Instituto Universitario de Investigación en Ingeniería de Aragón
Universidad Zaragoza

XIII JORNADA DE JÓVENES INVESTIGADORES/AS DEL I3A

TME Lab
Tissue Microenvironment Lab

Effect of glioblastoma tumour microenvironment on the modulation of the immune system

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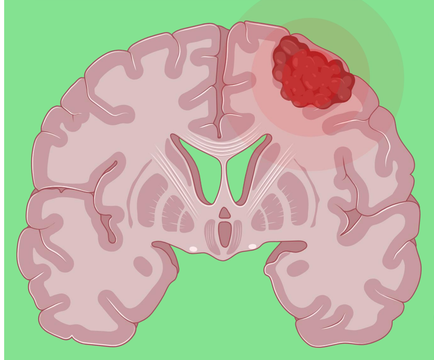
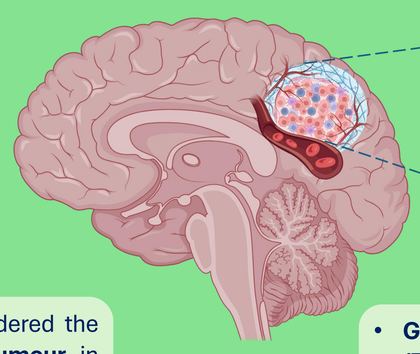
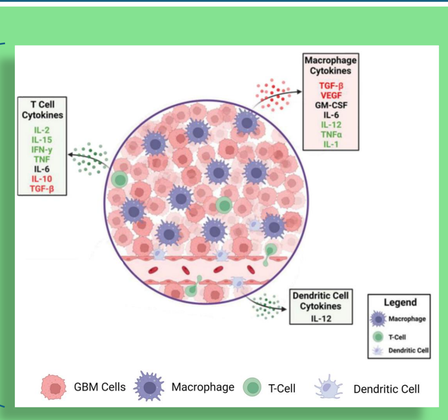
INTRODUCCTION





- **Glioblastoma** (GBM) is considered the **most lethal** primary **brain tumour** in adults.
- The **average survival** of patients **about 14 months.**
- Patients are treated with **radiotherapy and chemotherapy**, with concomitant and **adjuvant temozolomide after surgical resection** [1].

- **Glioblastoma tumour microenvironment (TME)** is characterized by being **highly immunosuppressive** and having **low immunogenicity**, highlighting it as a **'cold tumour'.**
- The **migration** of **immune system** cells **and access to tumour antigens** is an **important** aspect**.** This is **hindered** by the increased **stiffness** of the **extracellular matrix (ECM).**



[2]

- **Cytokines** are secreted proteins that **regulate** the **immune response.**
- **Effective** functioning of **immune system cells** is also **suppressed by** the secretion of diverse **cytokines**
- **Glioma cells** secrete **TGF-β, reducing** the **cytotoxic capacity** of immune cells

