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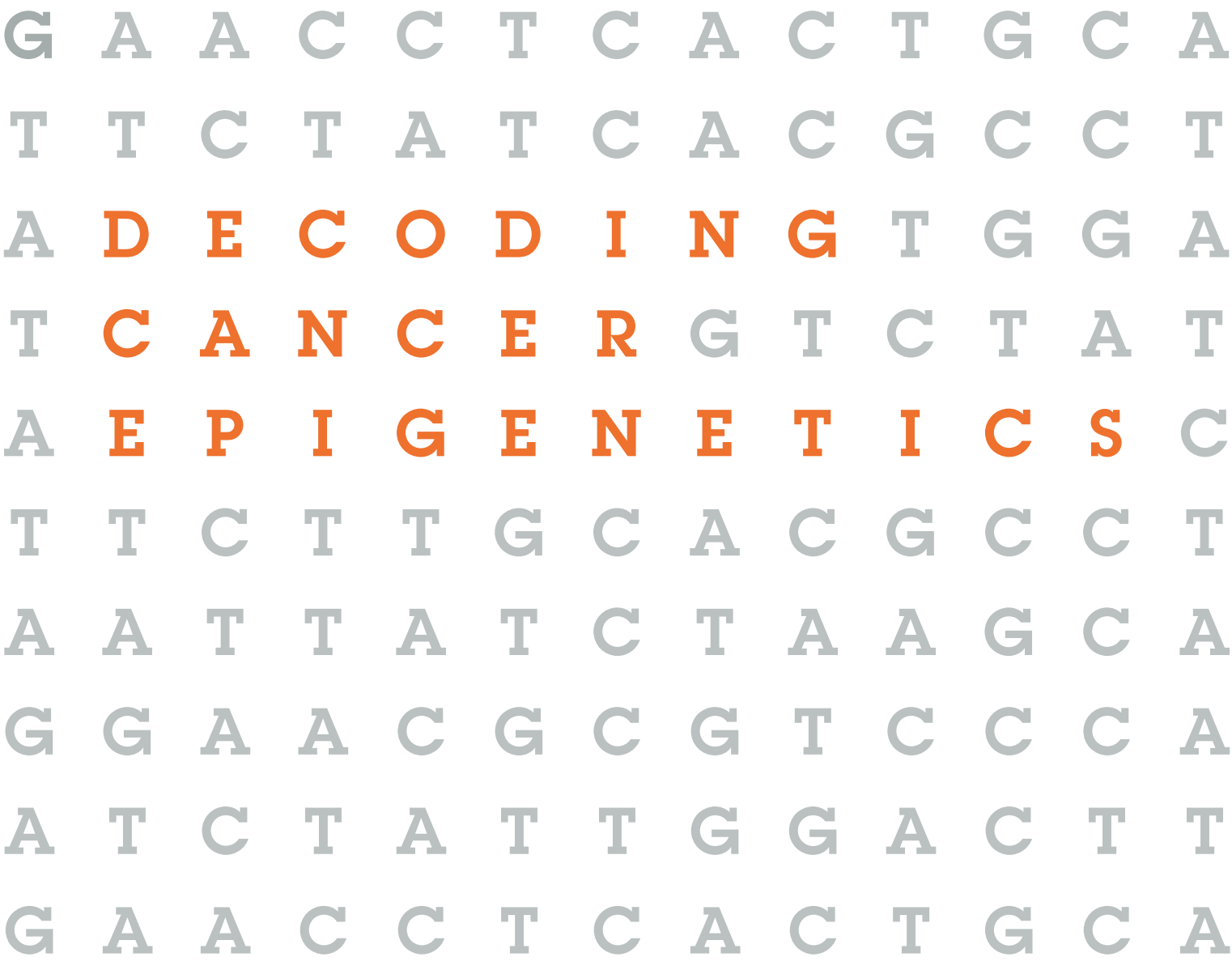
Guide to Cancer Epigenetics



Contents

Learn the role of various epigenetic targets and pathways in cancer with our comprehensive guide, which covers histone regulation, DNA and RNA modifications as well as polycomb and chromatin remodeling.

