Supplementary Figures

Fig.S1. Microfluidic device fabrication and design elements. **A-E** The manufacturing process of the microfluidic device involved elastomer casting on a two layer SU8 master and bonding of the elastomer to a glass slide. **F**. Fluids are introduced in the chip using pipette tips inserted in the elastomer. **G**. A sieve structure was used at the inlet for cells for removing debris from the cell suspension and to avoid channel clogging. **H**. A weir structure was used to trap silica particles in the separation column. **I-J** Nonspecific RNA binding inside the microfluidic has been avoided by treating the surface of the microchannels with fluorosilane. The efficacy of the treatment was evaluated by using a ribo-green RNA specific dye, following RNA solution flow through the device.

Fig.S2. Silica column characterization. **A**. Microscopy image of the silica particles inside the device. Scale bar is 500µm. **B**. Effect of silica type on the RNA isolation. The same amount of total RNA (10ng) was captured on microfluidic separation columns filled with three different types of silica (silica #1 – catalog number 40360, silica #2 – catalog number 0507, silica #3 – catalog number 227196, all from Sigma-Aldrich). Standard gel electrophoresis was used to compare the RNA amount eluted from each column and the original amount. **C**. Effect of silica preconditioning on RNA capture/elution. Pretreatment of the silica column with magnesium salt solutions improves the efficiency of RNA recovery from solution. Pretreatment the silica using diethylpyrocarbonate DEPC does not have a significant effect on the efficiency of separation, suggesting that the silica is not RNase contaminated. **D**. Effect of elution volume on the total RNA isolated from 50 cells. A volume of 20 µL of elution solution was used for better efficiency of RNA recovery from the silica column.

Fig.S3. Quality of the RNA isolated in the microfluidic device. The quality of the total RNA isolated and purified using the microfluidic chip, form a 2000 cells sample was measured using the “pico” electrophoresis chip (Agilent Bioanalyzer). The average amount of RNA in the sample in this example was calculated to 2.3pg/cell.

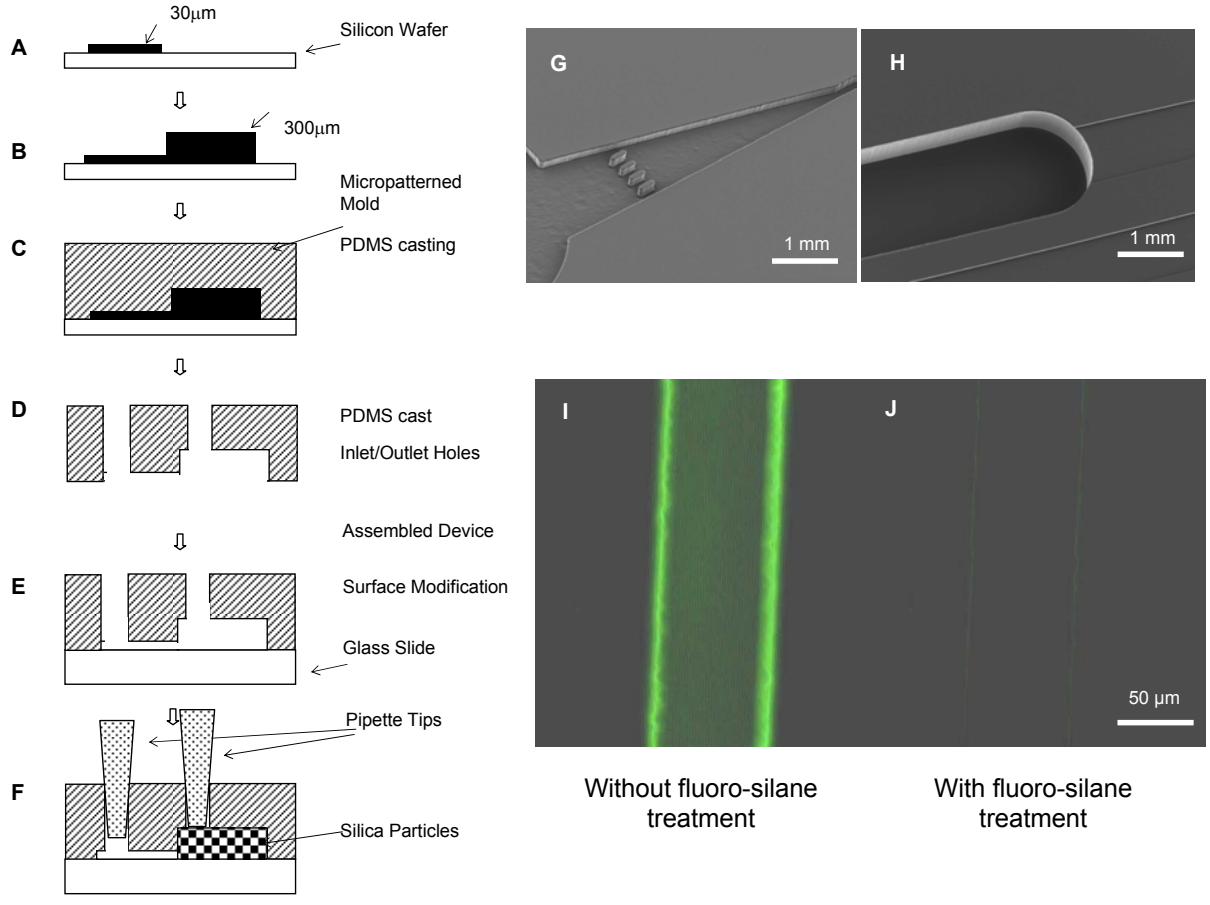
A
30μm
Silicon Wafer
B
300μm
Micropatterned Mold
C
PDMS casting
D
PDMS cast
Inlet/Outlet Holes
E
Assembled Device
Surface Modification
Glass Slide
F
Pipette Tips
Silica Particles
G
1 mm
H
1 mm
I
J
50 μm
Without fluoro-silane treatment
With fluoro-silane treatment

