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Role of Epigenetics and its Basic Mechanism in Health and Disease

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Abstract

An epigenetic trait is a stably heritable phenotype resulting from changes in a chromosome without alterations in the DNA sequence. Epigenetic mechanisms have a crucial role in regulation of gene activity and expression during development and differentiation or in response to environmental stimuli. Epigenetic research can help explain how cells carrying identical DNA differentiate into different cell types, and how they maintain differentiated cellular states. Epigenetics is thus considered a bridge between genotype and phenotype, a phenomenon that changes the final outcome of a locus or chromosome without changing the underlying DNA sequence. Multiple mechanistic steps lead to the stable heritance of the epigenetic phenotype. The molecular mechanisms of epigenetic phenomena are multiple. In the current work we reviewed recent updates the role of epigenetics and its basic mechanisms in health and disease.

Keywords: Epigenetic, Chromosome, DNA, Disease.

1. Introduction

Epigenetic trait is a stably heritable phenotype resulting from changes in a chromosome without alterations in the deoxyribonucleic acid (DNA) sequence.[1] Three major mechanisms have been categorized by biologists to instruct epigenetic regulations: DNA methylation, histone post-translational modifications (PTMs) and noncoding ribonucleic acid (RNA) (micro-RNA). [2] The interplay of DNA methylation and histone post-translational alterations, are key epigenetic players to rearrange chromatin into areas such as euchromatin, heterochromatin and nuclear compartmentalization. [3] The fact those numerous human diseases, including cancer, have epigenetic mechanisms has led the development of a new therapeutic option that might be termed ‘epigenetic therapy’. Numerous therapeutic agents have been discovered to alter methylation patterns on DNA or the modification of histones (histone acetylation and methylation patterns), and many of these agents are still being tested in clinical trials.[4] In the present work, we reviewed recent updates the role of epigenetics and its basic mechanisms in health and disease.

2. Mechanisms of Epigenetics

Three major mechanisms have been categorized by biologists to instruct epigenetic regulations; DNA methylation, histone post-translational modifications (PTMs) and noncoding RNAs (micro-RNA).[2]

3. DNA Methylation

Methylation of the C-5 position of cytosine residues in DNA has long been recognized as an epigenetic silencing mechanism of fundamental importance. A methyl (-CH3) group is added to the C-5 position of a cytosine adjacent to a guanine residue (cytosine-phosphate-guanine dinucleotides or CpG) that results in 5-methylcytosine, which inhibits DNA transcription leading to gene suppression. In human somatic cells, approximately 70–80% of cytosine in CpG sites is methylated. In some diseases, CpGs of some key genes are reported to be abnormally hyper- or hypomethylated, which further result in transcriptional misregulation. [5]

4. Histone Post-Translational Modifications (PTMs)

Each chromosome consists of thousands of nucleosomes. Histones have protruding N-terminal tails which can post-translationally undergo chemical modifications (so called post-translational histone modifications/marks)[6]. These modifications promote or are repressive against gene expression. As with DNA methylation, they also modulate the binding of transcriptional regulatory complexes to specific sequences. Histone modifications are dynamic and reversible; there are several epigenetic modifier enzymes (epi-enzymes) which are specifically responsible for adding (writers) or removing (erasers) histone modifications. [7]

5. Noncoding RNAs (micro-RNA)

Another mechanism of epigenetic regulation is the non-coding RNA (ncRNA), which also plays a critical role in gene regulation via a process called post-transcriptional gene silencing (PTGS). A ncRNA is a functional RNA molecule that is transcribed from DNA but not translated into proteins. Epigenetic-related ncRNAs are broadly divided into two main groups: long and short ncRNAs. Short ncRNAs (<30 nucleotides) include microRNAs (miRNA), short interfering RNAs (siRNA) and piwi-interacting RNAs (piRNA). A long ncRNA contains >200 nucleotides. Both groups are shown to play a role in heterochromatin formation, histone modification, DNA methylation targeting, and gene silencing[8].

MicroRNAs (miRNAs) are short ncRNAs (approximately 22 nucleotides in length) now recognized as one of the major epigenetic regulatory mechanisms in eukaryotes mainly via post-transcriptional mechanisms.[9]

6. Epigenetics and Human Diseases

The role of epigenetic modifications in the etiopathogenesis of some human diseases has been clarified. They are grouped into the following categories: neoplastic diseases (cancer epigenetics), autoimmune diseases, metabolic diseases, cardiovascular diseases and neurodegenerative diseases.

