Review Article

Research Progress of m⁶A RNA Methylation in Skin Diseases

Chang Liu [ID], Xin Wang [ID], Shengju Yang [ID], and Shuanglin Cao [ID]

Department of Dermatology, Affiliated Hospital of Nantong University, Medical School of Nantong University, Nantong 226001, China

Correspondence should be addressed to Shengju Yang; yangshengju@ntu.edu.cn and Shuanglin Cao; caoshuanglin@ntu.edu.cn

N^6-Methyladenosine (m⁶A) is the most common mRNA modification in eukaryotes and is a dynamically reversible posttranscriptional modification. The enzymes involved in m⁶A modification mainly include methyltransferases (writers), demethylases (erasers), and methylated readers (Readers). m⁶A modification is mainly catalyzed by m⁶A methyltransferase and removed by m⁶A demethylase. The modified RNA can be specifically recognized and bound by m⁶A recognition protein. This protein complex then mediates RNA splicing, maturation, nucleation, degradation, and translation. m⁶A also alters gene expression and regulates cellular processes such as self-renewal, differentiation, invasion, and apoptosis. An increasing body of evidence indicates that the m⁶A methylation modification process is closely related to the occurrence of various skin diseases. In this review, we discuss the role of m⁶A methylation in skin development and skin diseases including psoriasis, melanoma, and cutaneous squamous cell carcinoma.

1. Introduction

In recent years, RNA methylation-mediated epigenetic epitranscriptome regulation has emerged as a new field of RNA biology. There are several common internal modifications of mRNA including N^6-methyladenosine (m⁶A), N^1-methyladenosine (m¹A), 5-hydroxymethyl-cytosine (hm⁵C), N^7-methylguanosine (m⁷G), and pseudouridine (Ψ). Among them, N^6-methyladenosine (m⁶A) is the most frequent modification of eukaryotic mRNA with a dynamic and reversible posttranscriptional manner. The modifications of mRNA exist in the 5′ cap and 3′ poly-A regions of eukaryotes. The methylation in the 5′ untranslated region (5′UTR) of mRNA plays an important role in mRNA splicing, editing, stability, degradation, and polyadenylation, while the methylation in the 3′ untranslated region (3′UTR) contributes to mRNA outward transport, translation initiation, and mRNA structural stability maintenance with poly A-binding proteins [1]. N^6-Methyladenosine (m⁶A) is enriched near termination codon and 3′UTR and translated near 5′UTR or in long exons [2]. Recent studies have shown that m⁶A methylation regulates cell division and proliferation, death and apoptosis in nervous system development, circadian rhythm, DNA damage response, heat shock response, and tumorigenesis in mammals [3–8]. Therefore, m⁶A methylation defect or dysregulation may disrupt gene expression to result in RNA metabolism abnormality and disease.

Recently, an increasing body of evidence indicates that the m⁶A methylation modification process is closely related to the occurrence of various dermatological diseases. In this review, we summarized the m⁶A RNA methylation and the emerging progress in skin diseases including psoriasis, melanoma, squamous cell carcinoma, Merkel cell carcinoma, and atopic dermatitis.

2. m⁶A Methylation

The dynamic modification of m⁶A methylation requires the participation of three types of regulators. First, m⁶A methyltransferase with the encoding gene called "Writers" catalyzes the m⁶A methylation from RNA. Second, m⁶A demethylase with the encoding gene called "Eraser" removes the m⁶A methylation in RNA. Third, m⁶A-methylated reader proteins bind to m⁶A sites in RNA to perform specific biological

functions. Above enzymes play different roles of in dynamic modification of m^{6}A methylation (Figure 1).

2.1. Writers. RNA m^{6}A methyltransferases include methyltransferase-like protein 3 (METTL3), METTL5, METTL14, METTL16, Wilms' tumor 1-associated protein (WTAP), RNA-binding motif protein 15 (RBM15/15B), and zinc finger CCCH-type containing 13 (ZC3H13) [9]. The m^{6}A installation of methylation is catalyzed by several methyltransferase complexes (MTC). Methyltransferase-like 3 (METTL3) is an S-adenosylmethionine- (SAM-) binding protein, which is the key methyltransferase for m^{6}A methylation and the most important component of m^{6}A MTC. Abnormal expression of METTL3 can change the total methylation level of m^{6}A. As a structural support of METTL3, METTL14 forms a stable methyltransferase complex at a ratio of 1 : 1 to induce m^{6}A RNA [10]. METTL16, as a newly discovered independent RNA methyltransferase, may be involved in the regulation of mRNA splicing and stability and binding with the METTL3/METTL14 methylation complex [11]. RBM15/15B assists in the binding of METTL3 and WTAP by directing these two proteins to specific RNA sites for m^{6}A modification [12]. Other proteins, such as ZC3H13 and cofactors including WTAP, collaboratively control m^{6}A methylation [13]. Taken together, these enzymes form complexes and then cooperate with each other or cofactors to regulate m^{6}A methylation.

2.2. Erasers. Demethylase mainly includes fat mass and obesity-associated protein (FTO) and AlkB homologue 5(ALKBH5). FTO and ALKBH5 belong to α-ketoglutarate-dependent dioxygenase family and catalyze m^{6}A demethylation in Fe (II) and α-ketoglutaric acid-dependent manner. They mediate the demethylation process by oxidizing m^{6}A to form N^6-hydroxymethyladenosine (hm^{6}A), then converting hm^{6}A to N^6-formyladenosine (f^{6}A) and finally converting f^{6}A to adenosine [14]. FTO and ALKBH5 enzymes are neutral stable and can then be hydrolyzed into adenine. FTO is the first protein discovered to catalyze m^{6}A demethylation and is a member of the ALKB family with a highly conserved catalytic domain. ALKBH5 was shown to be an RNA demethylase that can oxidatively reverse m^{6}A modification. Unlike FTO, ALKBH5 with unique crystal structures is a specific demethylase during m^{6}A RNA methylation [15]. However, at present, there are relatively few demethylases, and some other components of m^{6}A methyltransferase remain to be verified. The mechanism to maintain dynamic balance with methylation and demethylation is not completely clear.

It is interesting that FTO has recently been found to preferentially target N^6, 2$'$-O-dimethyladenosine (m^{6}A$_m$) with its major target being m^{6}A$_m$ in small nuclear RNAs [16]. m^{6}A$_m$ is found at the 5$'$ end of mRNA, at the first encoded nucleotide adjacent to the 7-methylguanosine cap [17]. A recent study identified a cap-specific adenosine methyltransferase (CAPAM) was responsible for N^6-methylation of m^{6}A$_m$ and removed by FTO [18]. Thus, the methyltransferase to m^{6}A$_m$ is different from that of m^{6}A.

