Role of RING-Type E3 Ubiquitin Ligases in Inflammatory Signalling and Inflammatory Bowel Disease

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Ubiquitination is a three-step enzymatic cascade for posttranslational protein modification. It includes the ubiquitin-activating enzyme (E1), ubiquitin-conjugating enzyme (E2), and ubiquitin ligase (E3). RING-type E3 ubiquitin ligases catalyse the posttranslational proteolytic and nonproteolytic functions in various physiological and pathological processes, such as inflammation-associated signal transduction. Resulting from the diversity of substrates and functional mechanisms, RING-type ligases regulate microbe recognition and inflammation by being involved in multiple inflammatory signalling pathways. These processes also occur in autoimmune diseases, especially inflammatory bowel disease (IBD). To understand the importance of RING-type ligases in inflammation, we have discussed their functional mechanisms in multiple inflammation-associated pathways and correlation between RING-type ligases and IBD. Owing to the limited data on the biology of RING-type ligases, there is an urgent need to analyse their potential as biomarkers and therapeutic targets in IBD in the future.

1. Introduction

Ubiquitination is a crucial part of a diverse range of physiological and pathological processes, such as protein degradation and inflammation-associated signalling [1, 2]. It is a three-step enzymatic process that consists of ubiquitin-activating enzyme (E1), ubiquitin-conjugating enzyme (E2), and ubiquitin ligase (E3) [3]. E3 ligases transfer activated ubiquitin from E2 to specific substrates, thereby forming mono- or polyubiquitinated proteins to activate proteasome-mediated proteolysis, signal transduction, endocytosis, etc. [3]. E3 ligases are crucial as they catalyse target ubiquitination and enable the formation of polyubiquitin chains that enhance the complexity of ubiquitination in physiological and pathological processes. Although dysregulated ubiquitination is involved in the development of various types of immune pathologies (e.g., systemic lupus erythematosus, rheumatoid arthritis, and inflammatory bowel disease [IBD]) [1, 4], there is limited knowledge regarding the role of RING-type ligases in inflammation-associated pathways. In this review, we have focused on IBD owing to its complex pathogenesis involving a wide range of etiological factors, including dysregulated ubiquitination. The term IBD is used for a group of chronic autoimmune gastrointestinal disorders, including mainly Crohn’s disease (CD) and ulcerative colitis (UC) [5]. Chronicity of IBD often causes intestinal complications, hospitalisation, steroid dependency, and surgery in diagnosed patients [6]. Although significant progress has been made in understanding the nature of IBD, the underlying interacting mechanism involving ubiquitination, RING-type ligases, and onset of IBD remains to be fully understood.

2. Ubiquitination Mediated by RING-Type Ligases

Human cells express more than 600 E3 ubiquitin ligases that are classified into three types based on their catalytic domains: RING, HECT (homologous to the E6AP carboxyl terminus), and a recently identified RBR- (RING-between RING-RING-) type of E3 ligases [7, 8]. The RING domain has a crossbraced structure with two atoms of zinc that catalyse the direct transfer of ubiquitin from the E2-Ubiquitin
thioester to the substrate [9] (Figure 1). Apart from catalysing monoubiquitination, RING-type E3 ligases also elongate homotypic polyubiquitin chains with varying linkage specificities, such as that on Lys48 during the proteasomal targeting of substrates and Lys63 in signal transduction, thereby modulating proteins for proteolytic and nonproteolytic activity [10]. However, their roles in catalysing other (less common) types of ubiquitination, including atypical homotypic (e.g., Lys6, Lys11, Lys27, Lys29, Lys33, and Met1 [11]), heterotypic, and branched polyubiquitination, remain ambiguous. The efficiency of RING-type E3 ligases in ubiquitination depends on multiple factors, such as substrate modification, phosphorylation of E2/E3 enzymes, autoubiquitination by E3 ligases, and pseudosubstrate competition [10]. The role of the RING-type ligases and their sophisticated functional mechanism of ubiquitination will be discussed in the following sections.

3. Signalling Pathways Regulated by RING-Type Ligases

3.1. Pathogen Recognition. Under physiological conditions, pattern recognition receptors (PRRs) comprise toll-like receptors (TLRs), retinoic acid inducible gene- (RIG-) I-like receptors (RLRs), C-type lectin-like receptors, and nucleotide-binding oligomerisation domain-like receptors. PRRs recognise pathogen-associated molecular patterns (PAMPs) and trigger the activation of downstream effectors in innate immune responses [12]. For inflammatory diseases that are closely associated with microbiome dysbiosis, such as IBD [13, 14], dysregulation of PRRs and relevant RING-type ligases may be involved in pathogen-induced inflammation.

3.1.1. TLR Signalling. Upon recognising a wide range of microbial components, such as lipopolysaccharides, flagellin, and microbial nucleic acids, activated TLRs expressed on antigen-presenting cells trigger effector T cell responses in inflammatory diseases [15–17]. RING-type ligases modulate the activation of PAMP-induced TLRs. By directly binding with TLR3, ring finger protein 170 (RNF170) catalyses the Lys48-linked ubiquitination and proteasomal degradation of TLR3, thereby suppressing TLR3-mediated innate immunity in macrophages [18]. On the other hand, Fcγ receptor (FcγR) Iib, an inhibitory FcR on antibody-dependent monocyte phagocytosis, is targeted by MARCH3 (RNF173) for ubiquitination and degradation in lipopolysaccharide-induced TLR4 activation [19].

3.1.2. RIG-I Signalling. RIG-I is a cytoplasmic PRR that recognises viral RNA and triggers the activation of downstream immune responses that are associated with both viral infections and noninfectious autoimmune diseases, such as enterocolitis [20]. The deficiency of RIG-I aggravates virus-induced cell death in intestinal epithelial cells and induces susceptibility to chemically induced colitis in mice, suggesting the importance of RIG-I signalling in intestinal antiviral immune response [20, 21]. E3 ligases, like RNF122 and RNF125, mediate Lys48-linked RIG-I ubiquitination and proteasomal degradation, leading to the reduced expression of infection induced-proinflammatory cytokines, including IL-6 and type I interferons (α and β) [22, 23]. In contrast, independent of its E3 ligase activity, RNF123 binds with the CARD domain of RIG-I and melanoma differentiation-associated gene 5 to compete with the mutual downstream adaptor mitochondrial antiviral signalling protein (MAVS) and inhibit RLR-mediated antiviral response [24]. Unlike the aforementioned RING-type ligases that directly target RLRs, RNF114 negatively regulates RLR signalling by polyubiquitinating and inducing the proteasomal degradation of MAVS [25].

