Autologous organoid T cell co-culture platform as a powerful personalized model for Immunotherapy

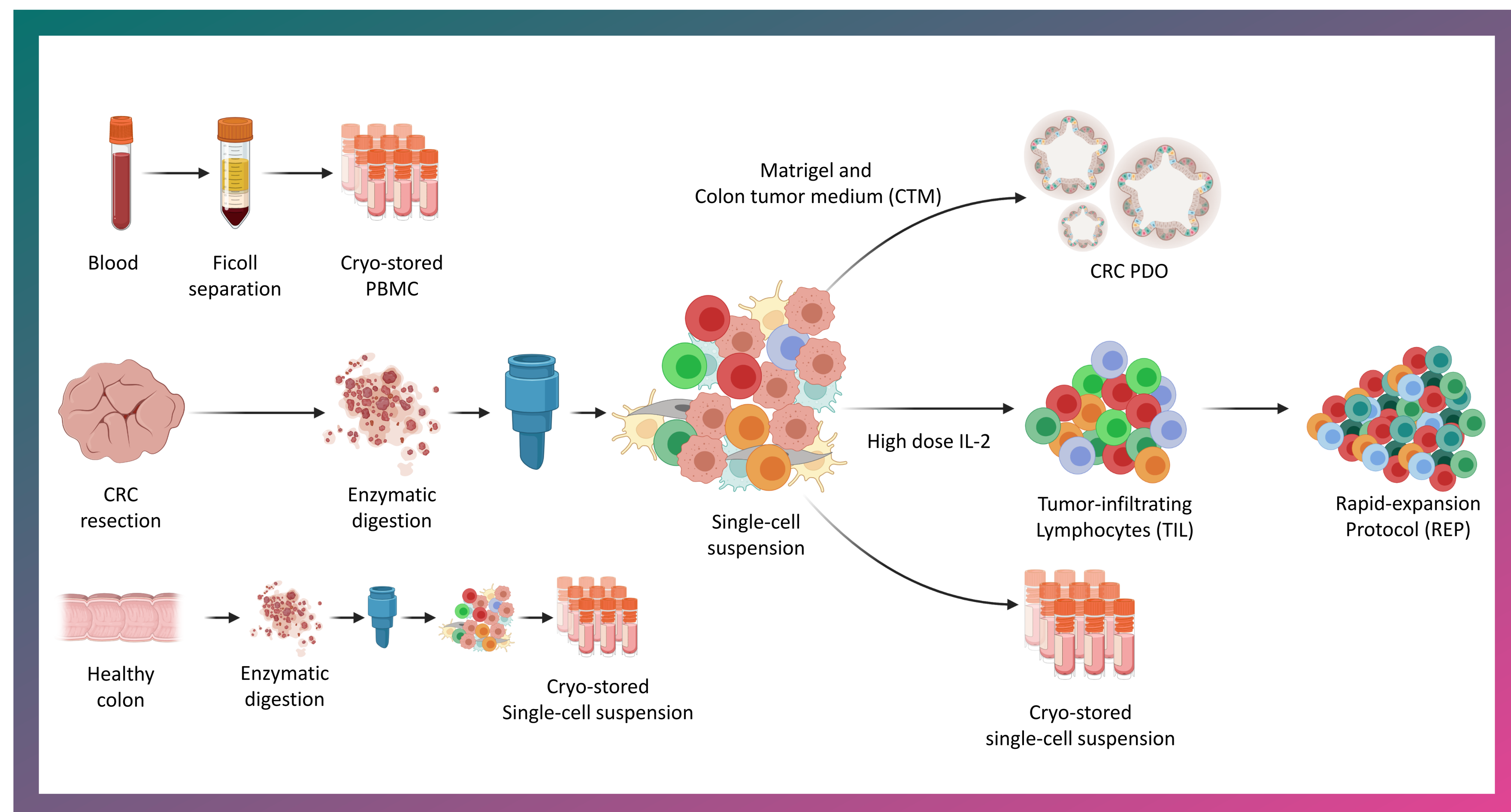
HUB ORGANOIDS

Soura Mardpour; Lorenz Jahn; Pleun Hombrink; Sylvia F. Boj and Farzin Pourfarzad
HUB Organoids (HUB), Yalelaan 62, 3584CM, Utrecht, The Netherlands

