Review Article

The Role of Posttranslational Modifications in DNA Repair

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The human body is a complex structure of cells, which are exposed to many types of stress. Cells must utilize various mechanisms to protect their DNA from damage caused by metabolic and external sources to maintain genomic integrity and homeostasis and to prevent the development of cancer. DNA damage inevitably occurs regardless of physiological or abnormal conditions. In response to DNA damage, signaling pathways are activated to repair the damaged DNA or to induce cell apoptosis. During the process, posttranslational modifications (PTMs) can be used to modulate enzymatic activities and regulate protein stability, protein localization, and protein-protein interactions. Thus, PTMs in DNA repair should be studied. In this review, we will focus on the current understanding of the phosphorylation, poly(ADP-ribosyl)ation, ubiquitination, SUMOylation, acetylation, and methylation of six typical PTMs and summarize PTMs of the key proteins in DNA repair, providing important insight into the role of PTMs in the maintenance of genome stability and contributing to reveal new and selective therapeutic approaches to target cancers.

1. Introduction

Base and nucleotide damage can be induced by changes in the external environment and by the products of cellular metabolism, such as reactive oxygen species. Single- or double-strand DNA breaks can be directly caused by ionizing radiation (IR) [1] or genotoxic agents [2]. To avoid or cope with the propagation of damaged DNA during cell division, eukaryotic cells utilize a complex and versatile network of mechanisms known as DDR [3]. When DNA damage is limited, the DDR mechanism detects the damage signal and activates the related signaling pathways involved in the relaxation of the chromatin structure. This action enables repair factors to examine and repair the damaged DNA sites. Furthermore, cell cycle arrest is promoted to enable DNA repair to occur. When the double-strand breaks (DSBs) cannot be repaired, the DDR will activate apoptosis-related signaling pathways, and the cell undergoes cell death [4]. However, if the DSBs are misrepaired, chromosomal translocations will occur and genomic instability will be affected [5].

Correspondingly, DNA-associated activities, such as replication or transcription, will be affected and result in mutation. DNA repair is a complex process that requires a series of damage and repair factors. These factors are sequentially recruited to the damage sites based on a specific order. Organisms have acquired many systematic and complex modes to repair damaged DNA, including direct repair, base excision repair (BER), nucleotide excision repair (NER), homologous recombination (HR), and SOS repair. Direct repair almost always plays a role in lower organisms [6]. DNA base and nucleotide damages induced by oxides or alkylates and spontaneous depurination are repaired by the BER pathway and the NER pathways [7]. The most serious form of DNA damage is DSBs, which is considered the most lethal form of DNA damage, which can be induced by exogenous agents, such as IR or topoisomerase inhibitor poisons. DSBs are repaired by HR and NHEJ [8]. These two DSB repair pathways act on different cell cycle phases: HR primarily acts in the S and G2 phases, whereas NHEJ primarily acts in the G1 phase.
SOS repair is a kind of DNA repair that occurs under extreme conditions and the repair consequence may leave many errors to maintain genome integrity. SOS repair is also called error-prone repair, and this type of repair is widely present in bacteria [10].

PTMs involve a series of transient, reversible covalent modifications of amino acidic residues, such as serine and threonine phosphorylation, lysine acetylation and ubiquitylation, and lysine and arginine methylation. PTMs have become increasingly important in DDR because they have the ability to alter protein activity (without requiring de novo protein synthesis) by attaching small molecules to substrate protein. PTMs of proteins play a crucial role in the first phase of DDR by mediating protein-protein interactions and regulating protein trafficking, localization, activity, and stability [11].

In this review, we discuss the PTMs of key proteins in base and nucleotide excision repair and double-strand break repair pathways (NHEJ and HR) by focusing on the best-known PTMs: phosphorylation, poly(ADP-ribosyl)ation, ubiquitination, SUMOylation, acetylation, and methylation (Figure 1).

1.1. Phosphorylation. Phosphorylation is ubiquitous and widely used posttranslational modification and is mediated by kinases attaching phosphate groups to the serine, threonine, histidine, or tyrosine residues of a substrate protein. Phosphorylation may occur at multiple amino acids. In signaling pathways, signal transduction from upstream sensors to downstream effectors occurs via the phosphorylation of a series of proteins, e.g., a first kinase phosphorylates and activates a second kinase, which then phosphorylates and activates a third kinase and so on. Thus, cellular responses are based on this series of signals; phosphorylation plays a major role in the regulation of enzyme activity [12]. Phosphorylation is also a reversible process. The dissociation of a phosphate group from a substrate protein is termed dephosphorylation. Phosphorylation and dephosphorylation have been involved in protein interaction [13], cell growth, differentiation, apoptosis, cell signaling in health condition and tumorigenesis [14], and so on.

1.1.1. Phosphorylation in Base and Nucleotide Excision Repair/Single-Strand Break Repair. Upon DNA base damages, the necessary repairs for these damages would require a BER/single-strand break repair (SSBR) system. X-ray cross-complementing protein 1 (XRCC1), a scaffold protein of the BER/SSBR system, functions as a component of a complex with DNA ligase IIIa (Lig III), which is involved in the ligation steps of BER and DNA single-strand repairs [15]. It also interacts with several other BER/SSBR factors and forms dense foci in the nucleus after DNA damage [16–18]. The formation of foci is critical for DNA repair. The phosphorylation of XRCC1 in chromatin has been reported to facilitate the BER incision step by promoting its dissociation from DNA [19]. The phosphorylation of XRCC1 in chromatin and the DNA damage-induced recruitment of XRCC1 to the nuclear matrix are critical for foci formation [20]. Chou et al. demonstrated that the phosphorylation of XRCC1 by Ataxia Telangiectasia Mutated-Checkpoint kinase 2 (ATM-Chk2) promoted BER by recruiting downstream BER proteins to the initial damage/incision step [21]. Moreover, BER and the stability of XRCC1 can be enhanced by phosphorylating serine 518 and threonine 519 and 523 by casein kinase 2 (CK2) [22]. In addition to XRCC1, several histones are also phosphorylated in BER, such as the H2AX histone.
variant at serine 139 (S139) and histone H2A at serine residues S2 (H2A-S2), S18 (H2A-S18), and S122 (H2A-S122) upon UV irradiation [23]. However, two histone residues of H3, serine 10 (S10), and threonine 11 (T11) are phosphorylated during NER and dephosphorylated by UV irradiation [24]. Thus, there is not a fixed pattern for histone phosphorylation in DNA nucleotide excision repair.

1.1.2. Phosphorylation in Double-Strand Break Repair. Unstable genomes activate DNA repair pathways, which are indicated by increased phosphorylation of numerous factors, including H2AX, ATM, ATR (ATM- and RAD3-related), Chk1, Chk2, DNA-dependent protein kinase, catalytic subunit (DNA-PKcs), and Ku70/Ku80 heterodimers. When a single-strand or double-strand break occurs in a cell, the phosphorylation of the carboxyl-terminal serine residues of thousands of H2AX molecules is observed; this produces phosphorylated H2AX, or γ-H2AX [25]. Subsequently, the γ-H2AX molecules form megabase chromatin domains surrounding the sites of DSBs. This enables the use of antibodies against γ-H2AX to detect the DSBs. Many components of DDR and repair are recruited directly or indirectly to these γ-H2AX foci, including BRCA1, Rad50, Rad51, 53BP1, mediator of DNA damage checkpoint protein 1 (MDC1), and Nijmegen breakage syndrome 1 (NBS1) [26]. ATM/Chk2 and ATR/Chk1 pathways are principal regulators of cell cycle arrest and activated DNA-PKcs that form a holoenzyme, called DNA-PK, which catalyzes the joining of nonhomologous ends [27]. LSH is a protein related to the SNF2 family of chromatin remodeling ATases. LSH-deficient cells exhibit reduced viability and repair DNA double-strand breaks after exposure to IR. This is because H2AX is less efficiently phosphorylated in LSH-deficient cells in response to DNA damage [28]. These reports suggest direct and indirect roles of H2AX and the phosphorylation of H2AX in DNA damage repair. MDC1, a key protein in DDR events, contains the fork-head-associated (FHA) domain at its N-terminus and a tandem key protein in DDR events, contains the fork-head-associated (FHA) domain at its N-terminus and a tandem repeat motifs of Ser-Asp-Thr (SDT) by CK2 in response to DNA repair. MDC1 is recruited to DNA damage foci by binding to γ-H2AX via its BRCT domains [29]. Additionally, NBS1, a component of the Mre11-Rad50-Nbs1 (MDC1) complex, carries a combination of FHA and BRCTs. NBS1 has been reported to interact with γ-H2AX via its FHA/BRCT domain [30]. MDC1 is constitutively phosphorylated on conserved repeat motifs of Ser-Asp-Thr (SDT) by CK2 in response to DNA damage agents. The phosphorylated forms of these motifs confer recognition sites and binding sites for the NBS1 and the MRN complex [31]. Several recent reports have demonstrated that NBS1 localizes to unpaired DSBs by interacting with phosphorylated MDC1 [31, 32]. Moreover, Aprataxin, an FHA domain-containing protein, localizes to sites of DNA damage by interacting with phosphorylated MDC1 [33]. Schizosaccharomyces pombe Ctp1, a DNA end-processing factor, is phosphorylated both basally and in a DNA damage-dependent manner [34]. Ctp1 is phosphorylated on a domain that resembles the SDT repeats of MDC1. This phosphorylation is required for Ctp1 to interact with the FHA domain of NBS1 and result in the subsequent localization to DSB sites [35]. Thus, the phosphorylation of MDC1 plays a pivotal role in the recruitment of NBS1 and other DNA repair factors to unpaired DSBs. Overall, by recognizing phosphorylated H2AX, MDC1 is recruited to DNA damage sites and is further phosphorylated by CK2 kinase to recruit other DDR factors to initiate DNA repair and delay cell cycle progression. In general, the role of phosphorylation can be stressed by generating binding interfaces at sites of DNA damage recognized by DNA repair factors.

