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PERSPECTIVE

The long journey to bring a Myc inhibitor to the clinic

Jonathan R. Whitfield[1] and Laura Soucek[1,2,3,4]

The oncogene *Myc* is deregulated in the majority of human tumors and drives numerous hallmarks of cancer. Despite its indisputable role in cancer development and maintenance, Myc is still undrugged. Developing a clinical inhibitor for Myc has been particularly challenging owing to its intrinsically disordered nature and lack of a binding pocket, coupled with concerns regarding potentially deleterious side effects in normal proliferating tissues. However, major breakthroughs in the development of Myc inhibitors have arisen in the last couple of years. Notably, the direct Myc inhibitor that we developed has just entered clinical trials. Celebrating this milestone, with this Perspective, we pay homage to the different strategies developed so far against Myc and all of the researchers focused on developing treatments for a target long deemed undruggable.

Introduction

2021 started with great expectations and the responsibility of holding hope for the entire world: everybody is looking forward to seeing the Sars-CoV-2 pandemic finally coming to an end, thanks to the newly developed vaccines that are competing to defeat "the beast" that has held us hostage for more than a year now and still threatens to keep us away from our normal life for a while longer. From a personal point of view, though, 2021 also comes with another huge expectation and milestone: the clinical assessment of a new first-in-class Myc inhibitor, which we have been developing for more than two decades, is finally beginning, with the hope of making a difference in the treatment of cancer patients. Indeed, in the last few decades, cancer survival has increased significantly, especially for certain oncological indications. However, there are still too many cancer deaths that we are not able to prevent. This motivates all of us to keep looking for new and effective therapeutic targets. Myc is one such target, infamous in the scientific community for a long time as the oncogene underpinning most human cancers. Nevertheless, no Myc inhibitor is yet approved for clinical use, and intense efforts around the world are underway to make this opportunity a reality for patients. We are just one of the many research groups driving toward this goal, contributing to it with the design and development of Omomyc, the best characterized direct Myc inhibitor known to date, which reached clinical trials this year.

With this Perspective, we acknowledge all those people who have not given up on the search for Myc inhibitors, and here briefly describe the multiple strategies that have been adopted so far to target Myc. Hopefully, 2021 will show a first glimpse of the potential of these approaches for cancer patient treatment.

Myc as master of all cancers

Myc is a nuclear transcription factor that coordinates intracellular and extracellular programs that allow cells to divide in an orderly manner. The Myc family consists of three functionally related genes: *c-myc* (*MYC*), *l-myc* (*MYCL*), and *n-myc* (*MYCN*; Massó-Vallés et al., 2020). *MYC* was the first gene to be discovered in this family, almost four decades ago, due to homology with the viral gene *v-myc*, carried by the avian virus myelocytomatosis (Beaulieu et al., 2020; Vennstrom et al., 1982). The other two members of the family, *MYCL* and *MYCN*, were found expressed in lung cancer and brain development, respectively. All three proteins (from now on, Myc) fall in the category of intrinsically disordered proteins, lacking a well-defined 3D structure in solution (Dang et al., 2017). However, the Myc C-terminus is characterized by the presence of a basic-loop-helix-leucine zipper domain that allows it to dimerize with the Myc-associated protein X (MAX) protein. This binding triggers a conformational change in Myc that enables binding to DNA, usually at sequences called enhancer boxes (E-boxes; Beaulieu et al., 2020; Tansey, 2014) that characterize high-affinity target genes, and at noncanonical sequences, which are instead recognized with lower affinity, especially in contexts where Myc is overexpressed (Lorenzin et al., 2016). In physiological conditions, Myc is transiently expressed to allow cells to efficiently progress through the cell cycle. However, in cancer, this regulation is lost. This alteration in Myc expression levels is rarely due to direct mutation, in contrast to what happens for other

[1]Vall d'Hebron Institute of Oncology, Edifici Cellex, Barcelona, Spain; [2]Institució Catalana de Recerca i Estudis Avançats, Barcelona, Spain; [3]Department of Biochemistry and Molecular Biology, Universitat Autònoma de Barcelona, Bellaterra, Spain; [4]Peptomyc S.L., Barcelona, Spain.

Correspondence to Laura Soucek: lsoucek@vhio.net.

common oncogenes. Instead, it is usually the consequence of upstream oncogenic signals, which all funnel through Myc in the nuclei, where it executes the transcriptional programs that ultimately lead to uncontrolled tumor growth (Dang, 2012). In this context, deregulated, tonic signaling through Myc can be as tumorigenic as elevated Myc levels (Murphy et al., 2008).

Importantly, the role of Myc in cancer is not only related to cell division. In fact, Myc has been shown to contribute to essentially all hallmarks of cancer, promoting angiogenesis, coordinating cross-talk with the tumor microenvironment (Whitfield and Soucek, 2012), blocking the antitumor immune response (Casey et al., 2016), and even conferring resistance to many standard-of-care therapies (Carabet et al., 2018).

For all these reasons, Myc is considered a particularly appealing target for cancer treatment and is currently considered one of the "most wanted" targets in cancer therapy (Dang et al., 2017). Nevertheless, no Myc inhibitor is available in the clinic yet, and the race is on to develop the first (Whitfield et al., 2017).