Introduction

Immunotherapy is a fast developing and effective treatment strategy to combat cancer. Immuno-oncology (IO) modulators such as checkpoint inhibitors, and bi-specific antibodies are being developed increasingly by biotech and pharmaceutical industry. Platforms that reliably model the tumour–immune-cell interaction will greatly contribute to our understanding of the critical factors that determine a successful IO therapy for solid tumours. HUB Organoids (HUB) has developed "living" biobanks of patient-derived tumour and normal 3D organoid from different epithelial organs, including but not limited to, gastrointestinal tract and lung, that can be cultured with autologous immune cells. HUB Organoid Technology allows for the in vitro expansion of patient-derived organoids (PDO) from healthy- and tumour tissues as three-dimensional primary cell cultures which retain the histological and mutational features of the original tumour tissue. HUB offers a co-culture system based on colorectal cancer (CRC) PDO and their paired immune cells such as tumour infiltrating lymphocyte (TIL).

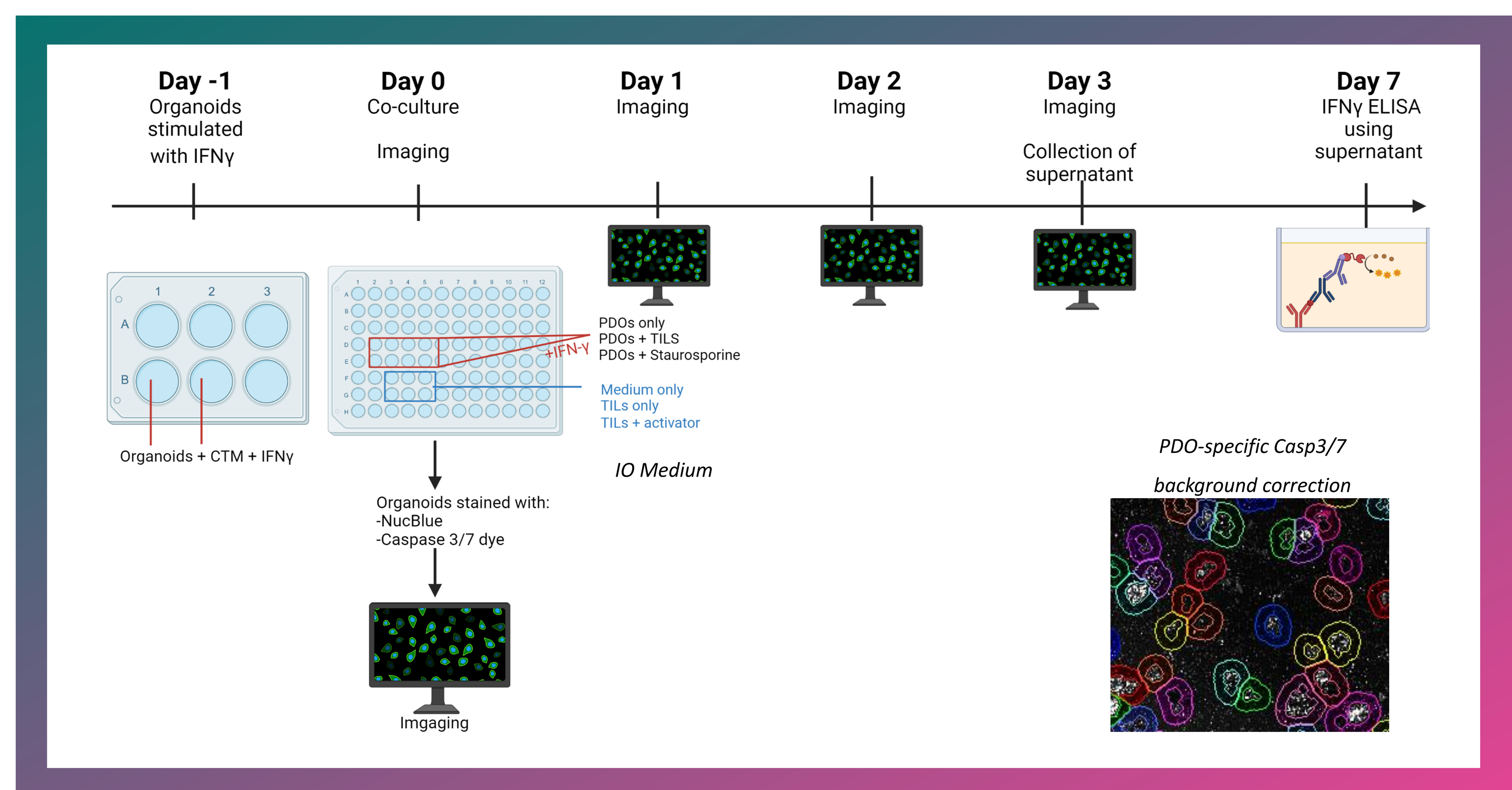
Figure 1. Biobanking of CRC-PDO and autologous TIL



