Challenges in obesity research

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Abstract

Obesity is the main nutritional problem and one of the most important health problems in developed societies. Central to the challenge of obesity prevention and management is a thoroughly understanding of its determinants. Multiple socio-cultural, socio-economic, behavioural and biological factors —often interrelated and many of them still unknown or poorly understood— can contribute to the establishment and perpetuation of obese phenotypes. Here, we address current research challenges regarding basic aspects of obesity and emerging science for its control, including brown adipose tissue thermogenesis and browning of white fat as possible therapeutic targets for obesity, the influence of the microbioma, and genetics, epigenetics, nutrigenomics and nutrigenetics of obesity. We also highlight hot topics in relation to food and lifestyle as determinants of obesity, including the brain mechanisms underlying environmental motivation to eat, the biological control of spontaneous physical activity, the possible role of concrete foods and food components, and the importance of early life nutrition and environment. Challenges regarding the connections of obesity with other alterations and pathologies are also briefly addressed, as well as social and economical challenges in relation to healthy food production and lifestyle for the prevention of obesity, and technological challenges in obesity research and management. The objective is to give a panoramic of advances accomplished and still ahead relevant to the different stakeholders engaged in understanding and combating obesity.

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Key words: *Obesity. Research challenges. Body weigth.*

RETOS DE INVESTIGACIÓN EN OBESIDAD

Resumen

La obesidad es el principal problema nutricional y uno de los principales problemas de salud en las sociedades desarrolladas. Conocer y entender los determinantes de la obesidad es clave para conseguir prevenirla y controlarla. Múltiples factores sociales, económicos, culturales, conductuales y biológicos —a menudo interrelacionados y muchos todavía desconocidos o mal conocidos— pueden contribuir al establecimiento y mantenimiento de fenotipos obesos. Aquí destacamos retos en relación con aspectos básicos de la obesidad y ciencia emergente para su control: la termogénesis en el tejido adiposo marrón y el "pardeamiento" *(browning)* de la grasa blanca como posibles dianas anti-obesidad, la infuencia del microbioma, y la genética, epigenética, nutrigenómica y nutrigenética de la obesidad. También destacamos temas abiertos en relación con la alimentación y el estilo de vida como determinantes de la obesidad, incluyendo los mecanismos neuronales que subyacen a la motivación a la ingesta por claves ambientales, el control biológico de la actividad física espontánea, el posible papel de alimentos y componentes concretos de alimentos, y la importancia de la alimentación y el ambiente en etapas tempranas de la vida. Nos referimos asimismo a los retos en relación con la conexión de la obesidad con otras alteraciones y patologías, los retos sociales y económicos en relación con el estilo de vida y la producción de alimentos saludables para la prevención de la obesidad, y los retos tecnológicos en la investigación y tratamiento de la obesidad.

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Palabras clave: *Obesidad. Retos de investigación. Peso corporal.*

Abbrevations

BAT: brown adipose tissue.
BMI: body mass index.
BRITE: brown-in-white.

GWAS: genome-wide association studies.
SNP: single nucleotide polymorphism.
UCP1: uncoupling protein 1.
WAT: white adipose tissue.

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Introduction

Obesity is the main nutritional problem and one of the most important health problems in developed societies. In fact, combating obesity constitutes a primary health objective, since many alterations and pathologies related to diet and lifestyle associate with obesity,

and strategies and treatments for obesity control have proved quite inefficient to date. The issue is complicated, most probably, by the fact that we are dealing with a set of "obesities", rather than with a single condition, since the aetiology of obesity can be very diverse.

Main challenges in relation to obesity relate to key aspects including its causes and mechanisms, the physiological processes affected, and possible treatments and preventive measures, and the definition of the main objectives to face for progress in these areas in upcoming years (fig. 1). These objectives can be grouped in 6 main blocks, which are very much interrelated and include many collateral aspects:

1) Basic aspects of obesity and the strategies for its control.
2) Food and lifestyle as determinants of body weight and its alterations.
3) The connections between obesities and associated clinical alterations and pathologies.
4) Social challenges in relation to environment, lifestyle and the prevention of obesity .
5) Economical and business challenges in relation to healthy food production and the improvement of diet.
6) Technological challenges in obesity research and management.

Here, we will try to define main challenges and research objectives in each of these blocks, with special focus on specific aspects that are being studied in our research group.

Basic aspects of obesity (causes, mechanisms, biochemical processes involved) and emerging science for its control

Central to the challenge of obesity prevention and management is a thoroughly understanding of its causes. Obesity can be most simply defined as the presence of excess body fat as adipose tissue and it is ultimately caused by chronic energy imbalance whereby energy intake exceeds energy expenditure. However, multiple endogenous and exogenous factors, i.e., a variety of socio-economic, socio-cultural, behavioral and biological factors —often interrelated and many still unknown or poorly understood— can contribute to the establishment and perpetuation of obese phenotypes reflecting this imbalance.

