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Plan de Recuperación, Transformación y Resiliencia
Next Generation Catalunya
Generalitat de Catalunya Departament de Recerca i Universitats
IBEC Institute for Bioengineering of Catalonia

PLAN COMPLEMENTARIO DE BIOTECNOLOGÍA APLICADA A LA SALUD EN CATALUÑA

PROYECTOS ScreenTech

INFORME FINAL

Proyecto: Discovery of anticancer molecular glue degraders (**glueDISC**)

Línea de Actuación: **LA3**

Entidad **Coordinadora**: IRB Barcelona

Entidades **Participantes**: IRB Barcelona

Fase de ejecución: **Fase 1** / Fase 2 (marcar la fase asignada)

Inicio proyecto: 27/02/2024

Fin Proyecto: 23/04/2025

CONTENIDO DEL INFORME

1. Estado final de los objetivos del proyecto
2. Contingencias y modificaciones del plan de trabajo
3. Publicaciones científicas
4. Resultados de transferencia
5. Difusión y comunicación
6. Resumen de los principales resultados para su publicación en la web del Plan Complementario

1. ESTADO FINAL DE LOS OBJETIVOS DEL PROYECTO

Describir el grado de alcance final en la ejecución del proyecto, aportando la siguiente información:

- Grado de cumplimiento del objetivo del proyecto (en %).
- Descripción resumida de las principales actividades realizadas.
- Próximos pasos
- Entidades y Comunidades Autónomas (CCAA) participantes (si aplica)

Tabla 1. Resumen del grado de cumplimiento del objetivo del proyecto indicados en el Convenio

OBJETIVO GENERAL	% CONSECUCIÓN FINAL	FECHA CONSECUCIÓN
Discovery of molecular glue degraders using a target-agnostic drug screening strategy	95%	23/04/2025

Tabla 2. Descripción de las principales actividades realizadas. ***Máximo 2 páginas***

Descripción resumida de las actividades realizadas para la consecución de los objetivos del proyecto. Estado final del proyecto en relación con la planificación inicial

Context: The project focused on the discovery of novel molecular glue degraders (MGs) that exploit vulnerabilities in chronic myeloid leukemia and pancreatic cancer, with a goal to address these diseases in a target-agnostic manner. The core approach harnessed comparative chemical profiling of the IRB drug library in engineered E3-off versus E3-on cell models. Our E3-off models have been engineered to disrupt key regulatory nodes within the ubiquitin-proteasome system, thereby allowing the simultaneous interrogation of more than 350 E3 ligases. This innovative design enabled the unbiased, large-scale screening of compounds for their ability to hijack the cellular machinery to induce targeted protein degradation, a process that is central to the function of MGs.

Actividades realizadas:

The project adopted a two-stage screening workflow.

- The first stage involved a pre-screening assay focused on measuring cell viability, which served to prioritize compounds from two extensive libraries: a diversity library comprising ~125,000 compounds and a focused library consisting of ~16,000 compounds. Both libraries were screened across two disease-relevant cell lines, KBM7 (representing CML) and CFPAC1 (representing pancreatic adenocarcinoma). Compounds that significantly impacted cell fitness were ranked according to their cell fitness impairment profiles, generating a prioritized list of candidates for the subsequent microscopy screening phase.

- The second stage featured a comparative high-throughput microscopy-based assay, where the prioritized compounds were profiled in mixed populations of E3-off and E3-on cells, each expressing a different fluorescent protein (Fig.1). For the CFPAC1 cell line, 2,400 compounds (15 × 384-well plates) from the diversity library and 3,000 compounds (19 × 384-well plates) from the focused library were selected for screening. Similarly, for the KBM7 cell line, 2,000 compounds (13

× 384-well plates) from the diversity library and 3,000 compounds (19 × 384-well plates) from the focused library were evaluated. This comparative approach allowed the identification of compounds whose activity specifically depends on the presence of functional E3 ligases, a hallmark of MG degraders.