Methods

- Co-isolation of colon PDO and their paired TILs from the resected tissue were performed using mechanical and enzymatic digestion (adapted from Dudley *et al,* 2003). A TIL master cell bank (MCB) was generated by expansion of TILs in presence of high dose IL-2. (Figure 1).
- PDO were further characterised in terms of expression of immune check-point molecules by flow cytometry. TILs tumour reactivity of was evaluated in co-culture with paired CRC-PDO. PDO killing and T cell activation was detected by image analysis of Caspase 3/7 activation and further confirmed by ELISA quantification of IFNγ secretion (Figure 2 and 3).
- TILs tumour reactivity of was enriched via repeated exposure to paired CRC PDO and subsequently CD137+ CD154+ activated TILs were selected and underwent a rapid expansion protocol (REP). TIL clonality was evaluated by TCRvβ analysis (Figure 4).

Figure 2. Schematic overview of CRC PDO-TIL co-culture assay



Results

Tumour PDO were further characterised for expression of immune regulatory receptors such as PD-L1, CD80 and CD86. We developed a robust protocol for simultaneous co-isolation and expansion of tumour PDO and their paired TILs with efficiency of 75%. Moreover, HUB developed a protocol to enrich tumour reactive TILs by repeated tumour PDO exposure. Enrichment of tumour reactive T cells led to skewed T cell receptor (TCR) repertoire and improved tumour organoid killing in co-culture with paired tumour PDO which was detected by several readouts such as image based apoptotic signal, expression of T cell activation markers, and secretion of proinflammatory cytokines.