OUR APPROACH

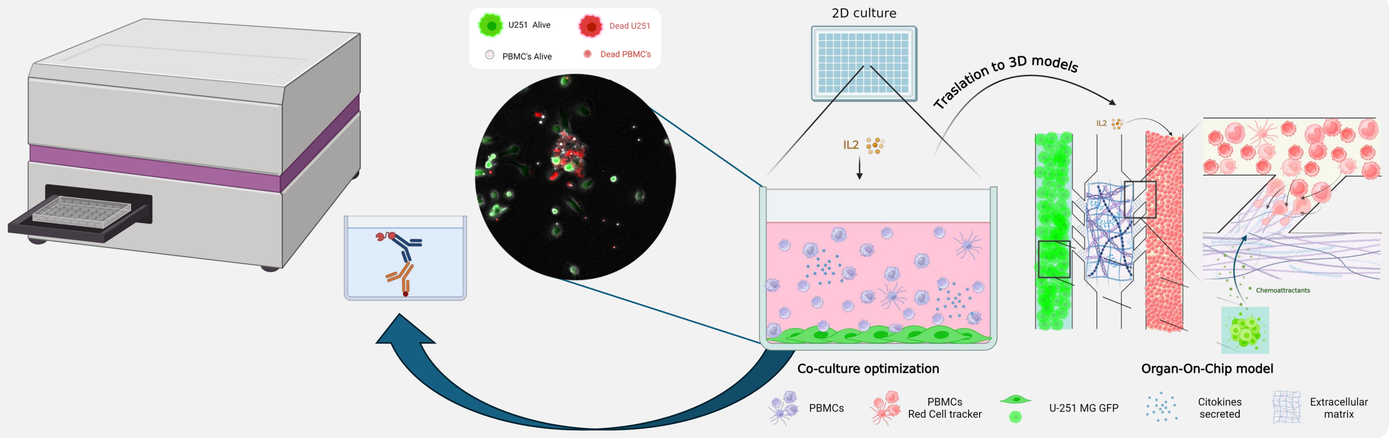
Our approach consists of generating **different co-culture conditions** to **recreate** a physiological environment similar to ***in vivo*** conditions and **apply** that **settings** in the **future** into an **organ-on-chip cell culture model**, that **represent** the **real physiology** of the **tumour.**

To carry this objective is very **important to find** the **optimal conditions** for all populations.

To optimize the conditions, **peripheral blood mononuclear cells (PBMCs)** and **tumoural cells (U-251 MG)** were **seeded** together and separately **in 24-well plates** and **different factors** were **studied**.

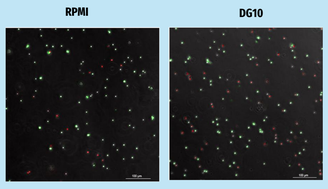
To observe that, **viability tests** were performed with **propidium iodide** (**PI**) and **calcein AM (CAM) stains.**

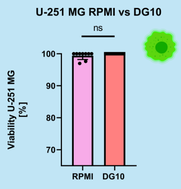
A study of **cytokine secretion** by the different populations in the culture conditions that were considered more physiological **was also carried out**.

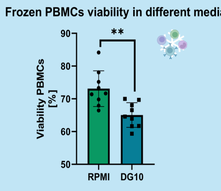


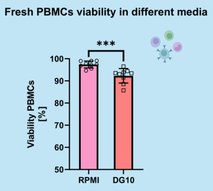
RESULTS

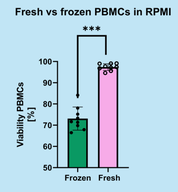
EFFECT OF THE CULTURE MEDIUM



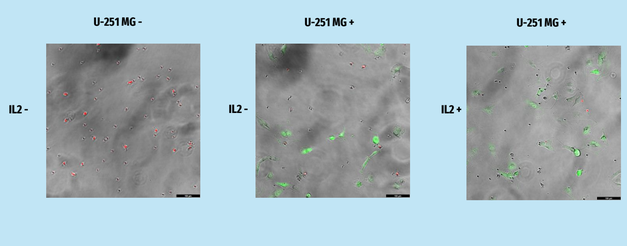


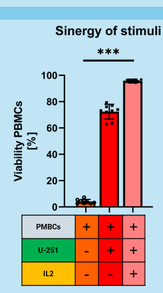




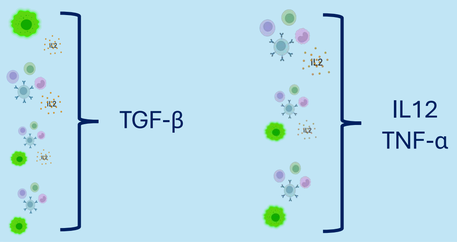


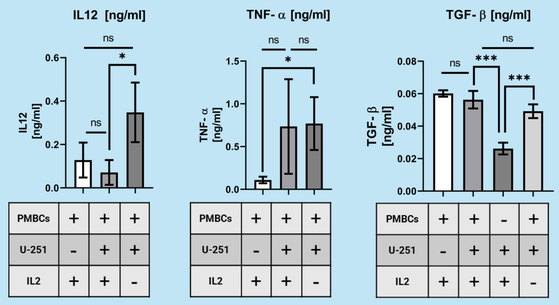
SYNERGISTIC EFFECT OF STIMULI (INTERLEUKIN 2 + U-251 MG)



