Therapeutic targeting of “undruggable” MYC

Victor Llombart,a and Marc R Mansourab,*

aUCL Cancer Institute, University College London, Department of Haematology, London WC1E 6DD, UK
bUCL Great Ormond Street Institute of Child Health, Developmental Biology and Cancer, London, UK

Summary
c-MYC controls global gene expression and regulates cell proliferation, cell differentiation, cell cycle, metabolism and apoptosis. According to some estimates, MYC is dysregulated in ≈70% of human cancers and strong evidence implicates aberrantly expressed MYC in both tumor initiation and maintenance. In vivo studies show that MYC inhibition elicits a prominent anti-proliferative effect and sustained tumor regression while any alteration on healthy tissue remains reversible. This opens an exploitable window for treatment that makes MYC one of the most appealing therapeutic targets for cancer drug development. This review describes the main functional and structural features of the protein structure of MYC and provides a general overview of the most relevant or recently identified interactors that modulate MYC oncogenic activity. This review also summarizes the different approaches aiming to abrogate MYC oncogenic function, with a particular focus on the prototype inhibitors designed for the direct and indirect targeting of MYC.

Introduction

The oncoprotein MYC is a pleiotropic transcription factor that modulates global gene expression and regulates critical cellular processes including proliferation, differentiation, cell cycle, metabolism and apoptosis. Strong evidence supports aberrant MYC expression as a driver of both tumor initiation and maintenance and is associated with all the “hallmark” features of cancer. The role of MYC as a general or specific transcription factor has been traditionally controversial, although recent studies further support the notion that MYC is a general amplifier of highly expressed genes. The MYC oncogene family includes c-MYC, MYCN (N-MYC) and MYCL (L-MYC). All three paralogs have a similar function but show distinct expression timings and tissue specificities during development. c-MYC is ubiquitously expressed during tissue development and in a broad variety of tumors. N-MYC is expressed in neural tissue and early hematopoietic development and is deregulated in different cancer types including neuroblastoma, rhabdomyosarcoma, medulloblastoma, Wilms tumor or retinoblastoma although it can functionally replace c-MYC in some contexts. L-MYC is expressed in lungs and particularly overexpressed in small cell lung carcinomas although it is considered to have lower transforming activity. MYC activity is usually tightly controlled at the transcriptional and protein level, but is estimated to be aberrantly expressed in up to 70% of human cancers, many of which are highly aggressive (e.g. acute leukemia and high-grade lymphoma) and/or show poor response to treatment (e.g., small-cell lung carcinoma and neuroblastoma). In support of the oncogene addiction model, it has been demonstrated that transient MYC inhibition in vivo results in sustained tumor regression by promoting proliferative arrest, differentiation, apoptosis and cellular senescence in cancer cells. Notably, the anti-proliferative effects on normal tissue remain minimal and reversible. Furthermore, MYC has an unforeseen major role in enabling tumors to escape immunosurveillance through various mechanisms including decrease of MHCI expression or upregulation of inhibitory cytokines and immune checkpoint proteins such as PD-L1 and CD47, providing a compelling rational for combining MYC inhibition and immune checkpoint blockade. Together, these data suggest small molecules targeting MYC have the potential to exploit a clinically meaningful therapeutic window making MYC one of the most enticing targets for cancer drug development.

Several approaches have attempted to inhibit MYC directly or indirectly at all levels of its regulation. Some strategies that have shown promising results in vivo have focused on preventing MYC gene expression using antisense oligonucleotides (ASOs) that target MYC mRNA or G4-quadruplex binders that stabilize these DNA secondary structures. Other strategies are based on inhibiting MYC synthetic lethal genes required for tumorigenesis or tumor maintenance such as BUD31.
NUAK1 (ARK5) or eIF4F. Other approaches receiving interest aim to reduce MYC stability and function at the protein level. However, despite massive efforts, targeting MYC with clinical grade small molecules still represents an intractable challenge, particularly at the protein level. The fact that important functional domains of MYC are intrinsically disordered and lack an enzymatically-active site, has precluded structure-guided drug design. In addition, the high affinity interaction between MYC and its obligate partner MAX along with the partial functional redundancy of the different MYC family members and their nuclear localization pose an enduring obstacle for the design of effective MYC inhibitors.

This review describes the main functional and structural features of the MYC protein, delineates the most relevant or recently identified MYC cofactors that modulate its oncogenic activity and summarizes recent advances on targeting MYC using small molecule inhibitors.

**MYC structure**

c-MYC (hereafter MYC) is a 439 amino acid oncoprotein that contains a well characterized C-terminal DNA-binding and an N-terminal transactivation domain (TAD). The C-terminal region (~100 residues, comprises a basic helix-loop-helix leucine zipper (bHLH-LZ) segment that regulates the heterodimerization between MYC and its bHLH-LZ partner MAX, mediating its binding to gene promoters. In contrast, the TAD is comprised of residues 1-143, conforming an intrinsically disordered domain that regulates MYC’s biological activity and is essential for MYC-mediated transcriptional activation. This intrinsically disordered nature of MYC structure has been linked to the formation of transcriptional condensates at super-enhancer sites and may represent, if confirmed, a previously overlooked opportunity to tackling MYC with certain antineoplastic agents that can be preferentially partitioned into such phase-separated condensates.

The protein sequence of MYC contains several evolutionarily-conserved segments including the so called MYC homology boxes (MB), that modulate MYC function and are mostly shared amongst all MYC paralogs, L-MYC and N-MYC. Each MB shows distinct, sometimes cell dependent, contributions to MYC activity although functional redundancies have been described. MBs are often involved in highly
dynamic protein-protein interactions with a wide number of cofactors that contribute to modulate MYC’s function as a master regulator (reviewed in27) (Figure 3). These interactions can be multivalent and occasionally involve distal binding sites outside these conserved domains. In fact, on many occasions MBs do not function as isolated regulatory entities and instead establish dynamic interactions that define complex patterns of cooperation modulating important aspects of the protein.

N-terminal Transactivation Domain

The N-terminal TAD of MYC comprises MB0, MBI and MBII subdomains involved in the regulation of the transcriptional activity and stability of the protein.26,28-31

MBI contains the canonical phosphodegron composed by residues S62 and T58 that regulates MYC protein stability through a sequential and hierarchical phosphorylation process. The residue S62 is first phosphorylated by RAS/MEK/ERK/CDK2 priming the subsequent phosphorylation by GSK3β at T58, followed by the dephosphorylation of S62 mediated by PIN-1 and PP2A (reviewed in32). Recent findings demonstrated the phosphorylation of MYC at S67, which is proximal to the S62 residue, by Aurora B Kinase counteracts GSK3β and promotes protein stability.33 These series of post-translational modifications govern the SCF{FBXW7}/Ub-proteasome-mediated degradation of MYC and ultimately has a prominent impact on its function. The deregulation of this pathway has strong effects on MYC stability and is frequently linked to malignant cell proliferation. For example, in Burkitt’s lymphoma, in which the MYC gene is translocated (t(8:14)), T58 in MBI is recurrently mutated, leading to protein stabilization.34

In addition, SCFFBXW7 is one of the most commonly

---

**Figure 2.** Structure of MYC protein. A) Intrinsically disordered region prediction for c-MYC (predictor tool PONDR). B) Organization of the main protein domains and conservation across different MYC super-family members. C) X-ray structure of MYC:MAX heterodimer binding DNA (PDB ID: 1NKP).
mutated proteins of the ubiquitin-proteasome system in human cancer.\textsuperscript{15}

MBII (residues 129-143) represents the most widely studied segment within the MYC TAD.\textsuperscript{26} This domain acts as a binding hub for a myriad of critical interactors (Figure 3) and is considered fundamental for MYC biological activity.\textsuperscript{31} In vivo, MBII is also indispensable for full MYC oncogenicity.\textsuperscript{30}

