Understanding DNA Methylation As An Epigenetic Factor In Type 2 Diabetes

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Abstract

Type 2 diabetes (T2D) is a complex metabolic condition that is influenced not only by genetics and lifestyle but also by DNA methylation, an epigenetic factor. This review investigates how irregular methylation patterns in pancreatic islets, adipose tissue, liver, and skeletal muscle can disrupt insulin signaling, impair glucose uptake, and alter metabolic pathways, which all have contributed significantly to the development of T2D. Environmental and lifestyle factors, including diet, physical activity, stress, and early-life exposures, are also explored, all of which can affect DNA methylation patterns. Many alterations in DNA methylation are reversible, creating potential for interventions such as healthy diets and targeted therapies to reverse these changes in the body. This review evaluates the current evidence on the role of DNA methylation in the pathogenesis of T2D, how it interacts with environmental factors, and the therapeutic potential of targeting these epigenetic modifications. There are many emerging therapeutic options, including nutritional epigenomics, pharmacological agents (e.g., metformin, hydralazine), and natural compounds (e.g., curcumin); these show promise in modifying methylation to prevent or manage disease. There are still challenges in determining causality, addressing tissue-specific variability, and identifying universal biomarkers. We think longitudinal studies tracking individuals from their early life, alongside personalized approaches based on individual epigenetic profiles, would be the most effective next steps in reducing T2D risk through methylation-based strategies.

Keywords

Biology; Epigenetics; DNA Methylation; Type 2 Diabetes; Insulin Resistance

Introduction

Type 2 diabetes (T2D) is a chronic disease that is currently affecting over 800 million people globally. T2D is one of the most common metabolic conditions, affecting approximately 1 in 9 adults.[1] T2D is recognised by many characteristics such as impaired insulin production, insulin resistance, and an inability to regulate blood glucose properly. Although positive lifestyle changes can help to prevent or even delay T2D, and help achieve remission, there is still no

definitive cure for T2D. Sustained progression of T2D can lead to complications and further issues arising, including retinopathy, neuropathy, and cardiovascular dysfunction.[2]

Aside from its physical impacts, disrupted cellular energy management contributes to metabolic dysfunction in T2D. Gene regulation is essential for maintaining metabolic balance by controlling enzyme production and nutrient utilization. If disruptions occur in these regulatory mechanisms, they can mark the onset of metabolic dysfunction and the onset of T2D.[3] Increasing evidence suggests that epigenetics, the heritable changes in gene activity that occur without altering the DNA sequence, is a critical factor in the development of T2D. Among the various epigenetic mechanisms, DNA methylation has emerged as a key factor.[4] Understanding how DNA methylation contributes to the development and progression of T2D may lead to the development of more personalized treatment plans or even targeted therapies. When methyl groups are added to DNA, it can effectively turn genes “on” or “off,” which influences how cells respond to metabolic stressors. Irregular DNA methylation patterns have been linked to insulin resistance and β-cell dysfunction, two key characteristics of T2D.[5,3,6]

This review will examine the role of DNA methylation in the development and progression of T2D. There will be a strong emphasis placed on tissue-specific methylation patterns, their contribution to insulin resistance, and the impact of lifestyle and environmental factors in shaping these epigenetic landscapes. This review will also discuss how insights into DNA methylation may guide personalized treatment strategies and highlight new treatments that aim to improve or reverse Type 2 Diabetes by changing gene activity.

DNA Methylation

Epigenetic changes are inheritable and also reversible; it is important to understand that these are changes in gene activity that do not alter the underlying DNA sequence. These changes can be passed down through cell divisions and sometimes across generations, yet they can be reversed, which allows genes to be turned on or off in response to environmental and developmental factors.[1] This reversible nature of epigenetic modifications, such as DNA methylation, plays a crucial role in regulating gene expression and cellular function. DNA methylation, an example of an epigenetic process, involves the addition of a methyl group (CH_3), a chemical tag, to cytosine bases, which are often followed by guanine (CpG dinucleotides). In DNA methylation, a methyl group is added to cytosine, a nitrogenous base in DNA; methylation occurs at the 5th carbon of its ring. DNA methyltransferases (DNMTs) are enzymes that add a methyl group to cytosine, converting it into 5-methylcytosine (5mC), a process that primarily occurs at CpG sites (cytosine followed by guanine) (Figure 1).

