Cell “phagocytosis” - a novel approach in cancer diagnostics

Brill-Karniely Y.*, Dror D, Duanis-Assaf T., Goldstein Y., Schwob O., Jaber M., Loyfer N., Benyamini H., Bielenberg D., Kaplan T., Buganim Y, Reches M., **Benny O**
Institute for Drug Research, The School of Pharmacy, Faculty of Medicine, The Hebrew University of Jerusalem, Jerusalem 9112001, Israel.

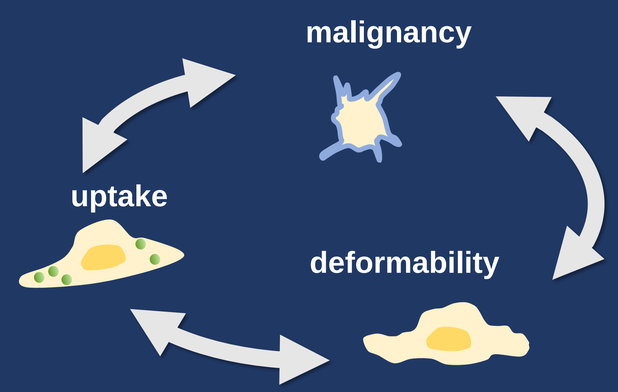
Introduction

Personalized therapy and diagnostics in cancer is often limited by the dynamic nature of cancer cells, their high heterogeneity and the dependence on specific biomarkers. This, making the data relevant for only a narrow time frame throughout the disease and for small sub-population patients.

Mechanical properties of cancer cells, i.e. their stiffness and their ability to change shape and deform are known to play a key role in cancer progression. Currently there are no tests for diagnostics based on that property. Mechanical measurements of cells are limited by a high degree of complexity making it not suitable for high throughput.

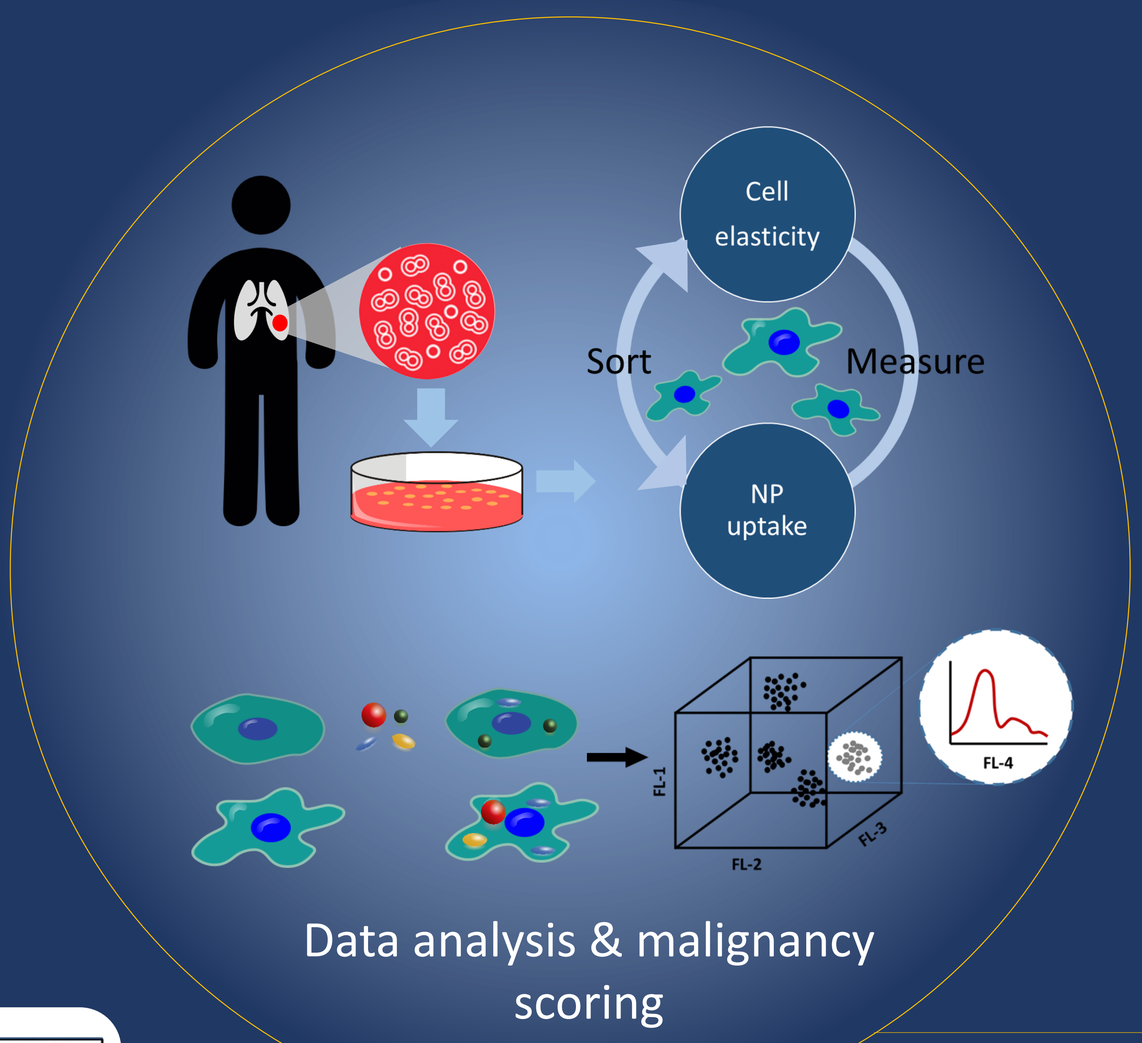
In our recently published paper in *Science Advances* we present a Triangular Correlation between cell deformability, “phagocytic capacity”, and cancer aggressiveness, suggesting that phagocytic measurements (particle uptake) can be a surrogate marker for malignancy.