3.2. Proinflammatory Pathways

3.2.1. Nuclear Factor Kappa B (NF-κB) Signalling. The NF-κB pathway is one of the most well-studied proinflammatory pathways regulated by ubiquitination [26]. TRAF6 (RNF85) ubiquitinates the evolutionarily conserved signalling intermediate in TLR activation that is essential for TLR4-dependent NF-κB activation [27]. RNF183 promotes NF-κB signalling by inducing the ubiquitin-dependent degradation of 1xBa [28]. TRAF2 (RNF117) and TRAF3 (RNF118) induce Lys48-linked ubiquitination and proteasomal degradation of c-Rel and interferon regulatory factor 5, thereby prohibiting the synthesis of proinflammatory cytokines in macrophages [29]. MKN2 (RNF62) mediates the polyubiquitination and degradation of the p65 subunit of NF-κB, thereby inhibiting NF-κB signalling [30]. RNF114 negatively regulates NF-κB signalling and T cell activation by ubiquitinating and stabilising NF-κB signalling inhibitors A20 and 1xBa [31, 32]. It has also been reported that RNF20 downregulation decreases histone H2B monoubiquitination and leads to the NF-κB-dependent transcription of proinflammatory cytokines, such as IL-6 and IL-8 [33]. Nevertheless, despite the formation of heterodimers of RNF40 with RNF20, RNF40 alone activates NF-κB signalling and upregulates NF-κB-dependent transcription by promoting 1xK kinase (IKK) phosphorylation and p65 nuclear translocation, indicating the involvement of NF-κB-dependent transcription in the ubiquitination of substrates other than H2B [34]. MARCH3 (RNF173) mediates Lys48-linked ubiquitination and lysosomal degradation of IL-1 receptor I and thereby inhibits IL-1β-triggered NF-κB activation [35]. Independent of its E3 ligase activity, RNF8 inhibits TNF-α-induced NF-κB activation by directly binding with the kinase domain of IKK and interfering with IκKα/β phosphorylation [36]. RNF1 also exerts a noncanonical role in negatively regulating NF-κB signalling. RNF11 has high affinity for the E2 enzyme Ubc13 and minimal E3 ligase activity that subsequently outcompetes E1 enzymes and other E3 enzymes, such as TRAF6 [37], and impedes the activation of NF-κB signalling [38].

3.2.2. Mitogen-Activated Protein Kinase (MAPK) Signalling. MAPKs are another family of proteins closely related to inflammation-associated pathologies, such as IBD [39]. Upon stimulation by TNF-α, TRAF2 is autoubiquitinated on the Lys63 residue that enables its translocation to the cytoskeletal fraction and activates JNK signalling [40, 41]. In vitro experiments have shown that JNK signalling is suppressed and enhanced in cells overexpressing RNF213
and RNF186, respectively [42, 43]; however, the mechanisms involved in this regulation remain to be understood. TRAF7 (RNF119) upregulates the kinase activity of mitogen-activated protein kinase 3 via the WD40 domain and potentiates cell apoptosis via the RING finger domain [44]. Similarly, RNF13 mediates endoplasmic reticulum (ER) stress-induced JNK activation and subsequent cell apoptosis by binding with and promoting the phosphorylation of the ER stress sensor endoplasmic reticulum to nucleus signalling 1 [45].

3.2.3. Janus Kinase (JAK)/Signal Transducer and Activator of Transcription 3 (STAT3) Signalling. JAK/STAT3 is one of the major proinflammatory signalling pathways that orchestrate

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**Figure 1**: Functional mechanisms of RING-type E3 ligase. Acting as a scaffolding, RING-type E3 ligase recruits a E2-ubiquitin thioester and a substrate and allows the lysine of substrate to attack the thioester for ubiquitin transfer. RING-type E3 ligases catalyse monoubiquitination or multiple monoubiquitination by transferring a single ubiquitin to one or several residues of the substrate. For polyubiquitination, ubiquitins form eight different linkage types including Met1, Lys6, Lys11, Lys27, Lys29, Lys33, Lys48, and Lys63. Apart from homotypic chains, RING-type E3 ligases also catalyse heterotypic chains and branched ubiquitin chains by adopting multiple linkage types and branched topology in the formation of polyubiquitin chains.
the progression of inflammatory and autoimmune diseases [46]. A number of RING-type ligases modulate JAK/STAT3 signalling. RNF6 and RNF38 function in catalysing the ubiquitination-induced proteasomal degradation of SH2-containing protein tyrosine phosphatase 1 that targets phosphorylated STAT3, thereby maintaining STAT3 phosphorylation and activating STAT3 signalling [47, 48]. In contrast, TRAF6 promotes Lys63-linked ubiquitination of STAT3 and represses STAT3-mediated transcription of downstream inflammation-related genes, such as C-reactive protein [49]. Interestingly, RNF41 modulates the cell surface expression of JAK2-associated cytokine receptors by blocking the cleavage of receptors and enhancing receptor shedding in a ubiquitination-dependent manner [50].

3.2.4. Phosphatidylinositol 3-Kinase (PI3K) Signalling. PI3K is another classical pathway involved in inflammation wherein RING-type ligases are of crucial importance. MKRN1 (RNF61) functions in the positive-feedback regulation of sustained PI3K/AKT activation upon stimulation by epidermal growth factor: AKT activation phosphorylates and stabilises the subunit of PI3K and downregulated AKT α [51]. MKRN2 (RNF62) induces the ubiquitin-dependent degradation of the p85α subunit of PI3K and downregulated AKT phosphorylation, suggesting a negative regulatory role of MKRN2 in PI3K/AKT signalling [52]. Downregulation of UHRF1 (RNF106) represses the phosphorylation of PI3K and AKT, which reveals an underlying interaction between UHRF1 and PI3K/AKT signalling [53].