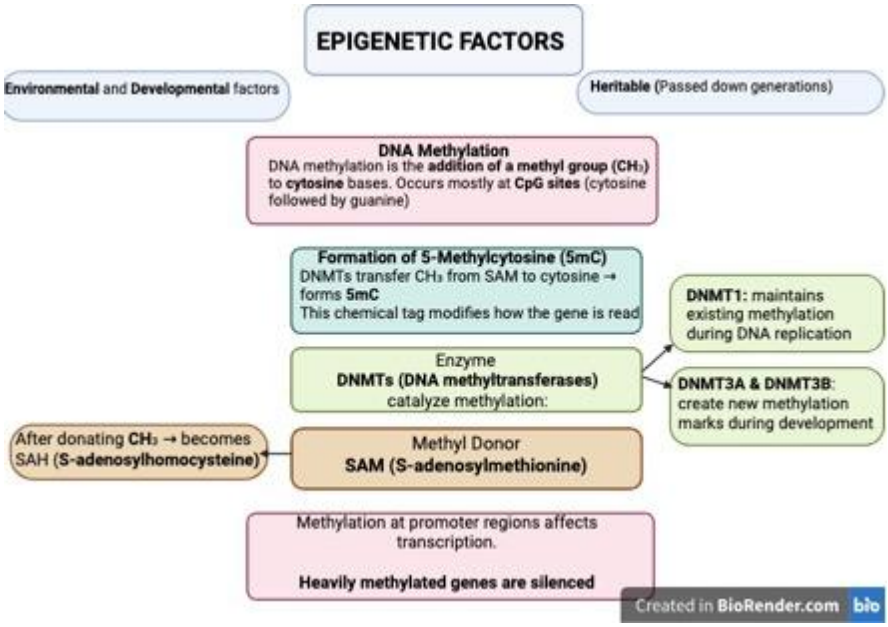

Figure 1. Process of DNA methylation. This figure was created in BioRender.com.

This process occurs by adding a methyl group to cytosine, which involves S-adenosylmethionine (SAM), a methyl donor used by DNMTs. After SAM donates its methyl group (CH_3), it becomes S-adenosylhomocysteine (SAH). S-adenosylmethionine is formed by the folate and methionine cycle. The gene *TXNIP* is consistently found to be hypomethylated in diabetic blood samples across multiple ethnicities, showing its importance in T2D pathophysiology.[7] Gene silencing happens when certain parts of DNA are switched off so they cannot be read or used. This process is carried out by DNA methyltransferases, including *DNMT1*, *DNMT3A*, and *DNMT3B*. *DNMT1* maintains existing methylation patterns during DNA replication, while *DNMT3A* and *DNMT3B* are responsible for creating new methylation marks during development. The presence of methyl groups in gene promoter regions can influence gene activity: when methyl groups are added to promoter regions, transcription factors cannot bind properly, making it hard for the cell to read the gene, so the gene becomes inactive. In addition, other proteins bind to the methylated DNA, helping pack it more tightly, which makes it even harder for the cell to access and use the gene. Conversely, if a promoter is not heavily methylated, the DNA remains open and readable, allowing transcription factors to bind and turn the gene “on” so it can be used when the cell needs it (Figure 2).

DNA methylation and histone modifications often affect whether chromatin is open or closed. Euchromatin refers to open chromatin, while heterochromatin refers to closed or condensed chromatin. Euchromatin is loosely packed, with active transcription in these regions and low DNA methylation levels, whereas heterochromatin is far more densely packed and transcription is silenced (Figure 2).[8] These heterochromatin regions are often found hypermethylated in the *INSR* promoter region and can worsen insulin resistance.

