THP-1 monocytes: transfection of siRNA

VG-01LB-00, VG-01LB-01

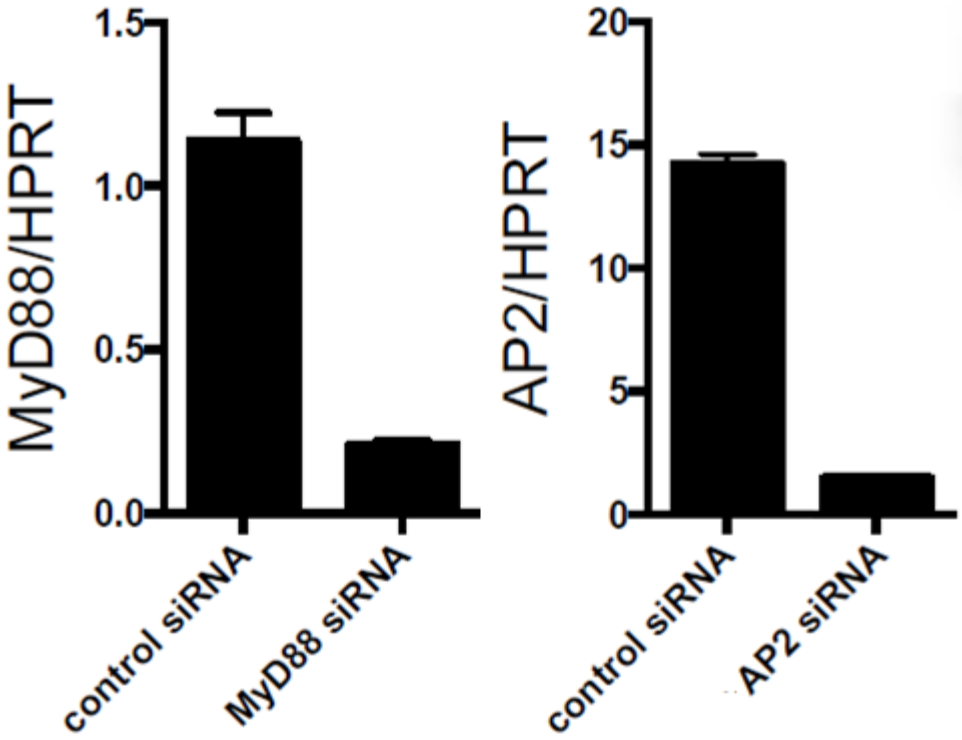


Fig. 1: Knockdown of MyD88 and AP2 in THP-1 monocytes after siRNA transfection with Viromer® GREEN.

- 12-well plate format set-up
- undifferentiated THP-1 monocytes
- MyD88 and AP2 siRNA conc.: **25nM**
- analysis by qPCR 72h post transfection

Up to 90% KD

Data from K. Pelka, University Hospital Bonn (Germany)