1.2. Poly(ADP-ribosyl)ation. Poly(ADP-ribosyl)ation is a phylogenetically ancient, reversible, posttranslational modification [36] and ubiquitously occurs in nearly all nucleated cells of mammals, plants, and lower eukaryotes, but not in yeast [37]. Cellular poly(ADP-ribose) (PAR) is synthesized by poly(ADP-ribose) polymerases. One of the most predominant polymerases, poly(ADP-ribose) polymerase-1 (PARP-1), transfers ADP-ribose from NAD+ onto specific amino acid residues of itself or substrate proteins to release nicotinamide. The known ADP-ribose acceptors in eukaryotic cells are lysine, arginine, glutamate, aspartate, cysteine, phosphoserine, and asparagine residues [36]. These positively charged amino acids interact with the negatively charged phosphate groups of PAR chains and thereby affect the structure, enzymatic activity, and interactions with other acceptor proteins [38]. Poly(ADP-ribose) and free ADP-ribose are simultaneously formed and generated, respectively. While PARP-1 catalyzes the synthesis of poly(ADP-ribose), another major enzyme PAR-degrading enzyme poly(ADP-ribose) glycohydrolase (PARG) hydrolyzes the glycosidic bond between the ADP-ribose units of poly(ADP-ribose) to produce free ADP-ribose. This maintains the balance of ADP-ribose in cells [39]. While poly(ADP-ribose) has been identified, the biological function of this protein modification is currently being investigated. Poly(ADP-ribosyl)ation is implicated in a wide range of processes including DNA damage, DNA repair, and the maintenance of genomic stability, transcriptional regulation, energy metabolism, mitosis, and cell death [40].

1.2.1. Poly(ADP-ribosyl)ation in Nucleotide Excision Repair. PARP-1 is highly efficient in sensing DNA nicks through its DNA-break-sensing motif, which contains two CX5CX33-34HX2C zinc fingers [41]. After binding to nicks, the catalytic activity of this protein is immediately activated and catalyzes the polymerization of poly(ADP-ribose) chains and other substrates [42]. The formation of poly(ADP-ribose) then promotes the dissociation of histones and nucleosomes from DNA and completely unravels polynucleosomes [43]. Poirier et al. first demonstrated that the poly(ADP-ribosyl)ation of histone H1 causes relaxation of chromatin fiber [44]. The dissociation of the H1-DNA and H3-B-DNA complex is accompanied by the formation of ADP-ribose polymers. Moreover, the incubation of these histones with free or PARP-1-bound PAR loosens the chromatin structure and makes it possible for the DNA repair machinery to access damaged DNA [45]. XRCC1, the scaffold protein of SSBR/BER, has been reported to interact with poly(ADP-
1.2.2. Poly(ADP-ribosyl)ation in Double-Strand Break Repair. PARP-1 senses double-strand breaks and provides a scaffold for chromatin remodelers and for binding DSB repair-associated proteins by synthesizing poly(ADP-ribose) on itself or other proteins. These proteins contain PAR binding motifs or modules and can be classified into three categories according to structural characteristics. The first category is characterized by a 20-25 amino acid motif with an N-terminal, basic, residue-rich cluster and alternately arranged hydrophobic amino acids. This motif has been reported in several proteins, such as p53, histones, XRCC1, and XPA [46, 47, 50]. The second category is characterized by highly conserved 130-190 residues (i.e., a “macro domain”) and is found in the poly(ADP-ribose) polymerases PARP9, PARP-14 and PARP-15 [51], LRP16 (MDO1), MDO2, and MacroH2A1.1 [52]. The final category is characterized by a PBZ domain. The PBZ domain is a putative C2H2 zinc finger structure separated by a 6–8 amino acid spacer and has [K/R]xxCx[F/Y] GxxCbbxxxxHxxxx [F/Y] xH sequence. Checkpoint proteins fork-head-associated and RING domains (CHFR) and DDR protein Aprataxin and PNK-like factor (APLF) have been reported to have PBZ domain [53]. CHFR, an E3 ubiquitin ligase, is recruited to DSBs by binding to poly(ADP-ribose) via its PBZ motif. Subsequently, CHFR ubiquitinates PAR polymerase 1 (PARP-1) and regulates chromatin-associated PARP-1 in vivo [54]. APLF is recruited to DNA damage sites by its PBZ motif and phosphorylated by ATM so as to facilitate DNA double-strand break repair through NHEJ [55]. Krietsch et al. demonstrated that RRM1, another motif found in RNA-binding protein NONO, is responsible for NONO binding to PAR; NONO localizes to damaged chromatin by binding to PAR and promotes NHEJ in response to double-strand breaks [56]. Amplified in liver cancer 1 (ALC1), a member of the SNF2 ATPase superfamily, is recruited to damaged chromatin via its C-terminal macro domain. The ATPase and chromatin remodeling activities are strongly activated by PARP-1 [57], so the role of ALC1 in controlling the chromatin structure in DNA repair is dependent on PARP-1. BAL1, another macro domain-containing protein, and its partner E3 ligase are PAR-dependent. Recruitment, local BBAP-mediated ubiquitylation, and subsequent recruitment of checkpoint mediators 53BP1 and BRCA1 are induced by DNA double-strand breaks [58]. These reports suggest that poly(ADP-ribosyl)ation in DNA double-strand break repair focuses on recruiting a series of proteins involved in DNA repair to damaged sites via different PAR binding motifs and furthermore contributes to the initiation of downstream events at different levels.

1.3. Ubiquitination. Compared with other PTMs, ubiquitination is a complicated and reversible covalent modification sequentially catalyzed by a series of enzymes. Ubiquitin is a highly conserved protein found in a number of species and is covalently attached to the lysine residues of target proteins [59]. Ubiquitination involves three steps. First, ubiquitin is activated by an activation enzyme (E1) with the required energy provided by ATP. Second, the activated ubiquitin is transferred to a ubiquitin-conjugation enzyme (E2). Finally, ubiquitin is attached via ubiquitin ligase (E3) to the target protein through an isopeptide bond between the C-terminal of ubiquitin and the [epsilon]-amino group of the lysine residue in the target protein [60]. Ubiquitin itself contains seven lysines that can be attached to another ubiquitin to form a polyubiquitin chain. The most common polyubiquitin chain is formed by linking Lys48 to another ubiquitin on target proteins. This chain mediates the recognition and degradation of proteins by proteasome complexes [61]. Meanwhile, Lys63-linked polyubiquitination does not undergo proteasome degradation of target proteins. Instead, Lys63-linked polyubiquitination has been implicated in regulating DNA repair, protein kinase activation, and other biological processes [60].