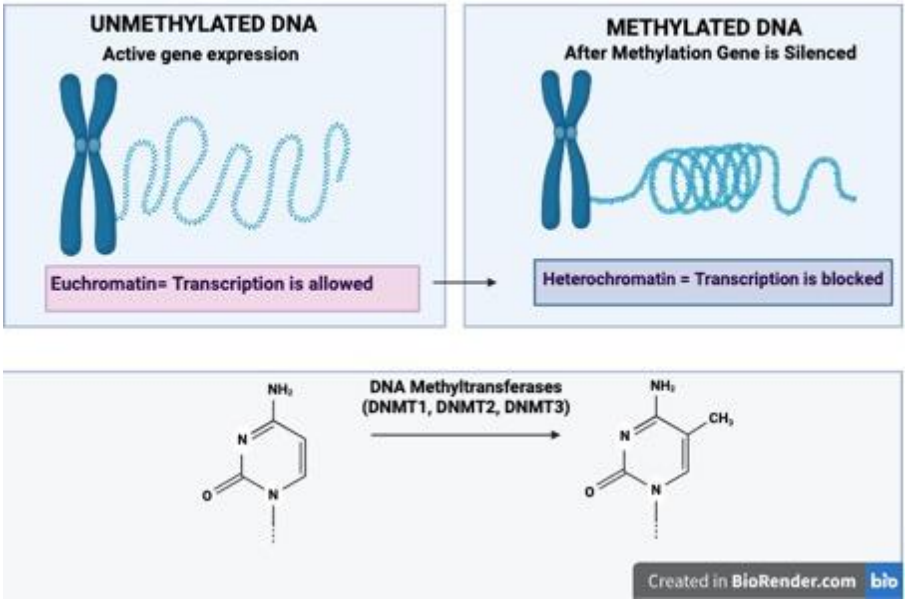

Figure 2. DNA methylation and gene expression regulation. This figure was created in BioRender.com.

DNA methylation is also affected by environmental factors, including diet, stress, and exercise, thereby altering gene expression without changing the DNA sequence. Multiple studies have suggested that changes in DNA methylation patterns can lead to altered gene expression.[7] Genome-wide association studies (GWAS) have identified altered DNA methylation patterns in pancreatic islets, adipose tissue, liver, and skeletal muscle in individuals with T2D compared to those without.[1]

Environmental factors are outside influences that people cannot control, such as air pollution, chemical contaminants, and early-life nutrition. These can influence DNA methylation patterns through consistent exposure over a long period of time.

In contrast, lifestyle factors involve an individual's personal choices and behaviours, including physical activity, diet, and stress management, which actively shape the epigenome and can either worsen or improve metabolic health. Environmental and lifestyle factors play a crucial role in triggering changes in DNA methylation that contribute to the development of T2D. Factors like obesity, unhealthy dietary habits, lack of physical exercise, and chronic stress are among the significant risk factors known to cause adverse methylation changes. These factors affect DNA methylation by increasing or decreasing the number of methyl groups at key gene regulatory regions, disrupting regular gene expression related to insulin signalling, glucose metabolism, and inflammatory pathways. For example, diets high in fat can alter methylation patterns, leading to dysregulated fat metabolism and increased inflammation, both key contributors to insulin resistance, a common trait of T2D.[1] Similarly, chronic stress can alter methylation and gene expression related to metabolic control.

In summary, environmental and lifestyle factors trigger dynamic changes in DNA methylation that affect gene expression involved in the development of insulin resistance and T2D. Early-life exposures may prime these epigenetic patterns, which can persist and increase risk later in life. However, these methylation changes are not permanent; they can be reversed through healthy lifestyle interventions that may reduce the risk of T2D and improve metabolic health.

DNA Methylation in Metabolic Tissues

Metabolic tissues, such as the pancreatic islets, liver, adipose tissue, and skeletal muscle, play crucial roles in type 2 diabetes by regulating and maintaining insulin production and glucose uptake. When metabolic dysfunction occurs in these tissues, whether it is impaired insulin secretion by pancreatic β-cells or excessive glucose production by the liver, it contributes directly to the development of insulin resistance and elevated blood sugar levels, both commonly associated with T2D (Table 1). Therefore, it is important to understand these tissue-specific changes for developing effective treatments (Figure 3).