Challenges and strategies in targeting Myc

Designing a clinically viable Myc inhibitor has been considered extremely difficult, if not impossible, as Myc was deemed essentially "undruggable" by most of the scientific community. This connotation is mainly due to technical reasons related to its intrinsically disordered nature, but also to the preconceived notion that inhibiting it would cause severe side effects in normal proliferative tissues. From a technical point of view, standard molecule approaches have had limited success in tackling Myc with enough specificity. Such a task is complicated by the fact that Myc lacks a classic druggable enzymatic pocket and is harder to reach in the nuclear compartment, which many drugs do not enter efficiently (Beaulieu and Soucek, 2019). With regard to the fear of catastrophic side effects in normal proliferating tissues, this mainly arose from the initial study of constitutive *Myc* knockout mice, which present with embryonic lethality between embryonic day 9.5 and 10.5, with severe defects in vasculogenesis and erythropoiesis (Baudino et al., 2002). However, whether such a dramatic phenotype would be observed in adult animals was unclear until, many years later, we finally managed to demonstrate it was not the case (Soucek et al., 2008).

Here we summarize some of the best characterized strategies developed to date to inhibit Myc in vitro and in vivo and some new approaches that, in our view, hold promise for their future clinical application. For the various approaches listed herein, we can distinguish between direct and indirect strategies that focus on tackling Myc itself or its regulators, respectively. In addition, we discuss the parallel approach of synthetic lethality that targets entirely different proteins and pathways to generate lethality in cancer cells that overexpress or deregulate Myc.

Impairing Myc transcription

Direct strategy: G-quadruplex stabilizers

G-quadruplexes are four-stranded DNA structures formed in guanine-rich regions. They can act as silencer elements, repressing transcription of proximal genes, or activators (e.g., when acting instead on noncoding strands). The *Myc* promoter happens to have such an actionable structure (Yang and Hurley, 2006), and several studies have shown that some small-molecule ligands (e.g., cationic porphyrins and quindolines), such as CX-33543 or quarfloxin, can stabilize G-quadruplexes in the *Myc* promoter, resulting in Myc down-regulation (Brooks et al., 2010; Brown et al., 2011; Fig. 1 A). The phase III trials for quarfloxin were discontinued due to high albumin binding, but other G-quadruplexes are in the pipeline for further development (Asamitsu et al., 2019). One of the most advanced currently is APTO-253, developed by Aptose Biosciences, currently in a phase Ia/b trial in patients with relapsed or refractory acute myeloid leukemia or high-risk myelodysplastic syndrome. APTO-253 has been related to stabilization of G4 structures at least in telomeres, *Myc*, and *KIT* promoters (Local et al., 2018). The initial clinical trial (in 2014) was temporarily discontinued because of formulation issues, but it was recently resumed and is due to be completed in May 2022 (clinicaltrials.gov; NCT02267863).

New G4 stabilizers appearing in the last year include (a) a short peptide sequence (DM039; Minard et al., 2020); (b) a bisacridine derivative called a9, which binds with nM affinity to the Myc G4 and appears to be a dual G-quadruplex/i-motif binder effective in both oncogene replication and transcription (Kuang et al., 2020); (c) a new curcumin analogue (Pandya et al., 2021); and even (d) a PARP-1 inhibitor derived from 7-azaindole-1-carboxamide (Dallavalle et al., 2021). All of them need to be further characterized to properly assess their potential pharmacological application. For a comprehensive review of the topic, refer to Wang et al. (2020).

Indirect strategy: Bromodomain and extraterminal (BET) domain inhibitors

The BET domain inhibitors (BETis) have been found to be able to displace bromodomain chromatin regulators from gene super-enhancers. Their role is clearly not limited only to the *Myc* gene, but the first one to demonstrate potential for its inhibition was JQ1 (Fig. 1 B), initially in multiple myeloma, followed by Burkitt's lymphoma and acute myeloid leukemia, where the *Myc* gene is frequently amplified (Delmore et al., 2011; Lovén et al., 2013; Mertz et al., 2011). JQ1 and other BETis, though, clearly extend their activity beyond *Myc*, to affect multiple genes within cancer cells that might also contribute to the tumorigenic phenotype (Andrieu et al., 2016; Donato et al., 2017; Hogg et al., 2016). In fact, sensitivity to BETi displayed by various types of cancers does not show any correlation with Myc regulation. Nevertheless, despite their often poor selectivity for Myc, many BETis are currently in early phase clinical trials in various malignancies (for a review, see Alqahtani et al. [2019]) because of their generic antitumorigenic function.

Blocking Myc translation

Antisense oligonucleotides (ASOs)

One of the very early strategies used against Myc was ASOs, which, based on their sequence homology, can hybridize with and induce the degradation of *Myc* mRNA (Prochownik et al., 1988; Sklar et al., 1991; Fig. 2 A). This direct approach was effective in multiple cell lines and was already tested in the clinic

