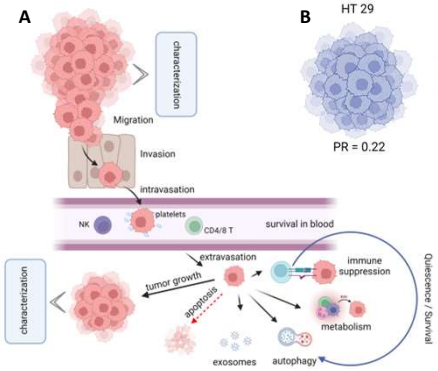
# Survival And Colonisation Axis Of Metastasised Cells In The Secondary Tissue: To Target Or Not To Target?

Samrat Roy*, Manoj Pandre*, Sundarajan Kannan, Rajesh Kumar RK, Chinmaya Amarkanth, Debabani Roy Chowdhury & Arnab Roy Chowdhury
Mestastop Solutions, Bangalore, India & Mestastop Inc, New Jersey, USA.
* Both authors contributed equally

## Introduction

- Between 2015 and 2020, the EMA issued 132 new indications for 62 oncology drugs (solid tumours), of which 46% were targeted therapy, 30% were immunotherapy and only 6% were chemotherapy[1]. Interestingly, though many were approved for metastatic conditions none of them treated the process of metastasis.
- Multiple clinical trial failures focused on targets involved in the early dissemination process of metastasis have led to the deprioritization of metastasis focused drug discovery[2].
- Several recent publications have shown that early dissemination was probably responsible for this failure, as none of the treatments was able to target the cells that were hiding in the tissues, post dissemination[3].
- We have previously shown that plasticity ratio (PR; the ratio of mesenchymal to epithelial markers), gives a true representation of a given cell's tumorigenic and metastatic potential[4].
- Here we highlight the importance of PR during the survival of the disseminated cells in the secondary tissue followed by subsequent colonisation and how one can manipulate it for drug discovery.

## Methods




*A.* Complete dissection of metastasis biology, with more focus on post-dissemination, e.g. survival in secondary tissues[5]

*B.* Genetically engineered cells with higher PR that have less tumorigenicity but more metastatic potential[6]

## Results

**Fig.2:** Representative changes in (A) PDL1 levels (B) Caspase activity (C) Autophagy (D) Metabolic profile & (E) Radial chart summary of functional assays

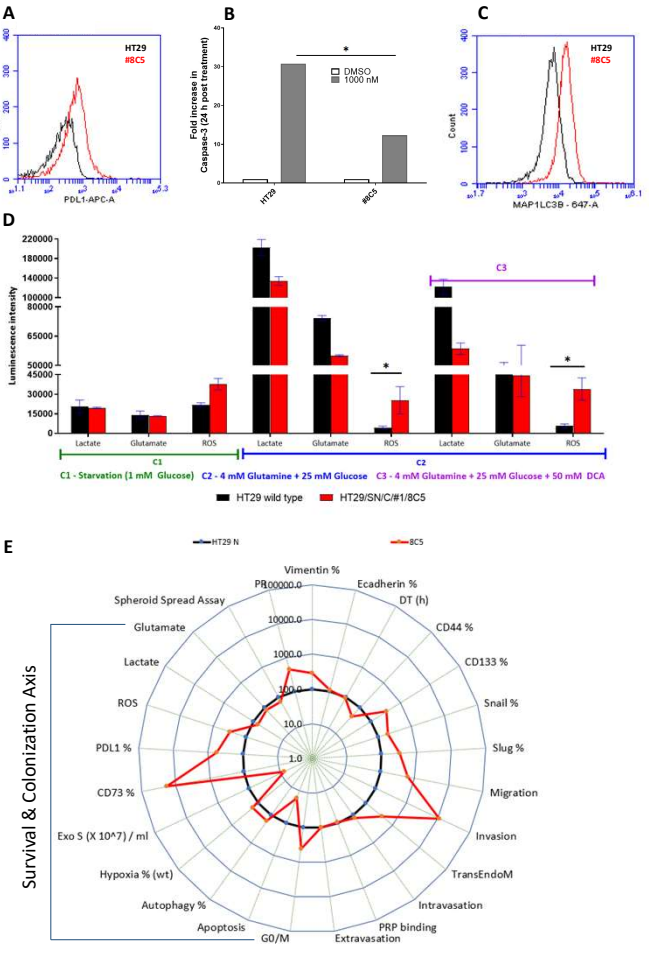



**Fig.3:** Effect of RA on 8C5 cells with respect to (A) PR, (B) Cell Cycle and (C) Growth and (D) % comparison of change, (E) Chemosensitivity

