BBA - Reviews on Cancer 1871 (2019) 138–159

Contents lists available at ScienceDirect

BBA - Reviews on Cancer

journal homepage: www.elsevier.com/locate/bbacan

Review

The emerging role for Cullin 4 family of E3 ligases in tumorigenesis

Ji Cheng[a,b,1], Jianping Guo[b,1], Brian J. North[b], Kaixiong Tao[a], Pengbo Zhou[c,*], Wenyi Wei[b,*]

[a] Department of Gastrointestinal Surgery, Union Hospital, Tongji Medical College, Huazhong University of Science and Technology, Wuhan 430022, China
[b] Department of Pathology, Beth Israel Deaconess Medical Center, Harvard Medical School, Boston, MA 02215, USA
[c] Department of Pathology and Laboratory Medicine, Weill Cornell Medicine, 1300 York Ave., New York, NY 10065, USA

ARTICLE INFO

Keywords:
CRL4, Cullin 4
E3 ligases
PROTACs
Tumorigenesis
Targeted therapy

ABSTRACT

As a member of the Cullin-RING ligase family, Cullin-RING ligase 4 (CRL4) has drawn much attention due to its broad regulatory roles under physiological and pathological conditions, especially in neoplastic events. Based on evidence from knockout and transgenic mouse models, human clinical data, and biochemical interactions, we summarize the distinct roles of the CRL4 E3 ligase complexes in tumorigenesis, which appears to be tissue- and context-dependent. Notably, targeting CRL4 has recently emerged as a noval anti-cancer strategy, including thalidomide and its derivatives that bind to the substrate recognition receptor cereblon (CRBN), and anticancer sulfonamides that target DCAF15 to suppress the neoplastic proliferation of multiple myeloma and colorectal cancers, respectively. To this end, PROTACs have been developed as a group of engineered bi-functional chemical glues that induce the ubiquitination-mediated degradation of substrates via recruiting E3 ligases, such as CRL4 (CRBN) and CRL2 (pVHL). We summarize the recent major advances in the CRL4 research field towards understanding its involvement in tumorigenesis and further discuss its clinical implications. The anti-tumor effects using the PROTAC approach to target the degradation of undruggable targets are also highlighted.

1. Introduction

1.1. The ubiquitin-proteasome system (UPS)

The ubiquitin-proteasome system (UPS) is an evolutionarily conserved apparatus that serves as a major regulator of proteostasis in eukaryotic cells [1]. The UPS generally consists of ubiquitin, ubiquitination enzymes and the 26S proteasome, which synergistically form an enzymatic cascade to transfer ubiquitin in a substrate-specific manner to promote subsequent proteolysis and degradation of the target protein [2]. The machinery of the ubiquitin-proteasome cascade has been reviewed extensively [3,4]. Briefly, the ATP-dependent activation of ubiquitin by the E1 activating enzyme is indispensable for the initiation of the enzymatic cascade [5]. As a result, activated ubiquitin is then transferred to an E2 conjugating enzyme, which then assists in the recruitment of an E3 ligase into a complex with the ubiquitin moiety. Subsequently, E3 ligase determines the substrate specificity and facilitates the formation of a covalent isopeptide bond between the ubiquitin and lysine residues of target proteins, leading to substrate ubiquitination. Additionally, E3 ligases also mediate the attachment of the ubiquitin moiety to existing ubiquitin chains on a substrate protein, resulting in poly-ubiquitination and diverse consequences to the target protein, such as degradation [2,6], altered activity, or subcellular localization of substrates [7].

1.2. Physiological significance and pathological roles of the UPS

Consistent with a broad central role that the UPS plays in proteostatic control, nearly every aspect of cellular biology is regulated in some manner by the UPS pathway, especially the cell cycle, cell growth, immune homeostasis, and metabolic stability [6]. For instance, proteasomal degradation of cell cycle regulators, such as p21 and p27, is a critical to control cell cycle progression [6]. In addition, proteolytic activities provide necessary raw materials for intracellular recycling and rebuilding, such as generation of amino acids [6,8]. Meanwhile, via proteasomal cleavage of endogenous proteins, the UPS governs the production of MHC (major histocompatibility complex) Class I antigens, which are critical for the functions of the immune response [9,10]. Dysregulation of the UPS is involved in the pathogenesis of multiple disorders, especially neurodegeneration, neoplastic transformation and autoimmune diseases [11]. Pathologically, neurodegenerative disorders often feature the accumulation of misfolded proteins, such as tau aggregates and Aβ plaques in *Alzheimer's* disease [12,13]. Autoimmune diseases are often induced by the mis-recognition of endogenous

---

[*] Corresponding authors.
E-mail addresses: pez2001@med.cornell.edu (P. Zhou), wwei2@bidmc.harvard.edu (W. Wei).

[1] **Note:** These authors contribute equally to the works.

https://doi.org/10.1016/j.bbcan.2018.11.007
Received 3 November 2018; Received in revised form 28 November 2018; Accepted 29 November 2018

proteins as exogenous antigens [14], which was tightly controlled by the UPS. For instance, increased generation of MHC Class I antigen HLA (human leukocyte antigen)-B27 closely correlates to Ankylosing Spondylitis [15,16].

1.3. Cancer relevant roles of the UPS

Dysregulation of the UPS is associated with tumorigenic events [17,18]. E3 ligases are involved in the final enzymatic activity leading to ubiquitination and dictates the specificity of substrate selection. Hence, depending on the tumorigenic properties of specific substrates, E3 ligases play a context-dependent role by degrading tumor suppressors or oncoproteins [19,20]. It is noteworthy that E3 ligase may also cause altered activity or subcellular re-localization of target proteins. For example, MDM2 (mouse double minute 2 homolog), serving as an E3 ligase to destabilize tumor suppressor p53, has been regarded as one of the most frequently mutated oncoproteins in lung and breast carcinoma [21–23]. On the other hand, SPOP (speckle-type POZ protein) is frequently mutated adaptor protein in prostate cancer serving as a tumor suppressor by targeting TRIM24 (tripartite motif-containing 24) and the androgen receptor [24,25]. Due to their critically important role in substrate selection and ubiquitin transfer during the UPS cascade, E3 ligases are broadly acknowledged as a key target for anti-cancer therapeutics, thus the use of small molecules targeting UPS for cancer treatment has been developed for diverse malignancies, especially multiple myeloma and lymphoma [26–28], which will be discussed in the following sections.

1.4. DUBs: The antagonizing force in counter-balancing the UPS

Similar to other post-translational modifications, there are also mechanisms in place to reverse and therefore antagonize the role of ubiquitination and thus maintain protein stability, which is largely mediated by a cluster of proteins termed deubiquitinating enzymes (DUBs) [29,30]. Currently, nearly 100 DUBs have been identified in humans [31], which are divided into 7 subgroups by a specific functional domain, including ubiquitin C-terminal hydrolase (UCH) DUBs, ubiquitin specific protease (USP) DUBs, ovarian tumor (OTU) DUBs, Josephin DUBs, JAB1/MPN+/MOV34 (JAMM) DUBs [32] and another two non-canonical DUBs, MINDY DUBs [33] and ZUFSP DUBs [34,35]. Consistent with the roles for the UPS participating in critically important cellular biological processes, DUBs are likewise involved in nearly every aspect of cellular biology [17,36]. For instance, USP9X (ubiquitin specific peptidase 9) is found to be negatively correlated with carcinogenesis, which deubiquitinates and functionally stabilizes its substrate LATS1 (large tumor suppressor kinase 1), leading to the inactivation of the Hippo pathway [37]. USP2 (ubiquitin specific peptidase 2) triggers malignant progression in prostate cancer cells via deubiquitination and stabilization of fatty acid synthase (FAS), which positively correlates with aggressiveness and negatively predicts survival [38].

2. CRL4 E3 ligases in tumorigenesis

2.1. E3 ligases and the Cullin-RING family

There are hundreds of E3 ligases identified in mammalian ubiquitination cascades [39,40]. Based on structural and functional differences, E3 ligases are subdivided into two groups, namely the HECT (homologous to the E6AP carboxyl terminus) family and the RING (really interesting new gene) family [41]. Specifically, RING E3 ligases bind to ubiquitin-E2 heterodimer and protein substrates in a simultaneous manner, facilitating an efficient transfer of ubiquitin onto selected substrates. Whereas, ubiquitination mediated by HECT E3 ligases is separated into two steps, formation of a thioester bond with ubiquitin followed by substrate recognition and ubiquitin transfer [40,42]. In addition, the RING ligases have been further categorized into several subfamilies, including BARD1 (BRCA1-associated RING domain 1), c-Cbl (Casitas B-lineage lymphoma), anaphase-promoting complex (APC), heterodimer of BRCA1 (breast cancer 1) and Cullin-RING ligases (CRLs), each harboring a RING catalytic domain [40].

Characterized by being composed of multi-subunit complexes, the Cullin-RING ligases are the most extensively studied subfamily of RING ligases. Structurally, a Cullin-RING ligase complex is composed of four essential subunits, including a Cullin scaffold, RING-finger protein, adaptor protein, and a substrate recognition protein [43]. As a central coordinator of complex formation, the Cullin scaffold provides the platform for both the RING-finger protein and adaptor protein. The RING-finger protein serves as a docking site for both the ubiquitin-E2 complex and the substrate recognition module to facilitate the transfer of ubiquitin to the recruited substrate. The substrate recognition module that associates within the complex through its interaction with the adaptor protein and dictate substrate specificity [43,44]. To date, eight Cullin scaffolds have been characterized in mammalian cells (Cullin 1, Cullin 2, Cullin 3, Cullin 4A, Cullin 4B, Cullin 5, Cullin 7, and Cullin 9) as well as two RING-finger proteins (RBX1 and RBX2), four adaptor proteins (SKP1, ElonginB, ElonginC, and DDB1) and more than 400 substrate recognition receptor proteins [45]. These subunits are assembled into hundreds of unique Cullin-RING ligase complexes that are responsible for nearly 20% of ubiquitination events mediated by the UPS [46,47].

2.2. Introduction of Cullin 4 E3 ligases

The Cullin 4-RING ligases (CRL4s) contain two homogenous scaffolds, defined as Cullin 4A (CUL4A) and Cullin 4B (CUL4B), respectively. Commonly, both scaffolds bind to damaged DNA binding protein 1 (DDB1) and the catalytic subunit RING-finger protein RBX1 through their N- and C-terminus respectively to support the structure of entire complex [48]. However, despite of 82% identity with regard to their genomic sequences, each scaffold targets a unique set of substrates. Compared to CUL4A, CUL4B features a longer N-terminus, which contains an extra nuclear localization signal (NLS) that governs its targeting to the nucleus, in addition to interaction with DDB1. On the other hand, the majority of CUL4A resides in the cytoplasm to regulate substrate ubiquitination [49], although a small fraction is found in the

Table 1
Pathological involvements of mutated components of CRL4.

Gene	Component category	Alteration	Consequence
Cul4b	Scaffold	Mutation	X-linked mental retardation [77,78]; Cerebral malformations [80]
Dcaf8	Substrate recognition protein	Mutation	Axonal hereditary motor and sensory neuropathy (HMSN2) [81]
Dcaf14	Substrate recognition protein	Mutation	Developmental retardation, intellectual defects, obesity and dysmorphic features [82]
Dcaf17	Substrate recognition protein	Mutation	Woodhouse Sakati syndrome [83]
Ddb2	Substrate recognition protein	Mutation	Xeroderma pigmentosum [84]
Gnb3	Substrate recognition protein	Mutation	Hypertension [85]
Wdr62	Substrate recognition protein	Mutation	Cerebral malformations [80]

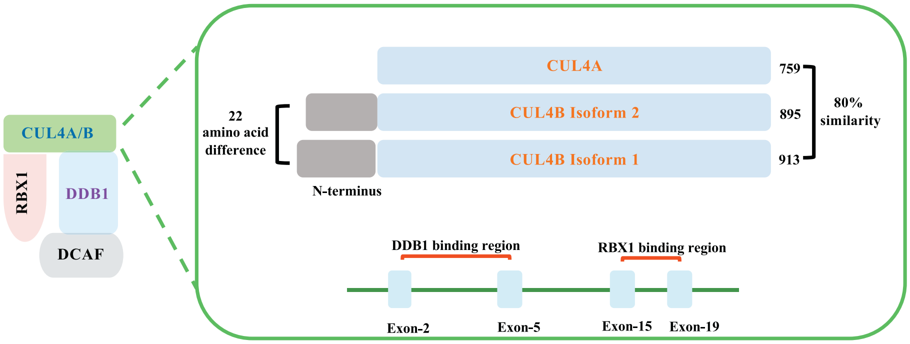


Fig. 1. Overview of neoplastic roles of CRL4 components.

nucleus to target nuclear proteins [50] (Table 1). CUL4 scaffolds are also neddylated by the ubiquitin-like protein NEDD8 (neural precursor cell expressed, developmentally down-regulated 8), which is essential to stimulate the activity of CRL4 complexes [51]. Moreover, RBX1 acts as a regulatory subunit of the CRL4 complex through recruitment of ubiquitin E2 complex and transferring the ubiquitin moiety. The adaptor DDB1 serves as the bridging factor between the Cullin scaffold and substrate recognition subunit, DDB1-CUL4-associated factor (DCAF) (Fig. 1) [51,52].

2.3. Physiological roles of Cullin 4 E3 ligases -evidence from knockout mouse models

According to the phenotypic studies derived from knockout mouse models on CRL4 components, CRL4s participate in embryonic development as well as maturation and homeostasis of multiple tissues and organs. Unlike the essential roles of most Cullins, germline knockouts of *Cul4a or Cul4b* are both viable and show no overt growth abnormalities [48,53–55], which is likely due to redundancy between CUL4A and CUL4B. Abrogation of both *CUL4A* and *CUL4B* in mouse embryonic fibroblasts and tumor cell lines led to growth arrest and loss of viability [55]. Moreover, germline knockout of the *Ddb1* adaptor is embryonic lethal [56,57], highlighting the essential role of the CRL4 ubiquitin ligase in maintaining growth and survival of mammals. Although a previous study suggested an embryonic requirement of *Cul4a* [58], other studies have shown that systemic ablation of *Cul4a* does not lead to embryonic lethality [55,59], and the embryonic phenotype seen in the earlier study may be attributed to an unintended deletion of the adjacent *Pcid2* gene on the complementary strand of DNA around the *Cul4a* locus [55].

The hematopoietic lineage is another system where CRL4 activity regulates protein homeostasis to control biological outcome. Earlier proteomic/yeast two-hybrid assays identified the HOX (Homeodomain transcription factors) to be targeted by the CRL4 ubiquitin ligase for ubiquitination and degradation [49,60]. As the hematopoietic stem cells (HSCs) and progenitors undergo differentiation, HOX genes are transcriptionally downregulated [61–63] and the HOX proteins are targeted for degradation via the CRL4 ubiquitin ligase to ensure proper differentiation [49]. Importantly, a conserved LXCXE motif was identified in the Helix I region of HOX HD which serves as the CRL4 degron motif that is conserved among all 39 HOX family members [60]. Failure of HOXA9 degradation by CRL4 blocks granulocytic differentiation [49], while transduction of an engineered degradation-resistant HOXB4 into adult HSCs could effectively promote ex vivo expansion of HSCs and multipotent progenitors, and enhance bone marrow engraftment of transduced human adult CD34+ HSCs [60].

The potential significance of CRL4 in the reproductive system was further revealed in the *Cul4a* and *Cul4b* knockout animals: *Cul4a-null* mice exhibit male infertility, yet had no major effect on reproduction of females was observed, revealing the essential roles of CRL4A in male reproduction [64–66]. *Cul4b*$^{-/Y}$ males are also infertile, while no *Cul4b*$^{-/-}$ females can be derived as CUL4B is x-linked. While the redundancy of the two CUL4s accounts for the viability and normal development of *Cul4a*$^{-/-}$ or *Cul4b*$^{-/Y}$ mice, *CUL4A* and *CUL4B* genes are differentially expressed at distinct stages of male meiosis [64–66]. As such, *Cul4a*$^{-/-}$ spermatocytes are arrested at pachytene to diplotene transitions of Meiosis I, while *Cul4b*$^{-/Y}$ sperms are defective at the later stage of spermiogenesis [64–66]. Accumulation of CRL4 target CDT1, as well as p53 was observed, consistent with the increased apoptosis among germ cells [64–66]. While female reproduction remains normal in individual *Cul4a*$^{-/-}$ mice, abrogation of the entire CRL4 ubiquitin ligase led to infertility in females [67]. Interestingly, conditional deletion of *Ddb1* or CRL4 substrate receptors *VprBP/DCAF1* or *DCAF2* cause female infertility possibly via diverse mechanisms including, massive DNA damage and disruption of the cell cycle, TET (Ten-eleven translocation methylcytosine dioxygenase) inactivation-mediated ovulation defects, oocyte loss, or repression on PI3K/AKT pathway [68–70].

Another core system that CRL4 regulates is the central nervous system. Conditional deletion of *Ddb1* in the murine brains leads to neonatal lethality by increased accumulation of p53, although p53 is not considered a direct substrate of CRL4 [57]. Meanwhile, specific ablation of *Ddb1* in the hippocampus and cerebral cortex generates an epileptic phenotype in experimental mice, mechanistically via changes in restriction on the activity of the BK (Ca^{2+} and voltage-activated K^{+}) channel, which further indicates the potentially pathological role of CRL4 in neuroelectrophysiological disorders [71].

CRL4s are also found to function in other organ systems. For instance, hepatocyte-specific deletion of *Ddb1* disables liver gluconeogenesis in part due to increased accumulation of the CRL4 substrate CRY1 (cryptochrome 1), which in turn leads to downregulation of the FOXO1 (forkhead box protein O1)-driven gluconeogenic responses [72]. However, CRL4s seem to play contradictory roles towards hepatic cell expansion under various circumstances. Deletion of *Ddb1* accelerates liver regeneration and trigger spontaneous onset of hepatic carcinogenesis, suggesting a role in controlling cellular proliferation [73]. This phenotypic disparity may be explained by DDB1 engaging in other regulatory networks besides CRL4, which could cloud our understanding of the effect of DDB1 loss due to reduction in CRL4 activity. Therefore, the phenotypic information from *Cul4a* knockout mouse models could be more informative. Moreover, CRL4s may have a role in regulating metabolism, since depletion of *Cul4b* in adipocytes and pancreatic δ cells produces opposing outcomes, namely enhanced insulin sensitivity by stabilizing PPARγ (peroxisome proliferator-activated receptor γ), or glucose intolerance by blocking PRC2 (polycomb repressive complex 2)-induced somatostatin secretion, respectively [74,75]. Meanwhile, CRL4s also demonstrate neoplastic involvement in skin carcinogenesis through restricting the capacity of nucleotide excision repair [55,76], which will be further discussed in subsequent sections (Table 2).

Table 2
Summarized phenotypes of knockout mouse models on CRL4 components.

Gene	Component category	Knockout mode	Phenotype
Cul4a$^{-/-}$	Scaffold	Germline	Normal development/lifespan. No gross abnormalities [55]. Male infertility [64,66]; mild Cardiac hypertrophy and hypertension in male but not female mice [85,249]
		Conditional (skin)	Resistant to UV-induced skin carcinogenesis [55]
		Conditional (liver)	Reduced hepatocyte proliferation after exposure to liver toxins [59]
Cul4b$^{-/Y}$	Scaffold	Germline (epiblast)	Normal development/lifespan; No obvious gross abnormalities [54]; Infertility [65]
		Conditional (pancreatic δ cell)	Decreased glucose intolerance and insulin secretion [75]
		Conditional (adipocyte)	Enhanced expansion of adipose tissue; Increased glucose tolerance and insulin sensitivity [74]
		Conditional (germ cell)	Male infertility [65]
		Conditional (hematopoietic cell)	Increased accumulation and activity of MDSCs post LPS [250]
		Conditional (nervous system)	Elevated amount of astrocytes [251]
		Conditional (myeloid system)	Enhanced peritonitis induced by LPS administration [252]
		Systematic	Embryonic lethality resulting from defective extra-embryonic development [54]; Growth retardation of embryos [253]
Ddb1$^{-/-}$	Adaptor	Germline	Embryonic lethality [57,254]
		Conditional (brain and lens)	Neonatal lethality [57]
		Conditional (skin)	Postnatal lethality [56]
		Conditional (hematopoietic cell)	Bone marrow failure [254]
		Conditional (oocyte)	Ovulation defect [69,70]
		Conditional (liver)	Impaired hepatic gluconeogenesis [72]; Liver regeneration and spontaneous development of hepatocellular carcinoma [73]
		Conditional (hippocampus and cerebral cortex)	Epilepsy [71]
Dcaf1$^{-/-}$	Substrate recognition protein	Germline	Embryonic lethality [255]
		Conditional (B cell)	Impaired B cell maturity and development [256]
		Conditional (T cell)	Insufficiency of T cells [257]
		Conditional (oocyte)	Oocyte death and infertility [69,70]
		Conditional (oocyte)	Female infertility [68]
Dcaf2$^{-/-}$	Substrate recognition protein	Germline	Susceptible to UV-induced skin carcinogenesis [76]

Abbreviations: MDSCs: myeloid-derived suppressor cells; LPS: lipopolysaccharide; UV: ultraviolet.

2.4. Pathological involvement of Cullin 4 E3 ligases-evidence from clinical studies

Consistent with their diverse physiological roles in mouse models, mutations of CRL4 components have also been linked to multiple human pathological conditions, especially in the nervous system. X-linked mental retardation (XLMR), also known as Cabezas Syndrome, is genetically characterized by specific mutations in the X chromosome, which causes short stature, hypogonadism, learning disability, obesity, aggressive outbursts, intentional tremor, pes cavus and seizures among adolescents [77,78]. Current studies have confirmed that mutations in X-linked *Cul4b* gene account for the onset of XLMR, which may be partially attributed to the stability of its substrate WDR5 (WD repeat-containing protein 5) that facilitates the epigenetic silencing of neuronal genes and thus inhibits neurite outgrowth [77–79], although WDR5 accumulation was not observed in *Cul4b*$^{-/-}$ MEFs [54]. Genetic rescue of *Cul4b*-mutant mice significantly decreases the occurrence of XLMR, suggesting a potential of *Cul4b*-targeted therapy [53]. In addition, mutation of *Cul4b* also leads to cerebral malformations, possibly through dysregulation of the function of WDR62 (WD repeat-containing protein 62), a substrate recognition receptor (DCAF) of CRL4 and also a protein whose mutation is frequently detected in patients with microcephaly [80]. Additionally, a specific mutation in *Dcaf8,* leading to the amino acid substitution R317C, disrupts its binding to the DDB1 adaptor and the formation of a functional CRL4 complex, leading to axonal hereditary motor and sensory neuropathy (HMSN2) and giant axons [81]. Moreover, patients bearing a mutant of *Dcaf14* feature developmental retardation, intellectual defects, obesity and dysmorphic characteristics [82]. *Dcaf17* is the causative gene of Woodhouse Sakati Syndrome (WSS), which is characterized by progressive extrapydamidal symptoms, together with hearing loss, hypogonadism, diabetes and learning disability [83]. This experimental evidence verifies the close correlation between CRL4 mal-function and pathogenesis in the central nervous system.

Meanwhile, mutated *Gnb3*, encoding a substrate recognition receptor GNB3 (G protein subunit beta 3), pathologically associates with hereditary hypertension while mutation on *Ddb2* results in xeroderma pigmentosum, which features impaired DNA repair upon UV exposure and higher susceptibility to skin carcinogenesis [84,85]. Considering the diverse physiological roles of CRL4, we may predict that more pathological correlations between CRL4 dysregulation and human disorders may be identified in the near future (Table 3).

