFI Bull* **19:**1-4

(6) Reddy KS, Bela Shah, Varghese C, Ramadoss (2005). A Responding to the threat of chronic diseases in Indians. *Lancet* **366:** 1744-1749

(7) WHO Expert consultation -Appropriate BMI for Asian populations and its implications for policy and intervention strategies *lancet* **363:**157-163, 2004.

(8) Prentice AM, Jebb SA (1995). Obesity in Britain- Gluttony or sloth *BMJ* **311:** 437-439

(9) Comuzzie AG ,Allison DB (1998). The search for human obesity genes Science **29:** 1374-1377, 1998

(10) Wing RR et al (2001). Lifestyle changes related to obesity, eating behavior and physical activity *Diabetes Care* **24:** 117-123

(11) Sorensen TIA The changing lifestyle in the world-Body weight and what else? *Diabetes Care* 23 (suppl 2) B1-B4, 2000

(12) Neel JV, Weder AB, Julius S (1998). Type 2 Diabetes essential hypertension and obesity as syndromes of impaired genetic homeostasis: the "Thrifty genotype hypothesis" enters the 21st century *Perspectives in Biological Medicine* **42:** 44-74

(13) Hill JO, Peters JC (1998). Environmental contributions to the obesity epidemic *Science* **280:** 1371-1374

(14) Popkin BM (2001) The Nutrition transition and obesity in the developing world *J Nutr* **131:** 8718-8738

(15) Wilson P, O'Meara S, Summerbell C, Kelly S (2003). The prevention and treatment of childhood obesity *Qual Saf health Care* **12:** 65-74

(16) Swinburn B, Bell C A (2005). Comprehensive approach to obesity prevention. In: Clinical obesity in adults and children Eds. Kopelman PG, Caterson ID, Dietz WH. 2nd Edition, Blackwell Publishing.

(17) Popkin BM (1998). The Nutrition Transition and its health implications in lower income countries *Pub Health Nutr* 1: 5-21

(18) Registrar general of India Census of India 1991 New Delhi Registrar general of India, 1998

(19) United Nations, Population division. World urbanization prospects; the 1994 revision. Estimates and projections of urban and rural populations and of urban agglomerations. New York: Population division, Deptt. For economic and social information and policy analysis, United Nations, 1995

(20) Diabetes Prevention Program Research Group Reduction in the incidence of Type 2 diabetes with life style intervention or metformin N Engl *J Med* **346:** 393-403, 2002

(21) Eckel RH et al (2004). Prevention conference VII Obesity, A worldwide epidemic related to heart disease and stroke -Executive summary *Circulation* **110:** 2968-75

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Energy Homeostasis and Obesity
Current Concepts

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Energy Homeostasis : Obesity is a complex metabolic disorder resulting from dysregulation of energy homeostasis which essentially reflects an abnormality in the balance between energy intake and energy expenditure. Such dysregulation may have genetic and /or life style as well as behavioural origins, involving the quality and quantity of food intake on the one hand, and sedentary life style on the other. As obesity is commonly associated with insulin resistance and hyperinsulinemia, such a constellation increases the risk of glucose intolerance, type 2 diabetes mellitus (T2DM), dyslipidemia and cardiovascular disease (CVD). The all-inclusive term, metabolic syndrome, has been suggested for this constellation.

The Regulation of Energy Balance : There is a general consensus that a physiological system maintains homeostasis of energy stores in response to varying availability of food and changing demands of energy expenditure. The energy demands of the body under resting basal, active and stressful conditions are adequately and appropriately responded to in a short period of a few minutes, by glucose which constitutes a most dependable energy supply source on a short term basis, although in the long term, body adipose tissue responds to the needs for the maintenance of energy balance. Based on our collaborative studies nearly three decades ago, Bajaj et al. (1) proposed the existence of an entero-hypothalamo-insular axis. Subsequently Bajaj (2) summarized the evidence for its metabolic role delineating neuroendocrine mechanisms involved in the regulation of energy balance. In this publication, it was specifically observed : 'the rate of glucose utilisation seems to be the set point in the regulation of entero-hypothalamo-insular axis. However, this may be so for the maintenance of energy balance on a short term basis. Adipose tissue functions as the major source of energy fuel; during starvation, glycogen stores in the human body may sustain life for less than 24 hours while energy stored as triglycerides can maintain supplies to vital organs for 30-60

days. It is therefore possible that control of triglyceride storage may be of considerable influence as a long range regulator of entero-hypothalamo-insular axis'. The subject has been recently reviewed (3) and the validity of the hypothesis proposed earlier has been established.

Hypothalamic Regulation of Energy Homeostasis : The hypothalamus is the principal brain region that acts as a key determinant in the integrated control of feeding, energy homeostasis, and regulation of body weight. Hypothalamus senses neural, endocrine, and metabolic signals, integrates these inputs, and engages distant effector pathways, resulting in behavioural, autonomic, and endocrine responses (4). The hypothalamic control and regulatory mechanism is mediated through a complex array of neuroendocrinal signaling pathways involving synthesis and release of several neurotransmitters and neuropeptides. These include monoamine neurotransmitters, such as 5-hydroxytryptamine, norepinephrine, as well as orexigenic neuropeptides (neuropeptide Y, orexins A and B) and anorectic peptides (cocaine-and amphetamine-regulated transcript: CART; pro-opiomelanocortin : POMC and related peptides i.e. αMSH) (5). A brief overview of the action of these peptides is provided in the following paragraphs.

The positional cloning of ob gene in 1994 (6), and the subsequent discovery that the encoded protein, named leptin, functions as an adipocyte derived signal for the regulation of feeding behaviour, set the direction and pace of research during the last decade. Ob receptor gene was cloned a year later, in 1995 (7), and leptin receptors were demonstrated in the arcuate nucleus of hypothalamus (8). The arcuate nucleus is located in the mediobasal hypothalamus adjacent to the floor of the third ventricle. It contains neurons that respond to afferent signals, predominately hormonal, which reflect and relate to the *size* and *state* of adipose tissue stores. Although leptin is secreted primarily from adipocytes and insulin is released from the endocrine panacreas, both circulate at levels proportionate to body fat mass and exert relatively long-lived inhibitory effects on food intake via actions on their receptors in the arcuate nucleus. These actions are mediated through a set of neurons in the arcuate nucleus which coexpress neuropeptide Y (NPY) and agouti-related peptide (AgRP). These two peptides are potent stimuli of food intake; moreover, these peptides also reduce energy expenditure and thus promote weight gain. In contrast, the arcuate nucleus also contains neurons that synthesise αMSH (Melanocyte stimulating hormone) that exerts a powerful anorectic effect. αMSH is synthesized from its precursor proopiomelanocortin (POMC). Many POMC neurons also coexpress another peptide called CART (cocaine and

amphetamine related transcript). Both αMSH and CART reduce food intake. α-MSH is an agonist for melanocortin 4 receptor (MC4R) which also appears to be involved in the regulation of appetite and body weight mediated through 5-hydroxytryptamine receptors (5HT2C), activation of which causes weight loss and deletion of which causes adult-onset obesity in mice (9).

Insulin and leptin signals, which are enhanced in a state of excess adipose tissue mass i.e. obesity, are inhibitory to NPY and AgRP neurons and facilitatory to POMC and CART neurons, the net effect of these hormones results in inhibition of feeding behaviour (10). It is through this reciprocal regulation of anabolic and catabolic neuronal circuits that insulin and leptin mediate their effects on energy balance. It was hypothesized that a decrease in plasma levels of insulin and leptin which follows a reduction in body fat mass, results in the activation of NPY/AgRP and inhibition of POMC neurons. While this may be partially true, the search for orexigenic peptides initiating feeding behaviour was intensified during the last decade but has met with remarkable success only recently. The recent discoveries, in this context, include the orexins in the lateral hypothalamus, the gastrointestinal hormone, Ghrelin in the gastric mucosa, and the Endocannabinoid system.

Orexins and Ghrelin : Two groups of investigators independently described peptides expressed exclusively in the LHA, zona incerta and PFA. While deLecea et al. named these peptides as hypocretins 1 and 2 (11), Sakurai et. al. designated them as Orexins A and B (12)(*G. orexis*, appetite). Orexin A consists of 33 aminoacids (Mol. wt. 3562 Da). Orexin A cDNA was made by reverse transcriptase treatment of rat brain mRNA followed by PCR. The structure of prepro-orexin gene has been determined, and it has been located on chromosome 17q21. Both the 33 aminoacid Orexin A and 28-aminoacid Orexin B (Mol. wt. 2937 Da) are encoded by a single mRNA transcript. Also identified were two Orexin receptors. Orexin A receptor (OX_1R) showed a 26% identity with NPY. While Orexin A was a specific high affinity agonist for OX_1R stimulation, a second receptor OX_2R with 64% identity to OX_1R and high affinity binding of Orexin B was sequenced. OX_2R also binds Orexin A (13). Impressed by the 'striking localisation' of Orexin containing neurons in the lateral hypothalamus and some of its adjacent areas, and none in the VMH-Arcuate-PVN area, Sakurai et. al. investigated the effect of Orexin administration through preimplanted indwelling catheters (12). Within one hour of intracerebroventricular administration of Orexin A bolus, food consumption was stimulated in a dose-dependent manner. These actions of Orexin were similar to, although of lesser order of magnitude, than those observed following NPY administration.

Ghrelin, an acylated peptide secreted by cells in the gastric mucosa, stimulates

food intake. Ghrelin activates previously identified orphan receptor (growth hormone secretagogue receptor) in the hypothalamus leading to release of growth hormone from the pituitary (14). It is now being increasingly realized that this peptide may be more important for initiating feeding behaviour through actions in the arcuate nucleus. Indeed, ghrelin is somewhat unique amongst gut hormones in *stimulating* food intake, and its chronic administration in rats causes obesity (15). Plasma levels of ghrelin rise significantly before meals, and fall following the intake of food (16). Colocalisation of ghrelin receptor and NPY mRNA in a group of neurons in acruate nucleus has been demonstrated (17). Thus ghrelin exerts its orexigenic effects by binding to its receptor on NPY/AgRP neurons, thereby activating this circuit.

The Endocannabinoid System : To the previously defined components of central and peripheral control mechanism regulating energy homeostasis as described above, has been added one of the great discoveries of the current decade, namely the existence of an endocannabinoid system.

Cannabis sativa or *cannabis indica* grows wild in several parts of the world, and is found all over India. *Marijuana* cigarettes are prepared from the leaves and flowering tops of the plant while *Hashish* is prepared from concentrated plant resin. While smoking is the most common mode of use, the oral intake of *Bhang* as a concoction is practiced in several socio-religious groups, more so during festivals. As a part of folk-lore medicine, cannabis has been used over millennia for disorders as varied as joint pains and epileptic convulsions. It was only in 1964 that its active psychoactive constituent Δ^9-tetrahydrocannabinol (Δ^9-THC) was isolated, its structure identified, and partial synthesis accomplished (18). Δ^9-THC alongwith other naturally occurring and synthetic cannabinoids, bind with two separate G protein-coupled receptors. Cannabinoid receptor 1(CB_1) (19) is localized in the CNS including neurons in lateral hypothalamus, and in the periphery especially in the liver and adipose tissue. In contrast, cannabinoid receptor 2 (CB_2) is found primarily on cells of the immune system (20). A complex biochemical pathway for the synthesis, release, transport, and degradation of endocannabinoids alongwith their receptors CB_1 and CB_2, constitutes the new signaling system. Since the discovery of fatty acid amide, arachidonoy-lethanolamide, by Devane et al in 1992 (21), who also coined the term 'Anandamide' from the Sanskrit root 'Ananda', meaning 'internal bliss', more than 3500 scientific reports have been published exploring diverse aspects of endocannabinoid system. The subject has been recently reviewed (22).

Endocannabinoid system, particularly in the brain, is generally 'silent' under normal conditions. It becomes activated under certain circumstances of stress in the

body. For example, it helps the body to relax; it reduces pain and anxiety; it causes sedation, or a slowing down; it tends to eliminate the memory of unpleasant things in life. It works at several levels of the body to do this. Importantly, this system, when it gets turned on, is thought to increase appetite through a number of rewarding pathways. In other words, *'relax, rest, forget, protect, and eat'* constitute the 'panchsheel' of peaceful coexistence of internal and external environments. However, increased endocannabinoid system activity is associated with excessive food intake, obesity, and insulin resistance.

The neurometabolic effects of endocannabinoids can be better understood in the context of neuroregulation of feeding behaviour. Endocannabinoids stimulate anabolic circuits, namely the NPY and AgRP in the arcuate and their receptors in the lateral hypothalamus where melanin-concentrating hormone (MCH) and orexins are located. MCH binds to a receptor MCH-R1, which is a G-protein coupled receptor (GPCR). Deletion of MCH-R1 produces a lean mouse due largely to increased energy expenditure, rather than suppression of feeding (23). Likewise, a recent study demonstrates significant antiobesity effect of a MCH-R1 antagonist in diet-induced obesity in mice (24). In addition, chronic administration of the antagonist ameliorated obesity-induced hypercholesterolemia, hyperinsulinemia, hypertriglyceridemia and hyperlipidemia. This study focuses sharply on the role of MCH in lateral hypothalamus. Finally, neuroanatomical studies reveal a connection between arcuate neurons expressing POMC and AgRP/NPY, and MCH neurons in lateral hypothalamus (25).

Endocannabinoids, in addition to stimulating anabolic circuits, also inhibit catabolic circuits consisting of CART and α-MSH, and their projections into PVN. The net effect is excessive food intake, obesity, and insulin resistance. Finally, evidence has been generated during the last decade indicating this signaling system as a modulator of physiological functions not only in the central nervous system, but also in the autonomic nervous system, neuroendocrinal network, the immune system, the gastrointestinal tract, the reproductive system both in the male and the female, and in microcirculation. CB_1 knock-out mice are lean as compared to wild type littermates (26). Specific antagonists of CB_1 such as rimonabant decrease food intake, reduce peripheral and hepatic lipogenesis, and lead to weight reduction. Clinical trials with rimonabant in obesity, dyslipidemia and type 2 diabetes mellitus have shown promise both in terms of efficacy and safety. The clearance from drug regulatory agencies (i.e. FDA) is awaited (22).

Control of Energy Expenditure : Rapid advances during the current decade have not only delineated the molecular mechanism underlying neural regulation of energy intake (feeding) as also of energy storage (adipogenesis and lipogenesis), but

have also provided new insights into the cellular mechanisms that control and regulate energy expenditure. Energy expenditure can be viewed as occurring in three major compartments. The first is the obligatory energy expended on basic cellular and physiologic functions that require ATP. The second is the energy spent in physical activity; some of this is noexercise activity thermogenesis (NEAT). It is due to physical activities other than volitional exercise. The third is referred to adaptive thermogenesis. The components of energy expenditure subject to ready alteration include physical activity and adaptive thermogenesis. It is the latter which has became the subject of current research, and subsequently of major scientific advance.

To facilitate easy understanding, it is useful to analyse energy expenditure from a thermodynamic perspective. Total energy expenditure reflects the conversion of oxygen and food (or fuels stored in the body as fat, glycogen and protein) into carbondioxide, water, heat, and work on the environment. Work performed on the environment plus heat produced during biological combustion of food or body fuels equals the amount of heat measured as calories. Adaptive thermogenesis, sometimes called facultative thermogenesis, is defined as heat production in response to environmental temperature or dietary intake.

Two main aspects of recent work on adaptive thermogenesis include the neural circuitory that activates thermogenesis and the peripheral tissues where oxidation of fuels takes place. As part of a unifying concept of energy homeostasis, it is not entirely unexpected that the hormonal signaling and neural pathways that control food intake are closely interlinked with these involved in energy expenditure. The role of insulin in diet-reduced thermogenesis is an illustrative example. Insulin injection into the ventromedial and paraventricular nucleus in the hypothalamus resulted in an increase in body temperature and energy expenditure, *in addition* to a reduction in food intake (27,28). In rats treated with diazoxide (a potent inhibitor of insulin secretion from pancreatic β-cells), there is a significant attenuation of the thermogenic response to a carbohydrate meal (29). Based on these studies, it can be surmised that insulin action in the hypothalamus not only reduces food intake, but also increases SNS (sympathetic nervous system) outflow to BAT (brown adipose tissue) to produce heat from fatty acid oxidation, thereby increasing energy expenditure.

These physiological and pharmacological observations may provide a rational basis for understanding obesity-associated hypertension, with hyperinsulinemia in obesity mediating the increase in SNS activity (30). Indeed, systemic insulin administration increases plasma catecholamine levels. An interesting corollary may be that elevated free fatty acids (FFA) commonly seen in

obesity may result from insulin activation of sympathetic nervous system. Such an increase in FFA may be causally linked with insulin resistance in the muscle. With this perspective, the association of obesity, insulin resistance, and type 2 diabetes mellitus, becomes readily understandable (30). However, more data need to be generated to substantiate such a linkage.

Besides insulin, leptin is the other major signal emanating from adipose tissue. Mutations in leptin, leptin receptor, melanocortin 4-receptor (MC4-R) and MCH all strongly influence both food intake and energy expenditure. Exogenous administration of leptin in ob/ob mice not only reduces food intake, but also increases energy expenditure. The mitochondrion ATP metabolism and thermogenesis mediated through one or more uncoupling proteins (UCP) not only in the adipose tissue but also in the skeletal muscle, constitute other focal points of recent investigations.

In the cold-induced thermogenesis in BAT in rodents, the primary molecule involved is UCP-1, a mitochondrial inner-membrane protein that uncouples proton entry from ATP synthesis (31). Two homologues of UCP-1 have been identified. These are UCP-2 (32) and UCP-3 (33). While all three uncoupling proteins are abundantly expressed in BAT, UCP-3 is also predominantly expressed in skeletal muscle whereas varying levels of UCP-2 are expressed in most tissues. These proteins do possibly play a major role in whole body energy expenditure outside of BAT; nevertheless, none of the UCP alone has been shown to exhibit an unequivoral anti-obesity effect. Moreover, in adult humans, the amount of BAT is minimal and UCP-1 expression is physiologically unmeaningful. Nevertheless, UCP-2 and UCP-3 were considered possible candidates in the human. Whether sympathetic drive plays a role has been investigated by developing 'betaless' (deletion of β_1, β_2 and β_3 receptors) mice which develop severe obesity even when food intake is identical with wild-type mice. Although direct evidence for such an effect in human obesity is lacking, studies on the expression of UCP-3 in different physiological situations in humans (and animals) tend to provide some support for its role in energy balance and lipid metabolism (34).

The recent discovery of PPARγ coactivator (PGC)-1 that can coactivate many transcription factors that participate in adaptive therogenesis, has provided a new insight linking adaptive thermogenesis with adipocyte biology (35). The cold-induced PGC-1 is the result of interaction of β_3 –adrenergic receptors and cAMP (36). When PGC-1 is expressed in white fat or skeletal muscle, a broad programme of thermogenesis is initiated which includes mitochondrial biogenesis, expression of UCP-1 in fat cells and UCP-2 in muscle, and an increase in cellular respiration. The ability of PGC-1 to promote mitochondrial biogenesis is due to its ability to turn on the expression of nuclear

respiratory factors NRF-1 and NRF-2, which in turn regulate mitochondrial transcription factor A (mt TFA) which directs the transcription and replication of mitochondrial genome (37). In addition to PPARγ, PGC-1 also binds to other nuclear receptors including those of retinoic acid and thyroid hormone, which in turn positively regulate expression of UCP-1.

UCP-2 and UCP-3 are also regulated by various nutritional perturbations in a tissue-selective manner. Present insights into the transcriptional regulation of UCP-2 and UCP-3 have come from *in vivo* administration to animals or *in vitro* treatment of cells with PPAR ligands such as thiozolidinediones. These studies have indicated that PPAR-α and PPAR-γ are positive regulators of UCP-2 and UCP-3, with specificity defined by the tissue or cell-type being examined. Given that fatty acids and/or their derivatives are ligands for these receptors, the PPARs may account for much of the nutritional regulation of UCP-2 and UCP-3.

Human obesity being a multifactorial syndrome, there may be a possibility that a combination of genetic defects may significantly enhance the susceptibility to obesity as compared to that due to independent gene defects. β_3 –adrenergic receptor (ADR β_3) is expressed in adipocytes and mediates the rate of lipolysis in response to catecholammies, whereas PPARγ_2 regulates adipogenesis. Although Trp64 Arg mutation of ADR β_3 has only a modest association with human obesity (38) . a recent study was undertaken to evaluate evidence for interaction between ADR β_3 Trp64 Arg mutation and PPARγ_2 Ala variants with respect of obesity in a cohort of 453 Mexican-American subjects. Interestingly, only those subjects who carried both the variants (32/453) exhibited increased obesity, in contrast to those with only PPARγ2 variant. The ADR β_3 variant was not significantly associated with any of the obesity-related traits (39). Further population studies aimed at investigating more than one gene variant shall be of interest.