1.3.1. Ubiquitination in Nucleotide Excision Repair. DNA damage excision is carried out by six repair factors, Replication Protein A (RPA), XPA, XPC, TFIIH, xeroderma pigmentosum complementation group G protein (XPG), and XPF-ERCC1 in mammalian cells [62]. The XPC protein is involved in recognizing lesions and plays an important role in initiating global-genome NER [63]. Ultraviolet light-(UV-) damaged DNA binding protein (UV-DDB), comprising DDB1 and DDB2 subunits, is another complex that appears to be involved in the recognition of NER [64]. DDB2 has been reported to be involved in the recruitment of XPC to UV-induced damaged DNA [65]. Moreover, Sugawara et al. demonstrated that upon UV irradiation, XPC was ubiquitylated by a functional UV-DDB-ubiquitin ligase complex [66]. Ubiquitylation XPC increases its affinity for DNA while ubiquitylation xeroderma pigmentosum complementation group E protein (XPE) leads to its degradation [62]. A recent report has suggested that XPC could enhance the E3 activity of the DDB complex and further promote the recruitment of XPA to damaged sites [67]. The XPA protein has also been reported to be rate-limiting to NER in response to UV [68] and plays an important role in a secondary
recognition step in conjunction with RPA for the verification of DNA lesions [69].

RNAPII is a huge complex and is involved in masking DNA lesions to prevent association with NER enzymes [70]. The rapid ubiquitination of RNAPII has been reported to be induced by UV irradiation and repaired by NER [71]. Ubiquitinated RNAPII degrades by 26S proteasome and enables DNA repair [72].

1.3.2. Ubiquitination in Double-Strand Break Repair. Ubiquitination plays a key role in the assembly and repair of signaling proteins at sites of double-strand DNA breaks. Upon DNA damage, RNF8, a RING-finger ubiquitin ligase, has been reported to rapidly recruit to DSBs by recognizing a cluster of highly conserved T-Q-X-F motifs in MDC1 via its FHA domain [73]. Subsequently, RNF8 interacts with HERC2, an E3 ubiquitin ligase with a giant HECT domain. This interaction increases the probability of forming K63-linked polyubiquitin chains on histones at sites of DNA damage [74]. RNF8 ubiquitylates the DSBR-flanking chromatin and H2A histones surrounding the DNA lesion and enables the assembly of 53BP1 and BRCA1 by the DSBR-flanking chromatin [73]. The ubiquitylated histone H2A further facilitates the recruitment of RNF168 at sites of damaged DNA. RNF168, another E3 ligase, cooperates with UBC13 to amplify the RNF8-dependent histone ubiquitylation by targeting histone H2A and promoting the formation of lysine 63-linked ubiquitin conjugates. These RNF168-dependent chromatin modifications trigger a second wave of accumulation of 53BP1 and BRCA1 to DNA lesions [75]. However, another report has suggested that the initial ubiquitination signal is derived from another FHA-containing E3 ligase CHFR. This ligase is recruited to DNA damage sites in the early stage of DNA damage by binding to PAR via its C-terminal PBZ motif, different from RNF8 [54]. The E3 ligases, RNF8 and CHFR, are recruited to DSBs by different means, i.e., phosphorylation or poly(ADP-ribosylation). While these two E3 ligases are known to play important roles in recruiting DSB regulators to DSBs in the first wave of ubiquitination, precise mechanisms have not been reported. Furthermore, these two E3 ligases trigger the second wave of ubiquitination, which amplifies the signal to recruit more DNA repair-related proteins. Smeenk et al. demonstrated that SMARCA5/SNF2H, the catalytic subunit of ISWI chromatin remodeling complexes, is recruited to DSBs with similar kinetics as that of GFP-MDC1 but in a different manner. SMARCA5/SNF2H binds to PARP-1 and regulates ubiquitin response by promoting the binding of E3 ubiquitin ligase RNF168. Ubiquitin conjugates the ubiquitin-binding factors RAD18 and the RAP80-BRCA1 complex throughout DSBR-flanking chromatin [76].

A report demonstrated that RNF8 induces the formation of Lys48-linked polyubiquitin chains at DSB sites in response to IR and subsequently ubiquitinates and degrades KU80 to facilitate NHEJ [77]. This report further suggested that RNF8 may have two distinct roles at DNA damage sites when forming different ubiquitin chains. The first role is to ubiquitinate H2A and to recruit RNF168 to accumulate other DNA damage repair proteins to DSB sites; this is achieved by forming a Lys63-linked polyubiquitin chain; the second role is to ubiquitinate and to remove a number of proteins from DNA damage sites by forming a Lys48-linked polyubiquitin chain.

1.4. SUMOylation. SUMOylation is a three-step enzyme pathway that attaches SUMO to a substrate through an isopeptide bond between the C-terminal carboxyl group of SUMO and the ε-amino group of a lysine residue in the substrate [78]. As with many other ubiquitin and ubiquitin-like proteins (UBL proteins), all eukaryotic SUMO proteins are translated as immature precursors that must first be processed by a protease to generate the mature protein form. The enzymes of the SUMO pathway, although analogous to those of the ubiquitin pathway, are specific for SUMO. SUMOylation begins with a SUMO-activating enzyme (E1), which activates the SUMO C-terminus in an ATP-dependent manner. An activated SUMO is transferred to a SUMO-conjugating enzyme Ubc9 (E2). Under the assistance of one of several SUMO-protein ligases (E3s), SUMO is then transferred from Ubc9 to the substrate. Substrate specificity is ensured by UBC9 and the E3s. Many identified SUMO-modified proteins contain a Lys acceptor within a ψKX(D/E) consensus motif, where ψ is a large hydrophobic amino acid (generally isoleucine, leucine, or valine), K is the modified lysine residue, X is any residue, and E is glutamic acid [79]. This motif is bound directly by UBC9. E3s likely enhance specificity by interacting with other features of the substrate [80]. SUMO conjugation is a reversible modification through proteases, including UBL-specific proteases and sentrin-specific proteases (i.e., ULPs and SENPs, respectively) [81]. SUMOylation and one member of the UBLs (ubiquitin-like proteins) become increasingly important in multiple biological processes, such as transcription, replication, and events following DNA damage [82].

1.4.1. SUMOylation in Nucleotide Excision Repair. The production of cyclobutane pyrimidine dimers (CPDs) and pyrimidine pyrimidone photoproducts (6-4PPs) is induced by UV irradiation [83]. These photoproducts are primarily repaired by the nucleotide excision repair (NER) pathway [84]. Many proteins have been reported to be SUMOylated in this process. In Saccharomyces cerevisiae, the vast majority of SUMOylation are catalyzed by two Siz/PIAS SUMO E3 ligases, Siz1 and Siz2. Siz1 and Siz2 mutants are sensitive to ultraviolet (UV) light and show delayed CPD repair [85]. Mms21 is another SUMO ligase in Saccharomyces cerevisiae; the abolition of the SUMO E3 activity of Mms21 leads to DNA damage sensitivity [86]. These reports have suggested that SUMOylation plays certain roles in the nucleotide excision repair pathway. Thymidine DNA glycosylase (TDG) is a BER enzyme that removes thymine or uracil from T-G or U-G mismatched base pairs. The SUMOylation of TDG dramatically reduces its DNA substrate and ATP binding site affinity. This is associated with significant increases in enzymatic turnover in reactions with a G+U substrate and the loss of G+T processing activity [87]. XPC is a key protein in the recognition step in the NER pathway, and its degradation is necessary for recruiting downstream XPG and for promoting
an efficient NER. The SUMOylation of XPC increases its stability by inhibiting degradation via a ubiquitin-proteasome system in response to UV irradiation to initiate DNA damage recognition [88]. A later report by the same group in 2007 suggested that XPC is degraded by the 26S proteasome and is independent of ubiquitylation upon UV irradiation. Moreover, the UV-induced XPC degradation is controlled by XPC SUMOylation. This prevents the excessive depletion of XPC from cells [89]. Overall, the SUMOylation of XPC is indispensable for the proper functioning of the NER pathway.