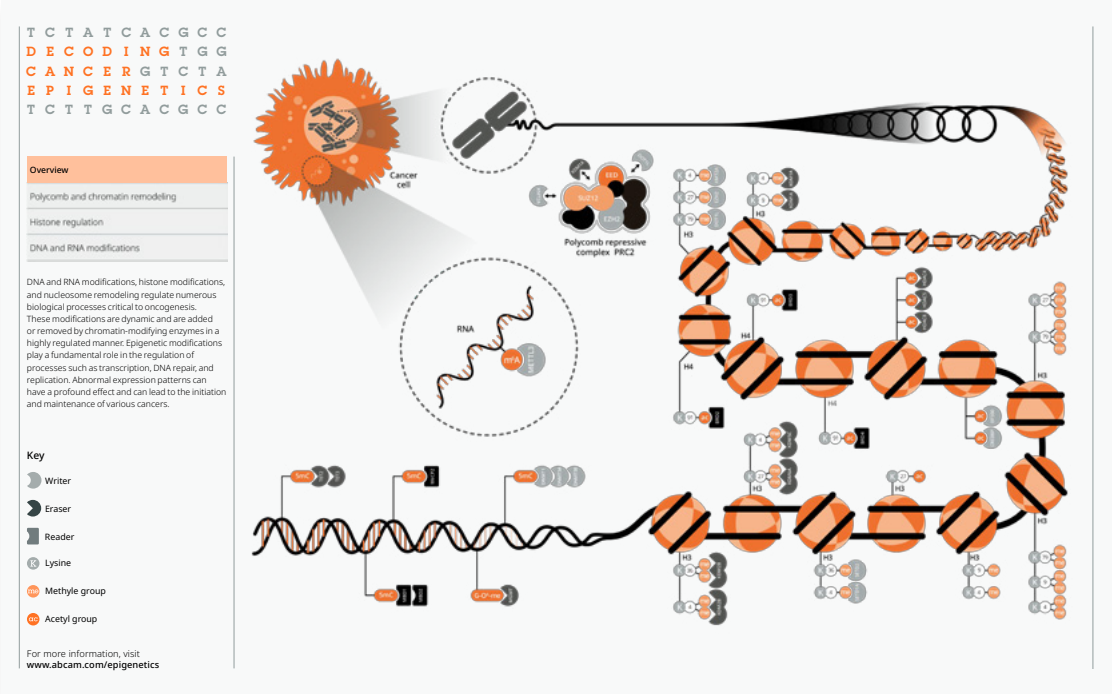
The link between epigenetics and cancer

Epigenetic mechanisms are essential for the normal development and maintenance of tissue-specific gene expression patterns in mammals. Epigenetic modifications play a fundamental role in the regulation of processes such as transcription, DNA repair, and replication. Alterations of genes regulating epigenetic processes are frequently found as cancer drivers and may be caused by changes in DNA and RNA modifications, histone modifications, and nucleosome remodeling. Abnormal gene expression patterns can lead to the initiation and maintenance of various cancers.

Whole-genome sequencing studies demonstrated the presence of recurrent somatic mutations in numerous epigenetic regulators in various types of cancer[1,2]. Histone modifications such as acetylation and methylation are among the most widely affected epigenetic pathways in cancer. For example, mutations in a histone demethylase UTX (KDM6A) are shown in up to 12 histologically distinct cancers[3].

The reversible nature of epigenetic modifications has led to the emergence of the promising field of epigenetic therapy. Multiple small-molecule epigenetic inhibitors are being developed for use in cancer therapies. Three of these inhibitors, targeting DNMTs, HDACs, and JAK2, have already gained approval by the US Food and Drug Administration (FDA).

Key cancer epigenetics targets – interactive poster



Study key cancer epigenetics targets with our interactive poster

Epigenetic pathways in cancer

Histone regulation

Histone modifications play a major role not only in transcription but in all DNA-templated processes[4]. The activity of many proteins that modify or bind certain histone modifications is misregulated in cancer. We will discuss below essential histone modifications related to cancer (Figure 1).

