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CDK9 Inhibitor KB-0742 Is Active in Preclinical Models of Small-Cell Lung Cancer

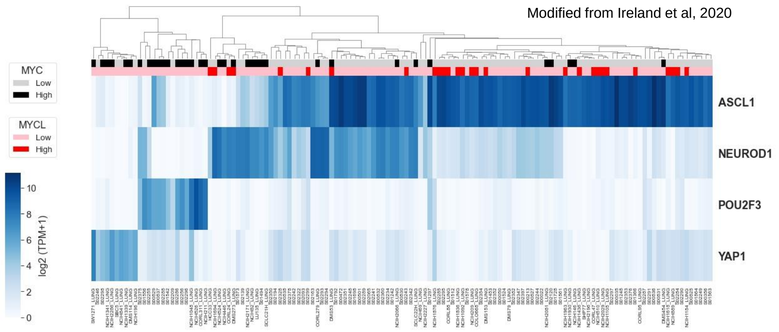
Melinda A. L. Day, Douglas C. Saffran, Tressa Hood, Nikolaus Obholzer, Akanksha Pandey, Akul Singhania, Charles Y. Lin, Pavan Kumar, Jorge DiMartino

Kronos Bio, Inc., San Mateo, CA

Abstract

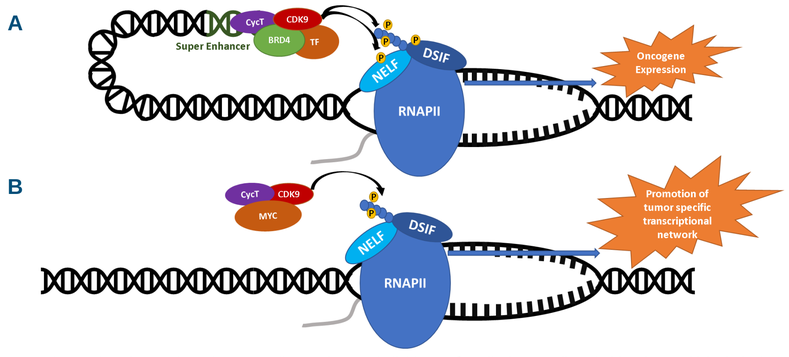
- Recent advancements in understanding the pathology of the disease has shown that small-cell lung cancer (SCLC) tumorigenesis and evolution are governed by increased expression of neuroendocrine-associated and other proto-oncogenic transcription factors. Thus, targeting transcription may be an effective therapeutic strategy. Cyclin-dependent kinase 9 (CDK9) is a serine-threonine kinase involved in transcriptional elongation through the phosphorylation of the RNA polymerase II (RNAPII), and it interacts with transcription factors to promote the activation of target genes. We developed KB-0742, a highly selective and orally bioavailable inhibitor of CDK9.
- Evaluation of KB-0742 activity in cell lines showed a correlation between *MYC* copy-number amplification (CNA) and sensitivity, with amplified lines having smaller area under the curve (AUC) values than nonamplified lines. In a panel of 6 patient-derived organoid (PDO) SCLC models with different treatment histories, KB-0742 was more active than the standard-of-care (SOC) compounds. In a separate study of 4 treatment-naive PDO models, KB-0742 was active in 3 different transcription factor-driven subtypes of SCLC, and the response correlated significantly with *c-MYC* and *MYCL* expression. Lastly, we used 4 patient-derived xenograft (PDX) models to evaluate KB-0742 activity in vivo. The tumor growth inhibition (TGI) rate ranged from 54% to 92%, with tumor regressions observed in 2 of the 4 models, and 1 model showed greater TGI with KB-0742 when compared with SOC.
- These data support the evaluation of KB-0742 as a potential treatment for SCLC. Patients with relapsed or refractory solid tumors or non-Hodgkin lymphoma are currently being enrolled in a phase 1/2 clinical trial of KB-0742 (NCT04718675) with an expansion arm for SCLC being planned after the recommended phase 2 dose is identified.

SCLC Is Governed by Proto-oncogenic Transcription Factors



SCLC comprises multiple molecular subtypes defined by the expression of specific transcription factors: ASCL1, NEUROD1, POU2F3, and YAP1. Evolution between subtypes is driven by the expression of MYC family proteins. *MYCL* is amplified/highly expressed in ASCL1-driven tumors, whereas *MYC* is amplified/highly expressed in the other subtypes.

