

- + Delivery of functional protein, antibody & peptide into the cell
- + Delivery to a wide variety of cells including primary cells
- + Highly efficient transfer
- + Easy-to-use

The delivery of protein, antibody and peptide using **PULSin™** represents a powerful approach for functional studies. For example, **PULSin™** enables the study of lethal proteins by controlling the level and time course of protein delivery into the cells. Similarly, delivery of blocking antibodies may provide additional information to traditional RNA interference experiments. With **PULSin™**, it is possible to target intracellular proteins with antibodies in living cells without fixation.

+ Efficient delivery of proteins and antibodies to the cytoplasm

PULSin™ was shown to deliver R-phycoerythrin, a fluorescent protein (240 kD) to the cytoplasm of up to 98% cells. As shown in Figure 1, the protein is evenly distributed in the cytoplasm and excluded from the nucleus due to its large size.

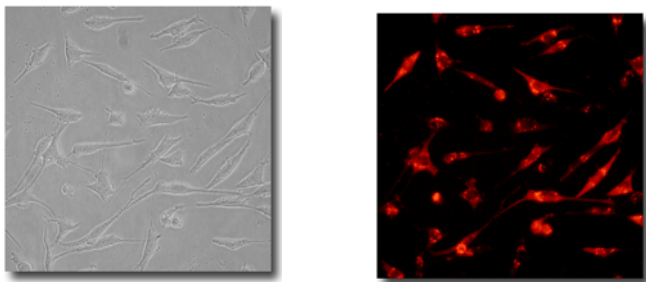
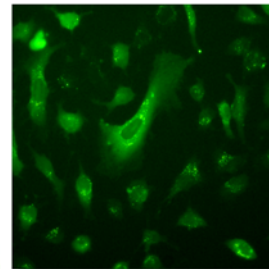


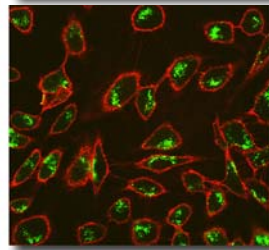
Fig. 1. **PULSin™**-mediated intracellular delivery of R-phycoerythrin to NIH-3T3 cells. R-phycoerythrin (1 µg) was complexed with 4 µl of **PULSin™** for 15 min and added to NIH-3T3 cells in a 24-well plate. Live cells were observed by fluorescence microscopy after 16 h.

Antibodies were also successfully delivered to HeLa cells and able to recognize their target protein inside the cytoplasm.



For example, **PULSin™** permits the delivery of FITC-labeled anti-α-tubulin to the cytoplasm of 85% HeLa cells (Fig.2).

Fig. 2. Delivery of a fluorescein-conjugated anti-α-tubulin antibody with **PULSin™** to HeLa cells.

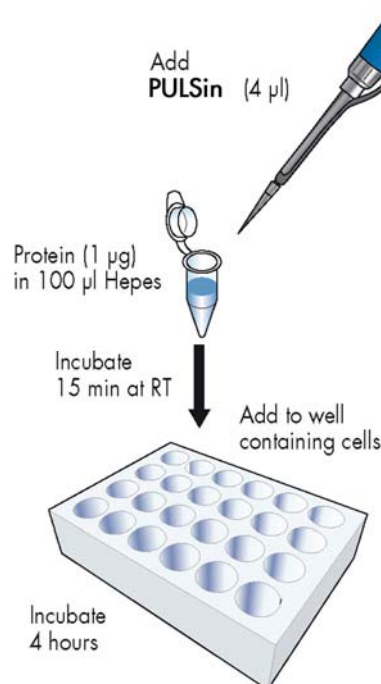


Similarly, anti-giantin Alexa Fluor® 488 was delivered to the cytoplasm of 98% of live HeLa cells, labeling the Golgi apparatus (Fig.3).

Fig. 3. Golgi labeling (green) of HeLa cells 24 h after delivery of 1 µg Alexa Fluor® 488 anti-Giantin using **PULSin™**. Plasma membrane was stained with ConA-rhodamine. Cells were observed by confocal microscopy.

+ **PULSin™** is easy to use and fast

PULSin™ will save you time and effort compared to other techniques using viral transduction or chemical conjugation.



+ **PULSin™** reagent is ready-to-use and provided with a dilution buffer and a fluorescent control protein (R-phycoerythrin).

+ The protocol is fast: simply add the reagent to the protein, incubate and add to the cells.

+ Cells can be analyzed starting 4 hours post-delivery.

+ Highly efficient transfer

The comparison of PULSin™ with two other protein delivery reagents showed a higher efficiency of protein delivery (Figure 4). Moreover, the amount of protein delivered per cell was higher with PULSin™ as measured for R-phycoerythrin protein and for FITC- α -tubulin antibody (Fig. 5).