1.4.2. SUMOylation in Double-Strand Break Repair. Protein inhibitor of activated STAT protein 4 (PIAS4) and protein inhibitor of activated STAT protein 1 (PIAS1), E3 ligase enzymes, are activators of transcription (PIAS) family and are recruited to damage sites via their SAP domains. SUMO E2 (UBC9), after SUMO1 and SUMO2/3 conjugation, relocates to sites of DSBs and forms SUMO2/3 and SUMO1 conjugate-coordinates via PIAS1 and PIAS4, respectively, upon various genomic agents [90]. Depletions of the PIAS proteins affect the ability of damaged cells to effectively relocalize 53BP1 and BRCA1 to sites of DNA breaks [91]; this significantly impairs histone H4 ubiquitylation and the accumulation of K63-linked ubiquitin conjugates at sites of DSBs [91]. These reports have suggested the important roles of SUMO E3 ligases in DNA repair. In response to genotoxic stress, RAP80 was recruited to DNA damage sites by binding specifically to SUMO2/3 via its SIM domain [92]. The SUMO-associated RAP80 recognizes RNF4-synthesized hybrid SUMO-ubiquitin chains and promotes BRCA1 recruitment [93]. MDC1 and BRCA1 are also modified by small ubiquitin-like modifiers (SUMOs) following DNA damage [94, 95]. RNF4 targets SUMOylated MDC1 and SUMOylated BRCA1 and degrades MDC1. This removes MDC1 and 53BP1 from sites of DNA damage and loads Rad51, an enzyme required for HR repair, to sites of DNA damage [94, 96]. The recruitment of Rad51 to DNA damage foci is facilitated by a hypoSUMOylated 70 kDa subunit of RPA (RPA70) in response to Camptothecin (CPT) [97]. HERC2 is a target of the SUMO E3 ligase PIAS4 and contains a novel ZZ zinc finger SUMO-binding domain. HERC2 is dependent on both SUMOylation and its SUMO-binding domain to interact with RNF8 and to stabilize the RNF8–UBC13 association [98]. When cooperating with PIAS1 and PIAS4, HERC2 facilitates an effective ubiquitin adduct formation to recruit more DSB proteins to promote DSB repair.

1.5. Acetylation. Acetylation is involved in regulating kinases, transcriptional activity, and protein stability [99]. The N-terminal acetylation of proteins is one of the most common protein modifications, occurs in approximately 85% of eukaryotic proteins, and is catalyzed by NATs that transfer acetyl groups from acetyl-coA to the termini of α-amino groups [100]. A less common but more important form of protein acetylation occurs on the ε-amino group of lysine and is catalyzed by histone acetyltransferases (HATs) that transfer an acetyl group from acetyl-coenzyme A (acetyl-CoA) to the ε-amino group of certain lysine side chains within the basic N-terminal tail region of a histone [101]. Acetylation is a highly reversible process; acetylated histone proteins can be deacetylated by histone deacetylases (HDACs). HDACs have been classified into four groups based on their homology to yeast histone deacetylases. The balance between acetylation and deacetylation is an important modulation mechanism for many cellular functions in diverse proteins [102].

1.5.1. Acetylation in Nucleotide Excision Repair. XPA is a key protein in nucleotide excision repair and has a high affinity for damaged DNA. XPA plays a central role in correctly positioning the repair machinery around the injury site by interacting with many NER factors [103]. Recently, XPA has been reported to be phosphorylated by checkpoint kinase ATR in cells in response to UV radiation [104]. Fan and Luo et al. showed that XPA could be acetylated at lysines 63 and 67. The acetylated XPA can be deacetylated by SIRT1 upon UV irradiation [105]. The acetylation of histone H3 lysine 9 (H3K9) in response to UV irradiation increases accessibility to the NER machinery [106]. Apurinic/apyrimidinic endonuclease 1 (APE1) is the main basic endonuclease. The acetylation of APE1 contributes to BER by modulating APE1 subnuclear distribution and enzymatic activity in vivo [107]. A recent report demonstrated that Tip60 acetylate XPF so as to mediate the XPF-ERCC1 complex assembly and activation in NER [108].

1.5.2. Acetylation in Double-Strand Break Repair. MRN is a proximal DSB sensor that senses DNA double-stick breaks and transmits damage signals downstream by phosphorylating proteins involved in cell cycle checkpoints and DNA repair. Tip60 has been reported to form a complex with MRN. This interaction is required for its binding to DSBs [109]. The recruitment of Tip60 to DSBs is dependent on Fe65 [110]. Given that Tip60 is an acetyltransferase, the activation of ATM has been reported to be dependent on acetylation modifications mediated by Tip60 histone acetyltransferase (HAT) in response to DSBs (the acetylation site of ATM is lysine 3016) [111, 112]. The acetylation of H4AX (K5) has been observed in the very early stages of DDR and is mediated by Tip60 [113]. Another report indicated that H4AX is constitutively acetylated on lysine 36 (H4AX K36Ac) by CBP/p300 acetyltransferases independently of H4AX phosphorylation [114]. Tip60-dependent H4 acetylation promotes DNA HR by decreasing 53BP1 DSB chromatin occupancy [115]. CBP/p300-dependent H3 and H4 acetylation facilitates NHEJ and chromatin relaxation by recruiting Ku70 and Ku80 [116]. BRG1, the catalytic subunit of the SWItch/Sucrose Nonfermentable (SWI/SNF) chromatin remodeling complex, facilitates DSB repair by interacting with acetylated H3 through its bromodomain [117]. Thus, we can conclude that histone acetyltransferase plays an important role in this process. HAT1 has been reported to facilitate efficient HR by enriching the binding of H4K5/K12-acetylated H3.3 (H3.3-H4K5/12ac) to the DSB sites and the subsequent recruitment of key repair factor RAD51 [118]. Therefore, acetylation plays a seemingly
earlier role in the early stages of DNA damage than the formation of γ-H2AX. Moreover, acetylation facilitates the recruitment of DSB-associated proteins. Additionally, the acetylation of core histones also mediates proteasomal degradation in DNA repair [99].

1.6. Methylation. Methylation occurs predominantly on arginine, lysine residues, and is catalyzed by S-adenosylmethionine-(AdoMet-) dependent enzymes that donate a methyl group to the side-chain nitrogen atoms of these residues. Arginine methylation is a posttranslational modification that is involved in a variety of cellular functions such as RNA processing, signal transduction, transcriptional regulation, and DNA repair [119]. Protein arginine methylation is a covalent modification that results in the addition of methyl groups to the nitrogen atoms of the arginine side chains and is catalyzed by a family of protein arginine methyltransferases (PRMTs), but only one arginine demethylase has been identified, namely, the Jumonji domain-containing 6 (JMJD6) [120]. Studies on lysine methylation have focused on histone proteins, which are regulated by histone methyltransferase (HMT) and histone demethylase (HDME) enzymes that add and remove methyl groups on lysine and arginine residues within proteins, respectively. Increasing reports suggested that histone protein methylation plays a major role in DNA damage repair.

1.6.1. Methylation in Base and Nucleotide Excision Repair. DNA polymerase β (Pol-β) is a critical player in BER, which is involved in step (iii) and step (iv) of BER. Recent findings indicate that the Pol-β lyase domain can associate with PRMT6 and PRMT1. These two PRMTs play distinct, nonredundant roles in regulating Pol-β function. PRMT6 methylates Pol-β R83 and R152 and augments its polymerase activity by enhancing its DNA binding and processivity while PRMT1 methylates R137, which in turn blocks the Pol-β-PCNA interaction [121].

Flap endonuclease-1 (PEN1) is a Rad2 nuclease family member that plays a vital role in DNA replication and repair. FEN1 methylation promotes the interaction between FEN1 and PCNA [122] so as to favor NER.

1.6.2. Methylation in Double-Strand Break Repair. RUVBL1 is a cofactor of the TIP60 complex. During DSB repair, methylation of RUVBL1 by PRMT5 is required to activate TIP60α, promoting histone H4K16 acetylation, which facilitates 53BP1 displacement from DSBs [123]. The mammalian MRN complex plays a critical role in HR. Follow-up studies indicated that Mre11 was indeed methylated by PRMT1. In cells treated with a global methyltransferase inhibitor followed by the DNA-damaging agent etoposide, few γH2AX foci formed, and Mre11 was not recruited to DSBs [124]. 53BP1 is a substrate of ATM and functions as a mediator protein when it accumulates at DSB. Similar to MRE11, treatment with methylase inhibitors interfered with 53BP1 recruitment at DSBs [122]. Many HMTs and HDTs are recruited to DNA damage sites where they act to modify chromatin to orchestrate chromatin-based DDR activities. Lysine demethylase 5B (KDM5B) mediated the H3K4me3 demethylation required for DSB repair factors Ku70 and BRCA1 accumulation at DSBs. The interaction between H3K9me3 and Tip60 increases the HAT activity of TIP60, which acetylates ATM and H4 to support HR repair [9]. Methylation of H3K6 improves the association of early DNA repair components, including NBS1 and Ku70, with the induced DSB, and enhanced DSB repair [125]. These reports imply that methylation regulates DNA damage repair protein relocalization to DSBs and plays important roles at multiple nodes in DNA repair.