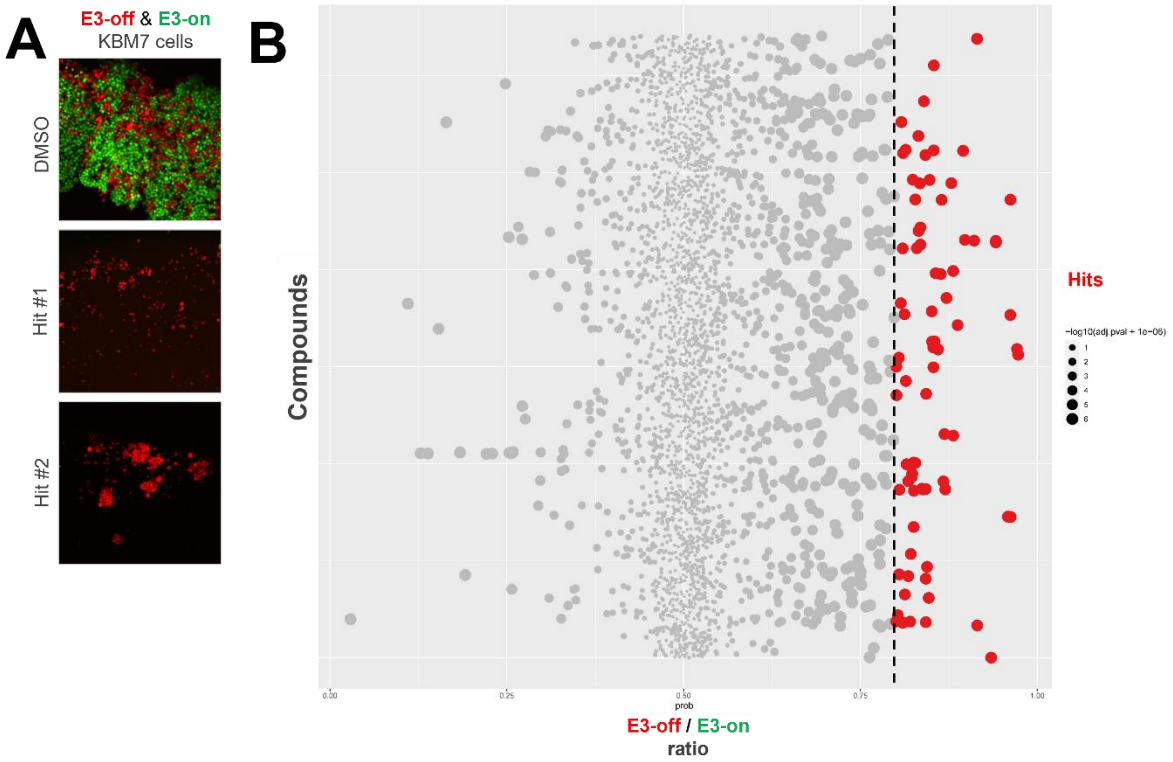

Figure 1 High-throughput microscopy-based drug screenings. a) E3-on-GFP cancer cells were mixed in a 1 to 1 ratio with E3-off-mCherry cells. After 6-day treatment, fluorescence was measured. E3-off-mCherry cells survive in the presense of molecular glue treatment given its resistance to degraders. b) Result of one of the microscopy drug screens (IRB diversity library, KBM7 cells).

Ultimately, the screening campaign yielded a set of **200 compounds** that exhibited E3-dependent activity, which have been selected for further dose-resolved validation. This validation will be essential to confirm the MG mechanism and to establish the specificity and potency of the identified candidates. Due to several technical problems with the Echo drug dispenser and Envision plate reader in the IRB Drug Screening Platform, all the screenings were delayed. Thus, the validation phase of these 200 compounds is still pending. Additionally, we need to advance the subsequent mechanistic studies, including proteomic analysis to identify the degraded targets and the specific E3 ligases involved, as well as structure-activity relationship (SAR) studies to refine the chemical scaffolds of the most promising compounds.

In summary, the integration of comparative chemical profiling in engineered E3-off/E3-on models not only streamlined the discovery of novel MGs but also provided a robust platform for interrogating the vast network of E3 ligases, yielding 200 potential new MG degraders with anticancer properties.

