Molecular Therapy
Nucleic Acids
Review

Rise of TRIM8: A Molecule of Duality

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The human tripartite motif containing protein 8 (TRIM8), a member of TRIM family proteins, is known to play a dual role as both tumor suppressor and oncogene, and to function at the crosstalk of cancer and innate immunity. In this review, in addition to accumulating recent corroborations that endorse this dual character of TRIM8, we appraise the game-changing capacity of TRIM8 under stress conditions against the backdrop of cell proliferation, apoptosis, and cancer, and also highlight the duality of TRIM8 in multiple contexts like cellular localization, stress-induced conditions, and E3 ubiquitin ligase activity. Finally, we discuss the emerging role of TRIM8 during bipolar spindle formation and mitotic progression, and its growing sphere of influence across multiple human cancers and pathologies, and suggest TRIM8-linked axes that can be modulated further for anti-cancer therapeutics development.

TRIM8: The Background

The human tripartite motif containing protein 8 (TRIM8) or RING finger protein 27 (RNF27), originally reported as glioblastoma expressed RING-finger protein (GERP),[1] is encoded by the *TRIM8* gene (Ensembl: ENSG00000171206) and positioned on the chromosome 10q24.32, a region that is known to show frequent deletion or loss of heterozygosity in glioblastomas.[1] Since its discovery studies on TRIM8 have impacted multiple areas of cell and disease biology (Figure 1). In humans, *TRIM8* is known to have a total of 15 splice-variant transcripts, and interestingly, three of them are uncharacterized long non-coding RNAs (lncRNAs) (Ensembl). TRIM8 is ubiquitously expressed in 27 human tissues considered in the Human Protein Atlas (HPA) RNA-Seq Project and produces one major transcript (Ensembl: ENST00000643721.1) of 7,290-bp length that codes for the 551-amino acid (aa) TRIM8 protein with a molecular mass of 61.489 kDa (UniProtKB: Q9BZR9 TRIM8 HUMAN).

In the context of ubiquitin system, it is widely known that the conjugation reaction of ubiquitin to a substrate is catalyzed by E1 ubiquitin-activating enzyme, E2 ubiquitin-conjugating enzymes, and E3 ubiquitin ligases.[2] The E3 ubiquitin ligases can be divided between two major classes: the homologous to E6-AP COOH terminus (HECT) E3 ubiquitin ligase family and the RING-finger-containing E3 ubiquitin ligase family. Although, TRIMs are considered among this RING-finger-containing E3 ubiquitin ligase family, not all TRIM E3 ubiquitin ligases have RING domains. For instance, in humans so far, the number of RING-less TRIM proteins stands at 9 out of more than 70 annotated TRIM proteins.[3] TRIM proteins play an important role in carcinogenesis and are highly associated with DNA repair, metastasis, tumor-suppressive regulation, and oncogenic regulation.[3] Further, some of the TRIM family proteins serve as critical regulators of autophagy and innate immunity and control important cellular processes, like intracellular signaling and transcription.[3–5]

TRIM8 belongs to this TRIM family of proteins (also called as RBCC proteins), which are known to maintain a common structural feature of having a tripartite motif, distinguished by the presence of a RING domain (R), one or two B-boxes (B), and a coiled-coil domain (CC).[6] The human TRIM8, an E3 ubiquitin ligase protein, contains an N-terminal C3HC4-type RING-finger domain, followed by two B-boxes (Bbox1 and Bbox2), a coiled-coil domain,[6] and a proline-rich no-domain C-terminal region with a monopartite nuclear localization signal (NLS)[7] (Figure 2A). TRIM8 comes structurally under class V of TRIM-type proteins, along with TRIM19, TRIM31, TRIM40, TRIM56, TRIM73, TRIM74, RNF207, TRIM52, and TRIM61, which are also observed to have no known domains in their C-terminal region so far.[5,7] The RING domain of TRIM8 is crucial for its activity to regulate stabilization and activation TP53, degradation of MDM2,[8] and destabilization of ΔNp63α,[9] and thus plays an important role in the context of cell proliferation, whereas the conserved B-box and coiled-coil domain region is known to mediate the interaction with SOCS1,[10] a tumor suppressor gene.[11] The coiled-coil domain of TRIM8 is required for homodimerization,[6] and deletion of the C-terminal domain of TRIM8 can result in protein mislocalization.[6] TRIM8 can also interact with other TRIM proteins in humans, like TRIM2, TRIM8, TRIM9, TRIM15, TRIM21, TRIM24, TRIM25, TRIM27, TRIM39, TRIM43, TRIM44, and TRIM47.[3]

Recent studies on TRIM8 strongly advocate for the critical role of TRIM8 as a regulator of cell proliferation[9,12] and as an important player in cancer,[13–16] immunity,[17] and inflammation.[18] Here, the duality of TRIM8 has been reviewed in multiple contexts, like its function and cellular localization, and the emerging role of TRIM8 during the course of cell cycle and bipolar mitotic spindle formation has been discussed. Further, TRIM8's growing sphere of influence has been assessed in the context of multiple human pathologies, and plausible