2.3. Readers. Methylated reader proteins mainly include members of YTH-domain families (YTHDF1, YTHDF2, YTHDF3, YTHDC1, and YTHDC2), heterogeneous ribonucleoproteins (HNRNPC, HNRNPG, and HNRNPA2B1), and insulin-like growth factor 2 mRNA-binding proteins (IGF2BPs). There is a conservative YTH domain with the properties of single-stranded RNA-binding proteins in members of YTH-domain family. YTHDF1 facilitates the translation of m^{6}A-methylated proteins by recruiting eukaryotic translation initiation factor 3 (eIF3) translation initiation complexes. YTHDF2 selectively binds to 3$'$UTR of m^{6}A and regulates mRNA degradation to affect mRNA stability. YTHDF3 promotes protein synthesis and translation through synergetic effects with YTHDF1 to augment YTHDF2-mediated mRNA degradation [19]. YTHDF1, YTHDF2, and YTHDF3 (DF1, DF2, and DF3) and all the DF paralogs regulate the same mRNAs based on the m^{6}A sites [20]. YTHDC1 is an m^{6}A reader protein mediated by RNA splicing. YTHDC2 interacts with RNA helicase to provide topological regulation of translation and extension [21]. HNRNPC and HNRNPG adjust splicing and abundance of mRNA after m^{6}A recognition. The phenomenon is called the "m^{6}A switch" [22]. In addition, IGF2BPs identify m^{6}A and enhance mRNA stability and translation in m^{6}A-dependent ways [23]. These methylated reader proteins may activate downstream regulatory pathways such as RNA degradation by identifying m^{6}A-modified critical sites during methylation.

3. Roles of m^{6}A Methylation in Skin Development

Skin with rich blood vessels and a complex structure is the most easily exposed organ to the external environment. Increasing evidence demonstrates that m^{6}A is closely related to skin development and regeneration. The mouse embryonic skin epithelium is initially a layer of pluripotent epithelial progenitor cells that later develop into three tissues consist of epidermis, hair follicles, and sebaceous glands. A research analyzed the potential effect of m^{6}A deficiency on mouse embryonic skin epithelial progenitor by single-cell RNA sequencing and functional studies. It demonstrated that both signaling and canonical translation pathways such as Wnt signal, actin regulators, cell polarity, extracellular matrix- (ECM-) receptor interaction, and NOTCH signal were significantly downregulated after m^{6}A loss [24]. Immunofluorescence imaging revealed that METTL3 deficiency in epidermal progenitor cells result in hair follicle morphology defection. After METTL3 knockout, translation initiation factors in mRNA were downregulated, while RNA metabolism and compensatory mechanisms were activated. Skin regeneration and wound repair in mammalians are maintained by epidermal progenitor cells [25, 26]. Genome editing and mouse genetics data reveal that deletion of m^{6}A methyltransferase impairs the skin's ability to self-renew and heal wounds. The above findings suggested that m^{6}A methylation is indispensable during skin development.
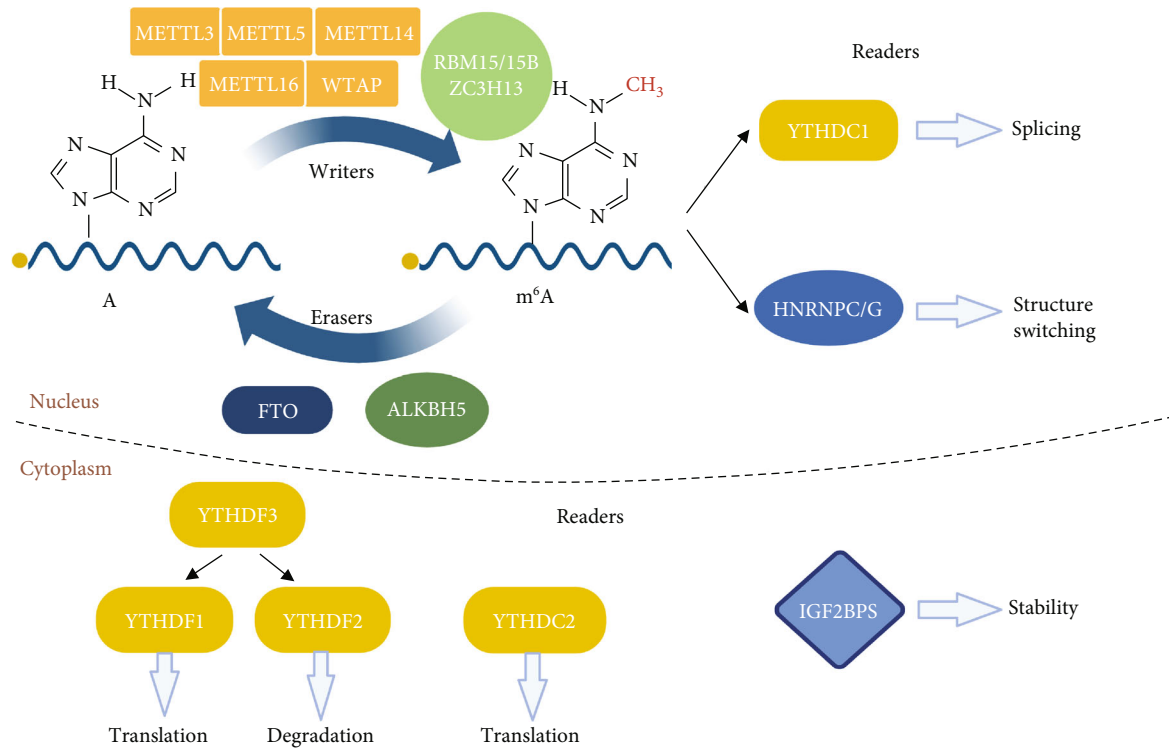
FIGURE 1: The types and roles of enzymes in dynamic modification of m^6A methylation. The m^6A RNA methylation is dynamically regulated by writers, erasers, and readers to add, remove, or recognize m^6A, respectively. Writers include METTL3, METTL5, METTL14, METTL16, WTAP, RBM15/15B, and ZC3H13. Erasers, FTO and ALKBH5, are two known demethylases. Readers are proteins that recognize the m^6A sites and perform multiple functions in nucleus or cytoplasm, including YTHDF1, YTHDF2, YTHDF3, YTHDC1, YTHDC2, HNRNPC, HNRNPG, and IGF2BPs. These enzymes are involved in RNA splicing, translation, structure switching, and RNA decay during m^6A RNA methylation.

Previous studies have discovered the possible mechanism of epitranscriptome dynamics occur during epidermal differentiation. The study demonstrated that plasmacytoma variant translocation 1 (Pvt1) methylation enhanced the interaction with myelocytomatosis oncogene (MYC) and stabilized MYC protein in epidermal progenitors. It identified m^6A methylation of Pvt1 as a key step in skin tissue homeostasis and wound repair [27]. In a recent study on skin development in goats, researchers verified 9085 m^6A sites with differential RNA methylation in cashmere fiber growth by methylated RNA immunoprecipitation followed by high-throughput sequencing (MeRIP-seq) and RNA-seq [28]. The results proved compelling evidence that these m^6A-modified genes were highly expressed in the skin tissue of goats. Keratins, the major components of the epithelial cytoskeleton, are responsible for maintaining the structural stability and integrity of keratinocytes, protecting cells from mechanical damage and affecting the formation of skin accessory organs. Furthermore, GO enrichment analysis showed that methylated genes were mainly involved in keratin filaments and intermediate filaments [29]. Therefore, these findings provide a theoretical basis for further research on the role of m^6A modification in skin development and growth. It proposes a possible strategy for wound healing via regulating m^6A modification.

