# Chapter 2

# Nutritional Models of Type 2 Diabetes Mellitus

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## Summary

In order to better understand the events which precede and precipitate the onset of type 2 diabetes (T2DM) several nutritional animal models have been developed. These models are generated by manipulating the diet of either the animal itself or its mother during her pregnancy and, in comparison to traditional genetic and knock out models, have the advantage that they more accurately reflect the aetiology of human T2DM. This chapter will discuss some of the most widely used nutritional models of T2DM: Diet-induced obesity (DIO) in adult rodents, and studies of prenatal and postnatal nutrition in offspring of mothers fed a low-protein diet or overnourished during pregnancy. Several common mechanisms have been identified through which these nutritional manipulations can lead to metabolic disease, including pancreatic beta-cell dysfunction, impaired insulin signalling in skeletal muscle and the excess accumulation of visceral adipose tissue and consequent deposition of non-esterified fatty acids in peripheral tissues resulting in peripheral insulin resistance. The following chapter will discuss each of these nutritional models, their application and relationship to human aetiology, and will highlight the important insights these models have provided into the pathogenesis of T2DM.

**Key words:** Type 2 diabetes, Obesity, Insulin resistance, Animal models, Nutrition, High-fat diet, Programming

## 1. Introduction

Whilst the human is undoubtedly the model of choice when studying the pathophysiology of human disease, the study of the underlying mechanisms of disease in living humans has a number of logistical and ethical limitations. Therefore, to better understand the events which precede and precipitate the onset of type 2diabetes (T2DM) there is a need to develop in vivo animal models of this disease. The commonly used genetic models of T2DM, including (*ob/ob* and *db/db* mice and Zucker *fa/fa* rats), have been useful in understanding some of the mechanisms which

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may contribute to altered glucose and insulin metabolism; however, none of these is an ideal disease model, since these gene mutations are extremely rare in human populations *(1)*. Similarly, experimental animal models of T2DM induced by chemical destruction or removal of a portion of the pancreas *(2)* are not representative of the aetiology of T2DM in humans, which, is typically preceded by obesity *(3, 4)*. This has led to the development of several nutritional animal models, by manipulating the diet of either the animal itself, or its mother during her pregnancy. The following chapter will present an overview of the most widely utilised nutritional models, their application and relationship to human aetiology, including some of the most important insights into the pathogenesis of T2DM which have been provided by each. The limitations of each model will also be discussed in each case.

## 2. Diet-Induced Obesity

A significant proportion of T2DM in human populations is linked to an excessive accumulation of body fat, particularly in the abdominal region *(4–6)*. This fact makes the diet-induced obesity model (DIO) particularly relevant for studying the underlying mechanisms through which an excessive accumulation of body fat and/or an excessive dietary fat intake can lead to the development of insulin resistance and T2DM. The two most widely used models for the study of obesity-induced T2DM are high-fat feeding in rodents and the sand rat (*Psammomys obesus*).

### *2.1. Diet-Induced Obesity: Rats and Mice*

The DIO rodent model involves a regimen in which healthy, non-obese mice or rats are provided with ad libitum access to a highly palatable high-fat, high-energy diet. C57BL/6J mice were originally selected for this model because in early studies these mice developed clear-cut diabetes more rapidly than other strains fed the same high-fat diet, suggesting that this mouse had a genetic predisposition to T2DM *(7)*. Typically, C57BL/6J (B6) males are maintained on these diets for 8 to 12 weeks and, as a result, become obese, mildly to moderately hyperglycaemic and develop impaired glucose tolerance *(7)*. Similarly, Wistar and Sprague Dawley rats have been the model of choice for studying the metabolic effects of diet-induced obesity in the rat *(1)*.

The use of high-energy diets to induce obesity in laboratory animals for the purpose of studying obesity-related disorders has been widely adopted, and the scope of these diets is now quite extensive, ranging from an oil-based diet to free access to a variety of human junk foods, including pies, cakes and chocolate *(8–10)*.

Despite this variation in the amount and source of dietary fat, all high-fat diets are associated with the emergence of hyperphagia, rapid weight gain and excess body fat accumulation *(8–10)*. Irrespective of the precise nature of the diet, diet-induced obesity results in many of the same changes as seen in human obesity, including the development of central and peripheral leptin and insulin resistance *(8)*, and in the altered expression of adipokines which are known to contribute to the regulation of peripheral insulin sensitivity, in particular adiponectin and resistin *(11)*.

DIO rodent models have been widely utilised in order to investigate the defects within specific tissues which may contribute to the development of insulin resistance in obese individuals. By comparing the profile of gene expression and protein content in specific tissues of DIO mice with those from lean controls using micro-array or 2D gel electrophoresis, it is possible to very quickly identify genes which may be involved in the development of T2DM. Using this approach, DIO was shown to be associated with a reduced pancreatic abundance of enzymes involved in clearance of reactive oxygen species, providing a potential mechanism for the deterioration of pancreatic function seen in the later stages of T2DM *(12)*, with adipocyte hypertrophy *(13)* and the induction of hepatic steatosis and hepatic insulin resistance *(14)*, which again resembles the human obese state **(Fig. 1)**.