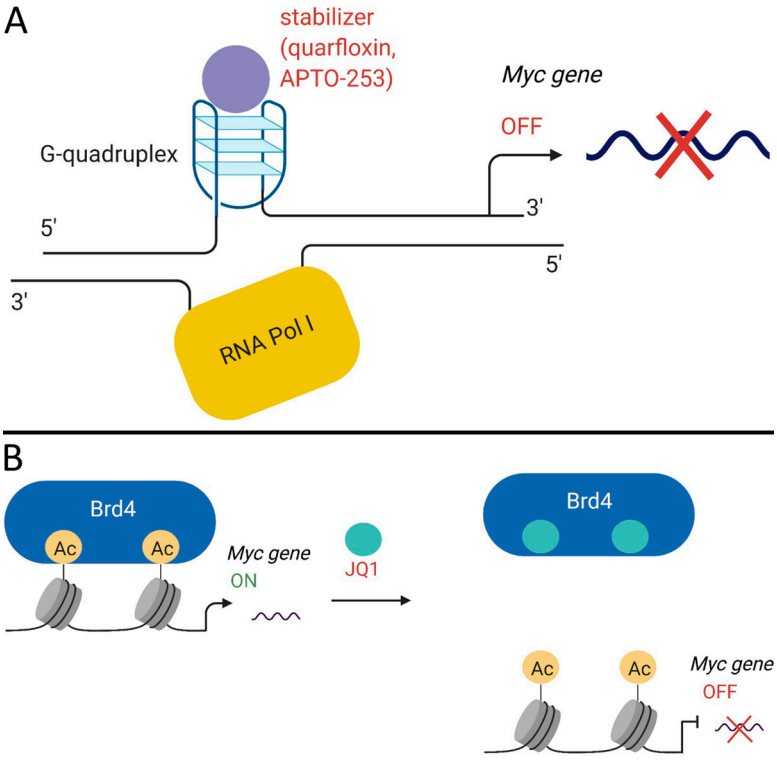

Figure 1. **Strategies to inhibit Myc transcription. (A)** Mechanism of action of G-quadruplex stabilizers on the Myc promoter. G4 stabilizers, such as quarfloxin or APTO-253, interfere with transcription of Myc by RNApol I. **(B)** Mechanism of action of BETis on Myc transcription. BETis, such as JQ1, prevent protein–protein interaction between BET proteins and acetylated histones, interfering with Myc gene transcription. Figure created with BioRender.

two decades ago, where AVI BioPharma (now Sarepta) took AVI-4126 as far as phase II clinical trials, although for coronary artery disease (Kipshidze et al., 2007). Not much information is available regarding why it has not been pursued further. Recently, the use of a Myc-specific ASO based on an oligonucleotide phosphoramidate conjugated with lipid groups has been shown to display high target specificity and was proposed as therapy in mouse models of Myc-driven hepatocellular carcinoma and renal cell carcinoma, demonstrating that new incarnations of this strategy still hold promise for potential future clinical application (Dhanasekaran et al., 2020).

siRNA or shRNA

RNA is also the tool for another therapeutic approach based on the use of siRNA or shRNA. They are usually delivered in vivo thanks to encapsulation in nanocarriers, and once inside the cells, they are both processed by Dicer and the RNA-induced silencing complex to become suitable for targeting of *Myc* mRNA before it can be translated into a functional protein (Fig. 2 B). This direct approach recently reached clinical trials sponsored by Dicerna, where it was tested as *Myc* RNAi (DCR-MYC) encapsulated in lipid nanoparticles for the treatment of patients with solid tumors, multiple myeloma, or lymphoma (Miller et al., 2020). Unfortunately, the trial did not meet expectations of *Myc* knockdown or efficacy and was discontinued. For a recent review on the subject, please refer to Habib et al. (2020).

Interference with internal ribosome entry site (IRES)-dependent translation

Myc mRNA can be translated both by 5′Cap-dependent and IRES-dependent mechanisms (Nanbru et al., 1997). Hence, to prevent *Myc* translation, one possibility is to inhibit mTOR or its upstream controllers (phosphatidylinositol 3-kinase; phosphatase and tensin homolog; protein kinase B and Ras/Raf/Mitogen-activated protein kinase, ERK kinase; and extracellular-signal-regulated kinase; Fig. 2 C). In this context, a small-molecule inhibitor of eukaryotic initiation factor-4A (eIF4A), silvestrol, showed efficacy in reducing *Myc* translation and inhibiting tumor growth (Wiegering et al., 2015). The good news is that multiple mTOR and mTORC1/2 kinase inhibitors are currently approved for clinical use (Whitfield et al., 2017), although their specific impact on Myc is dubious, since they impinge on a general cellular process involving multiple targets.

Another small-molecule inhibitor called saracatinib was also found to inhibit the ERK1/2-MNK1-eIF4E-mediated Cap-dependent translation of Myc (Jain et al., 2015), and more recently, rocaglates have shown to hold some promise in inhibiting *Myc* translation initiation in Myc-driven lymphomas through stabilization of RNA-eIF4A interaction (Zhang et al., 2020). Again, Myc is only one of their targets, making it difficult to estimate how much of their activity is actually due to Myc inhibition only.