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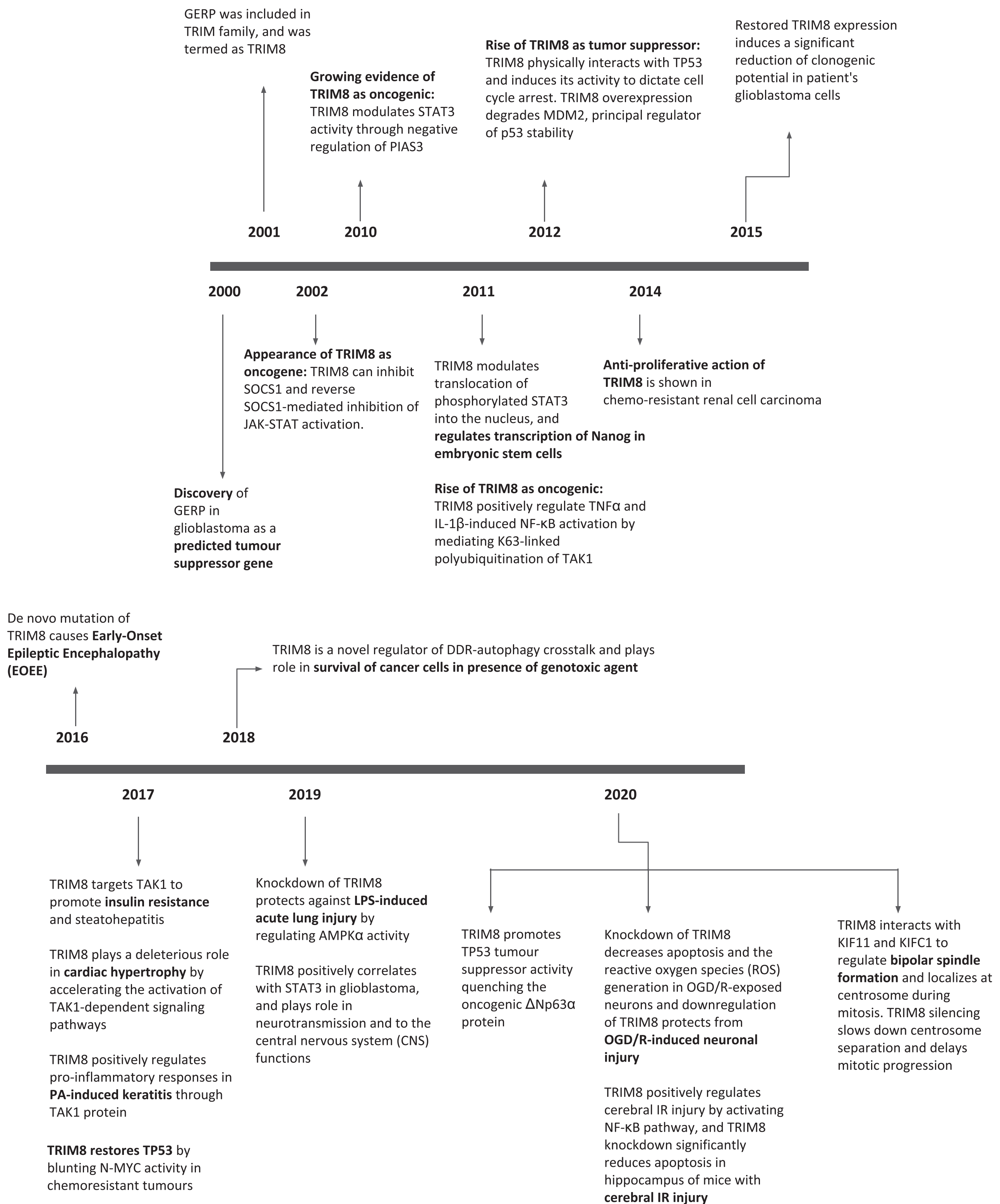

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approaches toward the development of anti-cancer therapeutics have been suggested.

TRIM8 Is a Molecule of Duality (MoD)

TRIM8 Can Act as Both Anti-proliferative and Pro-oncogenic

We showed that TRIM8 can act as a tumor suppressor by inducing TP53-dependent cell cycle arrest. TRIM8, a direct target gene of TP53, induces TP53 expression and tumor suppressor activity through a positive feedback loop-forming mechanism with TP53 during UV-instigated stress conditions by augmenting CDKN1A (p21) and GADD45 expression.[8] TRIM8 is highly downregulated in clear cell renal cell carcinoma (ccRCC), and the recovery of TRIM8 expression can lead to the enhancement of efficacy of chemotherapeutic drugs by re-activating the TP53 pathway, suggesting that TRIM8 can be used as an enhancer of chemotherapy efficacy in a TP53 wild-type background.[19] Further, TRIM8 can restore the TP53 activity by blunting N-MYC activity in chemo-resistant tumors, like ccRCC and colorectal cancer (CRC), upon inhibition of miR-17-5p (also known as MIR17) and/or miR-106-5p. The inhibition of miR-17-5p and/or miR-106-5p leads to the recovery of TRIM8-mediated TP53 tumor suppressor activity and inhibits N-MYC-dependent cell proliferation through miR-34a upregulation.[15] Overall, these studies established an anti-proliferative image of TRIM8 during the course of cell proliferation and cancer, and, following the same trend, recent studies have also provided further evidence toward the anti-proliferative activity of TRIM8. It has recently been shown that TRIM8 can blunt the pro-proliferative action of oncogenic ΔNp63α in a TP53 wild-type background.[9] Altogether, based on the current knowledge, it can be stated that TRIM8 has the capacity to exercise its anticancer power in three ways: by inducing the TP53 tumor suppressor activity through a positive feedback loop formation, restoring TP53 functions by blunting N-MYC activity in chemo-resistant tumors, and quenching the ΔNp63α oncogenic activity by forming a negative feedback loop. However, it should not go unnoticed that, in all three cases, TRIM8's anti-proliferative property is subject to the TP53 functional or wild-type background, suggesting a strong demand for research on TRIM8 and its anti-cancer capacity in a TP53 mutant background, as most of the TP53-associated axes (e.g., TP53-MDM2 axes) based on E3 ubiquitin ligase-targeting drugs fail in this caveat.[20] Indeed, it would be of high importance to look at the capacity of TRIM8 to restore the native conformation of TP53 mutants and reactivate the tumor-suppressor function.