4. Roles of m^6A Methylation in Skin Tumors

Many studies have shown that m^6A modification plays an important role in a variety of malignant tumors [30, 31]. The methyltransferase catalyzes the m^6A modification on oncogenes or tumor suppressor genes. m^6A reader proteins recognize corresponding markers through a series of biological effects, thereby to upregulate oncogene expression or downregulate tumor suppressor gene expression. On the other hand, the demethylase accelerates tumor progression via tumor-specific oncogenes [32]. These observations indicate that enhancing m^6A modification to regulate these genes may be a novel therapeutic strategy for cancer.

Among them, one group found that FTO was upregulated while m^6A RNA methylation was downregulated in arsenic-related human skin lesions. Moreover, FTO removal significantly inhibited arsenic-induced skin tumorigenesis [33]. Further m^6A-seq and RNA-seq analysis revealed that neural precursor cell-expressed developmentally downregulated 4 like (NEDD4L) was the critical target of FTO during m^6A RNA methylation, and FTO regulated NEDD4L mRNA stability via IGF2BPs. Another study revealed FTO-mediated dysregulation of mRNA m^6A methylation as an epitranscriptomic mechanism to promote arsenic tumorigenesis. Analysis of gene expression profiles showed higher expressions of m^6A effectors, especially METTL3, in

arsenic-exposed groups than low-arsenic-exposed populations. Further experiments found that arsenic promoted m^{6}A methylation by up-regulating METTL3 to increase the secretion of inflammatory factors including interleukin- (IL-) 6, IL-17, and IL-10 in human keratinocytes. Keratin 1 (KRT1) and keratin 10 (KRT10) reflecting skin injury were also significantly increased [34]. Another study demonstrated that METTL14 facilitated global genome repair (GGR) by regulating m^{6}A RNA methylation-mediated DNA damage-binding protein 2 (DDB2) translation and inhibits ultraviolet B- (UVB-) induced skin tumorigenesis. METTL14 knockout suppressed m^{6}A RNA methylation and translation in transcripts of DDB2 [35]. Therefore, METTL14 is an important specific autophagy target to regulate GGR and attenuate UVB-induced skin tumorgenesis.

4.1. Melanoma.
Melanoma is a serious and life-threatening skin malignancy [36]. About 75% of patients suffering skin tumor die of melanoma. Several m^{6}A regulators including METTL3, YTHDF1, HNRNPA2B1, FTO, and IGF2BP3 are involved in melanoma (Figure 2). Studies have shown that the expression of METTL3 is upregulated in human melanoma cell lines, and it promotes melanoma cell proliferation. Further research suggested METTL3 accelerated invasion and migration of melanoma cells by upregulating matrix metallopeptidase 2 (MMP2) [37]. In addition, the growth speed of YTHDF1-deficient melanoma cells was lower than that of wild type (WT) cells in mice. Moreover, the control rate of melanoma by immunotherapy in YTHDF1 knockout mice increased to nearly 100%, while the rate was only 40% in WT mice [38]. FTO was the first found RNA m^{6}A demethylase among the m^{6}A regulators. Studies have shown that FTO stimulates melanoma growth. FTO knockout increased m^{6}A methylation of genes including PD-1 (programmed cell death protein 1), CXCR4 (C-X-C motif chemokine receptor), and SOX10 (Sry-related HMG-box-10) in melanoma cells and thereby enhanced the susceptibility to interferon therapy [39]. METTL3 could modulate the mRNA level of uridine cytidine kinase 2 (UCK2) through m^{6}A modification to enhance its stability in melanoma. In addition, UCK2 enhanced the migration and invasion ability of melanoma cells by activating the Wnt/β-catenin pathway [40]. Both YTHDF1 and HNRNPA2B1 were upregulated in melanoma. The diagnosis sensitivity for melanoma with these two gene-combined screening increased approximately 10% compared to single-gene screening. The expression of p53 and associated genes including cyclin-dependent kinase 1 (CDK1) and CDK2 was positively correlated with upregulation of YTHDF1 or HNRNPA2B1 [41]. A three-gene prognostic marker including IGF2BP3, RBM15B, and METTL16 was constructed by analyzing the differently expression of m^{6}A regulatory factors in tumor samples and normal samples. It was demonstrated that IGF2BP3 promoted proliferation and migration of melanoma cells in vitro. The expression of IGF2BP3 was positively correlated with lymph node metastasis and immune cells infiltrating [42]. These results provided potential targets of novel drugs for melanoma.

Some researchers analyzed the gene expression profiles, copy number variation (CNV), and single-nucleotide polymorphism (SNP) data from The Cancer Genome Atlas (TCGA) databases. The study showed that 15 genes were changed on mRNA expression and m^{6}A regulatory proteins such as RBM15, YTHDF1, WTAP, and METTL14 genes in melanoma with a higher degree of genomic variation and a worse prognosis. Then, they identified a subset of molecules with immunological effects based on m^{6}A regulatory proteins [43]. The study provided a new and effective strategy for the treatment of cutaneous melanoma. The above results suggested that the regulatory proteins of m^{6}A methylation, including methylation on writing proteins, methylation reading proteins, and demethylases such as METTL3, YTHDF1, and FTO, play an important role in the development of melanoma. It provides a reliable theoretical basis for the treatment of melanoma and the development of drugs.

4.2. Cutaneous Squamous Cell Carcinoma.
Cutaneous squamous cell carcinoma (cSCC) is the second most common skin malignancy, accounting for 20% of nonmelanoma skin cancers. The most important physical factor causing the incidence of cSCC is sunlight ultraviolet rays especially UVB. Excessive sunlight exposure could lead to cell mutation and tumorgenesis. Studies have shown that exposure to UVB and arsenic resulted in upregulation of m^{6}A-binding protein YTHDF1 expression in skin keratinocytes, even skin mutation and oncogenesis to cause cSCC [44].

The m^{6}A methyltransferase METTL3 is also a key gene to regulate the occurrence of cSCC genesis. METTL3 was upregulated in cSCC, and METTL3 knockout disrupted the characteristics of stem cells in cSCC. METTL3 deficiency decreased colony-forming ability and suppressed tumorigenicity in vivo by reducing m^{6}A levels and ΔNp63 expression in cSCC. Exogenous ΔNp63 overexpression partially restored the proliferation of METTL3-knockout cSCC cells [45].

m^{6}A RNA methylation in cSCC was not studied as extensively as other related squamous cell carcinomas such as oral squamous cell carcinoma (OSCC), hepatocellular carcinoma (HCC), and gastric cancer (GC). Studies have shown that METTL3 promoted BMI1 translation in OSCC under the cooperation with m^{6}A reader IGF2BP1 and promoted OSCC proliferation and metastasis [46]. Combination of YTHDF1 silencing and epidermal factor growth receptor (EGFR) inhibition synergically suppressed the malignancies of HCC cells [47]. The expression profile variation of YTHDF1 was significantly associated with the high-risk subtype of GC patients, suggesting that YTHDF1 might be a potential target in GC early diagnosis [48]. These studies verified the detailed mechanism of m^{6}A methylation in cutaneous squamous cell carcinoma.

