



Single CTC isolation for genetic analysis using Parsortix™ and VyCAP

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Introduction

Isolating individual CTCs for genetic analysis, is currently associated with significant losses of these very rare cells. Here we demonstrate the combined use of ANGLE's CTC enrichment platform "Parsortix" with VyCAP's single cell Puncher to obtain single cell genetic information.

Materials, Methods and Results

Single CTCs were isolated from whole blood according to the presented experimental flow.

Blood was collected in Transfix CTC collection tubes and spiked with pre-labelled (CellTracker Green) SKBR-3 and MCF-7 cells. Two different IF staining methods were performed.

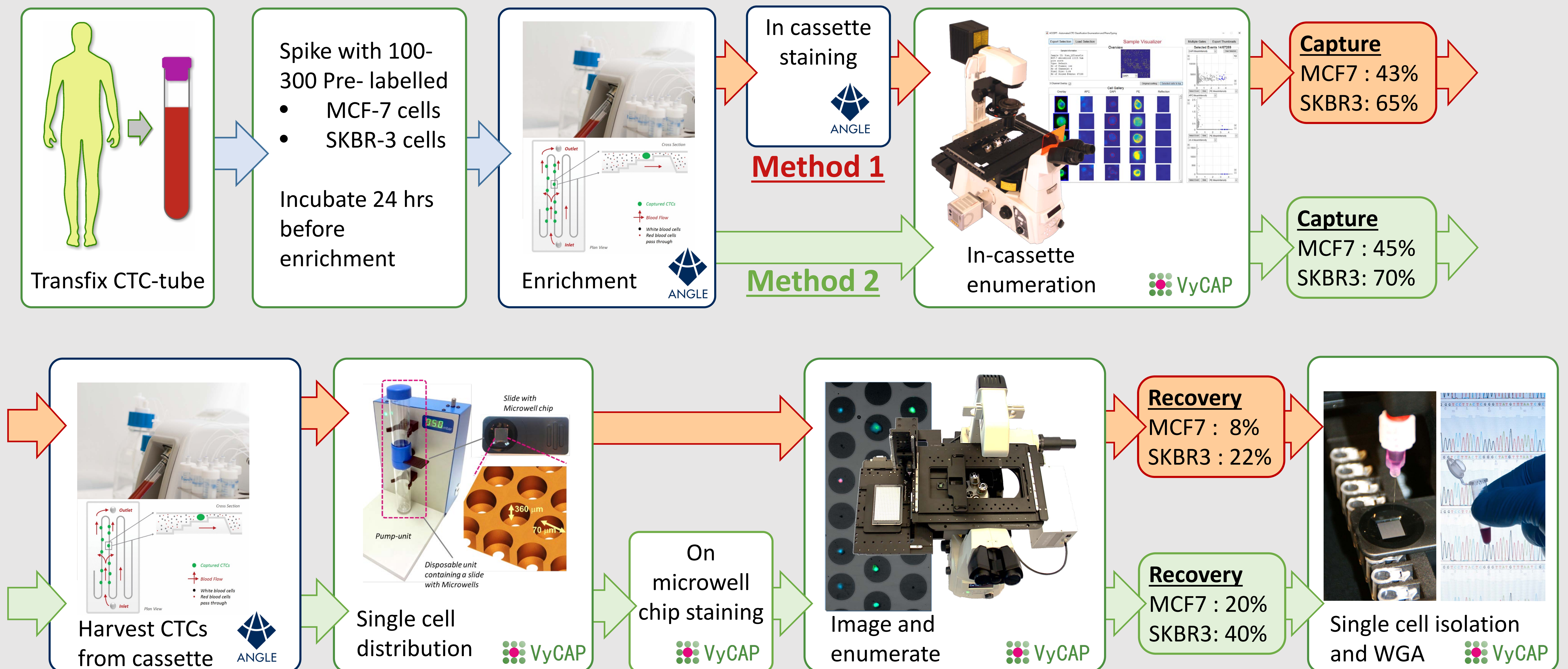
Method 1, in Parsortix cassette staining:

CTCs were captured with Parsortix and **IF stained** for Cytokeratin, CD45 and DNA **inside the Parsortix separation cassette**. CTCs were then enumerated inside the cassette before being harvested into VyCAP's microwell chip for single cell isolation.