In contrast to the anti-proliferative activity, TRIM8 can also work as an oncogenic protein.[7] TRIM8 serves as a critical regulator of tumor necrosis factor alpha (TNF-α)- and interleukin (IL)-1β-induced nuclear factor κB (NF-κB) activation by mediating K63-linked polyubiquitination of TAK1, and overexpression of TRIM8 activates NF-κB and potentiates TNF-α- and IL-1β-induced activation of NF-κB, whereas knock down of TRIM8 brings opposite effects. Silencing of TRIM8 potentially inhibits *TNFα* gene expression in U937 cells (histiocytic lymphoma), indicating the oncogenic capacity of TRIM8.[21] Further, TRIM8 positively regulates the TNF-induced NF-κB pathway at the p65 level by inducing the translocation of PIAS3 (protein inhibitor of activated STAT-3) from the nucleus to the cytoplasm.[22] TRIM8 also regulates the clonogenic and migration ability of the cells through the NF-κB pathway, and knock down of TRIM8 in the breast cancer MCF7 cell line significantly decreases the cell proliferation and clonogenic potential of cells.[22] Once activated, NF-κB promotes cell proliferation and protects cells from entering apoptosis. Thus, by positively regulating the TNF-induced NF-κB pathway, TRIM8 plays the role of an important oncogene that drives cell proliferation. Overall, from these two highly contrasting features, it can be stated that TRIM8 can function as both a tumor suppressor and an oncogenic molecule (Figure 2B).

Duality of TRIM8 under Stress Conditions

The duality of TRIM8 comes out under different stress conditions, like UV-instigated stress or genotoxic stress. Under exposure to UV, TP53 induces the expression of TRIM8, which, in turn, stabilizes TP53, leading to cell cycle arrest and reduction in cell proliferation through upregulation of *CDKN1A* (*p21*) and *GADD45*. Notably, TRIM8 silencing prevents TP53 activation after UV radiation. Further, the overexpression of TRIM8 reduces the half-life of MDM2, the key negative regulator of TP53, without altering MDM2 mRNA expression, which in turn results in an increased TP53 protein expression. Finally, it has also been proven that the TRIM8-RING domain is essential to regulate TP53 and MDM2 stability and activity. Overall, it suggests that, under UV-instigated stress conditions, TRIM8 plays an important role as a tumor suppressor to dictate cell cycle arrest.[8] In contradiction to this, a recent study has reported that TRIM8 can provide a survival advantage to cancer cells by enhancing autophagy flux through lysosomal biogenesis during genotoxic stress conditions. The study has shown TRIM8-regulated autophagy degrades the cleaved Caspase-3 subunit to inhibit genotoxic stress-induced cell death. TRIM8 knockdown reduces the expression of X-linked inhibitor of apoptosis protein (XIAP), whereas the enhanced expression of TRIM8 stabilizes XIAP during genotoxic stress conditions. XIAP also strongly activates NF-κB via BIR (baculovirus inhibitor of apoptosis protein repeat) domain-mediated dimerization and binding to TGF-β-activated kinase 1 (MAP3K7) binding protein 1 (Table 1). This XIAP-mediated NF-κB activation also induces expression of genes involved in autophagy, like Beclin-1. Interestingly, during genotoxic stress, TRIM8-mediated XIAP stabilization can also initiate inactivation of Caspase-3, one of the primary executioners of apoptotic cascade. Therefore, TRIM8-mediated XIAP stabilization has the capacity to bring two important oncogenic outcomes during the course of tumorigenesis. First, TRIM8-mediated XIAP stabilization can activate NF-κB, leading to expression of genes

Figure 1. TRIM8 through the Lens of History

Timeline showing major events in the scientific history of TRIM8—from discovery until recently, when it developed its growing sphere of influence across different areas of cell and disease biology; from proliferation to bipolar spindle formation, to multiple human pathologies.

A TRIM8 protein structure representation

B Key Figure: TRIM8 is a Molecule of Duality

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Table 1. A Summary of TRIM8-Linked Injuries and Apoptosis

Type of Injury	Neuronal Injury	Cerebral Injury	Hepatic Injury
Exposure to the pathogenic stimulus	oxygen-glucose deprivation/ re-oxygenation (OGD/R)	ischemia/reperfusion (I/R) injury	I/R injury
Changes in TRIM8 expression due to the exposure	upregulation	upregulation	upregulation
Impact of TRIM8 knockdown on apoptosis during the exposure	decreases apoptosis	decreases apoptosis	decreases apoptosis
Key molecules/pathways involved in the mode of action of TRIM8	Nrf2/ARE antioxidant signaling via AMPK	IκB kinase alpha (IKKα), inhibitory κB α (IκBα) and nuclear factor kappa B (NF-κB)	transforming growth factor β-activated kinase 1 (TAK1)-p38/JNK signaling pathway

involved in autophagy and cell proliferation. Second, TRIM8 mediated stabilized XIAP prevents activation of Caspase-3, leading to the suppression of apoptosis. Through this novel mechanism, TRIM8 prevents cell death during genotoxic stress and radiation therapy,[14] and this suggests TRIM8's highly potential oncogenic caliber can provide survival assistance to cancer cells.