Central region

The MBIII regulates protein stability and contributes to MYC cell transformation.\textsuperscript{26,36} MBIIIb encompasses a 10-amino-acid segment that is part of a PEST domain rich in proline, glutamic acid, threonine and serine residues that expands from positions 226-270 and is involved in the regulation of MYC stability but not in ubiquitination.\textsuperscript{37} MBIV domain may only be required for transformation in some biological contexts although there is a high \textit{in vitro} assay variability amongst different cellular models. The main nuclear localization signal (NLS) sequence in MYC lies on residues 320-328 and, although it regulates the transport of MYC into the nucleus through the association with Importin \(\alpha\), it is not indispensable for MYC nuclear localization suggesting that other subdomains might be marginally involved in the process.\textsuperscript{38,39}

\begin{figure}
\centering
\includegraphics[width=\textwidth]{myc_interactors.png}
\caption{Schematic of MYC protein interactors. The minimal interaction region between both proteins is mapped. Green boxes represent activators of MYC function. Red boxes represent repressors of MYC activity.}
\end{figure}
C-terminal bHLH-LZ domain

The C-terminal region of the MYC protein represents the most widely studied domain. It contains the basic helix-loop-helix leucine zipper (bHLH-LZ) spanning residues 357-439 and plays a key role in DNA binding. At physiological levels, MYC preferentially binds at the canonical E box sequence 5'-CACGTG-3' of target gene promoters whereas at deregulated levels it also binds the open chromatin containing the more prevalent non-canonical 5'-CANNTG-3' E-boxes. The bHLH-LZ domain also stabilizes the interaction between MYC and MAX forming an obligate heterodimer that is essential for MYC binding to DNA and regulation of gene expression.41,42

The structural model of the MYC-MAX heterodimer in the apo form has been recently resolved providing important structural information of the bHLH-LZ and, in contrast to older models, avoiding the conformational changes induced by DNA binding.43-45 This new model facilitates structure-based design of drugs that could target the formation of the MYC-MAX dimer, or impede its binding to DNA, providing at the same time relevant structural information related to its interactions with other dimer-associated cofactors.

Not surprisingly, the bHLH-LZ domain is considered an attractive therapeutic target and significant efforts have been made over the last decades aiming to develop small molecule compounds that target the MYC-MAX association. Although clinical grade has not yet been achieved for such compounds, these efforts have provided important insights into MYC biology and represent the basis for the generation of improved therapeutic agents.

Targeting MYC

Direct inhibition of the MYC-MAX heterodimer and targeted degradation of MYC

The role of MAX as an obligate partner of MYC has led to significant efforts towards the development of MYC-MAX heterodimer inhibitors (Figure 2c; Table 1). Over the last decade, new chemically refined compounds such as KJ-Pyr-9 or SaJM589 have been identified with improved overall in vivo properties in comparison to the prototypic small molecules 10058-F4 and 10074-G5. SaJM589 suppressed cell proliferation in diverse cancer cell lines by disrupting MYC-MAX heterodimerization and potentially by promoting proteasome-mediated MYC degradation.43 However, to fully realize the therapeutic potential of SaJM589, in vivo studies in different cancer models are still required. Another recently identified small compound, MYCMI-6, also inhibits MYC-MAX heterodimerization by binding to the bHLH-LZ domain of MYC and abrogates MYC-mediated transcription.46 In vitro, MYCMI-6 suppresses MYC-dependent cell growth and this effect correlates with the level of MYC expression in tumor cells. The administration of MYCMI-6 in mouse bearing xenografts of SK-N-DZ neuroblastoma cells resulted in the reduction of tumor cell proliferation and microvasculature density, as well as the induction of apoptosis. In contrast to the previous small molecules that inhibit heterodimerization, the compound KSI-3716 blocks MYC-MAX binding to DNA.47 KSI-3716 is able to inhibit tumor growth in bladder murine xenograft models and has proven effective even in xenografts resistant to Gemcitabine chemotherapy.48,49

The small molecule MYGi975 was recently identified following a screen of 16 million compounds using an in silico five-point pharmacochemical model.50 MYGi975 directly binds to the HLH domain of MYC, enhancing T58 phosphorylation, in turn reducing MYC stability, transcriptional activity and cancer cell viability. In vivo, MYGi975 showed reasonable pharmacokinetic properties, with the capacity to reduce tumor growth, at the same time potentiating immune cell infiltration at the site of the tumor. Most notably, when combined with anti-PD-1 immunotherapy, MYGi975 significantly reduced tumor growth suggesting combining MYC inhibition and immune checkpoint blockade is an enticing potential therapeutic strategy to test in future trials.

Omomyc is a 90 amino acid MYC mini-mutant that comprises the bHLH-LZ domain and competes with c-MYC, N-MYC and L-MYC for binding to DNA displacing the MYC/MAX heterodimers and inhibiting transcription of target genes.50-52 The role of Omomyc as an effective dominant negative form able to inhibit MYC has been evidenced in several cellular models showing anti-proliferative and pro-apoptotic effects.53,54 In vivo, Omomyc-mediated MYC inhibition elicited a potent anti-proliferative effect and sustained tumor regression with no detrimental effect on healthy tissue, and denoted, for the first time, MYC inhibition as a feasible anti-cancer therapeutic strategy.55 Recently, Omomyc mini-protein showed cell-penetrating activity and partial localization to the nucleus reverting the expression of MYC-regulated genes,56 although the MYC/Omomyc interaction is reported to occur primarily in the cytoplasm where Omomyc can bind excess MYC monomers and prevent it entering the nucleus.57 Co-treatment with Omomyc and Paclitaxel abrogated tumor growth of human lung cancer xenografts in mice.58 Despite its short effective half-life,59 a dose-escalation phase I/II clinical trial started in 2021 making OMO-103 (Omomyc) the first direct MYC inhibitor to reach clinical phase studies in patients with advanced solid tumors including non-small cell lung, colorectal and triple-negative breast cancer.

Omomyc inhibits MYC in part by promoting its proteasomal degradation.60 In recent years, the notion of directly targeting undruggable oncoproteins like MYC using Proteolysis-targeting chimeras (PROTACs) has grown in interest and the idea of enhancing the affinity
| Target | Compound | Malignancy | Phase | Trial number | Main adverse effects |
|--------|----------|------------|-------|--------------|----------------------|
| MYC-MAX | Omomyc | Advanced solid tumors. | Phase I/II | NCT04808362 | - |
| | KJ-Pyr-9 | Breast cancer xenografts. | Pre-clinical | | |
| | SaJMS89 | B cell line P493-6, Ramos (Burkitt's lymphoma cell line), HL-60 and KG1a (acute myeloid leukemia cell lines). | Pre-clinical | | |
| | MYCMI-6 | Neuroblastoma | Pre-clinical | | |
| | KSI-3716 | Bladder cancer xenograft. | Pre-clinical | | |
| | KI-M52-008 | T-cell acute lymphoblastic leukemia and hepatocellular carcinoma mouse models. | Pre-clinical | | |
| Aurora A kinase | MLN8237/ Alisertib | Relapsed/refractory peripheral T-cell lymphoma, non-Hodgkin lymphoma, advanced-non hematological malignancies, lung, breast, head and neck, gastrointestinal malignancies, and advanced or metastatic sarcoma. | Phase I/II/III | NCT01482962, NCT00807495, NCT01045421, NCT01653028 | Neutropenia, anemia or gastrointestinal disorders. |
| | CDS32 | Neuroblastoma xenografts. | Pre-clinical | NCT01121406 | Anemia, neutropenia and thrombocytopenia. |
| | B6727 (Volasertib) | Ovarian cancer. | Phase II | NCT01121406 | Anemia, neutropenia and thrombocytopenia. |
| PP2A | DT-061/(AZD6244) | KRAS-driven lung cancer mouse models, lung adenocarcinoma xenografts, non-small cell lung cancer xenografts. | Pre-clinical | | |
| | FTY720 | leukemia, colon cancer, non-small cell lung cancer, breast cancer, hepatocellular carcinoma, prostate cancer. | Pre-clinical | | |
| | OP449 | AML, breast cancer, and pancreatic cancer. | Pre-clinical | | |
| | Perphenazine | Brain, breast, colon, pancreatic, liver, skin and lung cancer, ovarian and oral carcinoma and leukemia and lymphoma cell lines. T-ALL and melanoma xenografts. | Pre-clinical | | |
| | LB-100 | Myelodysplastic syndromes, extensive stage lung small-cell carcinoma and astrocytoma, glioblastoma multiforme, giant cell glioblastoma, glioma and oligodendrogliomas. | Phase I/II | NCT03886662, NCT04560972, NCT03027388 | - |
| | Pin-1 | Pancreatic, lung, prostate, and breast cancer cell lines. Lung cancer xenografts. | Pre-clinical | | |
| | KPT-6566 | Advanced adenoid cystic carcinoma, pancreatic ductal adenocarcinoma, breast cancer. | Phase I/II | NCT03999684, NCT03307148, NCT04113863 | - ** |
| | ATRA | Pancreatic ductal adenocarcinoma. | Pre-clinical | NCT04113863 | |
| | BIP-06-005-3 | Neuroblastoma and pancreatic mouse model, and neuroblastoma zebrafish model. | Pre-clinical | | |
| | Sulfitop | Neuroblastoma and pancreatic mouse model, and neuroblastoma zebrafish model. | Pre-clinical | | |
| | PIM1 | AZD1208 | Phase I | NCT01489722, NCT01588548 | Guillain–Barré syndrome, increased blood |