It is now widely accepted that the susceptibility to developing obesity and T2DM varies between individuals, and there is considerable interest in understanding the genetic and physiological basis of this difference. This question has been addressed using an adaptation of the DIO model in which adult Sprague Dawley rats are fed a purified diet with a moderately high fat content. The rats are subsequently selected as either obesity prone or obesity resistant according to their response (i.e. degree of weight gain and increase in percentage body fat) on the high-energy diet *(15)*. The obesity-prone rats typically eat approximately 16% more calories over the first 30 days compared to the obesity-resistant strain and exhibit a phenotype comparable to human metabolic syndrome, including glucose intolerance, hyperinsulinaemia and T2DM *(15, 16)*. Selective breeding has enabled these researchers to generate obesity-resistant and obesity-prone strains, and to study these two populations in order to determine factors, which may underlie an increased propensity to obesity *(17, 18)*. This is a useful model, in that it provides a means of distinguishing between the effects due to high dietary fat content as distinct from those related to effects of excess body fat.

The major advantage of the DIO model lies in the fact that these rats share many of the same characteristics as humans with obesity-related diabetes ('diabesity'). These therefore provide a model for measuring the cellular events through which excess accumulation of body fat and/or excessive dietary fat intake

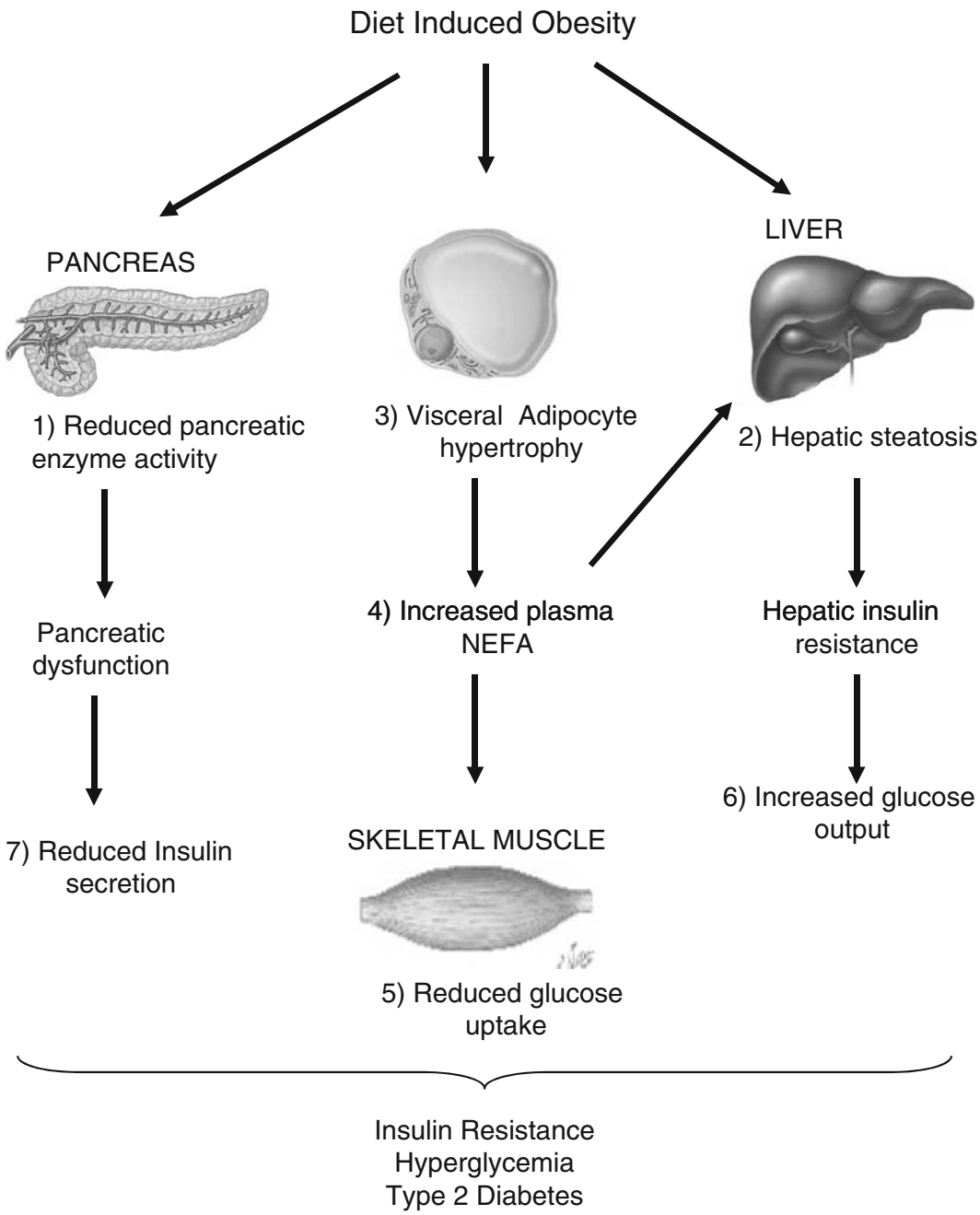


Fig. 1. Summary of proposed mechanisms involved in the development of type 2 diabetes in models of diet-induced obesity in adult rodents. Increased dietary fat intake is associated with (1) reduced activity of pancreatic enzymes, leading to impaired pancreatic function and reduced insulin secretion, and (2) increased accumulation of lipids in the liver (hepatic steatosis) which results in hepatic insulin resistance and increased hepatic glucose output. In addition, increased accumulation of adipose tissue in the visceral compartment (3) results in increased plasma concentrations of non-esterified free fatty acids (NEFAs) (4), and deposition of these NEFAs in liver and skeletal muscle, further contributing to insulin resistance in these tissues. Together, this results in an impaired glucose uptake by skeletal muscle (5), increased hepatic glucose output (6) and impaired insulin secretion (7) and precipitates the development of whole-body insulin resistance and type 2 diabetes.