Biochemically speaking, the level of fat reserves in mammals depends on the interplay between several interconnected processes, including the control of feeding behaviour, energy expenditure, partitioning of nutrients among tissues and anabolic *versus* catabolic
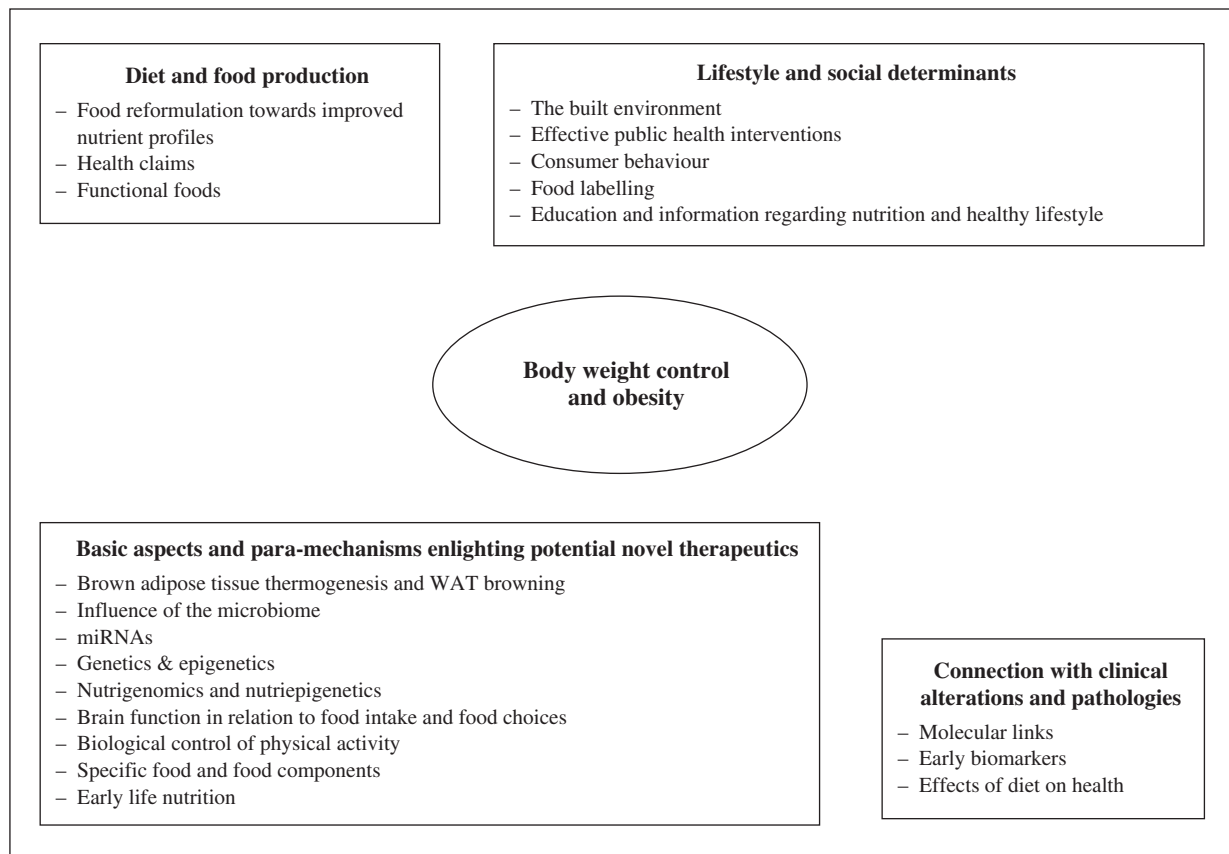

Diet and food production
– Food reformulation towards improved nutrient profiles
– Health claims
– Functional foods

Lifestyle and social determinants
– The built environment
– Effective public health interventions
– Consumer behaviour
– Food labelling
– Education and information regarding nutrition and healthy lifestyle

Body weight control and obesity

Basic aspects and para-mechanisms enlighting potential novel therapeutics
– Brown adipose tissue thermogenesis and WAT browning
– Influence of the microbiome
– miRNAs
– Genetics & epigenetics
– Nutrigenomics and nutriepigenetics
– Brain function in relation to food intake and food choices
– Biological control of physical activity
– Specific food and food components
– Early life nutrition

Connection with clinical alterations and pathologies
– Molecular links
– Early biomarkers
– Effects of diet on health

Fig. 1.—Key issues and challenges in the control of body weight and obesity.

pathways, and on-going processes within fat depots determining adipocyte number (e.g., preadipocyte proliferation, adipocyte differentiation, adipocyte apoptosis).[1] Each of these processes involves many signals, receptors, signal transduction cascades and downstream targets including metabolic enzymes, transport proteins, and additional regulatory factors. Alterations in key molecular elements of this complex system can favour a positive energy balance leading to obesity, as can subtle alterations in several "minor" elements, when presenting in a concerted way. These alterations can be inherited or acquired as a consequence of environmental exposritions, particularly early life experience as part of developmental programming. Importantly, once identified, processes determining body fat and key molecular elements in them constitute new potential therapeutic targets for obesity management.