TRIM8 Can Act in Both an E3 Ubiquitin Ligase-Dependent and -Independent Manner

Historically, TRIM8 is considered to be among E3 ubiquitin ligases due to the presence of RING domain. Although the mechanism of activation of its E3 ubiquitin ligase activity is not known yet, TRIM8 has been shown to function as an E3 ubiquitin ligase in several important biological pathways. It is demonstrated that TRIM8 mediates K63-linked polyubiquitination of TGF-β-activated kinase 1 (TAK1), triggered by TNF-α and IL-1β, and, through this, TRIM8 serves as a critical regulator of TNF-α- and IL-1β-induced NF-κB activation.[21] During *Pseudomonas aeruginosa* (PA)-induced keratitis infection, TRIM8 promotes K63-linked polyubiquitination of TAK1, leading to its activation and enhanced inflammatory responses.[23] Ye et al.[24] reported that TRIM8 can interact with Toll/IL-1 receptor domain-containing adaptor-inducing IFN-β (TRIF) and mediates its K6- and K33-linked polyubiquitination, which leads to the disruption of the TRIF-TANK-binding kinase-1 association. In general, it is known that K63-linked ubiquitination is involved in regulating proteasome-independent functions, including cellular processes, like endocytosis and inflammatory immune responses, innate immunity, protein trafficking, and NF-κB signaling, whereas K6-linked polyubiquitination is known to be associated with DNA damage response and Parkin-mediated mitophagy, and K33-linked polyubiquitination is associated with TCR signaling, post Golgi-trafficking, and AMPK-related kinase signaling.[25] Currently, it is experimentally well established that TRIM8 can perform K63-, K6-, and K33-linked polyubiquitination. TRIM8 also plays an important role in proteasomal degradation of SOCS1,[10] although it has not been proven yet whether it is through TRIM8-mediated K48-linked ubiquitination or in association with other protein complexes. Nevertheless, TRIM8's E3 ubiquitin ligase activity and its involvement in cancer and immunity lies much beyond doubt. But, against this running flow, recent studies have reported that TRIM8 can not only act in an E3 ubiquitin ligase-independent manner, but it can also protect phosphorylated IRF7 (pIRF7) from proteasomal degradation through an E3 ubiquitin ligase-independent path by preventing its recognition by the peptidyl-prolyl isomerase Pin1.[17]

Duality in Localization: Nucleus and Cytoplasm

TRIM8 can function at two subcellular sites—nucleus and cytoplasm—to regulate NF-κB, one of the central signaling pathways that plays a critical role in carcinogenesis and inflammatory diseases. PIAS3 is known to negatively regulate the NF-κB pathway via its interaction with p65 in the nucleus. Expression of TRIM8 enhances NF-κB activity even in the presence of PIAS3, suggesting TRIM8 can inhibit PIAS3-mediated negative regulation of NF-κB.[22] But TRIM8's ability to positively regulate NF-κB activity is not limited within the nucleus. Li et al.[21] observed earlier that TRIM8 positively regulates the NF-κB pathway by K63-linked polyubiquitination of cytoplasmic protein TAK1. Following this, Tomar et. al.[22] showed that TRIM8 can also regulate NF-κB activity in the cytoplasm under the influence of TNF-α. The study revealed that TNF-α induces the translocation of TRIM8 from the

Figure 2. TRIM8 Protein Structure and Function

(A) Structural representation of TRIM8. The illustration shows TRIM8 as an RBCC protein, made up of an N-terminal C3HC4-type RING-finger domain, followed by two B-boxes (Bbox1 and Bbox2), a coiled-coil domain, and a proline-rich no-domain C-terminal region with a monopartite nuclear localization signal (NLS). Like the class V TRIM family proteins, TRIM8 also does not have any known motifs/domains (e.g., B30.2/SPRY domain or RFP-like domain, COS domain, etc.) in its C-terminal region. (B) TRIM8 is a molecule of duality (MoD). TRIM8 can function in both a pro-proliferative and anti-oncogenic manner. TRIM8 primarily exercises its anticancer power by inducing TP53, negatively regulating N-MYC and quenching ΔNp63α while it functions as oncogenic due to its ability to induce NF-κB, and facilitate STAT3-mediated expressions of cancer related genes, leading to cell proliferation. In cytoplasm, TRIM8 inhibits the activity of PIAS3, which helps the translocation of STAT3 in the nucleus, where it exerts its oncogenic role. Interestingly, during UV-instigated stress, overexpression of TRIM8 inhibits MDM2, the master negative regulator of TP53, leading to transcriptional activation of cell-cycle arrest genes. In contrast, during genotoxic stress, TRIM8 induces XIAP, leading to inhibition of apoptosis and survival assistance for cancer cells. TRIM8-induced XIAP expression also leads to the activation of NF-κB and positive regulation of cell proliferation.

nucleus to the cytoplasm, which positively regulates NF-κB. A time-course study for TRIM8 nucleo-cytoplasmic translocation upon TNF treatment showed that TRIM8 translocates to the cytoplasm within 15 min and re-translocates back to the nucleus after 12 h.[22] Another instance of TRIM8's nucleo-cytoplasmic translocation can be found in the regulation of STAT3 by PIAS3. In the cytoplasm, the ectopic expression of TRIM8 promotes proteasomal degradation of PIAS3, leading to the nuclear translocation of STAT3, whereas, in the nucleus, TRIM8 facilitates the recruitment of STAT3 to the STAT3-inducible element (SIE) regions of several brain- and cancer-related genes and binds to the SIE regions of the same genes.[13]