4.3. Merkel Cell Carcinoma (MCC).
MCC is a highly aggressive and rare skin cancer [49]. Merkel cell polyomavirus (MCPyV or MCV) can be detected in 80% of cases with MCC [50]. MCPyV is related to the animal tumor virus simian virus 40 (SV40). The differential splicing of the mRNA sequence produces the oncoprotein T antigen, which encodes the large T, small T, and 57 kT antigens [51]. One group used genomic screening of copy number aberrations
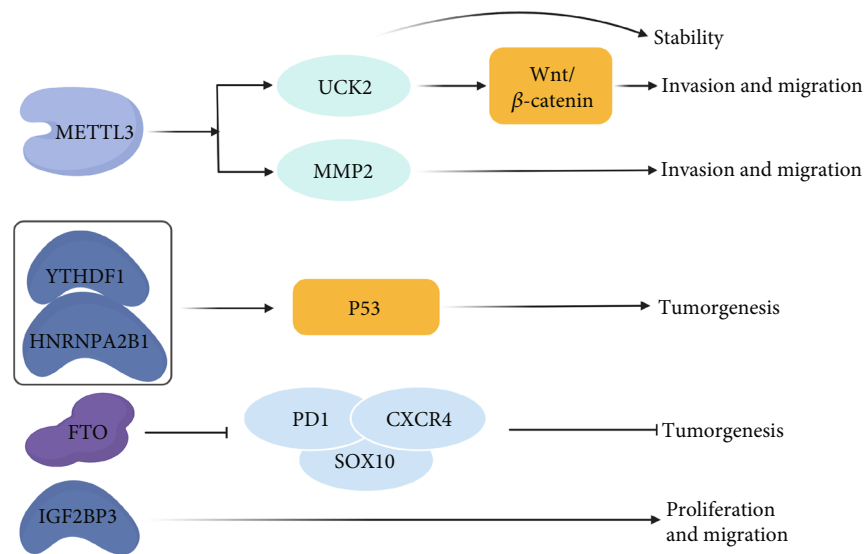
FIGURE 2: Mechanistic illustrations of the regulatory and functional roles of m⁶A in melanoma. Several m⁶A regulators including METTL3, YTHDF1, HNRNPA2B1, FTO, and IGF2BP3 are involved in melanoma. METTL3 increases mRNA level of UCK2 through m⁶A methylation to enhance its stability in melanoma. Then, UCK2 promotes the migration and invasion ability of melanoma cells by activating the Wnt/β-catenin pathway. METTL3 also accelerates invasion and migration of melanoma cells by elevating MMP2 expression. In addition, YTHDF1 or HNRNPA2B1 promotes p53 signal pathway via m⁶A methylation to tumorgenesis. FTO reduces m⁶A methylation to inhibit gene expressions including PD-1, CXCR4, and SOX10 but stimulate melanoma growth in melanoma cells. IGF2BP3 promotes proliferation and migration of melanoma cells.

along with transcriptomic analysis to investigate regions with amplification that harbor differentially expressed genes. They found that YTHDF1 was highly amplified and expressed in Merkel cell lines. YTHDF1 activated cap-dependent translation. Then, the translation initiation factors eIF3A and eIF3B cooperated synergistically, leading to high tumorigenicity. Moreover, YTHDF1 knockdown inhibited eIF3 to attenuate proliferative and clonal capacity. Survival data analysis showed that MCC patients with more YTHDF1 expression had lower overall survival compared with those with less YTHDF1. In other words, YTHDF1 level was negatively correlated with MCC patient prognosis and overall survival. Furthermore, the study identified m⁶A modifications on small T antigen mRNAs [52]. The study suggested a possible relationship between YTHDF1 amplification and MCPyV gene expression. YTHDF1 could be a novel prognostic marker for MCC.

In summary, m⁶A-binding protein FTO, METTL3, METTL14, and YTHDF1 expression were involved in skin lesion and oncogenesis. FTO promotes oncogenesis via IGF2BPs. METTL3 increases inflammatory factor secretion in human keratinocytes and enhanced ΔNp63 expression in cSCC. METTL14 facilitates GGR to suppress UVB-induced skin tumorigenesis. YTHDF1 activates translation initiation factors to high tumorigenicity in MCC (Figure 3).

5. Roles of m⁶A Modification in Inflammatory Skin Diseases

5.1. Psoriasis. Psoriasis is a common, complex, and chronic inflammatory disease that severely reduces patients' quality of life. The hallmark of psoriasis is uncontrolled proliferation and differentiation of keratinocytes by persistent inflammation. Previous studies have shown that methylated transcripts were mainly related to the Wnt signaling pathway in skin lesions of psoriasis patients. The Wnt gene family encodes a group of highly conserved secretory signal proteins involved in cell differentiation, proliferation, and immune-mediated inflammatory responses [53–55]. It is noted that m⁶A methylation regulated these key pathogenic processes including dendritic cell (DC) activation in psoriasis by altering Wnt gene expressions [56, 57]. Tumor necrosis factor- (TNF-) α/IL-23/Th17 axis is regarded as a key factor in inflammation exacerbation and lesion aggravation in psoriasis, being critical for the expansion of inflammation and aggravation of skin lesions in psoriasis vulgaris [58]. One study detected m⁶A methylation in psoriasis patients by MeRIP-Seq and RNA-Seq. The results showed that IL-17A and TNF-α, as two key genes of the TNF-α/IL-23/Th17 axis, are upregulated m⁶A methylation in skin lesions of psoriasis patients [59].

Additionally, the mRNA and protein levels of m⁶A writer WTAP were significantly increased in psoriasis patients especially in the epidermis. Overexpression of WTAP promoted the proliferation of keratinocytes, which may be possibly related to the upregulation of cyclinA2 and CDK2 [60]. The above findings suggest that m⁶A may be an important trans-epigenetic modifier to regulate proliferation, differentiation, and inflammation of psoriatic keratinocytes.