2. Conclusions and Perspectives

Following the induction of DNA damage, proteins surrounding chromatin at break sites undergo different types of PTMs. The poly(ADP-ribosyl)ation modification of proteins at damage sites relaxes the chromatin structure and enables DNA repair machinery to access damaged DNA. Acetylation likely occurs earlier than other modifications; the activity of ATM benefits from acetyltransferase Tip60 and Tip60 partner Fe65, activated ATM, and PIKK family other members lead to the phosphorylation of H2AX. MDC1, a mediator protein that plays a crucial role in the DDR pathway, is then immediately recruited to sites of DSBs to directly interact with γ-H2AX via BRCT domains. Subsequently, histones
H2A and H2AX are ubiquitylated in the chromatin surrounding DSBs by the E3 ubiquitin ligases RNF8 and RNF168 in an MDC1-dependent manner. RNF8 triggers the formation of two different ubiquitin chains, i.e., a Lys63-linked polyubiquitin chain that mediates the recruitment of new damage factors and a Lys48-linked polyubiquitin chain that mediates the degradation of associated proteins. RNF168 amplifies signals from RNF8, triggers the formation of a second wave of ubiquitin chain formations, and further recruits more proteins involved in DNA damage repair (Figure 2). These early steps in the DDR pathway are primarily controlled by phosphorylation, whereas ubiquitination appears to be a key player in later stages.

DNA damage repair is a complicated process; different types of PTMs are required to accomplish each step in an orderly and coordinatively fashion. For example, in the recruitment of DNA damage factors to damage sites, phosphorylation renders binding sites appropriate for damaging factors; methylation is also required for the recruitment of DNA repair proteins. Poly(ADP-ribosylation), ubiquitination,
and SUMOylation form long, branched chains for damage factors to bind. SUMOylation and ubiquitination, beyond the formation of these chains, also play pivotal roles in the turnover of DNA damage-associated proteins at damage sites (Figures 3 and 4). In general, the roles of PTMs in DNA damage repair can be categorized into three groups: to recruit DNA damage repair-related factors to damage sites, to prepare binding scaffolds and facilitate the formation of damage repair complexes, and to renew proteins at damage sites to complete the DNA damage repair.

In the present work, we have addressed the best-known protein PTMs related to excision and DNA DSB repair mechanisms. The PTMs of proteins play a critical role in DNA damage repair; mutations within the genes encoding effectors of these components lead to genomic instability and in selected cases human radiosensitivity and cancer susceptibility syndromes. Targeting enzymes involved in PTMs in DNA repair like PARPs, which cooperate and complement molecular defects of the DDR process, induces a specific lethality in DDR defective cancer cells and represents an anti-cancer strategy [108]; PARP inhibitors are the first clinically approved drugs targeting DDR to have entered the clinic. E3 ubiquitin ligases NEDD4-1 likely acts as a novel drug target or diagnostic marker in the battle against cancer [126]. Recent studies suggest that treatment with small molecule inhibitors of HDAC activity results in antitumor effects in a variety of transformed cell lines; several HDAC inhibitors are in clinical development and show antitumor activity in cancer patients [127]. PRMT inhibitors and KDM inhibitors have recently entered or are about to enter the clinic, mainly for cancer therapy [119, 128], etc. Therefore, inhibition of the key enzymes of PTMs in DNA repair would make a contribution to new cancer therapy strategies, and further study is required to create more inhibitors specific to enzymes in DNA repair.

In addition to the six types of PTMs mentioned above, recently unknown PTMs that are involved in DNA damage repair will be identified in the future. With the development of life science technology and chemical synthesis as well as the application of computer image modeling, PTMs will be better studied, which would contribute to a better understanding of the role of PTMs in DNA repair; thus, elucidating the molecular mechanisms of DNA repair pathways involving PTM signaling could reveal new and selective therapeutic approaches to target cancers.

Data Availability

All data generated or analyzed during this study are included in this article.

Conflicts of Interest

The authors have declared that no competing interests exist.

Authors’ Contributions

Miaomiao Bai and Dongdong Ti contributed equally to this work.

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References

[1] W. L. Santivasi and F. Xia, "Ionizing radiation-induced DNA damage, response, and repair," *Antioxidants & Redox Signaling*, vol. 21, no. 2, pp. 251–259, 2014.
[2] Z. Wu, C. Wang, M. Bai et al., "An LRP16-containing preassembly complex contributes to NF-κB activation induced by DNA double-strand breaks," *Nucleic Acids Research*, vol. 43, no. 6, pp. 3167–3179, 2015.
[3] J. W. Harper and S. J. Elledge, "The DNA damage response: ten years after," *Molecular Cell*, vol. 28, no. 5, pp. 739–745, 2007.
[4] I. Schonn, J. Hennesen, and D. C. Dartsch, "Cellular responses to etoposide: cell death despite cell cycle arrest and repair of DNA damage," *Aptosis*, vol. 15, no. 2, pp. 162–172, 2010.
[5] L. F. Povirk, "Biochemical mechanisms of chromosomal translocations resulting from DNA double-strand breaks," *DNA Repair*, vol. 5, no. 9-10, pp. 1199–1212, 2006.
[6] N. L. Vasilenko and G. A. Nevinskii, "Enzymes of direct, excision, and corrected systems of repair of higher and lower organisms and their biological roles," *Molekuliarnaia Biologii*, vol. 37, no. 6, pp. 944–960, 2003.
[7] J. A. Marteijn, H. Lins, W. Vermeulen, and J. H. J. Hoeijmakers, "Understanding nucleotide excision repair and its roles in cancer and ageing," *Nature Reviews Molecular Cell Biology*, vol. 15, no. 7, pp. 465–481, 2014.
[8] S. Bhattacharjee and S. Nandi, "Choices have consequences: the nexus between DNA repair pathways and genomic instability in cancer," *Clinical and Translational Medicine*, vol. 5, no. 1, p. 45, 2016.
[9] F. Gong and K. M. Müller, "Histone methylation and the DNA damage response," *Mutation Research*, vol. 780, pp. 37–47, 2019.
[10] B. Michel, "After 30 years of study, the bacterial SOS response still surprises us," *PLoS Biology*, vol. 3, no. 7, article e255, 2005.
[11] M. S. Y. Huen and J. Chen, "The DNA damage response pathways: at the crossroad of protein modifications," *Cell Research*, vol. 18, no. 1, pp. 8–16, 2008.
[12] C. Smal, D. Vertommen, L. Bertrand et al., "Identification of in vivo phosphorylation sites on human deoxycytidine kinase. Role of Ser-74 in the control of enzyme activity," *The Journal of Biological Chemistry*, vol. 281, no. 8, pp. 4887–4893, 2006.
[13] S. Mihlan, C. Reiß, P. Thalheimer et al., "Nuclear import of LASP-1 is regulated by phosphorylation and dynamic protein–protein interactions," *Oncogene*, vol. 32, no. 16, pp. 2107–2113, 2013.
[14] V. Singh, M. Ram, R. Kumar, R. Prasad, B. K. Roy, and K. K. Singh, "Phosphorylation: implications in cancer," *The Protein Journal*, vol. 36, no. 1, pp. 1–6, 2017.
[15] G. L. Dianov and J. L. Parsons, "Co-ordination of DNA single strand break repair," DNA Repair, vol. 6, no. 4, pp. 454–460, 2007.

[16] L. Lan, S. Nakajima, Y. Oohata et al., "In situ analysis of repair processes for oxidative DNA damage in mammalian cells," Proceedings of the National Academy of Sciences of the United States of America, vol. 101, no. 38, pp. 13738–13743, 2004.

[17] R. M. Taylor, D. J. Moore, J. Whitehouse, P. Johnson, and K. W. Caldecott, "A cell cycle-specific requirement for the XRCC1 BRCT II domain during mammalian DNA strand break repair," Molecular and Cellular Biology, vol. 20, no. 2, pp. 735–740, 2000.