*Table 1 (Continued)*
| Target | Compound | Malignancy | Phase | Trial number | Main adverse effects |
|--------|----------|------------|-------|--------------|----------------------|
| TP-3654 | Advanced solid tumors. | Phase I | NCT02078609 | - |
| PIM447 | Myelofibrosis, acute myeloid leukemia, high risk myelodysplastic syndrome, multiple myeloma. | Phase I | NCT01456689 | Thrombocytopenia, anemia or fatigue. |
| SGI-1776 | Refractory prostate cancer and relapsed/refractory non Hodgkin's lymphoma, relapsed/refractory leukemias, | Phase II | NCT00848601 | Cardiac toxicity. |
| SEL24/MEN1703 | Acute myeloid leukemia. | Phase I/II | NCT03008187 | - |
| SKZ-P1-41 | Prostate and long tumor xenografts. | Pre-clinical | NCT02696476 | |
| Diocin | Colorectal adenocarcinoma xenografts. | Pre-clinical | NCT02259114 | |
| FKA | Synovial sarcoma, osteosarcoma xenografts and prostate cancer in vivo models. | Pre-clinical | NCT01713582 | |
| BRD4 (and other BET proteins) | Uveal melanoma xenografts. | Phase II | NCT02698189 | Thrombocytopenia, anemia or gastrointestinal disorders. |
| SKPin C1 | Acute leukemia, solid tumors. | Phase II | NCT02696176 | |
| OTX015/MK-8628 | Pre-clinical | NCT02295114 | |
| GSK2820151 | Solid tumors. | Phase I | NCT02630251 | Eye disorders, nausea or fatigue. |
| ZEN-3694 | Metastatic castration-prostate cancer and triple-negative breast cancer. | Phase I/II | NCT02711956 | |
| CPI-0610 | Lymphoma, multiple myeloma, myelofibrosis leukemia, myelocytic, acute myelodysplastic/myeloproliferative neoplasm myelodysplastic syndrome (MDS). | Phase I/II | NCT03901469 | Gastrointestinal disorders, fatigue, anemia or thrombocytopenia. |
| GSK525762/r-BET762 | Hematological malignancies. | Phase I/II | NCT01943851 | Thrombocytopenia and gastrointestinal events. |
| INCB057643 | Any advanced malignancy. | Phase I/II | NCT01587703 | Nausea, thrombocytopenia or fatigue. |
| JQ1 | Multiple myeloma, T-ALL, B-ALL, acute myeloid leukemia cell lines. Medulloblastoma, glioblastoma, neuroblastoma, colorectal carcinoma and breast cancer cells. | Pre-clinical | NCT02711137 | |
| USP7 | Neuroblastoma and hepatocellular carcinoma xenografts. | Pre-clinical | NCT02711137 | |
| XL177A | Ewing Sarcoma, acute myeloid leukemia, and multiple myeloma cell lines. | Pre-clinical | NCT02711137 | |
| GNE-6640 | Acute myeloid leukemia cell lines and eosinophilic leukemia xenografts. | Pre-clinical | NCT02711137 | |
| GNE6776 | Eosinophilic leukemia xenografts. | Pre-clinical | NCT02711137 | |
| FT671 | Multiple myeloma xenografts. | Pre-clinical | NCT02711137 | |

Table 1 (Continued)
of MYC-specific ubiquitin ligases through heterobifunctional small molecules to promote its degradation seems plausible but remains to be shown. Some of the high-affinity molecules that target MYC could potentially be an excellent initial platform for the development of future anti-MYC PROTAC strategies. The PROTAC-mediated degradation of MYC partners, in contrast, has proven successful in pre-clinical models. The compound ARV-771 robustly degrades BET proteins in castration-resistant prostate cancer subseqently depleting the expression of c-MYC.55 Similarly, in neuroblastoma, ARV-825 targets BET proteins resulting in inhibition of both c-MYC and N-MYC.56 The bacterial protease Lon has also recently been described to directly degrade c-MYC in bladder and colon cancer murine models, presenting an additional potential tool to treat MYC-dependent tumors based on MYC degradation.57

Indirect inhibition of MYC
To date, most efforts for developing clinically applicable small molecule inhibitors of MYC have focused on targeting the MYC/MAX heterodimer, but the majority of compounds developed thus far show low cellular or in vivo potency, suboptimal pharmacological properties or unacceptable off-target effects.11 Due to the intrinsically disordered nature of its protein structure, MYC has been traditionally considered “undruggable”. However, the MYC protein presents transient secondary structure elements that become conformationally stable and can be structurally resolved when bound to other cofactors. These heterodimeric crystal models, such as those obtained for MYC-WDR558 or MYC-Aurora A,59 provide a platform for the structure-based design of MYC-inhibitors potentially with a greater specificity compared to compounds that recognize disordered polypeptides.28,60 Notably, some of these cofactors are functionally essential for MYC oncogenicity and can be potentially targeted for the indirect inhibition of MYC (Table 1: Figure 3).

Targeting MAX monomers and MAX-MAX homodimers. Apart from heterodimerizing with MYC, MAX can form homodimers or bind the proteins MGA and MXD.61 This MAX extended network antagonizes MYC function by sequestering MAX from MYC. The compound NSC13728 stabilizes MAX/MAX homodimers, thereby decreasing MYC-mediated transcription and oncogenic transformation in chicken embryonic fibroblasts.62 KI-MS2-008 is another compound that has been recently reported to stabilize the MAX-MAX homodimers in vitro.63 KI-MS2-008 reduces the expression of MYC target genes and cancer cell growth in MYC-dependent cell lines and suppresses tumor growth in mouse models of T-cell acute lymphoblastic leukemia (T-ALL) and hepatocellular carcinoma (Table 1). These encouraging results obtained with MAX stabilizers supports the feasibility of modulating MAX dimerization as an alternative to targeting the MYC-MAX heterodimer. In addition, other interactors of the extended MAX network may provide the foundations for the development of similar therapeutic strategies.

Targeting MYC post-translational modifiers. MYC protein stability is tightly controlled by the Ub-proteasome system, a process in which the MBI phosphodegron is critically but not exclusively involved. Some of the proteins implicated in the regulation of MYC protein stability are considered as potentially exploitable therapeutic targets (Table 1).
leukemia and lymphoma cells.64 Aurora A kinase prevents SCF^{FBXW7}_{-mediated MYC proteolysis by binding MBI through an interaction that relies on the phosphorylation status of T58 and S62.65,66 The inhibition of Aurora A kinase using small molecule inhibitors has been explored as a strategy to target MYC function indirectly. Compounds such as MLN8237/alisertib and CD532 render the Aurora A-c-MYC/N-MYC complex dysfunctional and promote tumor regression in neuroblastoma and human hepatocarcinoma cells.65,66 CD532 promotes N-MYC protein degradation in neuroblastoma xenografts,67 while MLN8237/alisertib showed a compelling antitumor effect in various Phase I/II clinical trials including patients with non-Hodgkin lymphoma, advanced non-hematological malignancies and metastatic sarcomas, although it is associated with significant side effects, particularly hematological.68 Unfortunately, most of the small molecule inhibitors that target this complex have not reached Phase III clinical trials, and for those that have, such as alisertib (tested in relapsed/refractory peripheral T-cell lymphoma), have not shown encouraging results.69