result in the degradation of insulin action in key insulin-sensitive tissues. Another advantage of this model is that DIO mice and rats can now be ordered directly from commercial suppliers. Whilst this is obviously more expensive than an in-house option, it does have the potential to save the researcher the 8-12 weeks that it would normally take to generate the DIO rodents. In addition, a variety of high-fat diets are now available from commercial animal feed manufacturers.

Whilst the DIO model has many positive attributes, it also has some potential limitations. Perhaps the major limitation is the lack of standardisation of the feeding regimen of different studies, which has meant that the phenotype of fat-fed animals varies between experiments. The high-fat diets which have been used consist of anywhere from 20 to 60% energy content from fat, and the type of fat (i.e. animal versus plant) also varies between studies *(1)*, and there is no clear indication of which type of high-fat feeding represents the best model of the metabolic disturbances seen in human obesity. It is therefore important to consider both the type of dietary intervention (i.e. the proportion of energy derived from fat and type of fat in the diet) and the precise phenotype of the strain under investigation when interpreting results and extrapolating these data to humans.

### 2.2. Diet-Induced Obesity: Psammomys obesus

The sand rat (*Psammomys obesus*) has been another popular model for studying the degradation of metabolic function associated with obesity. This is an attractive model because the sand rat under laboratory conditions rapidly develops obesity when fed a standard laboratory chow, which is considerably less expensive than custom-made high-fat diets. Many of the pathophysiological changes found in the obese sand rat are similar to those seen in human type 2 diabetic patients *(19)*, and this model therefore appears to provide an appropriate nutritional model of human T2DM. When provided with ad libitum access to laboratory chow the adult sand rat progresses through from normoglycaemia and normoinsulinaemia, to hyperinsulinaemia with marked insulin resistance, followed by pronounced hyperinsulinaemia with hyperglycaemia and, after a further 6-10 weeks of chow feeding, gradual beta-cell degradation and disappearance of beta-cell insulin which eventually results in severe insulin deficiency and overt diabetes *(20)*. The obese sand rat exhibits profound insulin resistance in both skeletal muscle and liver, and provides an attractive model for studying the mechanisms, which underlie the development of diabetes in human obesity *(21)*. Studies utilising this model have reported that GLUT4 protein content is reduced in skeletal muscle, resulting in a reduced peripheral glucose uptake and hyperglycaemia *(20)*. In addition, hepatic PEPCK activity is increased, suggesting that the ability of insulin to inhibit hepatic glucose production is impaired *(22)*. Extensive use has been made of the obese sand rats for the purpose of testing potential T2DM

drugs, including tyrosine phosphatase inhibitors and glucagon-like peptide-1 (GLP-1) analogues *(20)*.

The major limitations of this model are that the sand rat exhibits insulin resistance even when fed on a low-energy diet, suggesting that the physiology of this desert-adapted animal may not be entirely comparable with humans. The link between weight gain and the development of insulin resistance in this model has yet to be clearly defined; however, the phenotype is normalised by dietary restriction. Nevertheless, this model has been utilised extensively for the study of obesity-induced diabetes and continues to provide important insights into the cellular defects, which contribute to the development of central and peripheral insulin resistance **(Fig. 1)**.

## 3. Prenatal Nutritional Models of T2DM

The foetal or developmental origins of adult disease hypothesis states that insults during critical windows of development results in adaptive changes within foetal tissues and organ systems, which have lifetime consequences for the health of an individual. The hypothesis was first derived from studies of the Hertfordshire birth cohort by Professor David Barker in the early 1990s which showed that there was an inverse relationship between birth weight and the incidence of cardiovascular disease in adult life *(23)*. This was followed by a series of studies in human populations which all demonstrated that being of low birth weight was associated with an increased incidence of adult metabolic and cardiovascular disease *(24, 25)*.

The Dutch Winter Hunger Famine was a 5-month period in World War II during which the food supply to Amsterdam, Holland, was severely restricted, resulting in a substantial decrease in the daily energy intake of the population *(26)*. Subsequent studies of the children of women who were pregnant during the famine showed that those exposed to famine in the last 5 months of gestation had a significantly greater incidence of glucose intolerance, obesity, and T2DM in adult life compared to those children whose mothers were not exposed to the famine *(26, 27)*. Similarly, early studies in infants of diabetic mothers clearly showed that exposure to high glucose levels in the pre- and perinatal period was associated with an increased incidence of hyperglycaemia and T2DM in the offspring in postnatal life *(28)*. These studies provided the first evidence that exposure to an inappropriately low, or inappropriately high, nutrient supply during early development was associated with a permanent alteration of metabolic function in the offspring. An increasing number of

epidemiological and experimental animal studies have since continued to highlight the importance of both the prenatal and early postnatal nutritional environment for the determination of later metabolic health *(25, 29, 30)*.