Future Perspective : Obesity is emerging as a worldwide epidemic. Basic researches have yielded a rich harvest of knowledge enhancing our understanding of hormonal regulation of feeding behaviour and energy intake, the adipocyte biology underlying adipogenesis and adiposity, and the role of adaptive thermogenesis in the regulation of energy expenditure. New therapeutic strategies in the management of obesity are now emerging, targeting all these potential sites, through the use of modern tools of molecular pharmacology and pharmacogenomics. Nevertheless, life-style changes and positive behavioural alterations constitute the most safe and cost effective strategies to-date.

References :

1. Bajaj JS, Chhina GS, Mohankumar V, et al. (1975). Evidence for the existence of an entero-hypothalamic-insular axis. *Diabetologia* **11**:331-333.
2. Bajaj JS. (1976). Entero-hypothalamo-insular axis. In: Diabetes. Bajaj JS (ed), Amsterdam, Excerpta Medica . 18-31.
3. Bajaj JS (1998). Entero-hypothalamo-insular axis revisited. *Ann Natl Acad Med Sci* **34**:57-74.
4. Schwartz MW, Woods SC, Porte D, Jr. Seeley RJ, Baskin DG (2000). Central nervous system control of food intake. *Nature* **404**:661-671.
5. Williams G, Bing C, Cai XJ, Harrold JA, Kind PJ, Liu XH (2001). The hypothalamus and the control of energy homeostasis : different circuits, different purposes. *Physiol Behav* **74**:683-701.
6. Zhang Y, Proenca R, Maffei M, Barone M, Leopold L, Friedman JM (1994). Positional cloning of the mouse obese gene and its human homologue. *Nature* **372**:425-432.
7. Tartaglia LA, Dembski M, Weng X, Deng N, Culpepeer J, Devos R et. al. (1995). Identification and expression cloning of a leptin receptor, OB-R. *Cell* **83**:1263-1271.
8. Mercer JG, Hoggard N, Williams LM, et. al. (1996). Localization of leptin receptor mRNA and the long form splice variant (Ob-Rb) in mouse hypothalamus and adjacent brain regions by in situ hybridization. *FEBS Lett.* **387**:113-116.
9. Heisler, L K, Cowley, M A, Tecott, L H et. al. (2002). Activation of central melanocortin pathways by fenfluramine. *Science* **297**, 609-611.
10. Spiegelman BM, Flier JS (2001). Obesity and the Regulation of Energy Balance. *Cell* **104**:531-543.
11. de Lecea, L, Kilduff T S, Peyron C, et al.(1998). The hypocretins : hypothalamus-specific peptides with neuroexcitatory activity. *Proc Natl Acad Sci* USA **95**, 322-327.
12. Sakurai T, Amemiya A, Ishii M, et. al. (1998). Orexins and orexin receptors:a family of hypothalamic neuropeptides and G-protein-coupled receptors that regulate feeding behaviour. *Cell* **92**:573-585.
13. Date Y, Ueta Y, Yamashita H, et. al. (1999). Orexins, orexigenic hypothalamic peptides, interact with autonomic, neuroendorcrine and neuroregulatory systems. *Proc Natl Acad Sci* USA **96**: 748-753.
14. Kajima, M., Hosoda, H., Date, Y., Nakazato, M., Matsuo, H., and Kangawa, K. (1999). Ghrelin is a growth-hormone-releasing acylated peptide from stomach. *Nature* **402**, 656-660.
15. Tschop, M., Smiley, D.L., and Heiman, M.L. (2000). Ghrelin induces adiposity in rodents. *Nature* **407**, 908-913.
16. Cummings DE, Foster KE (2003). Ghrelin-leptin tango in body-weight regulation. *Gastroenterology* **124**:1532-1535.
17. Willesen MG, Kristensen P, Romer J (1999). Co-localisation of growth hormone secretagogue receptor and NPY mRNA in the arcuate nucleus of the rat. *Neuroendocrinology* **70**: 306-316.
18. Gaoni Y, Mechoulam R (1964). Isolation, structure, and partial synthesis of an active constituent of hashish. *J Am Chem Soc* **86**:1646-1647.

19. Matsuda LA, Lolait SJ, Brownstein MJ, Young AC, Bonner TI (1990). Structure of a cannabinoid receptor and functional expression of the cloned cDNA. *Nature* **346**:561-564.

20. Munro S, Thomas KL, Abu-Shaar M (1993). Molecular characterization of a peripheral receptor for cannabinoids. *Nature* **365**:61-65.

21. Devane WA, Hanus L, Breuer A, et. al. (1992). Isolation and structure of a brain constitutent that binds to the cannabinoid receptor. *Science* **258**:1946-1949.

22. Bajaj JS. (2006). Selective CB_1 Receptor Antagonists : An evolving therapeutic revolution. In: Medicine Update. Sahay B K (ed). The Association of Physicians of India, **16**:3-7.

23. Marsh , D J, Weingarth D T, Novi, D E et. al. (2002). Melanin-concentrating hormone 1 receptor-deficient mice are lean, hyperactive, and hyperphagic and have altered metabolism. *Proc Natl Acad Sci. USA* **99**, 3240-3245.

24. Mashiko S, Ishihara A, Gomori A (2005). Antiobesity effect of a melanin-concentrating hormone 1 receptor antagonist in diet-induced obese mice. *Endocrinology* **146**:3080-3086.

25. Elias, C F, Aschkenasi C, Lee C et. al. (1999). Leptin differentially regulates NPY and POMC neurons projecting to the lateral hypothalamic area. *Neuron* **23**, 775-786.

26. Cota D, Marsicano G, Tschop M, Grubler Y, Flachskamm C, Schubert M et. al. (2003). The endogenous cannabinoid system affects energy balance via central orexigenic drive and peripheral lipogenesis. *J Clin Invest* **112**:423-431.

27. Menendez JA, Atrens DM (1991). Insulin and the paraventricular hypothalamus : modulation of energy balance. *Brain Res* **555** : 193-201.

28. McGowan MK, Andrews KM, Grossman SP (1992). Chronic intrahyphothalamic infusions of insulin or insulin antibodies alter body weight and food intake in the rat. *Physiol Behav* **51** : 753-766.

29. Rothwell NJ, Saville ME, Stock MJ (1983). Role of insulin in thermogenic responses to refeeding in 3-day-fasted rats. *Am J Physiol* **245** : E160-E165.

30. Landsberg L (2001). Insulin-mediated sympathetic stimulation : role in the pathogenesis of obesity-related hypertension (or, how insulin affects blood pressure, and why). *J Hypertens* **19**:523-528.

31. Klingenberg M and Huang SG (1999). Structure and function of the uncoupling protein from brown adipose tissue. *Biochim Biophys Acta* **1415**:271-296.

32. Fleury C, Neverova M, Collins S, et al. (1997). Uncoupling protein-2 : a novel gene linked to obesity and hyperinsulinemia. *Nature Genet* **15**:269-272.

33. Vidal-Puig A, Salanes G, Grujic D, et al. (1997). UCP3 : an uncoupling protein homologue expressed preferentially and abundantly in skeletal muscle and brown adipose tissue. *Biochem Biophys Res. Commun* **235**:79-82.

34. Samec S, Seydoux J, Dulloo AG, et al. (1999). Post-starvation gene expression of skeletal muscle uncoupling protein 2 and uncoupling protein 3 in response to dietary fat levels and fatty acid composition - A link with insulin resistance. *Diabetes* **48**:436-441.

35. Puigserver P, Wu Z, Park CW, et al. (1998). A cold-inducible coactivator of nuclear receptors linked to adaptive thermogenesis. *Cell* **92**:829-839.

36. Boss O, Bechman E, Vidal-Puig A, et al. (1999). Role of the beta (3)-adrenergic receptor and/or a putative beta (4)-adrenergic receptor on the expression of uncoupling proteins and peroxisome proliferators-activated receptor-gamma coactivator-1. *Biochem Biophys Res Commun* **261**:870-876.

37. Wu Z, Pugiserver P, Andersson U, et al. (1999). Mechanism controlling mitochondrial biogenesis and respiration through the thermogenic coactivator PGC-1. *Cell* **98**:115-124.

38. Allison DB, Heo M, Faith MS, et al. (1998). Meta-analysis of the association of the Trp64Arg polymorphism in the β3 adrenergic receptor with body mass index. *Int J Obes Relat Metab Disord* **6**:559-566.

39. Hsueh Wen-Chi, Shelly A. Cole, Alan R. Shuldiner, et al. (2001). Interactions between variants in the β3-adrenergic receptor and peroxisome proliferators-activated receptor-γ2 genes and obesity. *Diabetes Care* **24**:672-677.

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Measurement of Insulin Resistance in Vivo

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Insulin resistance is defined as a subnormal biological response to endogenous or exogenous insulin. This subnormal biological response is considered for ability of insulin to decrease the blood glucose. However, the dose of insulin for optimal biological response is quite variable and has marked intra- and interindividual variations (1). In clamp studies it has been shown that subjects who are obese but non-diabetic are insulin resistant and it progressively increased with occurrence of glucose intolerance (1). It has been also described that the minimum amount of physiological concentration of insulin is required for prevention of ketosis, whereas the maximum amount of insulin concentration is required for muscle glucose uptake. The clinical markers for insulin insensitivity include BMI>25kg/m^2 , waist line>102 cm in men and 88 cm in women, acanthosis nigricans and skin tags (2).

Insulin resistance in vivo can be measured by a computerized based model named as Homeostatic Model Assessment (HOMA) assessing insulin resistance at periphery called as HOMA-IR and at β-cell called as HOMA-β.[3] It provides an idea about the basal insulin resistance where the formula for HOMA- IR includes FPG (mmol/L)x IRI(μU/ml) /22.5 and formula for HOMA-β is 20 x IRI (μU/ml) / FPG(mmol/L) -3.5. The HOMA correlates with clamp studies and is good for epidemiological surveys. Another test is short insulin tolerance test where insulin is injected at a does of 0.1 U/kg body weight as IV bolus and samples for glucose are taken at 15 and 5 minute before and 3, 6, 9, 12, 15, 20 and 30 minutes after insulin injection (1). The rate constant for plasma glucose disappearance is calculated from the formula 0.693/T ½ and the plasma glucose T ½ is calculated from the slope of the least square analysis of the plasma glucose concentration from 3-15 minute after IV insulin injection. The other method to assess insulin resistance is oral glucose tolerance test which includes following ingestion of 75 gm of glucose, plasma glucose and insulin samples are taken at 0,30,60,90,120 minutes. It correlates with hyperglycemic clamp technique ($r=0.67$, $p< 0.05$). The clamp studies are the 'gold standards' for assessing insulin resistance both at β cell as well as in peripheral tissues (4). The euglycemic clamp technique assesses peripheral insulin resistance where glucose is given at a variable rate with the aim of maintaining plasma glucose at or

between 90-110 mg/dl and insulin is given at a fixed dose infusion. Samples for blood glucose are collected at 3-5 minute interval and insulin resistance is calculated by one upon the amount of glucose infused to maintain the steady state. Similarly the hyperglycemic clamp is used to assess the β cell secretory function where glucose is infused at variable rate with the aim to keep the plasma glucose between 180-200 mg/dl and at a steady state sample for glucose and insulin are taken and glucose insulin ratio gives the value of resistance at a β cell level (4). The clamp studies are 'gold standard' to assess insulin resistance but they are invasive, expensive and requires a lot of efforts, therefore, these are used only for research purposes. However, in clinical practice, HOMA and insulin tolerance test are good modalities to assess insulin resistance (1).

The insulin resistance is associated with various metabolic abnormalities including visceral adiposity, hypertension, impaired glucose tolerance and dyslipidemia and virtually all these are surrogate markers of insulin resistance. ATPIII and WHO both have defined the insulin resistance (2) syndrome by including these parameters and at least 3 of them have to be present for a diagnosis of insulin resistance syndrome(IRS).

The abdominal or visceral adiposity is the hallmark of insulin resistance syndrome and whether it is a cause or effect of insulin resistance is a million dollar question (5). Possibly the visceral obesity is influenced by genetic factors to begin with and later on environmental factors predominate. The waist circumference of more than 102 cm in men and more than 88 cm in women has been defined as detrimental (6,7). The other putative mechanism for abdominal obesity include increased 11-beta hydroxysteroid dehydrogenase type 1 activity which converts cortisone to cortisol, that by inducing lipoprotein lipase activity results in increased visceral fat (6). Hyperinsulinemia as a consequence of insulin resistance also increases visceral fat (6). Visceral fat being more metabolically active and rich in blood and β_3 adrenergic supply results in excess release of free fatty acids (FFAs). FFA's not only result in impaired insulin action at target tissues including muscle but impaired insulin secretion by β cells (8,9). Impaired insulin action at muscle is largely attributed to interference with insulin signaling mechanism including tyrosine kinase and PI3 pathway (10). Similarly impaired insulin secretion is due to decrease in glucose uptake and utilization by the β cells thereby requisite amount of ATP is not produced resulting in decreased insulin secretion (9). These effects of FFA's are collectively called as lipotoxicity. Increasing insulin resistance progressively affects the β cell function and subsequently results in inexorable β cell exhaustion (12). Hypertension associated with insulin resistance is attributed to lack of insulin mediated vasodilatation because of decreased nitric oxide synthase (NO synthase), insulin mediated increased

absorption of sodium from the renal tubule, increased sympathetic nervous system activity and increased angiotensinogen and angiotensin converting enzyme activity from adipocytes are other contributory factors for hypertension (2). The glucose intolerance associated with insulin resistance is the result of increased insulin secretion from the beta cells subsequent to insulin resistance and later on leading to beta cell exhaustion and occurrence of hyperglycemia (13). The dyslipidemia associated with insulin resistance is characterized by increased VLDL, increased small dense LDL and decreased HDL. These abnormalities of lipid profile are consequent to impaired lipoprotein lipase (LPL) activity, increased hormone sensitive lipase (HSL) activity, and decreased degradation of LDL leading to their longer stay in circulation and making them vulnerable to become small dense LDL (10).

Therefore, insulin resistance is the characteristic abnormality of insulin resistance syndrome and various clinical and biochemical predictors are surrogate markers of it. It has a relevance in predicting the future onset of T2DM and cardiovascular diseases.

References :

1. Wallace TM, Matthews DR (2002). Assessment of insulin resistance in man. *Diabetic Medicine*, **19:** 527-534.
2. Reaven GM: Banting lecture 1988. Role of insulin resistance in human disease. *Diabetes* **37:** 1595-607.
3. Matthews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF, Turner RC. Homeostasis model assessment: insulin résistance and beta-cell function for fasting plasma glucose and insulin concentration in man. *Diabetologia* 1985; **28:** 412-419.
4. Defronzo RA, Tobin JD, Andres R (1979). Glucose clamp technique a method for quantifying insulin secretion and resistance. *Am J Physiol* **237:** E 214-223.
5. Kissebah AH, Peiris AN (1989). Biology of regional body fat distribution: relationship to non-insulin-dependent diabetes mellitus. *Diabetes Metab Rev* 5:83-109.
6. Albu JB, Kovera AJ, Johnson JA (2000). Fat distribution and health in obesity. *Ann N Y Acad Sci* **904:** 491-501
7. Carey DG, Jenkins AB, Campbell LV, et al (1996). Abdominal fat and insulin resistance in normal and overweight women. *Diabetes* **45:** 633-638.
8. Pratley RE,Weyer C, Bogardus C (2000). Metabolic abnormalities in the development of type 2 diabetes mellitus. In Diabetes Mellitus: A fundamental and clinical Text. Edited by LeRoith D, Taylor SI. Philadelphia: *Lippincott*; 2-11.
9. Pratley RE, Weyer C (2001). The role of impaired early insulin secretion in the pathogenesis of type 2 diabetes mellitus. *Diabetologia* **44:** 929-945.
10. Groop LC, Saloranta C, Shank M, et al (1991) the role of free fatty acid metabolism in the pathogenesis of insulin resistance in obesity and non insulin-dependent diabetes mellitus. *J Clin Endocrinol Metab* **72:** 96-107.

11. Radle PJ, Garland PB, Hales CN, Newsholme EA (1963). The glucose fatty acid cycle: its role in insulin sensitivity and metabolic disturbances of diabetes mellitus. *Lancet* **1**: 785-789.

12. DeFronzo RA : Llilly Lecture (1987). The triumvirate: B-cell muscle liver. A collusion responsible for type 2 diabetes. *Diaebtes* 1988; **37**: 667-687.

13. Porte Jr D: Banting Lecture (1990). beta-cells in type 2 diabetes mellitus. *Diabetes* 1991, **40**: 166-180.

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Genetics of Metabolic Syndrome

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Summary

The metabolic syndrome (MS) is characterized by the clustering of several metabolic disorders such as increased body weight, insulin resistance, elevated plasma triglyceride levels, low HDL-cholesterol, high blood pressure and altered glucose homeostasis. Genetic and environmental factors such as low physical activity and unhealthy diet are strong determinants of the MS. Association and linkage studies on candidate genes for MS have revealed significant associations between the MS and polymorphisms in several different genes such as adiponectin, Plasma Cell Membrane Glycoprotein 1 (PC1), Hepatic Lipase, ApoA1/A5/C3/A4 Cluster, Lymphotoxin, interleukin 6 , lamin A/C, peroxisome proliferator-activated receptor-gamma 2 (PPAR γ2), angiotensinogen-1-converting enzyme (ACE), low density lipoprotein-related protein-associated protein, and Beta 2-adrenergic receptor (ADRB2) genes . In contrast, no significant associations have been reported for PPAR α, PPAR co-activator 1 α, protein tyrosine phosphatase 1 B, ACE , fatty acid binding protein 2 , tumor necrosis factor alpha (TNF-α) , insulin receptor and ADRB3 genes. Altogether, the data supports the hypothesis that complex genetic factors may be implicated in the pathogenesis of the MS.

Introduction

The metabolic syndrome (MS) is characterized by the clustering of several metabolic disorders, such as increased body weight, insulin resistance, elevated plasma triglyceride levels, low HDL-cholesterol, high blood pressure, and altered glucose homeostasis. The prevalence of the MS ranges between 15 and 25%. This prevalence increases with age, affecting 40% of subjects aged 60 years or more. Environmental factors such as low physical activity and unhealthy diet are strong determinants of the MS.

The metabolic syndrome is a disease with multifactor etiologies. Genetic factors have been found to influence the individual susceptibility to the MS. This is supported by the observation that metabolic disorders of the MS tend to clusters in families. For instance, 45% of first-degree relatives of type 2 diabetes patients are insulin resistant compared to 20% of individuals without a family history of diabetes. Patients with

type 2 diabetes also have an increased waist-to-hip ratio compared to subjects without a family history of type 2 diabetes. The heritability of obesity varies from 20 to 90% depending on whether the estimates are based on twin, adoption, or family studies. It has been reported that heritability also influences other components of the MS such as hypertension, triglyceride, and HDL-cholesterol concentrations.

There are a number of approaches available for investigating genes that confer susceptibility to a disease, including genome scanning by positional cloning and the candidate gene approach. Genome-wide linkage analyses have identified several chromosomal regions where major susceptible genes to MS may exist. Table1 shows the range of LOD scores and locations on various chromosomes where such susceptibility loci have been discovered. These loci are known to harbor several candidate genes for metabolic syndrome.

Table 1

S.No	Chromosome	LOD Signal	Location(cM)
1	2	1.0-3.4	122-180.6
2	11	1.1-3.0	131-143.1
3	16	1.1-3.2	46-78.6
4	19	1.0-3.3	80-86.4
5	22	1.0-3.4	19-20.9

Mutations and Polymorphisms in the genes

Several studies have reported significant associations between the MS and polymorphisms (SNPs) in several different genes associated with lipid metabolism, insulin resistance, adipocyte abnormality, chronic inflammation, autonomic imbalance etc. Promising candidate genes include PPAR γ2, PC1, Adiponectin, Hepatic Lipase, Apo A1, C3, A4, A5, Fatty Acid Binding Protein 2, BAR 2, RAS, Insulin Receptor substrate 1 and 2, lymphotoxin-α, interleukin 6, lamin A/C, peroxisome proliferator-activated receptor-γ (PPARγ), angiotensinogen-1-converting enzyme (ACE), low density lipoprotein-related protein-associated protein 1, β_2-adrenergic receptor (ADRB2) genes, USF1, CAPN10, HNF4a and PPARc etc.