5.2. Atopic Dermatitis (AD). AD is a common and chronic inflammatory skin disease affecting approximately 10-25% of children and 7-10% of adults. It is characterized by disturbances in epidermal structure and keratinocyte differentiation, as well as excessive T cell-mediated inflammation
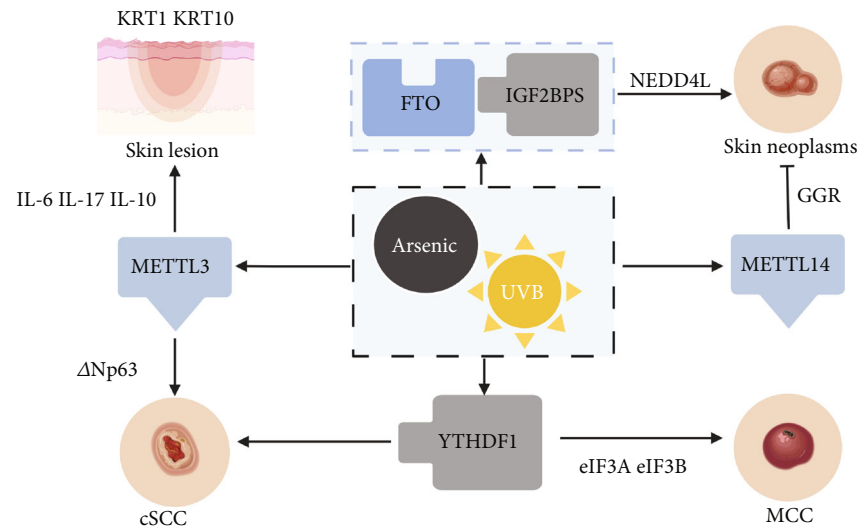
FIGURE 3: Process of m⁶A methylation induced by UVB and arsenic in skin. Several m⁶A regulators, including methylation on writing protein METTL3, methylation reading protein YTHDF1, and demethylase FTO, play an important role in the development of melanoma, even skin mutation and canceration to causing cSCC especially after UVB and arsenic exposure. FTO regulates NEDD4L mRNA stability to promote skin neoplasms via IGF2BPs. METTL3 increases inflammatory factor secretion including IL-6, IL-17, and IL-10 in human keratinocytes; then, KRT1 and KRT10 reflecting skin injury are also significantly elevated. METTL3 also enhanced ΔNp63 expression in cSCC. METTL14 facilitates GGR to suppress UVB-induced skin tumorigenesis. YTHDF1 activates cap-dependent translation. Then, the translation initiation factors eIF3A and eIF3B cooperate synergistically to possibly lead to high tumorigenicity in MCC.

with a complex genetic condition [61]. The pathogenesis is related to mutations in genes encoding epidermal structural proteins, barrier enzymes, and their inhibitors. Recent studies demonstrated a critical role of epigenetic changes in AD [62]. One study demonstrated that circRNAs regulated macrophage-mediated inflammation via analyzing differential expression profile of circRNAs by circRNA microarray. Further results also showed that Hsa_circ_0004287 reduced the stability of MALAT1 by competitively binding to IGF2BP3 and MALAT1 in an m⁶A-dependent manner in 2,4-dinitrochlorobenzene- (DNCB-) induced dermatitis. Low levels of MALAT1 promoted ubiquitinated degradation of S100A8/S100A9, thereby prevented p38/mitogen-activated protein kinase phosphorylation and macrophage-mediated inflammation, and finally inhibited M1 macrophage activation [63]. The existing studies on m⁶A RNA methylation in atopic dermatitis proposed a possible role of macrophage-mediated inflammation in disease progression.

6. Roles of m⁶A Modification in Other Skin Diseases

Scleroderma is an autoimmune connective tissue characterized by organ fibrosis, immune abnormalities, and vascular damage [64]. Although scleroderma is a rare disease with a low prevalence and incidence, it has a high mortality rate [65]. A recent study identified m⁶A-tagged mRNAs in a mouse model of bleomycin-induced scleroderma by m⁶A supratranscriptomic microarrays and m⁶A RNA immunoprecipitation qPCR. The results showed that there were differences in the m⁶A methylation of 843 mRNAs and include the methylation and the expression of Hras, Saa1, Ccl3, Ccl9,

and Il1b [66]. This study revealed the involvement of m⁶A methylation in scleroderma.

Cutaneous T-cell lymphomas (CTCL) are a rare type of non-Hodgkin lymphoma characterized by cutaneous infiltration of malignant lymphocytes [67]. Recent studies have uncovered a role for METTL3-mediated m⁶A modification in CTCL progression. The results showed that METTL3 significantly downregulated CTCL cells both *in vivo* and *in vitro*. Furthermore, METTL3 small interfering RNAs and RIP assay indicated that cyclin-dependent kinase inhibitor 2A (CDKN2A) was a key regulator in CTCL, and insufficient methylation blocked the interaction between CDKN2A and m⁶A reader IGF2BP2, resulting in mRNA degradation [68]. This is the first study to describe the role of m⁶A in CTCL development and provide a potential biological target for therapy.

Sequestosome 1 (SQSTM1) is a vital protein marker serving as a selective autophagy receptor [69]. SQSTM1/p62 was significantly downregulated in the epidermis of diabetic patients and in the db/db mouse model with chronic hyperglycemia. Knockdown of SQSTM1 led to the impairment of autophagic flux. In addition, the m⁶A reader protein YTHDC1 interacting with SQSTM1 mRNA was attenuated in keratinocytes during hyperglycemia. *In vivo*, knockout of endogenous YTHDC1 or SQSTM1 suppressed epidermal autophagy and impaired the biological functions of keratinocytes including promoted apoptosis and delayed wound healing. The results showed that YTHDC1 interacted with ELAV-like RNA-binding protein 1/HuR (ELAVL1) and cooperatively regulated the expression of SQSTM1 [70]. This study reveals that YTHDC1 regulated autophagy by regulating the stability of SQSTM1 nuclear mRNA in diabetic keratinocytes.

One group described m⁶A modification patterns in hyperplasia scars (HS) and normal skin (NS) tissues by m⁶A sequencing and RNA sequencing. Several targets of m⁶A-associated RNA were immunoprecipitated and verified by real-time quantitative PCR. The results showed that 14791 new m⁶A peaks appeared in the HS samples, while 7835 peaks disappeared. It suggested that the m⁶A-related genes in HS are associated with fibrosis-related pathways. In addition, they identified differentially expressed mRNA transcripts in HS samples with hypermethylated or hypomethylated m⁶A peaks [71]. This study maps the m⁶A epitranscriptome of human HS, which may help to elucidate the possible mechanisms of m⁶A-mediated regulation of gene expression.

7. Limitations and Challenges of Current Research on m⁶A Methylation

There is no study to exactly confirm the specific mechanism in dynamic m⁶A regulation in skin disease. Only a few studies were published on the mechanisms that control the expressions and activities of m⁶A "Writers," "Erasers," and "Readers" in the skin, especially the "reader." Previous studies have confirmed that m⁶A methylase has dual effects of cancer promoting or suppressing. METTL3 serves as an apoptosis driver in high glucose-reliant human lens epithelial cells (HLEC), while WTAP acts as an inhibitor in human natural killer/T cell lymphoma (NKTCL) cell lines [13, 27]. This study demonstrates the existence of a two-way regulatory mechanism for methylation. Therefore, the research on the roles of m⁶A methylation in skin tumors and skin inflammatory diseases still needs to be further explored.

The total amount of m⁶A in RNA can be probed by several methods, including two-dimensional thin layer chromatography [72], m⁶A dot-blot [73], and HPLC-MS/MS (high-performance liquid chromatography-tandem mass spectrometry) [74]. However, these approaches are not suitable for high throughput identification and localization of modified sites. MeRIP-seq, also known as m⁶A-seq (m⁶A RNA immunoprecipitation sequencing), relies on RNA fragmentation but incapable of single-nucleotide resolution detection in methylation sites [75]. Although various methods have been found currently, there are still so many challenges and difficulties to detect m⁶A methylation. Some more effective methods especially in the skin are urgently needed.