Brown adipose tissue thermogenesis and browning of white fat as therapeutic targets for obesity

The role of uncoupling protein 1 (UCP1)-mediated brown adipose tissue (BAT) thermogenesis in the regulation of energy balance and maintenance of body weight is well known in rodents. In adult humans, BAT has long been believed to be absent or negligible, but recent studies using fluorodeoxyglucose-positron emission tomography in combination with computed tomography demonstrated the existence of metabolically active BAT in healthy adult humans.[2] Intriguingly, functional and active BAT is inversely correlated with age and body mass index (BMI) in humans. Furthermore, in addition to purported regulatory roles of BAT in energy expenditure and body fat content, thermogenic BAT is a major site for lipid breakdown and glucose uptake, and there is recent evidence from animal studies that BAT controls blood triglyceride clearance[3] as well glucose homeostasis and insulin sensitivity.[4] These and other findings have suggested that BAT may be involved in the etiology of diabetes mellitus and dyslipidemias, independently of and/or secondly to obesity.

Another phenomenon long known for which there is a renewed interest nowadays is WAT-to-BAT remodelling or *browning* of white fat, which implies stimulating catabolic versus anabolic lipid metabolism and, eventually, UCP1 expression and function in fat depots traditionally considered white adipose tissue (WAT). Brown-like adipocytes —called "BRITE" (from brown-in-white) or "beige"— can be induced in WAT depots of rodents in response to appropriate stimuli. The origin of these cells, i.e. whether they result from trans-differentiation of pre-existing white adipocytes or from the recruitment of different resident precursor cells, is controversial. Regardless of this, available data from animal studies indicate that WAT browning can confer protection from obesity and related diseases and be induced by a variety of physiological, pharmacological and nutritional agents, as well as in response to certain targeted genetic manipulations[5]. Molecular mechanisms involved in WAT browning and their control constitute an active area of research. From these studies, synthetic chemicals and humoral factors able to induce BAT activation and/or WAT browning (many factors have proven to do both in animal models) can be designed/implemented. Alternatively, the possibility is envisaged to engineer synthetic brown adipocytes and BRITE cells *ex vivo* for autologous transplantation.[6]

Many research questions and challenges remain open in this area, among them: Does BAT function significantly impact energy balance and human obesity? What are the bases for the differences in BAT amount found among humans? What are the causes for the observed age-related decline in BAT activity in humans? What are the pathways linking BAT function to systemic insulin sensitivity and pancreatic beta cell function? What is the physiological significance if any of WAT browning in animals or humans? Is BAT activity or WAT browning activatable by dietary factors in a meaningful way? To what extent are pathways and factors involved in BAT development and WAT browning identified in animal research conserved in humans? What is the molecular signature of brown adipocyte and BRITE progenitor cell populations (this knowledge is a pre-requisite for cell-based therapies for the treatment of obesity and diabetes)? In addition, potential counterregulatory mechanisms (e.g., increased appetite) and side-effects (e.g. exaggerated heat production; ectopic body fat deposition) associated with anti-diabesity strategies based on activation of BAT or WAT browning should be rigorously considered. These are open questions that are being addressed by running projects such as the EU-funded DIABAT (http://www.diabat.org/).

Influence of the microbioma

Interest has surged in the last years regarding the possible role of the intestinal microbiota as potential novel contributors to the increased prevalence of obesity, metabolic syndrome, and type 2 diabetes. Mechanisms by which the gut microbiome may influence metabolism and energy homeostasis include regulation of energy uptake from diet, interaction with signalling molecules involved in host metabolism, modification of gut permeability, release of gut hormones, and low-grade, chronic inflammation, the latter being a hallmark of obesity-related diseases.[7] For instance, changes in the composition and metabolic function of the gut microbiota in obese individuals have been described which appear to enable the "obese microbiota" to extract more energy from the diet.[8] Additionally, various host pathways, mainly emanating from epithelial cells, have been characterized in the last years that might mediate the effects of microbiota on metabolism, including Fiaf, Ampk, Gpr41, Gpr43, Glp2, and the endocannabinoid system, among others.[9]

Recent advances in microbial DNA sequencing technologies have enabled the application of whole-genome sequencing technologies for metagenomic DNA analysis of complex ecosystems such as the human gut. Dietary factors and caloric intake appear to affect the composition of the gut microbiome, which also appears to be shaped by genetics and other environmental factors.[9] However, this research is still in its infancy. Additionally, most findings in the field of microbiome and obesity are based on rodent studies, and the relevance to human biology requires further investigation. Challenges are to gain a proper understanding of genetic and environmental influences on the microbiota and of the consequences of structural and functional changes within the microbiota on metabo-inflammatory diseases. It will also be important to reveal potential long-term consequences of antibiotic therapies at various ages of life, which could contribute to some forms of iatrogenic obesity.[9]

Genetics of obesity

Most cases of human obesity —aside from rare cases of monogenic obesity— are thought to be complex, i.e. to depend on genetic variation at several susceptibility loci (each entailing usually only a modest effect *per se*), with a variable contribution from environmental factors such as diet and physical activity.