Method 2, in microwell chip staining:

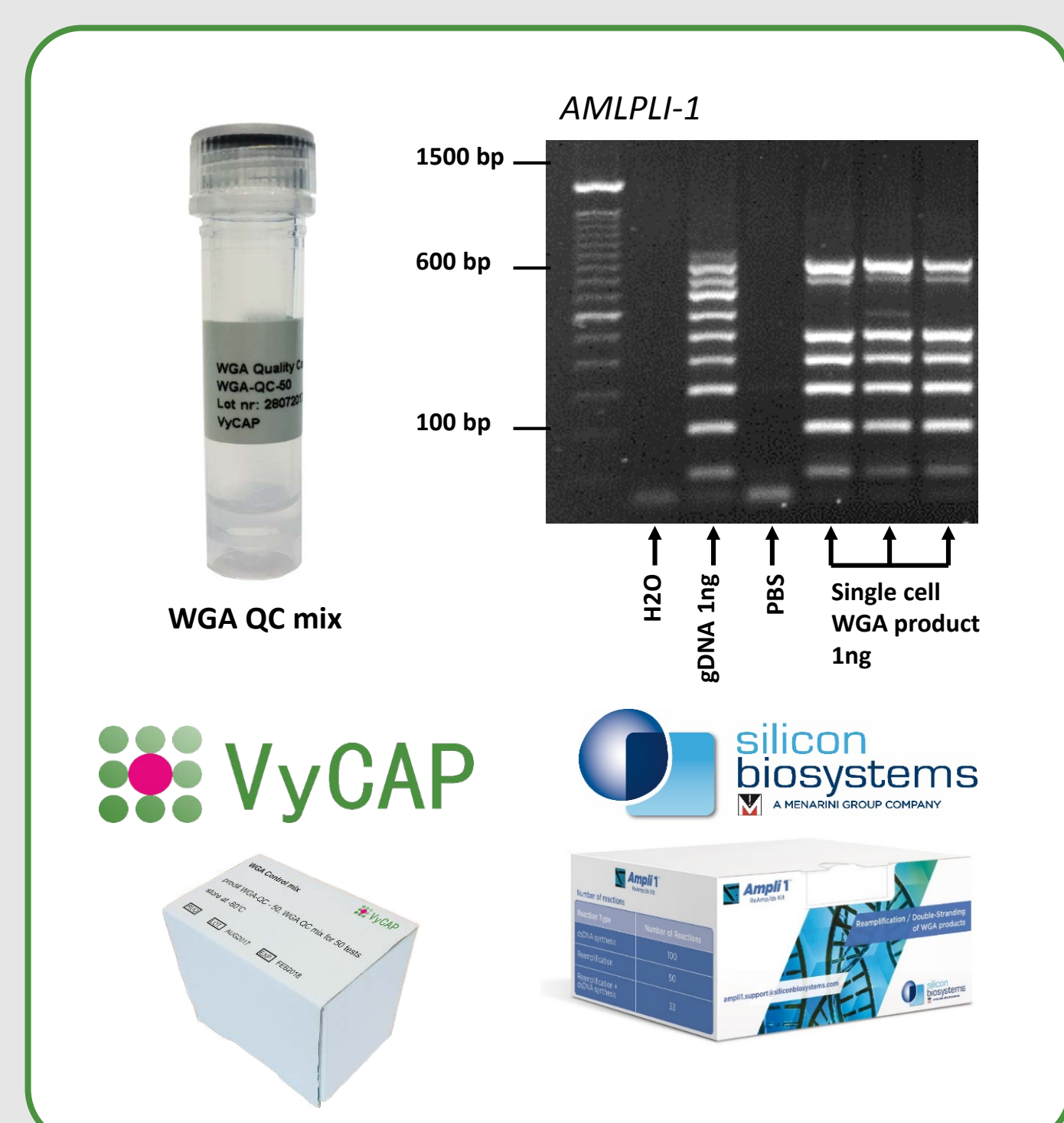
CTCs were captured with Parsortix and enumerated inside the Parsortix enrichment cassette before being harvested into VyCAP's microwell chip. **IF staining** for Cytokeratin, CD45 and DNA was performed **on the microwell chip** before single cell isolation.

Capture (enumeration inside the Parsortix cassette) and Recovery (enumeration on the microwell chip) are presented as percentages of spiked cells. After single CTC were isolated using VyCAP's Puncher platform, WGA was performed and the quality was determined using VyCAP's WGA QC mix.



WGA Quality control

WGA was performed using the AMPLI-1 WGA kit (Silicon Biosystems). The quality was determined using VyCAP's WGA control mix. Maximum DNA quality (7 bands of 7 bands visible) was obtained for all isolated single cells.



Conclusion

- The combination of Parsortix CTC enrichment with VyCAP's single cell platform is able to isolate single CTC from whole blood in a controlled way.
- Good yields of CTCs were obtained (up to 40% of spiked cells were recovered as single cells).
- Optimizing of the process is possible and could result in even higher single CTC yields.
- Maximum DNA quality obtained after WGA.
- Combination of cost effective platforms