There are already some drugs targeting methylations. The nonsteroidal anti-inflammatory agent meclofenamic (MA) has a strong inhibitory effect on FTO and is expected to become a demethylase inhibitor. MA2, an ethylester derivative of MA is hydrolyzed inside the cell to its active form MA. MA2 increased m⁶A methylation in HeLa cells and had better cellular permeability than MA [76, 77]. However, these drugs have not been tested in clinic for their effectiveness. It is well-known that immunotherapy plays a significant role in the treatment of skin diseases such as melanoma. Various studies have confirmed that m⁶A methylation regulates innate immune response [56, 77]. It was demonstrated that m⁶A methylation was a crucial regulator of T cell homeostasis. T cells carrying m⁶A modifying agents may be an effective target for the treatment of autoimmune diseases [78]. These studies showed the potential application of this mechanism in immunotherapy. Although they can provide potential pharmacological targets, the clinical effects should be further verified.

8. Conclusions

Taken together, m⁶A is dynamically regulated in many physiological and pathological processes of dermatological diseases. RNA methylation is regulated by methylase, demethylase, and methylation reader proteins, and the process is reversible. RNA methylation is a relatively new field. In recent years, m⁶A has been prominently discovered in various skin diseases. However, many unknowns still exist in the epitranscriptome modification of RNA. The detection method of m⁶A methylation needs improvement. The efficacy of methylation-modifying agents needs further clinical studies. Epitranscriptome modification has attracted more and more attention in regulating immune response and immunotherapy. Exploring key epitranscriptome variation sites and developing corresponding immunotherapy drugs have become research hotspots and have good development prospects.

Data Availability

All data are available on request.

Conflicts of Interest

The authors declare that they have no competing interests.

Authors' Contributions

All authors had made substantial contributions for this manuscript. Chang Liu and Xin Wang contribute equally to this work and are co-first authors.

Acknowledgments

References

[1] D. P. Patil, C. K. Chen, B. F. Pickering et al., "m⁶A RNA methylation promotes *XIST*-mediated transcriptional repression," *Nature*, vol. 537, no. 7620, pp. 369–373, 2016.

[2] K. D. Meyer, Y. Saletore, P. Zumbo, O. Elemento, C. E. Mason, and S. R. Jaffrey, "Comprehensive analysis of mRNA methylation reveals enrichment in 3′ UTRs and near stop codons," *Cell*, vol. 149, no. 7, pp. 1635–1646, 2012.

[3] Y. Pan, P. Ma, Y. Liu, W. Li, and Y. Shu, "Multiple functions of m⁶A RNA methylation in cancer," *Journal of Hematology & Oncology*, vol. 11, no. 1, p. 48, 2018.

[4] K. J. Yoon, F. R. Ringeling, F. Jacob et al., "Temporal Control of Mammalian Cortical Neurogenesis by m⁶A Methylation," *Cell*, vol. 171, no. 4, pp. 877–889, 2017.

[5] Y. Wang, Y. Li, J. I. Toth, M. D. Petroski, Z. Zhang, and J. C. Zhao, "N^6-methyladenosine modification destabilizes developmental regulators in embryonic stem cells," *Nature Cell Biology*, vol. 16, no. 2, pp. 191–198, 2014.

[6] F. Yu, J. Wei, X. Cui et al., "Post-translational modification of RNA m6A demethylase ALKBH5 regulates ROS-induced DNA damage response," *Nucleic Acids Research*, vol. 49, no. 10, pp. 5779–5797, 2021.

[7] J. Zhou, J. Wan, X. Gao, X. Zhang, S. R. Jaffrey, and S. B. Qian, "Dynamic m6A mRNA methylation directs translational control of heat shock response," *Nature*, vol. 526, no. 7574, pp. 591–594, 2015.

[8] J. Zhou, J. Wang, B. Hong et al., "Gene signatures and prognostic values of m6A regulators in clear cell renal cell carcinoma - a retrospective study using TCGA database," *Aging*, vol. 11, no. 6, pp. 1633–1647, 2019.

[9] R. Su, L. Dong, Y. Li et al., "METTL16 exerts an m6A-independent function to facilitate translation and tumorigenesis," *Nature Cell Biology*, vol. 24, no. 2, pp. 205–216, 2022.

[10] X. Wang, J. Huang, T. Zou, and P. Yin, "Human m6A writers: two subunits, 2 roles," *RNA Biology*, vol. 14, no. 3, pp. 300–304, 2017.

[11] H. Shima, M. Matsumoto, Y. Ishigami et al., "S-adenosyl-methionine synthesis is regulated by selective N^6-adenosine methylation and mRNA degradation involving METTL16 and YTHDC1," *Cell Reports*, vol. 21, no. 12, pp. 3354–3363, 2017.

[12] P. Knuckles, T. Lence, I. U. Haussmann et al., "Zc3h13/Flacc is required for adenosine methylation by bridging the mRNA-binding factor Rbm15/Spenito to the m6A machinery component Wtap/Fl^{2}d," *Genes & Development*, vol. 32, no. 5-6, pp. 415–429, 2018.

[13] J. Wen, R. Lv, H. Ma et al., "Zc3h13 regulates nuclear RNA m6A methylation and mouse embryonic stem cell SELF-RENEWAL," *Molecular Cell*, vol. 69, no. 6, pp. 1028–1038.e6, 2018.

[14] J. D. W. Toh, S. W. M. Crossley, K. J. Bruemmer et al., "Distinct RNA N-demethylation pathways catalyzed by nonheme iron ALKBH5 and FTO enzymes enable regulation of formaldehyde release rates," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 117, no. 41, pp. 25284–25292, 2020.

[15] H. Huang, H. Weng, and J. Chen, "m6A modification in coding and non-coding RNAs: roles and therapeutic implications in cancer," *Cancer Cell*, vol. 37, no. 3, pp. 270–288, 2020.

[16] S. Zaccara, R. J. Ries, and S. R. Jaffrey, "Reading, writing and erasing mRNA methylation," *Nature Reviews Molecular Cell Biology*, vol. 20, no. 10, pp. 608–624, 2019.

[17] J. Mauer, X. Luo, A. Blanjoie et al., "Reversible methylation of m^6A_m in the 5′ cap controls mRNA stability," *Nature*, vol. 541, no. 7637, pp. 371–375, 2017.

[18] S. Akichika, S. Hirano, Y. Shichino et al., "Cap-specific terminal N^6-methylation of RNA by an RNA polymerase II-associated methyltransferase," *Science*, vol. 363, no. 6423, 2019.

[19] X. Wang, Z. Lu, A. Gomez et al., "N^6-methyladenosine-dependent regulation of messenger RNA stability," *Nature*, vol. 505, no. 7481, pp. 117–120, 2014.

[20] S. Zaccara and S. R. Jaffrey, "A Unified Model for the Function of YTHDF Proteins in Regulating m6A-Modified mRNA," *Cell*, vol. 181, no. 7, pp. 1582–1595.e18, 2020.

[21] Y. Mao, L. Dong, X. M. Liu et al., "m6A in mRNA coding regions promotes translation via the RNA helicase-containing YTHDC2," *Nature Communications*, vol. 10, no. 1, p. 5332, 2019.

[22] N. Liu, K. I. Zhou, M. Parisien, Q. Dai, L. Diatchenko, and T. Pan, "N6-methyladenosine alters RNA structure to regulate binding of a low-complexity protein," *Nucleic Acids Research*, vol. 45, no. 10, pp. 6051–6063, 2017.