The field of the genetics of obesity was dominated till 2006 by candidate gene studies examining the association of concrete polymorphisms (usually single nucleotide polymorphisms, SNPs) in one or a few *candidate* genes with obesity and/or obesity-related phenotypes such as BMI or percent body fat as continuous traits.[10] Candidate genes are those with higher prior probability for phenotypic involvement on the basis of different criteria including biology, pharmacology, transgenic and knockout murine models relevant to obesity, and positional information, among others. These studies resulted in the suggestion of numerous *obesity* genes (at least 127), related for instance to the central regulation of energy balance, adipogenesis, lipid turnover, adaptive thermogenesis, and signaling by insulin and other extracellular signals impinging on energy metabolism.[11] However, none of these genes or combinations could be firmly validated, usually due to inadequate statistical power of the studies and insufficient biological and genomic knowledge. Since 2006, genome-wide association studies (GWAS) have been implemented which allow scanning the entire genome for common disease-associating SNPs in a hypothesis-free manner in large cohorts of familial-unrelated people, and are ideally suited to detect common (frequent) variants with small effect sizes. These unbiased, powerful studies have allowed the identification of about 40 loci related to human obesity, among which the fat mass and obesity associated gene (FTO) is the most replicated and the one showing the highest statistical significance.[12]

Complete understanding of identified associations in either candidate gene or GWA studies is often hampered by lack of data on the specific functional significance of the polymorphism(s) under investigation. Functional assessment remains a challenge in many instances, especially for variants located in non-coding areas of the genome including intronic or intergenic regions, which could either be linkage disequilibrium markers of the causal variants or true causal variants influencing for instance gene function or transcript regulation. The vast majority of obesity susceptibility variants identified in GWAS lie in non-coding regions.[12]

Additional challenges come from the fact that the contribution of a given SNP to obesity/adiposity can be modulated by the presence of other SNPs in the same gene or other genes. Therefore, the influence of haplotypes (comprising specific combinations of genetic polymorphisms in a single allele) and gene-gene (epistatic) interactions need to be considered. Epistatic interactions relevant for obesity development are largely unknown and yet to be analysed and mapped in a systematic way, using appropriate tools.

Importantly, despite the advances in the genetics of obesity, the combined effect of all loci identified so far account for only about 2-4% of the total heritability of common forms of obesity (estimated to be between 40-70%).[12] These numbers confirm the complex nature of obesity and the challenge to identify additional factors that may unravel some of the missing or hidden heritability of obesity. Such factors may include interactions between multiple genes and environmental factors (see the heading on Nutrigenomics below)[12] and the contribution of other types of variants not covered by current GWAS design, including low-frequency and rare variants, copy number variations and epigenetic modifications.[12] The latter may also be part of the mechanisms by which the environment could induce biological changes with lasting effects on obesity-related parameters (see next).

Epigenetics of obesity

Epigenetics refer to mitotically heritable modifications that regulate gene activity and/or expression rather than its DNA sequence.[13] Epigenetic marks are tissue specific and include, though are not limited to, DNA methylation and histone modifications. Epigenetic marks can be programmed already in the intrauterine environment and can be modulated by environmental influences including diet, besides genetic influences.[14] The epigenome is thus seen as a malleable interface between the environment and the genome. Changes in the epigenome at critical developmental stages can be shaped by the environment and have long-lasting effects affecting health and susceptibility to disease in later life, including obesity and metabolic syndrome.[10]

To what extent epigenetic modifications contribute to the above findings and to the total heritability of com-

mon forms of obesity is presently unknown. However, several lines of evidence suggest their contribution. At least one "obesity gene" codes for an enzyme involved in the control of epigenetic marks (the FTO locus, encoding a DNA-demethylase enzyme[15]), while genetic disruption of one of such enzymes (the histone H3k9-specific demethylase Jhdma2a) has been shown to result in obesity and hyperlipidemia in mice.[16] Many "obesity genes" critical to energy balance are regulated by epigenetic mechanisms depending on nutritional clues. Examples are the MC4R gene, which has reduced methylation in the brain following long-term exposure to a high fat diet in mice;[17] the POMC gene, whose promoter methylation in the hypothalamus is sensitive to early overfeeding[18] and, more specifically, to early leptin administration;[19] the leptin gene, whose promoter methylation in adipose tissue is modified by a high-fat diet;[20] and the PGC1a gene, whose promoter methylation in human umbilical cord and human muscle has been related, respectively, to maternal pregestational BMI[21] and to high-fat overfeeding in a birth-weight-dependent manner.[22] Finally, methylation at five candidate gene promoters at birth has been shown to be associated with child's later adiposity in humans[23].

