



**University of
Zurich**^{UZH}

**Zurich Open Repository and
Archive**

University of Zurich
University Library
Strickhofstrasse 39
CH-8057 Zurich
www.zora.uzh.ch

Year: 2023

MSC.sensor: Capturing cancer cell interactions with stroma for functional profiling

Huang, Yun ; Drakul, Aneta ; Sidhu, Jasmeet ; Rauwolf, Kerstin K ; Kim, James ; Bornhauser, Beat ; Bourquin, Jean-Pierre

DOI: <https://doi.org/10.1016/j.slasd.2023.08.003>

Posted at the Zurich Open Repository and Archive, University of Zurich

ZORA URL: <https://doi.org/10.5167/uzh-252405>

Journal Article

Published Version



The following work is licensed under a Creative Commons: Attribution 4.0 International (CC BY 4.0) License.

Originally published at:

Huang, Yun; Drakul, Aneta; Sidhu, Jasmeet; Rauwolf, Kerstin K; Kim, James; Bornhauser, Beat; Bourquin, Jean-Pierre (2023). MSC.sensor: Capturing cancer cell interactions with stroma for functional profiling. *SLAS Discovery*, 28(7):350-354.

DOI: <https://doi.org/10.1016/j.slasd.2023.08.003>



Short Communication

MSC.sensor: Capturing cancer cell interactions with stroma for functional profiling

Yun Huang^{a,b,*,#}, Aneta Drakul^{a,#}, Jasmeet Sidhu^c, Kerstin K. Rauwolf^a, James Kim^a, Beat Bornhauser^a, Jean-Pierre Bourquin^{a,*}

^a Division of Oncology and Children's Research Centre, University Children's Hospital Zurich, Zurich, Switzerland

^b Moores Cancer Center, University of California, San Diego, La Jolla, CA, United States

^c Tata Translational Cancer Research Centre, Tata Medical Center, Kolkata, India

ARTICLE INFO

Keywords:

Biosensor
Synthetic Notch receptor
Mesenchymal stromal cell
Leukemia
Ex vivo drug screen
Drug synergy

ABSTRACT

Mesenchymal stromal cells (MSCs) contribute to the microenvironment regulating normal and malignant hematopoiesis, and thus may support subpopulations of cancer cells to escape therapeutic pressure. Here, we engineered bone marrow MSCs to express a synthetic CD19-sensor receptor to detect and display interacting primary CD19+ leukemia cells in coculture. This implementation provides a versatile platform facilitating *ex vivo* drug response profiling of primary CD19+ leukemia cells in coculture with high-sensitivity and scalability.

1. Introduction

Ex vivo drug testing of primary tumor cells from individual patients is being evaluated in clinical trials to guide therapeutic decisions for patients with resistant disease [1]. However, primary tumor cells are generally difficult to maintain viable *ex vivo* in monoculture. Coculture of primary tumor cells with representative components of the *in vivo* microenvironment can maintain metabolic homeostasis and viability of primary tumor cells to extended periods. The complexity of the multicellular systems involved is nicely recapitulated using organoids derived from human induced pluripotent stem cells [2].

Two-dimensional models have been used to assess the impact of stromal cells in co-culture with malignant cell lines, but it remains challenging to specifically measure the viability of primary tumor cells in co-culture [3]. Conventional viability assays based on metabolic activity (such as ATP present) or cytotoxicity (such as lactate dehydrogenase release) do not distinguish tumor cells from stromal cells. In addition, assays that can specifically measure the tumor cells (for example, flow cytometry) require laborious steps with limited scalability. Bone marrow mesenchymal stromal cells (MSCs) maintain the viability of primary acute lymphoblastic leukemia (ALL) cells in *ex vivo* culture and influence the response to anti-leukemic agents [4–6]. Even in these simplified models it remains difficult to follow the fate of

leukemia cell subpopulations depending on their contact with the microenvironment. Here, we engineer a stroma-based biosensor for specific detection of tumor cells in a coculture model of bone marrow. Using this system, we performed drug response profiling to explore promising drug combinations to target high-risk leukemia subtypes in a protective context.