Peroxisome Proliferator-Activated Receptors

The three peroxisome-proliferator-activated receptor (PPAR) subtypes, PPAR-gamma, PPAR-alpha and PPAR-delta, are nuclear receptors that have been the focus of extensive research during the past decade. These receptors function as lipid sensors that coordinately regulate the expression of large gene arrays and, thereby, modulate important metabolic events. Peroxisome proliferator-activated receptor (PPAR) gamma is involved mainly in adipocyte differentiation and has been suggested to play an important role in the pathogenesis of insulin resistance and atherosclerosis. The most frequently occurring PPARγ polymorphism is substitution of proline to alanine (Pro12Ala) in exon B of the PPARγ2 gene, and although many studies have been

performed on the association between this polymorphism and type 2 diabetes mellitus, insulin resistance, and obesity, the significance of such associations remains an issue of debate Many studies explored the association between the Pro12Ala polymorphism in PPAR gamma and obesity, insulin sensitivity, and type 2 diabetes (1). It appears that the Ala12 allele confers modest protection against the onset of type 2 diabetes and is also associated with an increased BMI in overweight individuals.

PPAR alpha L162V polymorphism, alone or in combination or in interaction with dietary fat intake has been also found to be associated with metabolic syndrome (2).

Plasma Cell Membrane Glycoprotein 1 (PC1)

PC1 is a class II transmembrane glycoprotein that inhibits IR tyrosine kinase activity. The A K121Q polymorphic variant in exon 4 of this PC-1 gene has been associated with insulin resistance or hyperglycemia (3, 4).

Adiponectin

Adiponectin, is an adipose-derived plasma protein and has been well established to be an important biomarker for metabolic syndrome and its complications in humans. Hypoadiponectinemia is associated with insulin resistance, type 2 diabetes, obesity, and dyslipidemia. Some of the common polymorphisms in the promoter region, exon and intron 2, and the rare nonsynonymous mutations in exon 3 of the human adiponectin gene were repeatedly shown in many studies from many different ethnic populations to associate with the phenotypes related to body weight, glucose metabolism, insulin sensitivity, and risk of type 2 diabetes mellitus and coronary artery disease. The SNP276 may affect impaired glucose tolerance and hypoadiponectinemia (5).

The association of adiponectin genetic variations with dyslipidemia and blood pressure have been less explored. The common polymorphisms and rare mutations of the human adiponectin gene itself were demonstrated to associate with differential expression of adiponectin at the plasma protein level and mRNA level in adipose tissue. The PPARgamma2 Pro12Ala variants were also shown to influence insulin sensitivity in interaction with adiponectin genotype or to influence plasma adiponectin levels. However, the results were not consistent. Three genome-wide scans for the loci that regulate plasma adiponectin concentration suggest that further exploration on chromosomes 5, 9, 14, 15, and 18 is required. These human genetic studies on adiponectin and the metabolic syndrome strongly suggest that adiponectin is one of the causative factors in its pathogenesis and provide significant insights into the genetic makeup of the metabolic syndrome

Hepatic Lipase (HL)

High HL activity is associated with reduced HDL2 cholesterol levels and is

affected by dietary fat intake and selected medications. There is evidence for an interaction of the HL promoter polymorphism with visceral obesity and dietary fat intake. Several polymorphisms, G 250 A , C 514 T, T 710 C, A 763 G are known to be associated with HL activity (6).

A recent linkage and association analysis revealed variation at the APOA1/C3/A4/A5 gene cluster contributes to Familial hypercholesterolemia transmission in a substantial proportion of Northern European families. Apo CIII: TG-rich lipoproteins are often increased in metabolic syndrome. The T-455C polymorphism of Apo CIII are shown to be associated with increased Apo CIII and TG levels. Apo CIII-455C was found to increases risk of CAD in MS. Obesity was less frequent in MS carriers of the -455C allele than in MS non carriers (21.6% vs. 34.8%, $P < 0.05$)

APOA V

Several polymorphisms of Apo V (-1131C (originally referred to as SNP3), Ser19Trp , have been shown to be have associations with TG in healthy and non-smoking subjects. T-1131C was found to be associated with higher concentrations of plasma TG

Fatty Acid Binding Protein 2(FABP 2) and Beta Adrenergic Receptor 2 (BAR 2)

Both these proteins link the inflammatory and lipid-mediated pathways and have been are implicated in Atherosclerosis, Obesity, Insulin resistance, Type 2 diabetes and Fatty liver disease . Ala54Thr polymorphism of FABP2 and Trp64Arg polymorphism of BAR 2 has been implicated in MS (7).

Renin Angiotensin System (RAS)

Helps maintain blood pressure and salt homeostasis. Polymorphisms of RAS genes, namely ACE insertion/deletion (I/D), Angiotensinogen (AGT). M235T, Angiotensin II type 1 receptor (AT1R). A1166C polymorphisms have been studied for their association with various cardiovascular disorders, including metabolic syndrome. D-allele and DD genotype of ACE I/D has been found to be associated with development of obesity, insulin resistance, type 2 diabetes, dyslipoproteinemias and ischemic heart disease and myocardial infarction in some of the studies whereas many studies have failed to show any such association (8).

There has been some evidence to support an association between the AGT polymorphism and insulin resistance (9).

Angiotensin II type I receptor (AGTR1) A1166C appears to predispose to favourable anthropometric and metabolic traits, relative to cardiovascular risk (10).

USF1

USF1 is a transcription factor of the c-myc family [11] that is known to regulate several genes involved in glucose and lipid metabolism, including apolipoproteins CIII, AII and E, hormone sensitive lipase, fatty acid synthase, glucokinase, the

glucagon receptor, ATP binding cassette A1 and rennin (12). The USF1 gene is located on human chromosome 1q22–q23 (13). This locus on chromosome 1 has been linked to familial combined hyperlipidaemia (FCHL), a condition characterized by elevated total cholesterol and / or triglycerides, in Finnish families (14) and is also located close to type 2 diabetes linkage peaks in other populations (15, 16). Subsequently, the USF1 gene has been linked and associated with FCHL, and haplotypes of USF1 explain the linkage peak associated with disease, triglycerides, cholesterol, small dense low-density lipoprotein and apolipoprotein B. Furthermore, target genes for this transcription factor include many other genes associated with hypertension and diabetes. Subsequent candidate gene studies will determine if there is an association between components of metabolic syndrome and SNPs of the USF1 gene.

CAPN10

Calpains are ubiquitously expressed cysteine proteases that regulate a variety of cellular functions. CAPN10 is expressed in B cells where evidence suggests it mediates apoptosis and insulin exocytosis (17, 18), in fat and muscle CAPN10 modifies insulin-mediated glucose transport (18). It also appears to be involved in myoblast / pre-adipocyte differentiation (19, 20). CAPN10 was the first type 2 diabetes gene identified by a genome-wide scan of Mexican–American families. Initially, linkage was found on chromosome 2 (LOD 4.03) (21) and the gene was identified as calpain 10. Three intron variants account for most of the haplotype diversity and for 14% of the population-attributable risk of type 2 diabetes in Mexican–Americans (22). Meta-analyses of association studies assessing CAPN10 and type 2 diabetes risk have confirmed a role for CAPN10 polymorphisms in type 2 diabetes susceptibility (23), increasing risk by _30% for type 2 diabetes (24). In one of the analyses, SNP44, a rare CAPN10 allele, was shown to be over-transmitted from heterozygous parents to their affected offspring with type 2 diabetes. In Pima Indians, a CAPN10 polymorphism (SNP43) correlates with impaired insulin action and reduced expression of CAPN10 in the skeletal muscle of prediabetic subjects (25). CAPN10 polymorphisms have also been associated with insulin secretion, adipocyte biology and microvascular function. This suggests that CAPN10 plays a role in the metabolic syndrome.

HNF4a

HNF4a is a hepatic nuclear factor that controls expression of many essential genes in the liver, gut, kidney and B cells, and plays an important role in maintaining glucose homeostasis (26). It is also involved in B-cell development, and mutations of HNF4a are a rare cause of MODY (27). The gene for HNF4a has been mapped to chromosome 20. Chromosome 20 has shown evidence of linkage to type 2 diabetes (LOD score 2.48) (28), and an

association between a common HNF4a polymorphism in the upstream promoter region of the gene has been found in an Ashekenazi Jewish population. Furthermore, a case control association study searching for diabetes susceptibility variants at 20q13 found 10 associated polymorphisms located in the promoter regions and exons 1–3 of the HNF4a gene (28). These data suggest variants located near or within the HNF4 gene increase susceptibility to type 2 diabetes.

PPARc

Variants of the PPARc gene are strong candidates for conferring susceptibility to type 2 diabetes and obesity, because PPARc regulates adipocyte differentiation, and lipid and glucose metabolism (29). Two PPARc isoforms, PPARc1 and PPARc2, have been characterized, and are encoded by a single PPAR gamma gene. The PPARc gene has been mapped to human chromosome 3p25 (30). One of the first examples of a meta-analysis in complex disease demonstrated that the Pro12Ala variant of PPARc2 is associated with predisposition to type 2 diabetes (31). Evidence suggests a gene–nutrient interaction at the PPARc2 locus in nondiabetic subjects(32). BMI was shown to vary with the ratio of dietary polyunsaturated to saturated fat (P:S) – in Ala12 carriers, BMI was low when the P:S ratio was high and was elevated when the P:S ratio was low. This gene–nutrient interaction suggests that effects of the Pro12Ala polymorphism could depend on the most common diets in the populations studied and may explain the conflicting results of previous studies. Furthermore, in a single study, the Finnish diabetes prevention study, the Ala12 allele was associated with increased risk of progression to type 2 diabetes. However, the Ala12Ala genotype was found to be associated with increased weight loss in response to lifestyle intervention and less progression from IGT to diabetes (33). Thus, lifestyle intervention can reverse the diabetogenic risk of the Ala allele.

In contrast, no significant associations have been reported for PPARα [2], PPAR co-activator 1α (34), protein tyrosine phosphatase 1 B (35), ACE (36), fatty acid binding protein 2 (37) tumor necrosis factor-α (TNF-α) (38) , insulin receptor (39) and ADRB3 (40) and (41) genes. Altogether, these data support the hypothesis that complex genetic factors may be implicated in the pathogenesis of the MS.

UCP3

UCP3 is a mitochondrial membrane transporter mainly expressed in skeletal muscle. This protein uncouples oxidative ATP phosphorylation and if dysregulated, might lead to thermogenesis and energy balance disorders. The UCP3 -55 C/T SNP is associated with higher UCP3 mRNA expression levels (42) and fat mass (43-47). The relation between this SNP and type 2 diabetes is controversial, with a lower risk of type 2 diabetes in this population sample (48, 27) and a higher risk in Chinese (49). Despite the major contribution of increased body mass in the clustering of metabolic disorders and the persistent association

between the UCP3 -55 C/T SNP and increased fat mass, there was no significant association between this SNP and the MS

Leptin

It is an adipocyte-secreted hormone acting in the brain to control energy homeostasis. Its main effect is to regulate appetite and energy balance (50). Pathogenic mutations in the leptin (LEP) gene leading to the absence of any functional hormone are associated with early-onset morbid obesity (51, 52). The LEP 5'UTR +19 G/A SNP is associated with lower leptin concentrations in obese individuals (53). In the present population sample, this SNP was not significantly associated with the MS.

G proteins mediate many pathways including the β-adrenergic signalling pathways. A polymorphism in the β_2-adrenergic receptor is associated with the MS (54) , suggesting a possible implication of the adrenergic pathway in the clustering of MS. The GNB3 C825T SNP is functional and has been associated with several phenotypes such as dyslipidemia, hypertension, obesity, and diabetes mellitus (55, 56). Furthermore, a study including 806 Japanese subjects (80% men) showed the CC genotype was protective against the onset of the clustering of obesity, hypertriglyceridemia, hypertension, and diabetes mellitus (56).

Given the fact that the prevalence of coronary heart disease stroke and all-cause mortality is increased threefold in subjects with the MS (57), even in the absence of baseline CVD and diabetes (58), it is important to better understand the factors that increase the susceptibility to the development of the MS. Genetic association studies may help in this respect. We could not reveal major associations between any of the studied polymorphisms in FATP1, UCP3, TNF-α, LEP, and GNB3 genes and the MS, suggesting that these polymorphisms are not major risk factors for the MS.

As is evident, association of gene variants, insulin resistance and dyslipidaemia is complex and has resulted in inconsistent findings in different studies. Moreover, the genetic studies till date generally have been of a small size and cross sectional in approach which limits analysis of trends in insulin sensitivity and lipid and lipoprotein levels in association with different gene variants. Ethnic differences in prevalence of various alleles have further led to a large variability in the results. However, even taking these limitations into account, gene variants and their interaction with environment appear to modulate glucose and lipoprotein metabolism.

Given the fact that the prevalence of coronary heart disease, stroke and all-cause mortality is increased several folds in subjects with the MS, even in the absence of baseline cardiovascular diseases and diabetes, it is important to better understand the factors that increase the susceptibility to the development of the MS. Genetic association studies may help in this respect

References

1. Meirhaeghe A, Amouyel P (2004). Impact of genetic variation of PPARgamma in humans. *Mol Genet Metab* **83**(1-2): 93-102.
2. Robitaille J, Brouillette C, Houde A, Lemieux S, Perusse L, Tchernof A, Gaudet D, Vohl MC (2004). Association between the PPARalpha-L162V polymorphism and components of the metabolic syndrome. *J Hum Genet* **49**(9): 482-489.
3. Gu HF, Almgren P, Lindholm E, Frittitta L, Pizzuti A, Trischitta V, Groop LC (2000). Association between the human glycoprotein PC-1 gene and elevated glucose and insulin levels in a paired-sibling analysis. *Diabetes* **49**(9): 1601-1603.
4. Pizzuti A, Frittitta L, Argiolas A, Baratta R, Goldfine ID, Bozzali M, Ercolino T, Scarlato G, Iacoviello L, Vigneri R *et al* (1999). A polymorphism (K121Q) of the human glycoprotein PC-1 gene coding region is strongly associated with insulin resistance. *Diabetes* **48**(9): 1881-1884.
5. Gonzalez-Sanchez JL, Zabena CA, Martinez-Larrad MT, Fernandez-Perez C, Perez-Barba M, Laakso M, Serrano-Rios M (2005). An SNP in the adiponectin gene is associated with decreased serum adiponectin levels and risk for impaired glucose tolerance. *Obes Res* **13**(5): 807-812.
6. Deeb SS, Zambon A, Carr MC, Ayyobi AF, Brunzell JD (2003). Hepatic lipase and dyslipidemia: interactions among genetic variants, obesity, gender, and diet. *J Lipid Res* **44**(7): 1279-1286.
7. Ishii T, Hirose H, Kawai T, Hayashi K, Maruyama H, Saito I, Saruta T (2001). Effects of intestinal fatty acid-binding protein gene Ala54Thr polymorphism and beta3-adrenergic receptor gene Trp64Arg polymorphism on insulin resistance and fasting plasma glucose in young to older Japanese men. *Metabolism* **50**(11): 1301-1307.
8. Lee YJ, Tsai JC (2002). ACE gene insertion/deletion polymorphism associated with 1998 World Health Organization definition of metabolic syndrome in Chinese type 2 diabetic patients. *Diabetes Care* **25**(6): 1002-1008.
9. Sheu WH, Lee WJ, Jeng CY, Young MS, Ding YA, Chen YT (1998). Angiotensinogen gene polymorphism is associated with insulin resistance in nondiabetic men with or without coronary heart disease. *Am Heart J* **136**(1): 125-131.
10. Abdollahi MR, Gaunt TR, Syddall HE, Cooper C, Phillips DI, Ye S, Day IN (2005). Angiotensin II type I receptor gene polymorphism: anthropometric and metabolic syndrome traits. *J Med Genet* **42**(5): 396-401.
11. Gregor PD, Sawadogo M, Roeder RG (1990). The adenovirus major late transcription factor USF is a member of the helix-loop-helix group of regulatory proteins and binds to DNA as a dimer. *Genes Dev* **4**(10): 1730-1740.
12. Pajukanta P, Lilja HE, Sinsheimer JS, Cantor RM, Lusis AJ, Gentile M, Duan XJ, Soro-Paavonen A, Naukkarinen J, Saarela J *et al* (2004). Familial combined hyperlipidemia is associated with upstream transcription factor 1 (USF1). *Nat Genet* **36**(4): 371-376.
13. Shieh BH, Sparkes RS, Gaynor RB, Lusis AJ (1993). Localization of the gene-encoding upstream stimulatory factor (USF) to human chromosome 1q22-q23. *Genomics* **16**(1): 266-268.

14. Pajukanta P, Nuotio I, Terwilliger JD, Porkka KV, Ylitalo K, Pihlajamaki J, Suomalainen AJ, Syvanen AC, Lehtimaki T, Viikari JS *et al* (1998). Linkage of familial combined hyperlipidaemia to chromosome 1q21-q23. *Nat Genet* **18**(4): 369-373.

15. Xiang K, Wang Y, Zheng T, Jia W, Li J, Chen L, Shen K, Wu S, Lin X, Zhang G *et al* (2004). Genome-wide search for type 2 diabetes/impaired glucose homeostasis susceptibility genes in the Chinese: significant linkage to chromosome 6q21-q23 and chromosome 1q21-q24. *Diabetes* **53**(1): 228-234.

16. Das SK, Hasstedt SJ, Zhang Z, Elbein SC (2004). Linkage and association mapping of a chromosome 1q21-q24 type 2 diabetes susceptibility locus in northern European Caucasians. *Diabetes* **53**(2): 492-499.

17. Johnson JD, Han Z, Otani K, Ye H, Zhang Y, Wu H, Horikawa Y, Misler S, Bell GI, Polonsky KS (2004). RyR2 and calpain-10 delineate a novel apoptosis pathway in pancreatic islets. *J Biol Chem* **279**(23): 24794-24802.

18. Sreenan SK, Zhou YP, Otani K, Hansen PA, Currie KP, Pan CY, Lee JP, Ostrėga DM, Pugh W, Horikawa Y *et al* (2001). Calpains play a role in insulin secretion and action. *Diabetes* **50**(9): 2013-2020.

19. Patel YM, Lane MD (1999). Role of calpain in adipocyte differentiation. *Proc Natl Acad Sci U S A* **96**(4): 1279-1284.

20. Yajima Y, Kawashima S (2002). Calpain function in the differentiation of mesenchymal stem cells. *Biol Chem* **383**(5): 757-764.

21. Hanis CL, Boerwinkle E, Chakraborty R, Ellsworth DL, Concannon P, Stirling B, Morrison VA, Wapelhorst B, Spielman RS, Gogolin-Ewens KJ *et al* (1996). A genome-wide search for human non-insulin-dependent (type 2) diabetes genes reveals a major susceptibility locus on chromosome 2. *Nat Genet* **13**(2): 161-166.

22. Horikawa Y, Oda N, Cox NJ, Li X, Orho-Melander M, Hara M, Hinokio Y, Lindner TH, Mashima H, Schwarz PE *et al* (2000). Genetic variation in the gene encoding calpain-10 is associated with type 2 diabetes mellitus. *Nat Genet* **26**(2): 163-175.

23. Weedon MN, Schwarz PE, Horikawa Y, Iwasaki N, Illig T, Holle R, Rathmann W, Selisko T, Schulze J, Owen KR *et al* (2003). Meta-analysis and a large association study confirm a role for calpain-10 variation in type 2 diabetes susceptibility. *Am J Hum Genet* **73**(5): 1208-1212.

24. Song Y, Niu T, Manson JE, Kwiatkowski DJ, Liu S (2004). Are variants in the CAPN10 gene related to risk of type 2 diabetes? A quantitative assessment of population and family-based association studies. *Am J Hum Genet* **74**(2): 208-222.

25. Baier LJ, Permana PA, Yang X, Pratley RE, Hanson RL, Shen GQ, Mott D, Knowler WC, Cox NJ, Horikawa Y *et al* (2000). A calpain-10 gene polymorphism is associated with reduced muscle mRNA levels and insulin resistance. *J Clin Invest* **106**(7): R69-73.

26. Stoffel M, Duncan SA (1997). The maturity-onset diabetes of the young (MODY1) transcription factor HNF4alpha regulates expression of genes required for glucose transport and metabolism. *Proc Natl Acad Sci U S A* **94**(24): 13209-13214.

27. Sander M, German MS (1997). The beta cell transcription factors and development of the pancreas. *J Mol Med* **75**(5): 327-340.

28. Silander K, Mohlke KL, Scott LJ, Peck EC, Hollstein P, Skol AD, Jackson AU, Deloukas P, Hunt S, Stavrides G *et al* (2004). Genetic variation near the hepatocyte nuclear factor-4 alpha gene predicts susceptibility to type 2 diabetes. *Diabetes* **53**(4): 1141-1149.

29. Auwerx J (1999). PPARgamma, the ultimate thrifty gene. *Diabetologia* **42**(9): 1033-1049.

30. Greene ME, Blumberg B, McBride OW, Yi HF, Kronquist K, Kwan K, Hsieh L, Greene G, Nimer SD (1995). Isolation of the human peroxisome proliferator activated receptor gamma cDNA: expression in hematopoietic cells and chromosomal mapping. *Gene Expr* **4**(4-5): 281-299.