The Polo-Like Kinase-1 (PLK1) directly binds and phosphorylates FBXW7 promoting its autopolyubiquitination and degradation resulting in stabilization of N-MYC. In a feedforward loop, stabilized N-MYC activates PLK-1 transcription further supporting N-MYC oncogenicity. The selective inhibition of PLK-1 using BI6727 in combination with the BCL2 inhibitor ABT199 reduces neuroblastoma tumor volume in vivo supporting the clinical potential of combined PLK1/BCL2 inhibitors for MYC-over expressing malignancies.70 In a phase II clinical trial, single agent BI6727 (Volasertib) has shown anti-tumor activity in patients with ovarian cancer.71

PP2A activation and Pin-1 abrogation. PP2A is frequently inactivated in human cancers.72 The important role played by PP2A in the SCF^{FBXW7}_{-mediated degradation of MYC prompted the study of either the direct or indirect therapeutic activation of PP2A to potentially enhance the dephosphorylation of MYC pS62 and its subsequent proteasomal degradation. Small molecule activators of PP2A (SMAPs) have emerged as potential therapeutic drugs for the indirect inhibition of MYC specially in kinase-inhibition resistant tumors.72–75 In a recent study, the treatment of non-small cell lung cancer xenografts with the SMAP DT-061 suppressed tumor growth accompanied by abrogation of c-MYC protein.76 The combined treatment of DT-061 with the MEK inhibitor AZD6244, or compounds such as FTY720 and FTY720 analogues OP449 or perphenazine, have shown effective in multiple pre-clinical models and are considered promising for cancer therapeutics.72–75 Despite its role as a tumor suppressor, inhibition of PP2A-C by the small molecule inhibitor LB-100 has a potent sensitization effect for chemotherapy and radiotherapy that is currently being tested alone or in combined regimes in Phase I clinical trials including patients with myelodysplastic syndromes, small-cell lung cancer and recurrent glioblastoma.77

The proline isomerase PIN1 is essential in regulating MYC proteolysis via the Ub-proteasome system. PIN1 expression is increased in most cancers and is associated with poor outcome.80 KPT-6556 is a recently described small molecule that binds and inhibits the PIN1 active site and targets it for degradation.81 At the same time, remnant chemical species that promote DNA damage originate from the interaction between PIN1 and KPT-6556. Through this dual mechanism of action, KPT-6556 induces cell death specifically in cancer cells in vitro and limits lung metastasis growth in vivo. The retinoid ATRA, was identified following a mechanism-based screen as a new specific binder to the PIN1 active site that inhibits PIN1 function and induces its degradation in acute promyelocytic leukemia and breast cancer cells.82 Moreover, ATRA was recently reported to suppress Tamoxifen-resistant breast cancer cell growth in vitro and in tumor xenografts.83 Recently, the highly selective compound BJT-06-005-3 was designed to probe Cys113 in the PIN1 active site and suppressed cell viability in pancreatic ductal adenocarcinoma.84 However, despite being a promising tool, further chemical optimization may be needed to improve BJT-06-005-3 bioavailability and in vivo performance.85 Similarly, the compound Sulfopin has recently been reported to target Cys113 downregulating MYC-target genes and reducing tumor progression in N-MYC driven neuroblastoma in vivo, as well as pancreatic cancer models. Despite the moderate effect on tumor viability in vitro, Sulfopin shows activity and very low toxicity in vivo suggesting that higher doses or combination with other treatments might be suitable for future treatments in MYC-driven malignancies.86

Inhibition of PIM1 kinase. PIM1 promotes T58 dephosphorylation and S62/S329 phosphorylation, stabilizing the MYC protein and increasing both its oncogenic transcriptional activity and transforming capacity.87 PIM1 directly interacts with MBI and is recruited to MYC target genes where it phosphorylates nucleosomes at H3S10 contributing to MYC-dependent transcriptional activation and cellular transformation.88 PIM1, along with other PIM kinases, is aberrantly expressed in several malignancies and is considered an attractive target for therapy. Accordingly, a number of small molecule PIM inhibitors have been tested in clinical trials suggesting a potential therapeutic role.89 The small molecule AZD1208 is a pan-PIM kinase inhibitor that inhibits cell growth, induces apoptosis and sensitizes to radiation.90 AZD1208 has been tested in phase I clinical trials including acute myeloid leukemia (AML) and advanced solid tumors;91 however, despite showing on-target engagement, it lacked clinical efficacy as a
monotherapy, with a relatively high toxicity, particularly gastrointestinal and hematological.\textsuperscript{96} However, the combination of AZD1208 with other agents such as immunotherapeutics, immune checkpoint blockade or synthetic lethal targets might have potential to improve functional outcomes.\textsuperscript{97} Toxicity has also proven an issue with the PIM kinase inhibitor SGI-1776, where QT prolongation was observed on a phase I clinical trial, attributed to inhibition of the cardiac potassium channel hERG, leading to trial termination (NCT00848601). TP-3654 (SGI-9481), a second-generation pan-PIM kinase inhibitor that lacks off-target activity against hERG, has shown higher potency and lower toxicity in clinical trials for solid tumors.\textsuperscript{94} Other examples of PIM kinase inhibitors under evaluation are PIM447 which has activity in multiple myeloma, and SEL24/MEN1701, currently tested in a Phase I/II clinical trial in patients with AML.\textsuperscript{91-95} Targeting PIM kinases using small molecule inhibitors remains a promising anti-tumor strategy, and the outcome of ongoing clinical trials in terms of efficacy and toxicity are eagerly awaited.

Targeting SKP2. The E3 ligase SKP2 binds MBII along with the HLH-LZ domain, and targets MYC for ubiquitination and proteasome degradation. Distinct from SCF\textsuperscript{FBXW7}, SKP2 increases MYC turnover activating transcription of some MYC target genes.\textsuperscript{96,97} High expression of SKP2 correlates with poor outcome in multiple malignancies\textsuperscript{96} and its inactivation is considered a potential therapeutic strategy. Several SKP2 inhibitors, including compounds such as SZL-Pt-41,\textsuperscript{96} FKA\textsuperscript{100,101} or Dioscin,\textsuperscript{102} have been developed and show suppression of SKP2 oncogenic function in pre-clinical models, providing proof-of-principle for therapeutic targeting of SKP2 in human cancer therapy with associated high efficacy and safety. Further improvement on metabolic and pharmacokinetic properties of these compounds may eventually boost the development of candidates with clinical potential.

USP7 inhibition. USP7 is a multi-substrate deubiquitinating enzyme that plays a key role in proliferation and tumorigenesis, partly because it prevents the proteolysis of c-MYC and N-MYC.\textsuperscript{103} In neuroblastoma patients, USP7 overexpression is associated with a poor outcome and its inhibition in neuroblastoma xenografts by the small molecule P22077 prevents tumor growth via N-MYC destabilization suggesting the potential of USP7 inhibition as an exploitable therapeutic strategy.\textsuperscript{104} In other malignancies such as hepatocellular carcinoma, USP7 overexpression correlates with disease progression and its inhibition by P22077 also suppresses tumor growth in vivo.\textsuperscript{105} Other small molecule USP7 inhibitors such as XL177A, GNE-6640, GNE6776 or FT671 suppress the growth of different cancer cell lines and inhibit tumor growth in xenograft models.\textsuperscript{106-108} It is also worth noting that USP7 has been shown to have an oncogenic and tumor suppressor role within different subtypes of T-ALL,\textsuperscript{109,110} indicating that careful patient selection and biomarker analyses will need to be considered when USP7 inhibitors are tested in clinical trials.

Aurora B kinase. Aurora B (AURKB), a kinase that plays an important role during cell division, is frequently overexpressed in different types of cancer, including ALL and AML, and is associated with poor prognosis in patients.\textsuperscript{110} A recent study in T-ALL shows that the direct interaction between MYC and AURKB promotes leukemogenesis and depicts a model by which AURKB increases MYC stability after the phosphorylation of S67 antagonizing the binding of GSK3\textsuperscript{b}.\textsuperscript{8} In the same study, AURKB depletion reduced MYC-induced gene expression and MYC protein levels in both primary cells and cell lines. The treatment with the highly selective Aurora B inhibitor AZD1152 combined with Vincristine suppresses tumor growth in FBXW7\textsuperscript{WT} T-ALL xenografts suggesting a new therapeutic strategy for T-ALL.\textsuperscript{33} In two Phase I/II clinical trials, treatment with the compound AZD1152 resulted in a hematologic response rate in 25% of patients with AML,\textsuperscript{111} but no responses were seen in advanced solid malignancies.\textsuperscript{112} Overall, AZD1152 showed good tolerability in both studies providing promising preliminary evidence of an exploitable therapeutic window for AURKB inhibition as a potential anticancer strategy in hematological malignancies (Table 1).