2. Results

To specifically monitor the live ALL cells that interact with MSCs in coculture, we engineered hTERT-immortalized MSC cells [4] to express a CD19 binding synthetic Notch receptor (CD19-SynNotch-TetRVP64) [7] that controls expression of an integrated reporter cassette TetO-BFP (MSC.CD19sensor.BFP, Fig 1A). When coculture with CD19+ ALL cells, the membrane tethered transcription activator TetRVP64 is released and drives the expression of a BFP reporter in MSC cells (Fig. 1B). As the synthetic Notch receptor only responds to surface tethered ligands [7], the reporter of the MSC.sensor can only be induced by CD19 bound beads but not soluble CD19 (Fig. 1C). Thus, measurement of the reporter signal from MSC.sensor provides a way to monitor the viable ALL cells in contact with cocultured stromal cells *in situ*.

While imaging-based analysis can collect many spatially and temporally resolved parameters, a bioluminescence-based readout is

* Corresponding authors.

E-mail addresses: YUH086@health.ucsd.edu (Y. Huang), Jean-Pierre.Bourquin@kispi.uzh.ch (J.-P. Bourquin).

These authors contributed equally.

<https://doi.org/10.1016/j.slasd.2023.08.003>

Received 23 May 2023; Received in revised form 20 July 2023; Accepted 9 August 2023

Available online 11 August 2023

2472-5552/© 2023 The Author(s). Published by Elsevier Inc. on behalf of Society for Laboratory Automation and Screening. This is an open access article under the CC BY license (<http://creativecommons.org/licenses/by/4.0/>).

more convenient for quantification in high-throughput viability assays. Therefore, we engineered a bioluminescence reporter of MSC.sens (MSC.CD19sensor.Lum) using the NanoLuc luciferase fused to a PEST domain [8]. This reporter yields signals with higher sensitivity and dynamic range than fluorescence signals. Single cell clones of MSC.sens cells were generated and displayed a similar capacity to support the viability of ALL patient-derived xenografts (PDXs) in coculture as the parental MSC population (Fig. 1D). Coculturing of a fixed number of MSC.CD19sensor.Lum cells (2,500 / well in 384 well plates) with an increasing number of ALL PDXs cells yielded a linear increase of

bioluminescence signal up to a ratio of ALL/MS (Fig. 1E). Thus, the bioluminescence signal from MSC.CD19sensor.Lum can be used as a surrogate readout for viable ALL cell count to scale functional screening.

Although MSCs usually have a higher tolerance to chemotherapy than primary ALL cells, their physiological status should not be neglected while profiling with drugs in cocultures. To evaluate the impact of drugs on the MSC.CD19sensor.Lum cells, we measured the bioluminescence signal (driven by CD19 bound beads) and viability (ATP-based assay) of MSCs treated with a selection of anti-cancer agents that are routinely included in contemporary functional precision