2.5. Cullin 4 E3 ligases in tumorigenesis

According to the canonical definition of cancer, there are six hallmarks of cancer pathology, including sustaining proliferative signaling, resisting cell death, evading growth suppression, activating invasion and metastasis, inducing angiogenesis, and enabling replicative immortality [86]. Any regulators that effects these hallmarks are believed to be involved in tumorigenesis, either in a positive or negative manner [87]. Due to the participation of CRL4 in multiple cellular processes, including cell cycle progression and regulation of apoptotic death, its potential role in tumorigenesis has drawn much attention. Current studies have verified a close, but complicated, and context-dependent correlation between CRL4 activity and malignancies, on basis of the evidence from genetic mouse models, clinical pathological specimens and cellular and molecular experiments, which will be discussed in detail below (also see a recent review on pathological role of the CRL4 ubiquitin ligase [88]).

2.6. Cullin 4 scaffold protein in tumorigenesis

As described above, there are two homologous CRL4scaffold proteins, namely CUL4A and CUL4B, which have shared substrates but possess non-overlapping activities. Current investigations implicate that both scaffold proteins exert oncogenic functions in a variety of malignancies, suggesting that the entire CRL4 system may play a role in

Table 3
Major mammalian substrates of CRL4 and their biological impacts.

Substrate recognition protein	Scaffold	Substrate	Modification	Biological impact
DCAF1	CUL4A	LATS1	Degradation	Oncogenic contribution [139,140]
	CUL4A	LATS2	Altered activity	Oncogenic contribution [139]
	CUL4A	HLTF	Degradation	Triggering HIV replication [110]
	CUL4A	Dicer1	Degradation	Enhanced HIV infection [258]; Colon cancer progression [109]
	CUL4A/B	FOXM1	Degradation	Cell cycle arrest [136]
	CUL4A/B	PP2A	Degradation	Crucial for oocyte meiosis and female fertility [67]
	CUL4A/B	MyoD	Degradation	Halted differentiation of skeletal muscle [137]
	CUL4A	MCM10	Degradation	Cell cycle arrest [259,260]
	CUL4B	TR4	Degradation	Resistant to high fat caused fatty liver [261]
	CUL4A/B	SAMHD1	Degradation	Facilitating HIV replication [262]
	CUL4A/B	TET1/2/3	Altered activity	Essential for ovary maturity [69]; Tumor suppression [263]
	CUL4A/B	RORα	Degradation	Transcriptional repression [138]
	CUL4A	UNG2	Degradation	Cell cycle arrest [264,265]
	CUL4A	SMUG1	Degradation	Cell cycle arrest [264]
	CUL4A	NF2	Degradation	Activation of oncogenic pathways [141]
DCAF2	CUL4A	CRY1	Degradation	Promotion of liver gluconeogenesis; Homeostasis of molecular circadian behavior [72,147]
	CUL4A/B	CDT1	Degradation	Stimulating the proliferation of melanoma [150,152]
	CUL4A/B	p21	Degradation	Stimulating the proliferation of melanoma [150]; Regulation of replication licensing [98]
	CUL4A	p53	Degradation	Cell cycle control [266]
	CUL4A/B	SET7	Degradation	Maintaining genomic integrity [143–145]
	CUL4A/B	SET8	Degradation	Stimulating the proliferation of melanoma [150]; DNA damage control [145,267]
	CUL4A/B	PCNA	Altered activity	Enhanced translesion DNA synthesis [268]
	CUL4A/B	p12	Degradation	DNA repair control [269]
	CUL4A	CHK1	Degradation	Cell cycle progression [146]
	CUL4A/B	MMSET	Degradation	Cell cycle arrest [270]
	CUL4A/B	XPG	Degradation	DNA repair [271]
	CUL4A/B	TDG	Degradation	Homeostasis of DNA replication [272]
	CUL4A	SDE2	Degradation	Protection of genomic stability against replication stresses [273]
	CUL4A/B	CDC6	Degradation	Cell cycle regulation [274]
	CUL4A	Tob	Degradation	Apoptotic response to DNA damage [275]
	CUL4A	GCN5	Degradation	Transcriptional suppression [148]
DCAF4L2	CUL4A	PPM1B	Degradation	Increased invasion of colorectal cancer [276]
DCAF7	CUL4A/B	LigI	Degradation	NS [277]
DCAF8	CUL4A	H3	Altered activity	Postnatal liver maturation [278]
	CUL4B	CDC25A	Degradation	NS [170]
DCAF9	CUL4A/B	H2A	Altered activity	Suppressed adipogenesis [279]
DCAF11	CUL4A	NRF2	Degradation	More sensitive to chemotherapies [171]
	CUL4A	SLBP	Degradation	Cell cycle homeostasis [280,281]
	CUL4B	p21	Degradation	Inhibiting cell cycle progression [172]
DCAF15	CUL4A/B	CAPERα	Degradation	Anti-growth effect in cancer cells [173]
AhR	CUL4B	ERα	Degradation	NS [282]
COP1	CUL4A	ETV5	Degradation	Lung homeostasis and tumor suppression [166]
	CUL4A	c-Jun	Degradation	Inhibition on oncogenic transcriptions [167]
COPS8	CUL4A	CENP-A	Subcellular relocalization	Homeostasis of mitosis [283]
CRBN	CUL4A/B	IKZF1/3	Degradation	Anti-tumor impacts against multiple myeloma cells [162]
	CUL4A	ZFP91	Degradation	NS [284]
	CUL4A	GSPT1	Degradation	Anti-tumor impacts against leukemic cells [163]
	CUL4A/B	APP	Degradation	Alleviation of neurodegeneration [159]
	CUL4A	GS	Degradation	NS [185]
	CUL4A/B	CK1α	Degradation	Inhibition on myelodysplastic syndrome [164]
	CUL4A/B	CLC-1	Degradation	Homeostasis of membrane excitability [160]
	CUL4A	SLO1	Altered activity	Preventing epileptogenesis [71]
DDB2	CUL4A	p27	Degradation	Cell cycle progression [156]
	CUL4A	AR	Degradation	Suppression of prostate cancer proliferation [154,285]
	CUL4A/B	H2A	Altered activity	DNA repair [84]
	CUL4A/B	HBO1	Degradation	Suppressed cell proliferation [158]
	CUL4A	DDB2	Degradation	Regulation of DNA damage [286,287]
FBXO44	CUL4B	RGS2	Degradation	NS [288]
FBXW5	CUL4A	DLC1	Degradation	Growth of lung cancer cells [289]
	CUL4A/B	TSC2	Degradation	Homeostasis of cell growth [290]
GNB2	CUL4A	GRK2	Degradation	Cardiovascular protection [249]
GNB3	CUL4A	GRK2	Degradation	Cardiovascular homeostasis [85]
HBx	CUL4A/B	SMC5/6	Degradation	Increased replication of HBV [128]
HOXB4	CUL4A	Geminin	Degradation	Elevated proliferation of hematopoietic stem and progenitor cells [291]
RBBP7	CUL4A/B	CENP-A	Subcellular relocalization	Homeostasis of mitosis [292]
STRAP	CUL4A	PNKP	Degradation	Susceptible to oxidative DNA damage [293]
WDR70	CUL4A/B	H2B	Altered activity	Stability of cell division [294]

(*continued on next page*)

Table 3 (*continued*)

Substrate recognition protein	Scaffold	Substrate	Modification	Biological impact
Other substrates[a]	CUL4A/B	γ-tubulin	Degradation	Stability of centrosome [295]
	CUL4B	HUWE1	Degradation	Reduced apoptosis after DNA damage [296]
	CUL4A/B	AMBRA1	Degradation	Autophagy termination [297]
	CUL4B	p53	Degradation	Impeding stress-induced cellular senescence [298]
	CUL4A	PEX7P	Degradation	Control of peroxisome biogenesis [299]
	CUL4A	HOXB4	Degradation	Decreased proliferation of hematopoietic stem cells [60]
	CUL4B	CSN5	Degradation	Regulation of bone morphogenetic signaling [300]
	CUL4A	p73	Altered activity	Transcriptional repression [301]
	CUL4A	GRK5	Degradation	NS [302]
	CUL4A	ORCA	Degradation	Cell cycle regulation [303]
	CUL4B	WDR5	Degradation	Promoting neurite outgrowth [79]
	CUL4B	Peroxiredoxin III	Degradation	Increased production of cellular reactive oxygen species [304]
	CUL4A	RASSF1A	Degradation	Cell cycle progression [305]
	CUL4B	cyclin E	Degradation	Cell cycle progression [306]
	CUL4A	CHK1	Degradation	Genomic instability [307]
	CUL4B	H3/H4	Altered activity	Facilitating cellular response to DNA damage [308]
	CUL4A	p27	Degradation	Cell proliferation [50]
	CUL4A	HOXA9	Degradation	Myelocytic maturation [49]
	CUL4A	REDD1	Degradation	Activation of mTOR signaling [309]

Abbreviations: COPS8: COP9 signalosome subunit 8; CENP-A: centromere protein A; DCAF2: alias CDT2; CRY1: cryptochrome 1; TR4: nuclear receptor subfamily 2 group C member 2 (alias NR2C2); CRBN: cereblon; ZFP91: zinc finger protein 91; NS: not specified; DCAF11: alias WDR23; CAPERα: coactivator of activating protein-1 and estrogen receptor α; NRF2: nuclear factor erythroid 2 like 2; LATS1: large tumor suppressor 1; FOXM1: forkhead box M1; ETV5: ETS variant 5; AR: androgen receptor; H3: histone H3 protein; ERα: estrogen receptor α; SDE2: SDE2 telomere maintenance homolog; HBx: hepatitis B virus regulatory protein X; SMC5: structural maintenance of chromosome 5; LigI: DNA ligase I; GNB3: G protein subunit beta 3; GRK2: G protein-coupled receptor kinase 2; GSPT1: G1 to S phase transition 1; CDT1: chromatin licensing and DNA replication factor 1; SET8: lysine methyltransferase 5A (alias KMT5A); APP: amyloid precursor protein; SLBP: stem-loop binding protein; PPM1B: protein phosphatase, Mg2+/Mn2+ dependent 1B; WDR70: WD repeat domain 70; GS: glutamine synthetase; HLTF: helicase-like transcription factor; MMSET: nuclear receptor binding SET domain protein 2 (alias NSD2); HBO1: lysine acetyltransferase 7 (alias KAT7); PP2A: phosphatase 2A; CK1α: casein kinase 1A1; MCM10: minichromosome maintenance 10 replication initiation factor; CLC-1: chloride voltage-gated channel 1; FBXO44: F-box protein 44; RGS2: regulator of G protein signaling 2; TET: tet methylcytosine dioxygenase; XPG: Xeroderma pigmentosum group G protein; TDG: thymine DNA glycosylase; SLO1: potassium calcium-activated channel subfamily M alpha 1 (alias KCNMA1); CDC6: cell division cycle 6; FBXW5: F-box and WD repeat domain containing 5; DLC1: DLC1 Rho GTPase activating protein; CHK1: checkpoint kinase 1; RORα: RAR related orphan receptor A; STRAP: serine-threonine kinase receptor associated protein; PNKP: polynucleotide kinase 3′-phosphatase; UNG2: uracil DNA glycosylase 2; SMUG1: single-strand-selective monofunctional uracil-DNA glycosylase 1; GCN5: lysine acetyltransferase 2A (alias KAT2A); PCNA: proliferating cell nuclear antigen; β-TRCP: beta-transducin repeat containing E3 ubiquitin protein ligase; REDD1: regulated in development and DNA damage responses 1; TSC2: TSC complex subunit 2; NF2: neurofibromin 2; AhR: aryl hydrocarbon receptor; ERα: estrogen receptor α; COP1: ring finger and WD repeat domain 2 (RFWD2); CDC25A: cell division cycle 25A; HUWE1: HECT, UBA and WWE domain containing 1; AMBRA1: autophagy and beclin 1 regulator 1; PEX7P: peroxisomal biogenesis factor 7p; HOXB4: homeobox B4; CSN5: COP9 signalosome subunit 5; ORCA: origin recognition complex subunit 1; RASSF1A: RAS association domain family 1 isoform A; CHK1: checkpoint kinase 1; SKP2: S-phase kinase associated protein 2; HOXA9: homeobox A9; SAMHD1: SAM and HD domain containing deoxynucleoside triphosphate triphosphohydrolase 1.

[a] Those proteins are assumed to be the substrates of CRL4 without detailed description of binding motif and interactions with substrate recognition proteins.

tumorigenesis despite having distinct interactions towards cellular substrates, since the scaffold protein exclusively corresponds to the activity of the whole complex [51,89–93].

Mounting evidence suggests critical roles that CUL4A plays in cellular response to DNA damage. Following genotoxic stress, CUL4A targets the CDT1 DNA replication licensing factor, the PR-Set7/Set8 histone methyltransferase, and the p21 cyclin dependent kinase inhibitor for ubiquitin-proteasomal degradation to ensure that damaged DNA is not replicated [94–100]. However, CUL4A also restricts DNA repair capacity post UV. Conditional knockout of *Cul4a* in skin rendered the mice hyper-resistant to UV-induced dermatological carcinogenesis [55]. In response to UV irradiation, cells activate the nucleotide excision repair (NER) pathway for removing UV-induced cyclobutane pyrimidine dimers and 6, 4-photoproducts, and the G1/S DNA damage checkpoint pathway to stop the cell cycle until DNA lesions are repaired by NER. Liu et al showed that CUL4A is a potent inhibitor of DNA damage response by ubiquitin-dependent degradation of DDB2 and XPC, two rate-limiting NER factors responsible for recognition of DNA damage, as well as the p21 effector of the G1/S DNA damage checkpoint pathway. As such, $Cul4a^{-/-}$ cells displayed dramatically enhanced DNA repair and DNA damage checkpoint activities, and *Cul4a* knockout mice are hyper-resistant to UV-induced skin carcinogenesis [55]. Therefore, blocking CUL4A activity represents an attractive new strategy for cancer prevention.

Genetic and pathological evidence suggests that CUL4A is frequently dysregulated in neoplastic events. Gene amplification and transcriptional upregulation are both shown to account for CUL4A overexpression in tumors (e.g. CUL4A and CUL4B are both targets of LEF/TCF transcription factors of the canonical Wnt signaling) [101] Overexpression of CUL4A leads to initiation and progression of lung cancer in mice [89–91]. Consistently, numerous pathological studies have detected upregulation of CUL4A in various human cancer specimens, such as gastric cancer [102–104], breast cancer [104–106], colorectal cancer [104,107–109] and lung cancer [93,110,111], which is also negatively correlated with prognostic survival [93,110–116]. As the core component of the CRL4 complex, the tumorigenic effects of CUL4A overexpression mainly depend on its interactions with downstream targets (details will be discussed in subsequent sections), which trigger cell cycle progression and/or epithelial mesenchymal transition, leading to tumor proliferation, invasion or drug resistance [106,107,117].

CUL4B has also been recognized as a tumorigenic protein in many malignancies. Transgenic upregulation of *Cul4b* induces spontaneous hepatic carcinogenesis [92]. Analyses from clinical samples show that aberrant expression of CUL4B has been observed in a wide spectrum of malignant tumors, especially lung cancer [93,118], colorectal cancer [115,116] and pancreatic cancer [119], serving as a negative indicator of patient survival as well [116,120]. Analogous to CUL4A, abnormal interactions between CRL4B and substrates directly link to activation of proliferative pathways (such as the Wnt/β-catenin pathway) and epigenetic silencing, participating in nearly every aspect of tumor progression [121,122]. On the other side, downregulation of certain

Table 4
Neoplastic roles of CRL4 components.

Component	Neoplastic role	Physiological evidence (knockout or transgenic mouse models)	Pathological evidence (cancer relevant human specimens)
CUL4A	Oncogenic	Lung cancer initiation and progression (transgenic overexpression of *Cul4a*) [89–91]; Resistant to UV-induced skin carcinogenesis (skin-specific knockout of *Cul4a*) [55]	Overexpressed in multiple myeloma [310], cholangiocarcinoma [113,114], gastric cancer [102–104], colorectal cancer [104,107–109], ovarian cancer [104,112,311], lung cancer [93,110,111], breast cancer [104–106], osteosarcoma [312,313], hepatocellular cancer [117,314], malignant pleural mesothelioma [315,316], pituitary adenoma [317], prostate cancer [318]
CUL4B	Oncogenic	Spontaneous development of liver cancer (transgenic overexpression of *Cul4b*) [92]	Overexpressed in cholangiocarcinoma [120], pancreatic carcinoma [119], lung cancer [93,118], colorectal cancer [115,116], glioma [319], liver cancer [122], cervical cancer [121], esophageal cancer [320], ovarian cancer [112]
RBX1	Oncogenic	N/A	Overexpressed in ovarian cancer [123], lung cancer [124], gastric cancer [125], bladder cancer [126]
DDB1	Context-dependent	Spontaneous development of liver cancer (hepatocyte-specific knockout of *Ddb1*) [73]	Overexpressed in ovarian cancer [123]

miRNAs such as miR-194 and miR-300 may explain, in part, the mechanisms inducing the overexpression of CUL4B overexpression in several cancers, although its upstream mechanisms remain largely unknown [118,119] (Table 4).

2.7. The RING-finger protein RBX1 and adaptor DDB1 in tumorigenesis

Similar to the Cul4A and Cul4B scaffold proteins, the catalytic component, RING-finger RBX1 is also regarded as an oncoprotein in ovarian cancer [123], lung cancer [124], gastric cancer [125], and bladder cancer [126]. Nevertheless, both genetically engineered mouse models and mechanistic studies are still limited.

The adaptor DDB1, it displays a context-dependent role in tumorigenesis as well. Results from knockout mouse models show that hepatocyte-specific ablation of *Ddb1* facilitates spontaneous development of liver cancer [73], whereas overexpression of DDB1 is detected in ovarian cancer, consistent with the oncogenic role of other CRL4 complex components [123]. This context-dependent role of DDB1 may reflect its diversity on neoplastic contributions of the CLR4 complexes, or is possibly a result of other non-CLR4 related activities, since DDB1 is not an exclusive protein for CRL4 and could involve other regulatory networks in cancer cells. In this regard, DDB1 was initially identified as a component of the UV-DDB complex that surveys the chromosomes for UV-induced DNA lesions and recruits the nucleotide excision repair apparatus to sites of DNA damage (Table 4) [127].

2.8. Cullin 4 substrate proteins in tumorigenesis

2.8.1. Oncogenic members

Similar to the oncogenic functions of the CUL4A/B scaffolds, the majority of the CRL4 substrate recognition receptors play tumorigenic roles. HBx (hepatitis B virus regulatory protein X) is a recombinant protein encoded by the HBV genome (hepatitis B virus) and expressed following infection in human hepatocytes. This protein can function as a substrate recognition receptor of CRL4 leading to the destabilization of SMC5/6 (structural maintenance of chromosome 5/6) to increase the replication of HBV [128]. Transgenic overexpression of *Hbx* induces liver carcinogenesis in rodent models [129,130], which is also found to be upregulated in human liver cancer specimens [131]. Increased HBx expression is also observed in intrahepatic cholangiocarcinoma [132] and adenoid cystic carcinoma [133], suggesting a general involvement of HBx in promoting tumorigenesis. Mechanistically, apart from its involvement in HBV replication, SMC5/6 also has a vital role in regulating cell division and proliferation while HBx could interact with apoptotic machinery to stimulate downstream oncogenes [134,135], which together explain the oncogenic impact of HBx in human cancers (Tables 3 and 5).

DCAF1 is one of the primary and well-defined substrate recognition receptors of CLR4 complexes, where more than 15 proteins have been identified as substrates of DCAF1. By interacting with target substrates, resulting in either degradation or non-degradation outcomes, DCAF1 participates in multiple physiological and pathological processes, including cell cycle arrest [136], germ cell meiosis [67], virus replication [110], skeletal muscle differentiation [137] and transcriptional repression [138]. As for its neoplastic roles, DCAF1 has been regarded as an important oncogenic component of the CRL4 complex, where it is found to be upregulated in both ovarian cancer [136] and colon cancer specimens [109]. This oncogenic role may be mechanistically attributed to its destabilizing effects on substrates such as LATS1 (large tumor suppressor 1) [139,140], Dicer1 [109], and NF2 (neurofibromin 2) [141], which normally act as tumor suppressors to inhibit the activity of multiple downstream oncogenic pathways, including Hippo/YAP [139,140] and JAK/STAT3 [109]. Interestingly, the tumor suppressor NF2 is a direct inhibitor of CRL4-DCAF1/VPRBP, and tumor-associated mutations of Merlin either fail to bind DCAF1 or are unable to move into the nucleus to block DCAF1 ubiquitin ligase activity [142]. However, genetically engineered mouse models are still necessary to fully appreciate the oncogenic roles of DCAF1 (Tables 3 and 5).

Besides DCAF1, DCAF2 is also a well-investigated substrate receptor of CRL4, with more than 15 defined downstream ubiquitin substrates. By destabilizing or activating specific substrates, it is involved in the regulation of various biological events, including genomic stability [143–145], cell cycle progression [146], gluconeogenesis [72,147] and transcriptional suppression [148]. As for a role in tumorigenesis, DCAF2 has been recognized as a major oncogenic receptor of CRL4. Elevated levels of DCAF2 are frequently detected in head and neck squamous cell carcinoma [149], melanoma [150], ovarian cancer [123] and Ewing sarcoma [151]. Mechanistic studies have confirmed that degradation of CDT1, p21, and SET8 (set-domain histone methyltransferase-8) by CRL4 (DCAF2) regulates histone methylation and stimulates proliferation of melanoma, serving as a potential target of anti-melanoma therapeutics [150,152]. However, biochemical evidence of a role for DCAF2 in other malignancies remains limited, let alone in rodent models (Tables 3 and 5). Recently, WDR4 (WD repeat 4-containing cullin-RING ubiquitin ligase 4) has been reported as an oncogenic receptor of CRL4 by promoting the degradation of the tumor suppressor PML (Promyelocytic leukemia) via ubiquitination to re-modulate an immunosuppressive tumor microenvironment in lung cancer setting [153].

In addition to the aforementioned receptors, there are CRL4 substrate recognition receptors responsible for triggering the onset and progression of malignancies, including overexpression of DCAF6 in prostate cancer [154], DCAF13 in hepatocellular carcinoma [155]. The majority of substrate-mediated mechanisms are centered on cell cycle progression, genomic instability, and regulation of apoptosis, suggesting a therapeutic potential in targeting CRL4 ubiquitin ligase

Table 5
Major oncogenic substrate recognition proteins.