Inhibitors of Myc dimerization and DNA binding

Small molecules

After antisense, the earliest attempts to inhibit Myc made use of small molecules to interfere with Myc/MAX interaction and/or prevent their binding to DNA (Prochownik and Vogt, 2010; Yin et al., 2003). Most small molecules, unfortunately, have been described (at least until recently) as frequently suffering from poor bioavailability (Fletcher and Prochownik, 2015; Prochownik and Vogt, 2010). However, some small-molecule Myc inhibitors selected to interfere with Myc/MAX dimerization (Fig. 3 B) have

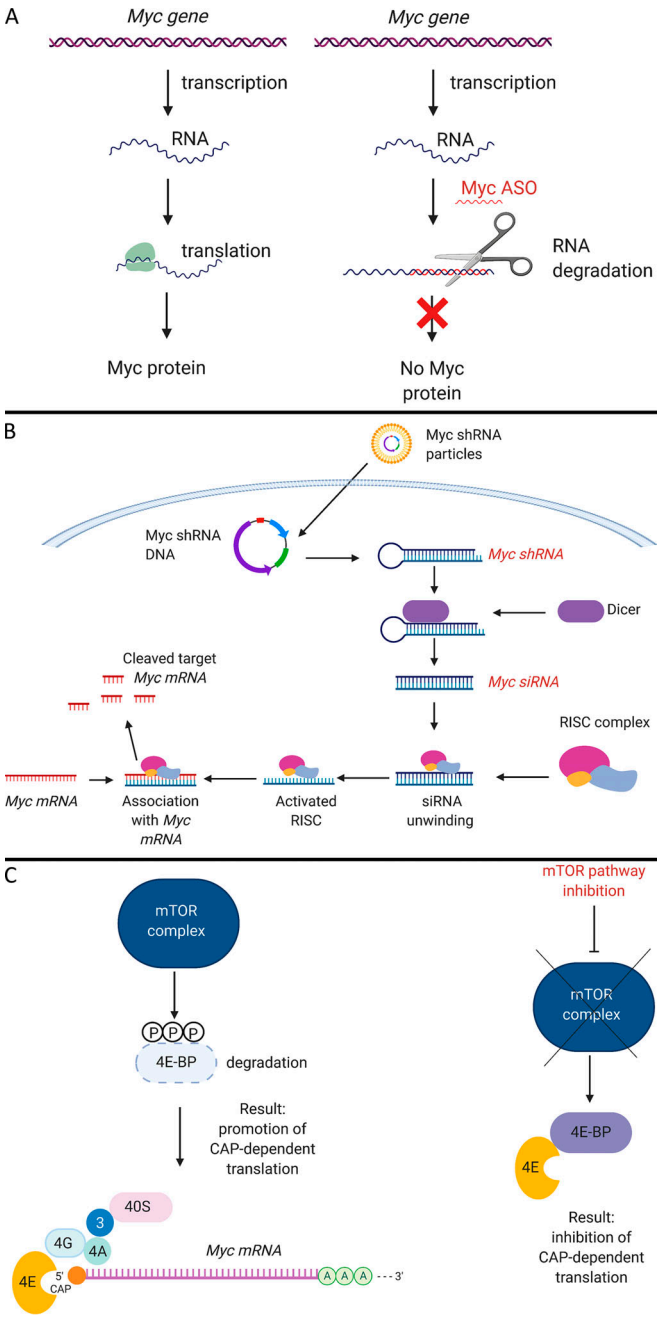

Figure 2. **Approaches to block Myc translation. (A)** Mechanism of action of Myc ASO. Myc ASO alters mRNA expression and translation through a variety of mechanisms, including decay of the pre-mRNA mediated by ribonuclease H (represented by scissors in the figure), direct steric blockage, or modulation of splicing by binding of the pre-mRNA. **(B)** Mechanism of action of Myc shRNA. Schematic of mode of action of a *Myc* shRNA, administered in the form of liposomal nanoparticles. The shRNA is processed intracellularly by Dicer and the RNA-induced silencing complex to become single-strand RNA capable of hybridizing with *Myc* mRNA and induce its degradation and/or prevent its translation into a Myc protein. **(C)** Schematic of 5′Cap-dependent *Myc* mRNA translation inhibition. mTOR pathway inhibition offers the opportunity of impairing Cap-dependent translation, the most common mechanism through which Myc protein is produced. Figure created with BioRender.

shown interesting in vivo capabilities in the last few years. Some are mentioned below.

3jc48-3. Small molecule 3jc48-3 is related to one of the earliest described compounds (10074-G5) and displayed improved activity as well as a 17-h intracellular half-life (Chauhan et al., 2014), although no further in vivo studies have been reported.

Mycro3. Mycro3 showed efficacy in vivo upon oral gavage administration, increasing survival in mouse models of pancreatic cancer (Stellas et al., 2014).

KJ-Pyr-9. KJ-Pyr-9 is an inhibitor found in a Kröhnke pyridine library; it has a very low dissociation constant (K_d; 6.5 nM) and blocks the growth of Myc-amplified human cancer cell line xenografts, even crossing the blood–brain barrier (Hart et al., 2014).

MYCMI-6. MYCMI-6 was shown to be able to inhibit tumor cell growth in a Myc-dependent manner, with half-maximal inhibitory concentrations (IC_{50}) as low as 0.5 µM, and to decrease proliferation and increase apoptosis in a Myc-driven tumor xenograft model (Castell et al., 2018). Novel computational techniques are also being used to virtually screen binding to different intrinsically disordered protein conformations, and compounds were identified with micromolar affinity for Myc (Yu et al., 2016).

MYCi975. MYCi975 was recently discovered through leveraging a large in silico library followed by a rapid in vivo screen to streamline the process of identifying small molecules already possessing tolerability and efficacy in vivo (Truica et al., 2021).