[18] J. Fan, M. Otterlei, H. K. Wong, A. E. Tomkinson, and Wilson DM 3rd, "XRCC1 co-localizes and physically interacts with PCNA," Nucleic Acids Research, vol. 32, no. 7, pp. 2193–2201, 2004.

[19] C. E. Ström, O. Mortusewicz, D. Finch et al., "CK2 phosphorylation of XRCC1 facilitates dissociation from DNA and single-strand break formation during base excision repair," DNA Repair, vol. 10, no. 9, pp. 961–969, 2011.

[20] Y. Kubota, T. Takanami, A. Higashitani, and S. Horiuchi, "Localization of X-ray cross complementing gene 1 protein in the nuclear matrix is controlled by casine kinase II-dependent phosphorylation in response to oxidative damage," DNA Repair, vol. 8, no. 8, pp. 953–960, 2009.

[21] W. C. Chou, H. C. Wang, F. H. Wong et al., "Chk2-dependent phosphorylation of XRCC1 in the DNA damage response promotes base excision repair," The EMBO Journal, vol. 27, no. 23, pp. 3140–3150, 2008.

[22] J. L. Parsons, I. I. Dianova, D. Finch et al., "XRCC1 phosphorylation by CK2 is required for its stability and efficient DNA repair," DNA Repair, vol. 9, no. 7, pp. 835–841, 2010.

[23] L. Méndez-Acuña, M. V. di Tomaso, F. Palitti, and W. Martínez-López, "Histone post-translational modifications in DNA damage response," Cytogenetic and Genome Research, vol. 128, no. 1-3, pp. 28–36, 2010.

[24] C. Dinant, A. B. Houtsmuller, and W. Vermeulen, "Chromatin structure and DNA damage repair," Epigenetics & Chromatin, vol. 1, no. 1, p. 9, 2008.

[25] P. L. Collins, C. Purman, S. I. Porter et al., "DNA double-strand breaks induce H2A histone phosphorylation domains in a contact-dependent manner," Nature Communications, vol. 11, no. 1, 2020.

[26] S. Bekker-Jensen and N. Mailand, "Assembly and function of DNA double-strand break repair foci in mammalian cells," DNA Repair, vol. 9, no. 12, pp. 1219–1228, 2010.

[27] S. H. Yang, T. C. Kuo, H. Wu et al., "Perspectives on the combination of radiotherapy and targeted therapy with DNA repair inhibitors in the treatment of pancreatic cancer," World Journal of Gastroenterology, vol. 22, no. 32, pp. 7275–7288, 2016.

[28] J. Burrage, A. Termanis, A. Geissner, K. Myant, K. Gordon, and I. Stancheva, "The SNF2 family ATPase LSH promotes phosphorylation of H2AX and efficient repair of DNA double-strand breaks in mammalian cells," Journal of Cell Science, vol. 125, no. 22, pp. 5524–5534, 2012.

[29] M. Stucki, J. A. Clapperton, D. Mohammad, M. B. Yaffe, S. J. Smerdon, and S. P. Jackson, "MDC1 directly binds phosphorylated histone H2AX to regulate cellular responses to DNA double-strand breaks," Cell, vol. 123, no. 7, pp. 1213–1226, 2005.

[30] J. Kobayashi, "Molecular mechanism of the recruitment of NBS1/hMRE11/hRAD50 complex to DNA double-strand breaks: NBS1 binds to gamma-H2AX through FHA/BRCT domain," Journal of Radiation Research, vol. 45, no. 4, pp. 473–478, 2004.

[31] F. Melander, S. Bekker-Jensen, J. Falck, J. Bartek, N. Mailand, and J. Lukas, "Phosphorylation of SDT repeats in the MDC1 N terminus triggers retention of NBS1 at the DNA damage-modified chromatin," Journal of Cell Biology, vol. 181, no. 2, pp. 213–226, 2008.

[32] J. R. Chapman and S. P. Jackson, "Phospho-dependent interactions between NBS1 and MDC1 mediate chromatin retention of the MRN complex at sites of DNA damage," EMBO Reports, vol. 9, no. 8, pp. 795–801, 2008.

[33] O. J. Becherel, B. Jakob, A. L. Cherry et al., "CK2 phosphorylation-dependent interaction between aprataxin and MDC1 in the DNA damage response," Nucleic Acids Research, vol. 38, no. 5, pp. 1489–1503, 2010.

[34] Y. Akamatsu, Y. Murayama, T. Yamada et al., "Molecular characterization of the role of the Schizosaccharomyces pombe nip1+/ctp1+ gene in DNA double-strand break repair in association with the Mre11-Rad50-Nbs1 complex," Molecular and Cellular Biology, vol. 28, no. 11, pp. 3639–3651, 2008.

[35] G. E. Dodson, O. Limbo, D. Nieto, and P. Russell, "Phosphorylation-regulated binding of Ctbp1 to Nbs1 is critical for repair of DNA double-strand breaks," Cell Cycle, vol. 9, no. 8, pp. 1516–1522, 2014.

[36] M. O. Hottiger, P. O. Hassa, B. Lüscher, H. Schüler, and F. Koch-Nolte, "Toward a unified nomenclature for mammalian ADP-ribosyltransferases," Trends in Biochemical Sciences, vol. 35, no. 4, pp. 208–219, 2010.

[37] O. Loseva, A. S. Jemth, H. E. Bryant et al., "PARP-3 is a mono-ADP-ribosylase that activates PARP-1 in the absence of DNA," Journal of Biological Chemistry, vol. 287, no. 41, pp. 34494–34499, 2012.

[38] M. Di Girolamo, N. Dani, A. Stilla, and D. Corda, "Physiological relevance of the endogenous mono(ADP-ribosyl)ation of cellular proteins," The FEBS Journal, vol. 272, no. 18, pp. 4565–4575, 2005.

[39] J. O’Sullivan, M. T. Ferreira, J.-P. Gagné et al., "Emerging roles of eraser enzymes in the dynamic control of protein ADP-ribosylation," Nature Communications, vol. 10, no. 1, pp. 1182, 2019.

[40] M. Platia, A. Ionescu, E. Ivan, J. F. Holland, J. Mandeli, and O. Platia, "PAR, a protein involved in the cell cycle, is functionally related to chromosomal passenger proteins," International Journal of Oncology, vol. 38, no. 3, pp. 777–785, 2011.

[41] M. F. Langelier and J. M. Pascal, "PARP-1 mechanism for coupling DNA damage detection to poly(ADP-ribose) synthesis," Current Opinion in Structural Biology, vol. 23, no. 1, pp. 134–143, 2013.

[42] F. Dantzer, J. C. Amé, V. Schreiber, J. Nakamura, J. Ménissier-de Murcia, and G. de Murcia, "Poly(ADP-ribose) polymerase-1 activation during DNA damage and repair," Methods in Enzymology, vol. 409, pp. 491–510, 2006.

[43] G. Yang, Y. B. Chen, J. X. Wu et al., "Poly(ADP-ribose)lation mediates early phase histone eviction at DNA lesions," Nucleic Acids Research, vol. 48, no. 6, pp. 3001–3013, 2020.

[44] G. G. Poirier, G. de Murcia, J. Jongstra-Bilen, C. Niedergang, and P. Mandel, "Poly(ADP-ribose)lation of poly(ADP-ribose)
causes relaxation of chromatin structure,” *Proceedings of the National Academy of Sciences of the United States of America*, vol. 79, no. 11, pp. 3423–3427, 1982.

[45] M. Tallis, R. Morra, E. Baraaksaite, and I. Ahel, “Poly(ADP-ribosylation) in regulation of chromatin structure and the DNA damage response,” *Chromosoma*, vol. 123, no. 1-2, pp. 79–90, 2014.

[46] P. A. Loeffler, M. J. Cuneo, G. A. Mueller, E. F. DeRose, S. A. Gabel, and R. E. London, “Structural studies of the PARP-1 BRCT domain,” *BMC Structural Biology*, vol. 11, no. 1, p. 37, 2011.

[47] J. M. F. Fischer, O. Popp, D. Gebhard et al., “Poly(ADP-ribose)-mediated interplay of OXPAn and PARP1 leads to reciprocal regulation of protein function,” *The FEBS Journal*, vol. 281, no. 16, pp. 3625–3641, 2014.