Together, these studies have clearly demonstrated that a low birth weight followed by a period of accelerated postnatal growth or a high birth weight, as a consequence of prenatal overnutrition, are each associated with an increased propensity towards the development of insulin resistance, glucose intolerance and T2DM in adult life *(31–33)*. As a result, both spontaneous and experimentally induced foetal growth restriction (in utero growth restriction (IUGR)) and prenatal overnutrition in animal models have been widely employed in order to understand the physiological basis of reduced insulin sensitivity. It has been demonstrated that global undernutrition in the pregnant rodent is also associated with later onset of T2DM in the offspring *(34)*. This is, however, considered to be largely a consequence of impaired insulin secretion rather than defects in insulin signalling *(35)*, and is therefore not the most appropriate model for the pathophysiological process which contribute to T2DM in the majority of human patients.

### *3.1. The Maternal Low-Protein Model*

The maternal low-protein model is one of the most extensively studied models of foetal growth restriction. In this model, pregnant dams are fed a diet containing approximately 8% of energy as protein, compared to approximately 20% in controls, and this is associated with low birth weight in the offspring, followed by accelerated postnatal growth *(29, 36)*. The period of rapid postnatal growth is due to enhanced insulin sensitivity in the period immediately after birth; however, this is not maintained beyond the early postnatal period, and the offspring develop insulin resistance by the age of 15 months and frank T2DM by 17 months *(33, 37)*, and the deterioration of metabolic function is accelerated by a postnatal high-fat diet *(38)*. The phenotype of the low-protein offspring has many similarities with that of human type 2 diabetics, including insulin resistance, altered regulation of hepatic glucose output and pancreatic dysfunction *(29)*. As a result, this model has been used extensively to explore and characterise the cellular defects within the insulin signalling pathway which may contribute to reduced insulin sensitivity. These studies have demonstrated that prenatal exposure to a low-protein diet results in defects in several peripheral insulin-sensitive tissues, which are central to the maintenance of glucose homeostasis. In the pancreas, the islets of Langerhans are smaller and exhibit a reduced insulin secretion in response to amino acid stimulation *(39)*. Whilst there is no obvious defect in glucose-stimulated insulin release in offspring fed a control chow diet post-weaning, a reduction in the capacity of glucose to stimulate insulin release emerges if these offspring are

fed on a high-fat diet post-weaning. The liver of the low-protein offspring is unresponsive to the actions of glucagon, and insulin stimulates, rather than suppresses, hepatic glucose output *(40)*. Moreover, adipose cells exhibit increased basal and insulin-stimulated glucose uptake, leading to an increased accumulation of fat in the visceral compartment and ectopic fat storage in liver and skeletal muscle, which further contributes to the peripheral insulin resistance *(41)* (**Fig. 2**).