3.3. Transforming Growth Factor-β (TGF-β) Signalling. TGF-β signalling functions in immunosuppression and inhibiting the activity of effector T cells, maintaining Treg differentiation, reducing B cell responsiveness, and inducing macrophage anergy [54]. RNF11 plays a dual role in the modulation of TGF-β signalling. By competing with Smad7 for Smurf2, RNF11 is a positive regulator for TGF-β signalling and reduces the formation of Smad7/Smurf2 complexes that degrade TGF-β receptors [55]. RNF11 is also responsible for the ubiquitination-mediated stabilisation of Smad4 that enhances Smad4-dependent TGF-β signalling by direct interaction [56]. Notably, RNF11 may negatively regulate TGF-β signalling by enabling the formation of Smurf2/RNF11 complexes and inducing the ubiquitination and degradation of the associated molecule with the SH3 domain of STAM that promotes TGF-β signalling [57]. PRAJA (RNF70) mediates the ubiquitination-induced proteasomal degradation of embryonic liver fodrin (a Smad4 adaptor protein), thereby negatively regulating TGF-β signalling [58].

3.4. Autophagy. Accumulating evidence reveals that autophagy contributes extensively to immune cell development and cell death, and its dysregulation has been implicated in many autoimmune diseases [59]. TRAF6 catalyses Lys63-linked ubiquitination of Lys63-linked ubiquitination of BECN1 and stimulates TLR-induced autophagy in macrophages upon proinflammatory stimulation [27, 60]. RNF166 has a novel role in antibacterial host defence owing to its function in inducing the Lys29- and Lys33-linked ubiquitination of autophagy adaptor p62, which mediates the recruitment of p62 to bacteria and initiates bacteria engulfment [61].

3.5. Noncoding RNAs. Since dysregulated noncoding RNAs are involved in the progression of inflammatory diseases [62], the posttranscriptional regulation of RING-type ligases by noncoding RNAs may play critical roles in potential inflammation-relevant signalling pathways. Until now, quite a few microRNAs have been proved to posttranscriptionally regulate RING-type ligases by hampering translation or inducing mRNA degradation (Table 1) [28, 63–71]. Nevertheless, although the other two major types of noncoding RNAs (long noncoding RNAs and circular RNAs) also have diverse functions in inflammatory diseases (e.g., competing endogenous RNA [ceRNA], transcription regulation, and RNA-binding protein sponges) [72, 73], to what extent they modulate RING-type ligases awaits further analysis.

4. RING-Type Ligases in IBD

4.1. Pathogenesis of IBD. The pathogenesis of IBD has been elucidated over the past years. More than 200 loci have been implicated in increased genetic risk for IBD that correlate with the functioning of cellular processes, such as innate/adaptive immune response, intestinal mucosal barrier homeostasis,
and autophagy, suggesting the involvement of multiple factors in shaping the procolitogenic environment during the development of IBD [74, 75]. IBD patients manifest with abnormalities in the composition of gut microbiota, such as decreased bacterial diversity, increased proportion of harmful bacterial strains, and decreased proportion of protective probiotics, which trigger proinflammatory intestinal pathogenic immune responses and contribute to the pathogenesis of IBD [14, 76]. Elevated levels of proinflammatory cytokines (e.g., IL-1, IL-6, and IL-23) and activation of adaptive (e.g., Th1, Th2, Th9, and Th17 cells) and innate immune cells (e.g., neutrophils and NK cells) constitute a synergistic inflammatory network that induces intestinal mucosal inflammation and sustained activation of multiple proinflammatory signalling pathways [17, 77]. IL-1 family-induced NF-xB, IL-6-induced STAT3, MAPK, and PI3K signalling pathways are pivotal in intestinal inflammation [39, 78, 79]. TGF-β signalling can mitigate immune cell hyperactivation but also causes the formation of intestinal strictures in chronic intestinal inflammation [54]. Autophagy is involved in the regulation of immune cell function; thus, defective autophagy also plays an important role in IBD pathogenesis [59]. A dysfunctional gut barrier and subsequent increased intestinal permeability are also considered important etiologic factors in the development of IBD that result in the uncontrolled exchange of materials between the intestinal lumen and internal environment. A compromised gut barrier is often attributed to the proinflammatory stimulation and subsequent downregulation of sealing tight junction proteins (e.g., claudin-5 and claudin-8) and upregulation of pore-forming tight junction proteins (e.g., claudin-2) [80–83]. Since multiple etiologic factors function in the pathogenesis of IBD in a synergistic manner, further research is warranted to understand the dynamics between these players.

### 4.2. Role of RING-Type Ligases in IBD

Despite the limited knowledge on RING-type ligases in IBD, research suggests a correlation between RING-type ligases and IBD pathogenesis (Table 2). Genome-wide association studies have identified RNF186 as one of the genes associated with susceptibility to UC; the disease-coding variant of RNF186 involves an altered RING domain [84, 85]. The truncated RNF186 lacking the second transmembrane domain is associated with protecting individuals against developing UC by inhibiting the ER localisation of RNF186 and subsequent Lys29- and Lys63-linked polyubiquitination of proapoptotic BCL2 interacting protein 1 under ER stress [86, 87]. Notably, RNF186 functions differently in a dextran sulfate sodium- (DSS-) induced mouse model of colitis: RNF186-deficient mice