31. Altshuler D, Hirschhorn JN, Klannemark M, Lindgren CM, Vohl MC, Nemesh J, Lane CR, Schaffner SF, Bolk S, Brewer C *et al* (2000). The common PPARgamma Pro12Ala polymorphism is associated with decreased risk of type 2 diabetes. *Nat Genet* **26**(1): 76-80.

32. Luan J, Browne PO, Harding AH, Halsall DJ, O'Rahilly S, Chatterjee VK, Wareham NJ (2001). Evidence for gene-nutrient interaction at the PPARgamma locus. *Diabetes* **50**(3): 686-689.

33. Lindi VI, Uusitupa MI, Lindstrom J, Louheranta A, Eriksson JG, Valle TT, Hamalainen H, Ilanne-Parikka P, Keinanen-Kiukaanniemi S, Laakso M *et al* (2002). Association of the Pro12Ala polymorphism in the PPAR-gamma2 gene with 3-year incidence of type 2 diabetes and body weight change in the Finnish Diabetes Prevention Study. *Diabetes* **51**(8): 2581-2586.

34. Ambye L, Rasmussen S, Fenger M, Jorgensen T, Borch-Johnsen K, Madsbad S, Urhammer SA (2005). Studies of the Gly482Ser polymorphism of the peroxisome proliferator-activated receptor gamma coactivator 1alpha (PGC-1alpha) gene in Danish subjects with the metabolic syndrome. *Diabetes Res Clin Pract* **67**(2): 175-179.

35. Dahlman I, Wahrenberg H, Persson L, Arner P (2004). No association of reported functional protein tyrosine phosphatase 1B 3' UTR gene polymorphism with features of the metabolic syndrome in a Swedish population. *J Intern Med* **255**(6): 694-695.

36. Costa LA, Canani LH, Maia AL, Gross JL (2002). The ACE insertion/deletion polymorphism is not associated with the metabolic syndrome (WHO Definition) in Brazilian type 2 diabetic patients. *Diabetes Care* **25**(12): 2365-2366; author reply 2366-2367.

37. Erkkila AT, Lindi V, Lehto S, Pyorala K, Laakso M, Uusitupa MI (2002). Variation in the fatty acid binding protein 2 gene is not associated with markers of metabolic syndrome in patients with coronary heart disease. *Nutr Metab Cardiovasc Dis* **12**(2): 53-59.

38. Lee SC, Pu YB, Thomas GN, Lee ZS, Tomlinson B, Cockram CS, Critchley JA, Chan JC (2000). Tumor necrosis factor alpha gene G-308A polymorphism in the metabolic syndrome. *Metabolism* **49**(8): 1021-1024.

39. Thomas GN, Tomlinson B, Chan JC, Lee ZS, Cockran CS, Critchley JA (2000). An insulin receptor gene polymorphism is associated with diastolic blood pressure in Chinese subjects with components of the metabolic syndrome. *Am J Hypertens* **13**(7): 745-752.

40. Thomas GN, Tomlinson B, Chan JC, Young RP, Critchley JA (2000). The Trp64Arg polymorphism of the beta3-adrenergic receptor gene and obesity in Chinese subjects with components of the metabolic syndrome. *Int J Obes Relat Metab Disord* **24**(5): 545-551.

41. Rissanen J, Kuopusjarvi J, Pihlajamaki J, Sipilainen R, Heikkinen S, Vanhala M, Kekalainen P, Kuusisto J, Laakso M (1997). The Trp64Arg polymorphism of the beta 3-Adrenergic receptor gene. Lack of association with NIDDM and features of insulin resistance syndrome. *Diabetes Care* **20**(8): 1319-1323.

42. Schrauwen P, Xia J, Walder K, Snitker S, Ravussin E (1999). A novel polymorphism in the proximal UCP3 promoter region: effect on skeletal muscle UCP3 mRNA expression and obesity in male non-diabetic Pima Indians. *Int J Obes Relat Metab Disord* **23**(12): 1242-1245.

43. Cassell PG, Saker PJ, Huxtable SJ, Kousta E, Jackson AE, Hattersley AT, Frayling TM, Walker M, Kopelman PG, Ramachandran A *et al* (2000). Evidence that single nucleotide polymorphism in the uncoupling protein 3 (UCP3) gene influences fat distribution in women of European and Asian origin. *Diabetologia* **43**(12): 1558-1564.

44. Damcott CM, Feingold E, Moffett SP, Barmada MM, Marshall JA, Hamman RF, Ferrell RE (2004). Genetic variation in uncoupling protein 3 is associated with dietary intake and body composition in females. *Metabolism* **53**(4): 458-464.

45. Halsall DJ, Luan J, Saker P, Huxtable S, Farooqi IS, Keogh J, Wareham NJ, O'Rahilly S (2001). Uncoupling protein 3 genetic variants in human obesity: the c-55t promoter polymorphism is negatively correlated with body mass index in a UK Caucasian population. *Int J Obes Relat Metab Disord* **25**(4): 472-477.

46. Lindholm E, Klannemark M, Agardh E, Groop L, Agardh CD (2004). Putative role of polymorphisms in UCP1-3 genes for diabetic nephropathy. *J Diabetes Complications* **18**(2): 103-107.

47. Otabe S, Clement K, Dina C, Pelloux V, Guy-Grand B, Froguel P, Vasseur F (2000). A genetic variation in the 5' flanking region of the UCP3 gene is associated with body mass index in humans in interaction with physical activity. *Diabetologia* **43**(2): 245-249.

48. Meirhaeghe A, Amouyel P, Helbecque N, Cottel D, Otabe S, Froguel P, Vasseur F (2000). An uncoupling protein 3 gene polymorphism associated with a lower risk of developing Type II diabetes and with atherogenic lipid profile in a French cohort. *Diabetologia* **43**(11): 1424-1428.

49. Shen H, Xiang K, Jia W (2002). [Effects of uncoupling protein 3 gene -55 C—>T variant on lipid metabolism, body fat, its distribution and non-insulin-dependent diabetes mellitus in Chinese]. *Zhonghua Yi Xue Yi Chuan Xue Za Zhi* **19**(4): 317-321.

50. Meier U, Gressner AM (2004). Endocrine regulation of energy metabolism: review of pathobiochemical and clinical chemical aspects of leptin, ghrelin, adiponectin, and resistin. *Clin Chem* **50**(9): 1511-1525.

51. Montague CT, Farooqi IS, Whitehead JP, Soos MA, Rau H, Wareham NJ, Sewter CP, Digby JE, Mohammed SN, Hurst JA *et al* (1997). Congenital leptin deficiency is

associated with severe early-onset obesity in humans. *Nature* **387**(6636): 903-908.

52. Strobel A, Issad T, Camoin L, Ozata M, Strosberg AD (1998). A leptin missense mutation associated with hypogonadism and morbid obesity. *Nat Genet* **18**(3): 213-215.

53. Hager J, Clement K, Francke S, Dina C, Raison J, Lahlou N, Rich N, Pelloux V, Basdevant A, Guy-Grand B *et al* (1998). A polymorphism in the 5' untranslated region of the human ob gene is associated with low leptin levels. *Int J Obes Relat Metab Disord* **22**(3): 200-205.

54. Dallongeville J, Helbecque N, Cottel D, Amouyel P, Meirhaeghe A (2003). The Gly16—>Arg16 and Gln27—>Glu27 polymorphisms of beta2-adrenergic receptor are associated with metabolic syndrome in men. *J Clin Endocrinol Metab* **88**(10): 4862-4866.

55. Siffert W (2005). G protein polymorphisms in hypertension, atherosclerosis, and diabetes. *Annu Rev Med* **56**: 17-28.

56. Yamamoto M, Abe M, Jin JJ, Wu Z, Tabara Y, Mogi M, Kohara K, Miki T, Nakura J (2004). Association of GNB3 gene with pulse pressure and clustering of risk factors for cardiovascular disease in Japanese. *Biochem Biophys Res Commun* **316**(3): 744-748.

57. Isomaa B, Almgren P, Tuomi T, Forsen B, Lahti K, Nissen M, Taskinen MR, Groop L (2001). Cardiovascular morbidity and mortality associated with the metabolic syndrome. *Diabetes Care* **24**(4): 683-689.

58. Lakka HM, Laaksonen DE, Lakka TA, Niskanen LK, Kumpusalo E, Tuomilehto J, Salonen JT (2002). The metabolic syndrome and total and cardiovascular disease mortality in middle-aged men. *Jama* **288**(21): 2709-2716.

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Insulin Resistance
Pathological Basis and Clinical Significance

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Prologue : In their classical landmark paper describing the radioimmunoassay of insulin, Solomon Berson and Rosalyn Yalow defined insulin resistance as a *"state of a cell, tissue, system or body in which greater than normal amounts of insulin are required to elicit a quantitatively normal response"*(1). David Kipins, in a chapter on insulin physiology, in the monumental monograph : Methods in Investigative and Diagnostic Endocrinology' edited by Berson and Yalow and published prior to the demise of Berson, clearly states(2) :

'Skeletal and cardiac muscle, adipose tissue and the liver are the principal insulin responsive tissues of the body. Exposure of these tissues to physiological levels of insulin *in vivo* results in prompt and readily detectable changes in various parameters of carbohydrate, lipid, and protein metabolism. Peripheral nerve, ciliary muscle, cartilage, fibroblasts, circulating lymphocytes and granulocytes, and the arterial wall have also been reported to respond to the hormone, but the physiological relevance of these observations with respect of either total body metabolic homeostasis or individual tissue viability remains to be elucidated — ——. It is well recognized that the insulin sensitivity of target tissues is influenced by a variety of metabolic states; insulin resistance is characteristically associated with obesity, trauma, pregnancy, acromegaly and hyperadrenocorticism.'

This descriptive narrative provides the essential context to understand the effects of differential insulin sensitivity (and resistance) in various insulin responsive tissues and organs, besides the possibility of differential sensitivity with regard to the effects on carbolydrate, lipid, and protein metabolism. It also alludes to the effects on arterial wall, and circulating elements such as lymphocytes, granulocytes and platelets.

Measurement of Insulin Sensitivity : The most frequently used method of measuring insulin sensitivity is by Euglycemic clamp technique(3). In all

research laboratories, use of this technique is considered essential to demonstrate impaired sensitivity i.e. insulin resistance. The technique is highly reproducible, although time consuming, requiring skill and expertise.

The other computer program based techniques include FSIVGTT (Frequently sampled intravenous glucose tolerance test) (4) and CIGMA (Continuous infusion of glucose with model assessment) (5). The values obtained by using these techniques generally correlate well with the results obtained with the euglycemic clamp technique.

A number of techniques have also been described using standard oral glucose tolerance tests (OGTT) wherein 75 gm. glucose load is given orally after a 10-16 hour fast (WHO, 1980)(6). Blood samples are collected at 30-minute intervals following the administration of glucose load. The test duration may be 2 hours (WHO, 1980) or may be extended to 3 or 4 hours. Glucose and insulin are measured in each sample. The data is analysed using various indices, some of which depend on $AUC_{glucose}$ (area under glucose curve) and $AUC_{insulin}$ (area under insulin curve)(7). Other investigators have used I_{mean} (mean plasma insulin during OGTT; mlU/L) and G_{mean} (mean glucose concentration during OGTT (mg/dl).

Mathematical models have been used in clinical practice wherein only fasting glucose concentration, and fasting insulin concentration have been used to calculate insulin sensitivity. For example, Raynaud Index uses the formula $R1 = 40/1_0$ where 1_0 is the fasting insulin level (μU/ml)(8). Likewise, fasting glucose to insulin ratio (FGIR) has been recommended using the formula :

$$FGIR = G_0/1_0$$ (G_0 = fasting glucose, mg/dl; 1_0 = fasting insulin, μU/ml)

The major problem with a single sample-based calculation of insulin sensitivity is due to the fact that insulin secretion occurs in an oscillatory manner, thereby rendering single value-based measurement amenable to possible error. Although it has been suggested that collection of three samples at intervals of five minutes (and using pooled sample for subsequent measurements) may minimize the error, it must be remembered that 'oscillations' or 'pulses' of insulin secretion occur at two different periodicities : (i) rapid small amplitude oscillations which occur every 10-15 minutes and are superimposed on (ii) slower, larger amplitude ultradian oscillations with periods ranging from 80-150 minutes(9).

In a general mathematical model which incorporates β-cell kinetics and a gastrointestinal absorption term for glucose insulin feedback, and comprises of a set of four nonlinear, coupled ordinary differential equations(10), numerical simulations showed that glucose and insulin levels oscillate before reaching a steady state. Interestingly, glucose oscillations are seen

to lead the insulin oscillations. In our study we observed that the time period of oscillations is about 90 minutes and the glucose peak precedes insulin peak by about 06 minutes(10).

Homeostasis Model Assessment (HOMA): Insulin resistance (and sensitivity as measured by this method) is one of the most commonly used parameters in clinical metabolic research and is based on a model of insulin-glucose interaction, with measurement of fasting glucose (G_0 : mmol/L) and fasting insulin (1_0 : μIU/ml).

$$IR_{HOMA} = \frac{1_0 \times G_0}{22.5}$$

In the mathematical model proposed by us(11) and referred to in the preceding paragraph, it was further shown that in obese controls with normal glucose tolerance, there was a four-fold increase in β-cell function, an increased peripheral resistance to insulin action, and an increased initial rate of gastro-intestinal absorption as compared to non-obese controls. In contrast, in both obese and non-obese subjects with type 2 diabetes mellitus (T2DM), the changes appear to occur as a result of decreased β-cell capacity and function and an increased peripheral resistance to insulin action(12).

Insulin Resistance : metabolic basis and clinical significance : The preceding mini-review makes it abundantly clear that although the term is frequently used to indicate impaired insulin-stimulated glucose disposal as generally measured with the hyperinsulinemic euglycemic clamp technique, it must be clearly understood that insulin resistance may occur at the level of one or more of the insulin sensitive tissues such as the skeletal muscle, liver, and the adipose tissue(13). Thus not only muscle glucose uptake but also inhibition of adipose tissue lipolysis and suppression of hepatic glucose production, all regulated by insulin, must be considered both individually and jointly for a comprehensive understanding of insulin action and insulin resistance. The subject is recently reviewed by us(14). The account that follows draws upon our published review.

There is a paucity of data examining the insulin dose response characteristics of stimulation of glucose uptake (muscle) and suppression of glucose production (liver) in normals and in subjects with T2 DM. A well designed study(15) showed in the normal subjects a significant shift to the right of the dose-response curve for glucose uptake with EC_{50} of 58 μU/ml. In contrast, the corresponding EC_{50} for suppression of glucose production was approximately one-half of this i.e. 26 μU/ml. Thus, with low physiological increments in plasma insulin, the liver is the primary determinant of whole body glucose homeostasis. In subjects with T2DM, there was a marked shift of a similar magnitude of both these curves to the right, with EC_{50} for glucose uptake of 118 μU/ml. and EC_{50} for glucose production of 66 μU/ml.

In contrast to EC_{50} for glucose uptake and glucose production, data on whole

body lipolysis during stepwise hyperinsulinemic-euglycemic clamp studies show that EC_{50} for suppression of lipolysis in normal subjects is much lower and ranges between 7 and 16 μU/ml(16,17). Thus, suppression of lipolysis seems to be the most sensitive of insulin actions, with the dose response curve of adipose tissue distinctly to the left of the corresponding curves for glucose production and glucose uptake. It is also obvious from these studies that inhibition of lipolysis can be achieved at a fasting insulin concentration which is well within the normal range, and suppression of glucose production can also be achieved within the physiological range of postparandial insulin concentration. Thus, while the term Insulin Resistance Syndrome (IRS) may provide a conceptual framework for diverse cardiometabolic risk factors grouped together as Metabolic Syndrome (MS), in depth study of insulin action and resistance at different sites (muscle, liver, adipocyte) is required to compare and contrast the metabolic defects in each of the clinical disorders including obesity, T2 DM, Coronary Heart Disease (CHD), Hypertenion, Polycystic Ovaries Syndrome (PCOS) and Non-Alcoholic Fatty Liver Disease (NAFLD), and finally to investigate their relationship, if any, with high and/or low birth weights.

The need of such studies becomes imperative in the light of information recently made available as a result of disruption of insulin receptor gene in specific target tissues. Mice without insulin receptors in skeletal muscle do not develop diabetes, suggesting that insulin sensitivity (and glucose uptake) in skeletal muscle might not always be a primary cause of diabetes(18). However, these mice develop increased fat mass, elevated serum triglycerides, and high levels of circulating FFA, suggesting that insulin resistance in the muscle contributes significantly to altered fat metabolism in these animals. Likewise, mice lacking hepatic insulin receptors also fail to develop diabetes, as compensatory hyperinsulinemia apparently maintains fasting normoglycaemia (19).

In summary, resistance to insulin may be viewed in the context of multisystem insulin resistance, and the clinical sequalae must depend not only on the system(s) exhibiting resistance in a more severe form, but also on the compensatory mechanism(s), such as β-cell function, operating to respond to the challenge posed by insulin resistance.

Free Fatty Acids and Insulin Resistance: The plasma FFA concentration is determined by: (i) the rate of FFA production (lipolysis) in the adipose tissue; and (ii) the rate of uptake from plasma either for oxidation or for reesterification to triglycerides. FFA from the visceral adipose tissue drain directly into the portal vein and reach the liver. The rate of lipolytic activity is higher in the abdominal visceral adipocytes(20). Subjects with central (visceral) adiposity have higher levels of FFA in portal vein, in addition to the daylong elevation of FFA in the peripheral

plasma. Chronically elevated plasma FFA can lead to insulin resistance in muscle and liver(21,22). The thesis that abnormalities of FFA metabolism may be involved in the etiology of T2 DM (then called maturity-onset diabetes mellitus) was first propounded by Randle et al in 1963 who suggested a fundamental role of glucose-fatty acid cycle in the regulation of energy balance. Based on their studies on rat cardiac muscle in vitro, it was concluded that the rate of fat oxidation increased relative to carbohydrate oxidation in response to elevated FFA concentration(23). It was suggested that in the fasting state with low basal insulin, there was an increase in the oxidation of fatty acids by muscle (with reciprocal reduction of glucose uptake). Opposite was the case in the fed-state where enhanced insulin secretion increases glucose uptake by muscle, in addition to inhibiting lipolysis in the adipose tissue. Concluding their classical paper in the Lancet, Randle et al stated(23) : 'We propose that the interactions between glucose and fatty-acid metabolism in muscle and adipose tissue take the form of a cycle, the glucose fatty-acid cycle, which is fundamental to control of blood glucose and fatty acid concentrations and insulin sensitivity'. Nearly three decades later, these observations were confirmed in healthy human skeletal muscle wherein a decrease in carbohydrate oxidation in association with an increase in fat oxidation was demonstrated following 1-hour of lipid infusion, under euglycaemic – hyperinsulinemic clamp(24). Although the precise molecular basis of the relationship between circulating FFA levels and insulin resistance in the muscle remains uncertain, it is recognized that chronically elevated FFA levels may also impair insulin secretion from the β – cells (lipotoxicity)(25), in addition to enhancing hepatic glucose output(26). The latter may be due to a combination of increased availability of FFA for gluconeogenesis, and decreased sensitivity (resistance) to the action of insulin at the hepatocyte.

Even though the conceptual framework of Randle's hypothesis remains intact, more recent studies have shown that FFAs and their metabolites also inhibit insulin signaling, and glucose transport, in addition to affecting the activities of several enzymes involved in glucose metabolism(27). Following a 5-h infusion of lipid in healthy volunteers, a significant decrease in intracellular glucose was demonstrated, supporting the hypothesis that elevated FFA induce insulin resistance principally at the level of glucose transport(28). This could be either due to a direct effect on glucose transporter GLUT 4, or mediated through an indirect effect by modifying upstream signaling events, notably at the level of phosphatidylinositol 3-kinase (P1 3-kinase). That the latter may be the case was demonstrated by the fact that increase in P 1 3-kinase activity in response to insulin stimulation was nearly completely abolished in human volunteers given lipid infusion(29). Additional studies

show that high FFA concentration may effect several upstream proteins in the signaling pathway, including IRS-1 and protein kinase C theta (PKC θ)(30). It has been proposed that elevated FFA may activate PKC θ, with the resulting decrease in IRS-1 tyrosine phosphorylation, a suppression of P1 3-kinase activity, reduced GLUT translocation, and cultiminating in a reduction of glucose transport(31). Such lipid infusions have, therefore, been shown to closely resemble the effects of chronically elevated FFA levels such as inhibition of glucose transport and phosphorylation, glycogen synthase and pyruvate dehydrogenase activity, and insulin signaling through the IRS-1, PKC θ, and P1 3-Kinase pathways.