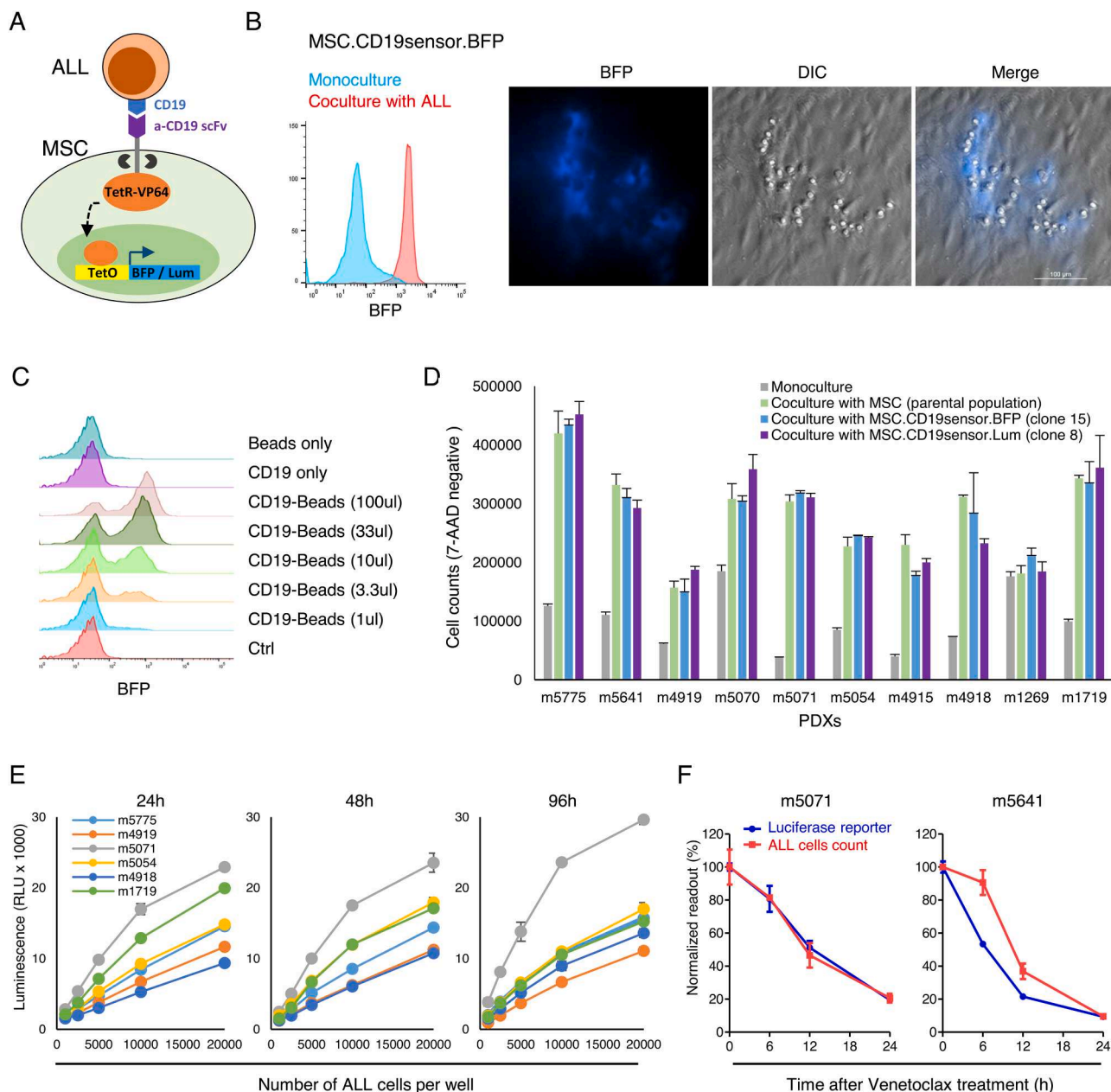


Fig. 1. MSC.CD19sensor for monitoring of ALL cells in coculture. (A) Schematic of the implementation of a CD19 SynNotch receptor in MSC cells. (B) MSC.CD19sensor.BFP-mediated displaying of ALL PDX cells in coculture measured by flow cytometry (left) and microscopy (right). (C) Reporter signal from MSC.CD19sensor.BFP cells exposed to increasing amounts of CD19-conjugated Dynabeads. (D) Viabilities of PDXs from 10 ALL patients after 3 days in monoculture or coculture with parental MSC population, single cell clones from MSC.CD19sensor.BFP or MSC.CD19sensor.Lum. Cell viability was measured by 7-AAD staining and flow cytometry. (E) MSC.CD19sensor.Lum-mediated displaying of ALL PDX cells in coculture measured by bioluminescence. Increasing amount of PDXs cells from 6 ALL patients were seeded together with 2500 MSC.CD19sensor.Lum cells per well in a 384-well-plate. The bioluminescence signals were acquired at 24 h, 48 h and 96 h after seeding. (F) Comparison of the bioluminescence signal decay from MSC.CD19sensor.Lum and ALL cell count measured by flow cytometry (7-AAD negative) after treatment with 1uM Venetoclax in 2 PDXs. PDXs from 2 ALL patients were seeded together with MSC.CD19sensor.Lum at a ratio of 4:1 one day before Venetoclax treatment.

hematology programs (Supplementary Fig S1). We tested 20 approved and investigational compounds in preclinical development with non-overlapping mechanisms of action. While most of the tested compounds (15 out of 20) did not perturb the viability or the reporter of MSC.CD19sensor.Lum cells in a dose-range of 1 nM – 1 μ M, 10 nM of vincristine, bortezomib and panobinostat, as well as 100 nM of cytarabine and idarubicin, provoked an elevated luminescence signal accompanied by a decrease of MSC viability (Supplementary Fig S1). Thus, highly bioactive compounds targeting pan-essential cellular functions may perturb the homeostasis of MSCs and confound the readout during drug profiling in coculture. To monitor the kinetics of the reporter signal after killing of ALL cells, we tested a BCL2 inhibitor Venetoclax in 2 PDXs (Fig. 1F). A progressive decay of the reporter signal matched the decrease of the PDX cell count detected by flow cytometry (7-AAD staining) upon 24 h treatment (Fig. 1F).