6.1 Cancer Epigenetics

6.1.1.1 Aberrant Reprogramming of The Epigenome in Cancer

The term “epimutations” has been used to describe cancer-associated epigenetic machinery changes. These epimutations, along with widespread genetic alterations, play an important role in cancer initiation and progression. The cancer epigenome with its epimutations result in global dysregulation of gene expression profiles

leading to the development and progression of cancer phenotype. The interplay between epimutations and genetic mutations and deletions that finally leads to initiation and progression of cancer genes has been studied.[10]

6.1.1.2 DNA Methylation Aberrations in Cancer

A cancer epigenome is marked by genome-wide hypomethylation and site-specific CpG island promoter hypermethylation.[11]
Global DNA hypomethylation plays a significant role in carcinogenesis. DNA hypomethylation at repeat sequences leads to increased genomic instability by promoting chromosomal rearrangements. In addition, DNA hypomethylation can lead to the activation of growth-promoting genes, such as R-Ras and MAPSIN in gastric cancer.[12]
Contrary to hypomethylation, site-specific hypermethylation contributes to carcinogenesis by silencing tumour suppressor genes.[13]

6.1.1.3 Changes in Histone Modifications in Cancer

Studies have revealed a global loss of acetylated H4-lysine 16 (H4K16ac). Such loss of histone acetylation, which is mediated by histone deacetylases (HDACs), results in gene repression. HDACs are often found overexpressed in various types of cancer and thus, have become a major target for epigenetic therapy.[14]
In addition to changes in histone acetylation, cancer cells also display extensive changes in histone methylation patterns. Alterations in H3K9 and H3K27 methylation patterns are associated with aberrant gene silencing in various forms of cancer.[15]
In addition to histone methyl transferases (HMTs), lysine-specific demethylases that work in coordination with HMTs to maintain global histone methylation patterns are also implicated in cancer progression. Lysine-specific demethylase 1 (LSD1), can effectively remove both activating and repressing marks (H3K4 and H3K9 methylation, respectively) thus, acting as either a corepressor or a co-activator After LSD1.[16]

6.1.1.4 Dysregulation of miRNAs in cancer

A miRNAs can act as either tumour suppressors or oncogenes depending on their target genes.[17] While the exact mechanism of action of miR-155 is still unclear, there are suggestions that it may play a role in the class switch recombination process by targeting activation-induced cytidine deaminase.[18]

6.1.2 Epigenetic Changes in Some Cancers

6.1.2.1 Colorectal Cancer

Three major mechanisms affecting gene function in colorectal cancer (CRC) have been recognized. These include microsatellite instability, chromosomal instability and CpG island methylator phenotype.[19]

6.1.2.2 Breast Cancer

Studies have shown that tumor suppressor genes BRCA1 and p16, DNA repair genes GSTP1 and CHD1, which all are linked with metastasis and invasion, were found hypermethylated in breast cancer samples. Specifically, promoter methylation of BRCA1 tumor suppressor gene is associated with developing aggressive breast tumors. In addition, the methylation status of ADAM23 gene, which is a member of surface molecules involved in the cell adhesion process, were analyzed.[20]
Also, post-translational histone modifications have a critical role in breast

tumorigenesis and aggressiveness of prognosis. Elsheikh et al. evaluated the relative levels of histone lysine acetylation in 880 primary operable invasive breast carcinoma cases. H4R3me2, H3K9ac and H4K16ac were detected at significantly low levels relevant to large tumor size.[21]

6.1.2.3 Cervical Cancer

Within the HPV16L1 region, 14 tested CpG sites have significantly higher methylation in CIN3+ than in HPV16 genomes of women without CIN3. Only 2/16 CpG sites tested in HPV16 upstream regulatory region were found to have an association with increased methylation in CIN3+. This suggests that the direct route from infection to cancer is sometimes detoured to a precancerous state in cervix intraepithelial neoplasia.[22]
In addition, a correlation exists between CIN3+ and increased methylation of CpG sites in the HPV16 L1 open reading frame. This could be a potential biomarker for future screens of cancerous and precancerous cervical disease.[23]

6.1.2.4 Leukemia

Studies have shown that mixed-lineage leukaemia (MLL) gene causes leukaemia. MLL protein is required for the epigenetic maintenance of gene activation during development and is also mutated in a subset of aggressive acute leukaemia's. MLL maintains gene activation in part by methylating histone 3 on lysine 4.[24]