Additional evidence for the role of intracellular lipid in mediating insulin resistance has been obtained from transgenic mice with muscle-specific and liver-specific overexpression of lipoprotein lipase (LPL). Muscle-LPL-overexpressing mice had a threefold increase in muscle triglyceride content and were insulin resistant due to (i) decrease in insulin-stimulated glucose transport and (ii) a reduced insulin activation of IRS-1 associated P1 3-kinase activity. In contrast, liver-LPL-overexpressing mice had a two-fold increase in liver triglyceride content. These mice were insulin resistant primarily due to the impaired ability of insulin to suppress endogenous glucose production(32). These defects in insulin signaling (and action) were associated with increases in intracellular fat metabolites (i.e. diacylglycerol, fatty acylcoenzyme A). Thus there is a causal relationship between intracellular accumulation of fat metabolites and insulin resistance mediated through changes in insulin signaling pathway.

In a recent study aimed at investigating the molecular mechanism (s) underlying the biochemical basis of insulin resistance due to the effect of elevated FFA on the muscle, a triglyceride emulsion was infused in healthy subjects for 48 hours, followed by muscle biopsis (vastus lateralis muscle), microarray analysis, quantitative real time PCR, and immunoblots. After lipid infusion, extracellular matrix genes and connective tissue growth factor were significantly over expressed. In contrast, nuclear encoded mitochondrial genes and PGC-1α (peroxisome proliferator activated receptor γ – coactivator –1α) expression were decreased(33). As PGC-1α is the transcriptional coactivator that initiates the expression of several genes coding for mitochondrial proteins, a decrease in PGC-1 expression may result in decreased expression of a number of metabolic and nuclear encoded mitochondrial genes involved in electron transport and oxidative phosphorylation. Indeed, a decreased expression of nuclear encoded mitochondrial genes, accompanied by a decreased expression of PGC-1α, has been demonstrated in insulin resistant subjects(34,35).

Notwithstanding the effect on nuclear encoded mitochondrial genes, a marked increase in expression of extracellular matrix-related genes following lipid infusion was of considerable interest(33). Such a pattern characterizes inflammatory

response leading to extracellular matrix remodelling and fibrosis. Thus chronic elevation of FFA may result in inflammation-associated extracellular matrix changes in the skeletal muscle. Such fibrotic inflammatory responses are mediated by the Connective tissue growth factor (CTGF), also termed CCN2, a $38KD_a$ protein belonging to the CNN family(36). Fig. 1 projects a conceptual model of the molecular basis of insulin resistance(14). Metabolic activity in the adipocytes, through changes in the circulating levels of adipokines and elevated levels of FFA may result in a decreased expression of PGC-1α and several nuclear encoded mitochondrial genes, thereby reducing oxidative phosphorylation, in addition to producing defects in glucose transport and insulin signaling pathways. The resultant insulin resistant state in turn increases lipolysis, further increasing the levels of circulating FFA. Adipokines, such as resistin, lead to increase in hepatic fat, causing insulin

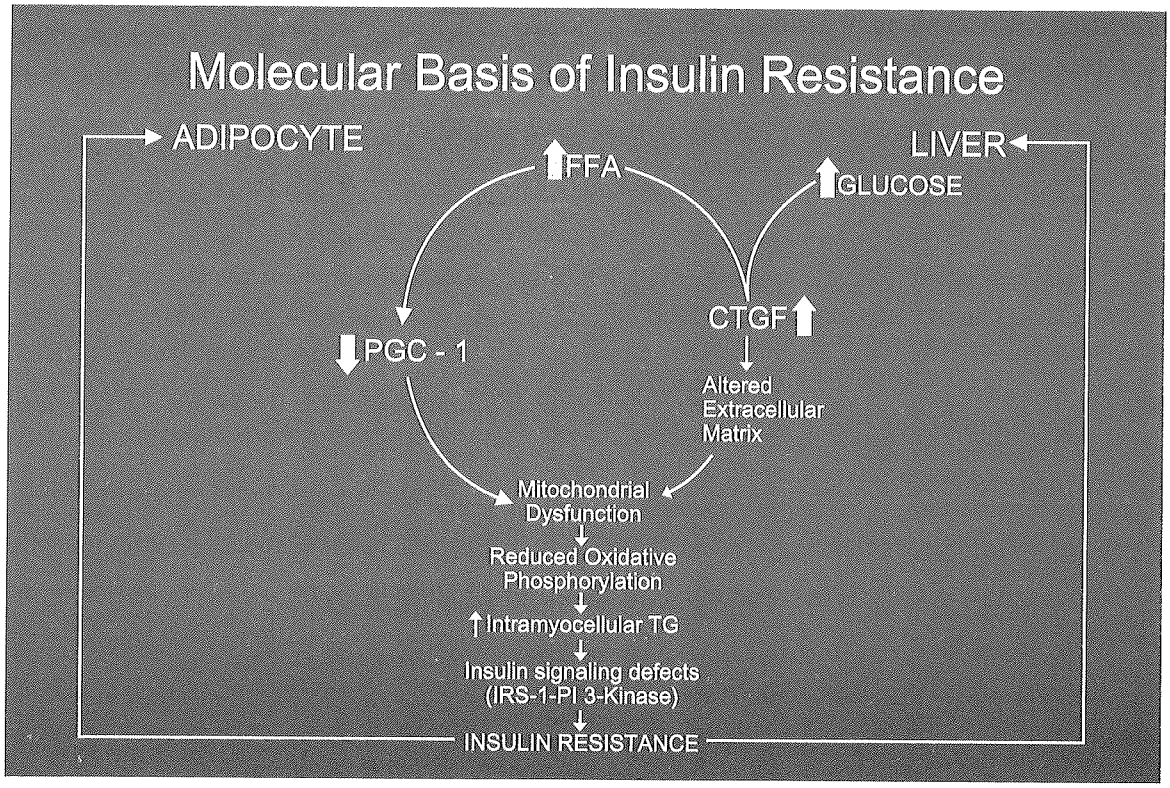

Fig. 1 : Decreased expression of nuclear encoded mitochondrial genes and increased expression of extracellular matrix - related genes may contribute to the molecular basis of insulin resistance (for explanation, see text). From Bajaj and Bajaj (14) with permission of Editor and Publisher

PGC - 1α : peroxisome proliferator activated receptor γ - coactivator - 1α; CTGF : connective tissue growth factor; FFA : free fatty acids.

resistance in the liver, and increased hepatic glucose output. In contrast, adiponectin reduces hepatic fat content. Increased levels of glucose and FFA enhance the expression of CTGF, leading to altered cellular matrix. It is obvious that new therapeutic targets aimed at increasing PGC-1α activity and reducing CTGF activity bear therapeutic potential.

There is evidence that CTGF mediates fibrotic changes at multiple sites i.e. atheromatous plaques(37); mesangium in the glomerulus(38); myocardium following ischemic injury(39) and activated hepatic stellate cells(40). As angiotensin II (acting through angiotensin receptor 1) increases the expression of CTGF, the current use of specific angiotensin receptor blockers in the prevention and management of diabetic nephropathy seems to be most rational. Of major interest is the observation that liver biopsis from nondiabetics and T2DM patients with non-alcoholic steatohepatitis (NASH) showed enhanced expression of CTGF that correlated with the degree of fibrosis(40). As the CTGF expression is increased in the liver from Zucker obese rats in association with lipid abnormalities and fatty liver in this animal model of

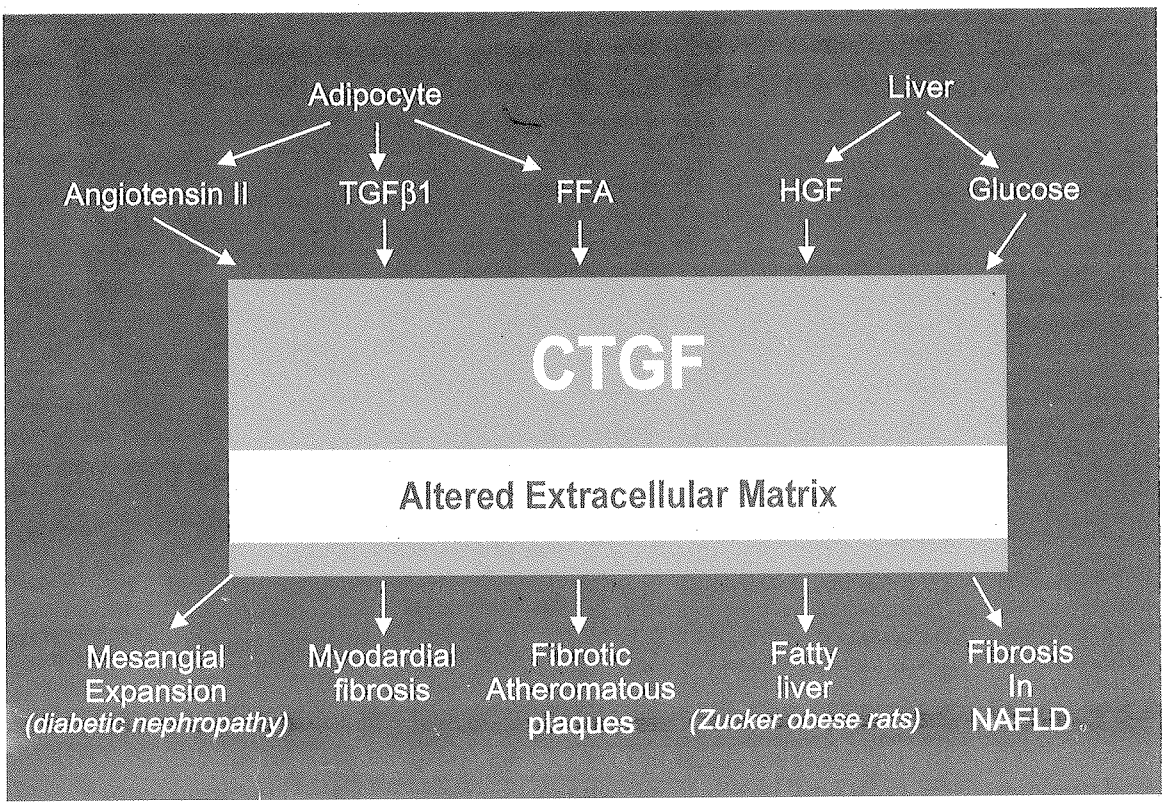

Fig. 2 : Enhanced expression of connective tissue growth factor and its possible consequences. From Bajaj and Bajaj (14) with permission of Editor and Publisher.

insulin resistance, it may be a rational molecular target for future drug development. Indeed, an angiotensin II type 1 receptor antagonist, olmesartan medoxomil, has been shown to improve experimental liver fibrosis by suppression of proliferation and collagen synthesis in hepatic stellate cells(41). A **unifying** model of several diseases included in the Metabolic Syndrome, and their complications, is shown in Fig. 2 which may also portend possible molecular sites for future drug development(14).

Adiponectin and Insulin resistance : Hypoadiponectinemia characterizes T2DM, a well recognized insulin-resistant state. Recent studies have provided evidence that increased hepatic fat content is an important determinant of hepatic insulin resistance in type 2 diabetic patients(42). Thiazolidinediones have been shown to reduce hepatic fat content and improve hepatic insulin sensitivity in patients with T2DM(42). Thiazolidinediones initiate their action by binding PPARγ, primarily located on adipocytes(43), and thereby increasing plasma adiponectin levels. Indirect evidence suggests that adiponectin might mediate some of the insulin-sensitizing effects of PPARγ agonists.

The first-ever clinical study aimed at investigating the effect of long term (14 weeks) administration of 45 mg. pioglitazone daily in subjects with T2DM resulted in a three-fold increase in plasma adiponectin which correlated inversely with endogenous (hepatic) glucose production. There was also a significant inverse correlation of plasma adiponectin with hepatic fat content. Higher the plasma adiponectin levels, lower the hepatic fat content. Thus the increase in plasma adiponectin following pioglitazone therapy is strongly associated with a decrease in hepatic fat content and enhanced hepatic and peripheral insulin sensitivity(44).

Put together with a similar study(45), there is unequivocal evidence that pioglitazone (thiozolidinedione) treatment of subjects with T2DM ***increases*** plasma adiponectin and ***decreases*** plasma resistin levels, resulting in a decrease in hepatic fat content and a reduction in hepatic glucose production. Indicators of net therapeutic benefit included a decrease in fasting plasma glucose as well as a lowering of the HbA1c and serum triglyceride levels(44).

In addition to its metabolic effects, adiponectin has also been shown to modulate endothelial inflammatory response through TNF-α-induced expression of endothelial adhesion molecules(46). *In vitro* studies in human aortic endothelial cells have shown that human recombinant adiponectin not only suppresses endothelial expression of adhesion molecules but also decreases the proliferation of vascular smooth muscle cells, and reduces lipid accumulation in macrophages, thereby modulating transformation of macrophages to foam cells(47).

Two clinical studies published recently provide interesting data linking the metabolic and anti-inflammatory roles of adiponectin. In a study of 77 subjects who had diabetes or were at high risk to develop diabetes, there was a significant negative correlation between circulating levels of adiponectin and C-reactive protein (CRP), plasminogen activator inhibitor-1 (PAI-1), and tissue plasminogen activator (tPA)(48). These negative associations remained significant after adjusting for gender and BMI. This study reinforces earlier observation regarding the protective role of adiponectin against inflammation and endothelial dysfunction, and provides evidence of its negative association with tPA, which is known to play a role in impaired fibrinolysis. A similar study in women with prior gestational diabetes mellitus (pGDM) who are known to be at higher risk of developing T2DM and associated cardiovascular complications, showed that plasma adiponectin was significantly lower in pGDM as compared to women with normal glucose tolerance during pregnancy. The differences remained statistically significant even after adjustment for body fat mass. Equally significant were the differences in the levels of PAI-1 and ultrasensitive CRP which were higher in the pGDM group. It was concluded that lower plasma adiponectin concentrations characterize women with previous GDM independently of the prevailing glucose tolerance, insulin sensitivity or the degree of obesity and are associated with subclinical inflammation and atherogenic parameters(49).

Thus, the role of adiponectin as a mediator of insulin resistance and an integrator of metabolic and inflammatory signals underlying obesity, T2DM, and coronary heart disease has assumed considerable significance, both in terms of its potential as a part of preventive strategies, and also as a prototype molecule for the development of new analogues and related compounds aimed at therapeutic intervention. Further, development of PPAR-γ agonists which increase endogenous adiponectin may be equally promising and rewarding.

Pathophysiological basis of metabolic syndrome : The foregoing narrative clearly delineates a constellation of risk factors of metabolic origin which have a close association with increased risk of T2DM, hypertension and atherosclerotic cardiovascular disease (CVD). Although there is a considerable agreement on the role and place of various cardiometabolic risk factors, the terminology of metabolic syndrome has raised intense debate(50). Nevertheless, the recognised risk factors include derangements of lipid metabolism (atherogenic dyslipidemia) comprising an increased levels of apoliprotein B, triglycerides, small LDL particles, and low levels of high density lipoproteins (HDL). Also present may be disturbances of

carbohydrate metabolism such as impaired fasting glucose, impaired glucose tolerance, or diabetes mellitus. A prothrombotic state comprising of an increase in procoagulant factors such as fibrinogen and factor VII, alongwith increased anti-fibrinolytic factors such as PAI-1, and endothelial dysfunction are important constituents. An associated proinflammatory state characterized by elevation of circulating cytokines i.e. TNF-α, resistin and IL-6 in addition to an increase in acute phase reactant (CRP) is generally observed. A significant reduction in circulating adiponectin is a contributory factor. The most important clincopathological basis is obesity, especially trunkal obesity or visceral adiposity, and the key molecular link is insulin resistance at various levels, and in different compartments *(vide supra)*.

The causes of the 'metabolic syndrome' are complex and as suggested above, involve hormonal and metabolic factors. Genetic susceptibility is described and gene-environment (lifestyle) interactions are well recognized. Although obesity, especially central adiposity remains a key clinical link, its definition based on ethnicity-specific cut-off values of waist circumference has been delineated only recently (Table(51)). A combination of accurately measured waist circumference, along with BMI {Wt. (kg) / Ht. (in meters)^2} may be more informative. It is however suggested that a single accurate measurement of waist circumference if culturally permissible, should be the preferred choice.

Ethnicity-specific values for waist circumference (51) :

Country/ ethnic group		Waist circumference (cm) (as measure of central obesity)
Europids	Male	≥ 94
	Female	≥ 80
South Asians	Male	≥ 90
	Female	≥ 80
Chinese	Male	≥ 90
	Female	≥ 80
Japanese	Male	≥ 90
	Female	≥ 80

While recommending these cut-off points for waist circumference, it is emphasized that 'these are pragmatic cut-off points and better data are required to link them to risk. It is also mentioned that ethnicity should be the basis for classification, not the country of residence' (51).

Epilogue : While the purists may continue to argue about the diagnostic criteria (WHO(52) or IDF(53) or NCEP : ATP III(54)) and the cell biologists may continue their efforts at localizing molecular lesion (s) underlying insulin resistance and deranged adipocyte biology, there is enough information already available to launch rationally sound public health measures at the *individual, community* and *national* levels(50). Time to act was yesterday : to-day may already be too late!

References :

1. Yalow RS, Berson SA (1960). Immunoassay of endogenous plasma insulin in man. *J Clin Invest.* **39**: 1157-1175.
2. Kipins DM (1973). Insulin - Physiology. In : Methods in Investigative and Diagnostic Endocrinology. Eds. Berson SA, Yalow RS. American Elsevier Publishing Company Inc. Newyork. Part III Non-Pituitary Hormones. *Peptide Hormones.* **2B** : 827-835.
3. DeFronzo RA, Tobin JD, Andres R (1979). Glucose clamp technique : a method for qualifying insulin secretion and insulin resistance. *Am J Physiol.* **237** : E214-223.
4. Bergman R, Prager R, Volund A, Olefsky J (1987). Equivalence of the insulin sensitivity index in man derived by the minimal model method and the euglycemic clamp. *J. Clin Invest.* **79** : 790-800.
5. Hosker JP, Matthews DR, Rudenski AS, Burnett MA, Darling P, Bown EG, Turner RC (1985). Continuous infusion of glucose with model assessment : measurement of insulin resistance and beta-cell function in man. *Diabetologia.* **28** : 401-411.
6. WHO Expert Committee on Diabetes Mellitus. Second Report. Geneva, World Health Organization, 1980 (WHO Technical Report Series, No. 646).
7. Belfiore F, Iannello S, Volpicelli G (1998). Insulin sensitivity indices calculated from basal and OGTT-induced insulin, glucose, and FFA levels. *Mol Genet Metab.* **63** : 134-141.
8. Raynaud E, Perez-Martin A, Brun JF, Benhadded AA, Mercier J (1999). Revised concept for the estimation of insulin sensitivity from a single sample. *Diabetes Care.* **22** : 1003-1004.
9. Polonsky KS, Sturis J, Byrne M, O'Meara N, Cauter EV (1994). Oscillatory insulin secretion: characterization and alterations in diabetes. Eds. Baba S, Kaneko T. Elsevier Amsterdam. *Diabetes.* 176-183.
10. Bajaj JS, Subba Rao G, Subba Rao J, Khardori R (1987). A mathematical model for insulin kinetics and its application to protein-deficient (Malnutrition-related) diabetes mellitus. *J Theor Biol.* **126** : 491-503.
11. Geevan CP, Subba Rao J, Subba Rao G, Bajaj JS (1990). A Mathematical Model for Insulin Kinetics III : Sensitivity Analysis of the Model. *J Theor Biol.* **147** : 255-263.
12. Rao GS, Bajaj JS, Rao JS (1997). Mathematical modeling of insulin kinetics. *Current Science.* **73**: 957-967.
13. Bajaj M, DeFronzo RA (2003). Metabolic and molecular basis of insulin resistance. *J. Nucl. Cardiol.* **10**: 311-323.
14. Bajaj JS, Bajaj M (2004). Hepatic Fat and Insulin Resistance. Causes and consequences of Non-alcoholic Fatty Liver Disease. In : Non-Alcoholic Fatty Liver Disease. Eds. Sarin SK, Sood OP. *Ranbaxy Science Foundation.* 175-197.
15. Campbell PJ, Mandarino LJ, Gerich JE (1988). Quantification of the relative impairment in actions of insulin on hepatic glucose production and peripheral glucose uptake in non-insulin-dependent diabetes mellitus. *Metabolism.* **37**:15-21.
16. Bonadonna RC, Groop L, Kraemer N, Ferrannini E, Del Prato S, DeFronzo RA (1990);. Obesity and insulin resistance in humans : a dose-response study. *Metabolism.* **39**:452-459.
17. Nurjhan N, Campbell PJ, Kennedy FP, Miles JM, Gerich JE (1986). Insulin dose-

response characteristics for suppression of glycerol release and conversion to glucose in humans. *Diabetes.* **35**:1326-1331.

