A Comprehensive Review of the Role of Genetics in Obesity in Dogs and Other Species

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INTRODUCTION

Obesity is a big problem. In recent years, the increasing incidence of obesity in companion animal species, particularly dogs, cats and horses, has attracted headlines. Near universally, poor owner management of diet and exercise are blamed for this increase in the press. However, there is a weight of evidence that obesity is perhaps best viewed as a complex homeostatic mechanism gone awry, influenced by genetics. In this lecture, I will review the role of genetics in influencing obesity susceptibility in dogs and other species and touch on what genetic studies have taught us about obesity biology.

EVIDENCE FOR THE ROLE OF GENETICS IN OBESITY

Obesity has become increasingly common over the last century, with the biggest change occurring in the last 40 years. There is a wealth of evidence that increased availability of inexpensive, calorie dense and highly palatable food, combined with an increasingly sedentary lifestyle, underpins the increase in human obesity and similar factors are likely to blame for our pet population. In recent years, the lifestyle of pets (as of humans) has changed such that most now live relatively inactive lives and have regular access to calorie dense food, and additional opportunities to scavenge. Most dogs now live indoors and don't use so much energy to keep warm. It is striking, however, that although this obesogenic environment is nearly ubiquitous, not every individual is overweight. In pets, owners' control of access to food and exercise in part explains that variability. But there is ample evidence that variation in susceptibility to obesogenic pressures are governed by genetics.

Heritability is the proportion of variation in a trait (e.g. body weight) attributable to genetic variation. Estimates for heritability of obesity in humans based on family, twin and adoption studies cumulatively show the heritability of obesity (body mass index, BMI) to be >70%. The heritability of adipose mass and related production traits are remarkably similar in pigs and broiler chickens. In dogs, clear breed predispositions to obesity suggest genetics are similarly important. Familial obesity has been described in cats and some breeds are predisposed. The heritability of physical activity is low in people (approximately 20%), and that of basal metabolic rate higher (approximately 45%) but these traits have been less well studied.

WHEN GETTING FAT WAS A GOOD THING

If sustained, obesity has adverse consequences but it is worth remembering that there are good physiological reasons for storing excess energy as fat – to build up energy reserves in case of future food scarcity. Given that the majority of evolution has occurred in a resource-poor environment, there could be evolutionary pressure to propagate genetic variants that promote the laying down of fat in times of plenty in preparation for periods of famine. This idea came to prominence as the 'thrifty gene hypothesis' and is a compelling explanation of why humans might be genetically prone to obesity. However, there are theoretical and data driven reasons to suggest genetic drift (random mutation and enrichment in different populations) may actually be responsible for high obesity susceptibility population wide.

OBESITY IS (USUALLY) A COMPLEX GENETIC DISEASE

Geneticists divide phenotypic traits into those with 'simple' or 'complex' forms of inheritance. Traits which are caused by the genetic segregation in a single gene are commonly considered 'simple' or 'Mendelian' disease. They tend to be inherited with clearly recognised patterns within families, and to be the consequence of mutations which severely disrupt the protein coding sequence of a gene. In contrast, complex traits are commonly quantitative and are the consequence of variation in many genes. Each individual in a population inherits a 'dose' of risk alleles which contribute to the overall phenotype observed.

Common obesity is a complex disease in all species. There are monogenic obesity syndromes recognised in humans (where they occur spontaneously) and laboratory animal models (genetically engineered and spontaneous). Study of those syndromes has vastly advanced our understanding of obesity biology. I will not focus on laboratory models of obesity but there is a wealth of valuable data about the mechanisms by which genes link to obesity resulting from those studies.

Identification of the genes responsible for complex obesity has advanced rapidly since the advent of genome-wide association studies which utilise data about bi-allelic single nucleotide polymorphism (SNP) markers along the genome to map obesity associated regions in a hypothesis free fashion. Such studies are vast – the most recent GWAS for BMI included approximately 700,000 people.

MONOGENIC OBESITY SYNDROMES ARE RARE BUT INFORMATIVE

Studies of patients with severe, early onset obesity tend to focus on candidate genes in small families in which severe obesity segregates. Modern high throughput DNA sequencing techniques have also been used to identify mutations in less well characterised genes. In general, these studies have pointed to the central role of the leptin-melanocortin pathway in regulating food intake (see previous lecture and references).