THP-1 monocytes: transfection of siRNA

VG-01LB-00, VG-01LB-01

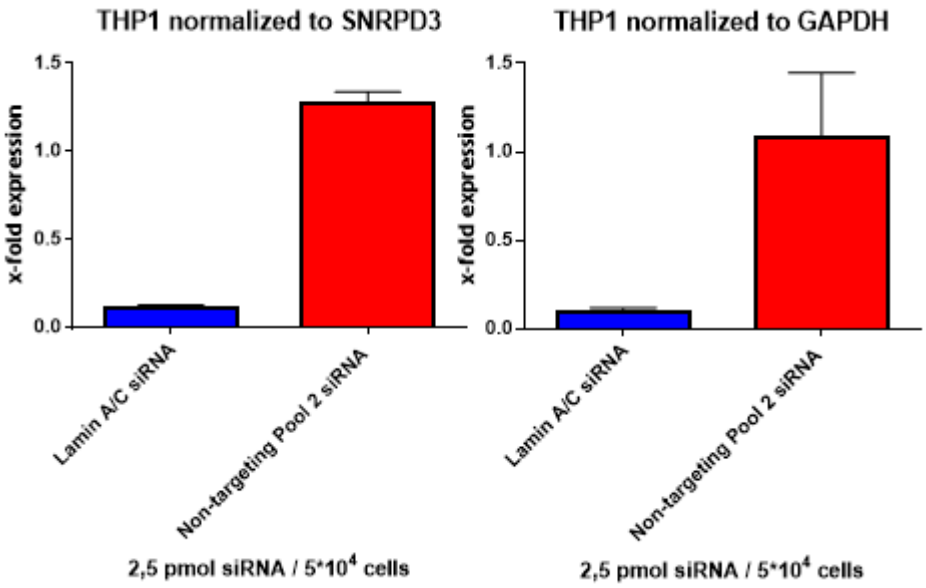


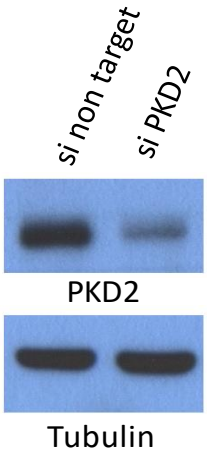
Fig. 2: Knockdown of Lamin A/C in undifferentiated THP-1 monocytes after siRNA transfection with Viromer® GREEN.

- 96-well plate format set-up, 50,000 cells/well
- undifferentiated THP-1 monocytes
- Lamin A/C siRNA conc.: **25nM**
- analysis by qPCR

Up to 90% KD

Data from J. Schupp, University Mainz (Germany)

THP-1 monocytes: transfection of siRNA

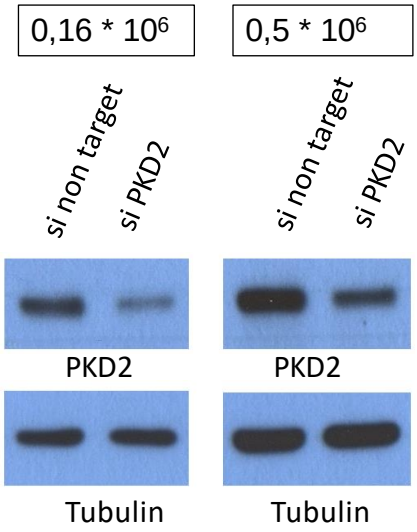


- 12-well format - 500,000 cells/well
- RPMI medium + 10% FCS
- siRNA conc. **100nM**
- 2h post-transfection: induction of differentiation with 500 nM PDBu (in complete medium)
- change of medium after 18h

VB-01LB-00, VB-01LB-01

Fig. 3: Knock-down of PKD2 in THP-1 monocytes transfected before differentiation with Viromer® BLUE.

Important note:
Time for complexation only 8 min at RT!



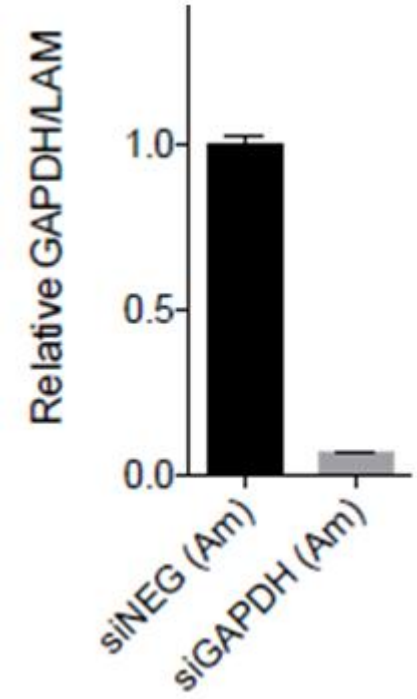
- 12-well format
- 160,000 or 500,000 cells/well
- RPMI medium + 10% FCS
- Induction of differentiation with 500 nM PDBu (in complete medium)
- Change of medium after 18h
- Transfection after 24h, with **100nM** siRNA
- Change of medium 7h post-transfection

Fig. 4: Knock-down of PKD2 in differentiated THP-1 monocytes transfected with Viromer® BLUE.

"Viability of cells was excellent (>90%)."

Data from Dr. A. Hausser & S. Heine, Univ. Stuttgart, Inst. Of Cell Biology and Immunology (Germany)

THP-1 monocytes: transfection of siRNA



VB-01LB-00, VB-01LB-01

Fig. 5: Knockdown of GAPDH in differentiated THP-1 monocytes after siRNA transfection with Viromer® BLUE