EN4. EN4 is possibly the most recent, identified by activity-based protein profiling; it is a functional covalent ligand that binds to disordered Myc domain, reduces its stability and transcriptional activity, and possesses in vivo activity, reducing tumor growth in a breast cancer xenograft model (Boike et al., 2021).

KSI-3716 and MYRA-A. Among compounds that can instead disrupt binding to DNA, KSI-3716 was effective in mouse models of bladder cancer (Jeong et al., 2014; Seo et al., 2014), and MYRA-A inhibits DNA binding of Myc family proteins without interfering with c-Myc/Max dimerization (Mo and Henriksson, 2006; Fig. 3 B).

MI1-PD. Given the number of effective small molecules out there that are hampered by in vivo delivery issues, it would also be interesting to see more attempts at incorporating these compounds into nanoparticles. To this end, MI1-PD is an integrin-targeted, lipid-encapsulated nanoparticle formulation of a Myc-MAX dimerization inhibitor that showed in vivo efficacy in a mouse model of multiple myeloma (Soodgupta et al., 2015).

KI-MS2-008. A complementary and maybe more indirect approach is instead to stabilize Myc's binding partner MAX, thus occupying DNA target sites with MAX homodimers and preventing Myc transcriptional activity (Jiang et al., 2009). Recently the compound KI-MS2-008 has been described as antagonizing Myc-dependent gene expression in cells and delaying growth of Myc-driven tumors (Struntz et al., 2019; Fig. 3 C).

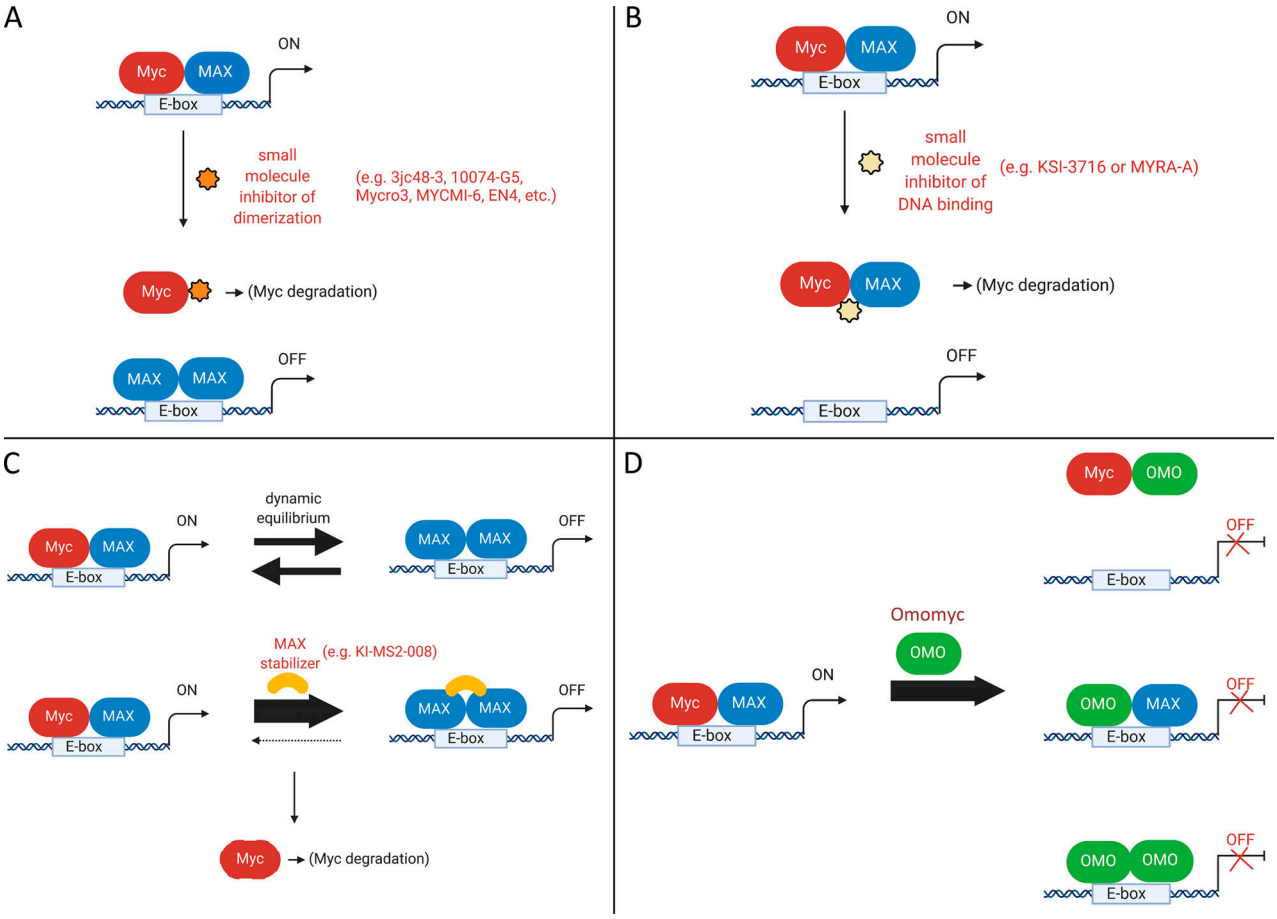

Figure 3. **Different potential mechanisms of action of small-molecule and peptide inhibitors. (A)** Schematic of the mechanism of action of a Myc/MAX dimer destabilizer. **(B)** An inhibitor of Myc/MAX binding to DNA. **(C)** A MAX/MAX dimer stabilizer. **(D)** Omomyc as Myc dominant-negative. Figure created with BioRender.