| RING-type ligase | Role | IBD patients | Animal model of colitis | Reference |
|------------------|------|--------------|------------------------|-----------|
| RNF186           | Controversial | Increase risk of UC and ER stress-induced apoptosis | Attenuate ER stress and maintain intestinal permeability in DSS-induced mouse model of colitis | [84–88] |
| RNF20            | Anti-inflammatory | Decrease in the colonic tissue from UC patients | Protect mice from DSS-induced colitis and maintain intestinal barrier | [33] |
| RNF40            | Proinflammatory | — | Activate NF-xB signalling in DSS-induced mouse model of colitis | [34] |
| RNF183           | Proinflammatory | Increase in the colonic tissue from CD and UC patients correlate with endoscopic index of disease severity | Increase in TNBS-induced mouse model of colitis | [28] |
| UHRF1            | Anti-inflammatory | — | Regulate TNF-α expression in mice with DSS-induced colitis and zebrafish | [89–94] |
| TRAF2            | Anti-inflammatory | Increase in the colonic tissue from CD and UC patients | Modulate the proliferation and differentiation of Treg cells and colonic microbiota composition, proinflammatory cytokine expression, and immune cell infiltration in mice with DSS-induced colitis | [29, 96, 97] |
| TRAF3            | Anti-inflammatory | Increase in the plasma and colonic tissue from CD and UC patients | Regulate proinflammatory cytokine expression and mitigate inflammatory damage in DSS-induced mouse model of colitis | [29, 95, 98] |
| TRAF5            | Anti-inflammatory | Increase in the plasma and colonic tissue from CD and UC patients | Control proinflammatory cytokine expression and protect mice against experimental colitis | [99–101] |
develop more severe colitis during DSS administration, and their colonic epithelial cell exhibits enhanced signs of ER stress and apoptosis [88]. RNF186 also modulates intestinal barrier function by mediating the Lys48-linked ubiquitination of tight junction protein occludin, and RNF186 deficiency increases intestinal permeability in RNF186 knockout mice [88]. Thus, RNF186 targets different substrates and has a complex association with gut inflammation.

Apart from the reduced expression of RNF20 in the colon samples from UC patients, homozygous RNF20-knockout mice die due to embryonic lethality, and heterozygous mice are susceptible to DSS-induced colitis with increased intestinal permeability, suggesting the anti-inflammatory role of RNF20 [33]. RNF40 knockout mice exhibit mitigated gut inflammation upon treatment with DSS; this can be attributed to the attenuated activation of NF-κB signalling [34]. The upregulation of RNF183 in the colonic tissue from IBD patients and 2,4,6-trinitrobenzenesulfonic acid- (TNBS-) induced mouse model of colitis indicates its proinflammatory function, probably by promoting the ubiquitination-induced degradation of 1βx [28].

Because UHRF1-catalysed histone H3 monoubiquitination recruits and stimulates DNA methyltransferase 1 to DNA methylation sites, and thereby maintains DNA methylation, UHRF1 participates in the epigenetic control of multiple genes, such as TNF-α [89, 90]. Mice with macrophages deficient for UHRF1 manifest with TNF-α overexpression and aggravated DSS-induced colitis. Also, the loss of function in UHRF1 reduces the methylation of the TNF-α promoter in macrophages, indicating the regulatory role of UHRF1 in the mouse model of colitis [91]. Similarly, an in vivo study in zebrafish has revealed that loss in function of UHRF1 leads to defects in the epigenetic regulation of TNF-α promoter methylation and elicits elevated TNF-α expression in inflammatory processes, including intestinal epithelial cell apoptosis, neutrophil recruitment, and weakened intestinal barrier function [92]. However, UHRF1 may affect differently among subtypes of regulatory T (T reg) cells, as UHRF1 maintains the proliferation and maturation of colonic T reg cells but inhibits the differentiation of peripheral induced T reg cells in the development of colitis [93, 94].

Studies have shown the diverse roles involved with the upregulation of TRAFs, including TRAF1/2/3/5, in the blood or colonic mucosa of IBD patients [95, 96]. DSS-induced colitis models of TRAF2- and TRAF3-deficient mice reveal similar functions of TRAF2 and TRAF3 as negative regulators of experimental colitis by decreasing proinflammatory cytokines and reducing the infiltration of immune cells in the colon [29]. In another study, TRAF2-deficient mice develop severe spontaneous colitis and exhibit altered colonic microbiota composition, indicating the anti-inflammatory role of TRAF2 in controlling colonic microbiota [97]. TRAF3 also acts as a colitis regulator by binding with the IL-17 receptor and interfering with the IL-17-mediated proinflammatory pathway in mice with TNBS-induced colitis [98]. Although TRAF5 (RNF84) promotes the ubiquitination and stabilisation of the retinoic acid-related orphan receptor γt that mediates proinflammatory Th17 cell differentiation and IL17A/IL17F expression [99, 100], TRAF5-deficient mice exhibit aggravated experimental colitis and upregulation of proinflammatory cytokines [101]. The complex function of TRAF5 needs further analysis.

5. Discussion

As the importance of RING-type E3 ligases is gradually unveiled, there are still problems to be solved. Firstly, apart from the canonical role of RING-type ligases in modulating key signalling pathways and their downstream adaptors as E3 ubiquitin ligases, some RING-type ligases interfere with the ubiquitination cascade by competition or direct interaction with other E3 ligases [102]. Owing to the variety of RING-type ligases and substrate specificity of E3 ligases, the potential competition among RING-type ligases in regulating immune response remains to be fully understood. Secondly, differences in RING E3 ligase-mediated target ubiquitination can also be attributed to the variance in length and linkage type of ubiquitin chains. Although RING-type ligase function in proteolytic degradation and signal transduction by catalysing Lys48-linked and Lys63-linked ubiquitination, respectively, their roles in catalysing less common linkage types of homotypic polyubiquitin chains, such as Lys11-linked ubiquitination [103], and the outcome of such polyubiquitination are still obscure.

RING-type ligases form a sophisticated but important ubiquitination network, wherein the expression and function of RING-type ligases are also influenced reciprocally in physiological and pathological processes. Understanding the mechanisms employed by RING-type ligases in modulating inflammation-associated pathways by catalysing atypical linkages and affecting signal transduction may further explain the interaction between RING-type ligases and IBD. Similarly, research on heterotypic polyubiquitin chains is also important to unveil these underlying mechanisms.