[23] H. Huang, H. Weng, W. Sun et al., "Recognition of RNA N^6-methyladenosine by IGF2BP proteins enhances mRNA stability and translation," *Nature Cell Biology*, vol. 20, no. 3, pp. 285–295, 2018.

[24] L. Xi, T. Carroll, I. Matos et al., "m6A RNA methylation impacts fate choices during skin morphogenesis," *eLife*, vol. 9, article e56980, 2020.

[25] D. Belokhvostova, I. Berzanskyte, A. M. Cujba et al., "Homeostasis, regeneration and tumour formation in the mammalian epidermis," *International Journal of Developmental Biologyl*, vol. 62, no. 6-7-8, pp. 571–582, 2018.

[26] A. N. Vagnozzi, J. F. Reiter, and S. Y. Wong, "Hair follicle and interfollicular epidermal stem cells make varying contributions to wound regeneration," *Cell Cycle*, vol. 14, no. 21, pp. 3408–3417, 2015.

[27] H. Ma, L. Shen, H. Yang, H. Gong, X. Du, and J. Li, "M6A methyltransferase Wilms' tumor 1-associated protein facilitates cell proliferation and cisplatin resistance in NK/T cell lymphoma by regulating dual-specificity phosphatases 6 expression via m6A RNA methylation," *IUBMB Life*, vol. 73, no. 1, pp. 108–117, 2021.

[28] Y. Wang, Y. Zheng, D. Guo et al., "m6A methylation analysis of differentially expressed genes in skin tissues of coarse and fine type Liaoning cashmere goats," *Frontiers in Genetics*, vol. 10, p. 1318, 2020.

[29] X. Zhang, M. Yin, and L. J. Zhang, "Keratin 6, 16 and 17-critical barrier alarmin molecules in skin wounds and psoriasis," *Cell*, vol. 8, no. 8, p. 807, 2019.

[30] L. He, H. Li, A. Wu, Y. Peng, G. Shu, and G. Yin, "Functions of N6-methyladenosine and its role in cancer," *Molecular Cancer*, vol. 18, no. 1, p. 176, 2019.

[31] J. Liu, B. T. Harada, and C. He, "Regulation of gene expression by N^6-methyladenosine in cancer," *Trends in Cell Biology*, vol. 29, no. 6, pp. 487–499, 2019.

[32] T. Sun, R. Wu, and L. Ming, "The role of m6A RNA methylation in cancer," *Biomedicine & Pharmacotherapy*, vol. 112, article 108613, 2019.

[33] Y. H. Cui, S. Yang, J. Wei et al., "Autophagy of the m6A mRNA demethylase FTO is impaired by low-level arsenic exposure to promote tumorigenesis," *Nature Communications*, vol. 12, no. 1, p. 2183, 2021.

[34] F. Yang and A. Zhang, "Role of N6-methyladenosine RNA modification in the imbalanced inflammatory homeostasis of arsenic-induced skin lesions," *Environmental Toxicology*, vol. 37, no. 8, pp. 1831–1839, 2022.

[35] Z. Yang, S. Yang, Y. H. Cui et al., "METTL14 facilitates global genome repair and suppresses skin tumorigenesis," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 118, no. 35, article e2025948118, 2021.

[36] P. A. Ascierto and A. M. M. Eggermont, "Neoadjuvant therapy in melanoma: the next step?," *The Lancet Oncology*, vol. 19, no. 2, pp. 151–153, 2018.

[37] U. Dahal, K. Le, and M. Gupta, "RNA m6A methyltransferase METTL3 regulates invasiveness of melanoma cells by matrix metallopeptidase 2," *Melanoma Research*, vol. 29, no. 4, pp. 382–389, 2019.

[38] D. Han, J. Liu, C. Chen et al., "Anti-tumour immunity controlled through mRNA m6A methylation and YTHDF1 in dendritic cells," *Nature*, vol. 566, no. 7743, pp. 270–274, 2019.

[39] S. Yang, J. Wei, Y. H. Cui et al., "m6A mRNA demethylase FTO regulates melanoma tumorigenicity and response to anti-PD-1 blockade," *Nature Communications*, vol. 10, no. 1, p. 2782, 2019.

[40] H. Wu, H. Xu, D. Jia, T. Li, and L. Xia, "METTL3-induced UCK2 m6A hypermethylation promotes melanoma cancer cell metastasis via the WNT/β-catenin pathway," *Annals of Translational Medicine*, vol. 9, no. 14, p. 1155, 2021.

[41] T. Li, M. Gu, A. Deng, and C. Qian, "Increased expression of YTHDF1 and HNRNPA2B1 as potent biomarkers for melanoma: a systematic analysis," *Cancer Cell International*, vol. 20, no. 1, p. 239, 2020.

[42] J. Liu, Z. Zhou, L. Ma et al., "Effects of RNA methylation N6-methyladenosine regulators on malignant progression and prognosis of melanoma," *Cancer Cell International*, vol. 21, no. 1, p. 453, 2021.

[43] Y. Lin, S. Wang, S. Liu, S. Lv, H. Wang, and F. Li, "Identification and verification of molecular subtypes with enhanced immune infiltration based on m6A regulators in cutaneous melanoma," *BioMed Research International*, vol. 2021, Article ID 2769689, 19 pages, 2021.

[44] Y. Sang and Y. Deng, "Current insights into the epigenetic mechanisms of skin cancer," *Dermatologic Therapy*, vol. 32, no. 4, article e12964, 2019.

[45] R. Zhou, Y. Gao, D. Lv, C. Wang, D. Wang, and Q. Li, "METTL3 mediated m6A modification plays an oncogenic role in cutaneous squamous cell carcinoma by regulating ΔNp63," *Biochemical and Biophysical Research Communications*, vol. 515, no. 2, pp. 310–317, 2019.

[46] L. Liu, Y. Wu, Q. Li et al., "METTL3 promotes tumorigenesis and metastasis through BMI1 m6A methylation in oral squamous cell carcinoma," *Molecular Therapy : The Journal of the American Society of Gene Therapy*, vol. 28, no. 10, pp. 2177–2190, 2020.

[47] T. Su, M. Huang, J. Liao et al., "Insufficient radiofrequency ablation promotes hepatocellular carcinoma metastasis through N6-methyladenosine mRNA methylation-dependent mechanism," *Hepatology*, vol. 74, no. 3, pp. 1339–1356, 2021.

[48] T. Liu, S. Yang, Y. P. Cheng et al., "The N6-methyladenosine (m6A) methylation gene *YTHDF1* reveals a potential diagnostic role for gastric cancer," *Cancer Management and Research*, vol. 12, pp. 11953–11964, 2020.

[49] D. Schadendorf, C. Lebbé, A. Zur Hausen et al., "Merkel cell carcinoma: epidemiology, prognosis, therapy and unmet medical needs," *European Journal of Cancer*, vol. 71, pp. 53–69, 2017.

[50] H. Feng, M. Shuda, Y. Chang, and P. S. Moore, "Clonal integration of a polyomavirus in human Merkel cell carcinoma," *Science*, vol. 319, no. 5866, pp. 1096–1100, 2008.