Duality of TRIM8 in Glioblastoma

The regulation of STAT3 by TRIM8 via PIAS3 is extensively important in the context of glioblastoma. Zhang et al.[26] reported that TRIM8 initiates STAT3 signaling to maintain stemness and self-renewal capacity in glioblastoma-like stem cells (GSCs) by suppressing the expression of PIAS3. Knockdown of TRIM8 reduces GSC self-renewal, whereas overexpression of TRIM8 leads to enhanced GSC self-renewal.[26] The study has further shown that STAT3 activation can also upregulate TRIM8 expression, even in the setting of hemizygous gene deletion in glioblastoma, and this bi-directional positive feedback mechanism facilitates stemness in GSCs. This suggests that TRIM8 acts as an oncogenic molecule by promoting the self-renewal capacity of GSCs. In contrast, we showed that the downregulation of TRIM8 in glioblastoma compared to its normal counterpart is indeed associated with a significant increase in the risk of disease progression in patients. Most importantly, TRIM8 overexpression and restored TRIM8 expression significantly reduce both cell proliferation and clonogenic potential in glioma cells, suggesting the anti-proliferative capacity of TRIM8 in glioma patients.[27] However, it is not at all clear how a molecule that helps the self-renewal capacity in GSCs can alter its mechanism to reduce the proliferation and clonogenic potential in glioma cells.

TRIM8: Growing Sphere of Influence

Emerging Role of TRIM8 in Bipolar Spindle Formation during Mitosis

The roles of different TRIM family proteins are known during the stages of mitosis and cell-cycle progression. TRIM28, TRIM19, TRIM69, TRIM22, TRIM37 are involved in prophase; TRIM32, TRIM19, and TRIM69 in prometaphase; TRIM36, TRIM17, and TRIM69 in metaphase; and TRIM17, TRIM21, TRIM47, and TRIM76 in cytokinesis.[28] We recently showed that TRIM8 interacts with KIF11 (also known as EG5) and KIFC1, two master regulators of mitotic spindle assembly and cytoskeleton reorganization, and localizes at the mitotic spindle during the stages of bi-polar spindle formation. Our study also suggested that TRIM8 is required for chromosomal stability, as we observed that silencing of TRIM8 resulted in a significant increase of cells with less than 46 chromosomes.[29] In particular, KIF11 inhibition and KIFC1 overexpression are known to be sufficient enough to induce a monopolar spindle phenotype in mitotic cells,[30–33] and thus the physical interactions of TRIM8 with KIF11 and KIFC1 strongly advocate for the plausible role of TRIM8 in coordinating cell polarity during mitosis. From our experimental data, some interesting clues are emerging. On one hand, TRIM8 localizes on centrosomes and colocalizes with Plk1 and directly interacts with CEP170-like centrosomal protein, and silencing of TRIM8 induces a delay of the mitosis progression with a cell accumulation in the G2/M phase.[29] Therefore, we suggest that TRIM8 can play an important function during the course of centrosome duplication. On the other hand, TRIM8 localizes at the mitotic spindle throughout all phases of mitosis and physically interacts with some of the most important mitotic assembly proteins like KIF11, KIFC1, KIF20B, and KIF2C. Altogether, it can be suggested that TRIM8 may play an important function in the bi-polar spindle formation from the very onset of centrosome duplication until the end of the division of one cell into two daughter cells, a signature process in eukaryotic life mediated by microtubules and kinesin family proteins.

Knockdown of TRIM8 Links Injuries and Apoptosis

Although earlier studies could not reveal much of the role of TRIM8 in regulating apoptosis during injuries, the emerging role of TRIM8 is highlighting its common characteristic feature in regulating apoptosis in different cell types when they are exposed to conditions like oxygen-glucose deprivation/re-oxygenation (OGD/R) or ischemia/reperfusion (I/R) injury. Recent studies on the role of TRIM8 in neural apoptosis identified that TRIM8 expression is upregulated in neurons when they are exposed to OGD/R. Knockdown of TRIM8 improves the cell viability and decreases the apoptosis and reactive oxygen species (ROS) generation in OGD/R-exposed neurons, whereas TRIM8 overexpression shows exactly the opposite effect. The elevated apoptotic neurons induced by OGD/R exposure gets remarkably decreased upon TRIM8 knockdown, suggesting that TRIM8 inhibition protects neurons from OGD/R-induced apoptosis and ROS production by reinforcing AMPK/Nrf2/ARE signaling.[34] TRIM8 is also significantly upregulated in the liver of mice exposed to hepatic I/R injury. The silencing of TRIM8 alleviates hepatic inflammation responses and inhibits apoptosis *in vitro* and *in vivo*.[35] TRIM8 deficiency plays a protective role in hepatic I/R injury by inhibiting the activation of TAK1-dependent signaling pathways. Further, it has also been recently identified that TRIM8 knockdown significantly reduced apoptosis in the hippocampus of mice with cerebral IR injury by reducing Caspase-3 cleavage. Suppression of TRIM8 reduces cerebral IR-induced inflammation and apoptosis, and TRIM8 positively regulates cerebral IR injury by activating the NF-κB pathway to enhance inflammation and apoptosis.[18] Overall, from these studies, it is crystal clear that, during exposure to pathogenic stimuli like OGD/R and I/R injury in different cell types, TRIM8 always gets upregulated, and knockdown of TRIM8 decreases apoptosis significantly. At the same time, it is also noteworthy that, although the impact of TRIM8 on apoptosis upon exposure to stimulus remains the same in different cell types, its mode of action varies across the cell types, indicating that