Miniproteins or protein domains

Several peptides and miniproteins based on domains from Myc family members have been described as potential therapeutic tools. The first and best characterized to date is Omomyc, a mutant Myc basic-loop-helix-leucine zipper domain that we designed and have validated over the past 20 years, that functions as a very efficient Myc dominant negative (Massó-Vallés and Soucek, 2020; Soucek et al., 1998; Soucek et al., 2002). Omomyc works through several mechanisms, forming heterodimers with Myc unable to bind DNA, and also homodimers and heterodimers with MAX, that occupy E-boxes with transcriptionally inactive complexes (Fig. 3 D). Hence, Omomyc displays two modes of action at once: it sequesters Myc away from DNA while also competing for DNA binding in the form of alternative dimers compared with Myc/MAX. It should be noted that Omomyc was initially used as a transgene in multiple mouse models of cancer to establish the proof of concept that Myc inhibition is feasible and effective and lacks significant side effects, serving as a paradigm shift for the applicability of systemic Myc inhibition in vivo (Alimova et al., 2019; Annibali et al., 2014; Duffy et al., 2021; Fiorentino et al., 2016; Sodir et al., 2011; Soucek et al., 2004; Soucek et al., 2008; Soucek et al., 2013). Recently though—and most relevant for clinical application—the recombinantly produced Omomyc miniprotein has been shown to have unexpected cell-penetrating properties and therapeutic activity in non–small-cell lung cancer, both in vitro and in vivo. Upon intranasal or intravenous administration, Omomyc reaches the tumor tissue and efficiently penetrates both cellular and nuclear membranes (Beaulieu et al., 2019). This miniprotein is being developed by Peptomyc S.L. as a drug (OMO-103) to be tested in clinical trials in 2021 (Beaulieu et al., 2019; Beaulieu and Soucek, 2019). To us, this is an extremely exciting time and we are looking forward to watching its performance in patients.

Some smaller peptides based on the Omomyc basic region have also been developed for interference with Myc/MAX binding to the E-box. Such peptides recapitulated at least one of Omomyc's mechanisms of action and have shown promise in vitro but remain to be tested in vivo (Brown et al., 2020).

Omomyc and other Myc domains have also been fused to additional sequences for targeting and delivery. Omomyc was fused with a functional penetrating "phylomer" peptide (FPPa) as a therapeutic strategy to inhibit Myc in triple-negative breast cancer (FPPa-OmoMYC; Wang et al., 2019). A similar approach was successful in delivering the Myc helix1 (H1) domain fused to an elastin-like polypeptide (ELP, a thermally responsive biopolymer that forms aggregates above a characteristic transition

temperature) and a cell-penetrating sequence (Bac) to treat a rat glioma model and an orthotopic model of breast cancer (Bac-ELP-H1; Bidwell et al., 2013; Bidwell et al., 2012). In the past year, the same H1 was fused to *Pseudomonas* exotoxin, which was employed as a peptide nuclear delivery device (PNDD) to inhibit Myc–dependent transcription at nanomolar concentrations in different tumor cell lines (PNDD1; Ting et al., 2020). However, to our knowledge, none of these approaches is close to clinical application yet.

Another miniprotein modulating the Myc network has been derived from MXD1: Mad. Like Omomyc, Mad dimerizes with MAX and binds the E-box, interfering with Myc-mediated transcription in cell models (Demma et al., 2020). Further studies will be needed to test its performance in vivo. Also, a small minimalist hybrid protein called ME47 was designed to disrupt MAX:E-box binding and block Myc transcriptional activity (Lustig et al., 2017). ME47 inhibited xenograft tumor growth upon induced expression but was not tested by direct protein delivery.

Induction of Myc degradation

In physiological conditions, Myc has a short half-life of ∼30 min in proliferating cells (Hann and Eisenman, 1984), mainly determined by its phosphorylation and consequent degradation by the ubiquitin-proteosome pathway (Sears, 2004). Hence, a potential strategy to decrease its activity in cells is to either increase the activity of ubiquitinases or interfere with deubiquitinases (Fig. 4 A). Below are some examples of these strategies, typically indirect approaches that rely on targeting molecules responsible for Myc ubiquitination or phosphorylation.

Oridonin has been associated with activation of FBXW7, which is an F-box protein that targets Myc for degradation (Huang et al., 2012; Yada et al., 2004). Some of its derivatives reached clinical trials (e.g., HAO472), although its anti-cancer activity has not necessarily been linked to Myc regulation.