In this review, we have highlighted the roles of RING-type ligases in PAMP recognition and modulation of inflammation-associated pathways that are crucial etiological factors in the development of autoimmune diseases. Accumulating evidence shows that many RING-type ligases are involved in inflammation-associated pathways, such as proinflammatory NF-κB, MAPK, JAK/STAT3, and PI3K signalling and anti-inflammatory TGF-β signalling. Subsequently, we have discussed the role of RING-type ligases in the pathogenesis of IBD via inflammation-related pathways. Patients with IBD exhibit the differential expression of specific RING-type ligases, such as TRAFs. However, there are limited studies on the potential clinical value of RING-type ligases in predicting or treating IBD. Thus far, there have been a few attempts to use RING-type ligases as predictive biomarkers and therapeutic targets in treating cancer; RNF43 modulates Wnt signalling and has been used to target colorectal cancer and pancreatic ductal carcinoma [104, 105]. Nevertheless, the potential of RING-type ligases in autoimmune diseases, especially in IBD, needs to be understood in greater detail. Therefore, future research on the expression profile of RING-type ligases in the gastrointestinal tract and the detailed mechanisms is warranted.
Conflicts of Interest

The authors have no conflict of interest to disclose.

Authors’ Contributions

The guarantor of the article is Shenghong Zhang. Shenghong Zhang designed the study. Shenghong Zhang, Liguo Zhu, Ying Li, and Longyuan Zhou wrote and revised the manuscript. Guang Yang, Ying Wang, Jing Han, and Li Li revised the important intellectual content of the manuscript. All the authors approved the final version. Liguo Zhu, Ying L, and Longyuan Zhou contributed equally to the work.

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References

[1] D. Popovic, D. Vucic, and I. Dikic, "Ubiquitination in disease pathogenesis and treatment," *Nature Medicine*, vol. 20, no. 11, pp. 1242–1253, 2014.
[2] D. Aki, Q. Li, H. Li, Y. C. Liu, and J. H. Lee, "Immune regulation by protein ubiquitination: roles of the E3 ligases VHL and itch," *Protein & Cell*, vol. 10, no. 6, pp. 395–404, 2019.
[3] C. M. Pickart, "Mechanisms underlying ubiquitination," *Annual Review of Biochemistry*, vol. 70, no. 1, pp. 503–533, 2001.
[4] H. Hu and S. S. C. Sun, "Ubiquitin signaling in immune responses," *Cell Research*, vol. 26, no. 4, pp. 457–483, 2016.
[5] R. Hodson, "Inflammatory bowel disease," *Nature*, vol. 540, no. 7634, p. 597, 2016.
[6] L. Peyrin-Biroulet, E. V. Loftus Jr., J. F. Colombel, and W. J. Sandborn, "The natural history of adult Crohn’s disease in population-based cohorts," *The American Journal of Gastroenterology*, vol. 105, no. 2, pp. 289–297, 2010.
[7] N. Zheng and N. Shabek, "Ubiquitin ligases: structure, function, and regulation," *Annual Review of Biochemistry*, vol. 86, no. 1, pp. 129–157, 2017.
[8] F. E. Morreale and H. Walden, "Types of ubiquitin ligases," *Cell*, vol. 165, no. 1, pp. 248–248.e1, 2016.
[9] C. E. Berndsen and C. Wolberger, "New insights into ubiquitin E3 ligase mechanism," *Nature Structural & Molecular Biology*, vol. 21, no. 4, pp. 301–307, 2014.
[10] R. J. Deshaies and C. A. P. Joazeiro, "RING domain E3 ubiquitin ligases," *Annual Review of Biochemistry*, vol. 78, no. 1, pp. 399–434, 2009.
[11] Y. Kulathu and D. Komander, "Atypical ubiquitylation - the unexplored world of polyubiquitin beyond Lys48 and Lys63 linkages," *Nature Reviews. Molecular Cell Biology*, vol. 13, no. 8, pp. 508–523, 2012.
[12] S. Akira, "Innate immunity to pathogens: diversity in receptors for microbial recognition," *Immunological Reviews*, vol. 227, no. 1, pp. 5–8, 2009.
[13] X. C. Morgan, T. L. Tickle, H. Sokol et al., "Disfunction of the intestinal microbiome in inflammatory bowel disease and treatment," *Genome Biology*, vol. 13, no. 9, p. R79, 2012.
[14] R. B. Sartor and G. D. Wu, "Roles for intestinal bacteria, viruses, and fungi in pathogenesis of inflammatory bowel diseases and therapeutic approaches," *Gastroenterology*, vol. 152, no. 2, pp. 327–339.e4, 2017.
[15] T. Kawai and S. Akira, "The role of pattern-recognition receptors in innate immunity: update on toll-like receptors," *Nature Immunology*, vol. 11, no. 5, pp. 373–384, 2010.
[16] J. Q. Chen, P. Szodoray, and M. Zeher, "Toll-like receptor pathways in autoimmune diseases," *Clinical Reviews in Allergy and Immunology*, vol. 50, no. 1, pp. 1–17, 2016.
[17] J. H. Park, L. Peyrin-Biroulet, M. Eisenhut, and J. I. Shin, "IBD immunopathogenesis: a comprehensive review of inflammatory molecules," *Autoimmunity Reviews*, vol. 16, no. 4, pp. 416–426, 2017.
[18] X. Song, S. Liu, W. Wang, Z. Ma, X. Cao, and M. Jiang, "E3 ubiquitin ligase RNF170 inhibits innate immune responses by targeting and degrading TLR3 in murine cells," *Cellular & Molecular Immunology*, vol. 17, no. 8, pp. 865–874, 2020.
[19] K. Fatechand, L. Ren, S. Elavazhagan et al., "Toll-like receptor 4 ligands down-regulate Fcy receptor IIb (FcyRllb) via MARCH3 protein-mediated ubiquitination," *The Journal of Biological Chemistry*, vol. 291, no. 8, pp. 3895–3904, 2016.
[20] T. Matsumiya and D. M. Stafforini, "Function and regulation of retinoic acid-inducible gene-I," *Critical Reviews in Immunology*, vol. 30, no. 6, pp. 489–513, 2010.
[21] Y. Hirata, A. H. Broquet, L. Menchén, and M. F. Kagnoff, "Activation of innate immune defense mechanisms by signaling through RIG-I/IPS-1 in intestinal epithelial cells," *Journal of Immunology*, vol. 179, no. 8, pp. 5425–5432, 2007.
[22] W. Wang, M. Jiang, S. Liu et al., "RNF122 suppresses antiviral type I interferon production by targeting RIG-I CARDs to mediate RIG-I degradation," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 113, no. 34, pp. 9581–9586, 2016.
[23] K. Arimoto, H. Takahashi, T. Hishiki, H. Konishi, T. Fujita, and K. Shimotohno, "Negative regulation of the RIG-I-signalizing by the ubiquitin ligase RNF125," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 104, no. 18, pp. 7500–7505, 2007.
[24] S. Wang, Y. K. Yang, T. Chen et al., "RNF123 has an E3 ligase-independent function in RIG-I-like receptor-mediated antiviral signaling," *EMBO Reports*, vol. 17, no. 8, pp. 1155–1168, 2016.
[25] B. Lin, Q. Ke, H. Li, N. S. Pheifer, D. C. Velliquette, and D. W. Leaman, "Negative regulation of the RLH signaling by the E3 ubiquitin ligase RNF114," *Cytokine*, vol. 99, pp. 186–193, 2017.
[26] J. Chen and Z. J. Chen, "Regulation of NF-κB by ubiquitination," *Current Opinion in Immunology*, vol. 25, no. 1, pp. 4–12, 2013.
[27] Y. Min, M. J. Kim, S. Lee, E. Chun, and K. Y. Lee, "Inhibition of TRAF6 ubiquitin-ligase activity by PRDX1 leads to inhibition of NFKB activation and autophagy activation," *Autophagy*, vol. 14, no. 8, pp. 1347–1358, 2018.
[28] Q. Yu, S. Zhang, K. Chao et al., "E3 ubiquitin ligase RNF183 is a novel regulator in inflammatory bowel disease".
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disease,” *Journal of Crohn’s & Colitis*, vol. 10, no. 6, pp. 713–725, 2016.