Substrate recognition protein	Physiological evidence (knockout or transgenic mouse models)	Pathological evidence (cancer relevant human specimens)	Biochemical evidence (cancer relevant substrates)
DCAF1	NA	Overexpressed in ovarian cancer [136], colon cancer [109]	LATS1/2 [139,140], Dicer1 [109], NF2 [141]
DCAF2	NA	Overexpressed in head and neck squamous cell carcinoma [149], melanoma [150], ovarian cancer [123], Ewing sarcoma [151]	CDT1 [150,152], p21 [150], SET8 [150]
DCAF4L2	NA	Overexpressed in colorectal cancer [276]	PPM1B [276]
DCAF6	NA	Overexpressed in prostate cancer [154]	NA
DCAF13	NA	Overexpressed in hepatocellular carcinoma [155]	NA
AhR	NA	Overexpressed in liver cancer [321], thyroid cancer [322]	NA
COPS8	NA	NA	CENP-A [323]
FBXO44	NA	Overexpressed in breast cancer [324]	NA
FBXW5	NA	NA	DLC1 [289], TSC2 [290,325]
HBx	Hepatocarcinogenesis in transgenic *Hbx* mice [129,130]	Overexpressed in liver cancer [131], intrahepatic cholangiocarcinoma [132], adenoid cystic carcinoma [133]	NA
RBBP7	NA	NA	CENP-A [292,323]
STRAP	NA	Overexpressed in colon cancer [326], lung cancer [326]	NA
HOXB4	NA	Overexpressed in acute myeloid leukemia [327], nephroblastoma [328]	Geminin [329]

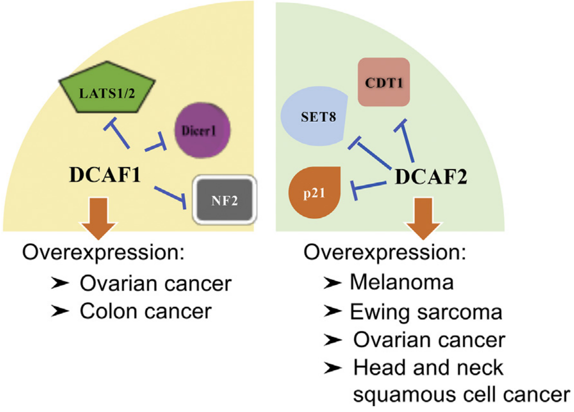


Fig. 2. Major oncogenic substrates of CRL4.

(Fig. 2, Tables 3 and 5).

2.9. *Tumor suppressive members*

Despite mounting evidence of an oncogenic role of CRL4, many substrate receptors of CRL4 have demonstrated tumor suppressive effects, adding diversity and complexity into the regulatory landscape of CRL4 in tumorigenesis.

2.9.1. *DDB2 (damaged DNA binding 2)*

DDB2 is recognized as one of the most consequential tumor suppressors in human cancer. Physiologically, the majority of CRL4 (DDB2)-substrate interactions lead to alterations in cell cycle regulation and DNA repair, hinting at an underlying role in neoplastic control [84,156]. Genetic ablation of *Ddb2* sensitizes rodent models to UV-induced skin tumorigenesis [76] while downregulated expression of DDB2 is commonly observed in cases of human prostate cancer [154], skin cancer [84] and colorectal cancer [157], which support the tumor suppressive role of DDB2. Moreover, studies have shown that destabilization of oncogenic substrates, including HBO1 (histone acetyltransferase bound to ORC 1) [158], may contribute to the cancer inhibitory effects of DDB2 (Tables 3 and 6).

2.9.2. *Cereblon (CRBN)*

Besides DDB2, cereblon (CRBN) is another vital substrate receptor of CRL4 found negatively correlated with tumorigenic behaviors. Owing to the variety of downstream substrates, CRL4 (CRBN) has multiple impacts on cellular biology, especially in neurodegeneration [159], anti-epileptogenesis [71] and homeostasis of membrane excitability [160]. The anti-tumor role of CRBN is usually centered on hematological malignancies, whose expression level is dramatically decreased in multiple myeloma cells [161]. Although IKZF1/3 (Ikaros family zinc finger protein 1/3), GSPT1 (G1-to-S phase transition 1) and CK1 (casein kinase 1α) may not be traditional substrates of CRBN, the induced turnover of IKZF1/3 [162], GSPT1 [163] and CK1α [164] by CRL4 (CRBN) accounts for the inhibitory effects of thalidomide-based molecules (thalidomide, lenalinomide, and pomalidomide) against multiple myeloma, leukemia, and myelodysplastic syndrome, respectively, which may mechanistically correlate to the inhibition on cell cycle progression (Tables 3 and 6). The role of thalidomide-based molecules inducing CRBN-dependent degradation of various key tumorigenic factors will be discussed further below.

2.9.3. *COP1 (constitutive photomorphogenesis 1)*

Expression of COP1 is suppressed in renal cell carcinoma [165]. Moreover, biochemical studies suggest that by destabilizing its substrate ETV5 (ETS variant 5) and c-Jun, CRL4 (COP1) suppresses multiple oncogenic transcription factors in lung tumorigenesis, suggesting a tumor suppressive function [166,167]. These results have indicated a role for COP1 in cancer regulation. *Cop1* knockout mice are embryonic lethal, whereas *Cop1*$^{+/-}$ mice are viable and fertile [168]. Through the generation of the Cop1 hypomorphic alleles, in which the Cop1 protein level was reduced by 90%, shows a 15–20% reduction in body weight compared to wild-type mice, coupled with decreased organ sizes [168] (Tables 3 and 6).

2.9.4. *WDR70 (WD repeat domain 70)*

Loss-of-function mutations of *WDR70* have been identified in ovarian cancer [169]. WDR70 mediates mono-ubiquitination of histone H2B, thereby ensuring H2B stability during cell division and preventing malignant transformation [169]. DCAF8, DCAF11 and DCAF15 have been identified as potential tumor suppressors due to their effects on regulating the stability of CDC25A (cell division cycle 25A) [170], NRF2 (nuclear factor erythroid 2-related factor 2) [171], p21 [172] and CAPERα (coactivator of activating protein-1 and estrogen receptor α) [173], respectively (Fig. 3, Tables 3 and 6).

2.10. *Context-dependent members*

Unlike those members with specific neoplastic contributions, context-dependent substrate recognition receptors are rarely identified. Both GNB2 (G protein subunit beta 2) and GNB3 are substrate receptors

Table 6
Major tumor suppressive substrate recognition proteins.

Substrate recognition protein	Physiological evidence (knockout or transgenic mouse models)	Pathological evidence (cancer relevant human specimens)	Biochemical evidence (cancer relevant substrates)
DCAF8	NA	NA	CDC25A [170]
DCAF11	NA	NA	NRF2 [171], p21 [172]
DCAF15	NA	NA	CAPERα [173]
COP1	NA	Downregulated in renal cell carcinoma [165]	ETV5 [166], c-Jun [167]
CRBN	NA	Downregulated in multiple myeloma [161]	IKZF1/3 [162], GSPT1 [163], CK1α [164]
DDB2	Susceptible to UV-induced skin carcinogenesis (systemic *Ddb2* knockout) [76]	Downregulated in prostate cancer [154], skin cancer [84], colorectal cancer [157]	AR [154,285], HBO1 [158]
WDR70	NA	Loss-of-function mutation in ovarian cancer [169]	H2B [294,330]

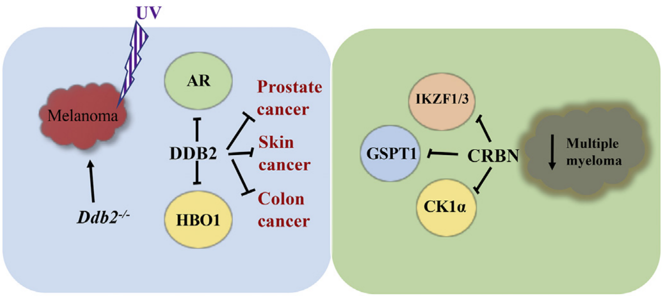


Fig. 3. Major tumor suppressive substrates of CRL4.

for CRL4, which degrade GRK2 (G protein-coupled receptor kinase 2) resulting in cardiovascular protective effects. However, due to the inconsistent roles of GRK2 in tumorigenesis, it is thought to be an oncoprotein in breast cancer [174] and tumor suppressor in hepatocellular carcinoma [175], these two receptors are accordingly considered as context-dependent members, despite a lack of clinical or mouse models studies [175–177] (Tables 3 and 7).

2.11. Degrons recognized by Cullin 4 substrate receptors

Degrons, specific short amino acid within a target protein, is critical for the interaction of target protein with their E3 ligase receptors [178]. Degrons have been identified within many E3 ligase receptor substrates, such as the ETGE degron motif for KEAP1 [179]; and the D-box, the KEN box and the ABBA motif for the APC/C E3 ligase complex [180]. More importantly, regulation of degron recognition and/or availability including through post-translational modifications such as phosphorylation, methylation, acetylation and hydroxylation, has been reported to promote or inhibit substrate recognition by E3 ligases in response to environmental stimuli. For instance, the substrate recognition receptor β-TRCP recognizes a phosphorylated DSG degron [181], and pVHL utilizes the prolyl hydroxylated degrons [182]. Conceivably, inherited/somatic mutations have been identified in degrons contributing to the accumulation of oncoproteins through escaping E3 ligase-mediated degradation, leading to various diseases, including cancer [178]. Thus, defining the degrons within substrates of E3 receptors is not only crucial to validate the direct interaction of a target protein with its E3 ligase receptor, but also highlight the potential to design therapeutics to target the oncoproteins for degradation.

CRL4 substrate receptors also display recognition of their substrates in a degron dependent manner. For instance, COP1 has been characterized to bind degrons within substrates with the consensus amino acid sequence EExxxVP[D/E] (Fig. 4A) [183], and Cdt2, also termed DCAF2, recognized its substrates containing a PCNA-interacting peptide (PIP) degron (Fig. 4B) [184]. Interestingly, DCAF1, harboring a putative chromo domain, has been demonstrated to read a “mono-methyl degron” (Fig. 4C) [138], whereas, Cereblon (CRBN), a direct thalidomide teratogenicity target protein, recognizes an “acetylated degron” in the presence of high glutamine (Fig. 4D) [185]. However, due to a limited number of substrates identified for CRL4 receptors, more effort is necessary in identifying CRL4 substrates, and degrons within those substrates, to clarify the exact degron requirements. These will benefit biomedical studies and further improve development of cancer treatments, especially in utilizing the PROTAC technology [186], which has been considered a novel mechanism targeting what has been long thought of as “undruggable” targets.

2.12. Harnessing the ubiquitin ligase for targeted degradation of cellular proteins

During the past two decades, targeted therapy has become the most promising strategy in modern medicine through the identification and development of small therapeutic molecules targeting mutated or modified factors driving cancer. In addition, biologics such as antibody-based therapeutics, RNA interference, and CRISPR-mediated genetic modifications, have gained much interest recently in drug discovery.

The primary mechanism behind antibody-based biologics is based on antigen-antibody interactions that may specifically and potently block the activity of pathogenic antigens to achieve therapeutic goals, which has been successful in cases such as bevacizumab for VEGFR (vascular endothelial growth factor receptor) and cetuximab for EGFR (epidermal growth factor receptor) [187]. However, since their ionic properties are naturally rejected by the non-ionic cell membrane, these antibody-based biologic drugs are only effective to extracellular or membrane proteins and also costly in nature, which greatly limits their clinical applications [186]. In addition, the RNAi (RNA interference) technology has emerged as a vital addition to antibody-based regimens, which focuses on degrading intracellular mRNA to restrict the translation and expression of specific proteins via pre-designed siRNA. Nevertheless, because of the instability of siRNA (small interfering RNA) under physiological circumstances, RNAi treatment displays unfavorably oral bioavailability and often poor tissue distribution, which has largely limited its usage thus far to liver diseases [188]. In consideration of these challenges, a better small molecule with intracellular permeability, increased bioavailability, and widespread distribution

Table 7
Major context-dependent substrate recognition proteins.

Substrate recognition protein	Pathological evidence (cancer relevant human specimens)	Biochemical evidence (cancer relevant substrates)
GNB2	NA	GRK2 [175–177]
GNB3	NA	GRK2 [175–177]

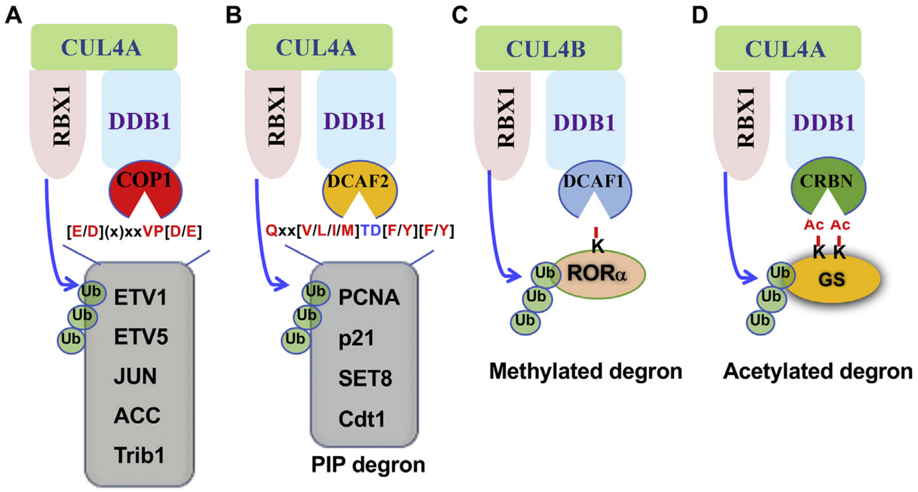


Fig. 4. A schematic illustration of degrons that are recognized by CRL4 receptors.

may therefore provide wider bioavailability and efficacy.

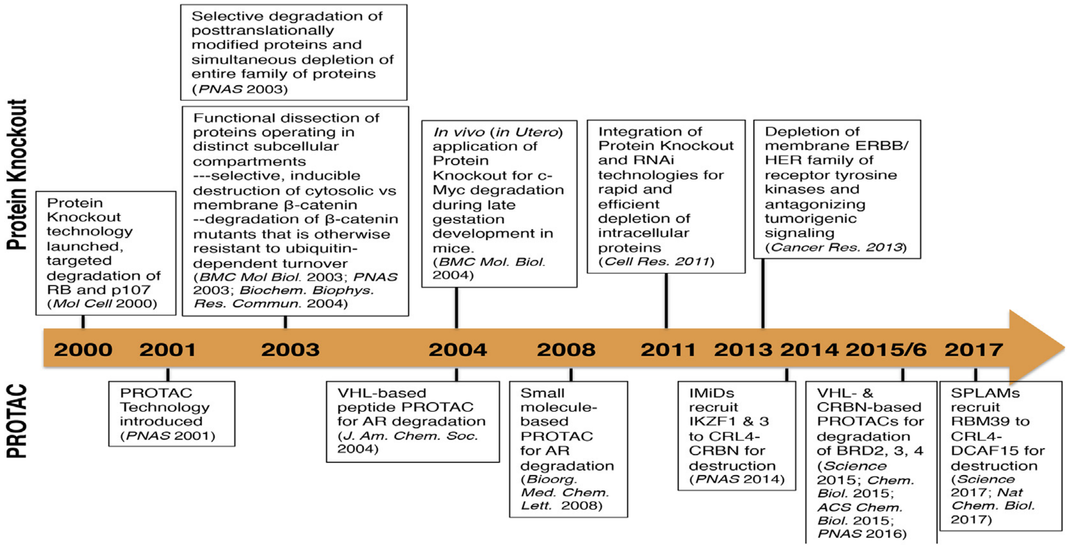
2.13. Protein knockout

Most intracellular proteins are directed to the ubiquitin machinery for proteolytic destruction. Owing to the outstanding selectivity and degradative proficiency of the ubiquitin machinery, nature has chosen ubiquitin ligases as a prime apparatus for eliminating key target proteins, thereby altering a desired cellular process or pathway (Fig. 5). In 1993, Scheffner and colleagues described the first case of viral hijacking of the ubiquitin ligase to promote tumorigenesis: the high-risk human papillomavirus (HPV) types 16 and 18 utilize their oncogenic E6 oncoprotein to recruit p53 tumor suppressor to the cellular E6AP ubiquitin ligase for ubiquitination and proteasomal degradation [189]. Here E6 functions as a bridging peptide that brings E6AP and p53 in close proximity to facilitate ubiquitin transfer onto p53.

Given that ubiquitin ligases are modular proteins and substrate specificity is dictated by protein-protein interactions, and inspired by the hijacking mechanism of the HPVs, Zhou et al. set out to engineer SCF$^{\beta\text{-TRCP}}$ ubiquitin ligase to target cellular proteins which are otherwise not substrates of β-TRCP or escaped recognition by β-TRCP due to oncogenic mutations of the DSG degron [190,191]. This is achieved by covalent attachment of specific binding peptide of the desired target to β-TRCP or truncated β-TRCP deleted of the substrate-binding WD40 domain, thereby enabling recruitment of desired target protein to the SCF machinery for ubiquitination and proteasomal degradation. This engineered ubiquitination machinery, designated protein knockout (PKO), was employed to successfully deplete a variety of intracellular or membrane proteins of interest, including pRB, p107, p130, β-catenin, c-Myc, EGFR, ErbB2/HER2, ErbB3/HER3, as well as viral oncoproteins (HIV16 E1) [191–197]. Moreover, the use of PKO demonstrated the remarkable versatility in selective targeting of post-translationally modified subpopulation of desired cellular proteins, a unique function that is not attainable by CRIPSPR-Cas9, RNAi and antisense technologies [191–197]. Furthermore, PKO can be integrated with RNAi to block protein synthesis and speed up protein destruction simultaneously, thereby achieving more rapid and effective depletion of stable proteins [198]. Taken together, these studies set the stage for harnessing ubiquitin ligases in degrading cellular proteins of interest, and opened up new avenues to design and develop therapeutic strategies in drug discovery.

2.14. PROTAC (PROteolysis TArgeting Chimera)

PROTAC technique has been established as an alternative to current small molecule therapies. PROTACs feature a bimodal molecule that



Selective degradation of posttranslationally modified proteins and simultaneous depletion of entire family of proteins (*PNAS* 2003)

Protein Knockout technology launched, targeted degradation of RB and p107 (*Mol Cell* 2000)

Functional dissection of proteins operating in distinct subcellular compartments ---selective, inducible destruction of cytosolic vs membrane β-catenin --degradation of β-catenin mutants that is otherwise resistant to ubiquitin-dependent turnover (*BMC Mol Biol.* 2003; *PNAS* 2003; *Biochem. Biophys. Res. Commun.* 2004)

In vivo (*in Utero*) application of Protein Knockout for c-Myc degradation during late gestation development in mice. (*BMC Mol. Biol.* 2004)

Integration of Protein Knockout and RNAi technologies for rapid and efficient depletion of intracellular proteins (*Cell Res.* 2011)

Depletion of membrane ERBB/HER family of receptor tyrosine kinases and antagonizing tumorigenic signaling (*Cancer Res.* 2013)

PROTAC Technology introduced (*PNAS* 2001)

VHL-based peptide PROTAC for AR degradation (*J. Am. Chem. Soc.* 2004)

Small molecule-based PROTAC for AR degradation (*Bioorg. Med. Chem. Lett.* 2008)

IMiDs recruit IKZF1 & 3 to CRL4-CRBN for destruction (*PNAS* 2014)

VHL- & CRBN-based PROTACs for degradation of BRD2, 3, 4 (*Science* 2015; *Chem. Biol.* 2015; *ACS Chem. Biol.* 2015; *PNAS* 2016)

SPLAMs recruit RBM39 to CRL4-DCAF15 for destruction (*Science* 2017; *Nat Chem. Biol.* 2017)

Fig. 5. Timeline illustrating crucial discoveries of the protein knockout and PROTAC techniques.

simultaneously connects an E3 ligase and the target protein, leading to ubiquitin-dependent degradation on specific substrates [199]. This method creates an opportunity to degrade specific proteins that currently could not be pharmaceutically targeted by small molecules or without characterized E3 ligases, and could theoretically lead to the proteolysis of any substrate via only a single E3 ligase. Therefore, identifying appropriate binding ligands for E3 ligases and substrates is the most critical and challenging step for PROTAC design (Fig. 5).

The first successful model of PROTAC was reported in 2001 by Sakamoto and colleagues, who creatively designed a chimeric molecule with a small ligand for MetAP2 (methionine aminopeptidase 2) and a 10-amino-acid phosphopeptide for β-TRCP, a substrate recognition receptor for CRL1 [200]. Later, both the Ivan and Jaakkola groups described a VHL (von Hippel-Lindau)-based PROTAC design which promoted the degradation of FKBP12 (the 12-kDa FK506-binding protein) and AR via the VHL E3 ligase [202]. The HIFα (hypoxia inducible factor α) moiety, which was a natural substrate for CRL2 (VHL), was therefore incorporated into the PROTAC molecule as a ligand for VHL, a substrate receptor of CRL2. The hydroxylation of Proline-564 on HIFα was prerequisite and necessary for recruiting VHL [182,201]. Therefore, this hydroxyproline-containing chimeric molecule became a classical representative of peptide-based PROTACs. Nonetheless, due to the peptidic nature of HIFα moiety in this PROTAC, its inefficient intracellular delivery limits in vivo applications. The hydroxyproline core is actually a non-ionic subunit of the HIFα moiety, and a re-designed VHL ligand featuring a sole hydroxyproline core greatly reduces the peptidic nature, while still retains binding affinity, which is therefore defined as small molecule-based PROTAC and more amenable for delivery to intracellular targets [203] (Details will be discussed in subsequent sections) (Fig. 6 and Table 9).

3. Small molecule-based PROTACs

3.1. CRL2 (VHL)-based PROTACs

The peptidic features of PROTACs limited its clinical application, which led to the design of less ionic and therefore a more bioavailable type of PROTAC, later defined as small molecule PROTACs [186]. The first report of a CRL2 (VHL)-based small molecule PROTAC emerged in 2012, which featured a hydroxyproline core inside the ligand for VHL with the unnecessary residues of HIFα moiety trimmed to minimize the molecular weight and peptide nature, allowing greater penetration of the cell plasma membrane [204–206]. In 2015, the degrading effects of this small molecule CRL2 (VHL)-based PROTAC was demonstrated in preclinical cancer models where this PROTAC displayed marked degradation of specific proteins of more than 90%, including RIPK2 (receptor interacting serine/threonine kinase 2), ERRα (estrogen-related receptor α) and HaloTag fusion proteins [203,207]. Therefore, these results demonstrate a potential for PROTACs as a novel therapeutic option against numerous diseases, especially cancers. Currently, more preclinical data is necessary before assessing therapeutic efficacy in clinical trials (Table 10).

3.2. Thalidomide derivatives and CRL4 (CRBN)-based PROTAC

As mentioned above, thalidomide and its derivatives are able to directly target the substrate receptor CRBN of CRL4, promoting the degradation of multiple substrates for anti-tumor effects especially in multiple myeloma [162,208,209]. Since these drugs feature non-ionic structures and are therefore suitable as small molecule ligands, many CRL4 (CRBN)-based PROTACs have been constructed via utilizing the moiety of thalidomide derivatives as the ligand for CRBN in conjugation with specific inhibitors as the substrate ligands.

In 2015, Winter and colleagues reported that by assembling phthalimide moiety and BET antagonist, a heterobifunctional PROTAC was formed which bridged CRBN and BRD4 (bromodomain-containing protein 4), leading to the highly selective degradation of BRD4 and consequently demonstrating anti-proliferative and tumor inhibitory effects in mouse models [210,211]. Additionally, with an FKBP12 inhibitor as the substrate ligand, the phthalimide-equipped PROTAC could also connect CRBN with FKBP12 to facilitate its ubiquitin-dependent degradation [211]. Apart from the phthalimide-characterized PROTACs, thalidomide-equipped PROTACs have also emerged, which lead to the degradation of Sirtuin 2 (SIRT2) [212] and CDK9 (cyclin-dependent kinase 9) [213], implicating their possible applications in regulating cell cycle and epigenetic stability (Table 10).

3.3. Other major E3 ligases for PROTAC technology

The MDM2-based PROTAC was the first reported example of small molecule PROTACs [214]. It was constructed by a nonsteroidal AR ligand for AR recognition, while the MDM2 ligand was a polyethylene glycol (PEG)-contained nutlin, a pharmaceutical inhibitor of MDM2. Schneekloth and colleagues discovered that the expression of AR was effectively inhibited in HeLa cells after administration of MDM2-based PROTAC which could be reversed by the proteasome inhibitor epoxomicin, displaying therapeutic potential against AR-positive human malignancies [214,215].