Approximately 5% of patients who present with early onset severe obesity carry a deleterious mutation in the melanocortin 4 receptor (MC4R). The signalling downstream of this G-protein coupled receptor can be tested *in vitro* and the severity of the cellular

signalling defect is associated with the severity and nature of the clinical syndrome observed. Hyperphagia and severe obesity are dominant clinical signs but more nuanced aspects of physiology have provided insight into other aspects of obesity pathophysiology. For instance, affected patients tend to have lower blood pressure than equivalently obese patients without MC4R mutations. That observation was critical to the recognition that leptin melanocortin signalling is instrumental in the sympathetic activation involved in development of obesity associated hypertension. Further work has identified that affected patients make distinct, unconscious choices about macronutrient preference, preferentially selecting higher fat but lower sugar foods than a control group.

Many other mutations which cause monogenic obesity affect the leptin melanocortin signalling pathway directly or indirectly. Severe, early onset obesity with other endocrine signs is present in patients who fail to produce leptin or its receptor, or who carry mutations in POMC or the enzymes responsible for its proteolytic cleavage to neuroactive peptides. Other proteins can modulate signalling at the MC4R. For instance, MRAP2, an accessory protein that interacts with MC4R is associated with obesity when disrupted in mice and mutations in the gene have been attributed as the cause of severe obesity in some human patients.

Genes responsible for mediating the development of the central nervous system have also been implicated in human monogenic obesity. One such is SIM1, a transcription factor involved in the development of key hypothalamic nuclei. Obesity also is frequently recognised as part of complex syndromes that involve autism or neurodevelopmental delay, implicating genes which govern development not just of the hypothalamus but more widely in the CNS.

Mutations which affect basal metabolic rate are very rare. Loss of function mutations in the cellular scaffolding protein *KSR2* (kinase suppressor of Ras2) have been identified in obese subjects. Affected patients have a low metabolic rate and at a molecular level the mutations are associated with impaired glucose oxidation and fatty acid oxidation in mitochondria. This confirms that defects in substrate utilisation can cause obesity but it is striking that other such mutations have not been reported.

THE GENES RESPONSIBLE FOR COMMON, POLYGENIC OBESITY IN HUMANS ARE LESS WELL UNDERSTOOD

The most recent GWAS for obesity in humans implicated over 700 regions of the genome (loci) in the control of BMI. The nature of GWAS means that the precise causative variants are hard to identify they often lie in areas of the genome which do not code for proteins. They probably alter expression or processing of nearby genes but the precise mechanisms for that can be hard to pin down. Emerging data suggests many causative SNPs have pleiotropic effects – altering expression of multiple genes – and therefore are likely to exert their effect via more than one effector pathway. Consequently, the gene labelled causative at a locus is often a 'best guess'. Nevertheless, some of the strongest associations with human BMI are in or near genes which have well characterised effects on energy homeostasis, including POMC, MC4R and PCSK1 (prohormone convertase 1, which is important in POMC processing).

In most cases, however, the nature of the cause-effect relationship between obesity and the genes in associated loci remains cryptic. The prime example of this is the FTO (fat mass and obesity related) locus which is consistently the region most strongly associated with human obesity. People who carry two copies of the risk allele are on average 3 kg heavier than individuals with two copies of the low risk allele. However, for a long time the function of FTO and the molecular basis of the association was a mystery. Subsequent human, murine and cellular studies suggested that FTO had a role in nutrient sensing and modification of appetite, possibly by varying epigenetic regulation. Those findings were later overshadowed by a study that showed the causative SNP formed long-range physical interactions with two other, further flung, genes and also affected mitochondrial respiration and adipocyte browning. The likely truth is that molecular pleiotropy means that all these mechanisms (and maybe others) play a role.

The effect of carrying common genetic variants on eating behaviour has been tested in people. Babies with high risk genotypes have a higher appetite that predates development of obesity. Specific variants have distinct effects on different aspects of eating behaviour. For instance, variants in the leptin gene (responsible for tuning background hunger) are associated with extreme snacking behaviour; in the cholecystokinin gene (usually responsible for rapid post prandial satiety) are related to extreme meal size; and variants that increase the perceived sweetness of food are inversely correlated with BMI.

We can also gain insight by considering the genes identified using GWAS collectively. For instance, the vast majority of genes associated with BMI exert their effect centrally, confirming the importance of the neural control of energy homeostasis. Another good example comes from a study which examined loci associated with insulin resistance; this showed they were linked to insulin resistance by an association with lower adipose mass in peripheral compartments. Whilst this might seem counterintuitive, it is an important piece of evidence supporting the theory that insulin resistance develops once adipose tissue has reached its limit of expandability (see previous lecture).