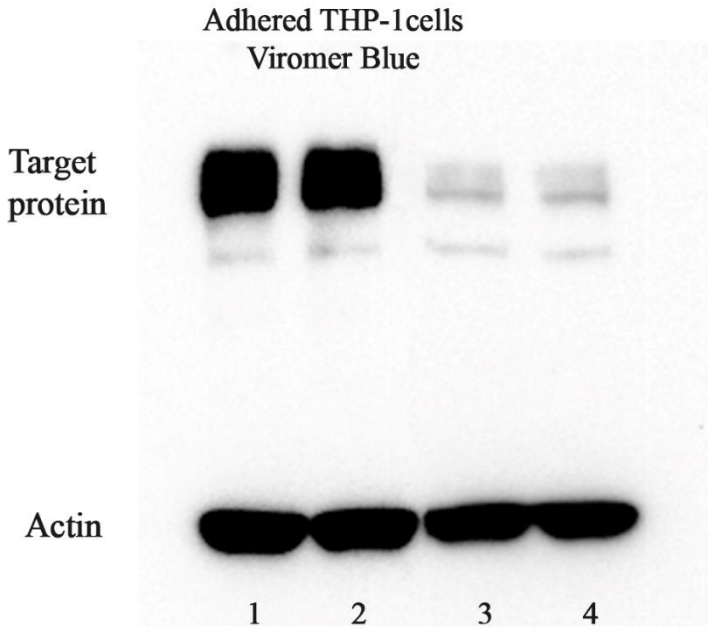
90% Knock-down

- activated for 24h with PMA (20ng/ml)
- GAPDH siRNA conc.: **20nM**
- analysis by qPCR 24h post-transfection

Data from Dr. M. Gantier, Hudson Institute (Australia)

THP-1 monocytes: transfection of siRNA

VB-01LB-00, VB-01LB-01



1, 2 - transfected with control siRNA, 50 nM
3, 4 - transfected with target siRNA, 50 nM

Fig. 6: Example of knockdown in differentiated THP-1 monocytes (adhered macrophage-like cells) after siRNA transfection with Viromer® BLUE

- 0.5 ml cells in 12-well plate wells with 1ml medium
- adhered by overnight incubation with PMA (100ng/ml)
- siRNA conc.: **50nM**
- analysis by qPCR 48h post-transfection

Data from A. Zhelankin, Bukrinsky's lab, The George Washington University (USA)

THP-1 monocytes: transfection of siRNA

VB-siBUNDLE-01

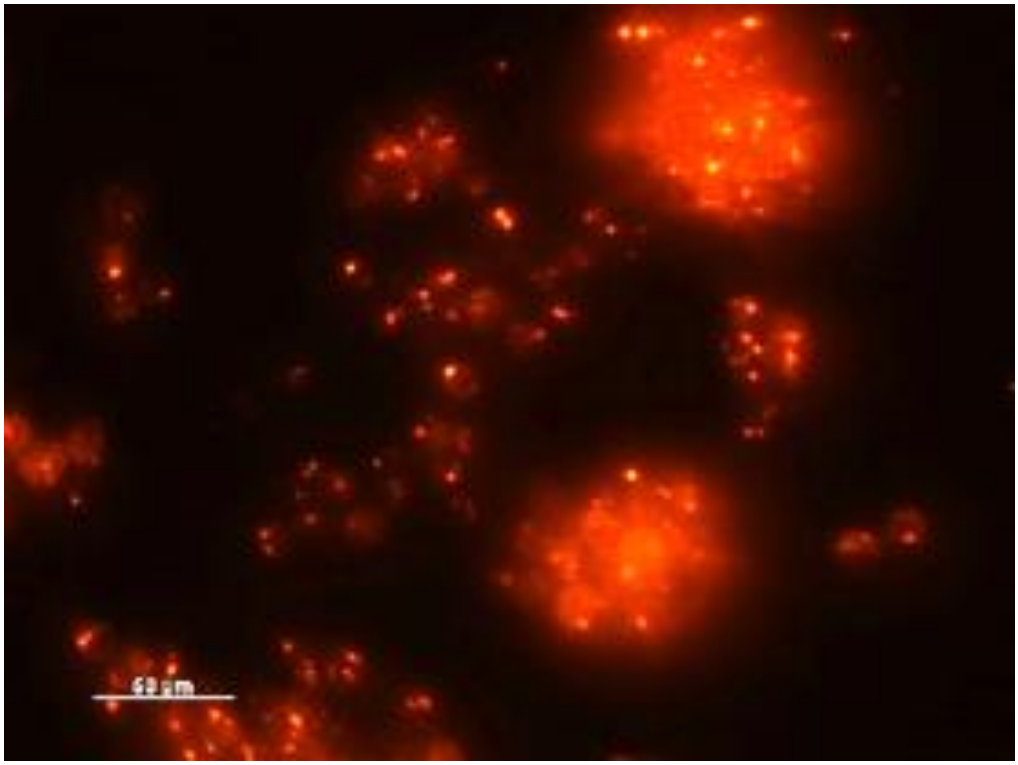
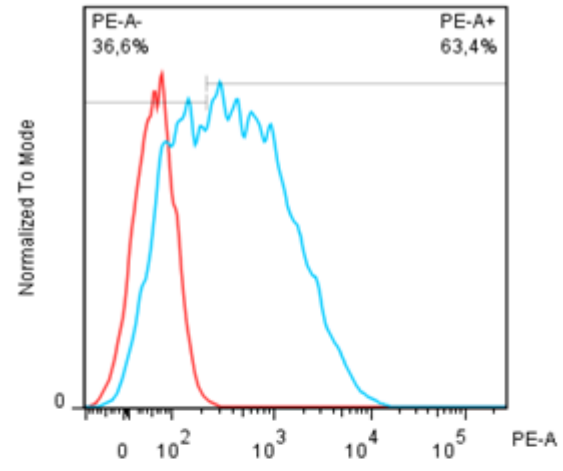
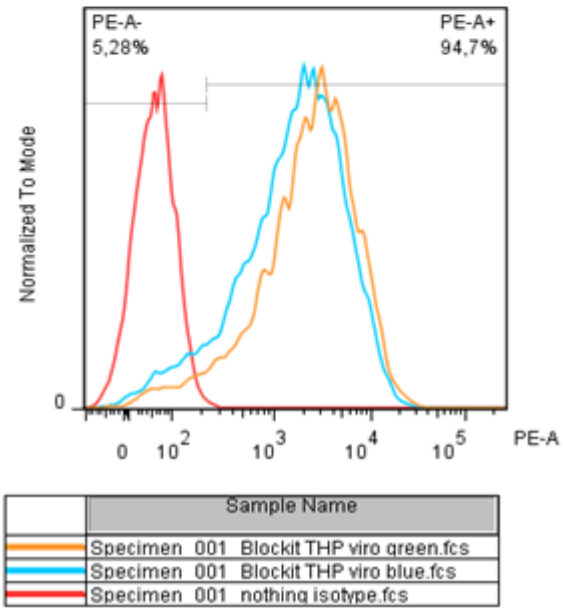