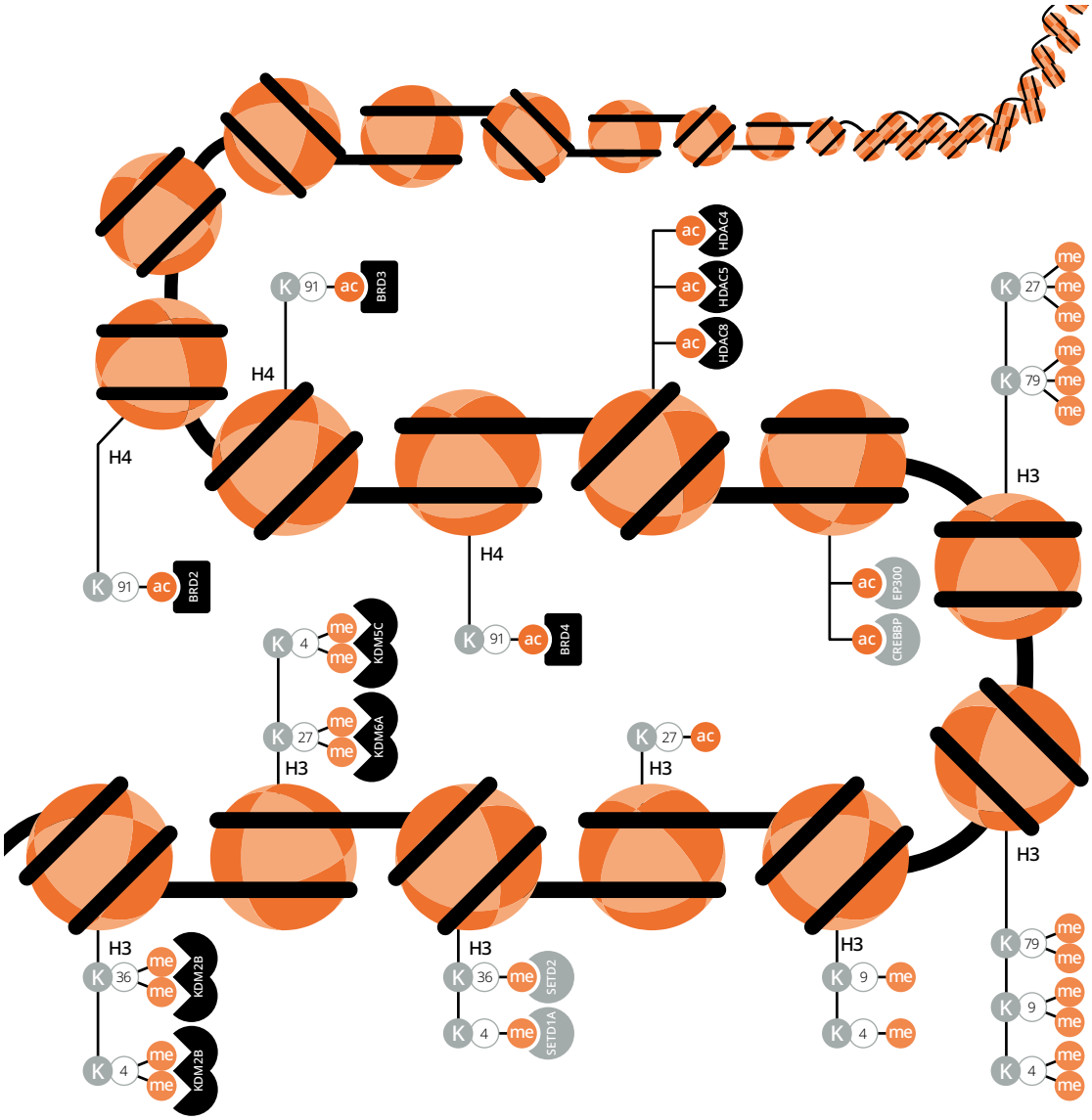


Figure 1. Key histone proteins and their modifications involved in cancer.

Histone acetylation

The N-terminal acetylation of lysine residues is a major histone modification involved in transcription, chromatin structure, and DNA repair. Acetylation adds a negative charge to lysine residues; and these negative charges repel negatively charged DNA, which results in a relaxed chromatin structure. The open chromatin conformation allows transcription factor binding and significantly increases gene expression[5].

Histone acetylation is a highly dynamic process regulated by two competing enzymatic families: the histone lysine acetyltransferases (KATs) and the histone deacetylases (HDACs).