EPIGENETICS OF OBESITY

There is accumulating evidence that the propensity toward adult obesity is influenced by the metabolic milieu in early development and that influence can affect subsequent generations. Epidemiological studies have shown that exposure to a suboptimal nutritional environment during development is associated with an increased risk of obesity and related disease. The mechanisms underlying this 'nutritional memory' are not clearly understood but epigenetic modification of DNA is likely to play a role.

Epigenetic changes are (typically) reversible chemical modifications to DNA (associated chromosomal proteins) that don't alter the protein coding sequence but affect gene regulation by modifying the physical packing and accessibility of DNA. Epigenetic marks are heritable and are emerging as important players in the regulation of energy homeostasis.

GENETICS OF OBESITY IN ANIMALS

Obesity related traits have been intensively studied in farm animal species where overall fat mass, fat distribution and food conversion efficiency have important implications for meat quality and production costs. Similar themes appear – metabolic traits are variable, highly heritable, subject to selection and the loci which are best understood relate to the control of food intake. A full review is outwith the scope of this lecture.

In dogs and cats, monogenic obesity syndromes have not been recognised. However, a research colony of cats amongst which there was clear segregation into lean and obese phenotypes has been reported; investigation suggested a major genetic modifier acting against a background of polygenic modifiers.

Canine genetics are interesting because the way dogs have been bred to produce marked homogeneity within breeds but retain huge diversity across the species means genetic disease is both common and readily easy to map to specific genes/loci. Recently, I discovered a mutation in Labrador retrievers which is associated with food-motivation and weight; a deletion in the gene POMC leads to reduced signalling through the leptin-melanocortin signalling pathway. The mutation is associated with food motivation, weight and body condition score in affected dogs. Approximately 25% of the Labrador population is affected (most heterozygotes) but the mutation is more common in flatcoat retrievers where it is similarly associated with weight and food motivation. This is the first time a major genetic modifier of weight and eating behaviour has been pinpointed in dogs. The finding was of particular interest to non-veterinary scientists because the biology of POMC in dogs is closer to humans than that of rodents and established the importance of the POMC derived peptide β-MSH in energy homeostasis in a way that not had previously been possible by studying an animal model.

Candidate gene studies in dogs have confirmed the effect of the POMC mutation as predisposing to obesity in Labradors and investigated a number of candidate regions and genes in different dog breeds. There was no evidence for association of obesity with polymorphisms at the FTO locus in Labradors. Common polymorphisms in MC4R have been investigated in several breeds with two variants reported as obesity associated in beagles although study reporting was poor. Since genetic studies to date are limited in scope, there is much to discover about which genes are major determinants of canine obesity and their effector mechanisms.

LESSONS LEARNED FROM THE GENETICS OF OBESITY

Advances in the understanding of obesity genetics by study of patients with monogenic obesity have been instrumental in establishing the role of the hypothalamus, particularly the leptin melanocortin axis, as a master regulator of energy homeostasis. Studies of genetically engineered mice and spontaneously occurring disease in humans allowed researchers to delineate nuanced aspects of the pathways responsible and related pathophysiology. Critically, genetic evidence has pointed to variability in eating behaviour as being the major effector pathway between risk genes and obesity. That likely reflects the necessity of basic maintaining metabolic rate within strict limits to facilitate basic cellular and physiological processes; it is possible (even likely) that GWAS variants of small effect are more commonly associated with variants that cause minor metabolic adjustments.

Overall, the current GWAS variants explain <10% of the observed variability in BMI, so only a fraction of that variation attributable to genetics. Although this has led to the value of GWAS to provide biological insight being questioned, those studies have value because (1) they revealed the role of genes previously unsuspected as having a role in energy homeostasis (e.g. NEGR1, CADM2, ADCY3), (2) although the effect size of common variants is small, they may point to genes where pharmaceutical interventions might have clinically relevant impacts, (3) they may provide useful ways to stratify patient populations to target clinical interventions, (4) they provide evidence that that the critical processes governing energy homeostasis are neurological because of the nature and sites of expression of associated genes.

In conclusion, the heritability of obesity is high and studies in lab animals, production animals, companion animals and humans point to neural regulation of appetite as being key, via moderating the susceptibility of an individual to an obesogenic environment. There is much to be discovered about the genes responsible and their mechanism of action. Studies in breeds of dog with high obesity risk have shown that companion animal studies not only have value for veterinary scientists, but those with a wider or human interest.

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