6.1.2.5 Sarcoma

Several oncogenes and tumour suppressor genes are epigenetically altered in sarcomas. These include APC, CDKN1A, CDKN2A, CDKN2B, Ezrin, FGFR1, GADD45A, MGMT, STK3, STK4, PTEN, RASSF1A, WIF1 as well as several miRNAs. Expression of epigenetic modifiers such as that of the BMI1 component of the PRC1 complex is deregulated in chondrosarcoma, Ewing's sarcoma and osteosarcoma.[25]

6.2 Autoimmune Diseases

Epigenetic homeostasis failure, as a response to environmental agents, can result in changes of gene expression in specific cells leading to dysregulated self-tolerance and the development of a wide spectrum of autoimmune diseases.[26]

6.2.1 Systemic Lupus Erythematosus (SLE):

Several studies have shown that there is a global hypomethylation of promoter regions, which contain the genes that are overexpressed in the disease such as ITGAL, CD40LG, PRF1, CD70, IFGNR2, MMP14, LCN2 and in the ribosomal RNA gene promoter (18S and 28S). This gene overexpression results in cell hyperactivity, perpetuation of the immune and inflammatory responses[27].
Moreover, histone modifications such as histone 3 lysine 4 trimethylation (H3K4me3), histone 3 lysine 8 (H4K8) triacetylation, histone 3 lysine 27 trimethylation (H3K27me3) and histone 2B lysine 12 acetylation (H2BK12ac) cause an increase in apoptotic nucleosomes which generate auto-immunogenicity that causes activation of antigen-presenting cells and autoantibody production with a subsequent inflammatory response.[28]

6.2.2 Rheumatoid Arthritis (RA)

In RA, global hypomethylation of these cells is responsible for the overexpression of inflammatory cytokines in synovial fluid.[29]Furthermore, RA is characterized by an imbalance between HAT and HDAC activity in synovial tissue.[30]

6.2.3 Type 1 Diabetes (T1D)

In T1D, there is a global hypermethylation activity caused by altered metabolism of homocysteine.[31]
Glucose and insulin levels are determinants of methylation. They modify homocysteine metabolism by increasing the production of homocysteine through its inhibition of trans-sulfuration. When there are elevated levels of homocysteine, intra-cellular methionine will be catalyzed by DNA methyl transferases (DNMTs) in S-adenosylmethionine. This will enhance DNMT activity that will, in turn, lead to increased global DNA methylation (hypermethylation).[32]

6.2.4 Multiple Sclerosis (MS)

Studies have shown that the promoter region of peptidyl arginine deiminase type II (PAD2) is hypomethylated in MS. PAD2 plays a key role in the citrullination process of myelin basic protein (MBP).[33]
A previous study found that miR-326 was significantly upregulated in patients with relapsing-remitting MS which produced an increase in Th-17 cell numbers and more severe symptoms. Other miRNAs involved in MS are miR-34a and miR-155, which are found upregulated in active MS lesions and take part in MS pathogenesis by targeting CD47.[28]

6.3 Metabolic Diseases

6.3.1 Obesity

In a large-scale study investigating DNA methylation in CD4+ T cells, eight CpG sites were associated with BMI and five with waist circumference. Those CpG sites are annotated to CPT1A, CD38 and PHGDH genes. CPT1A takes part in carnitine-dependent transport across the mitochondrial membrane when oxidation of long-chain fatty acids is initiated. In addition, DNA methylation in intron 1 of CPT1A was inversely associated with both BMI and waist circumference.[34]
The largest study to date, combining DNA methylation data from over 10,000 whole blood samples, identified 187 CpG sites significantly associated with BMI.[35]

6.3.2 Type 2 diabetes mellitus

Patients with T2D were found to have increased DNA methylation and decreased expression of these key genes (what key gene?), which were associated with impaired insulin secretion. In addition, high glucose and glycated hemoglobin (HbA1c) seemed to directly increase DNA methylation of these genes.[36]

6.4 Cardiovascular Diseases (CVDs)

6.4.1 Epigenetics in Congenital Heart Diseases (CHDs)

In a study examining the methylation profiles of DNA isolated from fetal cardiac tissue obtained from normal individuals, those with isolated CHD and those with Down syndrome with and without CHD, several intragenic sites within the muscle segment homeobox 1 (MSX1) gene and the GATA4 gene were identified as being hypermethylated which was associated with dysregulated expression of these genes.[37]

6.4.2 Epigenetics in Hypertension

In addition to Histone modification which is reported to be involved in the regulation of blood pressure[38] Hypomethylation of the promoter region of the Sic2a2 gene leads to increased expression of Na+, K+ and 2Cl- cotransporter 1 (NKCC1) which was positively correlated with hypertension.[39](more data).