Challenges regarding epigenetics of obesity include the translation of animal results to humans while dealing with the great tissue specificity of epigenetics marks (relevant tissue samples may be inaccessible, such as brain samples) and the integration of genetic and epigenetic information, for instance, linking the causally unexplained GWAS association signals with epigenetics and even further, at a large-scale genome —wide level.

miRNAs and obesity

MicroRNAs are endogenous small non-coding RNAs involved in the post-transcriptional regulation of gene expression by binding to complementary sequences often located in the 3' untranslated region of target mRNAs, leading to their translation repression or degradation. miRNas have been widely implicated in the fine-tuning of many physiological processes, including the pathogenesis of type 2 diabetes and obesity.[24] Microarray studies have highlighted an altered profile of miRNA expression in insulin target tissues in diabetic and obese models. Emerging evidences suggest that miRNAs play significant roles in insulin production, secretion and actions, as well as in diverse aspects of glucose homeostasis and lipid metabolism. They are also involved in many functional aspects of adipocyte differentiation and potentially contribute to the pathogenesis of obesity and its related medical complications. The identification of tissue-specific miRNAs implicated in type 2 diabetes and obesity might be useful for the future development of effective strategies for early diagnosis and therapeutic intervention of obesity-related medical complications.

Nutrigenomics and nutrigenetics of obesity

Nutrigenomics studies the links and interrelationships among diet, genetic makeup, and physiological responses, at a genome-wide level and in a systematic manner. From a nutrigenomics perspective, particular nutrients, non-nutritive food components, or nutritional regimes are dietary signals that are detected by cellular sensor systems that influence gene and protein expression and, subsequently, metabolite production.[25] Nutrigenomics attempts to study these "dietary signatures" in specific cells, tissues and organisms to understand how these signals influence homeostasis and thus, regulate health and the progress of diet-related chronic diseases, such as obesity and metabolic diseases. Features of such signatures are being investigated as biomarkers of early disruption of homeostasis, of interest in the context of public health strategies (disease prevention) as well as a basis for the substantiation of novel health claims made on food. This is the focus of a large collaborative EU funded project currently under development, BIOCLAIMS (http://bioclaims.uib.es/).

Nutrigenomics is very relevant for understanding obesity as well as the links of obesity with associated diseases. Dietary chemicals interact with the biochemical processes involved in the control of body fat (hunger and satiety, intestinal nutrient absorption, BAT thermogenesis and WAT browning, fat oxidation, lipogenesis), as well as with inflammation and stress pathways. Furthermore, specific food chemicals have been shown to ameliorate obesity in animals by molecularly defined mechanisms.[5,26,27] This knowledge has led to the proposal of nutritional strategies (functional foods or nutraceuticals) for weight management based on selected traditional foods (containing functional ingredients) or novel foods, although current knowledge in this area is based largely on animal and cell studies, with limited evidence from well designed, human intervention studies. The latter remain a challenge for most "anti-obesity" foods/compounds proposed so far (ethical considerations might be a limiting factor for such studies).

Nutrigenomics core concepts also imply that the individual genetic background can influence nutrient status, metabolic response to nutrients/diets and predisposition to diet-related diseases. This is at the foundations of Nutrigenetics, which has many implications in the field of obesity. Not everyone becomes overweight or obese in an obesogenic environment, suggesting that there are genetic or acquired factors interacting with actual environmental factors to predispose some individuals to obesity. Gene-environment interaction can also influence the outcome of weight-loss programs and weight-management strategies in overweight and obese subjects.[28,29] Not only SNPs in obesity-related genes may play a role; "metabotypes", i.e., an individual's distinct metabolic response to a specific intervention (e.g., calorie restriction), alone or in combination with genotypes, may also be a good predictor of personal outcomes.[30]

Ultimately, Nutrigenetics has the potential to provide, at least in part, the rationale for personalized dietary recommendations based on the individual's genetic constitution and biochemical individuality, to prevent and manage obesity and other diet-related diseases with maximal efficacy, and from an early stage. However, many challenges persist. Adequate statistical power is extremely hard to achieve in gene x environment interaction analysis, and better tools for the comprehensive and reliable capture of diet and other environmental exposures in timely and economically feasible approximations that can be effectively used in large-scale genetic epidemiological studies such as GWAS are needed.[12] Furthermore, the large majority of Nutrigenetics studies published so far have examined interactions in a simple scenario, in which single (or a few) dietary component and single (or a few) genetic polymorphism is/are analyzed in relation to well-defined single health outcomes/traits. Each individual possesses potentially hundreds of "at-risk" gene variants and consumes a highly-complex diet. Understanding gene x diet interactions and their relationship with health in general (or a range of relevant health outcomes) at a genome-wide level remains a big challenge, requiring strong investment, post-genomic approaches and adequate mathematical and bioinformatic tools.