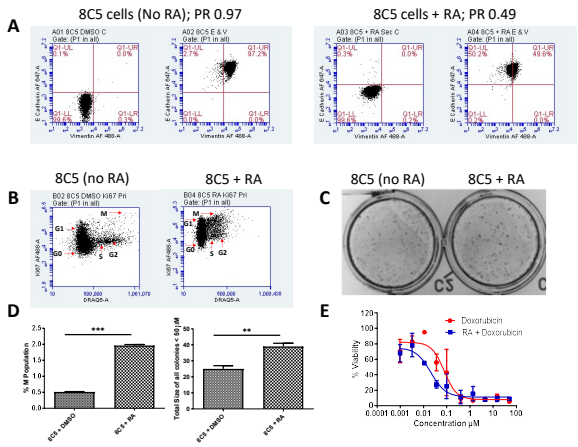
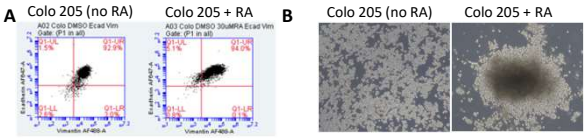


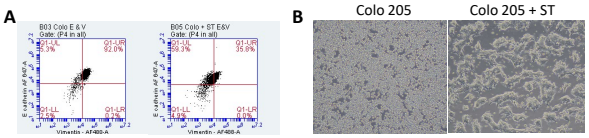

**Fig. 4**: Effect of RA on Colo 205 cells with respect to (A) PR, (B) Colony formation




PR changes from 1.01 to 0.94 (ns), BUT 30 fold decrease in free vimentin (Q1-LR), 3 fold increase in free E Cad (Q1-UL)

Clumping and clustering observed in 2D cultures

**Fig. 5**: Effect of Chemotherapy on Colo 205 cells with respect to (A) PR, (B) Colony formation




PR changes from 0.95 to 0.38 (**)

**Fig. 6**: Chemotherapy promotes a transition from (A) quiescence to (B) growth (a double-edged sword)

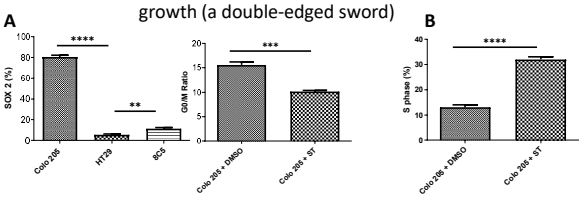
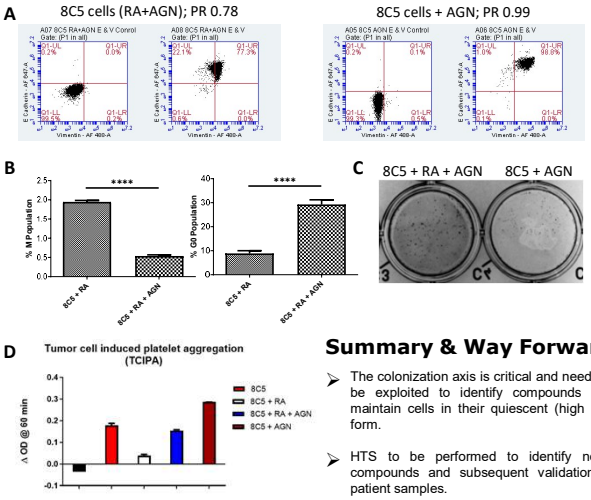



**Fig. 7**: AGN193109 reverses RA mediated PR change by (A) shifting cells to a more mesenchymal form, (B) slowing growth and (C) promoting quiescence and (D) changing functional properties




## Summary & Way Forward

- The colonization axis is critical and needs to be exploited to identify compounds that maintain cells in their quiescent (high PR) form.
- HTS to be performed to identify novel compounds and subsequent validation in patient samples.

1. Cancers, 2022, 14, 889
2. Nature Reviews Clinical Oncology, 2019,16, 185
3. Nature Genetics, 2019,51, 1113
4. EACR 2021, Poster #v-0134
5. PCT application published # WO 2022/059026 A1
6. PS application #202141061794