6.4.3 Epigenetics in Atherosclerosis

Studies have identified genes such as Collagen Type XV alpha 1 chain

(COL15A1), early B-cell factor 1 (EBF1) and nucleotide-binding oligomerization domain containing 2 (NOD2) that are hypomethylated while genes such as monocarboxylate transporter SLC16A3 (MCT3), human mitogen-activated protein kinase 4 (MAP4K4) and zinc finger E-box binding homeobox 1 (ZEB1) are hypermethylated in patients with atherosclerosis.[40]

6.5 Neurodegenerative Diseases

The association between epigenetic signature and neurologic disease has been established through observation of monogenic neurodevelopmental disorders.[42]

6.6 Epigenetics in Alzheimer's Disease

It has been shown that there is genome-wide DNA methylation of more than 900 CpG sites representing 918 unique genes associating late onset AD. The best candidate gene has been found to be a Transmembrane Protein 59 (TMEM59), whose promoter was hypomethylated in AD.[43]

Direct evidence of disturbed histone modifications in AD was proposed where elevated levels of phosphorylated histone H3 in AD hippocampal neurons were found and dislocalised to neuronal cytoplasm, as opposed to the nucleus as in actively dividing cells.[44]

miRNAs have been linked to posttranscriptional control of amyloid precursor protein expression, namely negative regulatory control by miR-101 and miR-16. Smith et al have shown that miR-124 is down-regulated in AD brain.[45]

6.7 Epigenetics in Parkinson's Disease

Depositions of misfolded α-synuclein constitute a pathologic hallmark of Parkinson's disease (Lewy bodies) and co-locate with sites of neuronal loss. In fact, studies have shown α-synuclein associates with histones and inhibits their acetylation, by means of its association with Sirtuin-2 (Sirt2) histone deacetylase.[46]

The role of miRNA regulatory system has also been implicated and surveyed in PD. A miRNA molecule (miR-133b) was detected with expression specific to midbrain dopaminergic neurons and reduction of expression level in PD patients' midbrain tissue samples. On the other hand, the role of DNA methylation in PD is unclear.[47]

7. Prospects for Epigenetic Therapy

Numerous therapeutic agents have been discovered to alter methylation patterns on DNA or the modification of histones.[4]

7.1 Targeting DNA Methylation

DNA methylation inhibitors can be divided into two groups: nucleoside analogues and non-nucleoside analogues.

Nucleoside analogues have a modified cytosine ring and can be converted into nucleotides and incorporated into newly synthesized DNA or RNA. DNA methyltransferases are bound by covalent bonds with the analogues, which inhibits DNA methylation. The prototype members of this nucleoside analogues group are 5-azacytidine (5-aza-CR) and 5-aza-2-deoxycytidine (5-aza-CdR). 5-aza-2-deoxycytidineis also known as Decitabine. 5-Azacytidine is currently used as an injectable suspension for the treatment of myelodysplastic syndromes. It promotes cell differentiation, demethylation and re-expression of inactivated genes.[48 49]

Many non-nucleoside analogues have been developed to prevent DNA from aberrant hypermethylation. These drugs are usually small molecular inhibitors and directly target catalytic sites rather than incorporating into DNA. Based on a three-dimensional model of DNMT1, RG108 was designed to block the activity of this enzyme and cause demethylation.[50]

8. Inhibitors of HMTs and HDMTs

EPZ004777 was the first identified selective inhibitor of DOT1L, a selective histone H3K79 methyltransferase. However, due to its poor pharmacokinetic features, a second generation (EPZ-5767) was developed with a cyclobutyl ring replacing the ribose moiety of the molecule. EPZ-5767 (Pinometostat) shows synergistic effects with cytarabine, daunorubicin and the DNMT inhibitor azacitidine in treatments for acute lymphoblastic leukemia with MLL rearrangement.[51]

Tranylcypromine (TCP) is an approved drug for depression due to its ability to inhibit monoamine oxidase (MAO) activity. TCP is acts as an inhibitor of lysine-specific histone demethylase 1A (LSD1) being capable of re-sensitizing non-acute promyelocytic leukemia cells to all-trans retinoic acid (ATRA) treatment via increasing H3K4me2 and the expression of myeloid-differentiation-associated genes leading to cancer cell differentiation and apoptosis.[52]

9. Conclusion

Epigenetic changes in gene expression not only create heritable phenotypic diversity within an individual but also within populations, independent of genetic variation. Changes in the regulation of gene expression levels have long been thought to play a vital role in evolution and adaptation. It has been hypothesized that epigenetic variants could be a novel substrate for natural selection and thus participate in the adaptation of species in changing the environment.

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