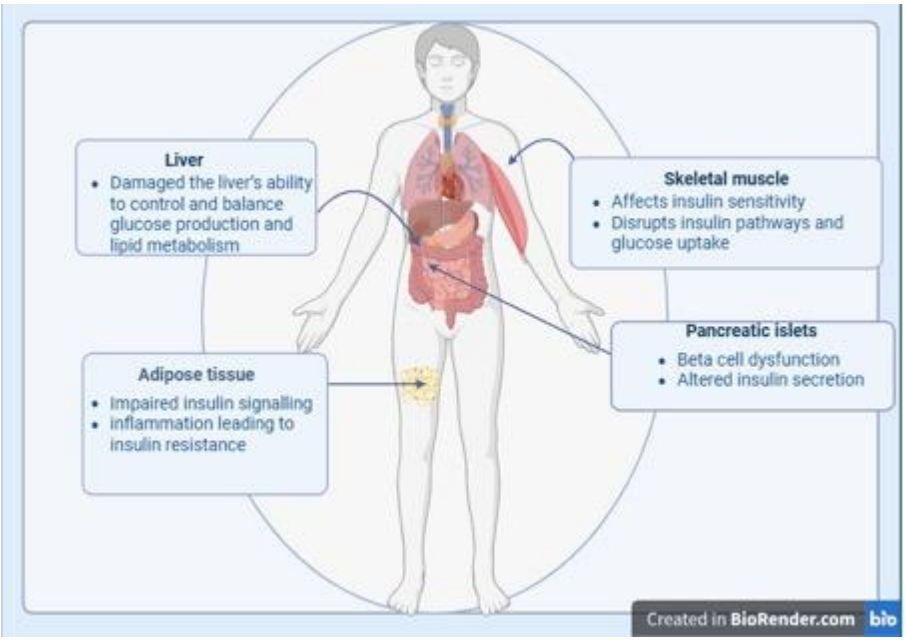

Figure 3. Organ-specific effects of metabolic dysfunction on T2D. This figure was created in BioRender.com.

Table 1. Tissue-specific genes affected by DNA methylation.

Tissue	Genes affected by DNA methylation	Functional impact	References
Pancreatic islets	*CDKN1A*, *PDE7B* and *SEPT9*	Impaired insulin and glucagon release due to β-cell dysfunction	3

Adipose tissue	***INSR* promoter, *FABP4*, *GPR210* and *LEP***	**Impaired insulin signalling, fatty acid transport, and hormone regulation**	**9**
Liver	***PPARGC1A*, *FGF21*, *GRB10*, *ABCC3*, *MOGAT1*, and *PRDM16***	**Key metabolic genes silenced, impairing glucose and lipid metabolism and worsening insulin resistance**	**10,1**
Skeletal muscle	***PDK4*, *INSR*, and *AKT2***	**Impaired insulin signalling and glucose uptake, reducing efficiency of glucose use**	**1**

The pancreatic islets play a crucial role in regulating metabolism and are key to the development of T2D. After eating a meal, insulin is released from pancreatic islet β-cells into circulation in response to increased blood glucose levels. The failure of β-cells can lead to impaired glucose tolerance, which results in T2D. Many studies have found that genes in diabetic islets that are excessively methylated are often associated with β-cell dysfunction. In T2D, genes related to the pancreatic islets, such as *CDKN1A*, *PDE7B*, and *SEPT9*, are influenced by changes in DNA methylation, disrupting the regular release of insulin and glucagon from these islets.[3]

Adipose tissue, also commonly known as body fat, is a type of connective tissue found throughout the body and plays a crucial role in regulating energy metabolism.[1] DNA methylation in adipose tissue has been recognised as a crucial factor that links obesity and insulin resistance, the most common features of T2D. DNA methylation helps regulate the behaviour of immune cells in adipose tissue, contributing to chronic inflammation that impairs insulin signalling. In several studies, methylation of the *INSR* promoter, which encodes the insulin receptor and facilitates cell response to insulin and glucose in fat tissue, has been linked to insulin resistance and systemic inflammation in T2D patients. Other genes affected directly by methylation include *FABP4*, where methylation negatively affects fatty acid transport, leading to altered lipid metabolism and contributing to metabolic dysfunction; *GPR210*, where methylation impairs anti-inflammatory fatty acid signalling and further contributes to insulin resistance; and *LEP*, the gene encoding leptin, where methylation alters leptin secretion and, in the long term, contributes to obesity-related insulin resistance.[9] Specifically, irregularly increased methylation in gluteal subcutaneous adipose tissue also correlates with greater insulin resistance, suggesting that excessive methylation alters gene expression and thereby regulates fat distribution, insulin sensitivity, and inflammation.

The liver is another crucial organ that plays a vital role in maintaining stable blood sugar levels. DNA methylation in the liver can silence key metabolic genes (e.g., *PPARGC1A*, *FGF21*), impairing the liver's ability to control glucose production and lipid metabolism, both crucial for maintaining insulin sensitivity.[1] Genes relevant to the development of T2D, such as *GRB10*,

ABCC3, *MOGAT1*, and *PRDM16*, are aberrantly methylated in the livers of T2D patients in several studies. Studies have also shown that in the livers of people who are severely obese, whether or not they have T2D, specific genes that control how the body processes sugar exhibit reduced DNA methylation in a regulatory region known as the ATF motif. This reduction in DNA methylation increases sugar breakdown via glycolysis; over time, these changes contribute to worsening insulin resistance, making it more difficult for the body to regulate blood sugar levels.[10]