[48] B. S. King, K. L. Cooper, K. J. Liu, and L. G. Hudson, “Poly(ADP-ribose) polymerase-1 and xeroderma pigmentosum complementation group A in nucleotide excision repair,” *Journal of Biological Chemistry*, vol. 287, no. 47, pp. 39824–39833, 2012.

[49] M. Robu, R. G. Shah, N. Petitclerc, J. Brind’Amour, F. Kandan-Kulangara, and G. M. Shah, “Role of poly(ADP-ribose) polymerase-1 in the removal of UV-induced DNA lesions by nucleotide excision repair,” *Proceedings of the National Academy of Sciences of the United States of America*, vol. 110, no. 5, pp. 1658–1663, 2013.

[50] J. M. Pleschke, H. E. Kleczkowska, M. Strohm, and F. R. Althaus, “Poly(ADP-ribose) binds to specific domains in DNA damage checkpoint proteins,” *The Journal of Biological Chemistry*, vol. 275, no. 52, pp. 40974–40980, 2000.

[51] G. I. Karras, G. Kustatscher, H. R. Buhecha et al., “The macro domain is an ADP-ribose binding module,” *EMBO Journal*, vol. 24, no. 11, pp. 1191–11920, 2005.

[52] G. Timinszky, S. Till, P. O. Hassa et al., “A macrodomain-containing histone rearranges chromatin upon sensing PARP1 activation,” *Nature Structural & Molecular Biology*, vol. 16, no. 9, pp. 923–929, 2009.

[53] I. Ahel, D. Ahel, T. Matsusaka et al., “Poly(ADP-ribose)-binding zinc finger motif in DNA repair/checkpoint proteins,” *Nature*, vol. 451, no. 7174, pp. 81–85, 2008.

[54] C. Liu, J. X. Wu, S. C. Paudyal, Z. You, and X. Yu, “CHFR is important for the first wave of ubiquitination at DNA damage sites,” *Nucleic Acids Research*, vol. 41, no. 3, pp. 1698–1710, 2013.

[55] C. J. Macrae, R. D. McCulloch, J. Ylanko, D. Durocher, and C. A. Koch, “APLF (C2orf13) facilitates nonhomologous end-joining and undergoes ATM-dependent hyperphosphorylation following ionizing radiation,” *DNA Repair*, vol. 7, no. 2, pp. 292–302, 2008.

[56] J. Krietsch, M. C. Caron, J. P. Gagné et al., “PARP activation regulates the RNA-binding protein NONO in the DNA damage response to DNA double-strand breaks,” *Nucleic Acids Research*, vol. 40, no. 20, pp. 10287–10301, 2012.

[57] A. J. Gottschalk, G. Timinszky, S. E. Kong et al., “Poly(ADP-ribose)ylation directs recruitment and activation of an ATP-dependent chromatin remodeler,” *Proceedings of the National Academy of Sciences of the United States of America*, vol. 106, no. 33, pp. 13770–13774, 2009.

[58] Q. Yan, R. Xu, L. Zhu et al., “BAL1 and its partner E3 ligase, BBAP, link poly(ADP-ribose) activation, ubiquitylation, and double-strand DNA repair independent of ATM, MDC1, and RNF8,” *Molecular and Cellular Biology*, vol. 33, no. 4, pp. 845–857, 2013.

[59] K. Hagiund and I. Dikic, “Ubiquitylation and cell signaling,” *The EMBO Journal*, vol. 24, no. 19, pp. 3353–3359, 2005.

[60] J. Terzic, I. Marinovic-Terzic, F. Ikeda, and I. Dikic, “Ubiquitin signals in the NF-κB pathway,” *Biochemical Society Transactions*, vol. 35, no. 5, pp. 942–945, 2007.

[61] R. Verma, R. Oania, J. Graumann, and J. R. Deshaies, “Multi-ubiquitin chain receptors define a layer of substrate selectivity in the ubiquitin-proteasome system,” *Cell*, vol. 118, no. 1, pp. 99–110, 2004.

[62] T. Nouspikel, “DNA repair in mammalian cells: nucleotide excision repair: variations on versatility,” *Cellular and Molecular Life Sciences*, vol. 66, no. 6, pp. 994–1009, 2009.

[63] F. C. Clement, U. Camenisch, J. Fei, N. Kaczmarek, N. Mathieu, and H. Naegeli, “Dynamic two-stage mechanism of versatile DNA damage recognition by xeroderma pigmentosum group C protein,” *Mutation Research*, vol. 685, no. 1-2, pp. 21–28, 2010.

[64] A. E. Koyen, M. Z.adden, D. Park et al., “EZH2 has a non-catalytic and PREC-independent role in stabilizing DDB2 to promote nucleotide excision repair,” *Oncogene*, vol. 39, no. 25, pp. 4798–4813, 2020.

[65] M. E. Fitch, S. Nakajima, Y. Okusa et al., “In vivo recruitment of XPC to UV-induced cyclobutane pyrimidine dimers by the DDB2 gene product,” *The Journal of Biological Chemistry*, vol. 278, no. 47, pp. 46906–46910, 2003.

[66] K. Sugasawa, Y. Okuda, M. Sajo et al., “UV-induced ubiquitylation of XPC protein mediated by UV-DDB2-ubiquitin ligase complex,” *Cell*, vol. 121, no. 3, pp. 387–400, 2005.

[67] A. Takedachi, M. Sajo, and K. Tanaka, “DDB2 complex-mediated ubiquitylation around DNA damage is oppositely regulated by XPC and Ku and contributes to the recruitment of XPA,” *Molecular and Cellular Biology*, vol. 30, no. 11, pp. 2708–2723, 2010.

[68] B. Kohrer, V. Roginskaya, and R. D. Good, “XPA protein as a limiting factor for nucleotide excision repair and UV sensitivity in human cells,” *DNA Repair*, vol. 5, no. 5, pp. 641–648, 2006.

[69] L. Borszkova Pulzova, T. A. Ward, and M. Chovanec, “Role of poly(ADP-ribose) polymerase-1 and xeroderma pigmentosum group A protein in nucleotide excision repair and UV sensitivity,” *Molecular and Cellular Biology*, vol. 11, no. 1, pp. 2182, 2000.

[70] S. Tornaletti, D. Reines, and P. C. Hanawalt, “Structural characterization of RNA polymerase II complexes arrested by a cyclobutane pyrimidine dimer in the transcribed strand of template DNA,” *The Journal of Biological Chemistry*, vol. 274, no. 34, pp. 24124–24130, 1999.

[71] D. B. Bregman, R. Halaban, A. J. van Gool, K. A. Henning, E. C. Friedberg, and S. L. Warren, “UV-induced ubiquitination of RNA polymerase II: a novel modification deficient in Cockayne syndrome cells,” *Proceedings of the National Academy of Sciences of the United States of America*, vol. 93, no. 21, pp. 11586–11590, 1996.

[72] J. N. Ratner, B. Balasubramanian, J. Corden, S. L. Warren, and D. B. Bregman, “Ultraviolet radiation-induced ubiquitination and proteasomal degradation of the large subunit of RNA polymerase II,” *The Journal of Biological Chemistry*, vol. 273, no. 9, pp. 5184–5189, 1998.

[73] N. Mailand, S. Bekker-Jensen, H. Fastrup et al., “RNF8 ubiquitylates histones at DNA double-strand breaks and
promotes assembly of repair proteins,” Cell, vol. 131, no. 5, pp. 887–900, 2007.

[74] S. Bekker-Jensen, J. R. Danielsen, K. Fugger et al., “HERC2 coordinates ubiquitin-dependent assembly of DNA repair factors on damaged chromosomes,” Nature Cell Biology, vol. 12, no. 1, pp. 80–86, 2010.

[75] C. Doil, N. Mailand, S. Bekker-Jensen et al., “RNF168 binds and amplifies ubiquitin conjugates on damaged chromosomes to allow accumulation of repair proteins,” Cell, vol. 136, no. 3, pp. 435–446, 2009.

[76] G. Smeenk, W. W. Wiegant, J. A. Marteijn et al., “Poly(ADP-ribosyl)ation links the chromatin remodeler SMARCA5/SNF2H to RNF168-dependent DNA damage signaling,” Journal of Cell Science, vol. 126, Part 4, pp. 889–903, 2013.

[77] L. Feng and J. Chen, “The E3 ligase RNF8 regulates KU80 removal and NHEJ repair,” Nature Structural & Molecular Biology, vol. 19, no. 2, pp. 201–206, 2012.