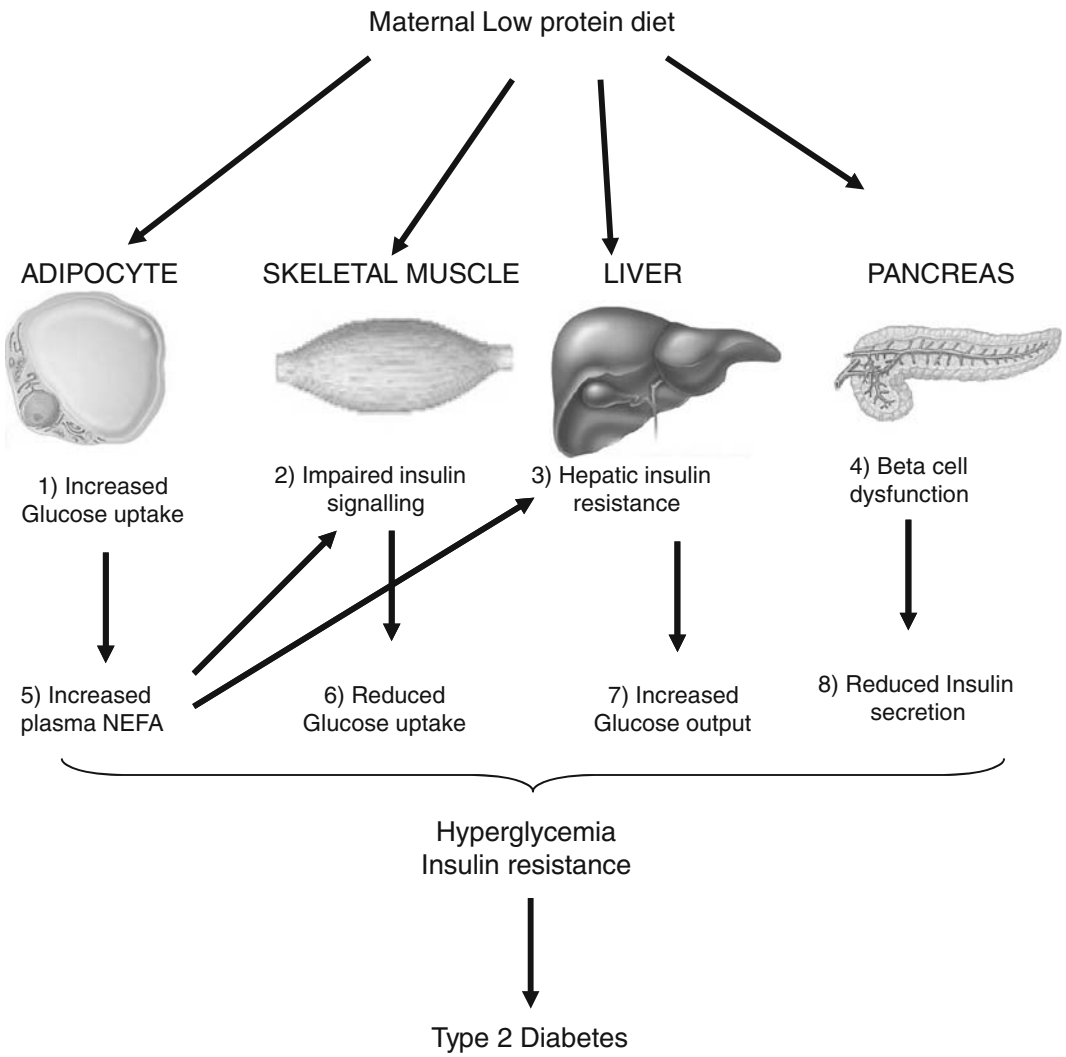


Fig. 2. Summary of mechanisms proposed to contribute to the programming of type 2 diabetes of offspring in the maternal low-protein model. Prenatal exposure to a low-protein diet results in (1) an increased capacity for glucose uptake by visceral adipocytes (2) impaired insulin signalling in skeletal muscle, (3) hepatic insulin resistance and (4) impaired pancreatic function. The increased glucose uptake by visceral adipocytes results in an increased accumulation of visceral adipose tissue, resulting in elevated plasma concentrations of non-esterified free fatty acids (NEFAs) (5) which are deposited in peripheral tissues (liver and skeletal muscle), further reducing insulin sensitivity. This reduced insulin sensitivity is associated with impaired glucose uptake by skeletal muscle (6), increased glucose output by the liver (7) and reduced insulin secretion by the pancreas (8), resulting in peripheral hyperglycaemia, insulin resistance and type 2 diabetes.

Given that muscle represents the major site of postprandial glucose disposal, it is not surprising that changes in the functional characteristics of muscle fibres during the perinatal period are important in the programming of insulin resistance and diabetes (**Fig. 2**). Isolated muscle strips from these low-protein animals exhibit enhanced basal and insulin-stimulated glucose uptake, and this increase in insulin sensitivity is associated with a twofold increase in the abundance of insulin receptors in muscle membranes *(42)*. By 15 months of age, however, there is a decrease in the insulin sensitivity of glucose uptake in skeletal muscle from the group exposed to the low-protein diet in utero *(43)*. This impaired insulin action is not associated with changes in the expression of either the insulin receptor or GLUT4, but is associated with a decrease in the abundance of signalling molecules downstream of the insulin receptor, including the zeta-isoform of protein kinase C, an isoform that is involved in GLUT4-mediated glucose transport *(43)* and p85 PI3K *(44)* (**Fig.** 2). In addition to defects in insulin signalling, it has also been reported that prenatal undernutrition is associated with impaired mitochondrial biogenesis and impaired mitochondrial oxidative capacity in skeletal muscle, which has previously been associated with reductions in insulin sensitivity in this tissue *(45, 46)*.

Whilst these studies have provided important insights into the cellular defects which contribute to deteriorations in insulin sensitivity in peripheral tissues, it is still unclear whether these defects are the same as those seen in human T2DM, which typically develops secondary to increased body fat accumulation. There is recent evidence that the mechanisms involved in the prenatal programming of insulin resistance may be quite distinct from those in insulin resistance which develops in response to diet-induced obesity *(35)*. Nevertheless, the obvious importance of the early environment in determining metabolic health in later life may well explain why it is that some individuals are more susceptible to insulin resistance than others at the same degree of body fatness.

### *3.2. Prenatal Overnutrition, Postnatal Obesity and T2DM*

There is increasing evidence that prenatal exposure to a high plane of nutrition is also associated with an increased risk of obesity and T2DM in postnatal life. The rat and the sheep represent the two main animal models which have been utilised thus far for investigating the effects of prenatal overnutrition on the offspring. Unlike the low-maternal low-protein model, however, the role of defects within the insulin signalling pathway in models of maternal overnutrition or high-fat feeding remains largely unexplored. Studies to date have suggested that the deterioration of metabolic function in these individuals is associated with altered development of pancreatic beta-cells, mitochondrial dysfunction

and programming of the central appetite-regulating network *(10, 47, 48)*. Several studies have also suggested that the deterioration of metabolic function may be secondary to the increased accumulation of adipose tissue in these offspring **(Fig. 3)** *(48, 49)*; however, it is not clear whether this is the case in all models of prenatal overnutrition.