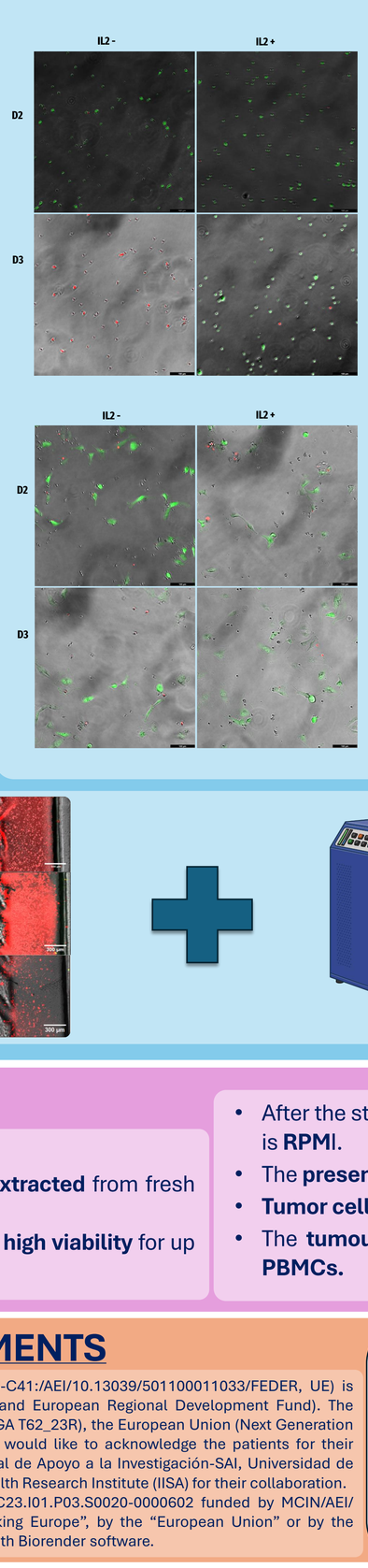


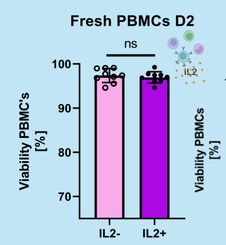
CYTOKINES SECRETED BY THE DIFFERENT MICROENVIRONMENTS

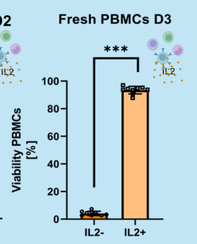


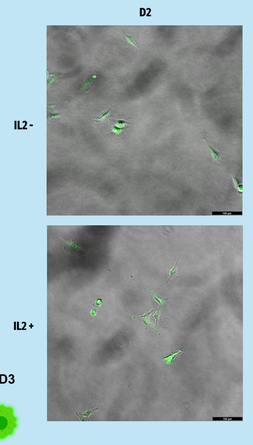


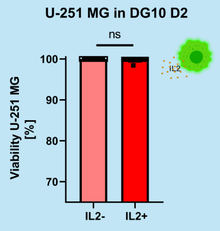
EFFECT OF INTERLEUKIN 2

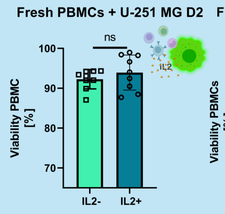


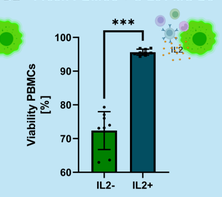






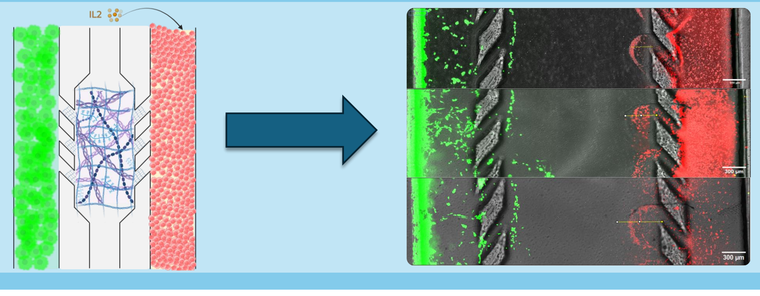


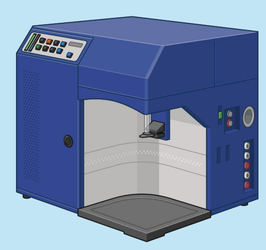


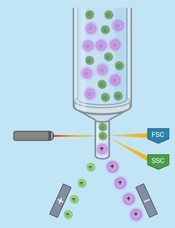


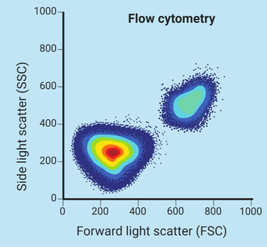
FUTURE INSIGHTS

The **future perspectives** of our research are to make a **translation to 3D models** and in addition to identify in a **more specific** way the interaction of the immune system with the tumor **microenvironment** by performing **flow cytometry** and seeing which **immune populations** are secreting the studied cytokines.









CONCLUSIONS

- It has been proven that the **most optimal medium** for co-culture is **RPM**I.
- **Immune system** cells that have been subjected to **cryogenic stress** have a **lower viability** than **PBMCs freshly extracted** from fresh blood.
- **Immune cells** that receive an **external stimulus**, from cytokines or other cell populations, are able to **maintain** their **high viability** for up to **24 hours longer** than those that are isolated from any type of stimulus.
- After the studies carried out, it has been proven that the **most optimal medium** for co-culture is **RPM**I.
- The **presence** of **tumor cells** (U-251 MG) induces **increased** secretion of **TNF- α** and **IL-12.**
