## Supporting information: volume dispensed

Different volumes can be dispensed from the reservoir on the chip or from larger droplets. The process and electrode actuation sequence has been described many times in the literature and in previous publications from our group (Fouillet<sup>21</sup>). The schematics below explain briefly where the different volumes can be dispensed. The volume is entirely determined by the electrode area, since the gap is constant throughout the chip. Very briefly, droplets are obtained by successive activations/deactivation of the electrodes between the reservoir (or a large electrode) and the dispense electrode.

From reservoirs "R" droplets of 256nL can be created. First, the electrode under the reservoir hole is activated, which pulls the liquid inside the chip towards the 256nL electrodes. The latter is then activated and the liquid proceeds onto it. To generate a droplet, the electrode from the reservoir is turned off which splits the liquid into a droplet on the 256nL electrode.

From each droplet, it is then possible to dispense smaller droplets, either of 64nL or 36nL. Larger volumes are obtained by merging several droplets together.



## Supporting information: expected Ct difference calculation for figure 3b

Extraction in tube on 10,000 cells (1000 cells/uL and 10uL were used): the mRNA collected was incorporated to the qRT-PCR mix (10 uL total volume)

Extraction on chip on 64 cells (1000 cells/uL and 64nLwere used): the mRNA collected on chip in 64nL was then incorporated to the qRT-PCR mix (10 uL total volume)

Supposing the same extraction yield for the two methods, the ratio between the initial numbers of cells  $N_{01}/N_{02}$  gives the ratio between the mRNA amounts and also between the cDNA at the beginning of the PCR.

During a qPCR, the Ct is used to compare reactions. The following equation is used (PCR efficiency set at 100%):  $N = N_0 x 2^{Ct}$ For all Ct, the numbers of DNA copy N are the same.  $N_{01} x 2^{Ct1} = N_{02} x 2^{Ct2}$ A Ct difference,  $\Delta$ Ct, can then be linked to the initial copy difference:  $\Delta$ Ct=Log( $N_{01}/N_{02}$ )/Log(2)

We know  $N_{01}/N_{02}$  which is 10,000/64=157

Therefore,  $\Delta Ct \sim 7.2$ 

This difference is close to the one found experimentally, which is 7 ( $N_{01}$ ~24 and  $N_{02}$ ~31)