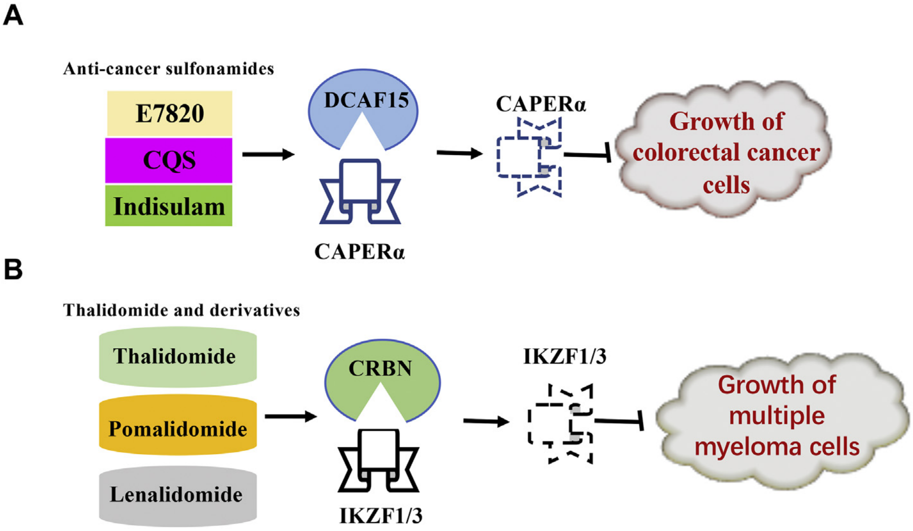


Fig. 6. Major CRL4 targeted drugs and pharmacological mechanisms.

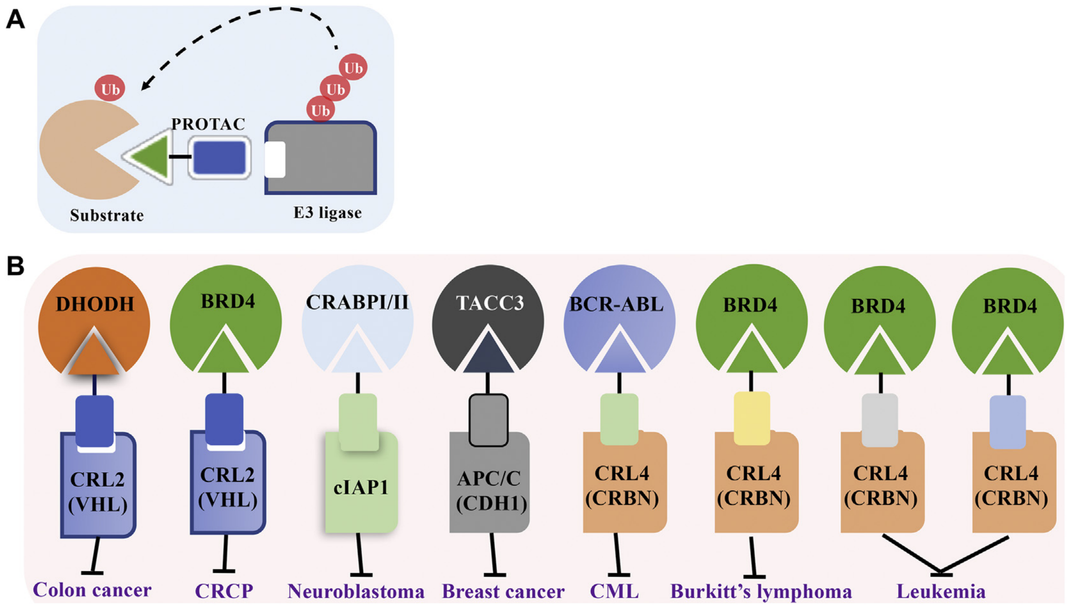


Fig. 7. Anti-tumor PROTACs and their working mechanisms.

Table 8
Major CRL4s targeted therapies.

Drug	Mechanism	Function	Preclinical evidence	Clinical trial	FDA approval
E7820	Promoting DCAF15-CAPERα degradation	Anti-growth in colorectal cancer cells [247]	Yes	Completed	Not yet
CQS	Promoting DCAF15-CAPERα degradation	Anti-growth in colorectal cancer cells [247]	Yes	Completed	Not yet
Indisulam	Promoting DCAF15-CAPERα degradation	Anti-growth in colorectal cancer cells [173,247]	Yes	Completed	Not yet
CC-220	Promoting CRBN- IKZF1 and IKZF3 degradation	Anti-tumor effect against multiple myeloma cells [331]	Yes	Ongoing	Not yet
Lenalidomide	Promoting CRBN- CK1α degradation	Anti-tumor effect against myelodysplastic syndrome [164]	Yes	Completed	Approved
	Promoting CRBN- IKZF1 and IKZF3 degradation	Anti-tumor effect against multiple myeloma cells [162,208,209]	Yes	Completed	Approved
Thalidomide	Promoting CRBN- IKZF1 and IKZF3 degradation	Anti-tumor effect against multiple myeloma cells [248]	Yes	Completed	Approved
Pomalidomide	Promoting CRBN- IKZF1 and IKZF3 degradation	Anti-tumor effect against multiple myeloma cells [248]	Yes	Completed	Approved
CC-885	Promoting CRBN- GSPT1 degradation	Anti-tumor effect against leukemic cells [163]	Yes	Not yet	Not yet
Pevonedistat	Inactivation of DCAF2-SET8/p21 degradation	Anti-tumor effect against melanoma cells [150]	Yes	Completed	Not yet

Abbreviations: CQS: chloroquinoxaline sulfonamide; CC-220: alias Compound 6.

Additionally, the cIAP1-based PROTAC has also been designed, featuring a bestatin-incorporated ligand for E3 ligase cIAP1 [215]. According to different target ligands for substrates, this set of PROTACs could successfully lead to the degradation of ERα [216,217], RAR (retinoic acid receptor) [216] and CRABP I/II (cellular retinoic acid binding protein I/II) [218,219], involved in multiple biological activities such as metastasis of neuroblastoma cells (Table 10).

4. PROTAC: Major biological impacts

4.1. Peptide PROTAC

As the first-generation of PROTAC technology, peptide PROTACs had limited tissue distributions and membrane penetration. However, these PROTACs still exhibited various biological effects via degrading specific substrates.

CRL2 (VHL) is the major E3 ligase that is utilized for experimental design of peptide PROTACs. Targeted turnover of ER (estrogen receptor), FRS2α (fibroblast growth factor receptor substrate 2 α) and tau, VHL-based PROTACs lead to inhibition of endothelial cell differentiation [220–222], neuronal differentiation [223] and reduced proteotoxic effects in AD (*Alzheimer's* Disease) mouse models [224], respectively, demonstrating vital physiological participation and therapeutic potential against pathological conditions. Additionally, the HBx-targeted PROTAC also displayed protective roles against HBV (hepatitis B virus) infection and liver carcinogenesis [225] while the Smad3-targeted PROTAC prevented the progression of renal fibrosis [226]. Meanwhile, despite a lack of in vivo evidence, the remaining PROTACs are believed to have crucial roles in multiple biological events, especially for those targeting AKT [227], AR [202] and MetAP-2 [220] for degradation.

Besides CRL2 (VHL), there are also two PROTACs, namely PROTAC-3 and PROTAC-2, that utilize CRL1 (β-TRCP) as E3 ligases to destabilize AR and ER, respectively [228]. Nonetheless, although the technology works well to degrade target proteins in vitro, more in vivo evidence are necessary to evaluate their potential efficacy.

Currently, although peptide PROTACs are no longer a primary focus in the development of PROTAC technology due to difficulties described above, there may still be instances where there is therapeutic value of peptide PROTACs under certain circumstances, which maybe act as alternative options to small molecule PROTACs in the future (Table 9).

Table 9
Major peptide PROTACs and their biological impacts.

Name	Target	Target ligand	E3 ligase	E3 ligase ligand	Biological impacts
Api-PROTAC	AHR	Apigenin	CRL2 (VHL)	A pentapeptide sequence derived from HIF1α	Potential roles against AHR positive disorders [332,333]
Tri_a-PROTAC	AKT	PCC	CRL2 (VHL)	A heptapeptide derived from HIF1α	Potential roles against AKT positive disorders [227]
PROTAC-3	AR	DHT	CRL1 (β-TRCP)	A phosphopeptide derived from IκBα	Potential roles against AR positive disorders [228]
PROTAC-5	AR	DHT	CRL2 (VHL)	A heptapeptide derived from HIF1α	Potential roles against AR positive disorders [202]
PROTAC-2	ER	Estradiol	CRL1 (β-TRCP)	A phosphopeptide derived from IκBα	Potential roles against ER positive disorders [228]
E2-SMPI	ER	Estradiol	CRL2 (VHL)	An octapeptide or pentapeptide derived from HIF1α	Inhibition of endothelial cell differentiation [220–222]
PROTAC-4	FKBP12	AP21998	CRL2 (VHL)	A heptapeptide derived from HIF1α	Potential roles against FKBP12 positive disorders [202]
phosphoPROTAC-1	FRS2α	Tyrosine phosphorylation sequences of TrkA	CRL2 (VHL)	A heptapeptide derived from HIF1α	Inhibition of neuronal differentiation [223]
HBx-PROTAC	HBx	The oligomerization domain of HBx	CRL2 (VHL)	ODD domain of HIF1α	Potential roles against HBV infection and development of liver cancer [225]
Fu-SMPI	MetAP-2	Fumagillol	CRL2 (VHL)	An octapeptide derived from HIF1α	Potential roles against MetAP-2 positive disorders [220]
phosphoPROTAC-2	PI3K	Tyrosine phosphorylation sequences of ErbB3	CRL2 (VHL)	A heptapeptide derived from HIF1α	Loss of viability of breast cancer cells [223]
Smad3-PROTAC	Smad3	SMC	CRL2 (VHL)	A pentapeptide derived from HIF1α	Potential roles against the development of renal fibrosis [226]
TH006	Tau	Sequences derived from β-tubulin	CRL2 (VHL)	A heptapeptide derived from HIF1α	Reduced proteotoxic effects among AD mice [224]

Abbreviations: NS: not specific; AHR: aryl hydrocarbon receptor; PCC: protein-catalyzed capture agent; DHT: dihydroxytestosterone; FRS2α: fibroblast growth factor receptor substrate 2 α; TrkA: tropomyosin receptor kinase A; PI3K: phosphatidylinositol-3-kinase; ErbB3: erythroblastosis oncogene B3; MetAP-2: methionine aminopeptidase-2; SMC: small molecule compound; ERK: extracellular regulated protein kinase; ODD: oxygen-dependent degradation; E2-SMPI: Estradiol-Small molecule proteolysis inducer; SNIPER: specific and nongenetic IAPs-dependent protein erasers; AR: androgen receptor; ER: estrogen receptor; AD: Alzheimer's Disease.

4.2. Small molecule PROTAC

Due to its limited molecule weight and non-ionic features, small molecule PROTAC is thought to provide greater bioavailability and efficacy. CRL2 (VHL) and CRL4 (CRBN) are two key E3 ligases for small molecule PROTACs. Currently, owing to the incurability of malignant diseases, all evidence regarding VHL-based PROTACs is tumor-associated, which serve as potential anti-cancer interventions (Table 10). Similarly, the pathological role of CRBN-based PROTACs is also centered on neoplastic relevant behaviors (Table 10). Besides this direct evidence, the biological functions of the remaining small molecule PROTACs still require in vivo verification.

In addition, other PROTAC members including cIAP1-based, APC/C (CDH1)-based and MDM2-based PROTACs also emerge as protein degraders for different targets, displaying potential therapeutic effects against RAR (retinoic acid receptor)-positive [216], TACC3 (transforming acidic coiled coil-containing protein 3)-positive [229] and AR-positive [214] disorders respectively. Moreover, apart from these traditional small molecule PROTACs, two types of novel PROTACs have been reported. The first type, HaloTag7-fused PROTACs (HT7-PROTACs), where unlike other PROTACs the E3 ligases (β-TRCP) is engineered and fused to an HT7 tag. And then a chloroalkane ligand in the PROTAC is applied to attach the E3 ligase and leads to substrate degradation [230]. This suggests that if it is difficult to design a small molecule ligand for certain E3 ligases, engineered modifications of E3 ligases may be an alternative approach for cell and molecular -based studies. Secondly, a homo-PROTAC has also been designed, which is a bivalent small-molecule dimerizer of VHL E3 ligase that induces its self-degradation [231]. This likewise provides therapeutic implications against those disorders with an elevated expression of undruggable E3 ligases (Table 10).

4.3. PROTAC: Anti-tumor functionality by degrading oncogenic proteins

Most of the PROTACs have thus far been designed with the purpose of treating cancers via degradation of specific oncogenic proteins, such as BRD4. Currently, investigations have confirmed that BRD4 acts as an oncoprotein in multiple malignancies such as prostate and hematological cancers, primarily by interacting with histones in order to trigger transcription of oncogenes [232]. Nevertheless, due to toxicities and progressive inefficacies of BRD4 inhibitors including BET, more specific and tolerable targeted medications are required, such as BRD4-targeted PROTACs. So far, there are six PROTACs reported to effectively target and destabilize BRD4, either VHL-based or CRBN-based. Among them, four PROTACs have shown anti-tumor efficacy, including ARV-771, compound 23, dBET1 and ARV-825, which display great inhibitory effects against the malignant progression of castration-resistant prostate cancer (CRPC) [233], xenograft leukemic models [234], leukemic cells [211] and Burkitt's lymphoma [210] respectively. All these PROTACs, especially ARV-771 and ARV-825, are waiting for verification in animal studies before being assessed in the clinic. Furthermore, despite a lack of direct evidence, the other two PROTACs including compound MZ1 [235,236] and CLIPTAC-1 [236,237] also target BRD4, which are believed to have potential contributions against BRD4-positive neoplasms.

In addition to BRD4, other PROTACs have also been regarded as anti-tumor PROTACs. As the only peptide PROTAC, phosphoPROTAC-2 directly degrades PI3K via recruiting CRL2 (VHL), culminating in reduced viability of breast cancer cells [223]. BCR-ABL, is an oncogenic fusion protein for chronic myelogenous leukemia, can be targeted by TKI-PROTAC-1 which has shown in vitro effects on survival of chronic myelogenous leukemic cells [238]. CRABP I/II are cellular binding proteins of retinoic acid and mediate the biologic impact of retinoic acid under diverse circumstances. The oncogenic role of CRABP I/II in the pathogenesis of neuroblastoma has been widely shown, and correspondingly, compound 4, a CRABP I/II-targeted PROTAC, has also been shown to have a tumor suppressive role against the migration of

Table 10
Major small molecule PROTACs and their biological impacts.

Name	Target	Target ligand	E3 ligase	E3 ligase ligand	Biological impacts
SNIPER-1	AR	DHT	cIAP1	BE04	Potential roles against AR positive disorders [216]
SARM–nutlin PROTAC	AR	SARM	MDM2	Nutlin	Potential roles against AR positive disorders [214]
TKI-PROTAC-1	BCR-ABL	TKI moiety (bosutinib and dasatinib)	CRL4 (CRB)	Pomalidomide	Reduced viability of chronic myelogenous leukemic cells [238]
Compound MZ1	BRD4	JQ1	CRL2 (VHL)	VHL binders	Suppression on oncogenic transcriptions associated with BRD4 [235]
ARV-771	BRD4	OTX015	CRL2 (VHL)	HIF1α-derived and hydroxyproline-contained ligand	Tumor suppression in CRPC mouse models [233]; Induced apoptosis in MCL cells [334]
Compound 23	BRD4	Azacarbazole	CRL4 (CRB)	Thalidomide	Growth suppression against xenograft leukemic tumors [234]
dBET1	BRD4	JQ1	CRL4 (CRB)	Phthalimide	Delayed leukemia progression in mice [211]
CLIPTAC-1	BRD4	TCO-tagged JQ1	CRL4 (CRB)	Thalidomide	Potential roles against BRD4 positive disorders [237]
ARV-825	BRD4	OTX015	CRL4 (CRB)	Pomalidomide	Suppressed proliferation and enhanced apoptosis in Burkitt's lymphoma [210]; Induced apoptosis in MCL cells [334]
TKI-PROTAC-2	c-ABL	TKI moiety (dasatinib)	CRL2 (VHL)	HIF1α-derived and hydroxyproline-contained ligand	Potential roles against c-ABL positive disorders [238]
TKI-PROTAC-3	c-ABL	TKI moiety (bosutinib and dasatinib)	CRL4 (CRB)	Pomalidomide	Potential roles against c-ABL positive disorders [238]
CDK9-PROTAC	CDK9	Aminopyrazole analog	CRL4 (CRB)	Thalidomide	Potential roles against CDK9 positive disorders [213]
Compound 4	CRABP I/II	ATRA	cIAP1	MeBS	Inhibited cell migration of neuroblastoma cells [218,219]
brequinar-PROTAC	DHODH	Brequinar	CRL2 (VHL)	HIF1α-derived and hydroxyproline-contained ligand	Cytotoxicity against cancer cells [239]
SNIPER-2	ER	Estrone	cIAP1	BE04	Potential roles against ER positive disorders [216,217]
CLIPTAC-2	ERK1/2	Probe 1	CRL4 (CRB)	Thalidomide	Potential roles against ERK1/2 positive disorders [237]
PROTAC_ ERRα	ERRα	Thiazolidinedione-based ligand	CRL2 (VHL)	HIF1α-derived and hydroxyproline-contained ligand	Potential roles against ERRα positive disorders [203]
HT7-PROTAC-1	FKBP12	NS	β-TRCP-HT7	Chloroalkane	Potential roles against FKBP12 positive disorders [230]
dFKBP	FKBP12	Steel factor	CRL4 (CRB)	Phthalimide	Potential roles against FKBP12 positive disorders [211]
HT7-PROTAC-2	FKBP12	NS	Parkin-HT7	Chloroalkane	Potential roles against FKBP12 positive disorders [230]
SNIPER-3	RAR	Ch55	cIAP1	BE04	Potential roles against RAR positive disorders [216]
PROTAC_ RIPK2	RIPK2	Vandetanib	CRL2 (VHL)	HIF1α-derived and hydroxyproline-contained ligand	Potential roles against RIPK2 positive disorders [203]
SirReal-PROTAC	Sirtuin 2	SirReal	CRL4 (CRB)	Thalidomide	Enhanced acetylation of the microtubule network [212]
SNIPER (TACC3)	TACC3	KHS101	APC/C (CDH1)	Bestatin	Cytotoxic effects against cancer cells [229]
PROTAC 3i	TBK1	Aminopyrimidine chemotype	CRL2 (VHL)	HIF1α-derived and hydroxyproline-contained ligand	Potential roles against TBK1 positive disorders [335]
Homo-PROTAC	VHL	VH032	CRL2 (VHL)	VH298	Self-degradation of E3 ligases [231]

Abbreviations: NS: not specific; ODD: oxygen-dependent degradation; E2-SMPI: Estradiol-Small molecule proteolysis inducer; SNIPER: specific and nongenetic IAPs-dependent protein erasers; AHR: aryl hydrocarbon receptor; AR: androgen receptor; ER: estrogen receptor; FKBP12: the 12-kDa FK506-binding protein; RAR: retinoic acid receptor; cIAP1: cellular inhibitor of apoptosis protein 1; SARM: non-steroidal androgen receptor ligand; MDM2: mouse double minute 2 homolog; BCR-ABL: TKI: tyrosine kinase inhibitor; BCR-ABL: breakpoint cluster region-abelson murine leukemia viral oncogene; BRD4: JQ1: a BET bromodomain inhibitor; BE04: a bestatin derivative that has affinity to cIAP1; CRPC: castration-resistant prostate cancer; CRABP: cellular retinoic acid-binding protein; ATRA: all-trans retinoic acid; MeBS: methyl bestatin; ERRα: estrogen-related receptor α; RIPK2: receptor interacting serine/threonine kinase 2; TACC3: transforming acidic coiled coil-containing protein 3; DHODH: dihydroorotate dehydrogenase; TBK1: TANK-binding kinase 1; MCL: mantle cell lymphoma; CDK9: cyclin-dependent kinase 9; SirReal: sirtuin rearranging ligand; SMC: small molecule compound; ERK: extracellular regulated protein kinase.

Table 11
Major anti-tumor PROTACs and their contributions.

Name	Category	Target	E3 ligase	Evidence[a]
phosphoPROTAC-2	Peptide	PI3K	CRL2 (VHL)	Direct roles against the viability of breast cancer cells [223]
TKI-PROTAC-1	Small molecule	BCR-ABL	CRL4 (CRB)	Direct roles against the viability of chronic myelogenous leukemic cells [238]
ARV-771	Small molecule	BRD4	CRL2 (VHL)	Direct roles against the tumor growth in CRPC mouse models [233] and MCL cells [334]
Compound 23	Small molecule	BRD4	CRL4 (CRB)	Direct roles against the growth of xenograft leukemic tumors [234]
dBET1	Small molecule	BRD4	CRL4 (CRB)	Direct roles against the progression of leukemia [211]
ARV-825	Small molecule	BRD4	CRL4 (CRB)	Direct roles against the proliferation of Burkitt's lymphoma [210] and MCL cells [334]
Compound 4	Small molecule	CRABP I/II	cIAP1	Direct roles against the migration of neuroblastoma cells [218,219]
brequinar-PROTAC	Small molecule	DHODH	CRL2 (VHL)	Direct roles against the viability of colon cancer cells [239]
SNIPER (TACC3)	Small molecule	TACC3	APC/C (CDH1)	Direct roles against the viability of breast cancer cells [229]
Api-PROTAC	Peptide	AHR	CRL2 (VHL)	Potential roles against multiple AHR positive cancers [332,333,336]
Tri_a-PROTAC	Peptide	Akt	CRL2 (VHL)	Potential roles against multiple Akt positive cancers [227,337]
PROTAC-3	Peptide	AR	CRL1 (β-TRCP)	Potential roles against multiple AR positive cancers [228,240]
PROTAC-5	Peptide	AR	CRL2 (VHL)	Potential roles against multiple AR positive cancers [202,240]
PROTAC-2	Peptide	ER	CRL1 (β-TRCP)	Potential roles against multiple ER positive cancers [228,242]
E2-SMPI	Peptide	ER	CRL2 (VHL)	Potential roles against multiple ER positive cancers [220–222,242]
phosphoPROTAC-1	Peptide	FRS2α	CRL2 (VHL)	Potential roles against multiple FRS2α positive cancers [223,338]
HBx-PROTAC	Peptide	HBx	CRL2 (VHL)	Potential roles against the development of liver cancer [225]
Fu-SMPI	Peptide	MetAP-2	CRL2 (VHL)	Potential roles against multiple MetAP-2 positive cancers [220,339]
SNIPER-1	Small molecule	AR	cIAP1	Potential roles against multiple AR positive cancers [216,240]
SARM–nutlin PROTAC	Small molecule	AR	MDM2	Potential roles against multiple AR positive cancers [214,240]
Compound MZ1	Small molecule	BRD4	CRL2 (VHL)	Potential roles against multiple BRD4 positive cancers [235,236]
CLIPTAC-1	Small molecule	BRD4	CRL4 (CRB)	Potential roles against multiple BRD4 positive cancers [236,237]
TKI-PROTAC-2	Small molecule	c-ABL	CRL2 (VHL)	Potential roles against multiple c-ABL positive cancers [238,340]
TKI-PROTAC-3	Small molecule	c-ABL	CRL4 (CRB)	Potential roles against multiple c-ABL positive cancers [238,340]
CDK9-PROTAC	Small molecule	CDK9	CRL4 (CRB)	Potential roles against multiple CDK9 positive cancers [213,341]
SNIPER-2	Small molecule	ER	cIAP1	Potential roles against multiple ER positive cancers [216,217,243]
CLIPTAC-2	Small molecule	ERK1/2	CRL4 (CRB)	Potential roles against multiple ERK1/2 positive cancers [237,342]
PROTAC_ ERRα	Small molecule	ERRα	CRL2 (VHL)	Potential roles against multiple ERRα positive cancers [203,343]
SNIPER-3	Small molecule	RAR	cIAP1	Potential roles against multiple RAR positive cancers [216,344]
PROTAC_ RIPK2	Small molecule	RIPK2	CRL2 (VHL)	Potential roles against multiple RIPK2 positive cancers [203,345]
SirReal-PROTAC	Small molecule	Sirtuin 2	CRL4 (CRB)	Potential roles against multiple Sirtuin 2 positive cancers [212,346]
PROTAC 3i	Small molecule	TBK1	CRL2 (VHL)	Potential roles against multiple TBK1 positive cancers [335,347]

[a] If there are direct descriptions or results demonstrating the anti-tumor efficacy of specific PROTAC, then these kinds of evidences are defined as “Direct”, while if there are no direct descriptions, the anti-tumor effects can only be predicted from the role of substrates, then these kinds of evidences are defined as “Potential”.

neuroblastoma cells [218,219]. Moreover, both brequinar-PROTAC and SNIPER (TACC3) are characterized by their tumor inhibitory effect among colon and breast cancer cells, respectively, via targeting DHODH (dihydroorotate dehydrogenase) [239] and TACC3 (transforming acidic coiled-coil-containing protein 3), respectively [229]. These results have shown the therapeutic potential of PROTACs as a novel class of anti-tumor therapeutics.