The MYC, HUWE1 and MIZ1 Trimer. The transcription factor MIZ1 binds MYC through the bHLH-LZ region and abrogates MYC transcriptional activity via two mechanisms.\textsuperscript{113} The first involves the trimerization of MYC, MIZ1 and DNMT3A followed by DNA methylation and subsequent inhibition of MYC target genes.\textsuperscript{114} The second mechanism is based on the competition for MYC binding between MIZ1 and the E2 ligase HUWE1.\textsuperscript{115,116} Depending on the cellular context, HUWE1 can act as a tumor suppressor or tumor promoter by regulating the turnover of MYC/MIZ1 and balancing the ratio of both proteins at gene promoters.\textsuperscript{115,117} Due to the profound functional implication of the MYC/MIZ1 interaction on MYC activity, fostering this association via small molecule inhibition of HUWE1 has been explored as a therapy.\textsuperscript{113,115,116,118,119} The small compounds BI8622 and BI8626 are specific inhibitors of HUWE1 that enhance MYC proteolysis and suppress MYC-dependent transactivation resulting in inhibition of cell growth in colon carcinoma and multiple myeloma.\textsuperscript{116,120} In vivo, HUWE1 knock-down clearly reduces tumor burden, but both BI8622 and BI8626 have suboptimal pharmacokinetics that prevents the assessment of their in vivo efficacy in MYC-driven tumor models. Nevertheless, these pre-clinical studies support the notion that it is possible to develop effective
antitumor strategies by targeting HUWE1. It is likely that other mechanisms of regulation like MIZ1-HUWE1 are involved in the interaction between MYC and other transcription factors that act as functional repressors. Extensive characterization of these mechanisms would allow the development of novel therapeutic strategies for MYC-related malignancies.

Other MYC modifiers. Apart from SCF^FBXW7, several other E3 ubiquitin ligases and proteases, such as CRY2-FBXL3, USP28, USP7 or USP37, SCF^FBXO23, FBXO29 and RNF115 (reviewed in1^24), are involved in modulation of c-MYC and/or N-MYC stability. PIA1 is involved in SUMOylation1^24 or ubiquitination of residues K51 and K52 via an SCF^FBXW7-independent mechanism1^25,1^26 albeit the functional effect of these modifications on MYC are likely to be context dependent. All these MYC post-translational modifiers could be explored as potential therapeutic targets for MYC-driven malignancies.

Targeting of chromatin modifiers. BRD4. BRD4 is a transcriptional regulator involved in DNA replication and reparation that plays a key role in the maintenance of cell proliferation.1^25 Apart from being a well-known transcriptional modulator of the MYC and MYCN genes, BRD4 directly interacts, phosphorylates and destabilizes MYC at the protein level through the formation of a ternary complex with ERK1 creating a regulatory network that balances MYC activity.1^26

Simultaneously, MYC regulates BRD4-mediated acetylation of histones establishing a negative feedback loop that controls its own transcription. Due to its important implication in controlling MYC homeostasis, BRD4 has been intensively investigated as a therapeutic target using small compounds in MYC-driven cancers. The small molecule bromodomain inhibitor JQ1 binds BRD2/4 and blocks the association of BRD4 at acetylated histones within the MYC locus decreasing the expression of all MYC superfamily members.1^27,1^28 As a result, anti-tumor effects are observed in diverse hematological malignancies and solid tumors.1^27,1^29 JQ1 is the first-in-class compound that established proof-of-principle for BET inhibition as a cancer therapy, however, its poor in vivo pharmacokinetic properties preclude its use in clinical trials. Alternatively, numerous other improved BRD4 inhibitors such as OTX015/MK-8628, GSK2828015, ZEN-3694, CPI-0610, GSK23762/1-BET762 and INCB057643 have been developed and their safety, pharmacokinetics and activity are currently under assessment in Phase I and Phase I/II clinical trials including several cancers1^30,1^31 (Table 1). Unfortunately, none of these inhibitors have yet received regulatory approval. More studies are necessary to optimize optimal dosing schedules and to explore potential combinations with other compounds that might improve efficacy.

HDACs and TRRAP. A myriad of components of the pre-replicative complex, including CDC6,1^32 ORC1,1^33 SNF5,1^34,1^35 CBP/300 and HDACs,1^36,1^37 associate with MYC through the bHLH-LZ domain, regulating DNA replication origin in response to cellular replication stress and genomic instability.1^38,1^39 Apart from their role in gene regulation, the deacetylases HDAC1/2 also modulate MYC stability through direct Sin3b-mediated deacetylation of the MYC peptidic chain accompanied with a reduction of MYC transcriptional activity.1^40 Similarly, HDAC3 attenuates MYC-mediated gene expression and MYC-associated pro-apoptotic activity through a direct protein-protein interaction.1^35,1^37 Over-expression of HDACs is observed in different cancer types and reversing epigenetic anomalies using HDAC inhibitors is considered an enticing therapeutic strategy in cancer. Accordingly, several compounds are currently tested in clinical trials (Table 1).1^41 The compound CUDC-907 is a dual inhibitor of both HDACs and PI3K that suppresses growth in MYC-dependent cancer cells1^42 and demonstrates efficacy against prostate cancer mouse xenografts.1^43 Results of phase I/II clinical trials reported anti-tumor activity of CUDC-907 in multiple myeloma, lymphoma, thyroid cancer and in solid tumors with MYC alterations, suggesting the therapeutic potential of CUDC-907 for the treatment of diverse MYC-driven malignancies.1^35,1^44,1^45

Transformation-transactivation domain-associated protein (Trrap) is one of the best characterized MYC cofactors. Trrap is the integral subunit of the GCN5-containing and the Tip60-containing histone acetyltransferase (HAT) complexes and it is crucial for their recruitment to MYC target genes, a pivotal element in regulating transcriptional activation.1^46,1^47,1^48 Trrap also directly acetylates MYC1^49,1^50 and the association of both proteins is indispensable for MYC oncogenic activity in most contexts.1^51 The recent in-depth structural characterization of the MYC-Trrap interaction may form the basis for the development of novel MYC small molecule inhibitors.1^48

WDR5 and MYC. The WDR5-repeat protein WDR5 promotes the assembly of multiple chromatin regulatory complexes responsible of the recruitment of MYC onto chromatin. The interaction between MYC and WDR5 depends on the hydrophobic residues of the highly conserved MBIIB core EEIDVV and is important for transcription of protein synthesis-related genes, gene target recognition and for somatic cell reprogramming (Table 1).1^53 However, how WDR5-MYC interaction regulates target genes is still not completely understood and the implications of MBIIB in cell transformation remains elusive due to contradictory results from in vitro transformation assays using different MYC deleterious mutants.1^26,1^53 In vivo, blocking the MYC-WDR5 interaction is emerging as a potential anti-tumor therapeutic strategy. In mice with established Burkitt’s
lymphoma tumors, exchanging wild type MYC with a WDR5-binding defective MYC mutant promotes tumor regression, offering a potentially exploitable therapeutic window. A series of high affinity small compounds that bind the MYC-site of WDR5 targeting the interaction between both proteins have been developed in vitro and are currently under optimization to improve their drug-like properties prior to cell or animal testing. It is not only the MYC-binding site of WDR5 that is an enticing therapeutic target; the small molecule C6 binds the WIN-site of WDR5 and displaces it from chromatin with the subsequent eviction of MYC and downregulation of genes associated with protein synthesis.

Outstanding Questions
Over the last decades, MYC has moved from being considered “undruggable” into one of the most appealing therapeutic targets for cancer treatment. Despite the major challenges in directly inhibiting MYC, particularly in translating promising in vitro results to meaningful in vivo efficacy, the first direct MYC inhibitors are now starting clinical trial evaluation. Omomyc, for instance, has promising in vivo properties in pre-clinical testing and the results of early phase trials in patients with advanced solid tumors will provide valuable insights into both the efficacy and toxicity of this approach. The complex protein structure of MYC still poses a substantial challenge for development and optimization of direct MYC inhibitors. Recent advances in the in silico prediction of a protein structure based on the polypeptidic sequence may represent an important breakthrough towards the refinement of incomplete crystallographic data. Nevertheless, the higher demand on computational resources may present an important limitation for many research facilities and no in silico model is yet capable of fully deciphering the complexities of intrinsic disordered domains.