Fig. 7: Monitoring of transfection complex uptake by undifferentiated THP-1 cells transfected with a Cy3-labeled siRNA using Viromer® BLUE Start Positive Controls (fluorescence microscopy).

Internal data Lipocalyx GmbH

THP-1 monocytes: transfection of siRNA





VB-01LB-00, VB-01LB-01

VG-01LB-00, VG-01LB-01

95%
Positive cells

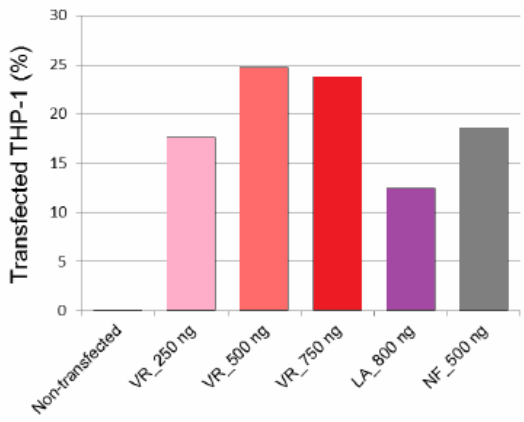
Fig. 8: Comparative uptake of a red-labeled siRNA (Blockit) in differentiated THP-1 monocytes transfected with Lipofectamine® RNAiMAX (upper graph) and Viromer® BLUE or Viromer® GREEN (lower graph).

"The transfection efficiency is very good. I am very satisfied."