Combinational drug screening is a valuable strategy to identify drug synergy to be further developed as safe and effective treatment options for cancer patients [9]. This approach usually requires high-throughput measurement of cell viability in large-scale combinational matrices [10]. We explored candidate drug synergisms targeting high-risk ALL subtypes using the MSC.CD19sensor.Lum coculture with ALL PDXs (Fig 2). We considered Venetoclax as an ideal backbone for exploring synergies with other compounds because: (1) Venetoclax has favorable pharmaceutical properties and efficacy against several high-risk ALL cohorts including *TCF3-HLF* and *MLL*-rearranged ALL [11–13]; (2) Combination of Venetoclax with other therapeutics achieves sustainable remission in many tumor types [14]; and (3) Genome-wide CRISPR analysis of chemotherapy–gene interactions in ALL cells uncovered that sgRNA targeting of *BCL2* scored broadly as a common target whose inhibition could enhance the response to many chemotherapy agents [15].

We established a high-throughput pipeline for combinational drug screening of Venetoclax with 12 therapeutic agents in high-risk ALL cells (Venetoclax^{plus} screen, Methods) (Fig 2, Supplementary Fig S2). Venetoclax showed the strongest synergy with the MCL1 inhibitor S63845, which is in consistent with the functional redundancy of BCL2 and MCL1 in cell death control and endorses the clinical development of this approach in ALL therapy [16] (Fig 3A). Our profiling also revealed potent synergistic combinations of Venetoclax with L-asparaginase and the XPO1 inhibitor Selinexor consistently across all tested PDXs (Figs 3A and 3B). Combinational treatment in a *TCF3-HLF* cell line (HAL-01) and a *TCF3-PBX1* cell line (697) recapitulated these synergies in monoculture (Supplementary Fig S3). The first-in-class nuclear transporter inhibitor Selinexor may represent a promising therapy for ALL patients and is currently in phase I/II clinical trial for pediatric

relapsed/refractory acute myeloid leukemia (AML) patients (Clinicaltrials.gov Identifier NCT04898894). To confirm the on-target effect on XPO1 we performed a CRISPR knockout of *XPO1* and a sgRNA competition assay in HAL-01 and 697 cells. We found that cells expressing *XPO1* sgRNAs were rapidly outcompeted by non-transduced cells shortly after infection, which was further pronounced under Venetoclax treatment (Fig 3C). Given the evidence of the synergy between Venetoclax and Selinexor in AML, diffuse large B-cell lymphoma and multiple myeloma [17], this combination may represent a paradigm of synthetic lethality in many hematologic malignancies.

3. Discussion

Although cell lines have been widely used in functional studies and preclinical drug screenings, cell lines have adapted to grow without niche support and do not represent the molecular complexity of the disease at presentation [3]. Experimental models that more faithfully reflect the primary disease may facilitate the development of next generation therapeutics. We and others have established coculture systems to support primary ALL *ex vivo* enabling functional profiling using imaging-based approaches [5,6]. While imaging records many morphological features of the cells, it generally requires complex analytical pipelines with elaborate software for quantification which require costly infrastructure [18]. Furthermore, inter-sample heterogeneity of leukemia cell morphology confers additional complexity for data analysis by automated imaging. Engineering of biosensor MSCs, which detect ALL cells that interact through CD19 as a surrogate biomarker confers several advantages: (1) Specific detection of leukemic CD19 blasts, which is particularly valuable for bone marrow samples with low leukemia cell infiltration (detection of a small subpopulation of cells without additional cell sorting steps); (2) Objective readout that captures the total amount of leukemia cells in contact in coculture; (3) Label-free and fast assay, which is suitable for high content exploration studies. Although CD19 is dispensable for ALL cells survival [19] and binding with CD19 antibody does not impact ALL cell viability (Fig 1D) [20], engagement with cell surface molecules may rewire downstream signaling and should be considered when designing sensors for other biomarkers in a new model.

We demonstrate the application of MSC.sensor platform to facilitate the exploration of clinically effective combination therapies in ALL. Using a 384-well plate holding 2,500 MSC / well, it is practical to detect 100 – 10,000 ALL cells / well. This format can be further miniaturized, for example by using a 1536 well plate or microfluidic systems to spare primary material. Since the MSC.sensor “reflect” the presence of ALL