The indirect inhibition of MYC through targeting binding proteins that are critical for its oncogenicity have emerged as a promising alternative. The identification of novel key cofactors for MYC function in promoting cancer opens new exploitable ways for the indirect inhibition of MYC (Figure 4). In addition, the
identification of novel critical binding partners may facilitate our understanding of the molecular mechanisms by which MYC promotes oncogenesis and tumor maintenance providing at the same time useful information for the development of future successful therapies. However, MYC interacts with a plethora of different proteins and these interactions are usually context dependent thus the effects of indirect MYC inhibition are likely to be tumor specific.

Given the importance of MYC to tumor maintenance, it is possible that a variety of resistance mechanisms might be engaged by tumor cells to escape therapeutic targeting. For instance, mutations interfering with drug engagement might be selected for with certain small molecule inhibitors, or genomic alterations of the E3 ligase complex might occur on chronic exposure to PROTACs, as recently described for BET-PROTACs. Moreover, indirect targeting of MYC may induce highly adaptive signaling pathways, potentially associated with therapeutic resistance. In contrast, direct targeting of MYC with compounds such as Omomyc might be less likely to promote drug resistance since MYC acts as a central regulatory node of gene expression without obvious bypass pathways. The fact that c-MYC, N-MYC and L-MYC can functionally replace each other in some contexts adds an additional layer of complexity to the development of specific MYC-inhibitors, particularly as their interactomes vary. Although some small molecules have shown the ability to inhibit different MYC family members, the future development of new pan-MYC inhibitors will increase the chances of success when translating the results from bench-to-bedside. Finally, combination strategies with small molecule MYC inhibitors and immunomodulators or indirect MYC inhibitors along with other antitumoral agents should be included in future studies. However, the fact that MYC inactivation can induce cellular senescence needs to be considered when designing such combination trials, particularly those that include chemotherapeutic drugs whose efficacy is cell cycle dependent. Significant progress has been achieved to date, but further research is still required to better understand the regulation mechanism of several MYC interactors, and to bring MYC inhibitors to daily clinical practice.

Search strategy and selection criteria
Data for this Review were identified by searches of PubMed, and references from relevant articles using the search terms “MYC small molecule inhibitor”, “MYC binding protein OR MYC interactor” and “(MYC-binding protein) small molecule inhibitor”. All relevant information and reference code for clinical trials was retrieved from Clinicaltrials.gov. Abstracts and reports from meetings were excluded. Only articles published in English were included and original reports published after 2016 were prioritized highlighting more novel findings. Nevertheless, references back to 1988 have been included for contextualization.

Contributors
V. Llombart contributed on the conceptualization, data curation, funding acquisition, investigation, project administration, resources, writing of original draft and editing. M. R. Mansour contributed on the conceptualization, funding acquisition, resources, supervision, validation, and review of the manuscript.

Declaration of interests
No conflicts of interest.

Acknowledgments
This work was funded by Cancer Research UK to V.L. and to M.M.

References
1 Carabet LA, Rennie PS, Cherkasov A. Therapeutic Inhibition of Myc in Cancer. Structural Bases and Computer-Aided Drug Discovery Approaches. Int J Mol Sci 2018;20(1).
2 Hanahan D, Weinberg RA. Hallmarks of cancer: the next generation. Cell 2011;144(6):646–74.
3 Gahay M, Li Y, Felsher DW. MYC activation is a hallmark of cancer initiation and maintenance. Cold Spring Harb Perspect Med 2014;4(6).
4 Nie ZQ, Guo CH, Das SK, Chow CC, Batchelor E, Simons SS, et al. Dissecting transcriptional amplification by MYC. Elife 2020;9.
5 Zeid R, Lawlor MA, Poon E, Reyes JM, Fulciniti M, Lopez MA, et al. Enhancer invasion shapes MYCN-dependent transcriptional amplification in neuroblastoma. Nature Genetics 2018;50(4):315–21.
6 Lin CY, Loven J, Rahl PB, Paranal RM, Burge CB, Bradner JE, et al. Transcriptional Amplification in Tumor Cells with Elevated c-Myc. Cell 2012;149(5):60–67.
7 Masso-Valles D, Beaulieu ME, Soucek LM, MYCN and MYCN as therapeutic targets in lung cancer. Expert Opin Ther Targets 2020;24(1):101–14.
8 Liu Z, Chen SS, Clarke S, Veschi V, Thiele CJ. Targeting MYCN in Pediatric and Adult Cancers. Front Oncol 2020;10:53679.
9 Felsher DW. MYC Inactivation Elicits Oncogene Addiction through Both Tumor Cell-Intrinsic and Host-Dependent Mechanisms. Genes Cancer 2010;1(6):597–604.
10 Soucek L, Whitfield J, Martins CP, Finch AJ, Murphy DJ, Sodir NM, et al. Modelling Myc inhibition as a cancer therapy. Nature 2008;453(7194):579–83.
11 Han H, Jain AD, Truica MI, Izquierdo-Ferrer J, Aaker JF, Lysy B, et al. Small-Molecule MYC Inhibitors Suppress Tumor Growth and Enhance Immunotherapy. Cancer Cell 2019;36(1):48–57.e15.
12 Dhanaasekaran R, Deutzmann A, Mahauad-Fernandez WD, Hansen AS, Gouw AM, Felsher DW. The MYC oncogene - the grand orchestrator of cancer growth and immune evasion. Nat Rev Clin Oncol 2021.
13 Schrier PI, Peitenburg LT. Relationship between myc oncogene activation and MHC class I expression. Adv Cancer Res 1995;63:241–246.
14 Dhanaasekaran R, Park J, Veytudioyken A, Bellovin DJ, Adam SJ, Kd AR, et al. MYC ASO Impedes Tumorigenesis and Elicits Oncogene Addiction in Autochthonous Transgenic Mouse Models of HCC and RCC. Mol Ther Nucleic Acids 2020;21:550–5.
Helander S, Montecchio M, Pilstal R, Su Y, Kuruvilla J, Herbst A, Hemann MT, Tworkowski KA, Salghetti SE, Lowe Kalkat M, Resetca D, Lourenco C, Chan PK, Wei Y, Shiah YJ, Tu WB, Helander S, Pilstal R, Hickman KA, Lourenco C, Duffy MJ, O'Connor R, Uhm SW, Ulbrich J, Muller J, Wustefeld T, Aeberhard L, Kress Dang CV, Lee WM. Identification of the human c-myc protein nuclear translocation signal. Mol Cell Biol 1998;18(2):404–8.

Conti E, Kuriyan J. Crystallographic analysis of the specific yet versatile recognition of distinct nucleolar localization signals by Ysc Nukrin alpha. Structure 2000;8(1):129–38.

Blackwell TK, Huang M, Ma A, Kretzner L, Alt FW, Eisenman RN, et al. Binding of myc proteins to canonical and noncanonical DNA sequences. Mol Cell Biol 1993;13(6):5216–24.

Nair SK, Burley SK. X-ray structures of Myc-Max and MadMax recognizing DNA. Molecular bases of regulation by proto-oncogenic transcription factors. Cell 2005;121(2):193–205.

Blackwood EM, Eisenman RN. Max: a helix-loop-helix zipper protein that forms a sequence-specific DNA-binding complex with Myc. Science 1993;259(5095):2141–7.

Sannikov S, Hamanishi N, Goretz F, Allen D, Freund SMV, Brycof M, et al. Crystal Structures and Nuclear Magnetic Resonance Studies of the Apo Form of the c-MYCMAX bHLHZip Complex Reveal a Helical Basic Region in the Absence of DNA. Biochemistry 2011;50(9):5144–54.

Hart JR, Garner AL, Yu J, Ito Y, Sun M, Ueno I, et al. Inhibitor of MYC identified in a Krohne pyridine library. Proc Natl Acad Sci U S A 2011;108(44):18355–61.

Choi SH, Mahankali M, Lee SJ, Häll M, Petrasri HM, Clatterjee AK, et al. Targeted Disruption of Myc-Max Oncoprotein Complex by a Small Molecule. ACS Chem Biol 2017;12(11):2757–63.