[29] J. Jin, Y. Xiao, H. Hu et al., “Proinflammatory TLR signalling is regulated by a TRAF2-dependent proteolysis mechanism in macrophages,” *Nature Communications*, vol. 6, no. 1, article 5930, 2015.

[30] C. Shin, Y. Ito, S. Ichikawa, M. Tokunaga, K. Sakata-Sogawa, and T. Tanaka, “MKRN2 is a novel ubiquitin E3 ligase for the p65 subunit of NF-κB and negatively regulates inflammatory responses,” *Scientific Reports*, vol. 7, no. 1, article 46097, 2017.

[31] M. S. Rodriguez, I. Egaña, F. Lopitz-Otsoa et al., “The RING ubiquitin E3 RNF114 interacts with A20 and modulates NF-κB activity and T-cell activation,” *Cell Death & Disease*, vol. 5, no. 8, article e1399, 2014.

[32] K. Heyninck and R. Beyaert, “A20 inhibits NF-κB activation by dual ubiquitin-editing functions,” *Trends in Biochemical Sciences*, vol. 30, no. 1, pp. 1–4, 2005.

[33] O. Tarcic, I. S. Pateras, T. Cooks et al., “RNF20 links histone H2B ubiquitylation with inflammation and inflammation-associated cancer,” *Cell Reports*, vol. 14, no. 6, pp. 1462–1476, 2016.

[34] R. L. Kosinsky, R. L. Chua, M. Qui et al., “Loss of RNF40 decreases NF-κB activity in colorectal cancer cells and reduces colitis burden in mice,” *Journal of Crohn’s & Colitis*, vol. 13, no. 3, pp. 362–373, 2019.

[35] H. Lin, D. Gao, M. M. Hu et al., “MARCH3 attenuates IL-1β-triggered inflammation by mediating K48-linked polyubiquitination and degradation of IL-1R,” *Proceedings of the National Academy of Sciences of the United States of America*, vol. 115, no. 49, pp. 12483–12488, 2018.

[36] S. Gao, J. Wu, L. Liang, and R. Xu, “RNF8 negatively regulates NF-κB activation by targeting IkappaB kinase: implications for the regulation of inflammation signaling,” *Biochemical and Biophysical Research Communications*, vol. 488, no. 1, pp. 189–195, 2017.

[37] R. Budhidarma, J. Zhu, A. J. Middleton, and C. L. Day, “TheRINGdomain ofRINGFinger 11 (RNF11) protein binds Ubc13 and inhibits formation of polyubiquitin chains,” *FEBS Letters*, vol. 592, no. 8, pp. 1434–1444, 2017.

[38] N. V. Dalal, E. L. Pranski, M. G. Tansley, J. J. Lah, A. I. Levey, and R. S. Betarbet, “RNF11 modulates microglia activation through NF-κB signalling cascade,” *Neuroscience Letters*, vol. 528, no. 2, pp. 174–179, 2012.

[39] O. J. Broom, B. Widjaya, J. Troelsen, J. Olsen, and O. H. Nielsen, “Mitogen activated protein kinases: a role in inflammatory bowel disease?,” *Clinical and Experimental Immunology*, vol. 158, no. 3, pp. 272–280, 2009.

[40] H. Habelhah, S. Takahashi, S. G. Cho, T. Kadoya, T. Watanabe, and Z. Ronai, “Ubiquitination and translocation of TRAF2 is required for activation of JNK but not of p38 or NF-κB,” *The EMBO Journal*, vol. 23, no. 2, pp. 322–332, 2004.

[41] J. Liu, J. Yan, S. Jiang et al., “Site-specific ubiquitination is required for relieving the transcription factor Miz1-mediated suppression on TNF-α-induced JNK activation and inflammation,” *Proceedings of the National Academy of Sciences of the United States of America*, vol. 109, no. 1, pp. 191–196, 2012.

[42] X. Wang, M. Ye, M. Wu et al., “RNF213 suppresses carcinogenesis in glioblastoma by affecting MAPK/JNK signaling pathway,” *Clinical & Translational Oncology*, vol. 22, no. 9, pp. 1506–1516, 2020.