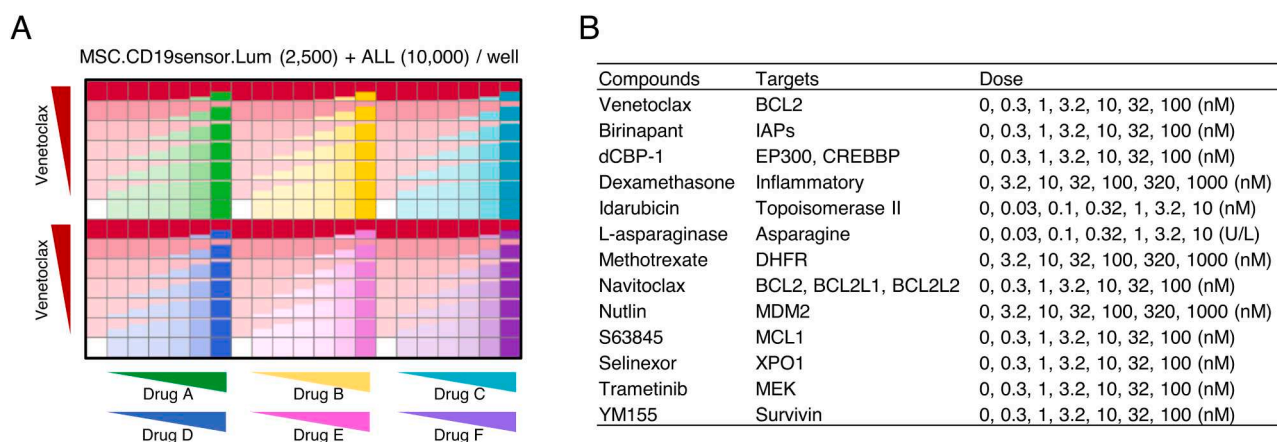


Fig. 2. Schematic of the combinational screen in ALL-MSC coculture in 384-well plates. (A) Illustration of dispense pattern of tested compounds creating 7 × 7 dose combinations in a 384-well plate. ALL cells were cocultured with MSC.CD19sensor.Lum cells for 24 h then treated with combinations for 72 h. Bioluminescence signals from MSC.CD19sensor.Lum cells were acquired at the end of treatment and transformed into normalized inhibition (%) over the dose matrix. (B) Dose ranges of selected compounds tested in Venetoclax^{plus} screen. (see methods section for more details).