Apart from those with direct evidence, the majority of current PROTACs are assumed to potentially play anti-neoplastic roles, which have not been shown yet in vivo despite their pro-degrading effects on certain oncogenic substrates in cells. Among those, AR and ER are the most popular targets for PROTACs. At present, there have been four PROTACs, either peptide or small molecule, believed to successfully and specifically destabilize AR in vitro and in vivo, including PROTAC-3 [228,240], PROTAC-5 [202,240], SNIPER-1 [216,240] and SARM-nutlin PROTAC [214,240]. The AR-targeted PROTACs are considered as potential therapeutic agents given the extensive oncogenic role of AR in a variety of cancers, especially prostate cancer. Moreover, ER, a notorious oncoprotein in gynecological malignancies [241], has been targeted by three PROTACs, namely PROTAC-2 [228,242], E2-SMPI [220–222,242] and SNIPER-2 [216,217,243], all demonstrating degrading effects of the ER protein despite limited studies showing suppression of tumorigenesis.

Taken together, although currently, no PROTAC has entered the clinic, the anti-tumor PROTACs are still expected to become important supplements to current targeted therapies. Compared to the direct evidence regarding the anti-tumor effects of small molecule PROTACs, only one peptide PROTAC has reported a direct effect on cancer cell viability, which highlights the advantage and therapeutic potential of small molecules PROTACs. Nevertheless, there are issues with the current status of PROTACs that should be addressed in future investigations. First, PROTAC related studies have lacked comparisons with standard of care agents, which may lower their credibility as an alternative to currently approved options. Second, although there has been direct evidence towards the anti-tumor impact of certain PROTACs, in vivo efficacy in animals has been limited, let alone clinical assessment, therefore more studies are necessary to demonstrate the clinical benefits of this promising technology (Fig. 7 and Table 11).

4.4. Perspective and therapeutic implications

As we have described, many substrate receptors demonstrate tumor-inhibitory effects by degrading oncogenic substrates, serving as potential therapeutic targets for cancer treatment. Currently, there are two major types of medications that specially aim at the interactions between substrates and recognition receptors in order to exert anti-cancer impacts, namely sulfonamides and thalidomide derivatives, each characterized by specific mechanisms.

Three members of anticancer sulfonamides E7820, CQS (chloroquinoxaline sulfonamide) and indisulam have displayed efficacy against various cancers, including colorectal cancer and melanoma [244–246]. However, their mechanisms of action remain poorly defined. Investigations have found that their anti-cancer contributions in colorectal cancer cells may rely on the interaction between CRL4 (DCAF15) and the substrate CAPERα, promoting its degradation [173,247]. Knockout of *DCAF15* or overexpression of CAPERα confers resistance to the effects by sulfonamides, further proving DCAF15 as the mechanistic target of sulfonamides [247]. So far, all these drugs have undergone clinical trials and are waiting for approval by the FDA for clinical application against colorectal cancer.

Thalidomide and its derivatives, known as immunomodulatory drugs (IMiDs), have drawn much attention owing to their great anti-tumor effectiveness against hematological malignancies, especially multiple myeloma [248]. Current investigations have confirmed that

IMiDs facilitates interaction between CRBN (cereblon) and its substrates. By binding to CRBN, IMiDs, including thalidomide, pomalidomide and lenalidomide, can trigger the degradation of substrates such as IKZF1/3 [248] and CK1α [164], thus halting tumor expansion of multiple myeloma and myelodysplastic syndrome. All these IMiDs have gained FDA approval against multiple myeloma. Besides IMiDs, there is another type of CRBN modulator CC-885 which promotes the interaction between CRBN and GSPT1 to trigger the degradation of GSPT1, showing tumor suppressive effects against leukemic cells [163]. In addition, DCAF2 seems to function as a therapeutic target since its interaction with substrates SET8 and p21 could be specifically inhibited by Pevonedistat [150] (Table 8).

As a lesser understood member of the Cullin-RING ligase family, CRL4 has drawn much attention due to its extensive involvement in physiological and pathological conditions, especially in tumorigenesis. Based on the evidence described above from knockout and transgenic mouse models, human clinical data, and biochemical interactions, the scaffold protein CUL4A/B and RING finger protein RBX1 are believed to serve as oncoproteins in a variety of malignancies. Nevertheless, the adaptor DDB1 and substrate recognition receptors seem to play context dependent roles in tumorigenesis, adding diversity to the actual role of CRL4 in tumorigenesis.

Moreover, owing to its critical role in cancer regulation, many CRL4-targeted medications have been identified, including thalidomide and its derivatives that target CRBN and the sulfonamides that target DCAF15, which significantly suppress proliferation of multiple myeloma and colorectal cancer, respectively. PROTACs are a group of artificially assembled protein degraders that induce the ubiquitination-mediated degradation of substrates via recruiting endogenous E3 ligases, such as CRL4 (CRBN) and CRL2 (VHL). Due to their ability to increase the number of druggable targets and potential for high specificity, more than 30 PROTACs have been designed and display anti-tumor effects, especially BRD4-targeted PROTACs. Current and future studies in animal tumor models and clinical trials would lead to mechanistic and functional understanding of PROTACs as drug candidates, and set the stage for their clinical usage in cancer patients.

Author information

The authors declare no competing interests. Correspondence and requests for materials should be addressed to W.W. (wwei2@bidmc.harvard.edu) or P.Z. (pez2001@med.cornell.edu).

Acknowledgements

The authors sincerely apologize to all those colleagues whose important work was not cited in this paper owing to space limitations. They thank the members of Wei laboratory for critical reading and discussion of the manuscript. W.W. is a Leukemia & Lymphoma Society (LLS) research scholar. This work was supported in part by Scientific Research Training Program for Young Talents (Union Hospital, Tongji Medical College, Huazhong University of Science and Technology) to J.C., by National Natural Science Foundation of China (81572413) to K.T., by the V Foundation for Cancer Research to P.Z., and by US National Institutes of Health (NIH) grants to P. Z. (CA159925 and CA213992) and W.W. (GM094777 and CA177910).

References

[1] E. Leithe, Regulation of connexins by the ubiquitin system: Implications for intercellular communication and cancer, Biochim. Biophys. Acta 1865 (2016) 133–146, https://doi.org/10.1016/j.bbcan.2016.02.001.

[2] A. Varshavsky, The ubiquitin system, autophagy, and regulated protein degradation, Annu. Rev. Biochem. 86 (2017) 123–128, https://doi.org/10.1146/annurev-biochem-061516-044859.

[3] J. Cheng, et al., Functional analysis of Cullin 3 E3 ligases in tumorigenesis, Biochim. Biophys. Acta 1869 (2017) 11–28, https://doi.org/10.1016/j.bbcan.2017.11.001.

[4] J. Guo, J. Cheng, B.J. North, W. Wei, Functional analyses of major cancer-related signaling pathways in Alzheimer's disease etiology, Biochim. Biophys. Acta 1868 (2017) 341–358, https://doi.org/10.1016/j.bbcan.2017.07.001.

[5] A. Ciechanover, The unravelling of the ubiquitin system, Nat. Rev. Mol. Cell. Biol. 16 (2015) 322–324, https://doi.org/10.1038/nrm3982.

[6] I. Dikic, Proteasomal and autophagic degradation systems, Annu. Rev. Biochem. 86 (2017) 193–224, https://doi.org/10.1146/annurev-biochem-061516-044908.

[7] S. Gilberto, M. Peter, Dynamic ubiquitin signaling in cell cycle regulation, J. Cell Biol. 216 (2017) 2259–2271, https://doi.org/10.1083/jcb.201703170.

[8] J.J.S. VerPlank, A.L. Goldberg, Regulating protein breakdown through proteasome phosphorylation, Biochem. J. 474 (2017) 3355–3371, https://doi.org/10.1042/BCJ20160809.

[9] N. Vigneron, J. Abi Habib, B.J. Van den Eynde, Learning from the proteasome: how to fine-tune cancer immunotherapy, Trends Cancer 3 (2017) 726–741, https://doi.org/10.1016/j.trecan.2017.07.007.

[10] S. Lazaro, D. Gamarra, M. Del Val, Proteolytic enzymes involved in MHC class I antigen processing: a guerrilla army that partners with the proteasome, Mol. Immunol. 68 (2015) 72–76, https://doi.org/10.1016/j.molimm.2015.04.014.

[11] A. Fierabracci, Proteasome inhibitors: a new perspective for treating autoimmune diseases, Curr. Drug Targets 13 (2012) 1665–1675.

[12] J.M. Deger, J.E. Gerson, R. Kayed, The interrelationship of proteasome impairment and oligomeric intermediates in neurodegeneration, Aging Cell 14 (2015) 715–724, https://doi.org/10.1111/acel.12359.

[13] Q. Zheng, et al., Dysregulation of ubiquitin-proteasome system in neurodegenerative diseases, Front. Aging Neurosci. 8 (2016) 303, https://doi.org/10.3389/fnagi.2016.00303.

[14] F. Coutant, P. Miossec, Altered dendritic cell functions in autoimmune diseases: distinct and overlapping profiles, Nat. Rev. Rheumatol. 12 (2016) 703–715, https://doi.org/10.1038/nrrheum.2016.147.

[15] A. Magnacca, et al., Characterization of a proteasome and TAP-independent presentation of intracellular epitopes by HLA-B27 molecules, J. Biol. Chem. 287 (2012) 30358–30367, https://doi.org/10.1074/jbc.M112.384339.

[16] G. Kaur, S. Batra, Emerging role of immunoproteasomes in pathophysiology, Immunol. Cell Biol. 94 (2016) 812–820, https://doi.org/10.1038/icb.2016.50.

[17] S.K. Kwon, M. Saindane, K.H. Baek, p53 stability is regulated by diverse deubiquitinating enzymes, Biochim. Biophys. Acta 1868 (2017) 404–411, https://doi.org/10.1016/j.bbcan.2017.08.001.

[18] L.N. Micel, J.J. Tentler, P.G. Smith, G.S. Eckhardt, Role of ubiquitin ligases and the proteasome in oncogenesis: novel targets for anticancer therapies, J. Clin. Oncol. 31 (2013) 1231–1238, https://doi.org/10.1200/JCO.2012.44.0958.

[19] K.I. Nakayama, K. Nakayama, Ubiquitin ligases: cell-cycle control and cancer, Nat. Rev. Cancer 6 (2006) 369–381, https://doi.org/10.1038/nrc1881.

[20] N. Zheng, Q. Zhou, Z. Wang, W. Wei, Recent advances in SCF ubiquitin ligase complex: Clinical implications, Biochim. Biophys. Acta 1866 (2016) 12–22, https://doi.org/10.1016/j.bbcan.2016.05.001.

[21] M. Wade, et al., Functional analysis and consequences of Mdm2 E3 ligase inhibition in human tumor cells, Oncogene 31 (2012) 4789–4797, https://doi.org/10.1038/onc.2011.625.

[22] R. Honda, H. Tanaka, H. Yasuda, Oncoprotein MDM2 is a ubiquitin ligase E3 for tumor suppressor p53, FEBS Lett. 420 (1997) 25–27.

[23] F. Ding, H. Xiao, M. Wang, X. Xie, F. Hu, The role of the ubiquitin-proteasome pathway in cancer development and treatment, Front. Biosci. (Landmark Ed) 19 (886–895) (2014).

[24] A.C. Groner, et al., TRIM24 is an oncogenic transcriptional activator in prostate cancer, Cancer Cell 29 (2016) 846–858, https://doi.org/10.1016/j.ccell.2016.04.012.

[25] C. Geng, et al., Androgen receptor is the key transcriptional mediator of the tumor suppressor SPOP in prostate cancer, Cancer Res. 74 (2014) 5631–5643, https://doi.org/10.1158/0008-5472.CAN-14-0476.

[26] Q.P. Dou, J.A. Zonder, Overview of proteasome inhibitor-based anti-cancer therapies: perspective on bortezomib and second generation proteasome inhibitors versus future generation inhibitors of ubiquitin-proteasome system, Curr. Cancer Drug Targets 14 (2014) 517–536.

[27] Y. Matsuo, et al., Proteasome inhibitor MG132 inhibits angiogenesis in pancreatic cancer by blocking NF-kappaB activity, Dig. Dis. Sci. 55 (2010) 1167–1176, https://doi.org/10.1007/s10620-009-0814-4.

[28] Y. Kravtsova-Ivantsiv, et al., KPC1-mediated ubiquitination and proteasomal processing of NF-kappaB1 p105 to p50 restricts tumor growth, Cell 161 (2015) 333–347, https://doi.org/10.1016/j.cell.2015.03.001.

[29] S.A.H. de Poot, G. Tian, D. Finley, Meddling with fate: The proteasomal deubiquitinating enzymes, J. Mol. Biol. (2017), https://doi.org/10.1016/j.jmb.2017.09.015.

[30] T.E.T. Mevissen, D. Komander, Mechanisms of deubiquitinase specificity and regulation, Annu. Rev. Biochem. 86 (2017) 159–192, https://doi.org/10.1146/annurev-biochem-061516-044916.

[31] M. He, et al., The emerging role of deubiquitinating enzymes in genomic integrity, diseases, and therapeutics, Cell Biosci. 6 (2016) 62, https://doi.org/10.1186/s13578-016-0127-1.

[32] Z.M. Eletr, K.D. Wilkinson, Regulation of proteolysis by human deubiquitinating enzymes, Biochim. Biophys. Acta 1843 (2014) 114–128, https://doi.org/10.1016/j.bbamcr.2013.06.027.

[33] S.A. Abdul Rehman, et al., MINDY-1 is a member of an evolutionarily conserved and structurally distinct new family of deubiquitinating enzymes, Mol. Cell 63 (2016) 146–155, https://doi.org/10.1016/j.molcel.2016.05.009.

[34] D. Kwasna, et al., Discovery and characterization of ZUFSP/ZUP1, a distinct deubiquitinase class important for genome stability, Mol. Cell 70 (2018) 150–164, https://doi.org/10.1016/j.molcel.2018.02.023.

[35] P. Haahr, et al., ZUFSP deubiquitylates k63-linked polyubiquitin chains to

promote genome stability, Mol. Cell 70 (2018) 165–174, https://doi.org/10.1016/j.molcel.2018.02.024.
[36] N. Kumari, et al., The roles of ubiquitin modifying enzymes in neoplastic disease, Biochim. Biophys. Acta 1868 (2017) 456–483, https://doi.org/10.1016/j.bbcan.2017.09.002.
[37] A. Toloczko, et al., Deubiquitinating enzyme USP9X suppresses tumor growth via LATS kinase and core components of the Hippo pathway, Cancer Res. 77 (2017) 4921–4933, https://doi.org/10.1158/0008-5472.CAN-16-3413.
[38] E. Graner, et al., The isopeptidase USP2a regulates the stability of fatty acid synthase in prostate cancer, Cancer Cell 5 (2004) 253–261.
[39] V. Fajner, E. Maspero, S. Polo, Targeting HECT-type E3 ligases - insights from catalysis, regulation and inhibitors, FEBS Lett. 591 (2017) 2636–2647, https://doi.org/10.1002/1873-3468.12775.
[40] N. Zheng, N. Shabek, Ubiquitin ligases: structure, function, and regulation, Annu. Rev. Biochem. 86 (2017) 129–157, https://doi.org/10.1146/annurev-biochem-060815-014922.
[41] L. Liu, C.C. Wong, B. Gong, J. Yu, Functional significance and therapeutic implication of ring-type E3 ligases in colorectal cancer, Oncogene (2017), https://doi.org/10.1038/onc.2017.313.
[42] C. Natarajan, K. Takeda, Regulation of various DNA repair pathways by E3 ubiquitin ligases, J. Cancer Res. Ther. 13 (2017) 157–169, https://doi.org/10.4103/0973-1482.204879.
[43] E. Bulatov, A. Ciulli, Targeting Cullin-RING E3 ubiquitin ligases for drug discovery: structure, assembly and small-molecule modulation, Biochem. J. 467 (2015) 365–386, https://doi.org/10.1042/BJ20141450.
[44] J.R. Lydeard, B.A. Schulman, J.W. Harper, Building and remodelling Cullin-RING E3 ubiquitin ligases, EMBO Rep. 14 (2013) 1050–1061, https://doi.org/10.1038/embor.2013.173.
[45] D. Cui, X. Xiong, Y. Zhao, Cullin-RING ligases in regulation of autophagy, Cell Div 11 (2016) 8, https://doi.org/10.1186/s13008-016-0022-5.
[46] Z. Chen, J. Sui, F. Zhang, C. Zhang, Cullin family proteins and tumorigenesis: genetic association and molecular mechanisms, J. Cancer 6 (2015) 233–242, https://doi.org/10.7150/jca.11076.
[47] Y. Zhao, Y. Sun, Cullin-RING Ligases as attractive anti-cancer targets, Curr. Pharm. Des. 19 (2013) 3215–3225.
[48] S. Jackson, Y. Xiong, CRL4s: the CUL4-RING E3 ubiquitin ligases, Trends Biochem. Sci. 34 (2009) 562–570, https://doi.org/10.1016/j.tibs.2009.07.002.
[49] Y. Zhang, et al., CUL-4A stimulates ubiquitylation and degradation of the HOXA9 homeodomain protein, EMBO J. 22 (2003) 6057–6067, https://doi.org/10.1093/emboj/cdg577.
[50] B. Li, N. Jia, R. Kapur, K.T. Chun, Cul4A targets p27 for degradation and regulates proliferation, cell cycle exit, and differentiation during erythropoiesis, Blood 107 (2006) 4291–4299, https://doi.org/10.1182/blood-2005-08-3349.
[51] J. Hannah, P. Zhou, Distinct and overlapping functions of the cullin E3 ligase scaffolding proteins CUL4A and CUL4B, Gene 573 (2015) 33–45, https://doi.org/10.1016/j.gene.2015.08.064.
[52] Y. Sang, F. Yan, X. Ren, The role and mechanism of CRL4 E3 ubiquitin ligase in cancer and its potential therapy implications, Oncotarget 6 (2015) 42590–42602, https://doi.org/10.18632/oncotarget.6052.
[53] C.Y. Chen, et al., Rescue of the genetically engineered Cul4b mutant mouse as a potential model for human X-linked mental retardation, Hum. Mol. Genet. 21 (2012) 4270–4285, https://doi.org/10.1093/hmg/dds261.
[54] L. Liu, et al., Essential role of the CUL4B ubiquitin ligase in extra-embryonic tissue development during mouse embryogenesis, Cell Res. 22 (2012) 1258–1269, https://doi.org/10.1038/cr.2012.48.
[55] L. Liu, et al., CUL4A abrogation augments DNA damage response and protection against skin carcinogenesis, Mol. Cell 34 (2009) 451–460, https://doi.org/10.1016/j.molcel.2009.04.020.
[56] Y. Cang, et al., DDB1 is essential for genomic stability in developing epidermis, Proc. Natl. Acad. Sci. U. S. A. 104 (2007) 2733–2737, https://doi.org/10.1073/pnas.0611311104.
[57] Y. Cang, et al., Deletion of DDB1 in mouse brain and lens leads to p53-dependent elimination of proliferating cells, Cell 127 (2006) 929–940, https://doi.org/10.1016/j.cell.2006.09.045.
[58] B. Li, J.C. Ruiz, K.T. Chun, CUL-4A is critical for early embryonic development, Mol. Cell. Biol. 22 (2002) 4997–5005.
[59] D. Kopanja, et al., Proliferation defects and genome instability in cells lacking Cul4A, Oncogene 28 (2009) 2456–2465, https://doi.org/10.1038/onc.2009.86.
[60] J. Lee, et al., Improved ex vivo expansion of adult hematopoietic stem cells by overcoming CUL4-mediated degradation of HOXB4, Blood 121 (2013) 4082–4089, https://doi.org/10.1182/blood-2012-09-455204.
[61] G. Sauvageau, et al., Differential expression of homeobox genes in functionally distinct CD34+ subpopulations of human bone marrow cells, Proc. Natl. Acad. Sci. U. S. A. 91 (1994) 12223–12227.
[62] A. Giampaolo, et al., HOXB gene expression and function in differentiating purified hematopoietic progenitors, Stem Cells 13 (Suppl. 1) (1995) 90–105.
[63] N. Pineault, C.D. Helgason, H.J. Lawrence, R.K. Humphries, Differential expression of Hox, Meis1, and Pbx1 genes in primitive cells throughout murine hematopoietic ontogeny, Exp. Hematol. 30 (2002) 49–57.
[64] Y. Yin, et al., The E3 ubiquitin ligase Cullin 4A regulates meiotic progression in mouse spermatogenesis, Dev. Biol. 356 (2011) 51–62, https://doi.org/10.1016/j.ydbio.2011.05.661.
[65] Y. Yin, et al., Cell autonomous and nonautonomous function of CUL4B in mouse spermatogenesis, J. Biol. Chem. 291 (2016) 6923–6935, https://doi.org/10.1074/jbc.M115.699660.
[66] D. Kopanja, et al., Cul4A is essential for spermatogenesis and male fertility, Dev. Biol. 352 (2011) 278–287, https://doi.org/10.1016/j.ydbio.2011.01.028.
[67] C. Yu, S.Y. Ji, Q.Q. Sha, Q.Y. Sun, H.Y. Fan, CRL4-DCAF1 ubiquitin E3 ligase directs protein phosphatase 2A degradation to control oocyte meiotic maturation, Nat. Commun. 6 (2015) 8017, https://doi.org/10.1038/ncomms9017.
[68] Y.W. Xu, et al., Maternal DCAF2 is crucial for maintenance of genome stability during the first cell cycle in mice, J. Cell Sci. 130 (2017) 3297–3307, https://doi.org/10.1242/jcs.206664.
[69] C. Yu, et al., CRL4 complex regulates mammalian oocyte survival and reprogramming by activation of TET proteins, Science 342 (2013) 1518–1521, https://doi.org/10.1126/science.1244587.
[70] C. Yu, Y.W. Xu, Q.Q. Sha, H.Y. Fan, CRL4DCAF1 is required in activated oocytes for follicle maintenance and ovulation, Mol. Hum. Reprod. 21 (2015) 195–205, https://doi.org/10.1093/molehr/gau103.
[71] J. Liu, et al., CRL4A(CRBN) E3 ubiquitin ligase restricts BK channel activity and prevents epileptogenesis, Nat. Commun. 5 (2014) 3924, https://doi.org/10.1038/ncomms4924.
[72] X. Tong, et al., DDB1-mediated CRY1 degradation promotes FOXO1-driven gluconeogenesis in liver, Diabetes 66 (2017) 2571–2582, https://doi.org/10.2337/db16-1600.
[73] S. Yamaji, et al., Hepatocyte-specific deletion of DDB1 induces liver regeneration and tumorigenesis, Proc. Natl. Acad. Sci. U. S. A. 107 (2010) 22237–22242, https://doi.org/10.1073/pnas.1015793108.
[74] P. Li, et al., Lack of CUL4B in adipocytes promotes PPARgamma-mediated adipose tissue expansion and insulin sensitivity, Diabetes 66 (2017) 300–313, https://doi.org/10.2337/db16-0743.
[75] Q. Li, et al., A cullin 4B-RING E3 ligase complex fine-tunes pancreatic delta cell paracrine interactions, J. Clin. Invest. 127 (2017) 2631–2646, https://doi.org/10.1172/JCI91348.
[76] T. Yoon, et al., Tumor-prone phenotype of the DDB2-deficient mice, Oncogene 24 (2005) 469–478, https://doi.org/10.1038/sj.onc.1208211.
[77] Y. Zou, et al., Mutation in CUL4B, which encodes a member of cullin-RING ubiquitin ligase complex, causes X-linked mental retardation, Am. J. Hum. Genet. 80 (2007) 561–566, https://doi.org/10.1086/512489.
[78] P.S. Tarpey, et al., Mutations in CUL4B, which encodes a ubiquitin E3 ligase subunit, cause an X-linked mental retardation syndrome associated with aggressive outbursts, seizures, relative macrocephaly, central obesity, hypogonadism, pes cavus, and tremor, Am. J. Hum. Genet. 80 (2007) 345–352, https://doi.org/10.1086/511134.
[79] T. Nakagawa, Y. Xiong, X-linked mental retardation gene CUL4B targets ubiquitylation of H3K4 methyltransferase component WDR5 and regulates neuronal gene expression, Mol. Cell 43 (2011) 381–391, https://doi.org/10.1016/j.molcel.2011.05.033.
[80] A.T. Vulto-van Silfhout, et al., Variants in CUL4B are associated with cerebral malformations, Hum. Mutat. 36 (2015) 106–117, https://doi.org/10.1002/humu.22718.
[81] C.J. Klein, et al., Ubiquitin ligase defect by DCAF8 mutation causes HMSN2 with giant axons, Neurology 82 (2014) 873–878, https://doi.org/10.1212/WNL.0000000000000206.
[82] E. Webster, et al., De novo PHIP-predicted deleterious variants are associated with developmental delay, intellectual disability, obesity, and dysmorphic features, Cold Spring Harb. Mol. Case Stud. 2 (2016) a001172, https://doi.org/10.1101/mcs.a001172.
[83] R.H. Ali, et al., Exome sequencing revealed a novel biallelic deletion in the DCAF17 gene underlying Woodhouse Sakati syndrome, Clin. Genet. 90 (2016) 263–269, https://doi.org/10.1111/cge.12700.
[84] M.G. Kapetanaki, et al., The DDB1-CUL4ADDB2 ubiquitin ligase is deficient in xeroderma pigmentosum group E and targets histone H2A at UV-damaged DNA sites, Proc. Natl. Acad. Sci. U. S. A. 103 (2006) 2588–2593, https://doi.org/10.1073/pnas.0511160103.
[85] Z. Zha, et al., Hypertension-associated C825T polymorphism impairs the function of Gbeta3 to target GRK2 ubiquitination, Cell Discov. 2 (2016) 16005, https://doi.org/10.1038/celldisc.2016.5.
[86] D. Hanahan, R.A. Weinberg, Hallmarks of cancer: the next generation, Cell 144 (2011) 646–674, https://doi.org/10.1016/j.cell.2011.02.013.
[87] D. Hanahan, R.A. Weinberg, The hallmarks of cancer, Cell 100 (2000) 57–70.
[88] J. Lee, P. Zhou, Pathogenic role of the CRL4 ubiquitin ligase in human disease, Front. Oncol. 2 (2012) 21, https://doi.org/10.3389/fonc.2012.00021.
[89] Y.L. Yang, et al., Lung tumourigenesis in a conditional Cul4A transgenic mouse model, J. Pathol. 233 (2014) 113–123, https://doi.org/10.1002/path.4352.
[90] Y. Wang, et al., PR-Set7 is degraded in a conditional Cul4A transgenic mouse model of lung cancer, Zhongguo Fei Ai Za Zhi 18 (2015) 345–350, https://doi.org/10.3779/j.issn.1009-3419.2015.06.15.
[91] Y. Wang, et al., Analysis of lung tumor initiation and progression in transgenic mice for Cre-inducible overexpression of Cul4A gene, Thorac. Cancer 6 (2015) 480–487, https://doi.org/10.1111/1759-7714.12257.
[92] J. Yuan, et al., Accelerated hepatocellular carcinoma development in CUL4B transgenic mice, Oncotarget 6 (2015) 15209–15221, https://doi.org/10.18632/oncotarget.3829.
[93] L. Jia, et al., Dysregulation of CUL4A and CUL4B ubiquitin ligases in lung cancer, J. Biol. Chem. 292 (2017) 2966–2978, https://doi.org/10.1074/jbc.M116.765230.
[94] J. Jin, E.E. Arias, J. Chen, J.W. Harper, J.C. Walter, A family of diverse Cul4-Ddb1-interacting proteins includes Cdt2, which is required for S phase destruction of the replication factor Cdt1, Mol. Cell 23 (2006) 709–721, https://doi.org/10.1016/j.molcel.2006.08.010.
[95] L.A. Higa, I.S. Mihaylov, D.P. Banks, J. Zheng, H. Zhang, Radiation-mediated