Próximos pasos previstos tras finalizar el Programa ScreenTech: Further validation of the 200 selected compounds will be conducted to confirm their activity as MG degrader. Additional mechanistic studies, including proteomic analysis and SAR refinement, are planned to further characterize the hits and advance the most promising candidates toward therapeutic development
Entidades y Comunidades Autónomas que han participado en las actividades realizadas (si aplica) IRB Barcelona (Barcelona, Cataluña)

2. CONTINGENCIAS Y MODIFICACIONES DEL PLAN DE TRABAJO

Breve descripción de problemas e incidencias que han impedido realizar las actividades previstas y/o alcanzar los objetivos inicialmente planteados en el plan de trabajo en el marco temporal previsto, así como las medidas que se han tomado para mitigar su impacto. Indicar, en su caso, desviaciones significativas del cronograma previsto.

Tabla 3. Contingencias y modificaciones del plan de trabajo

Descripción de los problemas e incidencias más relevantes que han impedido realizar las actividades previstas y/o alcanzar los objetivos inicialmente planteados en el plan de trabajo en el marco temporal previsto: During the project, several technical issues arose with the Echo drug dispenser and Envision plate reader in the IRB Drug Screening Platform. These instruments are essential for the accurate and high-throughput dispensing of compounds and for measuring the results of the cell-based assays. Problems included malfunctions in the automated liquid handling system of the Echo dispenser and inconsistencies in signal detection and data acquisition by the Envision plate reader. These technical setbacks significantly disrupted the planned workflow and resulted in delays for all screening activities.
Medidas tomadas para mitigar su impacto: To address these issues, troubleshooting and technical support were sought from the equipment manufacturers and the platform's technical team. Maintenance and calibration procedures were performed to restore the functionality of both the Echo drug dispenser and the Envision plate reader. Additionally, alternative scheduling was arranged to optimize the use of available equipment and to minimize downtime, although the delays were really big. We tried to adapt to all these issues in the best possible way, although cultured cells already expanded were trashed etc. We did try to use similar passages of our cell lines to diminish potential variability in that regard. In addition, many positional controls in the plates were added as we usually did. Thus, our team implemented additional quality control checks to ensure data integrity once the instruments were back in operation.

Desviaciones significativas del cronograma previsto:

As a consequence of the technical problems, all screenings were delayed. This required a revision of the project timeline to accommodate the extended time required for completing the high-throughput screening campaigns in close coordination with the IRB Drug Screening Platform team, who tried to be as diligent as possible.

Despite these setbacks, the project remained on track to achieve its main objectives, albeit with a shift in the expected completion dates for the screening phases.

3. PUBLICACIONES CIENTÍFICAS

Relación de las publicaciones científicas derivadas del proyecto, indicando cuáles han sido las entidades y Comunidades Autónomas participantes que han contribuido a dichas publicaciones. Indicar si se ha incluido agradecimiento al PRTR y si es de acceso abierto.

En el caso de falta de espacio, como mínimo debería haberse incluido la referencia: PRTR-C17.I1

"This work, integrated into the framework of PERTE for Vanguard Health, has been co-financed by the Spanish Ministry of Science, Innovation and Universities with funds from the European Union NextGenerationEU, from the Recovery, Transformation and Resilience Plan (PRTR-C17.I1) and from the Autonomous Community of Catalonia within the framework of the Biotechnology Plan Applied to Health."

Tabla 4. Publicaciones científicas

Publicación científica	Entidades y CCAA participantes	¿Se incluye agradecimiento al PRTR?	¿Es open Access?
No scientific publications have been published from this project yet. However, a manuscript detailing key related research (e.g., the screening approach) is currently in preparation and is expected to be submitted to Molecular Cell for peer review in the near future (Nieto-Barrado et al., *in preparation).*	All upcoming papers from our lab related to this project will acknowledge the funding and participating entities.	All upcoming papers from our lab related to this project will acknowledge the funding and participating entities.	All upcoming papers from our lab related to this project will be available with open access, as we always do.