Food and lifestyle as determinants of body weight control and its alterations

The increased availability and accessibility of palatable, energy dense foods and the reduced requirement for physical exertion during working and domestic life are usually seen as the two most critical factors underlying the obesity pandemic. However, food intake and physical activity behaviors might not always be a matter of conscious choice, and understanding causal pathways for them remains a challenge in obesity research. These aspects, together with the possible contribution to the current obesity pandemics of concrete food components, diet at critical developmental periods, and lifestyle factors other than total daily energy intake and physical activity are briefly addressed next.

Environmental and psychological factors as determinants of food intake

The brain plays a central role in the control of food intake and energy balance by coordinating on-going information about the quality and quantity of calories being consumed (mainly via satiation signals), the levels of fuels already in the plasma (via direct sensing by specialized cells in the brain and elsewhere), and the amount of energy present in the various storage depots (via adiposity signals, such as insulin and leptin).[31] However, factors unrelated to energy balance such as where and when food might be available, relevant aspects of the social situation, memory for key relevant information from past experiences (learned cues), and hedonic factors, among others, also control appetite and eating and are computed by the brain. One fundamental question in obesity research is to decipher the brain mechanisms underlying environmental (non-physiological) motivation to eat, and how these mechanisms are integrated with the physiological regulatory control of food intake.[32]

Limited investigations in obese subjects show that obesity is associated with alterations in neural and behavioural mechanisms of food reward and parallels have been drawn between drug and food addiction.[33] Internal mechanisms may favor neurophysiologic responses to food cues that result in overconsumption in a context of food abundance and food salience (through marketing and advertising).[34] This investigation is, however still in its infancy. It is unclear, for instance, whether genetic and other pre-existing differences in reward functions cause obesity, or if the obese state secondarily changes reward mechanisms, perhaps to contribute to accelerate/ perpetuate obesity, or both.

How is spontaneous physical activity determined?

Many environmental cues may play a role in determining voluntary exercise and spontaneous physical activity —defined as to include all energy expended due to activity, exclusive of volitional exercise and also called non-exercise activity thermogenesis— in the individual. These cues interact with a biological control of these activities, which remains poorly understood (reviewed in[35]). An important role of dopamine, in addition to other neural signalling networks (e.g. the endocannabinoid system), in the control of voluntary exercise has been proposed. Other reports point to a key role for orexins in the control of spontaneous physical activity. Brain reward centers are involved in both types of physical activities and eating behaviours, likely leading to complex interactions. Moreover, like eating, voluntary exercise can be addictive. Genes are being identified whose sequence variants have been associated with either the level of physical activity or indicators of sedentarism.[35,36] Future studies should explore the neurobiology, endocrinology and genetics of physical activity and sedentary behaviour by examining key brain areas, neurotransmitters and hormones involved in motivation, reward and/or the regulation of energy balance, as well as in the intertwined control of physical activity and eating.[35, 37]

Is there a role for specific foods or food components?

It has been speculated that not only overall excess energy intake but also excess of particular nutrients/foods may play a role in the aetiology of obesity. In particular, a role for fructose and fructose-derived sweeteners

(sucrose, high-fructose corn syrup in beverages) has been proposed, although the issue remains controversial.[38,39] Increasing intake of the omega 6 fatty acid linoleic acid has also been associated to obesity in both humans and rodents, through mechanisms that appear to include proadipogeneic effects,[40] and an elevation of the endocannabinoid tone (which is overall anabolic, increasing food intake, and causing increased lipogenesis and fat storage in adipose tissue and liver).[41] The specific role for leptin[42] and other nutrients in breast milk protecting from the development of overweight and obesity later on in life have been described (see next).

Early life nutrition and obesity in adult life

Epidemiological studies in humans and controlled intervention studies in animals have shown that nutrition in early periods of life programs a number of metabolic and physiological functions throughout life.[43] In this sense, gestation and lactation are disclosed as critical periods. Continuous food restriction during these stages, for instance, may lead to permanent adaptations with lasting effects on offspring metabolic mechanisms; they may alter the propensity to different chronic diseases, such as obesity and other features of the metabolic syndrome. However, the different outcomes of these adaptations on later health appear to depend on factors such as the type, duration, period and severity of the exposure to energy restriction conditions, and they are, at least in part, gender specific. A better understanding of the factors and mechanisms involved in metabolic programming, and their effects, may contribute significantly to the prevention of obesity.[43]

Mechanisms involved in developmental programming of obesity may include epigenetic changes (see previous section) and effects on the development of anatomical structures crucial to the control of energy balance and storage, such as regulatory brain centers[44] and the adipose depots themselves[45]. Many studies have addressed the role of calorie restriction, total energy intake and diet macronutrient composition during gestation or lactation in this programming.[43] Studies dealing with the impact of concrete nutrients and signals on the metabolic programming of obesity are less represented. Important effects in this sense have been confirmed experimentally for leptin,[42] nutrients known to participate in methyl transfer epigenetic reactions,[46] leucine,[47] vitamin A,[48] and maternal dietary fats.[40,49] A challenge in this area is the translation of results to humans in studies of sufficient duration so as to catch medium and long-term effects.