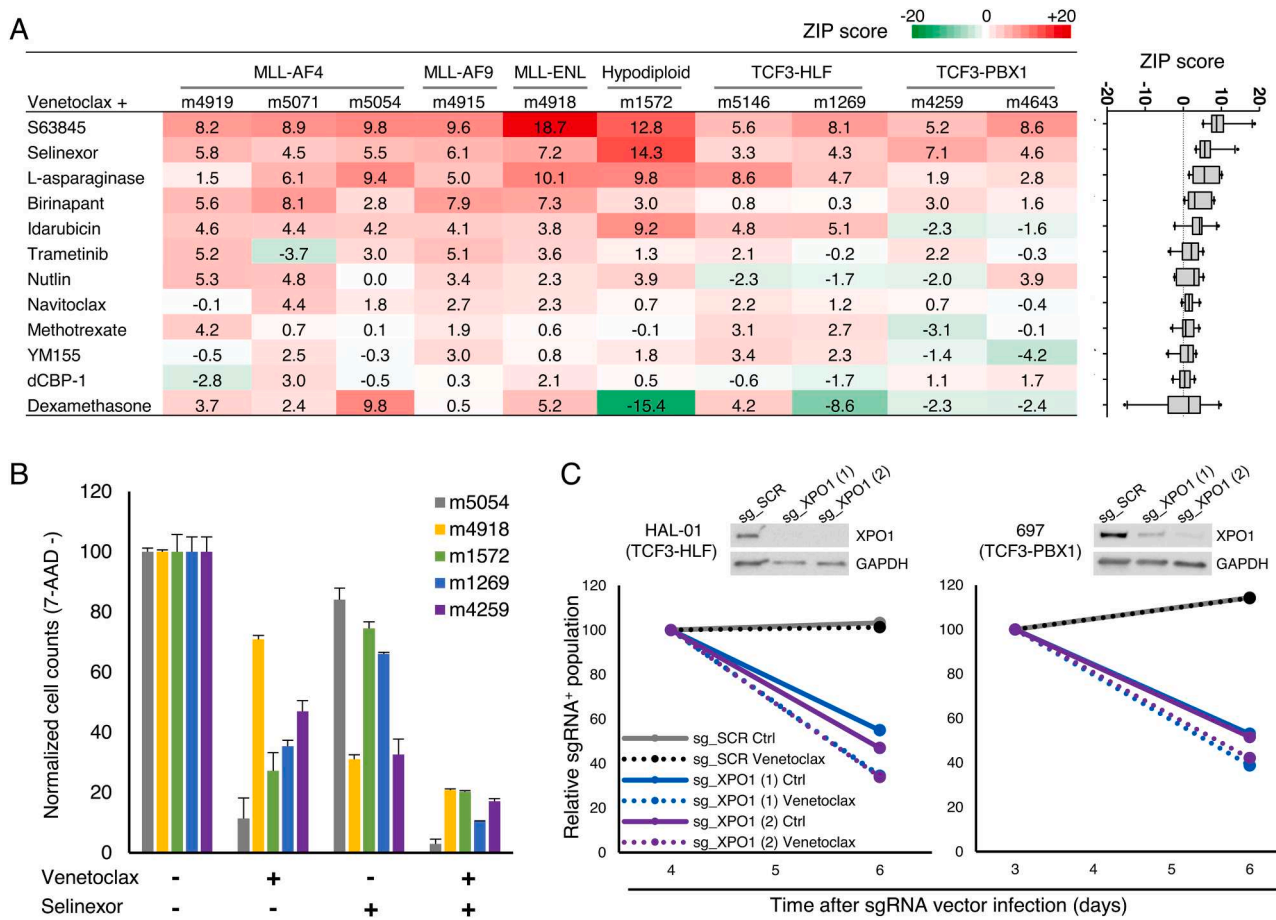


Fig. 3. Venetoclax^{plus} combinational screen in high-risk ALL. (A) Summary of the ZIP-scores[23] from Venetoclax^{plus} combinational screen in PDXs from 10 high-risk ALL patients. **(B)** Validation of the combinational effects of Venetoclax (10 nM) and Selinexor (10 nM) measured by flow cytometry with 7-AAD staining in 5 PDXs. **(C)** Competitive assay after infection of sgRNA vectors targeting *XPO1* (sg_XPO1) and a scramble control (sg_SCR) in HAL-01 (TCF3-HLF) and 697 (TCF3-PBX1) cells stably expressing Cas9 with or without 10 nM Venetoclax treatment.

through contact, this system may also constitute a feasible platform for pooled genetic screens interrogating non-cell-autonomous determinants supporting tumor survival [21].

4. Methods

4.1. Plasmids

The anti-CD19 SynNotch-TetRVP64 receptor vector pHR_PGK_antiCD19_synNotch_TetRVP64 (Addgene #79,126) was described previously [7]. The BFP reporter vector pHR_TetO_tBFP_PGK_mCherry was derived by subcloning the CTS-TRE3GV-TagBFP cassette from pLVX TRE 3 G BFP vector (Addgene #128,070) into the pHR_Gal4UAS_tBFP_PGK_mCherry vector (Addgene #79,130) using KflI and EcoRI sites. The NanoLuc-PEST reporter vector pHR_TetO_NLucPEST_PGK_mCherry was derived by swapping the BFP cassette of pHR_TetO_tBFP_PGK_mCherry vector with NLuc-PEST cassette (PCR from the Pci 9.0 vector (Addgene #74,229)) using MluI and EcoRI sites.

4.2. Lentivirus production and transduction

Lentivirus were produced by transfecting HEK 293T cells with transfer plasmids, psPAX2 (Addgene #12,260) and pCMV-VSV-G plasmids (Addgene #8454) in a ratio of 5:3:2 using polyethylenimine (Polysciences, 24,765–2). Viral supernatants were collected 30 h after transfection and used for transduction immediately.

4.3. Generation of MSC.CD19sensor cells

Human hTERT-immortalized primary bone marrow MSC were provided by Dario Campana (St Jude Children's Research Hospital, Memphis, Tennessee) and maintained in RPMI-1640 medium (Sigma-Aldrich, R0883) supplemented with 10% fetal bovine serum (Sigma-Aldrich), penicillin (100 u/ml)-streptomycin (100 mg/ml), L-glutamine (2 mM) and 1 μ M hydrocortisone. For lentiviral transduction, fresh virus' supernatant was added directly to the cells. Viral media was replaced with normal growth media 24 h after infection. MSC cells were first infected with pHR_PGK_antiCD19_synNotch_TetRVP64 vector encoding the anti-CD19 SynNotch receptor and a myc-tag and sorted by anti-myc A647 (cell-signaling #2233) staining. The MSC stably expressing the anti-CD19 SynNotch receptor were then infected with either pHR_TetO_tBFP_PGK_mCherry vector or pHR_TetO_NLucPEST_PGK_mCherry vector and sorted by mCherry to generate bulk populations of MSC.CD19sensor.BFP or MSC.CD19sensor.Lum cells. Single-cell clones from MSC.CD19sensor.BFP and MSC.CD19sensor.Lum cells were generated by seeding single cells in 384-well plates. Reporter signal induction of all single cell clones was detected by coculture with ALL cells and monitored by flow cytometry or bioluminescence assay.