- **Tumor cells** alone **secrete** detectable amounts of **TGF-β** .
- The **tumour microenvironment** (TME) has a **complex influence** and **interaction** with the **PBMCs.**

BIBLIOGRAPHY

[1]. OIKE, T. et al., Radiotherapy plus concomitant adjuvant temozolomide for glioblastoma: Japanese mono-institutional results. PLoS ONE 2013, 8(11), 6–11. doi: 10.1371/journal.pone.0078943

[2]. Adaptation from SOORESHJANI M. et al., The Use of Targeted Cytokines as Cancer Therapeutics in Glioblastoma. Cancers (Basel). 2023; 15(14):3739. doi: 10.3390/cancers15143739.

ACKNOWLEDGEMENTS

Financial support from the GBM_IMMUNE project (PID2021-126051OB-C41:/AEI/10.13039/501100011033/FEDER, UE) is gratefully acknowledged (Ministry of Science and Innovation, Agency and European Regional Development Fund). The authors are also grateful for the support of the Government of Aragón (DGA T62_23R), the European Union (Next Generation EU), and the intellectual and technical assistance of CIBER-BBN; and would like to acknowledge the patients for their participation, the Biobank of the Aragon Health System, Servicio General de Apoyo a la Investigación-SAI, Universidad de Zaragoza, and the Plataforma de Apoyo Preclínico (PAP) at the Aragon Health Research Institute (IISA) for their collaboration. The author's work is financed by the Investigo Program. Grant 2022-C23.I01.P03.S0020-0000602 funded by MCIN/AEI/ 10.13039/501100011033 and, as appropriate, by "ERDF A way of making Europe", by the "European Union" or by the "European Union NextGenerationEU/PRTR". Some figures were created with Biorender software.

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