The discovery of the new role of leptin intake during lactation as an essential nutrient during this period appears paradigmatic, as it deals on a protein which is present in breast milk but not in infant formula, and whose lack during lactation may be responsible for a number of obesities. The impact of the changes from breast-fed to formula fed needs further assessment.

Evidence for the essential role of leptin during the suckling period has been obtained from both animal and human studies. Firstly, studies in rats have shown that the intake of physiological doses of leptin during the suckling period prevents the animals from overweight and obesity and other metabolic alterations associated with feeding a high-fat diet during adulthood.[42,50,51] In addition, leptin treatment during lactation has lasting effects on the expression of the hypothalamic factors involved in the control of food intake, particularly POMC, leptin receptor and suppressor of cytokine signaling 3 (SOCS3).[42] Thus, leptin appears important during the lactation period in both regulating neonate food intake and affecting the developmental events involved in the control of energy balance in adulthood.[52] In addition, leptin during the suckling period has also been shown to program a better response of the adipose tissue under high fat diet conditions, by preventing the decrease of leptin receptor in internal depots and increasing the oxidative capacity of this tissue.[51] Therefore, leptin may exert regulatory effects, not only at a central level, but also peripherally. Interestingly, indirect evidence of the role of breast milk leptin during lactation in humans has also been obtained[53-55] and intervention studies should be performed to allow the inclusion of leptin in infant formulae to prevent a number of potential obesities.

Other lifestyle factors and a variety of incidents or circumstances impacting along life

Other factors related to environment and lifestyle that have been related to the obesity pandemics and deserve further study include increasing maternal age, sleep debt, reduction in variability of ambient temperatures, and increased presence in the environment and the food chain of "obesogen" chemicals such as endocrine disruptors, which appear to have estrogenic or other hormonal effects and could be especially active on the developing embryos.[56]

In addition, there are a variety of factors or circumstances (fever, different illness, injuries, infections, emotional periods, hormonal alterations, alterations of biological rhythms, etc.) that can conceivably alter the homeostasis of important control systems in the body leading to changes in the predisposition to obesity, through mechanisms that currently are out of our knowledge.

The connections between obesities and associated clinical alterations and pathologies

Many alterations and pathologies related to diet and lifestyle associate with obesity, including insulin resistance, diabetes, hypertension, dyslipidemias, cardiovascular disease and non-alcoholic fatty liver disease, which together are hallmarks of the metabolic syndrome, but also osteoarthritis of weight and non-weight

bearing joints,[55] several types of cancer,[56] neurodegenerative and autoimmnune diseases such as Alzheimer's disease[57], and others. Biological mechanisms underlying the relationship between obesity and these alterations/pathologies begin to be understood, with inflammatory, oxidative and endoplasmic reticulum stress in critical tissues including the adipose and key humoral mediators playing in general a prominent role. The main challenge is to get further insight into the molecular and cellular mechanisms linking obesity to these different forms of stress, and the later to pathologies, with the aim of understanding the system and developing therapeutics.

Social challenges in relation to environment, lifestyle and the prevention of obesity

Improving communication on food habits and different aspects of lifestyle is a key issue. The efficacy of public health messages to individuals to inform their lifestyle choices should be complemented, and thus enhanced, by environmental changes that facilitate healthy options. Education and information campaigns are an important mechanism for improving consumer understanding of food information.

The ultimate goal is to shape the physical and social-cultural food environment and improve education to facilitate people in making healthy food and lifestyle choices for effectively combating obesity and other diet-related diseases.[57] Many things can be done, and are being done around the world in this sense: architectural and urban design policies; legislative measures (e.g., taxes to certain foods, control of vending machines at schools); control on aggressive advertisement, especially to children; governmental educational campaigns to promote a healthy body weight; promotion of breastfeeding and of healthy pre(pregnancy) foetal and infant nutrition; school nutrition and physical education policies; interventions at the family, school, community, and population level, etc... The challenge is to understand the most effective ways to accomplish the goal. Studies on how individual, social, economic, cultural, gender and environmental factors affect dietary and physical activity behaviours are needed to help designing successful initiatives. Tools for evaluating the impact of institutional campaigns, legislative measures and interventions undertaken are also required.