4.4. Patient samples and patient-derived xenografts

Patient samples were collected from different countries within the International BFM Study Group (I-BFM-SG) as described previously [5]. Informed consent was given in accordance with the Declaration of

Helsinki. Approval for experiments with human samples in the mouse xenograft model was obtained from the ethics commission of the Canton Zurich (approval number 2014–0383). All animal studies were carried out in accordance with relevant guidelines and regulations and ARRIVE guidelines. Primary ALL cells were recovered from cryopreserved samples and transplanted intrafemorally or intravenously into 8–12 week old NOD.Cg-Prkdcscid Il2rgtm1Wjl/SzJ (NSG) mice. Leukemia progression was monitored by flow cytometry with rat anti-mouse CD45 (eFluor450, clone 30-F11, REF 48–451–82, eBioscience) and mouse anti-human CD19 (PE, clone HIB19, REF 302,208, BioLegend) antibodies. ALL cells recovered from spleens of NSG mice were used for molecular characterization *in vitro* and *in vivo* experiments.

4.5. ALL-MSc coculture and reporter assay

MSc or MSc.CD19sensor cells were co-cultured with ALL PDX cells in fixed ratios. The BFP reporter signals from MSc.CD19sensor.BFP cells were measured by flow cytometry and the bioluminescence reporter signals from MSc.CD19sensor.Lum cells were measured by a luminescence plate reader after adding a passive lysis buffer containing Furimazine directly in the coculture.

4.6. Drug screen assay

For drug testing, ALL PDX cells were maintained in coculture with MSc.CD19sensor.Lum in a physiologically relevant medium (Plasmax, Ximbio), which mimics the metabolic and physiological profile of human plasma [22], supplemented with 2.5% fetal bovine serum. 10,000 PDX ALL cells and 2,500 MSc CD19sensor.Lum (clone 8) cells were seeded together in 30ul medium per well in a white 384-well-plate. After a 24-hour incubation, drugs or the vehicle were added using a drug printer and incubated for 72 h. At the end of treatment, a Furimazine reaction mixture (2.5ul of PBS, 7.5ul of 5x Passive lysis buffer (Promega) and 0.04ul of 20 mM Furimazine (AOBIOUS)) was added directly to each well and incubated at room temperature in the dark for 10 min. The bioluminescence signals were read by a luminescence plate reader. Viability of ALL cells was determined by the normalized bioluminescence signals. For the combinational screen (Venetoclax^{plus} screen), the dose range of Venetoclax was set from 0.3 nM to 100 nM covering the IC50 values of the majority of the tested ALL PDX. The bioluminescence signals were acquired at the end of the treatment and the synergistic score was calculated based on the Zero interaction potency (ZIP) model [23].

4.7. CRISPR knockout and sgRNA competition assay

A TCF3-HLF⁺ ALL cell line (HAL-01) and a TCF3-PBX1⁺ ALL cell line (697) were firstly infected with a Cas9 vector (pL40C_PGKIntron_Cas9_Green [13]). Cells with stable Cas9 expression were then infected with a sgRNA vector (sg_shuttle_RFP657 [24]) coding the sgRNAs targeting *XPO1* (sg_XPO1(1): AGTGAGCTCTCAAAAAACGT; sg_XPO1(2): TCACACCAGCAATCTCAGTG) or a scramble sequence (sg_SCR: GTAGCGAACGTGTCCGGCGT). In the sgRNA competition assays, the multiplicity of infection (MOI) of the sgRNA vectors was controlled to achieve a transduction of ~50%. The infected populations (RFP657+) were monitored by flow cytometry at different timepoints with or without Venetoclax treatment.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

All data underlying the results are available as part of the article (see Supplementary Data) and no additional source data are required.

Acknowledgments

We thank Christopher J. Ott (Harvard Medical School) for providing dCBP-1. This work was supported by the Swiss Cancer League (KFS-3526-08-2014 and KFS-4237-08-2017) and the Swiss National Research Foundation SNF (310030-133108 and 323530-164223).

Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.slasd.2023.08.003.