MYCN has been specifically targeted because of its role in pediatric oncology and adult brain tumors, where it is often highly amplified. These approaches are described more extensively in a number of recent reviews (Borgenvik et al., 2021; Liu et al., 2021; Wolpaw et al., 2021). In this context, the role of Aurora-A kinase is particularly relevant, since it can complex with MYCN, enabling its escape from proteasomal degradation (Otto et al., 2009). Hence Aurora-A inhibitors (i.e., MLN8054 and MLN8237) have been developed to overcome this protection. Unfortunately, MLN8054 was terminated by Millennium in 2008 owing to side effects (Macarulla et al., 2010). A second-generation inhibitor, MLN8237 (Alisertib), was evaluated in multiple phase II and III studies, and some positive results were reported (Beltran et al., 2019; Falchook et al., 2019a; Falchook et al., 2019b), while others observed toxicity and disappointing responses (Mossé et al., 2019). Current trials are mostly focused on combining Alisertib with other therapeutics. Additional targeting options are also being explored, such as by chemical degradation (Adhikari et al., 2020).

Small-molecule inhibitors against another ubiquitin ligase, HUWE1, have also been shown to induce Myc and Miz1 degradation (Peter et al., 2014). These inhibitors (BI8622 and BI8626) have shown in vivo activity in multiple myeloma models (Crawford et al., 2020; Kunz et al., 2020), although in one case, Myc-dependent gene regulation was not altered (Kunz et al., 2020), and thus the involvement of Myc is not always clearly demonstrated.

Alternatively, since the phosphorylation status of Myc is key for its physiological turnover, one strategy focuses on the tumor suppressor protein phosphatase 2A (PP2A), which destabilizes Myc by targeting its serine 62 (Sears, 2004). It has been shown that cellular inhibitors of PP2A, the SE translocation (SET) oncoprotein and CIP2A, are increased in human cancers and lead to Myc stabilization (Junttila and Westermarck, 2008). Hence, inhibitors of SET (OP449) and CIP2A have been developed preclinically to reduce Myc levels and activity (Farrell et al., 2014; Janghorban et al., 2014). Once again, though, their specificity for Myc only is virtually impossible.

As a more direct approach, a recent report showed that an *Escherichia coli* protease could degrade Myc, and treatment of mouse models reduced tumor growth, notably without side effects or reduction of Myc levels in the intestine (Butler et al., 2021).

The field of "degraders" in general is growing, especially thanks to proteolysis-targeting chimeras (PROTACs) based on coupling a protein-targeting ligand with an E3 ubiquitin ligase (Sun et al., 2019; Fig. 4 C). Given the short half-life of Myc and its intrinsically disordered nature (which makes its high-affinity targeting more complicated), the applicability of this new approach to Myc is still to be demonstrated. So far, a promising result has been achieved by indirect targeting with pan-BETi PROTAC ARV-771, which reduced Myc expression and caused xenograft tumor regression in prostate cancer mouse models (Raina et al., 2016). Similarly, the BRD4 protein degrader ARV-825 reduced MYCN expression and decreased growth of neuroblastoma xenografts (Li et al., 2020), although it affects additional targets besides Myc (Saraswat et al., 2020). According to their developer, Arvinas Inc., these PROTACs are at the exploratory stage, and no clinical trials have yet been announced.

Interference with Myc transcriptional cofactors

The N-terminal segment of Myc contains the transactivating domain and several highly conserved regions termed Myc boxes (MBs; Beaulieu et al., 2020). MBII (aa 128–143), in particular, is involved in the assembly of transcriptional machinery through interaction with a wide range of transcription factors (Conacci-Sorrell et al., 2014). Although the MYC transactivating domain is inherently disordered, MBII may acquire a defined structure when complexed with these cofactors, offering an opportunity for therapeutic intervention that could significantly impair Myc transcriptional activity. Just to mention some examples: WDR5 plays a role in recruiting Myc to chromatin, and genetic disruption of WDR5/Myc interferes with tumor maintenance (Thomas et al., 2019), prompting the design of small-molecule inhibitors of this interaction. These compounds, however, still need substantial optimization to allow for their application in cell and animal studies (Chacón Simon et al., 2020; Thomas et al., 2020). The degrader approach has also been recently applied to WDR5 with encouraging results (Dolle et al., 2021).