Food labelling, in particular, has been the target of some interesting developments in Europe. The Commission White Paper of 30 May 2007 on a Strategy for Europe on Nutrition, Overweight and Obesity related health issues noted that nutrition labelling is one important method of informing consumers about the composition of foods and of helping them to make an informed choice. Knowledge of the basic principles of nutrition and appropriate nutrition information on foods would contribute significantly towards enabling the consumer to make such an informed choice. Considerable progress in Europe has been attained with the Regulation (EU) Nº 1169/2011, which will be largely in force in December 2014.

However, there are many other factors that can influence psychological and behavioural responses of the consumers. Key aspects related with the communication to the consumers of the "health-promoting" properties of foods have been addressed in recent EU legislation (see next section). However, the impact of communication on other aspects (cosmetics, biomedical devices, tools, etc.) needs further attention.

Scientists have a responsibility to provide the evidence base, on which to build policy and practice, but a range of other stakeholders, including governments, manufacturers and retailers, employers, schools, health professionals and parents, must each recognise their role and responsibilities. Effective communication between all parties is essential to build a national framework that facilitates the necessary changes in lifestyle.[58]

Economical and business challenges in relation to healthy food production and the improvement of diet

The food industry is faced with the challenge of producing tasty foods that are consistent with health status and lifestyle, and which meet consumer preferences, and to do it in a cost-effective and sustainable way.[57] This requires research and investment to develop reformulated food products with improved nutritional or nutrient profile, as well as functional food products with optimised levels of bioactive components. Incentives for such investment might be needed coming from outside the market, including protection of exploitation rights when novel food developments and claims are achieved.

Many functional foods and nutraceuticals for body weight control have been proposed which target food intake (by inhibiting hunger, stimulating satiety or limiting the bioavailability of nutrients), caloric content of foods (by including less caloric or less digestible substances substituting for sugars or fat), body composition or non-conscious energy expenditure (by stimulating thermogenesis). With the progress in Nutrigenomics and Nutrigenetics (see corresponding section above), new developments in the functional foods arena are envisaged shaped to specific groups of consumers. However, so far supportive human studies are scarce and, when available, the weight loss observed is small and should be considered mostly as a measure to prevent weight gain, or as co-adjuvant in more strict regimens. Properly designed human intervention studies and knowledge on the mechanism of action of food bioactives is required in order to support evidence based health claims.

The impact of the implementation in Europe of the Regulation (EC) No 1924/2006 that harmonises the provisions that relate to nutrition and health claims and

establishes rules governing the authorisation of health claims made on foods may be important in the following years. According to the Regulation, health claims should only be authorised for use in the Community after the European Food Safety Authority (EFSA) has carried out a scientific assessment of the highest possible standard. Moreover, article 21 of this Regulation protects the investments to obtain scientific data and other information required to substantiate a claim by stating that they may not be used for the benefit of a subsequent applicant for a period of five years from the date of authorisation, unless the subsequent applicant has agreed with the prior applicant that such data and information may be used. This clearly promotes R&D investments in the food sector. Among the health claims specified in this legislation are those related with overweight/obesity: *"health claims describing or referring to slimming or weight-control or a reduction in the sense of hunger or an increase in the sense of satiety or to the reduction of the available energy from the diet"*.

Regulation (EC) No 1924/2006 includes the provision that only those foods having an "appropriate nutrient profile" will be allowed to bear health claims. This provision – that would affect mainly the composition of saturated fat, salt and simple sugars— has not been implemented yet, due to its huge economic impact and related controversy. However, it is already producing deep changes in the food sector in Europe by stimulating food reformulation, which is also the subject of other initiatives over the world.

Technological challenges in obesity research and management

Genome-wide understanding of the genetics, epigenetics and nutrigenomics of obesity and the influence of microbiome largely depends on advances in mathematical modelling, bioinformatics and computational analysis, enabling interpretation and data meaning from high throughput analysis of genomes, metagenomes, transcriptomes, proteomes and metabolomes and the comparison between them in systems biology approaches. In parallel, there is also a need for better, feasible tools to capture and quantify diet and the "exposome", for use in large scale genetic epidemiological studies of obesity, such as GWAS, and devices enabling the control of energy expenditure in humans under free living conditions. The development of imaging technologies in relation to hunger, satiety, food preferences, taste perception and heat production applicable to human studies is also seen as a challenge in obesity research, as well as the development of novel non-invasive or micro-invasive techniques for tissue composition analysis and the provision of tissue biopsies for omics studies.

Technological solutions to obesity may include new drugs that do not have the side-effects and limited efficacy of current treatments. However, the relatively high costs, possible risks and lack of societal acceptability mean the use of medicines alone is not a long-term sustainable solution. Other technology to support healthy behaviours is envisaged, such as devices to monitor and provide feedback on energy intake and energy expenditure, along with biomarkers of health and functional status.

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