proteolysis of CDT1 by CUL4-ROC1 and CSN complexes constitutes a new checkpoint, Nat. Cell Biol. 5 (2003) 1008–1015, https://doi.org/10.1038/ncb1061.
[96] J. Hu, C.M. McCall, T. Ohta, Y. Xiong, Targeted ubiquitination of CDT1 by the DDB1-CUL4A-ROC1 ligase in response to DNA damage, Nat. Cell Biol. 6 (2004) 1003–1009, https://doi.org/10.1038/ncb1172.
[97] W. Zhong, H. Feng, F.E. Santiago, E.T. Kipreos, CUL-4 ubiquitin ligase maintains genome stability by restraining DNA-replication licensing, Nature 423 (2003) 885–889, https://doi.org/10.1038/nature01747.
[98] Y. Kim, N.G. Starostina, E.T. Kipreos, The CRL4Cdt2 ubiquitin ligase targets the degradation of p21Cip1 to control replication licensing, Genes Dev. 22 (2008) 2507–2519, https://doi.org/10.1101/gad.1703708.
[99] T. Abbas, et al., PCNA-dependent regulation of p21 ubiquitylation and degradation via the CRL4Cdt2 ubiquitin ligase complex, Genes Dev. 22 (2008) 2496–2506, https://doi.org/10.1101/gad.1676108.
[100] R.C. Centore, et al., CRL4(Cdt2)-mediated destruction of the histone methyltransferase Set8 prevents premature chromatin compaction in S phase, Mol. Cell 40 (2010) 22–33, https://doi.org/10.1016/j.molcel.2010.09.015.
[101] G.A. Miranda-Carboni, et al., A functional link between Wnt signaling and SKP2-independent p27 turnover in mammary tumors, Genes Dev. 22 (2008) 3121–3134, https://doi.org/10.1101/gad.1692808.
[102] Y. Gong, et al., CUL4A promotes cell invasion in gastric cancer by activating the NF-kappaB signaling pathway, Biologics 11 (2017) 45–53, https://doi.org/10.2147/BTT.S127650.
[103] J. Deng, et al., Cullin 4A (CUL4A), a direct target of miR-9 and miR-137, promotes gastric cancer proliferation and invasion by regulating the Hippo signaling pathway, Oncotarget 7 (2016) 10037–10050, https://doi.org/10.18632/oncotarget.7048.
[104] Y. Wang, et al., CUL4A induces epithelial-mesenchymal transition and promotes cancer metastasis by regulating ZEB1 expression, Cancer Res. 74 (2014) 520–531, https://doi.org/10.1158/0008-5472.CAN-13-2182.
[105] L.C. Chen, et al., The human homologue for the Caenorhabditis elegans cul-4 gene is amplified and overexpressed in primary breast cancers, Cancer Res. 58 (1998) 3677–3683.
[106] Y. Wang, et al., Involvement of CUL4A in regulation of multidrug resistance to P-gp substrate drugs in breast cancer cells, Molecules 19 (2013) 159–176, https://doi.org/10.3390/molecules19010159.
[107] X. Sui, et al., CUL4A promotes proliferation and metastasis of colorectal cancer cells by regulating H3K4 trimethylation in epithelial-mesenchymal transition, Onco Targets Ther. 10 (2017) 735–743, https://doi.org/10.2147/OTT.S118897.
[108] W. Ren, et al., Aberrant expression of CUL4A is associated with IL-6/ STAT3 activation in colorectal cancer progression, Arch. Med. Res. 47 (2016) 214–222, https://doi.org/10.1016/j.arcmed.2016.07.001.
[109] W. Ren, et al., Jak-STAT3 pathway triggers DICER1 for proteasomal degradation by ubiquitin ligase complex of CUL4A(DCAF1) to promote colon cancer development, Cancer Lett. 375 (2016) 209–220, https://doi.org/10.1016/j.canlet.2016.02.055.
[110] K. Hrecka, et al., HIV-1 and HIV-2 exhibit divergent interactions with HLTF and UNG2 DNA repair proteins, Proc. Natl. Acad. Sci. U. S. A. 113 (2016) E3921–E3930, https://doi.org/10.1073/pnas.1605023113.
[111] Y. Wang, et al., CUL4A overexpression enhances lung tumor growth and sensitizes lung cancer cells to erlotinib via transcriptional regulation of EGFR, Mol. Cancer 13 (2014) 252, https://doi.org/10.1186/1476-4598-13-252.
[112] P. Birner, et al., Human homologue for Caenorhabditis elegans CUL-4 protein overexpression is associated with malignant potential of epithelial ovarian tumours and poor outcome in carcinoma, J. Clin. Pathol. 65 (2012) 507–511, https://doi.org/10.1136/jclinpath-2011-200463.
[113] G. Huang, et al., CUL4A overexpression as an independent adverse prognosticator in intrahepatic cholangiocarcinoma, BMC Cancer 17 (2017) 395, https://doi.org/10.1186/s12885-017-3389-z.
[114] T.J. Zhang, et al., Cullin 4A is associated with epithelial to mesenchymal transition and poor prognosis in perihilar cholangiocarcinoma, World J. Gastroenterol. 23 (2017) 2318–2329, https://doi.org/10.3748/wjg.v23.i13.2318.
[115] B. Song, H. Zhan, Q. Bian, J. Li, Knockdown of CUL4B inhibits proliferation and promotes apoptosis of colorectal cancer cells through suppressing the Wnt/beta-catenin signaling pathway, Int. J. Clin. Exp. Pathol. 8 (2015) 10394–10402.
[116] T. Jiang, et al., Cullin 4B is a novel prognostic marker that correlates with colon cancer progression and pathogenesis, Med. Oncol. 30 (2013) 534, https://doi.org/10.1007/s12032-013-0534-7.
[117] Y. Pan, et al., CUL4A facilitates hepatocarcinogenesis by promoting cell cycle progression and epithelial-mesenchymal transition, Sci. Rep. 5 (2015) 17006, https://doi.org/10.1038/srep17006.
[118] J. Mi, et al., Dysregulation of the miR-194-CUL4B negative feedback loop drives tumorigenesis in non-small-cell lung carcinoma, Mol. Oncol. 11 (2017) 305–319, https://doi.org/10.1002/1878-0261.12038.
[119] J.Q. Zhang, et al., MicroRNA-300 promotes apoptosis and inhibits proliferation, migration, invasion and epithelial-mesenchymal transition via the Wnt/beta-catenin signaling pathway by targeting CUL4B in pancreatic cancer cells, J. Cell. Biochem. (2017), https://doi.org/10.1002/jcb.26270.
[120] P. Li, et al., Cul4B is a novel prognostic marker in cholangiocarcinoma, Oncol. Lett. 14 (2017) 1265–1274, https://doi.org/10.3892/ol.2017.6297.
[121] Y. Yang, et al., CRL4B promotes tumorigenesis by coordinating with SUV39H1/HP1/DNMT3A in DNA methylation-based epigenetic silencing, Oncogene 34 (2015) 104–118, https://doi.org/10.1038/onc.2013.522.
[122] J. Yuan, et al., CUL4B activates Wnt/beta-catenin signalling in hepatocellular carcinoma by repressing Wnt antagonists, J. Pathol. 235 (2015) 784–795, https://doi.org/10.1002/path.4492.
[123] W.W. Pan, et al., Ubiquitin E3 ligase CRL4(CDT2/DCAF2) as a potential chemotherapeutic target for ovarian surface epithelial cancer, J. Biol. Chem. 288 (2013) 29680–29691, https://doi.org/10.1074/jbc.M113.495069.
[124] R. Xing, et al., RBX1 expression is an unfavorable prognostic factor in patients with non-small cell lung cancer, Surg. Oncol. 25 (2016) 147–151, https://doi.org/10.1016/j.suronc.2016.05.006.
[125] K. Migita, et al., Prognostic impact of RING box protein-1 (RBX1) expression in gastric cancer, Gastric Cancer 17 (2014) 601–609, https://doi.org/10.1007/s10120-013-0318-y.
[126] W. Wang, et al., Overexpression of RING box protein-1 (RBX1) associated with poor prognosis of non-muscle-invasive bladder transitional cell carcinoma, J. Surg. Oncol. 107 (2013) 758–761, https://doi.org/10.1002/jso.23317.
[127] R. Dualan, et al., Chromosomal localization and cDNA cloning of the genes (DDB1 and DDB2) for the p127 and p48 subunits of a human damage-specific DNA binding protein, Genomics 29 (1995) 62–69, https://doi.org/10.1006/geno.1995.1215.
[128] C.M. Murphy, et al., Hepatitis B virus X protein promotes degradation of SMC5/6 to enhance HBV replication, Cell Rep. 16 (2016) 2846–2854, https://doi.org/10.1016/j.celrep.2016.08.026.
[129] C. Wang, et al., Hepatitis B virus X (HBx) induces tumorigenicity of hepatic progenitor cells in 3,5-diethoxycarbonyl-1,4-dihydrocollidine-treated HBx transgenic mice, Hepatology 55 (2012) 108–120, https://doi.org/10.1002/hep.24675.
[130] K. Koike, et al., High-level expression of hepatitis B virus HBx gene and hepatocarcinogenesis in transgenic mice, Hepatology 19 (1994) 810–819.
[131] R. Ding, et al., Overexpressed Id-1 is associated with patient prognosis and HBx expression in hepatitis B virus-related hepatocellular carcinoma, Cancer Biol. Ther. 10 (2010) 299–307.
[132] Y.M. Zhou, L. Cao, B. Li, X.Z. Zhang, Z.F. Yin, Expression of HBx protein in hepatitis B virus-infected intrahepatic cholangiocarcinoma, Hepatobiliary Pancreat Dis. Int. 11 (2012) 532–535.
[133] L. Xie, W. Wang, B. Xu, Y. Liu, Experimental study on hepatitis B-virus X gene expression in adenoid cystic carcinoma, Hua Xi Kou Qiang Yi Xue Za Zhi 32 (2014) 328–330.
[134] G. Hwang, et al., SMC5/6 is required for the formation of segregation-competent bivalent chromosomes during meiosis I in mouse oocytes, Development 144 (2017) 1648–1660, https://doi.org/10.1242/dev.145607.
[135] T. Jiang, M. Liu, J. Wu, Y. Shi, Structural and biochemical analysis of Bcl-2 interaction with the hepatitis B virus protein HBx, Proc. Natl. Acad. Sci. U. S. A. 113 (2016) 2074–2079, https://doi.org/10.1073/pnas.1525616113.
[136] X. Wang, et al., VprBP/DCAF1 regulates the degradation and nonproteolytic activation of the cell cycle transcription factor FoxM1, Mol. Cell Biol. 37 (2017), https://doi.org/10.1128/MCB.00609-16.
[137] E.S. Jung, et al., Jmjd2C increases MyoD transcriptional activity through inhibiting G9a-dependent MyoD degradation, Biochim. Biophys. Acta 1849 (2015) 1081–1094, https://doi.org/10.1016/j.bbagrm.2015.07.001.
[138] J.M. Lee, et al., EZH2 generates a methyl degron that is recognized by the DCAF1/DDB1/CUL4 E3 ubiquitin ligase complex, Mol. Cell 48 (2012) 572–586, https://doi.org/10.1016/j.molcel.2012.09.004.
[139] W. Li, et al., Merlin/NF2 loss-driven tumorigenesis linked to CRL4(DCAF1)-mediated inhibition of the hippo pathway kinases Lats1 and 2 in the nucleus, Cancer Cell 26 (2014) 48–60, https://doi.org/10.1016/j.ccr.2014.05.001.
[140] W. Ni, et al., A novel lncRNA uc.134 represses hepatocellular carcinoma progression by inhibiting CUL4A-mediated ubiquitination of LATS1, J. Hematol. Oncol. 10 (2017) 91, https://doi.org/10.1186/s13045-017-0449-4.
[141] J. Huang, J. Chen, VprBP targets Merlin to the Roc1-Cul4A-DDB1 E3 ligase complex for degradation, Oncogene 27 (2008) 4056–4064, https://doi.org/10.1038/onc.2008.44.
[142] W. Li, et al., Merlin/NF2 suppresses tumorigenesis by inhibiting the E3 ubiquitin ligase CRL4(DCAF1) in the nucleus, Cell 140 (2010) 477–490, https://doi.org/10.1016/j.cell.2010.01.029.
[143] H. Oda, et al., Regulation of the histone H4 monomethylase PR-Set7 by CRL4(Cdt2)-mediated PCNA-dependent degradation during DNA damage, Mol. Cell 40 (2010) 364–376, https://doi.org/10.1016/j.molcel.2010.10.011.
[144] M. Tardat, et al., The histone H4 Lys 20 methyltransferase PR-Set7 regulates replication origins in mammalian cells, Nat. Cell Biol. 12 (2010) 1086–1093, https://doi.org/10.1038/ncb2113.
[145] T. Abbas, et al., CRL4(Cdt2) regulates cell proliferation and histone gene expression by targeting PR-Set7/Set8 for degradation, Mol. Cell 40 (2010) 9–21, https://doi.org/10.1016/j.molcel.2010.09.014.
[146] J. Huh, H. Piwnica-Worms, CRL4(CDT2) targets CHK1 for PCNA-independent destruction, Mol. Cell. Biol. 33 (2013) 213–226, https://doi.org/10.1128/MCB.00847-12.
[147] X. Tong, et al., CUL4-DDB1-CDT2 E3 ligase regulates the molecular clock activity by promoting ubiquitination-dependent degradation of the mammalian CRY1, PLoS One 10 (2015) e0139725, , https://doi.org/10.1371/journal.pone.0139725.
[148] Y. Li, A. Jaramillo-Lambert, J. Hao, Y. Yang, W. Zhu, The stability of histone acetyltransferase general control non-derepressible (Gcn) 5 is regulated by Cullin4-RING E3 ubiquitin ligase, J. Biol. Chem. 286 (2011) 41344–41352, https://doi.org/10.1074/jbc.M111.290767.
[149] V. Vanderdys, et al., The Neddylation Inhibitor Pevonedistat (MLN4924) suppresses and radiosensitizes head and neck squamous carcinoma cells and tumors, Mol. Cancer Ther. (2017), https://doi.org/10.1158/1535-7163.MCT-17-0083.
[150] M. Benamar, et al., Inactivation of the CRL4-CDT2-SET8/p21 ubiquitylation and degradation axis underlies the therapeutic efficacy of pevonedistat in melanoma, EBioMedicine 10 (2016) 85–100, https://doi.org/10.1016/j.ebiom.2016.06.023.

[151] C. Mackintosh, et al., 1q gain and CDT2 overexpression underlie an aggressive and highly proliferative form of Ewing sarcoma, Oncogene 31 (2012) 1287–1298, https://doi.org/10.1038/onc.2011.317.

[152] L.A. Higa, et al., CUL4-DDB1 ubiquitin ligase interacts with multiple WD40-repeat proteins and regulates histone methylation, Nat. Cell Biol. 8 (2006) 1277–1283, https://doi.org/10.1038/ncb1490.

[153] Y.T. Wang, et al., Ubiquitination of tumor suppressor PML regulates prometastatic and immunosuppressive tumor microenvironment, J. Clin. Invest. 127 (2017) 2982–2997, https://doi.org/10.1172/JCI89957.

[154] H.H. Chen, et al., NRIP/DCAF6 stabilizes the androgen receptor protein by displacing DDB2 from the CUL4A-DDB1 E3 ligase complex in prostate cancer, Oncotarget 8 (2017) 21501–21515, https://doi.org/10.18632/oncotarget.15308.

[155] J. Cao, et al., The overexpression and prognostic role of DCAF13 in hepatocellular carcinoma, Tumour Biol. 39 (2017), https://doi.org/10.1177/1010428317705753.

[156] Y. Yan, X. Zhang, R.J. Legerski, Artemis interacts with the Cul4A-DDB1DDB2 ubiquitin E3 ligase and regulates degradation of the CDK inhibitor p27, Cell Cycle 10 (2011) 4098–4109, https://doi.org/10.4161/cc.10.23.18227.

[157] N. Roy, et al., DDB2 suppresses epithelial-to-mesenchymal transition in colon cancer, Cancer Res. 73 (2013) 3771–3782, https://doi.org/10.1158/0008-5472.CAN-12-4069.

[158] R. Matsunuma, et al., UV damage-induced phosphorylation of HBO1 triggers CRL4DDB2-mediated degradation to regulate cell proliferation, Mol. Cell. Biol. 36 (2015) 394–406, https://doi.org/10.1128/MCB.00809-15.

[159] D. Del Prete, R.C. Rice, A.M. Rajadhyaksha, L. D'Adamio, Amyloid precursor protein (APP) may act as a substrate and a recognition unit for CRL4CRBN and Stub1 E3 ligases facilitating ubiquitination of proteins involved in presynaptic functions and neurodegeneration, J. Biol. Chem. 291 (2016) 17209–17227, https://doi.org/10.1074/jbc.M116.733626.

[160] Y.A. Chen, et al., The Cullin 4A/B-DDB1-Cereblon E3 ubiquitin ligase complex mediates the degradation of CLC-1 chloride channels, Sci. Rep. 5 (2015) 10667, https://doi.org/10.1038/srep10667.

[161] A. Broyl, et al., High cereblon expression is associated with better survival in patients with newly diagnosed multiple myeloma treated with thalidomide maintenance, Blood 121 (2013) 624–627, https://doi.org/10.1182/blood-2012-06-438101.