In the rat, maternal high-fat feeding results in disturbed glucose homeostasis in the offspring at weaning and in adult life, including an impaired glucose tolerance and reduced whole-body insulin sensitivity. Patel and colleagues found that feeding rats on a high-fat chow during pregnancy and lactation resulted in offspring that were significantly heavier than controls, had elevated plasma concentrations of insulin, glucose, free fatty acids and triglycerides and exhibited glucose intolerance *(47)*. When they investigated the pancreatic islets in these animals, they found that those from male offspring exhibited an increased insulin secretary response at low glucose concentrations compared to controls, suggesting that high-fat feeding resulted in altered pancreatic development *(50)*. This was also the case in a separate study, in which feeding pregnant rats a high-fat diet during pregnancy and lactation was associated with hyperglycaemia and evidence of compromised beta-cell function in the weanling offspring *(51)*. Similarly, the offspring of Sprague-Dawley rats fed a lard-rich diet during pregnancy and lactation exhibited whole-body insulin resistance and a reduced glucose-stimulated insulin secretion in isolated islets at 9 months *(10)*, again supporting the thesis that maternal high-fat feeding is associated with impaired pancreatic function in the offspring **(Fig. 3)**.

In the sheep, feeding ewes approximately 55% above their maintenance energy requirements in late pregnancy results in an increase in foetal glucose and insulin concentrations in the last third of gestation *(52)*, and is associated with an increased accumulation of subcutaneous adipose tissue at the end of the first month of postnatal life *(52)*. The offspring of overfed ewes exhibit elevated glucose concentrations during the first month of postnatal life *(52)*, although whether this is associated with reduced insulin sensitivity during this period has yet to be determined.

Fig. 3. (continued) The increased accumulation of body fat contributes to metabolic dysfunction by elevating plasma non-esterified free fatty acid (NEFA) concentrations (4) which are deposited in skeletal muscle and liver, resulting in insulin resistance in these tissues (5) and (6). Recent evidence suggests that epigenetic changes which occur before birth (7) may also play a role in metabolic programming after prenatal overnutrition. Together, these changes result in peripheral insulin resistance and, ultimately, type 2 diabetes.

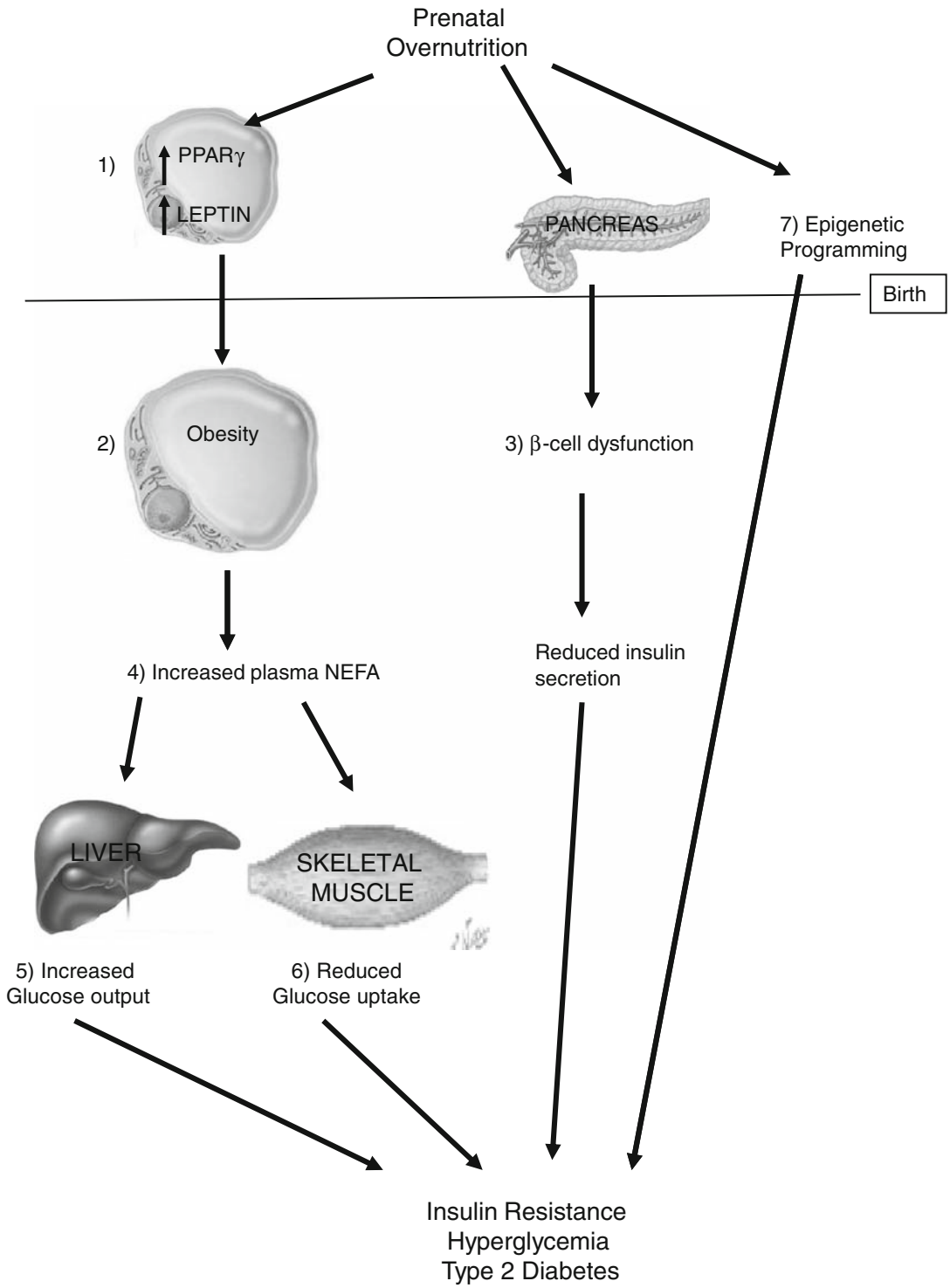


Fig. 3. Potential mechanisms contributing to the programming of type 2 diabetes following prenatal overnutrition. It has been demonstrated that prenatal overnutrition is associated with an increased lipogenic capacity of adipose cells (1), which results in increased fat accumulation after birth (2) and predisposes these offspring to obesity in later life. In addition, prenatal high-fat diets result in impaired pancreatic development and beta-cell dysfunction in postnatal life (3).