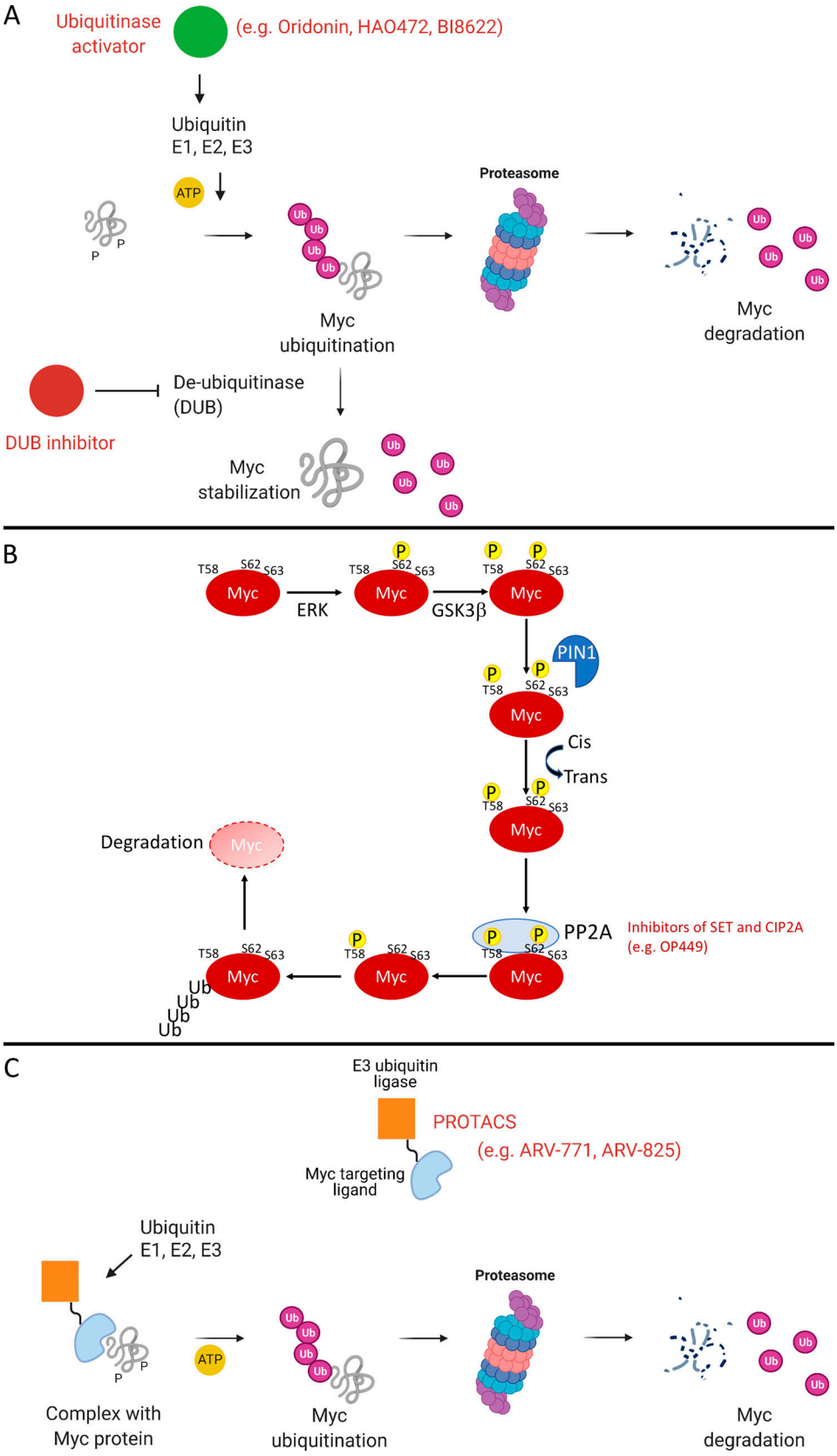

Figure 4. **Targeting Myc for degradation. (A)** Schematic of Myc degradation by modulation of the ubiquitin pathway. Myc is continuously subject to cycles of phosphorylation and dephosphorylation, strictly linked to its ubiquitination and degradation by the proteosome. This process can be enhanced by the use of ubiquitinase activators or inhibitors of deubiquitinases (DUB). **(B)** Interfering with Myc dephosphorylation to enhance degradation. **(C)** Targeting Myc for proteolytic degradation by PROTACs. Figure created with BioRender.

Another interaction that appears suitable for intervention is that of Myc/TRRAP, which has been described to have a lower free energy of association than the Myc/MAX interphase, and therefore could be a more desirable target for inhibition by small molecules (Feris et al., 2019). In this case too, development of small-molecule inhibitors is still at its beginning, and hence these approaches appear quite far from clinical application.

Synthetic lethality

In addition to the plethora of strategies described here, other indirect approaches have also been employed to get around the issues with direct Myc targeting. These include synthetic lethal approaches that could in theory target any specific dependence of Myc-driven tumor cells. Many diverse targets have already been identified, including SAE1/2 (Kessler et al., 2012), CDK2 (Hydbring and Larsson, 2010), CDK9 (Huang et al., 2014), PIM kinase (Horiuchi et al., 2016), and many more involved in metabolism (for a more complete discussion, see Thng et al. [2021]; Whitfield et al. [2017]). One advantage for this group is that novel synthetic lethal targets may already have a clinically approved inhibitor that could be quickly exploited for use in Myc-driven cancers. To date, there are few approved options, but many more are in current clinical trials (Thng et al., 2021).

Conclusion

We can all agree that this is an exciting time in the Myc inhibition world: many research groups (and now also biotechs and Big Pharma) are involved in the hunt for the first and best Myc inhibitor that can find its application in cancer patients. It has taken nearly four decades and thousands of publications to understand Myc, and there are still aspects of its biology that remain elusive. From a therapeutic point of view, it has been a long road to reach the point at which Myc inhibitors are more frequently being tested in the clinic. Our personal excitement is now specifically centered around Omomyc that we first published back in 1998 and that has been finally approved at the beginning of this year for phase I/IIa clinical trials. For us, it is a moment of truth, when this compound will show if it can keep its promise in patients as it did in mouse models.

The odds to tackle this previously undrugged and seemingly undruggable target now seem on our side. As summarized in this article, with so many different direct and indirect strategies being tested all the time, it finally feels like clinical success is fast approaching. We feel hopeful that such a historic milestone will revolutionize cancer treatment and potentially become applicable to multiple indications in oncology and beyond.

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L. Soucek is co-founder and shareholder of Peptomyc, currently conducting the clinical trials of the Omomyc mini-protein (OMO-103). J.R. Whitfield is a shareholder of Peptomyc.

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