[51] M. Shuda, H. J. Kwun, H. Feng, Y. Chang, and P. S. Moore, "Human Merkel cell polyomavirus small T antigen is an oncoprotein targeting the 4E-BP1 translation regulator," *The Journal of Clinical Investigation*, vol. 121, no. 9, pp. 3623–3634, 2011.

[52] E. Orouji, W. K. Peitsch, A. Orouji, R. Houben, and J. Utikal, "Oncogenic role of an epigenetic reader of m6A RNA modification: YTHDF1 in Merkel cell carcinoma," *Cancers*, vol. 12, no. 1, p. 202, 2020.

[53] R. A. Pollock, F. Abji, and D. D. Gladman, "Epigenetics of psoriatic disease: a systematic review and critical appraisal," *Journal of Autoimmunity*, vol. 78, pp. 29–38, 2017.

[54] N. Irrera, A. Bitto, M. Vaccaro et al., "PDRN, a bioactive natural compound, ameliorates imiquimod-induced psoriasis through NF-κB pathway inhibition and Wnt/β-catenin signaling modulation," *International Journal of Molecular Sciences*, vol. 21, no. 4, p. 1215, 2020.

[55] W. Wang, X. Yu, C. Wu, and H. Jin, "IL-36γ inhibits differentiation and induces inflammation of keratinocyte via Wnt signaling pathway in psoriasis," *International Journal of Medical Sciences*, vol. 14, no. 10, pp. 1002–1007, 2017.

[56] Y. N. Wang, C. Y. Yu, and H. Z. Jin, "RNA N^6-methyladenosine modifications and the immune response," *Journal of Immunology Research*, vol. 2020, Article ID 6327614, 6 pages, 2020.

[57] H. Wang, X. Hu, M. Huang et al., "Mettl3-mediated mRNA m6A methylation promotes dendritic cell activation," *Nature Communications*, vol. 10, no. 1, p. 1898, 2019.

[58] A. Rendon and K. Schäkel, "Psoriasis pathogenesis and treatment," *International Journal of Molecular Sciences*, vol. 20, no. 6, p. 1475, 2019.

[59] Y. N. Wang and H. Z. Jin, "Transcriptome-wide m6A methylation in skin lesions from patients with psoriasis vulgaris," *Frontiers in Cell and Developmental Biology*, vol. 8, article 591629, 2020.

[60] Y. Kong, R. Wu, S. Zhang et al., "Wilms' tumor 1-associating protein contributes to psoriasis by promoting keratinocytes proliferation via regulating cyclinA2 and CDK2," *International Immunopharmacology*, vol. 88, article 106918, 2020.

[61] L. Paternoster, M. Standl, J. Waage et al., "Multi-ancestry genome-wide association study of 21,000 cases and 95,000 controls identifies new risk loci for atopic dermatitis," *Nature Genetics*, vol. 47, no. 12, pp. 1449–1456, 2015.

[62] B. Nedoszytko, E. Reszka, D. Gutowska-Owsiak et al., "Genetic and epigenetic aspects of atopic dermatitis," *International Journal of Molecular Sciences*, vol. 21, no. 18, p. 6484, 2020.

[63] L. Yang, J. Fu, X. Han et al., "*Hsa_circ_0004287* inhibits macrophage-mediated inflammation in an N^6-methyladenosine -dependent manner in atopic dermatitis and psoriasis," *The Journal of Allergy and Clinical Immunology*, vol. 149, no. 6, pp. 2021–2033, 2022.

[64] C. P. Denton and D. Khanna, "Systemic sclerosis," *The Lancet*, vol. 390, no. 10103, pp. 1685–1699, 2017.

[65] J. G. Royle, P. C. Lanyon, M. J. Grainge, A. Abhishek, and F. A. Pearce, "The incidence, prevalence, and survival of systemic sclerosis in the UK clinical practice research datalink," *Clinical Rheumatology*, vol. 37, no. 8, pp. 2103–2111, 2018.

[66] L. Shen, Y. Yu, M. Jiang, and J. Zhao, "Alteration of the m6A methylation landscape in a mouse model of scleroderma," *Epigenomics*, vol. 13, no. 23, pp. 1867–1883, 2021.

[67] N. Bagherani and B. R. Smoller, "An overview of cutaneous T cell lymphomas," *F1000Research*, vol. 5, 2016.

[68] X. Wang, M. Hu, L. Yu et al., "The "m6A writer" METTL3 and the "m6A reader" IGF2BP2 regulate cutaneous T-cell lymphomas progression via CDKN2A," *Hematological Oncology*, vol. 40, no. 4, pp. 567–576, 2022.

[69] V. Rogov, V. Dötsch, T. Johansen, and V. Kirkin, "Interactions between autophagy receptors and ubiquitin-like proteins form the molecular basis for selective autophagy," *Molecular Cell*, vol. 53, no. 2, pp. 167–178, 2014.

[70] D. Liang, W. J. Lin, M. Ren et al., "m^{6}A reader YTHDC1 modulates autophagy by targeting SQSTM1 in diabetic skin," *Autophagy*, vol. 18, no. 6, pp. 1318–1337, 2022.

[71] J. Yang, J. Liu, S. Zhao, and F. Tian, "N^6-methyladenosine METTL3 modulates the proliferation and apoptosis of lens epithelial cells in diabetic cataract," *Molecular Therapy Nucleic Acids*, vol. 20, pp. 111–116, 2020.

[72] Y. Fu, G. Z. Luo, K. Chen et al., "$N\,^6$-Methyldeoxyadenosine marks active transcription start sites in *Chlamydomonas*," *Cell*, vol. 161, no. 4, pp. 879–892, 2015.

[73] A. Nagarajan, R. Janostiak, and N. Wajapeyee, "Dot blot analysis for measuring global N(6)-methyladenosine modification of RNA," *Methods in Molecular Biology*, vol. 1870, pp. 263–271, 2019.

[74] W. Xiao, S. Adhikari, U. Dahal et al., "Nuclear m^{6}A reader YTHDC1 regulates mRNA splicing," *Molecular Cell*, vol. 61, no. 4, pp. 507–519, 2016.

[75] Y. Niu, A. Wan, Z. Lin, X. Lu, and G. Wan, "$N\,^6$-Methyladenosine modification: a novel pharmacological target for anticancer drug development," *Acta Pharmaceutica Sinica B*, vol. 8, no. 6, pp. 833–843, 2018.

[76] Y. Huang, J. Yan, Q. Li et al., "Meclofenamic acid selectively inhibits FTO demethylation of m^{6}A over ALKBH5," *Nucleic Acids Research*, vol. 43, no. 1, pp. 373–384, 2015.

[77] C. Zhang, J. Fu, and Y. Zhou, "A review in research progress concerning m6A methylation and immunoregulation," *Frontiers in Immunology*, vol. 10, p. 922, 2019.

[78] H. B. Li, J. Tong, S. Zhu et al., "m^{6}A mRNA methylation controls T cell homeostasis by targeting the IL-7/STAT5/SOCS pathways," *Nature*, vol. 548, no. 7667, pp. 338–342, 2017.