KATs family has two major classes: type-A and type-B. Type-A KATs are primarily nuclear and are classified into the GNAT, MYST, and CBP/p300 families. Type-B KATs are predominantly cytoplasmic and modify free histones[6]. Many KATs are shown to be implicated in neoplastic transformation. Also, KATs function as transcription co-factors for several tumor suppressors, eg CBP/p300 acetylate p53[7].

Histone deacetylases (HDACs) are the family of 18 enzymes that reverse lysine acetylation and restore the positive charge on the histone side chain. The HDACs are overexpressed in numerous cancers, which results in a global loss of histone acetylation and silenced tumor suppressor gene expression.

The HDACs are divided into four major classes, depending on their sequence homology and structure. Class I HDACs (1, 2, 3, 8) and class II HDACs (4–7, 9, 10) have been reported to play roles in oncogenesis. HDAC4 is found to be a prognostic marker for pancreatic and ovarian cancers[8], whereas HDAC5 may be a marker for liver, renal and cervical cancers[9]. Furthermore, numerous HDACs inhibitors are currently being developed as potential anti-cancer therapies[10]. HDAC8 is a promising novel target in cancer, and many potent HDAC8-selective inhibitors have been reported to exhibit cellular activity and anticancer effects[11].

The histone acetylation marks on lysine residues are read by small protein modules, called "readers". These protein modules are called bromodomains and include BRD2, BRD3, and BRD4, which are members of the BET family. BET proteins are essential for transcriptional elongation and cell-cycle regression. BET bromodomains represent a promising therapeutic target in cancer. Several studies demonstrated the possibility of the development of highly specific and chemically distinct small molecules against the BET family[12–14].

Histone methylation

Histones are methylated on the side chains of arginine, lysine, and histidine residues. Methylation, unlike acetylation and phosphorylation, does not alter the overall charge of the molecule.

The best-studied histones, implicated in lysine methylation, include H3K4, H3K9, H3K27, H3K36, and H3K79. Methylation is associated with activation or deactivation of gene expression[14]. Importantly, different methylation states (eg monomethyl me1, dimethyl me2, trimethyl me3) of the same lysine residue may have distinct functional consequences. For example, H3K4me2/3 usually spans the transcriptional start site of active genes[15], whereas H3K4me1 modification is associated with active enhancers[16]. Similarly, H3K9me1 is found at active genes, while H3K9me3 is associated with gene repression[15].

Histone methylation of lysine residues is carried out by highly specific enzymes called the histone lysine methyltransferases (KMT). Cytogenetic and NGS studies of various cancer genomes have demonstrated recurrent translocations and/or coding mutations in many KMTs, including MLL family members. Also, recurrent coding mutations have been reported in several histone demethylases such as KDM5A (JARID1A), KDM5C (JARID1C), and KDM6A (UTX). These recurrent mutations may significantly alter the catalytic activity of the methyltransferases or demethylases. Additionally, as many of these enzymes contain chromatin-reader motifs, the mutations may affect the ability of these proteins to survey and bind epigenetic modifications[6].

Learn more about histone modifications

DNA modifications

Changes in both global and individual gene methylation patterns are often found in cancer, and methylation patterns can distinguish tumor types. Global DNA hypomethylation is commonly observed in cancer genomes. However, hypermethylation is frequently detected in specific CpG-rich regions leading to the silencing expression of tumor suppressors. 5%–10% of normally unmethylated CpG promoter islands become abnormally methylated in various cancer genomes. CpG island methylation plays an important role in transcriptional regulation and is commonly altered during malignant transformation[17,18].

Methylation of the 5-carbon of cytosine nucleotides is carried out by a family of DNA methyltransferases (DNMTs), which includes 3 active DNMTs in higher eukaryotes (Figure 2). DNMT1 is a maintenance methyltransferase that recognizes hemimethylated DNA generated during DNA replication and then methylates newly synthesized CpG dinucleotides, whose partners on the parental strand are already methylated[19]. DNMT3a and DNMT3b function primarily as de novo methyltransferases to establish DNA methylation during embryogenesis[20]. Recurrent loss-of-function mutations in DNMT3A have been reported in up to 25% of patients with acute myeloid leukemia (AML)[21].