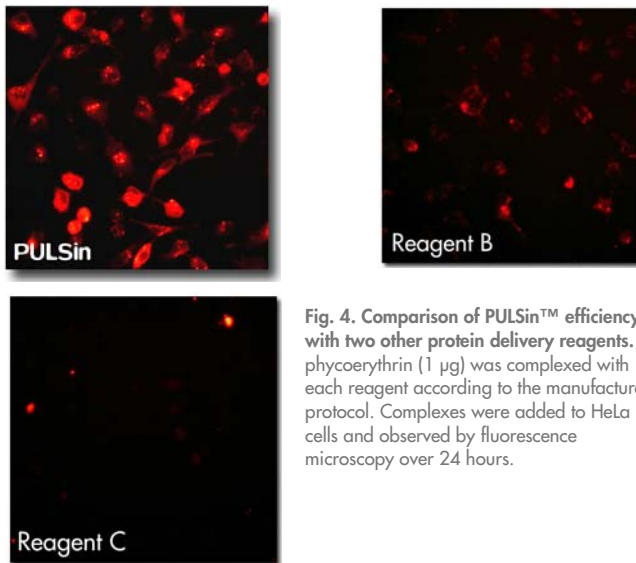


Fig. 4. Comparison of PULSin™ efficiency with two other protein delivery reagents. R-phycoerythrin (1 μ g) was complexed with each reagent according to the manufacturer's protocol. Complexes were added to HeLa cells and observed by fluorescence microscopy over 24 hours.

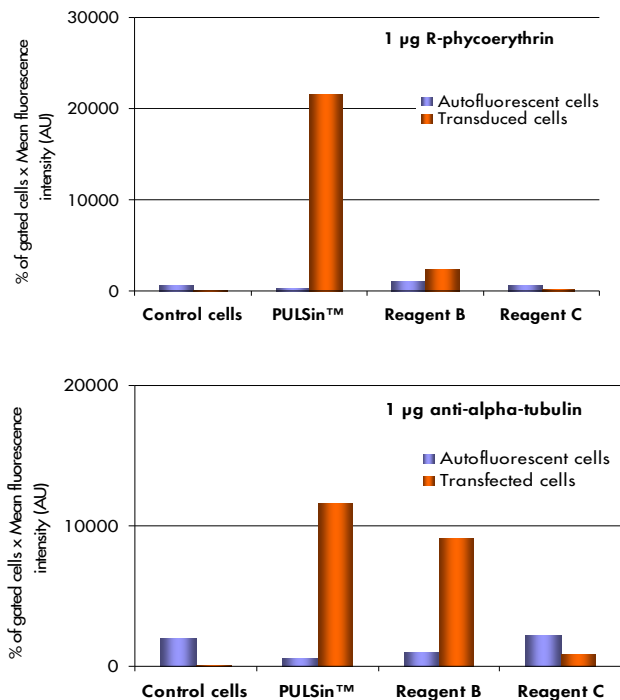


Fig. 5. FACS analysis of HeLa cells after delivery of R-phycoerythrin (upper graph) or FITC- α -tubulin antibody (lower graph) with PULSin™ or with competitor reagents B and C. Data are presented as histograms of the mean intensity of fluorescence for each cell population.

+ Delivery to a wide variety of cells

PULSin™ was shown to deliver proteins and antibodies to a large variety of cell lines and primary cells (Table 1, Fig. 1-3).

Table 1. Efficiency of R-phycoerythrin delivery using PULSin™ in selected cells.

Adherent cell lines			
3T3 L1	60-80%	HepaRG	60-70%
A549	80%	HepG2	20%
BHK-21	30-40%	MCF-7	50-60%
CaSki	80-90%	MLE-15	60-75%
CHO	80-90%	NIH-3T3	90-98%
CV-1	50%	RAW 264.7	40-50%
HEK-293	45-55%	Si Ha	60-70%
HeLa	80-90%		
Suspension cell lines			
HEK -293	30-40%	K-562	20-30%
Jurkat	20-30%	THP-1	10%
Adherent primary cells			
Primary human fibroblasts			60-70%
Primary human hepatocytes			50-40%
Primary human keratinocytes			55-70%
Primary human visceral preadipocytes			60-75%

Product	Cat N°	Reagent	Nb. of reactions in 6-well plates
PULSin™	501-01*	0.1 ml	6
	501-04*	0.4 ml	24
	501-16**	4 x 0.4 ml	96

* This kit contains 20 μ g of R-Phycoerythrin (positive control) and 20 ml of HEPES dilution buffer.
 ** This kit contains 20 μ g of R-Phycoerythrin (positive control) and 4 x 20 ml of HEPES dilution buffer.

For additional information, please contact our technical support at www.polyplus-transfection.com

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