18. Bruning JC, Michael MD, Winnay JN, Hayashi T, Horsch D, Accili D, Goodyear LJ, Kahn CR (1998). A muscle-specific insulin receptor knockout exhibits features of the metabolic syndrome of NIDDM without altering glucose tolerance. *Mol Cell.* **2**:559-569.
19. Michael MD, Winnay JN, Curtis SE, Kulkarni RN, Postic C, Magnuson MA, Kahn CR (1999). Liver-specific insulin receptor knockout mice are severely insulin resistant. *Diabetes (Suppl 1)* **48**:A10.
20. Wahrenberg H, Lonnqvist F, Arner P (1989). Mechanisms underlying regional difference in lipolysis in human adipse tissue. *J Clin Invest.* **84**:458-467.
21. DeFronzo RA (1997). Pathogenesis of type 2 diabetes mellitus: metabolic and molecular implications for identifying diabetes genes. *Diabetes Rev.* **5**:117-269.
22. Kelley DE, Mandarino LJ (2000). Fuel selection in human skeletal muscle in insulin resistance. A reexamination. *Diabetes.* **49**:677-683.
23. Randle PJ, Garland PB, Hales CN, Newsholme EA (1963). The glucose fatty-acid cycle : its role in insulin sensitivity and the metabolic disturbances of diabetes mellitus. *Lancet.* **1**:1785-1789.
24. Boden G, Jadali F, White J, Liang Y, Mozzoli M, Chen X et al (1991). Effects of fat on insulin-stimulated carbohydrate metabolism in normal men. *J Clin Invest.* **88**:960-966.
25. Carpentier A, Mittelman SD, Bergman RN, Giacca A, Lewis GF (2000). Prolonged elevation of plasma free fatty acids impairs pancreatic beta-cell function in obese nondiabetic humans but not in individuals with type 2 diabetes. *Diabetes.* **49**:399-408.
26. McGarry JD (2002). Banting lecture 2001: Dysregulation of fatty acid metabolism in the etiology of type 2 diabetes. *Diabetes.* **51**:7-18.
27. Roden M, Price TB, Perseghin G, Petersen KF, Rothman DL, Cline GW et al (1996). Mechanism of free fatty acid-induced insulin resistance in humans. *J Clin Invest.* **97**:2859-2865.
28. Cline GW, Petersen KF, Krssak M, Shen J, Hundal RS, Trajanoski Z et al (1999). Impaired glucose transport as a cause of decreased insulin-stimulated muscle glycogen synthesis in type 2 diabetes. *N Engl J Med.* **341**:240-246.
29. Dresner A, Laurent D, Marcucci M, Griffin ME, Dufour S, Cline GW et al (1999). Effects of free fatty acids on glucose transport and IRS-1 associated phosphatidylinositol 3-kinase activity. *J Clin Invest.* **103**:253-259.
30. Griffin ME, Marcucci MJ, Cline GW, Bell K, Barucci N, Lee D et al (1999). Free fatty acid-induced insulin resistance is associated with activation of protein kinase C theta and alterations in the insulin signaling cascade. *Diabetes.* **48**:1270-1274.
31. Shulman GI (2000). Cellular mechanisms of insulin resistance. *J Clin Invest.* **106**:171-176.
32. Kim JK, Fillmore JI, Chen Y, Yu C, Moore IK, Pypaert M et al (2001). Tissue-specific overexpression of lipoprotein lipase causes tissue-specific insulin resistance. *Proc Natl Acad Sci USA.* **98**:7522-7527.
33. Richardson DK, Kashyap S, Mandarino SJ, Bajaj M, Cusi K, Finlayson J, Jenkinson CP, Mandarino LJ (2004). Lipid infusion induces an inflammatory/fibrotic response

and decreases expression of nuclear encoded mitochondrial genes in human skeletal muscle. *Diabetes.* **53** *(Suppl 2)*:A369.

34. Mootha VK, Lindgren CM, Eriksson KF et al (2003). PGC-1alpha-responsive genes involved in oxidative phosphorylation are coordinately down regulated in human diabetes. *Nat Genet.* **34**:267-273.
35. Yang X, Pratley RE, Tokraks S, Bogardus C, Permana PA (2002). Microarry profiling of skeletal muscle tissues from equally obese, non-diabetic insulin-sensitive and insulin-resistant Pima Indians. *Diabetologia.* **45**:1584-1593.
36. Brigstock DR (2003). The CCN family : a new stimulus package. *J. Endocrinol.* **178**:169-175.
37. Candido R, Jandeleit-Dahm KA, Cao Z et al (2002). Prevention of accelerated atherosclerosis by angiotensin – converting enzyme inhibition in diabetic apolipoprotein E-deficient mice. *Circulation.* **106**:246-253.
38. Ruperez M, Ruiz-Ortega M, Esteban V et al (2003). Angiotensin II increases connective tissue growth factor in the kidney. *Amer. J. Path.* **163**:1937-1947.
39. Ohnishi H (1998). Increased expression of connective tissue growth factor in the infarct zone of experimentally induced myocardial infarction in rats. *Jour. Mol. Cell. Cardiol.* **30**:2411-2422.
40. Paradis V, Perlemuter G, Bonvoust F et al (2001). High glucose and hyperinsulinemia stimulate connective tissue growth factor expression : a potential mechanism involved in nonalcoholic steatohepatitis. *Hepatology.* **34**:738-744.
41. Kurikawa N, Suga M, Kuroda S, Yamada K, Ishikawa H (2003). An angiotensin II type 1 receptor antagonist, olmesartan medoxomil, improves experimental liver fibrosis by suppression of proliferation and collagen synthesis in activated hepatic stellate cells. *Brit. Jour. Pharmacol.* **139**:1085-1094.
42. Bajaj M, Suraamornkul S, Pratipanawatr T, Haridies LJ, Pratipanawatr W, Glass L, Cersosimo E, Miyazaki Y, DeFronzo RA (2003). Pioglitazone reduces hepatic fat content and augments splanchnic glucose uptake in patients with type 2 diabetes. *Diabetes.* **52**: 1364-1370.
43. Spiegelman BM (1998). PPAR-γ adipogenic regulator and thiazolidinedione receptor. *Diabetes.* **47**:507-514.
44. Bajaj M, Suraamornkul S, Piper P, Hardies LJ, Glass L, Cersosimo E, Pratipanawatr T, Miyazaki Y, DeFronzo RA (2004). Decreased plasma adiponectin concentrations are closely related to hepatic fat content and hepatic insulin resistance in Pioglitazone-Treated type 2 diabetic patients. *J Clin Endocrinol Metab.* **89**:200-206.
45. Bajaj M, Suraamornkul S, Hardies LJ, Pratipanawatr T, DeFronzo RA (2004). Plasma resistin concentration, hepatic fat content, and hepatic and peripheral insulin resistance in pioglitazone-treated type II diabetic patients. *Int. J. Obes. Relat. Metab. Disord.* **28**:783-789.
46. Ouchi N, Kihara S, Arita Y, Maeda K, Kuriyama H, Okamoto Y, Hotta K, Nishida M, Takahashi M, Nakamura T, Yamashita S, Funahashi T, Matsuzawa Y (1999). Novel modulator for endothelial adhesion molecules : adipocyte-derived plasma protein adiponectin. *Circulation.* **100** : 2473-2476.
47. Ouchi N, Kihara S, Arita Y, Nishida M, Matsuyama A, Okamoto Y (2001).

Adipocyte-derived plasma protein, adiponectin, suppresses lipid accumulation and class A scavenger receptor expression in human monocyte-derived macrophages. *Circulation.* **103** :1057-1063.

48. Shetty GK, Economides PA, Horton ES, Mantzoros CS, Veves A (2004). Circulating Adiponectin and Resistin Levels in Relation to Metabolic Factors, Inflammatory Markers, and Vascular Reactivity in Diabetic Patients and Subjects at Risk for Diabetes. *Diabetes Care.* ; **27**: 2450-2457.
49. Winzer C, Wagner O, Festa A, Schneider B, Roden M, Todesca DB, Pacini G, Funahashi T, Willer AK (2004). Plasma Adiponectin, Insulin Sensitivity, and Subclinical Inflammation in Women With Prior Gestational Diabetes Mellitus. *Diabetes Care.* **27** :1721-1727.
50. Bajaj JS (2006). Metabolic Syndrome : From Inert Facts to Informed Action. *Ann Natl Acad Med Sci (India),* 42 (Suppl.) : 5-10.
51. Zimmet P, Alberti G (2006). Diabetes and the metabolic syndrome. The IDF definition. *Diabetes Voice.* **51** : 11-14.
52. World Health Organisation (1999). Definition, diagnosis and classification of diabetes mellitus and its complications. Report of a WHO consultation. World Health Organisation, Geneva.
53. International Diabetes Federation Epidemiology Task Force Consensus Group (2005). The IDF consensus worldwide definition of the metabolic syndrome. International Diabetes Federation. Brussels.
54. Expert Panel of Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (2001). Executive Summary of the Third Report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III). *JAMA.* **285**: 2486-2497.

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The Metabolic Syndrome and Cardiovascular Risk

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Introduction:

The term "Syndrome X" was coined by Prof. Gerald Reaven at the Banting lecture of the American Diabetic Association in 1988 for a constellation of abnormalities that had insulin resistance as the underlying abnormality. The term metabolic syndrome (MetS) has been in extensive use after WHO formulated diagnostic criteria in 1999 to denote this entity although the expression per se was in use since the late 1970s. The syndrome encompasses a cluster of metabolic risk factors associated with an increased risk for Type 2 Diabetes and cardiovascular disease (CVD). Several expert groups have attempted to produce diagnostic criteria to define metabolic syndrome (see Table 1).

Table 1 Definitions of the metabolic syndrome

ATP III definition (15)

Any three or more of the following criteria:

(1) Waist circumference > 102 cm in men and > 88 cm in women.

(2) Serum triglycerides > 150 mg/dl

(3) Blood pressure > 130/85 mm Hg

(4) HDL cholesterol < 40 mg/dl in men and < 50mg/dl in women

(5) Serum glucose (fasting) > 110 mg/dl

WHO definition (2)

Diabetes, IFG, IGT or insulin resistance (assessed by clamp studies) and at least two of the following criteria:

(1) Waist-to-hip ratio > 0.90 in men or >0.85 in women

(2) Serum triglycerides > 150 mg/dl or HDL cholesterol < 35 mg/dl in men and <40 mg/dl in women

(3) Blood pressure > 140/90 mm Hg

(4) Urinary albumin excretion rate >20ug/min or albumin to creatinine ratio >30 mg/g

International Diabetes Federation (IDF) definition (3)

Central obesity

(Defined as waist circumference =94cm for Europid men and = 80cm for Europid

women, with ethnicity specific values for other groups) plus any two of the following four factors:

(1) Raised TG level: =150 mg/dL (1.7 mmol/L), or specific treatment for this lipid abnormality

(2) Reduced HDL cholesterol: <40 mg/dL (1.03 mmol/L*) in males and <50mg/dL (1.29 mmol/L*) in females, or specific treatment for this lipid abnormality.

(3) Raised blood pressure: systolic BP =130 or diastolic BP =85 mm Hg, or treatment of previously diagnosed hypertension.

(4) Raised fasting plasma glucose (FPG) =100 mg/dL (5.6 mmol/L), or previously diagnosed type 2 diabetes (If above 5.6 mmol/L or 100 mg/dL, OGTT is strongly recommended but is not necessary to define presence of the syndrome.)

* These values have been updated from those originally presented to ensure consistency with ATP III cut points.

The most widely accepted of the definitions for MetS have been produced by the World Health Organization (WHO) and the National Cholesterol Education Program —Third Adult Treatment Panel (NCEP ATP III) (1, 2). Both groups agree on the core components: obesity, insulin resistance, dyslipidaemia and hypertension. Unlike the WHO definition, the ATP III definition does not obligatorily require impaired glucose regulation or insulin resistance as an essential component. In addition, the levels set for each component and the combination of components required to diagnose the MetS are slightly different in the two recommendations.

Pathophysiology of cardiovascular disease in the metabolic syndrome

Abnormal fat distribution

Population studies have shown an increase in the risk of chronic non-communicable diseases associated with a progressive increase in total adiposity as assessed by the body mass index (BMI). Among equally obese individuals, those with an increase in abdominal fat (assessed by waist circumference) are at increased risk of Type 2 diabetes and CVD. This is independent of the risk predicted by increased BMI. For instance, the INTERHEART study (4) studied patients with initial myocardial infarction worldwide in over 55 countries and identified nine important risk factors for myocardial infarction which accounted for over 9/10th of the worldwide myocardial infarction risk. One of these risk factors was abdominal obesity. Yusuf et al. (5) subsequently explored the associations between different measures of obesity and risk of myocardial infarction in this study. Four different measurements of obesity were examined: BMI, waist-to-hip ratio, waist circumference and hip circumference. The waist-to-hip ratio was the obesity

measurement most strongly associated with myocardial infarction. The cut-off value for waist circumference is likely to be population specific as there are clear differences across various ethnic populations in the relationship between overall adiposity, abdominal obesity and visceral fat accumulation. This has been noted by several studies in India (6). In this regard waist circumference criteria for South Asians suggest a cut-off of 94 cm for males and 80cm for females (7).

Thus, although Reaven's original description did not include it, abdominal obesity as assessed by waist circumference is an important cardiovascular risk marker and the primary target for the treatment of metabolic syndrome. The International Diabetes Federation (IDF) (3) places even more emphasis on central obesity. This new emphasis on central obesity as opposed to BMI is based on growing evidence that waist circumference has a closer association with cardiovascular disease and mortality.

Insulin resistance

Insulin resistance is central to the pathophysiology of the metabolic syndrome and has been postulated as its underlying cause. It correlates univariately with the risk of Type 2 diabetes and CVD; however the association with hypertension is weak. The mechanisms underlying the link between insulin resistance and CVD still need further investigation but there is evidence that excess free fatty acids are involved. A recent meta-analysis (8) examined the relationship between plasma insulin and cardiovascular mortality in nondiabetic adults based on data from 11 prospective studies. After adjusting for various risk factors including age and smoking, the hazard ratio of cardiovascular mortality was highest in the subgroup with the highest fasting insulin. Although there is overwhelming evidence suggesting that the metabolic syndrome is significantly associated with insulin resistance, the ATP III definition of the metabolic syndrome does not include a measurement of insulin resistance.

Obesity and inflammation

Inflammation clearly plays a major role in atherogenesis (9). Several inflammatory markers: Tumour necrosis factor (TNF) α, Interleukin-6 (IL-6), fibrinogen and C-reactive protein (CRP) have often been found to be elevated in studies of patients with MetS. The most extensively studied inflammatory marker is high sensitivity C-reactive protein (CRP). It has been shown to be a predictor of both diabetes and cardiovascular disease risk. A proinflammatory state recognized by elevated C-reactive protein (CRP) levels is commonly present in people with the MetS . There is a significant relationship between plasma CRP levels and measures of adiposity and of insulin resistance. One contributory mechanism to this association is obesity, as adipocytes and macrophages release inflammatory cytokines which promote an inflammatory state.

CRP has also been shown to be an independent predictor of cardiovascular

events and its predictive value has been said to equal that of metabolic syndrome (10). The predictive strength of CRP levels on cardiovascular disease has, however, been questioned by Danesh et al. (11), who reported CRP levels in 2459 patients with major adverse cardiovascular events in comparison with a matched cohort of 3969 individuals. After adjusting for many known risk factors, CRP showed an odds ratio of only 1.45 for coronary artery disease. At present the American Heart Association (AHA) does not recommend using CRP for population screening or to monitor treatment.

Obesity and atherogenesis: multiple signals

Adipose tissue is not an inert organ but an important player in the integration of endocrine, metabolic, and inflammatory signals for the control of energy homeostasis. Several substances are secreted and several receptors have been identified on adipocytes. One important adipocytokine identified is adiponectin. Levels of adiponectin have been shown to have a strong and consistent inverse association with insulin resistance and inflammation (12,13). Adiponectin levels possibly, may be a marker for atherosclerosis and coronary heart disease. In a large case-control study that examined adiponectin levels it was found that men with the lowest quintile of adiponectin levels had a significantly decreased risk of myocardial infarction (relative risk 0.39) (14). Adiponectin may have anti-inflammatory effects that provide protection against atherosclerosis development, particularly in those clinical situations associated with low plasma concentrations of adiponectin. The future may possibly see recombinant adiponectin used in the prevention or treatment of cardio-vascular disease in selected patients.

Leptin and resistin are other markers associated with obesity. Obese individuals have been demonstrated to have elevated leptin concentrations (15). The West of Scotland Coronary Prevention Study (WOSCOPS) (16) showed that leptin independently increased the relative risk of coronary artery disease. The role of resistin in obesity and insulin resistance in humans is debated. Obese individuals have higher serum levels of resistin than lean individuals (17) but serum resistin has not been shown to be a significant predictor of insulin resistance (18,19).

Atherogenic dyslipidaemia

The dyslipidaemia associated with the MetS includes raised TGs and low concentrations of HDL-cholesterol. A more detailed analysis usually reveals other lipoprotein abnormalities, including elevated apolipoprotein B (Apo B), increased number of small dense low-density lipoprotein (LDL) particles and small HDL particles. All of these abnormalities are independently atherogenic.

Elevated blood pressure

MetS is usually associated with raised blood pressure. However, the strength of

association varies considerably from one population to another.

Prothrombotic state

Components of the MetS are associated with both coagulation and fibrinolytic proteins, with a link to an elevated plasminogen activator inhibitor-1 (PAI-1) and fibrinogen being the most consistent findings.

Endothelial dysfunction

There is evidence that patients with MetS have elevated mononuclear cell adhesion, and diminished endothelial-dependent vasodilata-tion that are markers of endothelial dysfunction and associated with cardiovascular adverse events.

Relationship to predictability of cardiovascular disease and diabetes mellitus

Cardiovascular disease

Since the metabolic syndrome comprises accepted CVD risk factors, it would be expected that the syndrome is a strong predictor of CVD. A substudy of the Botnia study, which involved over 4000 Finnish and Swedish adults, demonstrated that those with the MetS, as defined by the 1999 WHO criteria, were thrice as likely to have a history of CHD compared with those without the syndrome. Moreover, the presence of the syndrome was associated with a significantly increased cardiovascular mortality (12% vs. 2%) (20). Observational studies that have confirmed that the risks of developing CVD, cardiovascular mortality and all-cause mortality, are increased by the presence of the MetS, include the European DECODE study (8), the Finnish Kuopio study (21) and the San Antonio Heart Study (22). Similar findings are also reported from clinical trials, including the WOSCOPS trial, and, at least for insulin resistance, the VA-HIT study. Nonetheless, other studies have disputed whether the MetS gives any additional information over and above the individual well-known CVD risk factors (23). This may relate to an inadequate definition of the MetS; (clear measurements of inflammation are not included) and the cut-points used, rather than a problem with the overall concept. Indian studies have documented the prevalence of abnormal lipid distribution and waist-hip ratio (6); however, the predictive value of MetS for cardiovascular events has not yet been validated.

Diabetes

Non-diabetic people with the MetS are at a very high risk for the development of Type 2 diabetes. In one study, the risk for diabetes was five fold higher in patients with the syndrome (22). However, glucose dysregulation (IFG or IGT) is often already present in patients with MetS. Importantly, the greatest impact of diabetes is the two to four times greater risk of CHD and stroke.

Management

Lifestyle modification

The management of metabolic syndrome lies in the early detection and

treatment of the underlying risk factors which make up the syndrome, mainly abnormal blood pressure and glucose levels, dyslipidemia, being overweight and abdominal obesity. In individuals with a normal fasting glucose, the presence of IGT is a good surrogate marker for insulin resistance, and is easily measured using the oral glucose tolerance test. Once a clinical diagnosis is made, decreasing energy intake and increasing physical activity remains the cornerstone of therapy. Decreasing dietary glucose by reducing the intake of high-glycemic beverages and replacing refined grain products and potatoes with minimally processed plant based foods such as whole grains, fruits and vegetables may reduce coronary heart disease incidence among people with metabolic syndrome.

Mega doses of dietary antioxidants have not demonstrated protection against CV disease or diabetes. Smoking increases the risk of adverse CVD events and should be strongly discouraged.

Exercise has the potential to bring about weight loss, increase insulin sensitivity, alter the plasma lipoprotein profile and improve fibrinolytic activity. These favourable effects likely result from changes in the activity of insulin sensitive glucose transporters and of skeletal muscle lipoprotein lipase. The health benefits of regular physical activity as a primary preventive measure is beyond doubt. Accumulating at least 30 min of daily physical activity of moderate intensity is probably enough to maintain body weight.

Drug therapy

Appropriate drug therapy should be initiated to manage elevations in blood pressure and dyslipidemia among patients with the metabolic syndrome. The choice of medications prescribed should be based on the established guidelines for the management of these conditions. ACE inhibitors are a reasonable choice for the management of hypertension given its renal protection for patients with diabetes, and the possibility that it may prevent diabetes among patients with IGT. Statins or fibrate therapy should be initiated depending on the type of dyslipidemia.