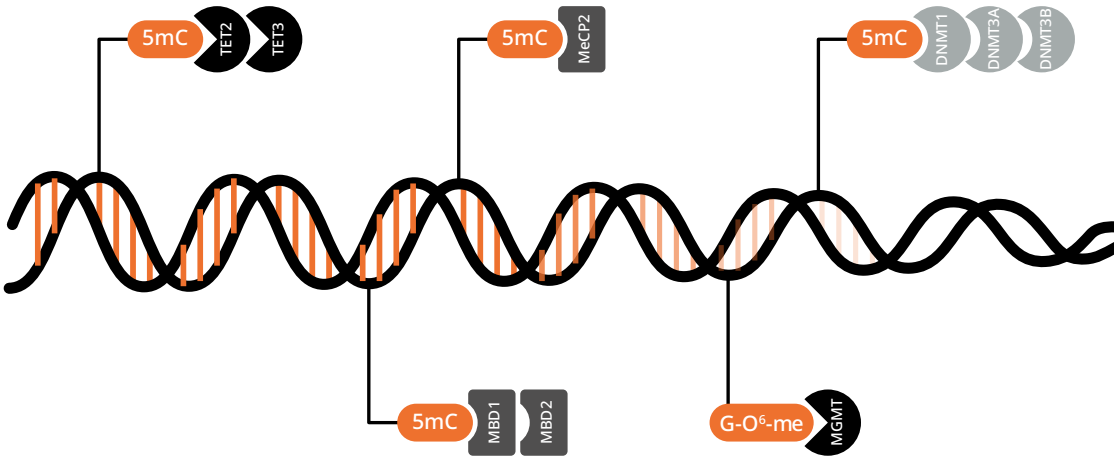


Figure 2. Key DNA modifications critical in cancer.

DNA methylation provides a platform for several methyl-binding proteins, including MBD1, MBD2, and MeCP2 (Figure 2). These proteins in turn function to recruit histone-modifying enzymes to coordinate the chromatin-templated processes[22]. MBD1, MBD2, and MeCP2 all act as transcriptional repressors and are implicated in various cancers. Thus, MeCP2 has been identified as a frequently amplified oncogene in several cancer types, including breast, lung, cervical, and uterine cancers[23]. MBD1 is involved in tumor development and progression of multiple cancers, including lung, prostate, and colorectal cancers[24]. Finally, MBD2 is essential for the maintenance and spread of DNA methylation at CpG islands and shores in cancer[25].

DNA hydroxymethylation is another type of DNA modification, in which the hydrogen atom at the C5-position in cytosine is replaced by a hydroxymethyl group. The ten-eleven translocation (TET 1–3) family of proteins carry out DNA hydroxymethylation in mammals by catalyzing the successive oxidation of 5-methylcytosine (5mC) to 5-hydroxymethylcytosine (5hmC), 5-formylcytosine (5fC), and 5-carboxylcytosine (5caC)[26,27]. TET genes, and especially TET2, are frequently mutated and show reduced expression in various cancers. Thus, TET2 was found to mutate recurrently in numerous hematopoietic malignancies, including AML, MPD, MDS, CMML[28–30]. However, it is unknown how the TET proteins contribute to preventing the onset and maintenance of these malignancies[31].

Learn more about DNA methylation and demethylation

RNA modifications

Non-coding RNAs are essential for normal development and may play a fundamental role in epigenetic regulation[32]. Small non-coding RNAs have a high degree of sequence conservation across species and are involved in transcriptional and post-transcriptional gene silencing through specific base-pairing with their targets. Long non-coding RNAs show poor sequence conservation across species and may act as molecular chaperones for various chromatin regulators[33].

Similar to DNA, RNA is subject to a covalent modification such as methylation (Figure 3). Among all RNA modifications, N6-methyladenosine (m6A) is the most common, abundant, and conserved internal transcriptional modification, especially within eukaryotic messenger RNAs (mRNAs). m6A RNA modifications play an important role in RNA transcription, processing, translation, and metabolism and are associated with multiple diseases such as obesity, infertility, and cancer. In cancer, these modifications have been linked to the oncogenesis of multiple malignancies, including AML, glioblastoma, lung cancer, and others[34].