Castell A, Yan Q, Fawkner K, Hydbring P, Zhang F, Verchut V, et al. A selective high affinity MYC-binding compound inhibits MYCMAX interaction and MYC-dependent tumor cell proliferation. Sci Rep 2018;8(1):10064.

Jeong KC, Ahn KO, Yang CH. Small molecule inhibitors of c-Myc transcriptional factor suppress proliferation and induce apoptosis of promyelocytic leukemia cell via cell cycle arrest. Mol Biotechnol 2010;48(5):1509–20.

Jeong KC, Kim KT, Seo HH, Shin SP, Ahn KO, Ji MJ, et al. Intravesical instillation of c-MYC inhibitor KSI-3716 suppresses orthotopic bladder tumor growth. J Urol 2014;191(2):310–8.

Seo HK, Ahn KO, Jung NR, Shin JS, Park WS, Lee KH, et al. Antitumor activity of the c-Myc inhibitor KSI-3716 in gemicitabine-resistant bladder cancer. Oncotarget 2014;5(2):336–37.

Sousek L, Helmer-Citterich M, Sacco A, Jucker R, Cesareni G, Nasi S. Design and properties of a Myc derivative that efficiently homodimerizes. Oncogene 1998;17(9):2463–72.

Demna M, Mapelli C, Sun A, Bodea S, Ruprecht B, Javad S, et al. Oromycin: Reveals New Mechanisms To Inhibit the MYC Oncogene. Mol Cell Biol 2019;39(2).

Fiorentino FP, Tokgoz E, Sole-Sanchez S, Giampaolo S, Tokgoz O, JauDET T, et al. Growth suppression by MYC inhibition in small cell lung cancer cells with TP53 and RB1 inactivation. Oncotarget 2015;6(24):21014–28.

Annnabali D, Whitfield JR, Favuzzi E, JauDET S, Serrano E, Cuartas I, et al. MYC inhibition is effective against glioma and reveals a role for Myc in proficient mitosis. Nat Commun 2014;5(1):62.

Beaulieu ME, JauDET T, Masso-Valles D, Martinez-Martin S, Rahl P, Maltais L, et al. Intrinsic cell-penetrating activity propels Oromycin from proof of concept to viable anti-MYC therapy. Sci Transl Med 2019;11(48).

Raina K, Liu J, Qian Y, Altimiri M, Gordon D, Rossii AM, et al. PROTAC-induced BET protein degradation as a therapy for castration-resistant prostate cancer. Proc Natl Acad Sci U S A 2016;113(16):5772–9.

Li Z, Lim SI, Tao Y, Li X, Xie Y, Yang C, et al. PROTAC Brosmod domain Inhibitor ARV-845 Displays Anti-Tumor Activity in Neuroblastoma by Repressing Expression of MYCN or c-Myc. Front Oncol 2020;10:5742.

Butler DSC, Cafaro C, Putze J, Wan MLY, Tran TH, Ambite L, et al. A bacterial peptide depletes c-Myc and increases survival in mouse models of bladder and colon cancer. Nat Biotechnol 2021;39(6):754–64.

Thomas LR, Adams CM, Fesik SW, Eschen CM, Tansey WP. Targeting MYC through WDPrs. Mol Cancer 2007;6(2):1709188.

Richards MW, Burgess SS, Poon E, Cartensen A, Eilers M, Chesler L, et al. Structural basis of N-Myc binding by Aurora-A and its destabilization by kinase inhibitors. Proc Natl Acad Sci U S A 2016;113(48):15726–31.
Brockmann M, Poon E, Berry T, Carstensen A, Deubzer HE, Huang HL, Weng HY, Wang LQ, Yu CH, Huang QJ, Zhao (5):539–53.

Allen-Petersen BL, Sears RC. Mission Possible: Advances in benefit assessment of the novel anti-cancer aurora a kinase inhibitor alisertib (MLN8237): A comprehensive review of transition in Aurora kinase A. Cancer Cell 2014;26(1):1–9.

Donnell EF, Wang J, Zaware N, et al. Activation of tumor suppressor protein MYC and its interactome. Crit Rev Oncol Hematol 2017;107:1–5.

Zhang F, Song M, Kong JD, et al. PIM kinase-dependent inhibition of MYC degradation. Oncogene 2020;39(15):2019–19.

Kirschner AN, Wang J, van der Meer R, Anderson PD, Zhao S, Kozono S, Kats L, Nechama M, Li W, Guarnerio J, Wang J, van der Meer R, Anderson PD, et al. Identification of a potent and selective covalent PIM inhibitor. Nat Chem Biol 2020;16(10):1189–200.

Farrington CC, Yuan E, Mazhar S, Izadmehr S, Hurst L, Tiwana D, et al. Targeting PIM kinase in cancer cells by a dual mechanism of action. Signal Transduct Target Ther 2019;4(1):74–53.

Zhao S, Kozono S, Kats L, Nechama M, Li W, Guarnerio J, Wang J, van der Meer R, Anderson PD, et al. Identification of a potent and selective covalent PIM inhibitor. Nat Chem Biol 2020;16(10):1189–200.

Donnell EF, Wang J, van der Meer R, Anderson PD, Zhao S, Kozono S, Kats L, Nechama M, Li W, Guarnerio J, Wang J, van der Meer R, Anderson PD, et al. Identification of a potent and selective covalent PIM inhibitor. Nat Chem Biol 2020;16(10):1189–200.

Kirschner AN, Wang J, van der Meer R, Anderson PD, Zhao S, Kozono S, Kats L, Nechama M, Li W, Guarnerio J, Wang J, van der Meer R, Anderson PD, et al. Identification of a potent and selective covalent PIM inhibitor. Nat Chem Biol 2020;16(10):1189–200.
invasion and lung metastasis in osteosarcoma. Sci Rep 2018; 8(1):14294.

102. Zhao L, Yu X, Li M, Gong G, Liu W, Li T, et al. CDH1-mediated Skp2 degradation by dioxin reprogrammes aerobic glycolysis and inhibits colorectal cancer cell growth. Cell Mol Med 2020; 24:102310.

103. Bhattacharya S, Ghosh MK, HAUSP regulates c-MYC expression via de-ubiquitination of TRRAP. Cell Oncol (Dordr) 2015; 38(4):465–77.

104. Tavona O, Li D, Dai C, Lopez G, Banerjee D, Kon N, et al. HAUSP deubiquitinates and stabilizes N-Myc in neuroblas-toma. Nat Med 2016; 22(10):1186–6.

105. Zhang W, Zhang JX, Xu CZ, Zhang SQ, Bian SY, Jiang F, et al. Ubiquitin-specific protease 7 is a drugable target that promotes hepatocellular carcinoma and chemoresistance. Cancer Cell Int 2020; 20(1).

106. Schauer NJ, Liu X, Maqin RS, Doherty LM, Chan WC, Ficarro SB, et al. Selective USP9 inhibition elicits cancer cell killing through a p35-dependent mechanism. Sci Rep 2020; 10(1):1324.

107. Katayama L, Di Lellio P, Rouge L, Pastor R, Clark KR, Drummond J, et al. USP9 small-molecule inhibitors interfere with ubiquitin binding. Nature 2017; 550(7677):534–8.

108. Turnbull AP, Ioannidis S, Krajewski WW, Pinto-Fernandez Crawford LJ, Campbell DC, Morgan JJ, Lawson MA, Down Peter S, Bultinck J, Myant K, Jaenicke LA, Walz S, Muller J, et al. HECT-domain ubiquitin ligase Huwe1 small molecule inhibitors of the HUWE1 ubiquitin ligase. Cell 2017; 168(3):409–21.

109. Baek J, Kim J, Suhan J, Zhao Q, Bae J, et al. The HECT-histidine ubiquitin ligase c-Myc represses transcription through recruitment of Miz-1. Mol Cell 2011; 43(6):1344–9.

110. Bae J, Kim J, Suhan J, Zhao Q, Bae J, et al. The HECT-histidine ubiquitin ligase c-Myc represses transcription through association with Miz-1. Mol Cell 2011; 43(6):1344–9.

111. Sun Y, Han J, Wang Z, Li X, Sun Y, Hu Z. Safety and Efficacy of Bromodomain and Extra-terminal Inhibitors for the Treatment of Hematological Malignancies and Solid Tumors: A Systematic Study of Clinical Trials. Front Pharmacol. 2020; 11:52093.