Fig.S1

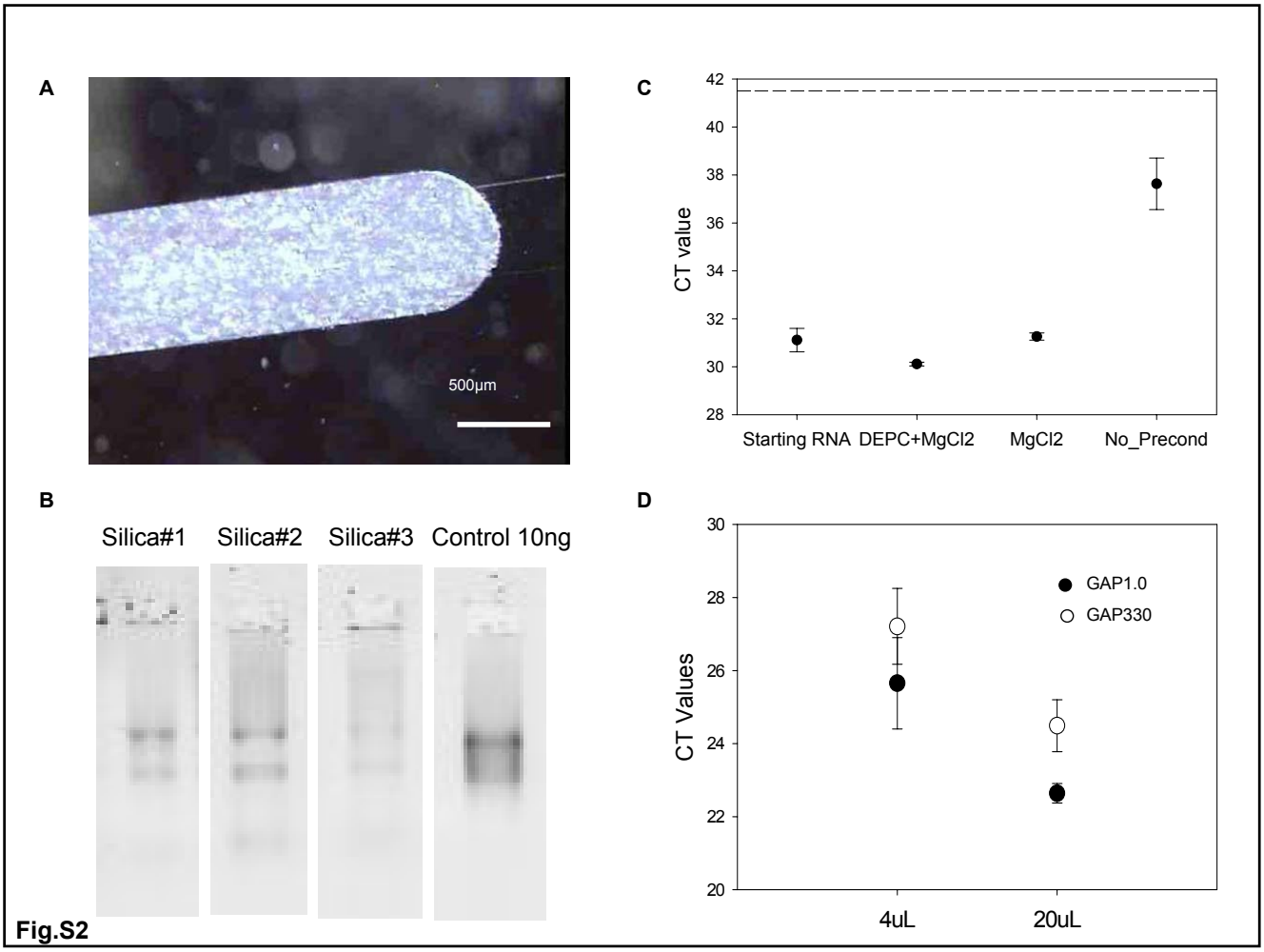
A
500µm
B
Silica#1
Silica#2
Silica#3
Control 10ng
C
CT value
42
40
38
36
34
32
30
28
Starting RNA
DEPC+MgCl2
MgCl2
No_Precond
D
CT Values
30
28
26
24
22
20
GAP1.0
GAP330
4uL
20uL

Fig.S2

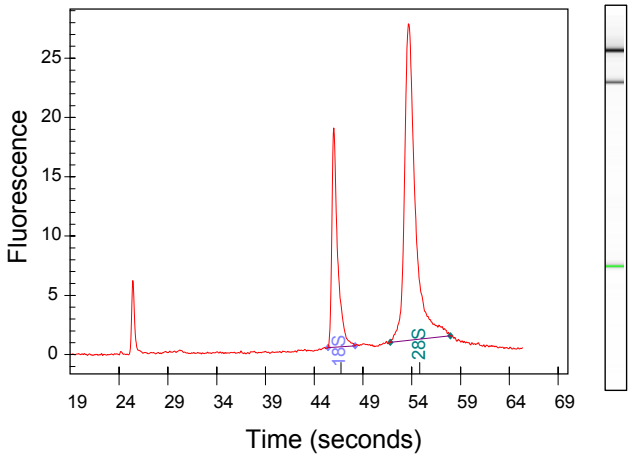
Fluorescence
25
20
15
10
5
0
19
24
29
34
39
44
49
54
59
64
69
Time (seconds)
18S
28S

Fig.S3