Mechanical Targeting as a principle for cancer selectivity

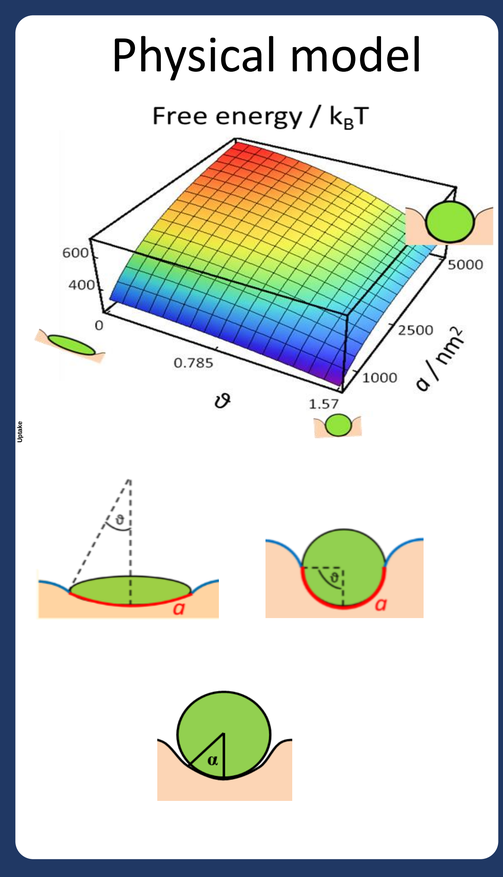


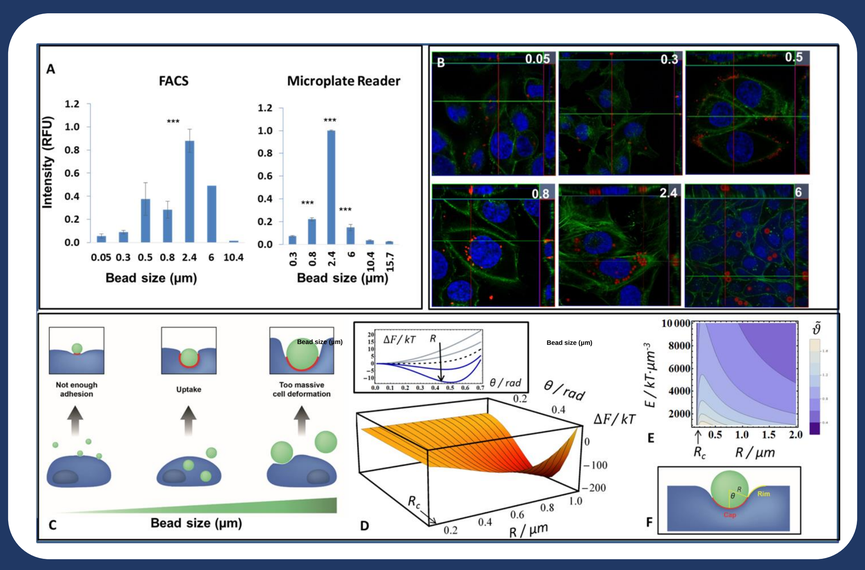
Results & Discussion

The Triangular Correlation was proved in human prostate cancer cells with different malignancy potential, and in human bladder cancer and melanoma cells that were sorted into subpopulations based solely on their phagocytic capacity. The more phagocytic subpopulations showed elevated aggressiveness ex vivo and in vivo. The uptake potential was preserved, and differences in gene expression and in epigenetic signature were detected. In all cases, enhanced phagocytic and aggressiveness phenotypes were correlated with greater cell deformability and predicted by a computational model. Our multidisciplinary study provides the proof of concept that phagocytic measurements can be applied for cancer diagnostics and precision medicine.