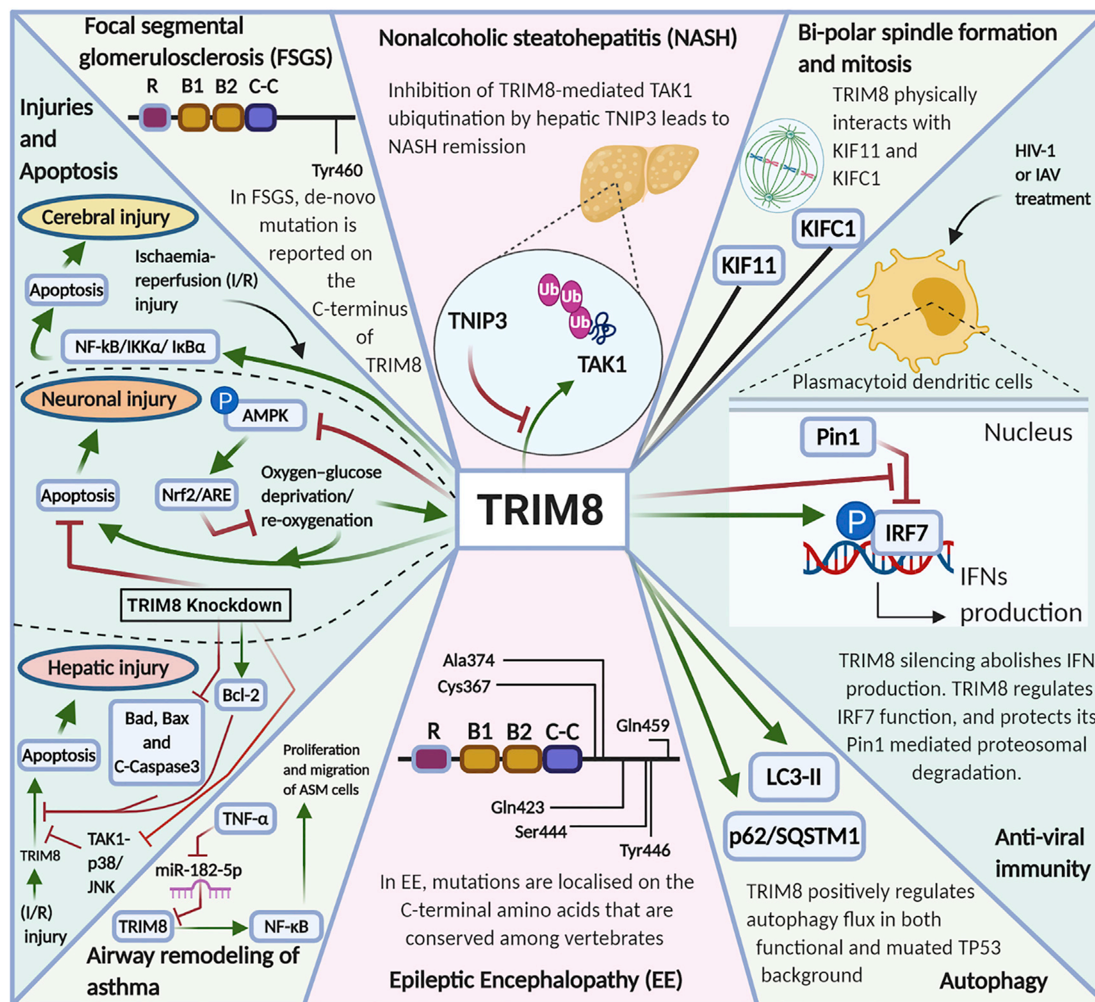

Figure 3. TRIM8: Growing Sphere of Influence

Alongside its emerging role in bipolar spindle formation, mitosis, and autophagy, TRIM8 is now known to be involved in multiple non-cancerous human pathologies, like nonalcoholic steatohepatitis (NASH), epileptic encephalopathy (EE), asthma, and focal segmental glomerulosclerosis (FSGS), among many others. TRIM8 is emerging as an important therapeutic target against injuries in different cell types as it promotes apoptosis, leading the positive regulation of injuries when exposed to pathogenic stimuli, like oxygen-glucose deprivation and reperfusion (OGD/R) or ischemia/reperfusion (I/R) injury. Most importantly, TRIM8 plays an important role in developing antiviral immunity against human immunodeficiency virus 1 (HIV1) and influenza A virus (IAV). Of note, many of the pathways or molecules like NF-κB, TAK1, or KIF11 that are emerging as partners of TRIM8 in multiple cellular mechanisms or pathologies are also known to be highly significant in the context of cancer.

TRIM8 can impact apoptosis via different pathways (Table 1; Figure 3).