Modulation of insulin resistance is possible by thiazolidinediones and metformin. Apart from their glucose lowering effects, the thiazolidinediones have multiple nonglucose metabolic effects. They also reduce insulin resistance through binding to and activation of the nuclear receptor, peroxisome proliferators-activated receptor - gamma (PPAR gamma), with subsequent effects on glucose and lipid homeostasis. Thus, diabetic patients must be treated with insulin sensitizers whenever possible. ACE-inhibitors should be the first-choice antihypertensives in patients with hypertension associated with the metabolic syndrome.

Other drugs that have the potential to control glucose abnormalities associated with the metabolic syndrome include the prandial oral antidiabetic agents such as alpha glucosidase inhibitors (Acarbose, voglibose, miglitol) and the rapidly acting

insulin secretagogues (Nateglinide, Repaglinide). These drugs improve the control of postprandial hyperglycemia.

The approach of treating patients with IGT with medications to prevent or delay diabetes is rapidly evolving and their benefits need to be confirmed in large prospective clinical trials.

Patients with diabetes derive substantial benefit from aspirin. Apart from its use in diabetics the role of aspirin in nondiabetics with metabolic syndrome is uncertain.

Antiobesity drugs

When attempts at lifestyle modification are inadequate, the use of anti-obesity drugs is warranted. Drug treatment is often indicated but is somewhat limited by the small number of safe and well tolerated drugs that are proven to have long-term efficacy in maintaining body weight loss. The currently available drugs, sibutramine and orlistat, appear modestly effective in promoting weight loss. Blockade of the endocannabinoid system with rimonabant appears to be a promising new strategy.

References :

1. Executive Summary of the Third Report of The National Education Program (NCEP) Expert Panel on Detection, and Treatment of High Blood Cholesterol In Adults (Adult Treatment Panel III). *JAMA* 2001; **285** : 2486 -2497.
2. World Health Organization. Definition, Diagnosis and Prevention of Diabetes Mellitus and its Complications. Report consultation. Geneva: World Health Organization 1999.
3. Ford ES (2005). Prevalence of the metabolic syndrome defined by the International Diabetes Federation among adults in the US. *Diabetes Care* **28:** 2745-2749.
4. Yusuf S, Hawken S, Ounpuu S, et al (2004). Effect of potentially modifiable risk factors associated with myocardial infarction in 52 countries (the INTER-HEART study): case-control study. *Lancet* **364:** 937-952.
5. Yusuf S, Hawken S, Ounpuu S, et al (2005). INTERHEART Study Investigators. Obesity and the risk of myocardial infarction in 27,000 participants from 52 countries: a case-control study. *Lancet* **366:** 1640-1649.
6. Misra A, Wasir JS, Vikram NK (2005). Waist circumference criteria for the diagnosis of abdominal obesity are not applicable uniformly to all populations and ethnic groups. *Nutrition* **21**:969-76).
7. (International Diabetes Federation (2005). New IDF worldwide definition of the metabolic syndrome. Press conference, 1st International congress on "Pre-Diabetes"and the Metabolic syndrome. Berlin, Germany. April 14, 2005 (www.idf.org).
8. Hu G, Qiao Q, Tuomilehto J, et al (2004). DECODE Insulin Study Group. Plasma insulin and cardiovascular mortality in non-diabetic European men and women: a meta-analysis of data from eleven prospective studies. *Diabetologia* **47:** 1245-1256.

9. Ross R (1999). Atherosclerosis: an inflammatory disease. *N Engl J Med* **340:** 115–126.

10. Pearson TA, Bazzarre TL, Daniels SR, et al (2003). American Heart Association guide for improving cardiovascular health at the community level: a statement for public health practitioners, healthcare providers, and health policy makers from the American Heart Association Expert Panel on Population and Prevention Science. *Circulation* **107:** 645–651.

11. Danesh J, Wheeler JG, Hirschfield GM, et al (2004). C-reactive protein and other circulating markers of inflammation in the prediction of coronary heart disease. *N Engl J Med* **350:** 1387–1397.

12. Chandran M, Phillips SA, Ciaraldi T (2003). Adiponectin: more than just another fat cell hormone? *Diabetes Care* **26:**2442–2450.

13. Weyer C, Funahashi T, Tanaka S, et al (2001). Hypoadiponectinemia in obesity and type 2 diabetes: close association with insulin resistance and hyperinsulinemia. *J Clin Endocrinol Metab* **86:** 1930–1935.

14. Pischon T, Girman CJ, Hotamisligil GS, et al (2004). Plasma adiponectin levels and risk of myocardial infarction in men. *JAMA* **291:** 1730–1737.

15. Rosicka M, Krsek M, Matoulek M, et al (2003). Serum ghrelin levels in obese patients: the relationship to serum leptin levels and soluble leptin receptor levels. *Physiol Res* **52:** 61–66.

16. Wallace AM, McMahon AD, Packard CJ, et al (2001). Plasma leptin and the risk of cardiovascular disease in the West of Scotland Coronary Prevention Study (WOSCOPS). *Circulation* **104:** 3052–3056.

17. Steppan CM, Bailey ST, Bhat S, et al (2001). The hormone resistin links obesity to diabetes. *Nature* **409:** 307–312.

18. Youn BS, Yu KY, Park HU, et al (200). Plasma resistin concentrations measured by enzyme-linked immunosorbent assay using a newly developed monoclonal antibody are elevated in individuals with type 2 diabetes mellitus. *J Clin Endocrinol Metab* **89:** 150–156.

19. Rea R, Donnelly R (2004). Resistin: an adipocyte-derived hormone. Has it a role in diabetes and obesity? *Diabetes Obes Metab* **6:** 163–170.

20. Isomaa B, Almgren P, Tuomi T etal (2001). Cardiovascular morbidity and mortality associated with the metabolic syndrome. *Diabetes Care* **24(4):** 683-9.

21. Lakka HM, Laaksonen DE, Lakka TA etal (2002). The metabolic syndrome and total cardiovascular disease mortality in middle-aged men. *JAMA* **288(21):** 2709-16

22. Hunt KJ, Resendez RG, Williams K etal (2004). National Cholesterol Education Program versus World Health Organization metabolic syndrome in relation to all-cause and cardiovascular mortality in the San Antonio Heart Study. *Circulation* Sep 7; **110(10):** 1251-7.

23. Stern MP, Williams K, Gonzalez-Villalpando C, etal (2004). Does the metabolic syndrome improve identification of individuals at risk of type 2 diabetes and/or cardiovascular disease? *Diabetes Care* **27(11):** 2676-81.

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Metabolic syndrome, microalbuminuria and chronic kidney disease

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The metabolic syndrome is defined by a constellation of risk factors, including abdominal obesity, impaired glucose tolerance in association with hyperinsulinaemia, insulin resistance, dyslipidaemia characterized by low high-density lipoprotein (HDL)-cholesterol and high triglyceride levels, and hypertension. The World Health Organization (WHO) and the Adult Treatment Panel (ATP) III of the National Cholesterol Education Program have developed clinical criteria for metabolic syndrome (1). Metabolic syndrome has gained a great deal of attention because it is considered a precursor to type 2 diabetes mellitus (DM) and also increases cardiovascular disease (CVD) risk, even with levels of glycaemia below that used to define diabetes (1). The connections between chronic kidney disease (CKD) and CVD are increasingly evident. Indicators of CKD, albuminuria (micro- or macro-) and loss of glomerular filtration rate (GFR) are independently associated with increased CVD risk in the general population, as well as high-risk subgroups (2,3). These connections are exceedingly complex and include a number of shared traditional risk factors (notably diabetes and hypertension), development of non-traditional risk factors (anaemia, hyperparathyroidism with disordered mineral metabolism, high levels of homocysteine and others) and more severe atherosclerosis.

Microalbuminuria It is defined as urinary albumin excretion of 30 - 300 mg/day (20 - 200 ug/mt.) or albumin creatinine ratio in untimed urine specimen of 30 mg/g. Microalbuminuria in type I DM is the earliest sign of overt nephropathy of these who have microalbuminuria within 10 years of the onset of DM, most will progress to overt proteinuria while in type-II DM progression of microalbuminuria to overt nephropathy occurs in 20-40% of patients. Microalbuminuria is a clinical criterion for metabolic syndrome by the WHO classification (1). The frequency of microalbuminuria increases across the spectrum from those with normal glucose tolerance (5-10%), to metabolic syndrome (12-20%), to type 2 diabetes (25-40%) (4). Enough evidence has been garnered confirming the association between microalbuminuria and each of hypertension (5) and central obesity (6). Also, studies confirm the association of microalbuminuria with salt sensitivity, the

absence of nocturnal drops in both systolic and diastolic blood pressure, dyslipidemia, and left ventricular hypertrophy (7).

More importantly, microalbuminuria is now established as a modifiable predictor of CVD and CVD mortality (5). Evidence has been garnered that microalbuminuria is a marker of generalized endothelial dysfunction and consequently a risk factor for CVD (8). In recent studies, this endothelial dysfunction has been characterized by the presence of transmembrane leakiness (9). It is presently unclear whether transmembrane leakiness should be viewed as the culminating event of different atherogenic factors acting in concert to promote endothelial dysfunction or whether it should be considered as the underlying substrate that enhances the atherogenicity. For one, the increase in vascular permeability can promote the penetration of atherogenic lipoprotein particles in the arterial wall. One possible explanation is that endothelial dysfunction might promote increased penetration of atherogenic lipoprotein particles in the arterial wall, but glucose control, insulin resistance, procoagulant stage, and adhesion molecules have all been implicated in the pathogenesis. In addition, microalbuminuria has also been associated with alterations in hemodynamic and vascular responses. This is exemplified by studies that have demonstrated that the compensatory vasodilation seen after relief from prolonged ischemia or infusion of vasodilators such as nitroglycerin is blunted in people with microalbuminuria (9).

In summary, microalbuminuria is a signal from the kidneys conveying the abnormalities in endothelial function and vascular response. It can be seen as a early marker of generalized endothelial dysfunction, althrosclerosis increased cardiovascular disease risk and progressive renal failure. More importantly microalbumniuria represents the stage of nephropathy at which treatment is often successful in preventing progressive renal disease. In this light, the reduction of microalbuminuria should be implemented as a therapeutic goal to reduce overall CVD and CKD risk (10).

Chronic kidney disease (CKD) as per position statement from Kidney Disease: Improving Global Outcomes (KDIGO) 2005 has been defined as GFR < 60 ml.mt. (11). There has been on alarming growth of the prevalence of CKD and end stage renal disease (ESRD) over the last decade, in concert with a striking increase in the burden of diabetes, the leading cause of ESRD. Furthermore US Renal Data Support System (USRDS) 15th annual report projects that by the year 2030, ESRD population will increase by 460,000 new cases annually and the prevalent population will reach 2.24 million, with two thirds of these numbers having diabetes as the primary cause of renal disease (12,13).

In parallel with the growth in kidney disease, the prevalence of obesity/insulin resistance and impaired glucose

metabolism has been rising rapidly, currently meeting the epidemic proportions (14). The prevalence of diagnosed and undiagnosed diabetes constitute a major portion of the insulin resistant population is estimated at 8% of adult population (15). An even greater number of patients have cardiometabolic syndrome (24%) (12).

In India, conservative estimates put the annual incidence of ESRD in India around 100 per million of the population this would mean approximately 100,000 new patients every year for a population of 1 billion. Diabetic nephropathy is the commonest cause of ESRD in persons over 40 years of age accounting for 34% of ESRD cases. The fate of ESRD patients in this country is quite dismal, with a majority (65.7%) not receiving any form of RRT or stopping the treatment due to lack of resources. Only 12.8% undergo renal transplantation, 16.3% receive maintenance hemodialysis for varying periods of time and 5% continuous ambulatory peritoneal dialysis (16).

The prevalence of metabolic syndrome in Indian population has been reported to be 11-40 % (17). If metabolic syndrome per se is the cause of CKD as has been shown in recent studies then a substantial population is going to have ESRD in near future. This burden of ESRD can neither be borne by individuals nor the state. Hence it is important to address the issue of CKD in metabolic syndrome so that preventive steps can be taken.

The two most important causes of ESRD viz. diabetes and hypertension are closely associated with excess body weight, but obesity is still not listed as a cause of ESRD in USRDS report.

Obesity and Kidneys

Obesity is the phenotypic hallmark of metabolic syndrome. In addition to adverse consequences to the health of obese individuals obesity during pregnancy has been linked to future risk for the development of type II Diabetes and hypertension in the offspring when they reach adulthood, thus risking the kidneys of next generation.

Obesity related glomerulopathy

Nephrotic syndrome associated with obesity was reported 3 decades ago. The prevalence of obesity related glomerulopathy which causes CKD has increased 10 fold over last 15 years as a consequence of the spread of obesity epidemic (18). Increased incidence of higher body mass index (BMI) and ESRD in Japanese men has been shown after adjusting for comorbid conditions (19).

In experimental animals a number of structural changes viz expansion of bowman's space, increased bowman's capsule surface area and glomerular tuft area, endothelial and mesangial cell proliferation and basement membrane thickening have been shown in early obesity (20). In human beings kidney biopsy from obese individuals showed glomerulomegaly in all, focal segmental glomerulosclerosis in 80% and increased mesangial matrix and cellularity in 45% of

individuals (21). Obesity has also been shown to accelerate the course of idiopathic glomerular disease, such as IgA nephropathy (18). The clinical course of obesity related glomerulopathy appears to be progressive. After a mean follow up of 27 months 14% of patients reached renal end point i.e. doubling of serum creatinine (21).

Renal hemodynamic changes in obesity

The elegant renal physiological studies of Chagnac et al (22) in obese patients have demonstrated that values of GFR and renal plasma flow (RPF) exceeded those of lean controls by 50% and 30% respectively. This results in increased filtration fraction, an indirect indicator of glomerular hypertension. Glomerular hyperperfusion, hyperfiltration and hypertension lead to stretching of the glomerular capillary wall, injury to endothelial and epithelial cell and transudation of macromolecular in the mesangium. This leads to mesangial expansion and sclerosis. This supported with experimental data has been proposed to contribute to progression of nephropathy.

Effect of weight loss on renal function

Obesity related glomerular hyperfiltration ameliorates after weight loss. In a study of 17 morbid obese individuals (BMI-48 kg/m^2) who lost 48 kg and 1 year with BMI decreasing to 32 kg/m^2 after gastroplasty, augmented GFR, RPF and albuminuria improved substantially after weight loss (23). The decreased albuminuria was not solely due to decrease in GFR as evidenced by decrease in the fractional albumin clearance. The decrease in fractional albumin excretion after weight loss could be due to improvement in glomerular hemodynamics or it could also be the result of improvement of generalised endothelial cell dysfunction and thus a better integrity of the systemic capillary network. Whether these improvements in general hemodynamics and endothelial dysfunction translates in prevention of nephropathy is yet to be established.

Pathophysiology of kidney involvement in metabolic syndrome

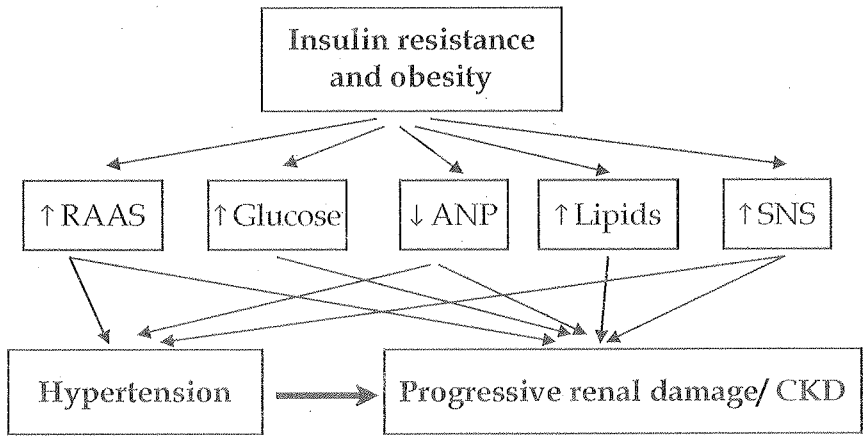
Kidney involvement in metabolic syndrome per se occurs due to interplay between number of mediators as shown in Fig.1.

Renin angiotensin aldosterone system (RAAS): Adipose tissue, especially visceral type, possesses a local RAAS, which has more significant local paracrine as well as systemic effects than the subcutaneous fat In insulin resistance/hyperinsulinemia, which is frequently associated with visceral obesity, RAAS activity is increased. In addition to the increased adipose tissue RAAS activity, systemic RAAS effects are also enhanced in the insulin-resistant/ hyperinsulinemic state, despite a state of sodium retention and volume expansion.

Sympathetic nervous system (SNS): Both animal and human studies suggest that increased SNS activity, where vascular

Figure 1

Pathophysiology of Chronic Kidney Disease



RAAS : renin angiotensin aldosterone system SNS : sympathetic nervous system
ANP : natriuretic peptide

and renal INS actions are selectively preserved, may be another mediator of hypertension in insulin resistance/ hyperinsulinemia state via stimulating renal sodium reabsorption with subsequent volume expansion and increasing cardiac output (9). In the Normative Aging Study, SNS activity was elevated with hyperinsulinemia and correlated with BMI (24).

Natriuretic peptide system: The natriuretic peptide system consists of the atrial natriuretic peptide(ANP), the brain natriuretic peptide (BNP), and the C-type natriuretic peptide (CNP), each encoded by a separate gene. They are synthesized predominantly in the heart, brain, and kidneys and work via specific receptors, namely NPr-A, NPr-B, and NPr-C.(3) The natriuretic peptides have a protective role on the development of hypertension due to their natriuretic and vasodilator effects as well as due to their inhibitory effect on the SNS and the RAAS. In obesity/ hyperinsulinemia, over expression of NPr-C receptor and lower levels and function of ANP with a possible role for a promoter variant at position -55 in the NPr-C gene, have been reported, contributing to increased sodium retention (7).

Dyslipidemia: It enhances renal dysfunction through filtered lipoproteins, damaging glomerular and tubular cells, in addition to enhancing endothelial dysfunction and atherosclerosis and participating in the deleterious renal functional and structural changes, eventually leading to nephron damage.

Dysglycemia: It is not only involved in the aforementioned renal changes, but it

also exerts a direct toxic effect on nephrons through glycosylation of glomerular proteins

Functional and Structural Changes, Compensatory Responses, and Late Nephronal Damage: Insulin resistance/ hyperinsulinemia state is accompanied by increased EC proliferation and intrarenal lipid and hyaluronate deposition in the matrix and inner medulla . These depositions increase intra-renal pressure and volume in the tightly encapsulated kidney, leading to parenchymal prolapse and urine outflow obstruction, which result in slow tubular flow and subsequently increased sodium reabsorption, especially in the loop of Henle . This leads to inappropriately small natriuretic response to saline load at mean and glomerular pressure, often referred to as "impaired pressure natriuresis" (6,7).

These functional and structural changes in the kidney provoke compensatory lowered renal vascular resistance, elevated kidney plasma flow, glomerular hyperfiltration, and stimulation of RAAS, despite volume expansion. Neurohumoral factors like Ang II, sympathetic system, and cytokines are synergistically involved in these compensatory mechanisms. For instance, AngII, in addition to its systemic effects on BP, directly contributes to increased glomerular capillary pressure through vasoconstriction of the efferent arterioles and upregulation of renal injury response. These alterations with the hypertension associated with the insulin-resistant state help overcome the increased tubular reabsorption and maintain sodium balance. The persistence of these compensatory responses, increasing glomerular wall stress, in the presence of hypertension, dyslipidemia and dysglycemia, will precipitate gradual nephron loss, glomerulosclerosis and eventually ESRD. This glomerulosclerosis in the hyperinsulinemic/insulin-resistant kidney is peculiar and characterized by lower rate of nephrotic syndrome, fewer lesions of segmental sclerosis and a greater glomerular size compared with the idiopathic variety (25).

Clinical evidence of CKD in metabolic syndrome

Theoretically as discussed above metabolic syndrome per se can and should lead to CKD. But the evidence was not so convincing because of the effect of overlapping variables which constitute this syndrome. Epidemiological studies have linked the metabolic syndrome with an increased incidence of microalbuminuria, few studies have evaluated its relationship with CKD (26,27).

In National Health and Nutrition Examination survey (NHANES III) prevalence of metabolic syndrome was 24.7%. There was a graded increase in the prevalence of CKD with more number of risk factor 0.9% with 1 risk factor to 9.2% with 5 factors. The prevalence of microalbuminuria also increased for 4.9% with 1 risk factor to 20.1% with 5 risk

factors. The multivariate-adjusted odds ratio of CKD and microalbuminuria in participants with metabolic syndrome 2.60 and 1.89 respectively (27). The cross sectional study however does not prove whether the syndrome is a cause or a consequence of reduction of kidney function and also whether the association is independent of future development of diabetes and hypertension.