[43] X. Tong, Q. Zhang, L. Wang et al., “RNF186 impairs insulin sensitivity by inducing ER stress in mouse primary hepatocytes,” *Cellular Signalling*, vol. 52, pp. 155–162, 2018.

[44] L. G. Xu, L. Y. Li, and H. B. Shu, “TRAF7 potentiates MEKK3-induced AP1 and CHOP activation and induces apoptosis,” *The Journal of Biological Chemistry*, vol. 279, no. 17, pp. 17278–17282, 2004.

[45] M. Arshad, Z. Ye, X. Gu et al., “RNF13, a RING finger protein, mediates endoplasmic reticulum stress-induced apoptosis through the inositol-requiring enzyme (IRE1α)/c-Jun NH2-terminal kinase pathway,” *The Journal of Biological Chemistry*, vol. 288, no. 12, pp. 8726–8736, 2013.

[46] Y. T. Yeung, F. Aziz, A. Guerrero-Castilla, and S. Arguelles, “Signaling pathways in inflammation and anti-inflammatory therapies,” *Current Pharmaceutical Design*, vol. 24, no. 14, pp. 1449–1484, 2018.

[47] Q. Liang, D. Ma, X. Zhu et al., “RING-finger protein 6 amplification activates JAK/STAT3 pathway by modifying SHP-1 ubiquitylation and associates with poor outcome in colorectal cancer,” *Clinical Cancer Research*, vol. 24, no. 6, pp. 1473–1485, 2018.

[48] J. Zhang, H. Wu, B. Yi et al., “RING finger protein 38 induces gastric cancer cell growth by decreasing the stability of the protein tyrosine phosphatase SHP-1,” *FEBS Letters*, vol. 592, no. 18, pp. 3092–3100, 2018.

[49] J. Wei, Y. Yuan, C. Jin et al., “The ubiquitin ligase TRAF6 negatively regulates the JAK-STAT signalling pathway by binding to STAT3 and mediating its ubiquitination,” *PLoS One*, vol. 7, no. 11, article e95967, 2012.

[50] J. Wauman, L. de Cevenick, N. Vanderroost, S. Lievens, and J. Tavernier, “RNF41 (Nrdp1) controls type 1 cytokine receptor degradation and ectodomain shedding,” *Journal of Cell Science*, vol. 124, no. 6, pp. 921–932, 2011.

[51] M. S. Lee, M. H. Jeong, H. W. Lee et al., “PI3K/AKT activation induces PTEN ubiquitination and destabilization accelerating tumourigenesis,” *Nature Communications*, vol. 6, no. 1, article 7769, 2015.

[52] J. Jiang, Y. Xu, H. Ren et al., “MKRN2 inhibits migration and invasion of non-small-cell lung cancer by negatively regulating the PI3K/Akt pathway,” *Journal of Experimental & Clinical Cancer Research*, vol. 37, no. 1, pp. 189, 2018.

[53] Y. Liu, G. Liang, T. Zhou, and Z. Liu, “Silencing UHRF1 inhibits cell proliferation and promotes cell apoptosis in retinoblastoma via the PI3K/Akt signalling pathway,” *Pathology Oncology Research*, vol. 26, no. 2, pp. 1079–1088, 2020.

[54] S. Ihara, Y. Hirata, and K. Koike, “The ubiquitin ligase RNF11 is overexpressed in breast cancer and is a target of SmurF2 E3 ligase,” *British Journal of Cancer*, vol. 89, no. 8, pp. 1538–1544, 2003.

[55] S. Azmi and A. K. Seth, “The RING finger protein11 binds to Smad4 and enhances Smad4-dependant TGF-beta signalling,” *Anticancer Research*, vol. 29, no. 6, pp. 2253–2263, 2009.

[56] H. Li and A. Seth, “An RNF11: Smurf2 complex mediates ubiquitination of the AMSH protein,” *Oncogene*, vol. 23, no. 10, pp. 1801–1808, 2004.
[58] L. Mishra, V. Katuri, and S. Evans, “The role of PRAJA and ELF in TGF-β signaling and gastric cancer,” Cancer Biology & Therapy, vol. 4, no. 7, pp. 694–699, 2014.

[59] Y. Matsuzawa-Ishimoto, S. Hwang, and K. Cadwell, “Autophagy and inflammation,” Annual Review of Immunology, vol. 36, no. 1, pp. 73–101, 2018.

[60] C. S. Shi and J. H. Kehrl, “TRA6 and A20 regulate lysine 63-linked ubiquitination of beclin-1 to control TLR4-induced autophagy,” Science Signaling, vol. 3, no. 123, p. ra42, 2010.

[61] R. J. Heath, G. Goel, L. A. Baxt et al., “RNFi66 determines recruitment of adaptor proteins during antibacterial autophagy,” Cell Reports, vol. 17, no. 9, pp. 2183–2194, 2016.

[62] J. L. Marques-Rocha, M. Sambas, F. I. Milagro, J. Bressan, J. A. Martínez, and A. Martí, “Noncoding RNAs, cytokines, and inflammation-related diseases,” The FASEB Journal, vol. 29, no. 9, pp. 3595–3611, 2015.

[63] U. Ashraf, B. Zhu, J. Ye et al., “MicroRNA-19b-3p modulates Japanese encephalitis virus-mediated inflammation via targeting RNF11,” Journal of Virology, vol. 90, no. 9, pp. 4780–4795, 2016.

[64] Y. Wu, X. Li, J. Jia et al., “Transmembrane E3 ligase RNF183 mediates ER stress-induced apoptosis by degrading Bcl-xl,” Proceedings of the National Academy of Sciences, vol. 115, no. 12, pp. E2762–E2771, 2018.

[65] Y. Chen, X. Y. Cao, Y. N. Li et al., “Reversal of cisplatin resistance by microRNA-139-5p-independent RNF2 downregulation and MAPK inhibition in ovarian cancer,” American Journal of Physiology-Cell Physiology, vol. 315, no. 2, pp. C225–C235, 2018.

[66] Y. Zhang, R. Sui, Y. Chen, H. Liang, J. Shi, and H. Piao, “Downregulation of miR-485-3p promotes glioblastoma cell proliferation and migration via targeting RNF135,” Experimental and Therapeutic Medicine, vol. 18, no. 1, pp. 475–482, 2019.