Role beyond Cancer: TRIM8 in Multiple Human Pathologies

Besides playing a significant role in cancer, TRIM8 is also emerging as an important player in multiple cellular and pathological phenomena (Figure 3). TRIM8 has been identified as a novel gene responsible for early-onset epileptic encephalopathy (EOEE) and epileptic encephalopathy (EE). Pathogenicity of TRIM8 mutations on its C terminus has been established as the causative agent for EE, possibly associated with nephrotic syndrome.[36,37] *De novo* mutation on the C-terminal region of TRIM8 is also associated with focal segmental glomerulosclerosis (FSGS).[38] Liu et al.[39] reported that inhibition of TRIM8-mediated TAK1 polyubiquitination by hepatic TNIP3 can lead to the blocking of the progression of nonalcoholic steatohepatitis (NASH), a subtype of nonalcoholic fatty liver disease (NAFLD). Most importantly, TRIM8 has recently been identified as an important driver of IFN production in plasmacytoid dendritic cells (pDCs), playing a crucial role in developing antiviral immunity against influenza A virus (IAV) and human immunodeficiency virus 1 (HIV-1) by positively regulating IFN regulatory factor 7 (IRF7) via inhibition of Pin1.[17]

TRIM8, KIF11, and Glioblastoma - a link yet to be discovered

TRIM8 expression is known to be downregulated in glioblastoma compared to its normal counterpart (Figure 4. A), and we showed earlier that this downregulation is associated with cell proliferation and patient survival. A statistically significant downregulation was observed in WHO Grade IV glioblastoma compared to Grade II and Grade III in both tumour tissues and cell lines, and upon overexpression of TRIM8, proliferation and clonogenic potential of glioma cells reduced significantly. We had also shown this downregulation of TRIM8 is highly correlated with the loss of its copy number. From Grade I to Grade IV, with the increase in grades of glioblastoma, a significant escalation in the loss of copy number was observed [27]. Recently, we showed that in neural stem cells TRIM8 physically interacts with KIF11 [29], an important driver of invasion, proliferation and self-renewal in glioblastoma [41]. TCGA data set analysis also reveals that KIF11 is highly exprepressed in glioblastoma compared to its normal condition (Figure 4. B). But whether TRIM8 ubiquitinates KIF11 is still subject to the availability of experimental evidence. Nevertheless, there is a possibility that in glioblastoma E3 ubiquitin ligase dependent interaction or E3 ubiquitin ligase independent interaction of TRIM8 with KIF11 gets perturbed, and due to which such a high expression of KIF11 is observed.

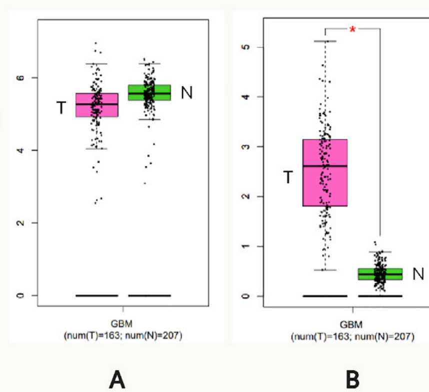

Figure 4. (A) TRIM8 is downregulated in glioblastoma. **(B)** KIF11 is downregulated in glioblastoma. Expression is calculated in log2(TPM + 1) scale and plotted on the y-axis. Pink and green represent tumour (T) and normal (N) samples respectively. *Data source: TCGA and GTEx. Credit: GEPIA.*

Figure 4. TRIM8, KIF11, and Glioblastoma: A Link Yet to Be Discovered
A proposed role of interaction between TRIM8 and KIF11 in the progression of glioblastoma.

TRIMming Cell Proliferation with TRIM8: A Game-Changer in Cancer?

Emerging concepts in "drugging the undruggable" have brought the large E3 ubiquitin ligase family of enzymes to the forefront of ubiquitin system-based drug development due to its anticipated possibility of having better specificity and lesser toxicity.[20] TRIM8, one of the most promising and emerging E3 ubiquitin ligases from the TRIM family, has the potential to be recognized in this very frame of modern E3 ubiquitin ligase-based drug discovery. TRIM8 is highly expressed in MCF7, and knockdown of TRIM8 decreases the clonogenic potential of MCF7. TRIM8 positively regulates NF-κB by inhibiting PIAS3 via inducing its nuclear-cytoplasmic translocation.[22] Modulating TRIM8-NF-κB-PIAS3 axes can be a therapeutic choice. TRIM8 is negatively regulated in chronic lymphocytic leukemia (CLL) by miR-17.[40] Inducing TRIM8 expression by silencing miR-17 can induce anti-proliferative gene expressions via TP53 axes in CLL. TRIM8 is downregulated in ccRCC, and this downregulation is associated with suppression of TP53. Restoration of TRIM8 expression in ccRCC renders the cells sensitive to chemotherapeutic treatments by reactivating the TP53 axes,[19] suggesting TRIM8 can be used as an enhancer to ablate the chemoresistance property of the aggressive ccRCC cells in TP53 wild-type background. Another study in human anaplastic thyroid cancer (ATC) tissues and cell lines reported that TRIM8 is significantly downregulated in ATC and miR-182 induces tumor growth by repressing TRIM8 expression. Overexpression of miR-182 delivers chemoresistance capacity to tumor cells and reduces TRIM8 expression.[16] Together, these data suggest the anti-proliferative property of TRIM8 can be used in the treatment of chemoresistant human thyroid papillary cancer.