CDK9 Is a Key Dependency in Tumor Transcriptional Reprogramming



BRD4 = bromodomain protein 4; CycT = cyclin T; DSIF = 5,6-dichloro-1-β-D-ribofuranosylbenzimidazole sensitivity-inducing factor; NELF = negative elongation factor; P = phosphate; TF = transcription factor.

As a transcriptional regulator, CDK9 is a key dependency in transcriptionally addicted tumors. CDK9 helps promote the tumor-associated transcriptional landscape through 2 mechanisms:

(A) Supporting expression of key oncogenes, and

(B) Working as a cofactor to oncogenic transcription factors, such as MYC, to promote high rates of transcription

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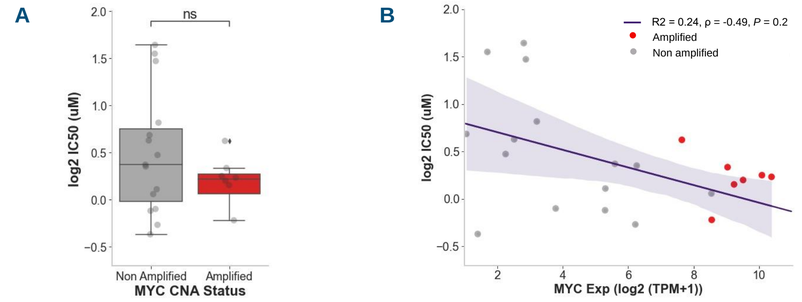
KB-0742 Sensitivity Trends With *MYC* Copy Number and Expression in SCLC



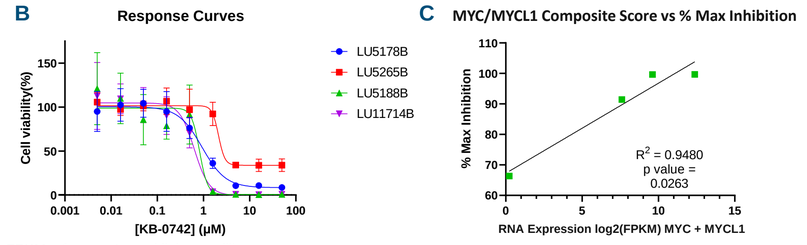
ns = not significant; TPM = transcripts per kilobase million.

KB-0742 shows greater activity in MYC-elevated cell-line models. **(A)** Twenty-four SCLC cell lines were evaluated using the Broad PRISM platform. Cell lines that had *MYC* copy-number amplifications (3 or more) trended with lower IC_{50} values when compared with nonamplified cell lines. **(B)** Higher *MYC* expression trended with IC_{50} values in the same screen. The amplified cell lines (*red dots*) clustered together, showing higher expression of *MYC* in general compared to nonamplified cell lines *(grey dots)*.

KB-0742 Activity in SCLC Organoid Models Correlates With MYC Family Expression

A

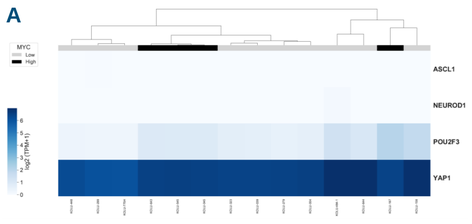
Model	Subtype	MYC Log2(FPKM)	MYCL1 Log2(FPKM)	Composite Score	IC_{50} (μM)	Max Inhibition (%)
LU5178B-P9	REST/YAP1	6.87	0.74	7.60	0.95	91.43
LU5188B-P21	NEUROD1	10.63	1.76	12.38	1.21	99.68
LU5265B-P5	ASCL1	–2	2.23	0.23	2.84	66.34
LU11714B-P5	ASCL1	0.47	9.14	9.61	0.65	99.64



FPKM = fragments per kilobase million.

KB-0742 activity in SCLC PDO models correlated with MYC protein family expression. **(A)** Four PDO models with different transcription-factor drivers were treated with a range of KB-0742 concentrations and cell viability measured using CellTiter-Glo® (Promega). **(B)** Dose-response curves of the 4 models treated with KB-0742. **(C)** Sensitivity to KB-0742 correlated with increased expression of *MYC* and *MYCL1*.