Skeletal muscle is where, after eating, most of the glucose from the blood is taken up, especially when insulin is present. Insulin helps muscle cells absorb glucose, allowing them to use it for energy or store it as glycogen. When skeletal muscle metabolism is functioning correctly, this process helps maintain stable blood sugar levels. However, if skeletal muscle becomes less responsive to insulin due to factors such as inflammation, fat accumulation, or changes in gene expression, glucose uptake slows down. This can lead to insulin resistance, in which the body requires increasing amounts of insulin to achieve the same effect, and blood sugar levels rise. DNA methylation in skeletal muscle alters the expression of essential genes involved in glucose uptake and insulin signalling. For example, DNA methylation changes in genes such as *PDK4*, *INSR*, and *AKT2* can disrupt insulin signalling pathways, making muscle cells less responsive to insulin. Hypermethylation of the *PPARGC1A* gene, mentioned previously, also reduces its expression and lowers the muscle's ability to use glucose efficiently. Over time, these epigenetic changes collectively contribute to the development of insulin resistance, ultimately leading to T2D.[1]

Epigenetic Modulators Target DNA Methylation to Treat T2D

There are several targeted therapy options relating to methylation, including nutritional epigenomics, which refers to how diet influences epigenetic modifications and how these modifications impact disease risk in T2D (Figure 4). Folic acid and B vitamins (B2, B6, B9/folate, and B12) play a crucial role in providing methyl groups; without these nutrients, the body struggles to regulate gene activity properly. Diets rich in bioactive nutrients (such as folate and B vitamins) and polyphenols (plant-based antioxidants) can help alter gene behaviour by affecting epigenetic signalling pathways and influencing gene expression related to insulin sensitivity, glucose metabolism, and inflammation.[11] For example, Mediterranean and Atlantic diets are rich in fibre, fruits, vegetables, and antioxidants; these nutrients have been shown to promote beneficial DNA methylation patterns that help regulate metabolic and inflammatory genes, potentially offering protection against T2D. This suggests that specific dietary approaches can offer therapeutic benefits by modifying gene activity related to obesity, insulin resistance, and inflammation.[12]

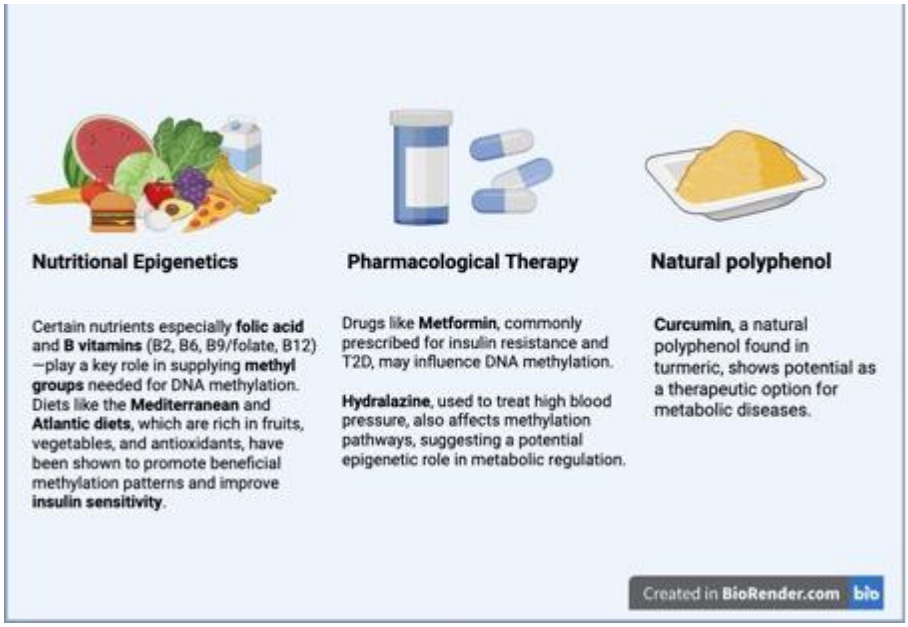

Figure 4. Therapeutic approaches targeting epigenetic regulation in T2D. This figure was created in BioRender.com.