4. RESULTADOS DE TRANSFERENCIA

Relación y breve descripción de los resultados de transferencia obtenidos (patentes, contratos de licencia, modelos de utilidad, contratos de explotación, spin off, colaboraciones con el sector productivo, etc.). Especificar cuáles han sido las entidades y Comunidades Autónomas participantes que han contribuido a los resultados indicados.

Tabla 5. Resultados de transferencia

Tipo de Resultado	**Breve descripción de los resultados más relevantes**	**Entidades y CCAA que han contribuido a los resultados**
To date, no transfer results have been generated from this project. Our teams will conduct the mechanistic deconvolution of validated druh hits as planned, and then, together with the Innovation department at IRB willevaluate the potential for future transfer activities.	Not applicable yet	IRB Barcelona, Cataluña

5. DIFUSIÓN Y COMUNICACIÓN

Resumen de las principales actuaciones realizadas para dar visibilidad al proyecto y al Plan Complementario (eventos de presentación, participación en congresos, notas de prensa, presencia en medios de comunicación y redes sociales, etc.). Indicar, si es el caso, cuáles han sido las entidades y Comunidades Autónomas participantes en cada una de las actuaciones y si se ha realizado alguna acción conjunta.

Tabla 6. Principales actuaciones de difusión del Plan Complementario de Biotecnología

Principales actuaciones de visibilización. Incluir, si está disponible, enlaces a las publicaciones, comunicaciones / abstracts en congresos, etc.	**Tipo de actuación**	**Entidades y CCAA que han contribuido a los resultados**	**¿Se ha incluido agradecimiento al PRTR?**
Project results have been presented at national and international scientific conferences and congresses multiple times, enabling dissemination to the research community and facilitating discussion and feedback.	Conferences, seminars & Congresses	IRB Barcelona, Cataluña	Yes, in the Acknowledgments final slide.
Engagement in outreach activities has allowed for the communication of project aims and results to the general public (e.g., IRB open day, Women in Science Day, Foro Teófilo Hernando from the Spanish Royal Academy of Medicine, etc.)	Outreach	IRB Barcelona, Cataluña	Yes, in the Acknowledgments final slide.

6. RESUMEN DE LOS PRINCIPALES RESULTADOS PARA SU PUBLICACIÓN EN LA WEB DEL PLAN COMPLEMENTARIO

Breve resumen de principales resultados que sea publicable. **Máximo 250-300 palabras**, y 1 imagen representativa (puede ser un tipo infografía resumen).

Se publicará en la pestaña Proyectos de la web del Plan. El contenido se utilizará también para en las redes sociales.

Web: https://planescomplementariossalud.es/proyectos-colaborativos/

LinkedIn: https://www.linkedin.com/company/ppccsalud

X: https://x.com/PpccSalud

Around 90% of human proteins, including many that are linked to life-threatening diseases, remain inaccessible to traditional inhibitors. In recent years, molecular glue degraders (MGs) have demonstrated that compound-induced proximity to E3 ubiquitin ligases can prompt the ubiquitination and degradation of disease-relevant proteins, providing a promising alternative for therapeutic intervention. Rather than relying on inhibition, these degraders function by binding proteins and orchestrating their elimination, thereby broadening the druggable proteome. While clinical proof for this approach already exists, as exemplified by lenalidomide and its analogs (currently used against multiple myeloma), the discovery of MGs has historically been largely serendipitous, limiting their broader application as a generalizable drug solution.

This project has addressed this challenge by developing a systematic platform to move MG discovery from serendipity to intentional design. By employing innovative screening strategies that interrogate E3 ligase activity, we have identified some chemical matter that can be classified as MG-like compounds. Our approach has yielded promising results, demonstrating the feasibility of uncovering new classes of degraders with anticancer potential and expanding the range of proteins that can be targeted therapeutically. These achievements position Catalonia at the front of next-generation drug discovery, with the potential to help transform the treatment landscape for intractable diseases.

Representative image of the high-throughput and automation needed for the drug discovery screening cascade applied in the project to interrogate large-scale compound libraries.