[78] E. S. Johnson, “Protein modification by SUMO,” Annual Review of Biochemistry, vol. 73, no. 1, pp. 355–382, 2004.

[79] K. Ei, G. Smeenk, W. W. Wiegant, J. A. Marteijn et al., “Poly(ADP-ribosyl)ation links the chromatin remodeler SMARCA5/SNF2H to RNF168-dependent DNA damage signaling,” Journal of Cell Science, vol. 126, Part 4, pp. 889–903, 2013.

[80] J. R. Gareau and C. D. Lima, “The SUMO pathway: emerging mechanisms that shape specificity, conjugation and recognition,” Nature Reviews Molecular Cell Biology, vol. 11, no. 12, pp. 861–871, 2010.

[81] H. Dou, C. Huang, T. van Nguyen, L. S. Lu, and E. T. H. Yeh, “SUMOylation and de-SUMOylation in response to DNA damage,” FEBS Letters, vol. 585, no. 18, pp. 2891–2896, 2011.

[82] M. M. Madsen, N. C. Jones, S. B. Nielsen, and S. V. Hoffmann, “On the wavelength dependence of UV induced thymine photolesions: a synchrotron radiation circular dichroism study,” Physical Chemistry Chemical Physics, vol. 18, no. 44, pp. 30436–30443, 2016.

[83] R. M. Costa, V. Chigaças, S. Galhardo Rda, H. Carvalho, and C. F. Mencêk, “The eukaryotic nucleotide excision repair pathway,” Biochimie, vol. 85, no. 11, pp. 1083–1099, 2003.

[84] H. R. Silver, J. A. Nissley, S. H. Reed, Y. M. Hou, and E. S. Johnson, “A role for SUMO in nucleotide excision repair,” DNA Repair, vol. 10, no. 12, pp. 1243–1251, 2011.

[85] X. Zhao and G. Blobel, “From The Cover: A SUMO ligase is part of a nuclear multiprotein complex that affects DNA repair and chromosomal organization,” Proceedings of the National Academy of Sciences of the United States of America, vol. 102, no. 13, pp. 4777–9086, 2005.

[86] U. Hardeland, R. Steinacher, J. Jiricny, and P. Schär, “Modification of the human thymine-DNA glycosylase by ubiquitin-like proteins facilitates enzymatic turnover,” The EMBO Journal, vol. 21, no. 6, pp. 1456–1464, 2002.

[87] Q. E. Wang, Q. Z. Zhu, G. Wani, M. A. el-Mahdy, J. Li, and A. A. Wani, “DNA repair factor XPC is modified by SUMO-1 and ubiquitin following UV irradiation,” Nucleic Acids Research, vol. 33, no. 13, pp. 4023–4034, 2005.

[88] Q. E. Wang, M. Prætorius-Ibba, Q. Zhu et al., “Ubiquitylation-independent degradation of xeroderma pigmentosum group C protein is required for efficient nucleotide excision repair,” Nucleic Acids Research, vol. 35, no. 16, pp. 5338–5350, 2007.

[89] Y. Galanty, R. Belotserkovskaya, J. Coates, S. Polo, K. M. Miller, and S. P. Jackson, “Mammalian SUMO E3 ligases PIAS1 and PIAS4 promote responses to DNA double-strand breaks,” Nature, vol. 462, no. 7275, pp. 935–939, 2009.

[90] A. Zlatanou and G. S. Stewart, “A PIAS-ed view of DNA double-strand break repair focuses on SUMO,” DNA Repair, vol. 9, no. 5, pp. 588–592, 2010.

[91] X. Hu, A. Paul, and B. Wang, “Rap80 protein recruitment to DNA double-strand breaks requires binding to both small ubiquitin-like modifier (SUMO) and ubiquitin conjugates,” Journal of Biological Chemistry, vol. 287, no. 30, pp. 25510–25519, 2012.

[92] C. M. Guzzo, C. E. Berndsen, J. Zhu et al., “SUMO4-dependent hybrid SUMO-ubiquitin chains are signals for Rap80 and thereby mediate the recruitment of BRCA1 to sites of DNA damage,” Science Signal, vol. 5, no. 253, article ra88, 2012.

[93] K. T. Luo, H. X. Zhang, L. W. Wang, J. Yuan, and Z. Lou, “Sumoylation of MDC1 is important for proper DNA damage response,” EMBO Journal, vol. 31, no. 13, pp. 3008–3019, 2012.

[94] J. R. Morris, C. Boullé, M. Keppler et al., “The SUMO modification pathway is involved in the BRCA1 response to genotoxic stress,” Nature, vol. 462, no. 7275, pp. 886–890, 2009.

[95] R. Vyas, R. Kumar, F. Clermont et al., “RNF4 is required for DNA double-strand break repair in vivo,” Cell Death and Differentiation, vol. 20, no. 3, pp. 490–502, 2013.

[96] H. Dou, C. Huang, M. Singh, P. B. Carpenter, and E. T. H. Yeh, “Regulation of DNA repair through deSUMOylation and SUMOylation of replication protein a complex,” Molecular Cell, vol. 39, no. 3, pp. 333–345, 2010.

[97] J. R. Danielsen, L. K. Povlsen, B. H. Villumsen et al., “DNA damage-inducible SUMOylation of HERC2 promotes RNF8 binding via a novel SUMO-binding zinc finger,” The Journal of Cell Biology, vol. 197, no. 2, pp. 179–187, 2012.

[98] M. X. Qian, Y. Pang, C. H. Liu et al., “Acetylation-mediated proteasomal degradation of core histones during DNA repair and spermatogenesis,” Cell, vol. 153, no. 5, pp. 1012–1024, 2013.

[99] K. T. Nguyen, S. H. Mun, C. S. Lee, and C. S. Hwang, “Control of protein degradation by N-terminal acetylation and the N-end rule pathway,” Experimental & Molecular Medicine, vol. 50, no. 7, pp. 1–8, 2018.

[100] S. Thao, C. S. Chen, H. Zhu, and J. C. Escalante-Semerena, “Ne-Lysine acetylation of a bacterial transcription factor inhibits its DNA-binding activity,” PLoS One, vol. 5, no. 12, article e15123, 2010.

[101] B. S. Dwarakanath, A. Verma, A. N. Bhatt, V. S. Parmar, and H. G. Raj, “Targeting protein acetylation for improving cancer therapy,” The Indian Journal of Medical Research, vol. 128, no. 1, pp. 13–21, 2008.

[102] G. Spivak, “Nucleotide excision repair in humans,” DNA Repair, vol. 36, pp. 13–18, 2015.

[103] X. Wu, S. M. Shell, Z. Yang, and Y. Zou, “Phosphorylation of nucleotide excision repair factor xeroderma pigmentosum group a by ataxia telangiectasia mutated and Rad3-related-dependent checkpoint pathway promotes cell survival in response to UV irradiation,” Cancer Research, vol. 66, no. 6, pp. 2997–3005, 2006.

[104] W. Fan and J. Y. Luo, “SIRT1 regulates UV-induced DNA repair through deacetylating XPA,” Molecular Cell, vol. 39, no. 2, pp. 247–258, 2010.
Acetylation limits 53BP1 association with damaged chromatin to promote homologous recombination, "Nature Structural & Molecular Biology," vol. 20, no. 3, pp. 317–325, 2013.

[107] H. Ogiwara, A. Ui, A. Otsuka et al., "Histone acetylation by CBP and p300 at double-strand break sites facilitates SWI/SNF chromatin remodeling and the recruitment of non-homologous end-joining factors," "Oncogene," vol. 30, no. 18, pp. 2135–2146, 2011.

[108] H. S. Lee, J. H. Park, S. J. Kim, S. J. Kwon, and J. Kwon, "A cooperative activation loop among SWI/SNF, γ-H2AX and H3 acetylation for DNA double-strand break repair," "EMBO Journal," vol. 29, no. 8, pp. 1434–1445, 2010.

[109] E. Guccione and S. Richard, "The regulation, functions and clinical relevance of arginine methylation," "Nature Reviews Molecular Cell Biology," vol. 20, no. 10, pp. 642–657, 2019.

[110] C. Poulard, L. Corbo, and M. Le Romancer, "Protein arginine methylation/demethylation and cancer," "Oncotarget," vol. 7, no. 41, pp. 67532–67550, 2016.