[162] J. Kronke, et al., Lenalidomide causes selective degradation of IKZF1 and IKZF3 in multiple myeloma cells, Science 343 (2014) 301–305, https://doi.org/10.1126/science.1244851.

[163] M.E. Matyskiela, et al., A novel cereblon modulator recruits GSPT1 to the CRL4(CRBN) ubiquitin ligase, Nature 535 (2016) 252–257, https://doi.org/10.1038/nature18611.

[164] J. Kronke, et al., Lenalidomide induces ubiquitination and degradation of CK1alpha in del(5q) MDS, Nature 523 (2015) 183–188, https://doi.org/10.1038/nature14610.

[165] L. Ta, C. Xuan, N. Xing, X. Zhu, COP1 is downregulated in renal cell carcinoma (RCC) and inhibits the migration of RCC ACHN cells in vitro, Mol. Med. Rep. 14 (2016) 1371–1378, https://doi.org/10.3892/mmr.2016.5373.

[166] Z. Zhang, et al., Transcription factor Etv5 is essential for the maintenance of alveolar type II cells, Proc. Natl. Acad. Sci. U. S. A. 114 (2017) 3903–3908, https://doi.org/10.1073/pnas.1621177114.

[167] I.E. Wertz, et al., Human De-etiolated-1 regulates c-Jun by assembling a CUL4A ubiquitin ligase, Science 303 (2004) 1371–1374, https://doi.org/10.1126/science.1093549.

[168] D. Migliorini, et al., Cop1 constitutively regulates c-Jun protein stability and functions as a tumor suppressor in mice, J. Clin. Invest. 121 (2011) 1329–1343, https://doi.org/10.1172/JCI45784.

[169] L.D. Guo, et al., Functional analysis of DNA damage repair factor WDR70 and its mutation in ovarian cancer, Sichuan Da Xue Xue Bao Yi Xue Ban 47 (2016) 501–506.

[170] Y. Wu, et al., A genome-scale CRISPR-Cas9 screening method for protein stability reveals novel regulators of Cdc25A, Cell Discov. 2 (2016) 16014, https://doi.org/10.1038/celldisc.2016.14.

[171] J.Y. Lo, B.N. Spatola, S.P. Curran, WDR23 regulates NRF2 independently of KEAP1, PLoS Genet. 13 (2017) e1006762, , https://doi.org/10.1371/journal.pgen.1006762.

[172] Z. Chen, et al., CRL4BDCAF11 E3 ligase targets p21 for degradation to control cell cycle progression in human osteosarcoma cells, Sci. Rep. 7 (2017) 1175, https://doi.org/10.1038/s41598-017-01344-9.

[173] T. Han, et al., Anticancer sulfonamides target splicing by inducing RBM39 degradation via recruitment to DCAF15, Science 356 (2017), https://doi.org/10.1126/science.aal3755.

[174] L. Nogues, et al., G protein-coupled receptor kinase 2 (GRK2) promotes breast tumorigenesis through a HDAC6-Pin1 axis, EBioMedicine 13 (2016) 132–145, https://doi.org/10.1016/j.ebiom.2016.09.030.

[175] Y. Ma, C.C. Han, Q. Huang, W.Y. Sun, W. Wei, GRK2 overexpression inhibits IGF1-induced proliferation and migration of human hepatocellular carcinoma cells by downregulating EGR1, Oncol. Rep. 35 (2016) 3068–3074, https://doi.org/10.3892/or.2016.4641.

[176] V. Rivas, et al., Developmental and tumoral vascularization is regulated by G protein-coupled receptor kinase 2, J. Clin. Invest. 123 (2013) 4714–4730, https://doi.org/10.1172/JCI67333.

[177] L. Zhou, et al., G-protein-coupled receptor kinase 2 in pancreatic cancer: clinicopathologic and prognostic significance, Hum. Pathol. 56 (2016) 171–177, https://doi.org/10.1016/j.humpath.2016.06.012.

[178] B. Meszaros, M. Kumar, T.J. Gibson, B. Uyar, Z. Dosztanyi, Degrons in cancer, Sci. Signal. 10 (2017), https://doi.org/10.1126/scisignal.aak9982.

[179] S.C. Lo, X. Li, M.T. Henzl, L.J. Beamer, M. Hannink, Structure of the Keap1:Nrf2 interface provides mechanistic insight into Nrf2 signaling, EMBO J. 25 (2006) 3605–3617, https://doi.org/10.1038/sj.emboj.7601243.

[180] L. Chang, D. Barford, Insights into the anaphase-promoting complex: a molecular machine that regulates mitosis, Curr. Opin. Struct. Biol. 29 (2014) 1–9, https://doi.org/10.1016/j.sbi.2014.08.003.

[181] A.W. Lau, H. Fukushima, W. Wei, The Fbw7 and betaTRCP E3 ubiquitin ligases and their roles in tumorigenesis, Front. Biosci. (Landmark Ed) 17 (2012) 2197–2212.

[182] M. Ivan, et al., HIFalpha targeted for VHL-mediated destruction by proline hydroxylation: implications for O2 sensing, Science 292 (2001) 464–468, https://doi.org/10.1126/science.1059817.

[183] S. Uljon, et al., Structural basis for substrate selectivity of the E3 ligase COP1, Structure 24 (2016) 687–696, https://doi.org/10.1016/j.str.2016.03.002.

[184] C.G. Havens, et al., Direct role for proliferating cell nuclear antigen in substrate recognition by the E3 ubiquitin ligase CRL4Cdt2, J. Biol. Chem. 287 (2012) 11410–11421, https://doi.org/10.1074/jbc.M111.337683.

[185] T.V. Nguyen, et al., Glutamine triggers acetylation-dependent degradation of glutamine synthetase via the thalidomide receptor cereblon, Mol. Cell 61 (2016) 809–820, https://doi.org/10.1016/j.molcel.2016.02.032.

[186] T.K. Neklesa, J.D. Winkler, C.M. Crews, Targeted protein degradation by PROTACs, Pharmacol. Ther. 174 (2017) 138–144, https://doi.org/10.1016/j.pharmthera.2017.02.027.

[187] C.J. Punt, J. Tol, More is less – combining targeted therapies in metastatic colorectal cancer, Nat. Rev. Clin. Oncol. 6 (2009) 731–733, https://doi.org/10.1038/nrclinonc.2009.168.

[188] M.L. Bobbin, J.J. Rossi, RNA interference (RNAi)-based therapeutics: delivering on the promise? Annu. Rev. Pharmacol. Toxicol. 56 (2016) 103–122, https://doi.org/10.1146/annurev-pharmtox-010715-103633.

[189] M. Scheffner, J.M. Huibregtse, R.D. Vierstra, P.M. Howley, The HPV-16 E6 and E6-AP complex functions as a ubiquitin-protein ligase in the ubiquitination of p53, Cell 75 (1993) 495–505.

[190] P. Zhou, R. Bogacki, L. McReynolds, P.M. Howley, Harnessing the ubiquitination machinery to target the degradation of specific cellular proteins, Mol. Cell 6 (2000) 751–756.

[191] F. Cong, J. Zhang, W. Pao, P. Zhou, H. Varmus, A protein knockdown strategy to study the function of beta-catenin in tumorigenesis, BMC Mol. Biol. 4 (2003) 10, https://doi.org/10.1186/1471-2199-4-10.

[192] J. Zhang, N. Zheng, P. Zhou, Exploring the functional complexity of cellular proteins by protein knockout, Proc. Natl. Acad. Sci. U. S. A. 100 (2003) 14127–14132, https://doi.org/10.1073/pnas.2233012100.

[193] Y. Su, S. Ishikawa, M. Kojima, B. Liu, Eradication of pathogenic beta-catenin by Skp1/Cullin/F box ubiquitination machinery, Proc. Natl. Acad. Sci. U. S. A. 100 (2003) 12729–12734, https://doi.org/10.1073/pnas.2133261100.

[194] J. Liu, J. Stevens, N. Matsunami, R.L. White, Targeted degradation of beta-catenin by chimeric F-box fusion proteins, Biochem. Biophys. Res. Commun. 313 (2004) 1023–1029.

[195] J.C. Cohen, et al., Transient in utero knockout (TIUKO) of C-MYC affects late lung and intestinal development in the mouse, BMC Dev. Biol. 4 (2004) 4, https://doi.org/10.1186/1471-213X-4-4.

[196] W. Chen, J. Lee, S.Y. Cho, H.A. Fine, Proteasome-mediated destruction of the cyclin a/cyclin-dependent kinase 2 complex suppresses tumor cell growth in vitro and in vivo, Cancer Res. 64 (2004) 3949–3957, https://doi.org/10.1158/0008-5472.CAN-03-3906.

[197] F. Kong, et al., Engineering a single ubiquitin ligase for the selective degradation of all activated ErbB receptor tyrosine kinases, Oncogene 33 (2014) 986–995, https://doi.org/10.1038/onc.2013.33.

[198] J. Hannah, P. Zhou, Maximizing target protein ablation by integration of RNAi and protein knockout, Cell Res. 21 (2011) 1152–1154, https://doi.org/10.1038/cr.2011.89.

[199] K.M. Sakamoto, Protacs for treatment of cancer, Pediatr. Res. 67 (2010) 505–508, https://doi.org/10.1203/PDR.0b013e3181d35017.

[200] K.M. Sakamoto, et al., Protacs: chimeric molecules that target proteins to the Skp1-Cullin-F box complex for ubiquitination and degradation, Proc. Natl. Acad. Sci. U. S. A. 98 (2001) 8554–8559, https://doi.org/10.1073/pnas.141230798.

[201] P. Jaakkola, et al., Targeting of HIF-alpha to the von Hippel-Lindau ubiquitylation complex by O2-regulated prolyl hydroxylation, Science 292 (2001) 468–472, https://doi.org/10.1126/science.1059796.

[202] J.S. Schneekloth, et al., Chemical genetic control of protein levels: selective in vivo targeted degradation, J. Am. Chem. Soc. 126 (2004) 3748–3754, https://doi.org/10.1021/ja039025z.

[203] D.P. Bondeson, et al., Catalytic in vivo protein knockdown by small-molecule PROTACs, Nat. Chem. Biol. 11 (2015) 611–617, https://doi.org/10.1038/nchembio.1858.

[204] D.L. Buckley, et al., Small-molecule inhibitors of the interaction between the E3 ligase VHL and HIF1alpha, Angew. Chem. Int. Ed. Engl. 51 (2012) 11463–11467, https://doi.org/10.1002/anie.201206231.

[205] D.L. Buckley, et al., Targeting the von Hippel-Lindau E3 ubiquitin ligase using small molecules to disrupt the VHL/HIF-1alpha interaction, J. Am. Chem. Soc. 134 (2012) 4465–4468, https://doi.org/10.1021/ja209924v.

[206] I. Van Molle, et al., Dissecting fragment-based lead discovery at the von Hippel-Lindau protein:hypoxia inducible factor 1alpha protein-protein interface, Chem. Biol. 19 (2012) 1300–1312, https://doi.org/10.1016/j.chembiol.2012.08.015.

[207] D.L. Buckley, et al., HaloPROTACs: use of small molecule PROTACs to induce degradation of HaloTag fusion proteins, ACS Chem. Biol. 10 (2015) 1831–1837,

https://doi.org/10.1021/acschembio.5b00442.

[208] J. Kronke, S.N. Hurst, B.L. Ebert, Lenalidomide induces degradation of IKZF1 and IKZF3, Oncoimmunology 3 (2014) e941742, , https://doi.org/10.4161/21624011.2014.941742.

[209] G. Lu, et al., The myeloma drug lenalidomide promotes the cereblon-dependent destruction of Ikaros proteins, Science 343 (2014) 305–309, https://doi.org/10.1126/science.1244917.

[210] J. Lu, et al., Hijacking the E3 ubiquitin ligase cereblon to efficiently target BRD4, Chem. Biol. 22 (2015) 755–763, https://doi.org/10.1016/j.chembiol.2015.05.009.

[211] G.E. Winter, et al., DRUG DEVELOPMENT. Phthalimide conjugation as a strategy for in vivo target protein degradation, Science 348 (2015) 1376–1381, https://doi.org/10.1126/science.aab1433.

[212] M. Schiedel, et al., Chemically induced degradation of Sirtuin 2 (Sirt2) by a proteolysis targeting chimera (PROTAC) based on Sirtuin rearranging ligands (SirReals), J. Med. Chem. (2017), https://doi.org/10.1021/acs.jmedchem.6b01872.

[213] C.M. Robb, et al., Chemically induced degradation of CDK9 by a proteolysis targeting chimera (PROTAC), Chem. Commun. (Camb) 53 (2017) 7577–7580, https://doi.org/10.1039/c7cc03879h.

[214] A.R. Schneekloth, M. Pucheault, H.S. Tae, C.M. Crews, Targeted intracellular protein degradation induced by a small molecule: En route to chemical proteomics, Bioorg. Med. Chem. Lett. 18 (2008) 5904–5908, https://doi.org/10.1016/j.bmcl.2008.07.114.

[215] P. Ottis, C.M. Crews, Proteolysis-targeting chimeras: induced protein degradation as a therapeutic strategy, ACS Chem. Biol. 12 (2017) 892–898, https://doi.org/10.1021/acschembio.6b01068.

[216] Y. Itoh, R. Kitaguchi, M. Ishikawa, M. Naito, Y. Hashimoto, Design, synthesis and biological evaluation of nuclear receptor-degradation inducers, Bioorg. Med. Chem. 19 (2011) 6768–6778, https://doi.org/10.1016/j.bmc.2011.09.041.

[217] Y. Demizu, et al., Design and synthesis of estrogen receptor degradation inducer based on a protein knockdown strategy, Bioorg. Med. Chem. Lett. 22 (2012) 1793–1796, https://doi.org/10.1016/j.bmcl.2011.11.086.

[218] Y. Itoh, M. Ishikawa, M. Naito, Y. Hashimoto, Protein knockdown using methyl bestatin-ligand hybrid molecules: design and synthesis of inducers of ubiquitination-mediated degradation of cellular retinoic acid-binding proteins, J. Am. Chem. Soc. 132 (2010) 5820–5826, https://doi.org/10.1021/ja100691p.

[219] K. Okuhira, et al., Specific degradation of CRABP-II via cIAP1-mediated ubiquitylation induced by hybrid molecules that crosslink cIAP1 and the target protein, FEBS Lett. 585 (2011) 1147–1152, https://doi.org/10.1016/j.febslet.2011.03.019.

[220] D. Zhang, S.H. Baek, A. Ho, K. Kim, Degradation of target protein in living cells by small-molecule proteolysis inducer, Bioorg. Med. Chem. Lett. 14 (2004) 645–648.

[221] P. Bargagna-Mohan, S.H. Baek, H. Lee, K. Kim, R. Mohan, Use of PROTACS as molecular probes of angiogenesis, Bioorg. Med. Chem. Lett. 15 (2005) 2724–2727, https://doi.org/10.1016/j.bmcl.2005.04.008.

[222] K. Cyrus, et al., Jostling for position: optimizing linker location in the design of estrogen receptor-targeting PROTACs, ChemMedChem 5 (2010) 979–985, https://doi.org/10.1002/cmdc.201000146.

[223] J. Hines, J.D. Gough, T.W. Corson, C.M. Crews, Posttranslational protein knockdown coupled to receptor tyrosine kinase activation with phosphoPROTACs, Proc. Natl. Acad. Sci. U. S. A. 110 (2013) 8942–8947, https://doi.org/10.1073/pnas.1217206110.

[224] T.T. Chu, et al., Specific knockdown of endogenous tau protein by peptide-directed ubiquitin-proteasome degradation, Cell Chem Biol 23 (2016) 453–461, https://doi.org/10.1016/j.chembiol.2016.02.016.

[225] K. Montrose, G.W. Krissansen, Design of a PROTAC that antagonizes and destroys the cancer-forming X-protein of the hepatitis B virus, Biochem. Biophys. Res. Commun. 453 (2014) 735–740, https://doi.org/10.1016/j.bbrc.2014.10.006.

[226] X. Wang, et al., New strategy for renal fibrosis: Targeting Smad3 proteins for ubiquitination and degradation, Biochem. Pharmacol. 116 (2016) 200–209, https://doi.org/10.1016/j.bcp.2016.07.017.

[227] R.K. Henning, et al., Degradation of Akt using protein-catalyzed capture agents, J. Pept. Sci. 22 (2016) 196–200, https://doi.org/10.1002/psc.2858.

[228] K.M. Sakamoto, et al., Development of Protacs to target cancer-promoting proteins for ubiquitination and degradation, Mol. Cell. Proteomics 2 (2003) 1350–1358, https://doi.org/10.1074/mcp.T300009-MCP200.

[229] N. Ohoka, et al., Cancer cell death induced by novel small molecules degrading the TACC3 protein via the ubiquitin-proteasome pathway, Cell Death Dis. 5 (2014) e1513, , https://doi.org/10.1038/cddis.2014.471.

[230] P. Ottis, et al., Assessing different E3 ligases for small molecule induced protein ubiquitination and degradation, ACS Chem. Biol. 12 (2017) 2570–2578, https://doi.org/10.1021/acschembio.7b00485.

[231] C. Maniaci, et al., Homo-PROTACs: bivalent small-molecule dimerizers of the VHL E3 ubiquitin ligase to induce self-degradation, Nat. Commun. 8 (2017) 830, https://doi.org/10.1038/s41467-017-00954-1.

[232] C. French, A. Small-Molecule, Targeting of BET proteins in cancer, Adv. Cancer Res. 131 (2016) 21–58, https://doi.org/10.1016/bs.acr.2016.04.001.

[233] K. Raina, et al., PROTAC-induced BET protein degradation as a therapy for castration-resistant prostate cancer, Proc. Natl. Acad. Sci. U. S. A. 113 (2016) 7124–7129, https://doi.org/10.1073/pnas.1521738113.

[234] B. Zhou, et al., Discovery of a small-molecule degrader of bromodomain and extra-terminal (BET) proteins with picomolar cellular potencies and capable of achieving tumor regression, J. Med. Chem. (2017), https://doi.org/10.1021/acs.jmedchem.6b01816.

[235] M. Zengerle, K.H. Chan, A. Ciulli, Selective small molecule induced degradation of the BET bromodomain protein BRD4, ACS Chem. Biol. 10 (2015) 1770–1777, https://doi.org/10.1021/acschembio.5b00216.

[236] M. Jung, K.A. Gelato, A. Fernandez-Montalvan, S. Siegel, B. Haendler, Targeting BET bromodomains for cancer treatment, Epigenomics 7 (2015) 487–501, https://doi.org/10.2217/epi.14.91.

[237] H. Lebraud, D.J. Wright, C.N. Johnson, T.D. Heightman, Protein degradation by in-cell self-assembly of proteolysis targeting chimeras, ACS Cent. Sci. 2 (2016) 927–934, https://doi.org/10.1021/acscentsci.6b00280.

[238] A.C. Lai, et al., Modular PROTAC design for the degradation of oncogenic BCR-ABL, Angew. Chem. Int. Ed. Engl. 55 (2016) 807–810, https://doi.org/10.1002/anie.201507634.

[239] J.T. Madak, C.R. Cuthbertson, W. Chen, H.D. Showalter, N. Neamati, Design, synthesis, and characterization of brequinar conjugates as probes to study DHODH inhibition, Chemistry 23 (2017) 13875–13878, https://doi.org/10.1002/chem.201702999.

[240] Z. Culig, F.R. Santer, Androgen receptor signaling in prostate cancer, Cancer Metastasis Rev. 33 (2014) 413–427, https://doi.org/10.1007/s10555-013-9474-0.

[241] I. Cristobal, et al., PP2A regulates signaling through hormonal receptors in breast cancer with important therapeutic implications, Biochim. Biophys. Acta 1868 (2017) 435–438, https://doi.org/10.1016/j.bbcan.2017.08.005.

[242] D. Huang, F. Yang, Y. Wang, X. Guan, Mechanisms of resistance to selective estrogen receptor down-regulator in metastatic breast cancer, Biochim. Biophys. Acta 1868 (2017) 148–156, https://doi.org/10.1016/j.bbcan.2017.03.008.

[243] D. Wu, A. Cheung, Y. Wang, S. Yu, F.L. Chan, The emerging roles of orphan nuclear receptors in prostate cancer, Biochim. Biophys. Acta 1866 (2016) 23–36, https://doi.org/10.1016/j.bbcan.2016.06.001.

[244] T.S. Bekaii-Saab, et al., A phase II study of chloroquinoxaline sulfonamide (CQS) in patients with metastatic colorectal carcinoma (MCRC), Investig. New Drugs 24 (2006) 343–346, https://doi.org/10.1007/510637-005-4827-3.

[245] M. Baur, M. Gneist, T. Owa, C. Dittrich, Clinical complete long-term remission of a patient with metastatic malignant melanoma under therapy with indisulam (E7070), Melanoma Res. 17 (2007) 329–331, https://doi.org/10.1097/CMR.0b013e3282ef4189.

[246] M. Mita, et al., Phase I study of E7820, an oral inhibitor of integrin alpha-2 expression with antiangiogenic properties, in patients with advanced malignancies, Clin. Cancer Res. 17 (2011) 193–200, https://doi.org/10.1158/1078-0432.CCR-10-0010.

[247] T. Uehara, et al., Selective degradation of splicing factor CAPERalpha by anticancer sulfonamides, Nat. Chem. Biol. 13 (2017) 675–680, https://doi.org/10.1038/nchembio.2363.

[248] E.S. Fischer, et al., Structure of the DDB1-CRBN E3 ubiquitin ligase in complex with thalidomide, Nature 512 (2014) 49–53, https://doi.org/10.1038/nature13527.

[249] Z. Zha, et al., A non-canonical function of gbeta as a subunit of E3 ligase in targeting GRK2 ubiquitylation, Mol. Cell 58 (2015) 794–803, https://doi.org/10.1016/j.molcel.2015.04.017.

[250] Y. Qian, et al., The CUL4B/AKT/beta-catenin axis restricts the accumulation of myeloid-derived suppressor cells to prohibit the establishment of a tumor-permissive microenvironment, Cancer Res. 75 (2015) 5070–5083, https://doi.org/10.1158/0008-5472.CAN-15-0898.

[251] W. Zhao, et al., Lack of CUL4B leads to increased abundance of GFAP-positive cells that is mediated by PTGDS in mouse brain, Hum. Mol. Genet. 24 (2015) 4686–4697, https://doi.org/10.1093/hmg/ddv200.

[252] M.H. Hung, Y.R. Jian, C.C. Tsao, S.W. Lin, Y.H. Chuang, Enhanced LPS-induced peritonitis in mice deficiency of cullin 4B in macrophages, Genes Immun. 15 (2014) 404–412, https://doi.org/10.1038/gene.2014.32.

[253] B. Jiang, et al., Lack of Cul4b, an E3 ubiquitin ligase component, leads to embryonic lethality and abnormal placental development, PLoS One 7 (2012) e37070, , https://doi.org/10.1371/journal.pone.0037070.

[254] J. Gao, et al., The CUL4-DDB1 ubiquitin ligase complex controls adult and embryonic stem cell differentiation and homeostasis, Elife 4 (2015), https://doi.org/10.7554/eLife.07539.

[255] C.M. McCall, et al., Human immunodeficiency virus type 1 Vpr-binding protein VprBP, a WD40 protein associated with the DDB1-CUL4 E3 ubiquitin ligase, is essential for DNA replication and embryonic development, Mol. Cell. Biol. 28 (2008) 5621–5633, https://doi.org/10.1128/MCB.00232-08.

[256] M.D. Kassmeier, et al., VprBP binds full-length RAG1 and is required for B-cell development and V(D)J recombination fidelity, EMBO J. 31 (2012) 945–958, https://doi.org/10.1038/emboj.2011.455.

