

**Supplementary Information 1 (S1). Process time of experimental procedure**

<b>Procedure</b>	<b>Time</b>	<b>On-chip/ Off-chip procedure</b>
Leukocyte depletion procedure	50 min	Off –chip
Staining and washing	40 min	
Filtration	10 min	On self-seeding microwells
Detection	20 min	
Punching (Including alignment)	10 min  1 sec per 1 cell	On-chip
Lysis	10 min	
Neutralization	10 min	
On-chip amplification	16 h	
Inactivation	10 min	Off-chip
Purification	15 min	
qPCR	2h 30 min	

## **Supplementary Information 2 (S2) . Detailed protocol for the leukocyte depletion procedure**

RosetteSep CTC Enrichment Cocktail Containing Anti-CD36 (Stemcell technology) was used for the depletion of leukocytes in 1 ml of whole blood. The protocol was performed as described by the manufacturer. In short 50  $\mu$ l of Rossetsep reagent was added to 1 ml of whole blood spiked with 1000 MCF7 cells and incubated for 20 min after mixing. 1 ml of 2% FBS in PBS was added onto the blood to dilute. 4.5 ml of Ficoll paque PLUS (GE healthcare) was prepared in a SepMate tube (Stemcell technology). Then, 2 ml of diluted blood was carefully loaded on the top of Ficoll layer and followed by centrifugation at 1200 X g for 10 min with break-on. After centrifugation, the top layer was poured into a new tube and 4 ml of 2% FBS in PBS solution was added and mixed. Then, the tube was centrifuged at 300 X g for 8 min and the supernatant aspirated. The pellet was resuspended by pipetting in 50  $\mu$ l of 1X PBS including 1 % BSA and the staining reagents.