[67] B. Zhu, J. Ye, Y. Nie et al., “MicroRNA-15b modulates Japanese encephalitis virus-mediated inflammation via targeting RNF125,” Journal of Virology, vol. 95, no. 5, pp. 2251–2262, 2015.

[68] S. Qiu, Y. Feng, G. LeSage et al., “Chronic morphine-induced microRNA-124 promotes microglial immunosuppression by modulating P65 and TRAF6,” The Journal of Immunology, vol. 194, no. 3, pp. 1021–1030, 2015.

[69] K. D. Taganov, M. P. Boldin, K. J. Chang, and D. Baltimore, “NF-kappaB-dependent induction of microRNA miR-146, an inhibitor targeted to signaling proteins of innate immune responses,” Proceedings of the National Academy of Sciences of the United States of America, vol. 103, no. 33, pp. 12481–12486, 2006.

[70] H. Li, T. Li, J. Fan et al., “miR-216a rescues dexamethasone suppression of osteogenesis, promotes osteoblast differentiation and enhances bone formation, by regulating c-Cbl-mediated PI3K/AKT pathway,” Cell Death and Differentiation, vol. 22, no. 12, pp. 1935–1945, 2015.

[71] Z. Cui, Z. Luo, Z. Lin, L. Shi, Y. Hong, and C. Yan, “Long non-coding RNA TTN-AS1 facilitates tumorigenesis of papillary thyroid cancer through modulating the miR-153-3p/ZNRF2 axis,” The Journal of Gene Medicine, vol. 21, no. 5, article e0083, 2019.

[72] J. Beermann, M. T. Piccoli, J. Vierecck, and T. Thum, “Non-coding RNAs in development and disease: background, mechanisms, and therapeutic approaches,” Physiological Reviews, vol. 96, no. 4, pp. 1297–1325, 2016.

[73] S. Qu, X. Yang, X. Li et al., “Circular RNA: a new star of non-coding RNAs,” Cancer Letters, vol. 365, no. 2, pp. 141–148, 2015.

[74] W. T. C. Uniken Venema, M. D. Voskuil, G. Dijkstra, R. K. Weersma, and E. A. M. Festen, “The genetic background of inflammatory bowel disease: from correlation to causality,” The Journal of Pathology, vol. 241, no. 2, pp. 146–158, 2017.

[75] B. Khor, A. Gardet, and R. J. Xavier, “Genetics and pathogenesis of inflammatory bowel disease,” Nature, vol. 474, no. 7351, pp. 307–317, 2011.

[76] I. Van den Berghe, E. Zimprich, A. H. Dickson et al., “Expression of the miR-16-1-5p cluster is lost in colon cancer and confers a poor prognosis,” Nature Genetics, vol. 40, no. 10, pp. 1189–1194, 2008.

[77] S. Qu, X. Yang, X. Li et al., “Circular RNA: a new star of non-coding RNAs,” Cancer Letters, vol. 365, no. 2, pp. 141–148, 2015.

[78] W. T. C. Uniken Venema, M. D. Voskuil, G. Dijkstra, R. K. Weersma, and E. A. M. Festen, “The genetic background of inflammatory bowel disease: from correlation to causality,” The Journal of Pathology, vol. 241, no. 2, pp. 146–158, 2017.

[79] B. Khor, A. Gardet, and R. J. Xavier, “Genetics and pathogenesis of inflammatory bowel disease,” Nature, vol. 474, no. 7351, pp. 307–317, 2011.

[80] I. Van den Berghe, E. Zimprich, A. H. Dickson et al., “Expression of the miR-16-1-5p cluster is lost in colon cancer and confers a poor prognosis,” Nature Genetics, vol. 40, no. 10, pp. 1189–1194, 2008.

[81] S. Qu, X. Yang, X. Li et al., “Circular RNA: a new star of non-coding RNAs,” Cancer Letters, vol. 365, no. 2, pp. 141–148, 2015.

[82] W. T. C. Uniken Venema, M. D. Voskuil, G. Dijkstra, R. K. Weersma, and E. A. M. Festen, “The genetic background of inflammatory bowel disease: from correlation to causality,” The Journal of Pathology, vol. 241, no. 2, pp. 146–158, 2017.

[83] B. Khor, A. Gardet, and R. J. Xavier, “Genetics and pathogenesis of inflammatory bowel disease,” Nature, vol. 474, no. 7351, pp. 307–317, 2011.

[84] S. Qu, X. Yang, X. Li et al., “Circular RNA: a new star of non-coding RNAs,” Cancer Letters, vol. 365, no. 2, pp. 141–148, 2015.

[85] W. T. C. Uniken Venema, M. D. Voskuil, G. Dijkstra, R. K. Weersma, and E. A. M. Festen, “The genetic background of inflammatory bowel disease: from correlation to causality,” The Journal of Pathology, vol. 241, no. 2, pp. 146–158, 2017.

[86] B. Khor, A. Gardet, and R. J. Xavier, “Genetics and pathogenesis of inflammatory bowel disease,” Nature, vol. 474, no. 7351, pp. 307–317, 2011.

[87] S. Qu, X. Yang, X. Li et al., “Circular RNA: a new star of non-coding RNAs,” Cancer Letters, vol. 365, no. 2, pp. 141–148, 2015.

[88] W. T. C. Uniken Venema, M. D. Voskuil, G. Dijkstra, R. K. Weersma, and E. A. M. Festen, “The genetic background of inflammatory bowel disease: from correlation to causality,” The Journal of Pathology, vol. 241, no. 2, pp. 146–158, 2017.

[89] S. Qu, X. Yang, X. Li et al., “Circular RNA: a new star of non-coding RNAs,” Cancer Letters, vol. 365, no. 2, pp. 141–148, 2015.
[98] C. Ma, W. Lin, Z. Liu et al., “NDR1 protein kinase promotes IL-17- and TNF-α-mediated inflammation by competitively binding TRAF3,” EMBO Reports, vol. 18, no. 4, pp. 586–602, 2017.