## 4. Possible Mechanisms for Metabolic Programming

### *4.1. The Adipocyte*

In large animal models there is evidence that exposure to prenatal overnutrition or undernutrition can act to permanently alter the function of adipocytes and result in an increased lipogenic capacity in adipose depots in postnatal life. In sheep, which have a similar profile of adipose cell development to humans, exposure to increased glucose concentrations in late foetal life increases the expression of genes within adipose cells that are responsible for promoting lipid storage and forming new adipocytes *(53)*. This is associated with increased adipose tissue mass by the end of the first month of life, due primarily to an increase in adipocyte cell size *(48)*. In pigs, adipocytes exposed to high glucose levels before birth also exhibit a dramatic increase in their capacity for lipogenesis *(54)* and this precedes their development of obesity *(55)*. These findings have since been supported by work in rodents, in which maternal junk food feeding during pregnancy and lactation in rats and an obesogenic diet during this same period in mice were each associated with increased adiposity and increased expression of lipogenic genes and insulin-independent glucose transporters in the perirenal adipose depot *(49, 56)*. It would therefore appear that fat cells exposed to an excess substrate supply during critical windows in their development have an increased capacity for storing lipid in postnatal life. This enhanced lipogeneic capacity would render these individuals more likely to store excess energy in the form of fat, and increase their susceptibility to weight gain, obesity and, consequently, to the excess deposition of fatty acids in liver and skeletal muscle, resulting in peripheral insulin resistance, hyperglycaemia and, ultimately, type 2 diabetes **(Fig. 3)**.

In individuals exposed to low nutrient levels before birth, adipocyte development is initially sacrificed in favour of essential organs *(25, 57)*. If an in utero 'restricted' individual is born into a postnatal environment where nutrient supply is no longer constrained, a period of 'catch-up' fat deposition ensues, mainly in the visceral adipose depot *(58)*. These individuals are therefore at increased risk of visceral obesity *(59)* and, consequently, to the development of insulin resistance and T2DM *(60)*. It is the increased accumulation of visceral adipose tissue that has also been demonstrated in animal models of IUGR, including the sheep, guinea pig and rodent *(61–63)*. Therefore, exposure of the developing adipocyte to sub-optimal nutrition, particularly of the visceral adipose tissue which is the first adipose depot to develop in sheep and humans *(64, 65)*, also appears to play a critical role in defining an individual's propensity for accumulating

visceral body fat later in life, i.e. the fat pattern linked to increased risk of metabolic dysfunction *(6)*.

### 4.2. Mitochondrial Biogenesis

Mitochondria play a central role in the regulation of cellular energy metabolism, and impaired mitochondrial function and reduction in mitochondrial oxidative capacity in skeletal muscle have been associated with the onset of insulin resistance and T2DM both in experimental animal models and in human subjects *(66)*. In experimental animal studies, both diet-induced obesity and prenatal undernutrition have been associated with impaired mitochondrial biogenesis and reduced abundance of oxidative enzymes in skeletal muscle. Importantly, there is evidence that the reduction of mitochondrial biogenesis in adult rats exposed to prenatal undernutrition precedes the development of insulin resistance and glucose intolerance in this model *(45, 46)*, therefore implicating impaired mitochondrial function in the causal pathway linking prenatal undernutrition to later metabolic disease. The role of mitochondrial dysfunction in metabolic programming has yet to be explored in large animal models, and the potential role of mitochondrial dysfunction in the development of T2DM remains an important area for investigation.

### 4.3. Epigenetics

The concept that changes in phenotype could be elicited by modification of the DNA in the absence of changes in DNA sequence, i.e. epigenetic changes, provides a new basis for understanding of phenotypic programming. The two most well-characterised epigenetic modifications include both DNA methylation and histone acetylation, which act to suppress gene expression *(67, 68)*. Recent evidence has demonstrated that exposure to an either an excession or deficient nutrient supply during early development may result in epigenetic modifications in the foetus which have the potential to permanently alter the function of important metabolic systems. Waterland and colleagues demonstrated that when mice with a genetic tendency towards obesity were fed a standard diet during pregnancy, the degree of obesity in the offspring increased progressively with each generation. This effect was, however, completely abolished if mice were fed a diet high in methyl groups (which increases DNA methylation) during pregnancy, and the offspring remained lean. These findings suggested that reduced DNA methylation in genes which play a role in the regulation of energy balance and fat storage, as a result of early nutritional programming, may contribute to the later development of obesity and metabolic disease *(69)*. There is also increasing evidence that exposure to a sub-optimal nutrient supply before birth can also alter methylation of a series genes involved in growth and energy homeostasis, and that this may explain the link between foetal growth restriction and later metabolic dysfunction *(67, 68)*.