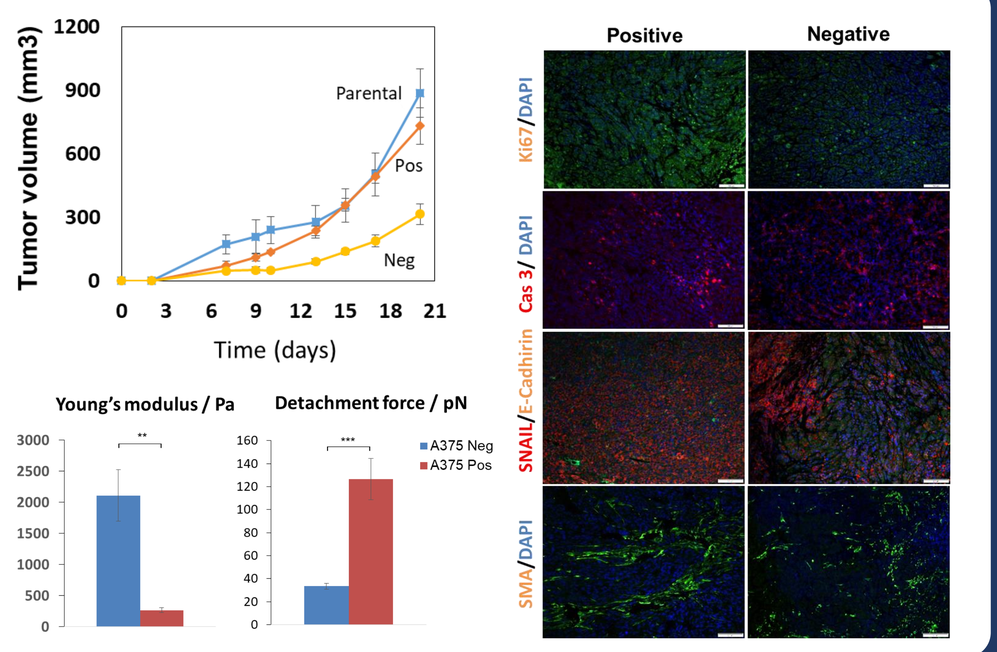


Physical model





Uptake of beads by cancer cells has a nonmonotonic dependence on the bead size. Cancer cell interaction with inert spherical fluorescent polystyrene beads increases with bead size, reaching a maximum around 2.4-μm bead diameter and then decreases. (A) Left: FACS analysis; average over normalized uptake values of eight cell lines. (B) Confocal fluorescent images (blue, DAPI; green, phalloidin (actin); red, polystyrene fluorescently labeled beads). Scale bars, 10 μm. (C) Physical model predicted the nonmonotonic dependence due to the interplay between adhesion and cell deformability. (D) The free energy of the wrapping model has a minimum as a function of both the bead radius and the wrapping angle, ϑ (bottom); however, for small beads of R < Rc, there is no stable adhesion [top; plots downward are for R = 0.05, 0.11, 0.14 (Rc, dashed), 0.18, and 0.23 μm]. (E) The stable adhesion angle, ϑ∼ (rad) is shown versus Young’s modulus and R. (F) Model geometry. Parameters used in plots of (D) and (E) are based on AFM calculations in cells.



Uptake-based sorted cells was validated in a xenograft study. Parental, positive (Highly phagocytic, Highly deformative), and negative (low phagocytic, low deformability) A375 cells were injected subcutaneously to athymic nude mice. Immunohistochemical staining for evaluating proliferation (anti-Ki67 in green), apoptosis [anti-cleaved caspase-3 (Cas 3) in red], and EMT (anti-SNAIL/SLUG in red versus anti–E-cadherin in green, as well as SMA in green). Positive tissues showed a higher level of proliferation (Ki67 staining), while negative tissues showed a higher level of apoptosis (cleaved caspase-3). SNAIL/SLUG versus E-cadherin show enhanced EMT in the positive tumors compared to the negative ones, while SMA values were similar. Scale bars, 100 μm.

Conclusion

- **There is a clear Triangular Correlation between cancer cell malignancy, cell phagocytosis and cell deformability.**
- **This TrC can be utilized alone with physical model to design optimal drug carrier for personalized therapy.**
- **The method may be adapted for diagnostics and for other diseases**

References

- Brill-Karniely Y.*, Dror D, Duanis-Assaf T., Goldstein Y., Schwob O., Millo T., Orehov N., Stern T., Jaber M., Loyfer N. Vosk R., Benyamini H., Bielenberg D., Kaplan T., Buganim Y, Reches M., Benny O. Triangular Correlation (TrC) between cancer aggressiveness, cell uptake capability and cell deformability. Science Advances, 2020, Vol. 6, no. 3, eaax2861
- provisional application (#62467349)

האוניברסיטה העברית בירושלים
THE HEBREW UNIVERSITY OF JERUSALEM

The Lab for Nanomedicine & Tumor Microenvironment