Metformin is one of the pharmacological therapy options for the treatment of T2D, prescribed for people with insulin resistance or T2D. Metformin helps activate AMPK, a protein that monitors cellular energy levels. Once AMPK is activated, the liver produces less glucose, allowing the muscles to take up more glucose, which improves insulin sensitivity. In addition to blood sugar control, metformin also alters DNA methylation at key genes commonly associated with T2D, such as *TCF7L2*, *KCNQ1*, and *ABCG1*, thereby supporting normal insulin secretion and fat metabolism. Ongoing studies are exploring metformin's benefits in prediabetes and metabolic syndrome.[13]

Hydralazine is a drug commonly used to treat high blood pressure, and it has been researched for its ability to influence DNA methylation. It works by inhibiting DNMTs, the enzymes responsible for adding methyl groups to DNA. Research has shown that hydralazine can reverse abnormal methylation patterns and reactivate genes that may help regulate inflammation and metabolism. While most studies have focused on cancer and cardiovascular disease, its potential role in treating metabolic conditions is being explored.

Another targeted therapy option is curcumin, a natural polyphenol and key ingredient in turmeric, which can influence gene expression without altering the underlying DNA. It works by affecting enzymes such as DNMTs and histone-modifying proteins, which play a crucial role in epigenetic regulation. Studies have shown that curcumin can reduce inflammation and oxidative stress, which are major drivers of T2D, and may help improve blood sugar and cholesterol levels. Curcumin also affects microRNAs, which play a crucial role in regulating gene

expression. While curcumin shows promise as a therapeutic option for metabolic diseases such as T2D, its low bioavailability presents a challenge for clinical use.[14]

Limitations and Future Directions

Limitations

A significant limitation currently is the difficulty in distinguishing whether DNA methylation changes are a cause of T2D or simply a consequence of changes that have occurred after insulin resistance and the condition have developed. Several studies have identified altered DNA methylation patterns in T2D patients; however, these studies remain unclear about whether these changes are causal factors in T2D or consequences of hyperglycaemia or obesity. Another limitation is accessing and retrieving samples from key tissues and cells such as pancreatic islets, liver, and skeletal muscle, since this is considered invasive, so much of the evidence comes from blood samples. While blood samples are helpful, they do not fully reflect organ-specific methylation changes.

Methylation-based biomarkers can differ across ethnicities, age groups, and environmental exposures, making it difficult to define universal biomarkers for T2D. Across ethnicities, individuals have different genetic backgrounds, diets, and cultural habits, all of which influence epigenetic patterns. Environmental factors, such as exposure to air pollution or toxins, can also lead to differences between study groups. DNA methylation can change in response to diet, stress, or medication, making it more challenging to define long-term, early-warning signs of T2D development.

While drugs such as metformin, curcumin, and hydralazine have shown beneficial effects in preclinical studies, more large-scale clinical studies or trials are needed before methylation-targeted therapies can be widely used. Focusing solely on DNA methylation overlooks the roles of histone modifications, chromatin remodelling, and non-coding RNAs, all of which significantly influence T2D pathogenesis.

Future Directions

Conducting long-term studies that follow individuals from early life to adulthood could help clarify how early-life exposures shape lifestyle and T2D risk via methylation. Such studies would also help address the limitation of distinguishing whether methylation is a cause or consequence, through consistent follow-up and records. Another direction involves developing personalized or population-specific epigenetic profiles. Studies that include large, diverse groups of individuals from different ethnicities, ages, genders, and environmental backgrounds could help researchers identify consistent biomarkers to predict T2D risk, addressing the limitation regarding biomarkers and ensuring greater relevance and reliability across genetic and environmental backgrounds.

Future treatments could utilize DNA methylation markers, such as *TXNIP* hypomethylation or *INSR* promoter methylation, to distinguish between different patient groups.[15] This would allow for personalized care, including specialised diets, specific exercise programs, or targeted medications depending on a patient's individual epigenetic profile, potentially improving treatment effectiveness and reducing the risk of T2D development or progression. Since DNA methylation is reversible, researchers should explore nutritional interventions, exercise programs, and DNMT inhibitors as safe and effective ways to reprogram the epigenome in T2D. Targeting specific DNA methylation during pregnancy and childhood could also help develop new strategies to reduce the risk of T2D through maternal nutrition and environmental control.

Conclusion

T2D is shaped by genetics, lifestyle, and epigenetic changes, such as DNA methylation, which can disrupt insulin signalling and glucose metabolism in key metabolic tissues, including the pancreas, liver, adipose tissue, and skeletal muscle. Environmental factors, including diet, activity, stress, and early-life exposures, further influence these patterns, many of which are potentially reversible. Lifestyle interventions and emerging therapies targeting DNA methylation hold promise, though challenges remain in establishing causality, achieving tissue specificity, and developing biomarkers. Future work in long-term studies and personalized approaches may advance the prevention, management, and even partial reversal of T2D.

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