Figure 3. CRC PDO-TIL co-culture assay

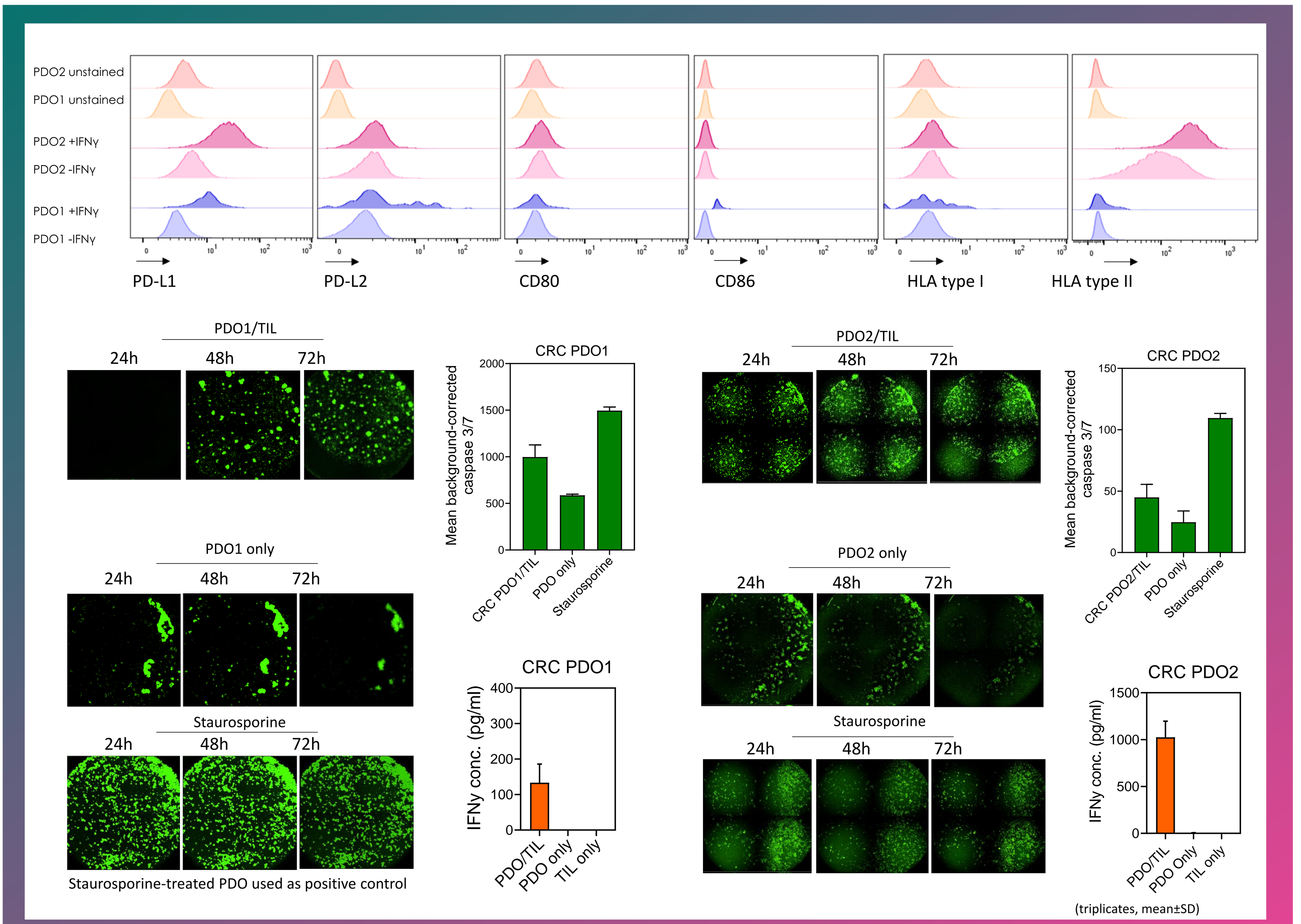
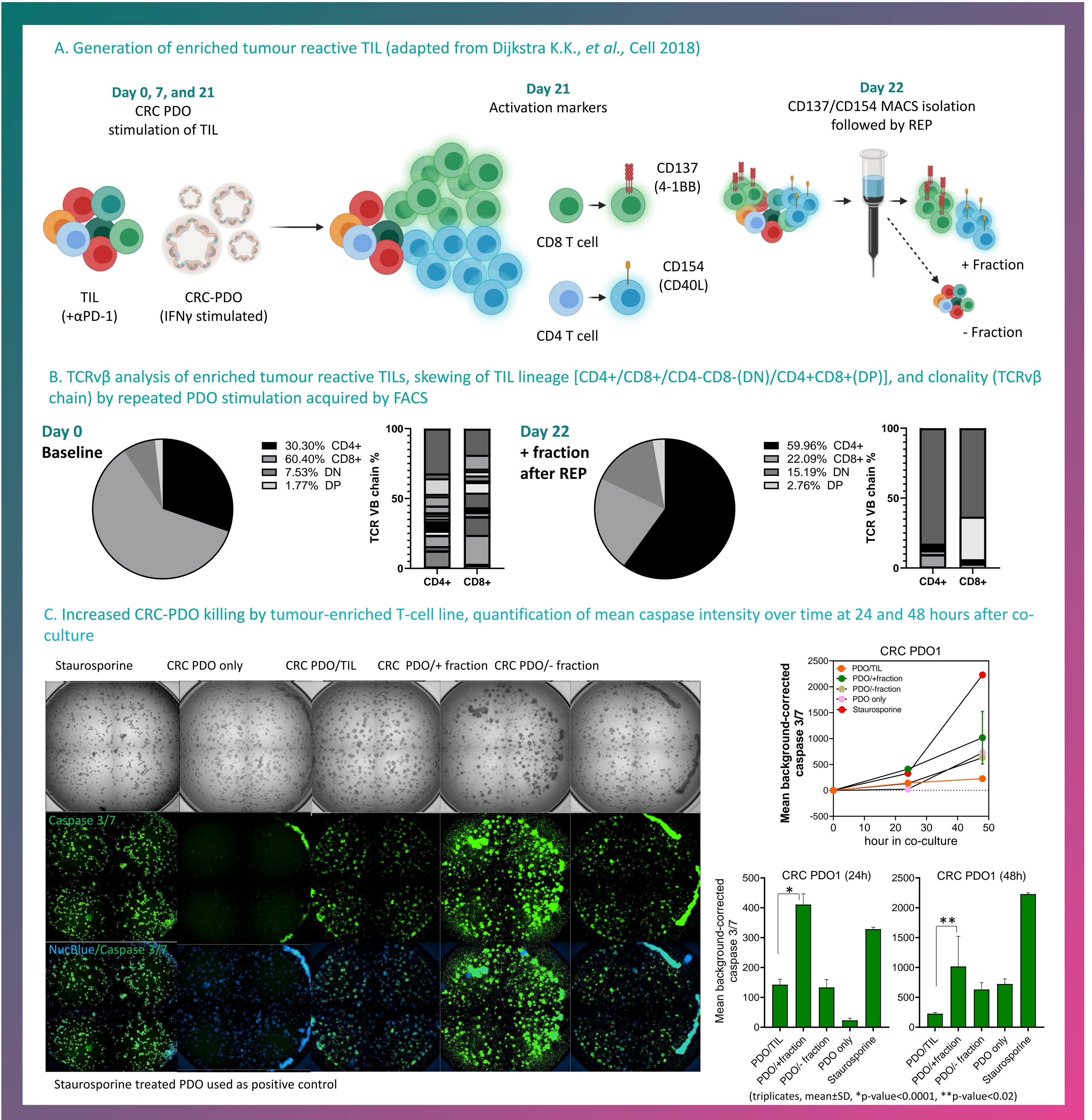


Figure 4. Generation, characterisation and tumour reactivity of enriched CRC PDO-reactive TIL



Conclusion

- CRC organoids can be generated with paired immune cells
- PDO-TIL co-cultures provide a screening platform for T-cell immunomodulators, αCD3-bispecific antibodies, or cancer vaccines

References

1. Dudley *et al*; Generation of Tumour-Infiltrating Lymphocyte Cultures for Use in Adoptive Transfer Therapy for Melanoma Patient; J Immunother. 2003; 26(4): 332–342.
2. Dijkstra *et al*; Generation of Tumour-Reactive T Cells by Co-culture of Peripheral Blood Lymphocytes and Tumour Organoids. Cell. 2018 Sep 6;174(6):1586-1598.e12. doi: 10.1016/j.cell.2018.07.009. Epub 2018 Aug 9.

© Copyright 2022. HUB Organoids. **All rights reserved.**

www.huborganoids.nl

Contact us