112. Takayama M, Taira T, Ichiugu-Ariga SM, Ariga H. CDC6 interacts with c-Myc to inhibit E-box-dependent transcription by abrogating c-Myc/Max complex. FEBS Lett 2000; 477(1-2):41–8.

113. Takayama MA, Taira T, Tani K, Ichiugu-Ariga SM, Ariga H. ORC1 interacts with c-Myc to inhibit E-box-dependent transcription by abrogating c-Myc-SNF5/IN1 interaction. Genes Cells 2000; 5(1):54–9.

114. Cheng SW, Davies KP, Yung E, Beltran RJ, Yu J, Kalpana GV, c-MYC interacts with INI1/hSNF5 and requires the SWI/SNF complex for transcriptional activation. Nat Genet 1999;24(1):102–5.

115. Sennens T, de Klerk CM, de Klerk CM, van der Vorst APC, Pelger FM, et al. The SWI/SNF complex for transactivation of INI1 by c-Myc. Cell 2005; 121(4):497–508.

116. Calviello R, Sabatino M, Scardello FA, et al. Epigenetic modification of INI1 by c-Myc. Genes Cancer 2012; 3(6):592–9.

117. Akiyama T, Maruyama M, et al. HDAC4, HDAC5, and HDAC6 interact to enhance c-Myc-mediated transcriptional activation in colon cancer cells. Mol Cancer Ther 2006; 5(5):1559–69.

118. Dufour B, Leveille D, et al. INI1/hSNF5 interacts with c-Myc to enhance its transcriptional activity. FEBS Lett 2008; 582(2):285–9.

119. Kato F, Fiorentino FP, Albes A, Peruchio M, Sanchez-Cespedes M, Kohn T, et al. MYCL is a target of a BET bromodomain inhibitor, JQ1, on growth suppression efficacy in small cell lung cancer cells. Oncotarget 2016; 7(24):37779–88.

120. Zhou S, Zhang S, Wang L, Huang S, Yuan Y, Yang J, et al. BET protein inhibitor JQ1 downregulates chromatin accessibility and suppresses metastasis of gastric cancer via inactivation of RhoA/ROCK signaling. Oncogene 2020; 39(3):11.

121. Algimian T, Choscaur K, Ashraf M, Hammond MD, Allogbhi A, Khan T, et al. Bromodomain and extra-terminal motif inhibitors: a review of preclinical and clinical advances in cancer therapy. Future Sci OA 2019; 5(1):FSO372.

122. Kato F, Fiorentino FP, Albes A, Peruchio M, Sanchez-Cespedes M, Kohn T, et al. MYCL is a target of a BET bromodomain inhibitor, JQ1, on growth suppression efficacy in small cell lung cancer cells. Oncotarget 2016; 7(24):37779–88.

123. Delmore JE, Issa GC, Lernieux ME, Rahi PB, Shi J, Jacobs HM, et al. BET bromodomain inhibition as a therapeutic strategy to target c-Myc. Cell 2017; 161(6):904–7.

124. Faiola F, Liu X, Lo S, Pan S, Zhang K, Lymar E, et al. Dual motif inhibitors: a review of preclinical and clinical advances in cancer therapy. Future Sci OA 2019; 5(1):FSO372.

125. Sun Y, Han J, Wang Z, Li X, Sun Y, Hu Z. Safety and Efficacy of Bromodomain and Extra-terminal Inhibitors for the Treatment of Hematological Malignancies and Solid Tumors: A Systematic Study of Clinical Trials. Front Pharmacol. 2020; 11:52093.
142 Sun K, Atoyian R, Borek MA, Dellarocca S, Samson ME, Ma AW, et al. Dual HDAC and PI3K Inhibitor CUDC-907 Downregulates MYC and Suppresses Growth of MYC-dependent Cancers. Mol Cancer Ther 2017;16(2):285–99.

143 Hu C, Xia H, Bai S, Zhao J, Edwards H, Li X, et al. CUDC-907, a novel dual PI3K and HDAC inhibitor, in prostate cancer: Antitumour activity and molecular mechanism of action. J Cell Mol Med 2020;24(13):7239–53.

144 Oki Y, Kelly KR, Flinn I, Gharavi R, Ma A, et al. CUDC-907 in relapsed/refractory diffuse large B-cell lymphoma, including patients with MYC-alterations: results from an expanded phase 1 trial. Haematologica 2017;102(11):1921–30.

145 Younes A, Berdeja JG, Patel MR, Flinn I, Gerecitano JF, Neelapu SS, et al. Safety, tolerability, and preliminary activity of CUDC-907, a first-in-class, oral, dual inhibitor of HDAC and PI3K, in patients with relapsed or refractory lymphoma or multiple myeloma: an open-label, dose-escalation, phase 1 trial. Lancet Oncol 2016;17(5):622–31.

146 Liu X, Tesfai J, Evrard YA, Dent SY, Martinez E. c-Myc transformation domain recruits the human STAGA complex and requires TRRAP and GCN5 acetylase activity for transcription activation. J Biol Chem 2003;278(22):20405–12.

147 Frank SR, Parisi T, Taubert S, Fernandez P, Fuchs M, Chan HM, et al. MYC recruits the TIP60 histone acetyltransferase complex to chromatin. EMBO Rep 2003;4(6):575–80.

148 Feris EJ, Hinds JW, Cole MD. Formation of a structurally-stable conformation by the intrinsically disordered MYC-TRRAP complex. PloS One 2019;14(12):e0225984.

149 Wei Y, Rescita D, Li Z, Johannson-Akhe I, Ahlner A, Helander S, et al. Multiple direct interactions of TBP with the MYC oncoprotein. Nat Struct Mol Biol 2019;26(11):1035–43.

150 McMahon SB, Van Buskirk HA, Dugan KA, Copeland TD, Cole MD. The novel ATM-related protein TRRAP is an essential cofactor for the c-Myc and E2F oncoproteins. Cell 1998;94(3):363–74.

151 Patel JH, Du Y, Ard PG, Phillips C, Carella B, Chen CJ, et al. The c-MYC oncoprotein is a substrate of the acetyltransferases hGCN5/PCAF and Tip60. Mol Cell Biol 2004;24(14):1026–34.

152 Nikiforov MA, Chandriani S, Park J, Kotenko I, Matheos D, Johnsson A, et al. TRRAP-dependent and TRRAP-independent transcriptional activation by Myc family oncoproteins. Mol Cell Biol 2002;22(14):5054–65.

153 Thomas LR, Adams CM, Wang J, Weissmiller AM, Creighton J, Lorey SL, et al. Interaction of the oncoprotein transcription factor MYC with its chromatin cofactor WDR5 is essential for tumor maintenance. Proc Natl Acad Sci USA 2010;107(50):21526–8.

154 Macdonald JD, Chacon Simon S, Han C, Wang F, Shaw JG, Howes JE, et al. Discovery and Optimization of Salicylic Acid-Derived Sulfonamide Inhibitors of the WD Repeat-Containing Protein 3-MYC Protein-Protein Interaction. J Med Chem 2019;62(24):11322–39.

155 Aho ER, Wang J, Gogliotti RD, Howard CC, Phan J, Acharya P, et al. Displacement of WDR5 from Chromatin by a WIN Site Inhibitor with Picomolar Affinity. Cell Rep 2019;26(11):2016–28.e13.

156 Senior AW, Evans R, Juniper J, Kirkpatrick J, Sifre L, Green T, et al. Improved protein structure prediction using potentials from deep learning. Nature 2020;577(7792):706–10.

157 Zhang L, Riley-Gillis B, Vijay P, Shen Y. Acquired Resistance to BET-PROTACs (Proteolysis-Targeting Chimeras) Caused by Genomic Alterations in Core Components of E3 Ligase Complexes. Mol Cancer Ther 2019;18(7):1302–11.

158 Soucek I, Whitfield JR, Sodir NM, Masso-Valles D, Serrano E, Karnezis AN, et al. Inhibition of Myc family proteins eradicates Kras-driven lung cancer in mice. Genes Dev 2013;27(5):704–13.

159 Wu CH, van Riggelen J, Yetil A, Fan AC, Bachireddy P, Felsher DW. Cellular senescence is an important mechanism of tumor regression upon c-Myc inactivation. P Natl Acad Sci USA 2007;104(12):15028–33.