Data from L. Pawig, IMCAR Aachen (Germany)

THP-1 monocytes: transfection of plasmid DNA

VR-01LB-00, VR-01LB-01

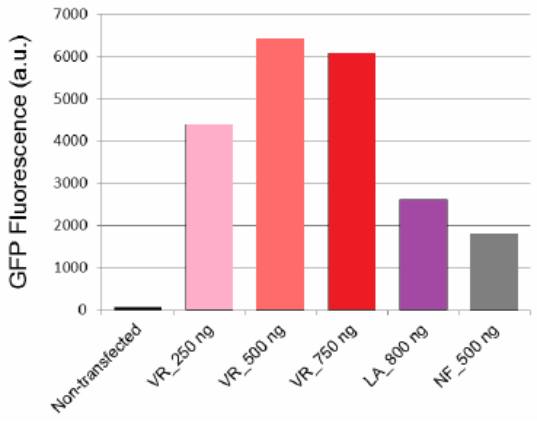


THP-1 cells (ATCC TIB-202) were transfected with:

- pmaxGF vector (Lonza; 3486 nt; stock: 1 µg/µl in 10 mM Tris pH 8.0)

using:

- Viromer RED (VR; Lipocalyx); $2.5x10^5$ cells + 250, 500 or 750 ng of pmaxGF
- Lipofectamine 2000 (LA; Invitrogen); $2.5x10^5$ cells + 800 ng of pmaxGF
- 4D-Nucleofection (NF; Lonza); $1x10^6$ cells + 500 ng of pmaxGF



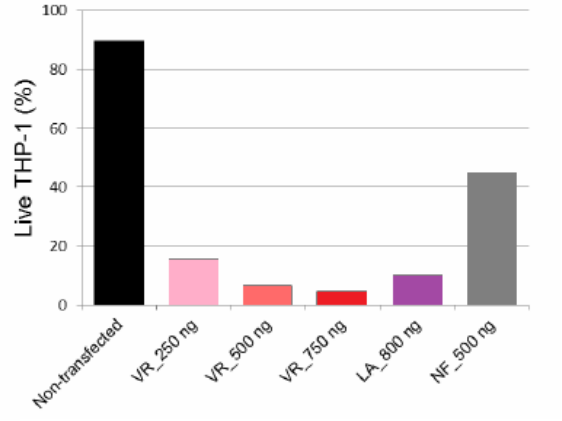
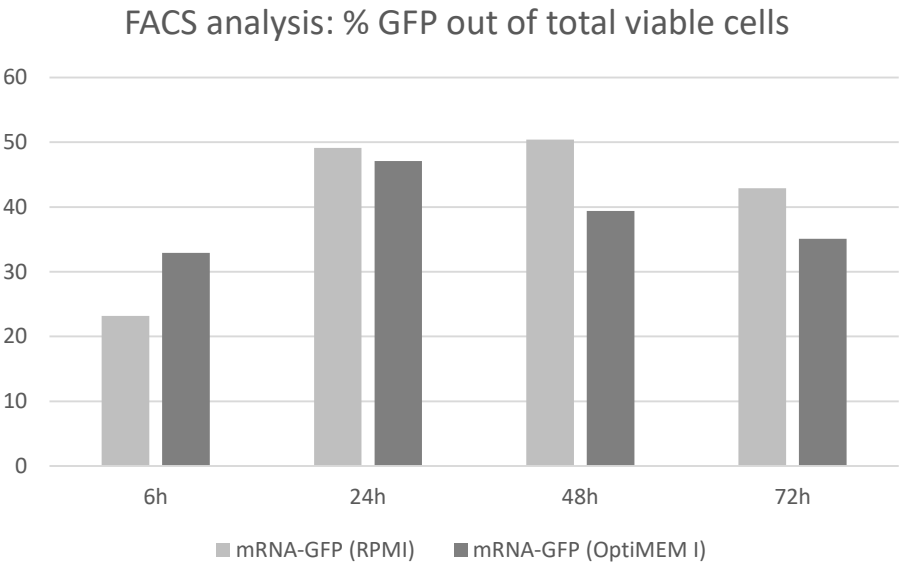


Fig. 9: Comparative transfection efficiency and cell viability of THP-1 cells transfected with a GFP-plasmid using Viromer® RED, Lipofectamine® 2000 or electroporation

Max. efficiency: 25% GFP+ cells

Anonymous data

THP-1 monocytes: transfection of mRNA



VR-BUNDLE-01

Fig. 10: Optimization of read-out endpoint and use of 2 different media for mRNA transfection in THP-1 monocytes with Viromer® RED.

Max. efficiency:
50% GFP+ cells
with RPMI at 48h

- cells seeded at 48,000 cells/96-well in either 100 µL RPMI (10% FCS) or OptiMEM® medium (0% FCS)
- transfection with mRNA-GFP/**Viromer® RED** control at 125 ng/well
- FACS analysis after 6, 24, 48 and 72h

Data from Dr. S. Fabb, Monash Inst. of Pharmaceutical Sciences, Victoria (Australia)