The Atherosclerosis Risk in Communities Study (ARIC), a large prospective, community-based longitudinal study has demonstrated that the metabolic syndrome, absent diabetes, is associated with an increased risk for incident CKD, defined as progression to estimated GFR<60ml/min per 1.73 m^2 over a 9-yr period. The risk was independent of potential confounding factors such as age, gender, race, education, BMI, alcohol and tobacco use, coronary heart disease, and physical activity. There were graded relations among the number of clinical traits of the metabolic syndrome, HOMA-insulin resistance, and fasting insulin levels and the risk for CKD, suggesting a pathophysiologic basis for these findings. Moreover, the increased risk for CKD was evident even after adjusting for hypertension (a potential cause and consequence of kidney disease) and incident diabetes, another known mediator of CKD. The association between metabolic syndrome and CKD in nondiabetic individuals remained robust even after accounting for the subsequent development of diabetes and hypertensionIn sum, these findings suggest that the metabolic syndrome directly contributes to the development of CKD (28).

Future directions In view of the emergence of evidence linking CKD with metabolic syndrome further work is required to address the issue whether weight reduction, exercise and other measures to improve insulin sensitivity as well as interventions that target biochemical components of metabolic syndrome result in the reduction of the risk of CKD. This would help in preventing a large number of individuals from progressing to ESRD.

References

1. Grundy SM, Brewer HB, Cleeman JI, Smith SC, Lenfant C (2004) for the conference participants. Definition of metabolic syndrome : report of the National Heart, Lung, and Blood Institute/American Heart Association Conference on scientific issues related to definition. *Circulation* **109**: 433-438.
2. Go AS, Chertow GM, Fan D, McCulloch CE, Hsu C (2004). Chronic kidney Disease and the risks of death, cardiovascular events, and hospitalization. *N Engl J Med* **351**: 1296-1305.
3. Hillege HL, Fidler V, Diercks GF et al (2002). Prevention of Renal and Vascular End Stage Disease (PREVEND) Study Group. Urinary albumin Excretion predicts

cardiovascular and noncardiovascular mortality in General population. *Circulation* **106:** 1777-1782

4. Tuttle KR (2005). Renal manifestations of the metabolic syndrome. *Neph Dial Transplant* **20:** 861-64.
5. Palaniappan L, Carnethon M, Fortmann SP (2003). Association between microalbuminuria and the metabolic syndrome : NHANES III. *Am J Hypertens* **16:** 952-958.
6. Mykkanen L, Zaccaro DJ, Wagenknecht LE, Robbins DC, Gabriel M, Haffner SM (1998). Mijcroalbuminuria is associated with insulin resistance in nondiabetic subjects : The insulin resistance atherosclerosis study. *Diabetes* **47:** 793-800.
7. El-Atat FA, Stas SN, Mcfarlane SI, Sowers JR (2004). The relationship between hyperinsulinemia, hypertension and progressive renal disease. *JASN* **15:** 2816-2827.
8. Yudkin JS (1996). Hyperinsulinemia, insulin resistance, microalbuminuria and the risk of coronary heart disease. *An Med* **28:** 433-438.
9. Castro JP, EI-Atat FA, McFarlane SI, Aneja A, Sowers JR (2003). Cardiometabolic syndrome: pathophysiology and treatment. *Curr Hypertens Rep* **5:** 393-402.
10. Mangrum A, Bakris GL (1997). Predictors of renal and cardiovascular mortality in patients with non-insulin-dependent diabetes : a brief overview of microalbuminuria and insulin resistance. *J Diabetes Complications* **11:** 352-357.
11. Levey AS,Eckardt K, Tsukamoto Y, Levine A et al (2005). Definition and classification of chronic kidney disease: A position statement from Kidney Disease: Improving Global Outcomes (KDIGO). *Kidney International* 67; 2089.
12. Ford ES, Giles WH, Dietz WH (2002). Prevalence of the Metabolic Syndrome among US adults : Findings from the Third National Health and Nutrition Examination Survey. *JAMA* **287:** 356-359.
13. U.S. Renal Data System. USRDS 2003 15th Annual Data Report : Atlas of end-stage renal disease in the United States. Bethesda (MD) : National Institute of Health, National Institute of Diabetes and Digestive and Kidney Diseases 2003.
14. Sowers JR (2003). Recommendations for special populations : Diabetes mellitus and the metabolic syndrome. *Am J Hypertens* **16:** 41S-45S.
15. Harris MI, Flegal KM, Cowie CC, Eberhardt MS, Goldstein DE, Little RR, Wiedmeyer HM, Byrd-Holt DD (1998). Prevalence of diabetes, impaired fasting glucose, and impaired glucose tolerance in U.S. adults. The Third National Health and Nutrition Examination Survey, 1988-1994. *Diabetes Care* **21:** 518-524, 10.
16. Sakhuja V, Sud K (2003). End stage renal disease in India and Pakistan: Burden of disease and management issues. *Kidney Int* 63 (supp.83); s115-s118.
17. Cameron JS, Shaw JE, Zimmet PZ (2004). The metabolic syndrome : prevalence in worldwide populations : *Endocrine Metabolic clinics of N America* **33:** 351-375.
18. Bonnet E, Deprele C, Sassolas A, Moulin P, Alamartine E, Berthezene F, Bertoux F (2001) Excessive body weight as a new independent risk factor for clinical and pathological progression in primary IgA nephritis. *Am J Kidney Dis* **37:** 720-727.
19. Iseki K, Ikemiya, Kinjo K, Inoue T, Iseki C, Takishita (2004). Body mass index and risk

of development of end stage renal disease in a screened cohort. *Kidney Int.* **65**: 1870-1876.

20. Henegar JR, Bigler SA, Henegar LK, Tyagi SC, Hall JE (2001). Functional and structural changes in the kidney in the early stages of obesity. *JASN* **12**: 1211-1217.

21. Kambham N, Markowitz S, Valeri AM, Lin J, D'Agati VD (2001). Obesity-related glomerulonephropathy : an emerging epidemic. *Kidney Int* **59**: 1498-1509.

22. Chagnac A, Weinstein T,Korzets A, Ramadan E, Hirsch J., Gafter U (2000). Glomerular hemodynamics in severe obesity. *Am J Physiol Renal Physiol* **278**: F817-F822.

23. Chagnac A, Weinstein T, Herman M, Hirsh J, Gafter U, Yaacov O (2004). The effects of weight loss on renal function in patients with severe Obesity. *J Am Soc Nephrol* **14**: 1480-1486.

24. Word KD, Sparrow D, Landsberg L, Young JB, Weiss ST (1993). The influence of obesity, insulin and sympathetic nervous system activity on blood pressure. *Clin Res* **4**: 168A-168A.

25. Bagby SP (2004). Obesity initiated metabolic syndrome and the kidney : a recipe for chronic kidney disease? *JASN* **15**: 2775-2791.

26. Fujikawa R, Okubo M, Egusa G, Kohno N (2001). Insulin resistance precedes the appearance of albuminuria in non-diabetic subjects : 6 years follow up study. *Diabetes Res Clin Pract* **53**: 99-106.

27. Chen J, Muntner P, Hamm LL, Jones DW, Batuman V, Fonseca V, Whelton PK, He J (2004). The metabolic syndrome and chronic kidney disease in U.S.adults. *Ann Intern Med* **140**: 167-174.

28. Kurella M, Lo J.C. and Chertow GM (2005). Metabolic Syndrome and the risk for chronic kidney disease among nondiabetic adults. *J Am Soc Nephrol* **16**: 2134-2140.

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Medical management of Obesity-Current status

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Pharmacotherapy can be used in conjunction with diet and physical activity to achieve and maintain a realistic degree of weight loss. Pharmacotherapy should be considered for patients with BMI greater than 27 kg/m^2 and associated obesity related complications (i.e. hypertension, CHD, hyperlipidemia, diabetes and sleep apnea) or for those with a BMI greater than 30 kg/m^2 with diagnosed complications in conjunction with diet and exercise.

Physicians are sometimes reluctant to prescribe medications for obesity, possibly because patients rapidly regain weight when treatment is discontinued. However, long-term pharmacotherapy for this chronic condition should not be dismissed. This is especially in view of the fact that the serious systemic adverse effects that have been associated with certain anti-obesity agents in the past can now be avoided with the selection of newer agents. Moreover, a landmark study by Weintraub *et al* (1) showed that appetite suppressant medication combined with diet and behaviour modification can lower body weight by as much as 10kg and maintain this weight loss for as long as 4 years. This achievement had not previously been accomplished by any method except gastric surgery. The drugs available for the treatment of obesity in the past have been outlined below.

In the 1950s and 1960s, amphetamines were the major prescription medications for weight loss (2). In the years after amphetamines were no longer approved for long-term weight loss, behavioural treatments and diet changes were used as the main strategy to achieve weight loss. No new medications were approved by the FDA for obesity treatment from 1973 to 1996 (2).

Weight loss medications again gained wide popularity in the mid 1990s with the introduction of fenfluramine and dexfenfluramine. However, these drugs were withdrawn from the market due to the continued reporting to the FDA of heart valve defects.

Currently, there are limited choices for pharmacotherapy for obesity, yet understanding of the disease and the individual neurochemical processes involved is rapidly developing. The introduction of the new agents, sibutramine, orlistat and rimonabant demonstrates that medications are evolving to treat obesity (Table-1).

Table -1 : Newer Antiobesity agents

Drug	Class	Dose
Sibutamine	Centrally acting appetite suppresent (Inhibits reputake of serotonin and nonadrenaline. This results in suppression of appetite and inducement of satiety)	Usual initial dose : 10mg OD Maximum dose : 15mg OD Patients who do not tolerate 10mg can be switched to 5mg OD
Orlistat	Lipase inhibitor (Inhibits lipase necessary for absorption of body fat)	120mg tid
Rimonabant	CB_1 receptor antagonist	5-20mg tid

A combination of drug therapy, dietary therapy and increased physical activity provides the most successful therapy for weight loss and weight maintenance.

Sibutramine:

Sibutramine blocks the presynaptic reuptake of both norepinephrine and serotonin, thereby potentiating the anorexic effect of these two neurotransmitters in the CNS. Unlike fenfluramine and dexfenfluramine, which also release serotonin from presynaptic sites, sibutramine did not cause valvular heart disease in 133 patients treated for a mean of 7.6 months (3). Sibutramine given at 2-5 times the therapeutic dose was found to lack acute abuse potential in comparison with 20mg of D-amphetamine (4).

The randomized, double-blind trial of sibutramine involved 605 obese subjects recruited from 8 European centers (5). After being treated for 6 months with a hypocaloric diet and 10 mg/d of sibutramine, 467 subjects achieving more than 5% weight loss were more randomized to 18 months of further treatment with 10-20 mg/d of sibutramine (n=352) or placebo (n=115). Forty-two percent of subjects in the sibutramine group and 50% in the placebo group dropped out. Of subjects completing the trial, 43% in the drug group as compared with 16% in the placebo group maintained 80% or more of their original weight loss. An absolute weight loss of 8.9 kg was documented from baseline. Allowing for dropouts and nonresponders, one of five subjects who received sibutramine for the entire 24 months of the study maintained 80% or more of their original weight loss.

In a number of studies lasting up to I yr, weight loss with a hypocaloric diet and 10-20 mg/d of sibutramine ranged from 4.7-7.3% of baseline or about 2-3 times that observed in placebo treated control subjects (6-9). The decline in fasting triglyceride levels ranged from 4.5-42 mg/dl (0.051-0.47 mmol/L), the increase in high-density lipoprotein cholesterol ranged from 3-9

mg/dl and changes in LDL were small and variable. Consistent side effects of sibutramine therapy included a 0.3-2.7 mmHg increase in systolic blood pressure, a 1.6-3.4 mmHg increase in diastolic blood pressure, and a 2-5 beat per minute increase in resting heart rate. Other side effects included headache, insomnia, dry mouth and constipation. One study found that the frequency of adverse events could be reduced without sacrificing efficacy if sibutramine was given intermittently as 12 wk of active drug alternating with 7 wk of placebo over a 44-wk period (10).

ORLISTAT:

Orlistat is the only approved inhibitor of the gastrointestinal lipases, predominantly pancreatic lipase, necessary for the hydrolysis of triglyceride to free fatty acids in the lumen of the gut. Because this agent can reduce the absorption of dietary fat by up to 30%, it produces weight loss comparable to or greater than that obtained by placing an individual on a fat restricted diet. Although there are no systemic side effects of orlistat due to its lack of absorption, supplementation of fat-soluble vitamins may be prudent to prevent the development of vitamin deficiency syndromes.

In a representative 2yr study, 1187 obese adult subjects were placed on a hypocaloric diet for 4wk (11). Of this group 892 subjects were randomized to receive placebo 3 times a day or orlistat, 120 mg 3 times a day, for 52 wk. At the end of this period, the orlistat group was again randomized to 3 times a day for an additional 52wk. During the first year, orlistat treated subjects lost more weight than placebo treated subjects (8.76 to 5.81 kg; $p < 0.001$). During the second year, subjects treated with orlistat 120mg 3 times a day regained 35.2% of lost weight; subjects treated with orlistat 60mg 3 times a day regained 51.3% of lost weight and subjects treated with placebo regained 63.4% of lost weight.

In contrast to sibutramine, orlistat causes significant reductions in total and low-density lipoprotein cholesterol and in systolic and diastolic blood pressure. Gastrointestinal side effects of orlistat, including loose stools, increased defecation, fecal urgency and oily discharge are significantly more common than observed with placebo and may lead to discontinuation of the drug. One study found that concomitant use of natural fibre (psyllium mucilloid) may reduce the incidence of these gastrointestinal side effects (12).

Because their mechanisms of action differ, it is reasonable to ask whether combined therapy with orlistat plus sibutramine might produce a greater degree of weight loss than is achievable with either agent alone. One study of 34 obese women addressed this issue (13). Subjects were treated with sibutramine for 1 yr and achieved a mean weight loss of 11.6% of initial weight. They were then randomly assigned in a double blind

fashion to 16 additional wk of treatment with either sibutramine plus placebo or sibutramine plus orlistat. The study demonstrated that addition of orlistat produced no additional weight loss during the 16wk of combined therapy. This finding suggests that weight loss with currently available agents may be limited to about 10% of starting weight. Only 20-30% of unselected individuals will come close to this degree of weight loss, body weight begins to rise again after 12-18 months of treatment. The possibility of long term failure of these agents must be borne, in mind and drug therapy should be discontinued if significant weight regain occurs.

RIMONABANT:

It is a cannabinoid receptor antagonist. Cannabinoids are well known for centuries for its appetite enhancing effect. Keeping this knowledge in mind people discovered the endo cannabinoid system. The cannabinoid (EC) system contributes to the physiological regulation of energy balance, food intake, and lipid and glucose metabolism through both central and peripheral effects. EC acts on various region of hypothalamus viz lateral hypothalamus, acute and paraventicular nucleus. At periphery EC has direct effect on fat cells through cannabinoid one (CB_1) receptor, it promotes, lipogenesis and positive energy balance. CB_1 receptor is also expressed in various other peripheral organs including autonomic nervous system, liver, muscle and gastrointestinal tract (14). Administration of the first endocannabinoid discovered, anadamide, in hypothalamus or of 2-arachidonoxyl-glycerol in the nucleus accumbens can provoke food intake in satiated rodents (15).

Rimonabant is a CB_1 receptor antagonist, it increases serotonin and dopamine levels by increasing their turn over. Centrally it blocks CB_1 receptor, inhibits orexigenic action of ghrelin and causes reduction in appetite. Peripherally, it causes lipolysis as well as increases basal metabolic rate. The effects of Rimonabant were studied in more than 1500 patients in Rimonabant in obesity Europe (RIO Europe) study (16). The results of this phase 3 study showed that weight loss at 1 year was significantly greater in patients treated with rimonabant 5mg (mean-3.4 kg, [SD 5.7]; $p=0.002$ vs placebo) and 20 mg (-6.6 kg, [7.2]; $p < 0.001$ vs placebo) compared with placebo (-1.8 kg [6.4]). Significantly more patients treated with rimonabant 20mg than placebo achieved weight loss of 5% or greater ($p < 0.001$) and 10% or greater ($p < 0.001$). Rimonabant 20mg produced significantly greater improvements than placebo in waist circumference, HDL-cholesterol, triglycerides and insulin resistance and prevalence of the metabolic syndrome. The effect of rimonabant 5 mg were of less clinical significance. Rimonabant was generally well tolerated with mild and transient side effects.

Rimonabant is prescribed in a doses of 5-20 mg/day. Common side effects are

nausea, vomiting, depression and anxiety. As seen with other antiobesity drugs, tolerance is quickly developed to its anorexic effect (16).

Many drugs are available and many more are under development for management of obesity *viz* leptin analogues, neuropeptide antagonist, amylin, melanocortin-4 receptor antagonist, and glucagon - like peptide agonists (GLP-1)

The need of new drugs arises because most of the available drugs are associated with limitations like can reduce up to 5-10% of body weight, rebound weight gain, associated side effects and lack of predictability.

In conclusion, available antiobesity drugs are effective only in presence of hypocaloric diet and physical activity. Most of the drugs can reduce up to 10% of body weight. So, quest for novel anti-obesity drug is not yet over.

References :

1. Weintraub M (1992). Long term weight control study: conclusions. *Clin Pharmacol Ther* **51:** 642-46.
2. David S W (2003). Pharmacological therapy of obesity: Past, Present and Future. *J Clin Endocrine) Metab* **88:** 2462-69.
3. Bach DS, Rissanen AM, Mandel CM, Shepherd G, Weinstein SP, Kelley F, Seaton TB, Patel B, Pekkarinen TA. Armstrong WF (1999). Absence of cardiac valve dysfunction in obese patients treated with sibutramine. *Obes Res* **7:** 363-69.
4. Schuh LM, Schuster CR, Hopper JA, Mendel CM (2000). Abuse liability assessment of sibutramine, a novel weight control agent. *Psychopharrna-cology* **147:** 339-46.
5. James WPT, Astrup A, Finer N, Hilsted J, Kopelman P, Roosner S, Saris WHM, Van Gaal LF (2000). Effect of sibutramine on weigh maintenance after weight loss: a randomized trial *Lancet* **356:** 2119-25.
6. Apfelbaum M, Vague P, Ziegler O, Hanotin C, Thomas F, Leutenegger E (1999). Long term maintenance of weight loss after a very low calorie diet: a randomized blinded trial of the efficacy and tolerability of sibutramine. *Am J Med* **106:** 179-84.
7. Smith IG, Goulder MA (2001). Randomized placebo controlled trial of long term treatment with with sulbutramine in mild to moderate obesity *J Fam Pract* **50:** 505-12.
8. Dujovne CA, Zavoral JH, Rowe E, Mendel CM (2001). Effects of sibutramine on body weight and werum lipids: a double blind, randomized, placebo controlled study in 322 overweight and obese patients with dyslipidemia. *Am Heart J* **142:** 489-97.
9. Hazenberg BP (2000). Randomized, double blind, placebo controlled with center study of sibutramine in obese hypertensive patients. *Cardiology* **94:** 152-48.
10. Wirth A, Krause J (2001). Long term weight loss with sibutramine a randomized controlled trial. *JAMA* **286:** 1331-39.
11. Davidson MH, Hauptman J, DiGirolamo M, Foreyt JP, Halsted CH, Mo D, Heimburger DC, Lucas CP, Robbins DC,

Chung J, Heymsfield SB. Weight control and risk factor reduction in obese subjects treated for 2 years with orlistat: a randomized controlled trial. *JAMA* **281:** 235-242.

12. Cavaliere H, Floriano I, Medeiros Neto G (2001). Gastrointestinal side effects of orlistat may be prevented by concomitant prescription of natural fibre (psylJium mucilloid) *Int J Obese* **25:** 1095-99.
13. Wadden TA, Berkowitz RI, Womble LG, Sarwer DB, Arnold ME, Steinberg CM (2000). Effects of sibutramine plus orlistat in obese women following 1 year of treatment by sibutramine alone: a placebo controlled trial Obes Res and 431-37.
14. Di Marzo V, Goparaju SK, Wang L, et al (2001). Leptin-regulated endocannabinoids are involved in maintaining food intake. *Nature* **410:** 822-25.
15. Di Marzo V, Bifulco M, De Petrocellis L (2004). The endocannabinoid system and its therapeutic exploitation. *Nat Rev Drug Dicov* **3:** 771-84.
16. Gaal Luc F van, Rissanen AM, Scheen AJ, Zieglar O, Rossner S (2005). Effects of the Cannabinoid-1 receptor blocker rimonabant on weight reduction and cardiovascular risk factors in overweight patients: 1-year experience from the RIO-Europe study. *Lancet* **365:** 1389-97.