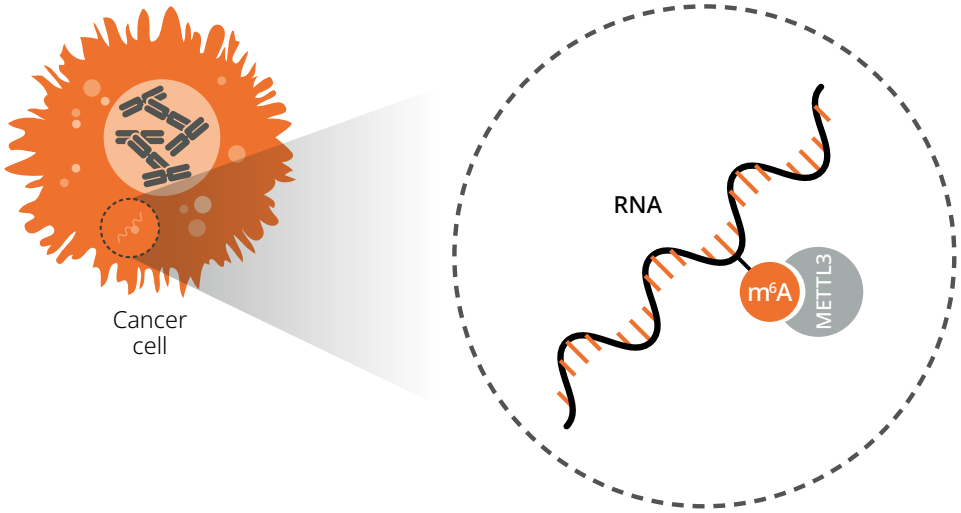


Figure 3. Key RNA modifications involved in cancer.

m6A methylation is a dynamic and reversible process coordinated by methyltransferases, such as METTL3, demethylases, and identifiers. METTL3 is involved in all stages of the life cycle of RNA. It plays a key role in pre-mRNA splicing, 3'-end processing, nuclear export, translation regulation, mRNA decay, and microRNA processing[35]. METTL3 affects tumor formation by regulating the m6A modification in the mRNAs of critical oncogenes or tumor suppressors. For example, METTL3 acts as an oncogenic signal in lung adenocarcinoma and as a tumor suppressor in glioblastoma[36,37]. METTL3 was also shown to be upregulated in various solid tumors, including hepatocellular carcinoma, associated with poor prognosis[38].

RNA epigenetics (or epitranscriptomics) can present new opportunities for early-stage diagnostics and treatment of cancer. Thus, since METTL3 have been detected in multiple cancer types, targeting m6A-associated proteins might constitute a new form of cancer treatment[39]. For instance, novel small molecule inhibitors of METTL3 have been demonstrated as an effective therapeutic strategy in AML[40].

You can find more detailed information on RNA modifications here.

If you are particularly interested in the m6A pathway, check out our poster:

m6A pathway and functions poster

If you want to learn more about the research on METTL3 in AML, watch our webinar presented by Dr Konstantinos Tzepelis from the Milner Therapeutics Institute (University of Cambridge, UK):

Drugging the epitranscriptome as an effective anti-cancer strategy

Polycomb and chromatin remodeling

Polycomb group (PcG) proteins are transcriptional repressors that regulate several crucial developmental and physiological processes. PcG proteins have been linked to cancer development and progression; several cancer types demonstrate the deregulation of the PcG protein expression and function.

PcG proteins selectively repress gene expression during embryonic development by the formation of multi-subunit complexes called polycomb repressive complexes (PRCs) (Figure 4). These complexes regulate chromatin organization and keep it in a transcriptionally inactive state[41]. PRC targets consist of diverse genes encoding transcription factors, signaling proteins, receptors, and regulators involved in all major developmental pathways. The PRCs comprise of PRC1 and PRC2 (with additional variants also being reported), which induce covalent post-translational histone modifications. While the PRC1 subunits catalyze the monoubiquitination of histone H2A at lysine 119 (H2AK119Ub1), the PRC2 subunits catalyze di- and trimethylation of histone H3 at lysine 27 (H3K27me2/3). Both histone modifications are associated with transcriptional silencing[42].