KB-0742 Was Active In Treatment Naïve and Post Treatment PDO Models of SCLC

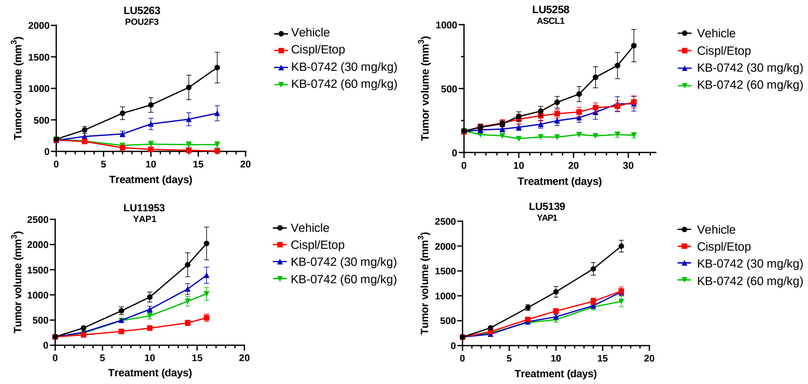


KB-0742 is active in PDO SCLC models regardless of treatment history. **(A)** SCLC PDOs were evaluated for transcription factor and MYC status. All available models were YAP1 driven. **(B)** Six of the PDO models with different treatment histories were tested for sensitivity to KB-0742. Treatment with KB-0742 resulted in over 90% maximum inhibition in 5 of the 6 models regardless of the treatment history.

B

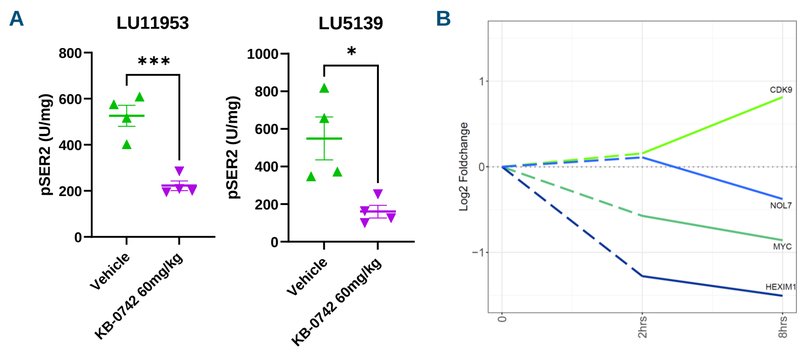
Model Number	Treatment History	MYC TPM	Subtype	Max Inhibition (%)
KOLU-045	Naive	70	YAP1	99.99
KOLU-299	Naive	30	YAP1	94.19
KOLU-448	Lobaplatin + Etoposide	30	YAP1	99.02
KOLU-775H	Cisplatin	20	YAP1	94.69
KOLU-545H	VP16 + Lobaplatin	68	YAP1	95.88
KOLU-643H	VP16 + Lobaplatin	88	YAP1	70.65

KB-0742 Is Active In Vivo



KB-0742 showed antitumor activity in 4 PDX models (different transcription-factor drivers) of SCLC. PDX models were treated with KB-0742 at 30 and 60 mg/kg on a dosing schedule of 3 days on/4 days off. KB-0742 was compared to a SOC of cisplatin plus etoposide. KB-0742 showed antitumor activity in a dose-dependent manner in all 4 models, with 60 mg/kg KB-0742 showing tumor regressions in 2 of the models. In 1 model, LU5258, KB-0742 at 60 mg/kg was more active than the SOC.

Target Engagement Observed in SCLC PDX Tumors



*P<0.05; ***P<0.001.

KB-0742 treatment reduced phosphorylation of RNAPII (pSER2) levels and altered RNA expression profiles in PDX tumors. **(A)** pSER2 protein levels were measured using a Meso Scale Discovery assay. KB-0742 60 mg/kg treatment resulted in a 50% or greater reduction after 3 days of dosing. **(B)** RNA sequencing of the LU11953 PDX tumors showed altered gene expression in key genes, including a reduction in *MYC* expression.

Conclusions

- Sensitivity to CDK9 inhibitor KB-0742 is associated with *MYC* expression/ amplification in SCLC cell lines.
- *MYC* and *MYCL* expression correlates with KB-0742 activity in PDO models.
- SCLC PDO models showed sensitivity to KB-0742 regardless of the treatment history.
- KB-0742 showed antitumor activity in multiple PDX models of SCLC, representing different tumor subtypes with tumor regressions observed in half of the models.
- KB-0742 activity in SCLC models corresponded with transcription-factor activity, whether it was MYC or MYCL.
- Together, these data support the development of KB-0742 as a potential treatment for SCLC.

A phase 1/2 clinical trial of KB-0742 (NCT04718675) is currently recruiting patients with relapsed or refractory solid tumors or non-Hodgkin lymphoma.

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