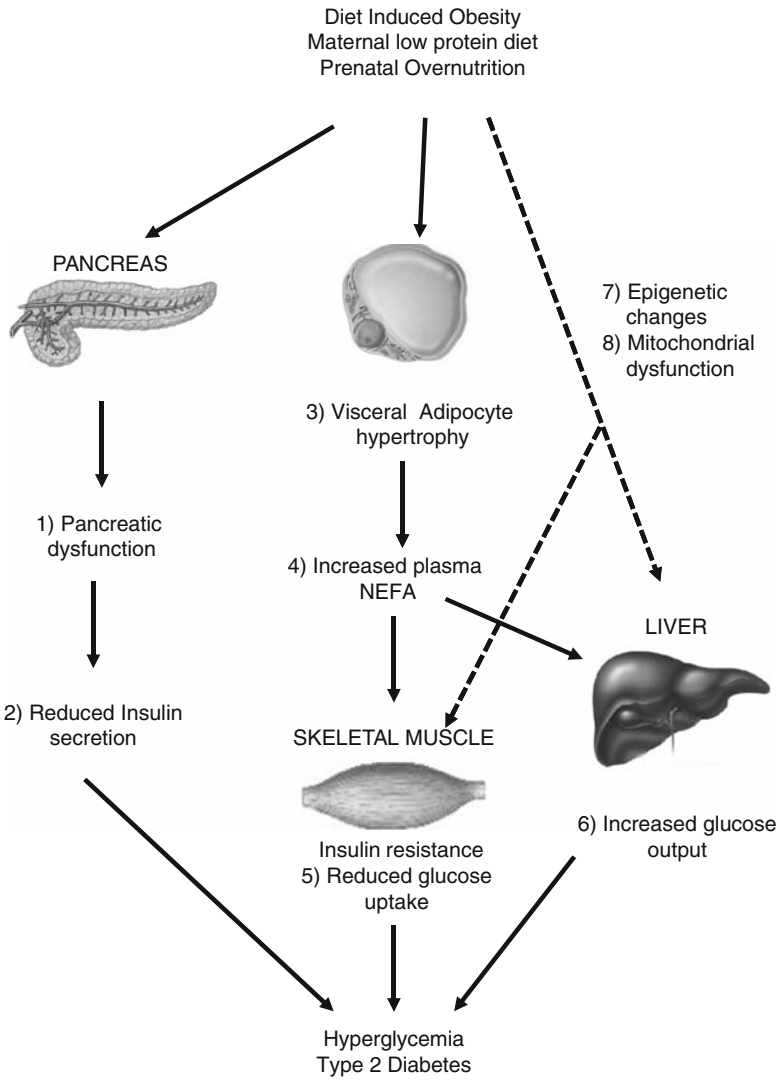


Fig. 4. Summary of common mechanisms identified in all nutritional models of type 2 diabetes. Both diet-induced obesity and prenatal nutritional interventions are associated with pancreatic dysfunction (1) and impaired insulin secretion (2), which contributes to peripheral hyperglycaemia. In addition, adipocyte hypertrophy, in particular within the visceral compartment (3), is associated with increased circulating concentrations of non-esterified free fatty acids (NEFA) (4) which, when deposited in liver and skeletal muscle, are associated with the development of insulin resistance in these tissues and, consequently, reduced glucose uptake by skeletal muscle (5) and increased hepatic glucose output (6). In addition, recent evidence suggests that epigenetic changes and mitochondrial dysfunction in liver and skeletal muscle may also contribute to development of metabolic disorders.

## 5. Summary

The dramatic increase in the incidence of T2DM over the past decade has highlighted the need to better understand the pathophysiology of this disease. The use of animal models which mimic

the disease progression in humans is essential in order to investigate the cellular mechanisms which contribute to the deterioration of insulin metabolism in key insulin-sensitive tissues. This criterion is not fulfilled by genetic models, or models in which the pancreas is surgically ablated, since these causes account for only a relatively minor proportion of the humans who will develop T2DM. A number of nutritional models of T2DM have now been developed, and have made a significant contribution to our current understanding of the cellular defects which emerge in insulin-sensitive tissues in response to an excess accumulation of body fat or excessive intake of dietary fat, and which result in the development of peripheral insulin resistance. It has also become clear that exposure to either inappropriately high or inappropriately low levels of nutrition *in utero* is associated with an increased susceptibility to T2DM in adult life, and that the detailed study of the cellular and molecular defects in these offspring has the potential to provide novel insights into why some individuals are more prone to the development of insulin resistance and T2DM than others. The study of diet-induced obesity and models of prenatal undernutrition and overnutrition has revealed several common mechanisms which contribute to the onset of insulin resistance and T2DM in these different models **(Fig. 4)**, and this has provided important insights into the aetiology of type 2 diabetes in humans and has identified possible targets for intervention. As with all animal models, it is important to recognise that animal data may not always be directly extrapolated to the human situation, where other factors, in particular socioeconomic status and demographic variations, also contribute to risk of metabolic disease *(70)*. It is therefore important to exercise caution when extrapolating results to human clinical practice. Nevertheless, it is clear that nutritional studies in the animal models discussed in this chapter have been critical in shaping our current understanding of the pathophysiology of T2DM and offer significant opportunities for identifying and testing potential clinical interventions.

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