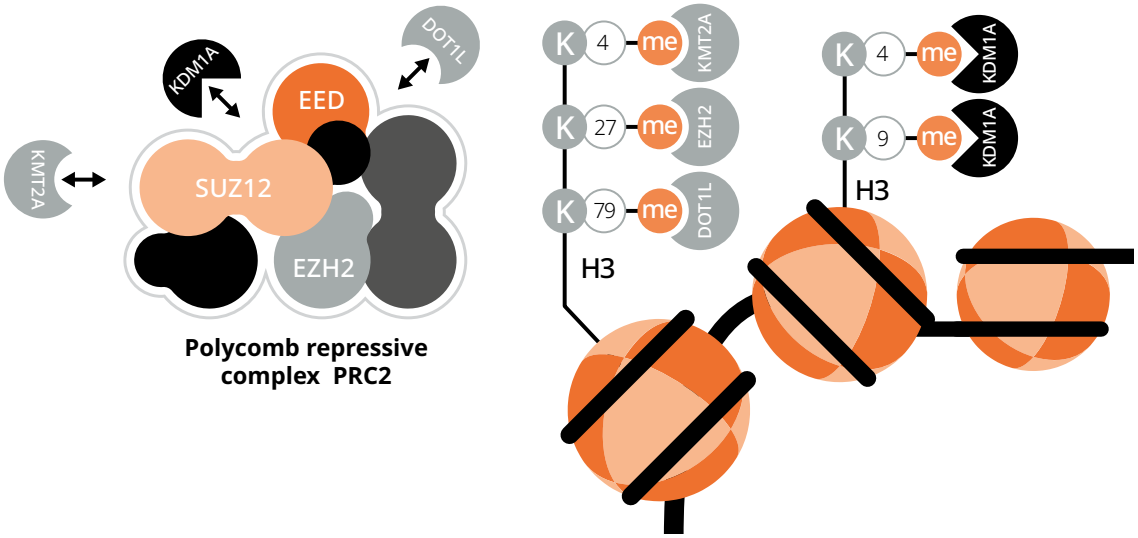


Figure 4. Key polycomb and chromatin remodeling proteins involved in cancer.

PRC2 activity is often misregulated in various human cancers and thereby became a focus for therapeutic drug development. PRC2 complex contains several proteins, including EZH2, EED, and SUZ12. EZH2 is a PRC2 component that catalyzes di- and trimethylation of H3K27. Extensive evidence has linked EZH2 to cancer development and progression. EZH2 overexpression and gain-of-function and loss-of-function mutations were identified in various cancer types. Furthermore, EZH2 overexpression is correlated with a worse progression of several cancers, including prostate cancer[43]. The overexpression of EZH2 and its mutations in various cancers motivated the development of small-molecule inhibitors of EZH2, which are currently a hot topic in drug development[9]. Thus, several EZH2 inhibitors have been developed and are being investigated in clinical trials.

Several other important epigenetic targets are involved in the interactions with PRC2 complex, including histone methyltransferases DOT1L and KMT2A and histone demethylase KDM1A. KDM1A is highly expressed in several cancers and is specifically required for the terminal differentiation of hematopoietic cells[44].

DOT1L, H3K79 methyltransferase, is a promising epigenetic target, which plays an important role in mixed lineage leukemia. Furthermore, deletion and somatic mutations of the DOT1L gene are found in several types of solid cancers including melanoma, colorectal cancer, and ovarian cancer[45,46].

Lysine methyltransferase 2 (KMT2, also known as MLL) methylates H3K4 to promote genome accessibility and transcription. KMT2 genes are frequently mutated in many human cancers and are associated with some of the most common and deadly solid tumors, such as lung and colon carcinomas[47–49].

Summary

The epigenetic modifications and proteins described in this guide are by far not the only ones involved in cancer. For an in-depth analysis of cancer epigenetics, its pathways, and epigenetic targets, we recommend the following review articles:

Dawson and Kouzarides 2012

Nebbioso et al 2018

Laugesen et al 2016

If want to learn more about the key techniques used in epigenetics research, refer to our Epigenetics application guide:

Epigenetics application guide

Also, you can check out our posters on histone modifications and RNA modifications:

Histone modifications

RNA modifications

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