[257] Z. Guo, et al., DCAF1 controls T-cell function via p53-dependent and -independent mechanisms, Nat. Commun. 7 (2016) 10307, https://doi.org/10.1038/ncomms10307.

[258] L. Casey Klockow, et al., The HIV-1 protein Vpr targets the endoribonuclease Dicer for proteasomal degradation to boost macrophage infection, Virology 444 (2013) 191–202, https://doi.org/10.1016/j.virol.2013.06.010.

[259] B. Romani, N. Shaykh Baygloo, M.R. Aghasadeghi, E. Allahbakhshi, HIV-1 Vpr protein enhances proteasomal degradation of MCM10 DNA replication factor through the Cul4-DDB1[VprBP] E3 ubiquitin ligase to induce G2/M cell cycle arrest, J. Biol. Chem. 290 (2015) 17380–17389, https://doi.org/10.1074/jbc.M115.641522.

[260] M. Kaur, M.M. Khan, A. Kar, A. Sharma, S. Saxena, CRL4-DDB1-VPRBP ubiquitin ligase mediates the stress triggered proteolysis of Mcm10, Nucleic Acids Res. 40 (2012) 7332–7346, https://doi.org/10.1093/nar/gks366.

[261] M.F. Karim, T. Yoshizawa, S.U. Sobuz, Y. Sato, K. Yamagata, Sirtuin 7-dependent deacetylation of DDB1 regulates the expression of nuclear receptor TR4, Biochem. Biophys. Res. Commun. 490 (2017) 423–428, https://doi.org/10.1016/j.bbrc.

2017.06.057.

[262] H.J. Sharifi, A.K. Furuya, R.M. Jellinger, M.D. Nekorchuk, C.M. de Noronha, Cullin4A and cullin4B are interchangeable for HIV Vpr and Vpx action through the CRL4 ubiquitin ligase complex, J. Virol. 88 (2014) 6944–6958, https://doi.org/10.1128/JVI.00241-14.

[263] T. Nakagawa, et al., CRL4(VprBP) E3 ligase promotes monoubiquitylation and chromatin binding of TET dioxygenases, Mol. Cell 57 (2015) 247–260, https://doi.org/10.1016/j.molcel.2014.12.002.

[264] X. Wen, L. Casey Klockow, M. Nekorchuk, H.J. Sharifi, C.M. de Noronha, The HIV1 protein Vpr acts to enhance constitutive DCAF1-dependent UNG2 turnover, PLoS One 7 (2012) e30939, , https://doi.org/10.1371/journal.pone.0030939.

[265] J. Ahn, et al., HIV-1 Vpr loads uracil DNA glycosylase-2 onto DCAF1, a substrate recognition subunit of a cullin 4A-ring E3 ubiquitin ligase for proteasome-dependent degradation, J. Biol. Chem. 285 (2010) 37333–37341, https://doi.org/10.1074/jbc.M110.133181.

[266] D. Banks, et al., L2DTL/CDT2 and PCNA interact with p53 and regulate p53 polyubiquitination and protein stability through MDM2 and CUL4A/DDB1 complexes, Cell Cycle 5 (2006) 1719–1729, https://doi.org/10.4161/cc.5.15.3150.

[267] S. Jorgensen, et al., SET8 is degraded via PCNA-coupled CRL4(CDT2) ubiquitylation in S phase and after UV irradiation, J. Cell Biol. 192 (2011) 43–54, https://doi.org/10.1083/jcb.201009076.

[268] K. Terai, T. Abbas, A.A. Jazaeri, A. Dutta, CRL4(Cdt2) E3 ubiquitin ligase monoubiquitinates PCNA to promote translesion DNA synthesis, Mol. Cell 37 (2010) 143–149, https://doi.org/10.1016/j.molcel.2009.12.018.

[269] S. Zhang, et al., A novel function of CRL4(Cdt2): regulation of the subunit structure of DNA polymerase delta in response to DNA damage and during the S phase, J. Biol. Chem. 288 (2013) 29550–29561, https://doi.org/10.1074/jbc.M113.490466.

[270] D.L. Evans, et al., MMSET is dynamically regulated during cell-cycle progression and promotes normal DNA replication, Cell Cycle 15 (2016) 95–105, https://doi.org/10.1080/15384101.2015.1121323.

[271] C. Han, et al., Cdt2-mediated XPG degradation promotes gap-filling DNA synthesis in nucleotide excision repair, Cell Cycle 14 (2015) 1103–1115, https://doi.org/10.4161/15384101.2014.973740.

[272] E. Shibata, A. Dar, A. Dutta, CRL4Cdt2 E3 ubiquitin ligase and proliferating cell nuclear antigen (PCNA) cooperate to degrade thymine DNA glycosylase in S phase, J. Biol. Chem. 289 (2014) 23056–23064, https://doi.org/10.1074/jbc.M114.574210.

[273] U. Jo, et al., PCNA-dependent cleavage and degradation of SDE2 regulates response to replication stress, PLoS Genet. 12 (2016) e1006465, , https://doi.org/10.1371/journal.pgen.1006465.

[274] L. Clijsters, R. Wolthuis, PIP-box-mediated degradation prohibits re-accumulation of Cdc6 during S phase, J. Cell Sci. 127 (2014) 1336–1345, https://doi.org/10.1242/jcs.145862.

[275] T. Suzuki, et al., Inhibition of DNA damage-induced apoptosis through Cdc7-mediated stabilization of Tob, J. Biol. Chem. 287 (2012) 40256–40265, https://doi.org/10.1074/jbc.M112.353805.

[276] H. Wang, et al., DCAF4L2 promotes colorectal cancer invasion and metastasis via mediating degradation of NFkappab negative regulator PPM1B, Am. J. Transl. Res. 8 (2016) 405–418.

[277] Z. Peng, Z. Liao, Y. Matsumoto, A. Yang, A.E. Tomkinson, Human DNA ligase I interacts with and is targeted for degradation by the DCAF7 specificity factor of the Cul4-DDB1 ubiquitin ligase complex, J. Biol. Chem. 291 (2016) 21893–21902, https://doi.org/10.1074/jbc.M116.746198.

[278] G. Li, et al., CRL4DCAF8 ubiquitin ligase targets histone H3K79 and promotes H3K9 methylation in the liver, Cell Rep. 18 (2017) 1499–1511, https://doi.org/10.1016/j.celrep.2017.01.039.

[279] B.S. Groh, et al., The antiobesity factor WDTC1 suppresses adipogenesis via the CRL4WDTC1 E3 ligase, EMBO Rep. 17 (2016) 638–647, https://doi.org/10.15252/embr.201540500.

[280] U. Djakbarova, W.F. Marzluff, M.M. Koseoglu, DDB1 and CUL4 associated factor 11 (DCAF11) mediates degradation of Stem-loop binding protein at the end of S phase, Cell Cycle 15 (2016) 1986–1996, https://doi.org/10.1080/15384101.2016.1191708.

[281] M.M. Brodersen, et al., CRL4(WDR23)-mediated SLBP UBIQUITYLATION ensures histone supply during DNA replication, Mol. Cell 62 (2016) 627–635, https://doi.org/10.1016/j.molcel.2016.04.017.

[282] F. Ohtake, et al., Dioxin receptor is a ligand-dependent E3 ubiquitin ligase, Nature 446 (2007) 562–566, https://doi.org/10.1038/nature05683.

[283] Y. Niikura, et al., CENP-A K124 ubiquitylation is required for CENP-A deposition at the centromere, Dev. Cell 32 (2015) 589–603, https://doi.org/10.1016/j.devcel.2015.01.024.

[284] J. An, et al., pSILAC mass spectrometry reveals ZFP91 as IMiD-dependent substrate of the CRL4CRBN ubiquitin ligase, Nat. Commun. 8 (2017) 15398, https://doi.org/10.1038/ncomms15398.

[285] S.W. Chang, et al., DDB2 is a novel AR interacting protein and mediates AR ubiquitination/degradation, Int. J. Biochem. Cell Biol. 44 (2012) 1952–1961, https://doi.org/10.1016/j.biocel.2012.07.023.

[286] M.A. El-Mahdy, et al., Cullin 4A-mediated proteolysis of DDB2 protein at DNA damage sites regulates in vivo lesion recognition by XPC, J. Biol. Chem. 281 (2006) 13404–13411, https://doi.org/10.1074/jbc.M511834200.

[287] A. Nag, T. Bondar, S. Shiv, P. Raychaudhuri, The xeroderma pigmentosum group E gene product DDB2 is a specific target of cullin 4A in mammalian cells, Mol. Cell. Biol. 21 (2001) 6738–6747, https://doi.org/10.1128/MCB.21.20.6738-6747.2001.

[288] B. Sjogren, S. Swaney, R.R. Neubig, FBXO44-mediated degradation of RGS2 protein uniquely depends on a Cullin 4B/DDB1 complex, PLoS One 10 (2015) e0123581, , https://doi.org/10.1371/journal.pone.0123581.

[289] T.Y. Kim, et al., CRL4A-FBXW5-mediated degradation of DLC1 Rho GTPase-activating protein tumor suppressor promotes non-small cell lung cancer cell growth, Proc. Natl. Acad. Sci. U. S. A. 110 (2013) 16868–16873, https://doi.org/10.1073/pnas.1306358110.

[290] J. Hu, et al., WD40 protein FBW5 promotes ubiquitination of tumor suppressor TSC2 by DDB1-CUL4-ROC1 ligase, Genes Dev. 22 (2008) 866–871, https://doi.org/10.1101/gad.1624008.

[291] Y. Ohno, et al., Hoxb4 transduction down-regulates Geminin protein, providing hematopoietic stem and progenitor cells with proliferation potential, Proc. Natl. Acad. Sci. U. S. A. 107 (2010) 21529–21534, https://doi.org/10.1073/pnas.1011054107.

[292] J. Mouysset, et al., CRL4(RBBP7) is required for efficient CENP-A deposition at centromeres, J. Cell Sci. 128 (2015) 1732–1745, https://doi.org/10.1242/jcs.162305.

[293] J.L. Parsons, et al., Phosphorylation of PNKP by ATM prevents its proteasomal degradation and enhances resistance to oxidative stress, Nucleic Acids Res. 40 (2012) 11404–11415, https://doi.org/10.1093/nar/gks909.

[294] M. Zeng, et al., CRL4(Wdr70) regulates H2B monoubiquitination and facilitates Exo1-dependent resection, Nat. Commun. 7 (2016) 11364, https://doi.org/10.1038/ncomms11364.

[295] A. Thirunavukarasou, et al., Cullin 4A and 4B ubiquitin ligases interact with gamma-tubulin and induce its polyubiquitination, Mol. Cell. Biochem. 401 (2015) 219–228, https://doi.org/10.1007/s11010-014-2309-7.

[296] J. Yi, et al., DNA damage-induced activation of CUL4B targets HUWE1 for proteasomal degradation, Nucleic Acids Res. 43 (2015) 4579–4590, https://doi.org/10.1093/nar/gkv325.

[297] M. Antonioli, et al., AMBRA1 interplay with cullin E3 ubiquitin ligases regulates autophagy dynamics, Dev. Cell 31 (2014) 734–746, https://doi.org/10.1016/j.devcel.2014.11.013.

[298] Z. Wei, et al., CUL4B impedes stress-induced cellular senescence by dampening a p53-reactive oxygen species positive feedback loop, Free Radic. Biol. Med. 79 (2015) 1–13, https://doi.org/10.1016/j.freeradbiomed.2014.11.010.

[299] Y. Miyauchi-Nanri, S. Mukai, K. Kuroda, Y. Fujiki, CUL4A-DDB1-Rbx1 E3 ligase controls the quality of the PTS2 receptor Pex7p, Biochem. J. 463 (2014) 65–74, https://doi.org/10.1042/BJ20130861.

[300] F. He, et al., X-linked intellectual disability gene CUL4B targets Jab1/CSN5 for degradation and regulates bone morphogenetic protein signaling, Biochim. Biophys. Acta 1832 (2013) 595–605, https://doi.org/10.1016/j.bbadis.2013.01.015.

[301] M. Malatesta, et al., The Cul4A-DDB1 E3 ubiquitin ligase complex represses p73 transcriptional activity, Oncogene 32 (2013) 4721–4726, https://doi.org/10.1038/onc.2012.463.

[302] Z. Wu, et al., Targeted ubiquitination and degradation of G-protein-coupled receptor kinase 5 by the DDB1-CUL4 ubiquitin ligase complex, PLoS One 7 (2012) e43997, , https://doi.org/10.1371/journal.pone.0043997.

[303] Z. Shen, S.G. Prasanth, Orc2 protects ORCA from ubiquitin-mediated degradation, Cell Cycle 11 (2012) 3578–3589, https://doi.org/10.4161/cc.21870.

[304] X. Li, et al., Cullin 4B protein ubiquitin ligase targets peroxiredoxin III for degradation, J. Biol. Chem. 286 (2011) 32344–32354, https://doi.org/10.1074/jbc.M111.249003.

[305] L. Jiang, R. Rong, M.S. Sheikh, Y. Huang, Cullin-4A.DNA damage-binding protein 1 E3 ligase complex targets tumor suppressor RASSF1A for degradation during mitosis, J. Biol. Chem. 286 (2011) 6971–6978, https://doi.org/10.1074/jbc.M110.186494.

[306] Y. Zou, et al., Characterization of nuclear localization signal in the N terminus of CUL4B and its essential role in cyclin E degradation and cell cycle progression, J. Biol. Chem. 284 (2009) 33320–33332, https://doi.org/10.1074/jbc.M109.050427.

[307] V. Leung-Pineda, J. Huh, H. Piwnica-Worms, DDB1 targets Chk1 to the Cul4 E3 ligase complex in normal cycling cells and in cells experiencing replication stress, Cancer Res. 69 (2009) 2630–2637, https://doi.org/10.1158/0008-5472.CAN-08-3382.

[308] H. Wang, et al., Histone H3 and H4 ubiquitylation by the CUL4-DDB-ROC1 ubiquitin ligase facilitates cellular response to DNA damage, Mol. Cell 22 (2006) 383–394, https://doi.org/10.1016/j.molcel.2006.03.035.

[309] S. Katiyar, et al., REDD1, an inhibitor of mTOR signalling, is regulated by the CUL4A-DDB1 ubiquitin ligase, EMBO Rep. 10 (2009) 866–872, https://doi.org/10.1038/embor.2009.93.

[310] Y. Yang, S. Wang, J. Li, S. Qi, D. Zhang, CUL4A as a marker and potential therapeutic target in multiple myeloma, Tumour Biol. 39 (2017), https://doi.org/10.1177/1010428317703923.

[311] X. Han, et al., CUL4A functions as an oncogene in ovarian cancer and is directly regulated by miR-494, Biochem. Biophys. Res. Commun. 480 (2016) 675–681, https://doi.org/10.1016/j.bbrc.2016.10.114.

[312] X. Li, R. Xu, H. Liu, K. Fang, CUL4A expression in pediatric osteosarcoma tissues and its effect on cell growth in osteosarcoma cells, Tumour Biol. 37 (2016) 8139–8144, https://doi.org/10.1007/s13277-015-4715-1.

[313] J. Song, J. Zhang, J. Shao, Knockdown of CUL4A inhibits invasion and induces apoptosis in osteosarcoma cells, Int. J. Immunopathol. Pharmacol. 28 (2015) 263–269, https://doi.org/10.1177/0394632015586656.

[314] K. Yasui, et al., TFDP1, CUL4A, and CDC16 identified as targets for amplification at 13q34 in hepatocellular carcinomas, Hepatology 35 (2002) 1476–1484, https://doi.org/10.1053/jhep.2002.33683.

[315] Y.L. Yang, et al., Cul4A overexpression associated with Gli1 expression in

malignant pleural mesothelioma, J. Cell. Mol. Med. 19 (2015) 2385–2396, https://doi.org/10.1111/jcmm.12620.
[316] M.S. Hung, et al., Cul4A is an oncogene in malignant pleural mesothelioma, J. Cell. Mol. Med. 15 (2011) 350–358, https://doi.org/10.1111/j.1582-4934.2009.00971.x.
[317] Y. Xu, Y. Wang, G. Ma, Q. Wang, G. Wei, CUL4A is overexpressed in human pituitary adenomas and regulates pituitary tumor cell proliferation, J. Neuro-Oncol. 116 (2014) 625–632, https://doi.org/10.1007/s11060-013-1349-2.
[318] S. Ren, et al., Oncogenic CUL4A determines the response to thalidomide treatment in prostate cancer, J Mol Med (Berl) 90 (2012) 1121–1132, https://doi.org/10.1007/s00109-012-0885-0.
[319] J. Dong, X.Q. Wang, J.J. Yao, G. Li, X.G. Li, Decreased CUL4B expression inhibits malignant proliferation of glioma in vitro and in vivo, Eur. Rev. Med. Pharmacol. Sci. 19 (2015) 1013–1021.
[320] H. Hu, et al., CRL4B catalyzes H2AK119 monoubiquitination and coordinates with PRC2 to promote tumorigenesis, Cancer Cell 22 (2012) 781–795, https://doi.org/10.1016/j.ccr.2012.10.024.
[321] Z. Liu, et al., AhR expression is increased in hepatocellular carcinoma, J. Mol. Histol. 44 (2013) 455–461, https://doi.org/10.1007/s10735-013-9495-6.
[322] C. Mian, et al., AHR over-expression in papillary thyroid carcinoma: clinical and molecular assessments in a series of Italian acromegalic patients with a long-term follow-up, PLoS One 9 (2014) e101560, , https://doi.org/10.1371/journal.pone.0101560.
[323] X.M. Gu, et al., Expression and prognostic relevance of centromere protein a in primary osteosarcoma, Pathol. Res. Pract. 210 (2014) 228–233, https://doi.org/10.1016/j.prp.2013.12.007.
[324] Y. Lu, et al., The F-box protein FBXO44 mediates BRCA1 ubiquitination and degradation, J. Biol. Chem. 287 (2012) 41014–41022, https://doi.org/10.1074/jbc.M112.407106.
[325] Y.J. Kang, M.K. Lu, K.L. Guan, The TSC1 and TSC2 tumor suppressors are required for proper ER stress response and protect cells from ER stress-induced apoptosis, Cell Death Differ. 18 (2011) 133–144, https://doi.org/10.1038/cdd.2010.82.
[326] S.K. Halder, et al., Oncogenic function of a novel WD-domain protein, STRAP, in human carcinogenesis, Cancer Res. 66 (2006) 6156–6166, https://doi.org/10.1158/0008-5472.CAN-05-3261.
[327] S. Umeda, et al., Prognostic significance of HOXB4 in de novo acute myeloid leukemia, Hematology 17 (2012) 125–131, https://doi.org/10.1179/102453312X13376952196250.
[328] K. Koller, et al., Identification of the transcription factor HOXB4 as a novel target of miR-23a, Genes Chromosomes Cancer 52 (2013) 709–715, https://doi.org/10.1002/gcc.22066.
[329] Z. Blanchard, et al., Geminin overexpression promotes imatinib sensitive breast cancer: a novel treatment approach for aggressive breast cancers, including a subset of triple negative, PLoS One 9 (2014) e95663, , https://doi.org/10.1371/journal.pone.0095663.
[330] K. Zhang, et al., Loss of H2B monoubiquitination is associated with poor-differentiation and enhanced malignancy of lung adenocarcinoma, Int. J. Cancer 141 (2017) 766–777, https://doi.org/10.1002/ijc.30769.
[331] M.E. Matyskiela, et al., A Cereblon Modulator (CC-220) with improved degradation of Ikaros and Aiolos, J. Med. Chem. (2017), https://doi.org/10.1021/acs.jmedchem.6b01921.
[332] H. Lee, D. Puppala, E.Y. Choi, H. Swanson, K.B. Kim, Targeted degradation of the aryl hydrocarbon receptor by the PROTAC approach: a useful chemical genetic tool, Chembiochem 8 (2007) 2058–2062, https://doi.org/10.1002/cbic.200700438.
[333] D. Puppala, H. Lee, K.B. Kim, H.I. Swanson, Development of an aryl hydrocarbon receptor antagonist using the proteolysis-targeting chimeric molecules approach: a potential tool for chemoprevention, Mol. Pharmacol. 73 (2008) 1064–1071, https://doi.org/10.1124/mol.107.040840.
[334] B. Sun, et al., BET protein proteolysis targeting chimera (PROTAC) exerts potent lethal activity against mantle cell lymphoma cells, Leukemia (2017), https://doi.org/10.1038/leu.2017.207.
[335] A.P. Crew, et al., Identification and characterization of Von Hippel-Lindau-recruiting proteolysis targeting Chimeras (PROTACs) of TANK-binding kinase 1, J. Med. Chem. (2017), https://doi.org/10.1021/acs.jmedchem.7b00635.
[336] S. Feng, Z. Cao, X. Wang, Role of aryl hydrocarbon receptor in cancer, Biochim. Biophys. Acta 1836 (2013) 197–210, https://doi.org/10.1016/j.bbcan.2013.05.001.
[337] J.M. Spangle, T.M. Roberts, J.J. Zhao, The emerging role of PI3K/AKT-mediated epigenetic regulation in cancer, Biochim. Biophys. Acta 1868 (2017) 123–131, https://doi.org/10.1016/j.bbcan.2017.03.002.
[338] T. Sato, N. Gotoh, The FRS2 family of docking/scaffolding adaptor proteins as therapeutic targets of cancer treatment, Expert Opin. Ther. Targets 13 (2009) 689–700, https://doi.org/10.1517/14728220902942330.
[339] S.Q. Yin, J.J. Wang, C.M. Zhang, Z.P. Liu, The development of MetAP-2 inhibitors in cancer treatment, Curr. Med. Chem. 19 (2012) 1021–1035.
[340] O. Hantschel, G. Superti-Furga, Regulation of the c-Abl and Bcr-Abl tyrosine kinases, Nat. Rev. Mol. Cell Biol. 5 (2004) 33–44, https://doi.org/10.1038/nrm1280.
[341] F. Morales, A. Giordano, Overview of CDK9 as a target in cancer research, Cell Cycle 15 (2016) 519–527, https://doi.org/10.1080/15384101.2016.1138186.
[342] A.A. Samatar, P.I. Poulikakos, Targeting RAS-ERK signalling in cancer: promises and challenges, Nat. Rev. Drug Discov. 13 (2014) 928–942, https://doi.org/10.1038/nrd4281.
[343] H.S. Ranhotra, Estrogen-related receptor alpha and cancer: axis of evil, J. Recept. Signal Transduct. Res. 35 (2015) 505–508, https://doi.org/10.3109/10799893.2015.1049362.
[344] L. Altucci, M.D. Leibowitz, K.M. Ogilvie, A.R. de Lera, H. Gronemeyer, RAR and RXR modulation in cancer and metabolic disease, Nat. Rev. Drug Discov. 6 (2007) 793–810, https://doi.org/10.1038/nrd2397.
[345] S. Wu, T. Kanda, S. Nakamoto, F. Imazeki, O. Yokosuka, Knockdown of receptor-interacting serine/threonine protein kinase-2 (RIPK2) affects EMT-associated gene expression in human hepatoma cells, Anticancer Res. 32 (2012) 3775–3783.
[346] S. Bruzzone, et al., Rejuvenating sirtuins: the rise of a new family of cancer drug targets, Curr. Pharm. Des. 19 (2013) 614–623.
[347] W. Chen, et al., TBK1 promote bladder cancer cell proliferation and migration via Akt signaling, J. Cancer 8 (2017) 1892–1899, https://doi.org/10.7150/jca.17638.