Earlier, we showed that TRIM8 downregulation in glioblastoma induces cell proliferation and is associated with patients' survival. In glioblastoma, restored TRIM8 expression can function as a tumor suppressor and reduce the clonogenic potential of cancer cells significantly,[27] although it is not clear so far whether restoration of TRIM8 inhibits cell proliferation in glioblastoma via TP53 axes. Nevertheless, it does not affect the demand on the search for a possible therapeutic path to induce the restoration of TRIM8 in glioblastoma and inhibit cell proliferation. Along with exploring TRIM8-TP53 axes in glioblastoma, we also suggest exploring the TRIM8-KIF11 axes, based on our

Box 1 Open Questions

1. What sort of structural, molecular, or epigenetic features act as the deciding factor to drive TRIM8 as a MoD?
2. How does the E3 ubiquitin ligase function of TRIM8 get activated?
3. Does TRIM8 ubiquitinate KIFC1 and KIF11, two master regulators of mitotic spindle assembly formation? Which signaling pathway is regulated by the interaction between TRIM8 and KIFC1/KIF11 during bi-polar spindle formation?
4. What are the direct target genes of TRIM8 during the bi-polar mitotic spindle formation?
5. Does silencing or overexpression of TRIM8 have any impact on the expression of KIF11, KIFC1, and other mitotic assembly-related TRIM8 interacting proteins, such as Cep170, Bicd2, Kif2c, etc.?
6. What is the effect of TRIM8 perturbation on cell shape, spindle assembly (size and shape), spindle robustness, and interactions between organelles?
7. Does TRIM8 play any essential role in centrosome duplication and deciding the number of poles during cell division?
8. Under stress conditions, TP53 induces TRIM8 expression, which in turn stabilizes the TP53, leading to cell-cycle arrest and reduction in cell proliferation; whereas TRIM8 overexpression leads to the degradation of MDM2. Why is the rise of TP53-mediated TRIM8 expression not able to reach the level to degrade MDM2 protein under the stress conditions that are possible during TRIM8 overexpression?

hypothesis in glioblastoma. In our recent study, we have shown, for the first time, that TRIM8 physically interacts with KIF11,[29] one of the master regulators of mitotic spindle assembly and cytoskeleton reorganization[30–33] that has also been established as an important driver of glioblastoma.[41] There has been no study so far that aims to connect KIF11 and TRIM8 in glioblastoma. Therefore, we strongly believe further research is needed to shed light on the connecting point of TRIM8, KIF11, and glioblastoma (Figure 4). It is noteworthy that, in some cancers, like breast cancer, TRIM8 is highly expressed and can function as a pro-proliferative or an oncogenic molecule, whereas, in other cancers, like ccRCC, it is downregulated and its capacity to exercise anticancer power is significantly suppressed. But TRIM8's game-changing capacity in cancer is unquestionable, and it can be utilized in either way–by suppressing or overexpressing—depending on its pattern of function in a specific cancer. Further we propose TRIM8 and other important TRIM proteins can be explored together in order to modulate TP53 axes for arresting cell proliferation.

One of the most important hallmarks of cancer is "limitless replicative potential" or proliferated cell division.[42] TP53 is known to have great potential to regulate cell proliferation, and the TP53 axes have been at the center of anti-cancer therapeutics search for several years. We showed earlier that TRIM8 plays a very important role in the regulation of TP53,[8] and along with TRIM8, some other TRIM proteins are also known to regulate TP53. For instance, TRIML2, TRIM3, TRIM8, TRIM13, and TRIM19 function as the positive regulators of TP53, whereas some other TRIMs, like TRIM24 and TRIM32, work as the negative regulators of TP53.[43] These TRIM proteins are also known to have homotypic and heterotypic interactions among them. For instance, TRIM8 can interact with TRIM2, TRIM8, TRIM9, TRIM15, TRIM21, TRIM24, TRIM25, TRIM27, TRIM39, TRIM43, TRIM44, and TRIM47.[3] Among these TRIM8-associated TRIMs, many are known to be oncogenic players in different cancers. Particularly, TRIM24 and TRIM25 are known to act as TP53-negative regulators and are overexpressed in castration-resistant prostate cancer (CRPC) and ovarian cancer, respectively.[3,43] We believe this kind of heterotypic interaction between TRIM8 and other TRIM proteins can be an applicable choice of future study in order to elucidate its impact on cell proliferation by modulating the TP53 axes and to develop anti-cancer therapeutics.

Conclusions

The experimental findings discussed in this review have shown concrete evidence for TRIM8 that can act as a "molecule of duality" (MoD), a new term we propose to objectify a protein molecule that has the capacity to play two opposite roles, and can showcase two highly contrasting features in one or more cellular or physiological conditions and/or pathophysiological context. Overall, it appears that TP53 lies at the center of TRIM8's tumor suppressor activity, whereas NF-κB lies at the center of TRIM8's oncogenic activity and its role in apoptosis. Although several outstanding questions need to be addressed (refer to Box 1), we feel it's highly crucial to understand how TRIM8-associated axes can be modulated further for anti-cancer therapeutics development, as pharmaceutical companies have entered the age of E3 ubiquitin ligase-targeted therapies and targeting E3 ligases is progressively becoming a considerable choice for therapeutic development against many fusion-driven cancers. Further studies on TRIM8 also have immense possibilities to open new vistas that can show the role of TRIM8 at the connecting point of apoptosis, cell proliferation and cell division, and the cross-roads of brain development and brain-associated disorders like epilepsy and glioblastoma. For instance, it would be interesting to investigate whether susceptibility to cancer increases in TRIM8-mutated EOEE or EE patients. Finally, we feel the open questions on TRIM8 hold significant importance in the forthcoming years of cell and cancer biology.

New Terminology

Molecule of duality (MoD): A protein molecule that has the capacity to play two opposite roles, and can showcase two highly contrasting features in one or more cellular or physiological conditions, and/or pathophysiological context.

CONFLICTS OF INTEREST

The authors declare no competing interests.

AUTHOR CONTRIBUTIONS

U.B. and G.M. conceived this review. U.B. reviewed the literature and wrote the manuscript along with G.M. U.B. designed all figures and illustrations. Both U.B. and G.M. contributed to the final version of the manuscript.

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