GENERAL SHORT MANUAL

Cell-IN
enhance cellular uptake

I. MEDIUM PREPARATION

HYPERTONIC MEDIUM:

Dissolve Cell-IN powder in a serum-free medium in amount according to **Table 1**.
If possible, use an ultrasonic bath (36°C) for 30 minutes to enhance the effectiveness

HYPOTONIC MEDIUM:

Mix serum-free low glucose medium with sterile water at the rate of **6:4**

II. DELIVERY PROTOCOL

1. Prewarm hypotonic medium and culture medium
2. Prepare a working solution by mixing the hypertonic medium with a desired compound in the ratio of **9:1**

   Please note that to obtain good efficiency, the concentration of a stock solution should be no less than 100 µM

3. Remove culture medium from cells
4. Add working solution in amount according to **Table 2A**
5. Incubate for precisely 10 minutes in 37°C

6. Remove working solution
7. Add hypotonic medium in amount according to **Table 2B**
8. Incubate for precisely 2 minutes in 37°C

9. Remove the hypotonic medium
10. Add culture medium in amount according to **Table 2C**
11. Incubate for at least 10 minutes in 37°C

12. Change the culture medium to a medium in which measurements are conducted

Table 1. The recommended amount of a medium for dissolving Cell-IN

PRODUCT NAME	The volume of medium [mL]
Cell-IN Basic 0.1	0.1
Cell-IN Basic 0.5	0.5
Cell-IN Basic 1	1
Cell-IN Basic 5	5

Table 2. The recommended amount of working solution, hypotonic medium and culture medium for different plates

CULTURE PLATE	Surface area per well [cm²]	A. The volume of working solution per well [µL]	B. The volume of hypotonic medium per well [µL]	C. The volume of culture medium per well [µL]
96-well	0.3	4	90	100
24-well	1.9	24	460	500
12-well	3.8	48	900	1 000
8-well	0.9	12	280	300
6-well / 35 mm	9.4	125	1 800	2 000
60 mm	21	280	4 600	5 000