References

- Letai A, Bhola P, Welm AL. Functional precision oncology: testing tumors with drugs to identify vulnerabilities and novel combinations. *Cancer Cell* 2022;40:26–35.
- Xu H, Jiao D, Liu A, Wu K. Tumor organoids: applications in cancer modeling and potentials in precision medicine. *J. Hematol. Oncol.* 2022;15:58.
- McMillin DW, et al. Tumor cell-specific bioluminescence platform to identify stroma-induced changes to anticancer drug activity. *Nat Med* 2010;16:483–9.
- Mihara K, et al. Development and functional characterization of human bone marrow mesenchymal cells immortalized by enforced expression of telomerase. *Br J Haematol* 2003;120:846–9.
- Boutter J, et al. Image-based RNA interference screening reveals an individual dependence of acute lymphoblastic leukemia on stromal cysteine support. *Oncotarget* 2014;5:11501–12.
- Frisman V, et al. Ex vivo drug response profiling detects recurrent sensitivity patterns in drug-resistant acute lymphoblastic leukemia. *Blood* 2017;129:e26–37.
- Morsut L, et al. Engineering customized cell sensing and response behaviors using synthetic notch receptors. *Cell* 164, 780–91 (2016).
- England, C.G., Ehlerding, E.B. & Cai, W. NanoLuc: a small luciferase is brightening up the field of bioluminescence. *Bioconjug Chem* 27, 1175–87 (2016).
- Mathews Griner LA, et al. High-throughput combinatorial screening identifies drugs that cooperate with ibrutinib to kill activated B-cell-like diffuse large B-cell lymphoma cells. In: *Proceedings of the National Academy of Sciences of the United States of America*. 111; 2014. p. 2349–54.
- Chory EJ, et al. High-throughput approaches to uncover synergistic drug combinations in leukemia. *SLAS Discov* 2023.
- Fischer U, et al. Genomics and drug profiling of fatal TCF3-HLF-positive acute lymphoblastic leukemia identifies recurrent mutation patterns and therapeutic options. *Nat Genet* 2015;47:1020–9.
- Khaw SL, et al. Venetoclax responses of pediatric ALL xenografts reveal sensitivity of MLL-rearranged leukemia. *Blood* 2016;128:1382–95.
- Huang Y, et al. The leukemogenic TCF3-HLF complex rewires enhancers driving cellular identity and self-renewal conferring EP300 vulnerability. *Cancer Cell* 2019;36. 630–644 e639.
- Yue X, Chen Q, He J. Combination strategies to overcome resistance to the BCL2 inhibitor venetoclax in hematologic malignancies. *Cancer Cell Int* 2020;20:524.
- Oshima K, et al. Mutational and functional genetics mapping of chemotherapy resistance mechanisms in relapsed acute lymphoblastic leukemia. *Nature cancer* 2020;1:1113–27.
- Ramsey HE, et al. A novel MCL1 inhibitor combined with venetoclax rescues venetoclax-resistant acute myelogenous leukemia. *Cancer Discov.* 2018;8:1566–81.
- Fischer MA, et al. Venetoclax response is enhanced by selective inhibitor of nuclear export compounds in hematologic malignancies. *Blood advances* 2020;4:586–98.
- Chandrasekaran SN, Ceulemans H, Boyd JD, Carpenter AE. Image-based profiling for drug discovery: due for a machine-learning upgrade? *Nat. Rev. Drug Discov.* 2021;20:145–59.
- Weiland J, et al. BCP-ALL blasts are not dependent on CD19 expression for leukaemic maintenance. *Leukemia* 2016;30:1920–3.
- Winterberg D, et al. Engineering of CD19 antibodies: a CD19-TRAIL fusion construct specifically induces apoptosis in B-cell precursor acute lymphoblastic leukemia (BCP-ALL) cells *in vivo*. *J. Clin. Med.* 2021;10.
- Qu Y, et al. Tumor microenvironment-driven non-cell-autonomous resistance to antineoplastic treatment. *Mol. Cancer* 2019;18:69.
- Vande Voorde J, et al. Improving the metabolic fidelity of cancer models with a physiological cell culture medium. *Sci. Adv.* 2019;5:eaau7314.
- Ianevski A, Giri AK, Aittokallio T. SynergyFinder 2.0: visual analytics of multi-drug combination synergies. *Nucleic Acids Res.* 2020;48:W488–93.
- Huang Y, et al. Rapid generation of leukemogenic chromosomal translocations *in vivo* using CRISPR/Cas9. *Hemasphere* 2020;4. e456.