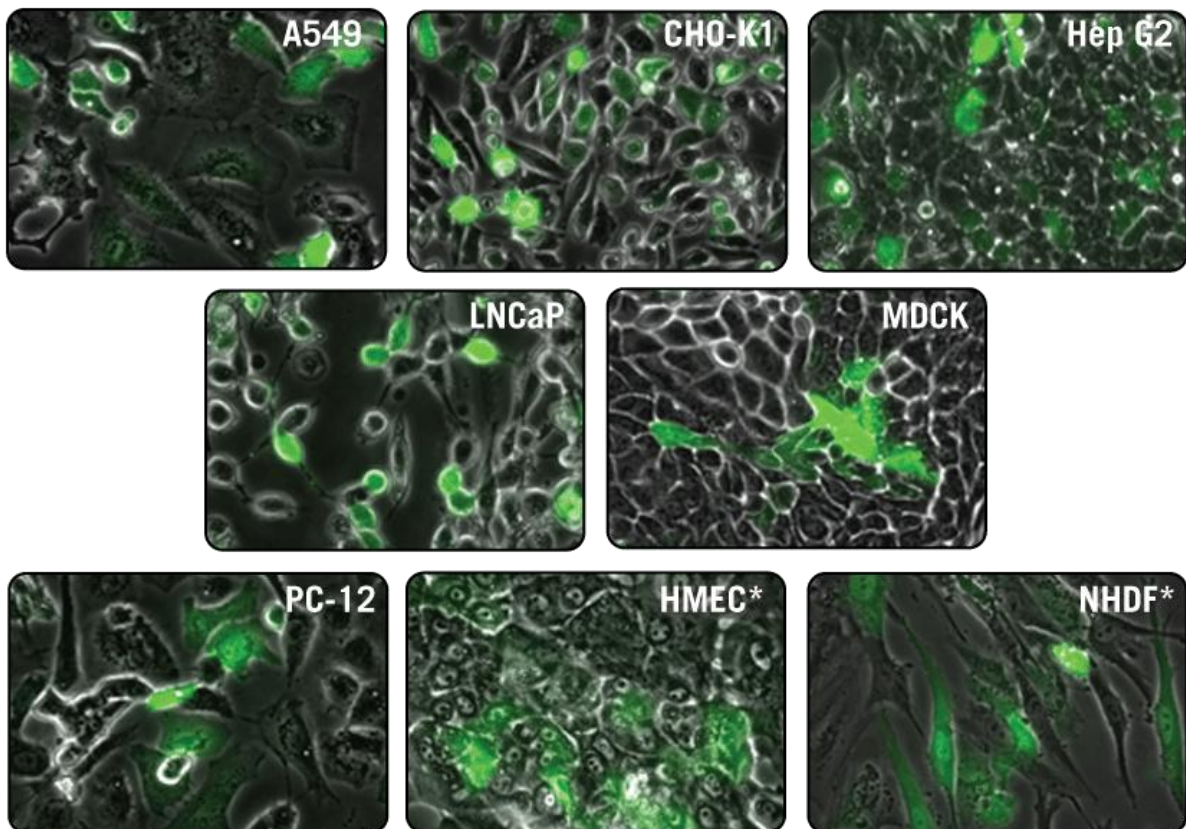


## NEW! *TransIT-X2*<sup>™</sup> Dynamic Delivery System

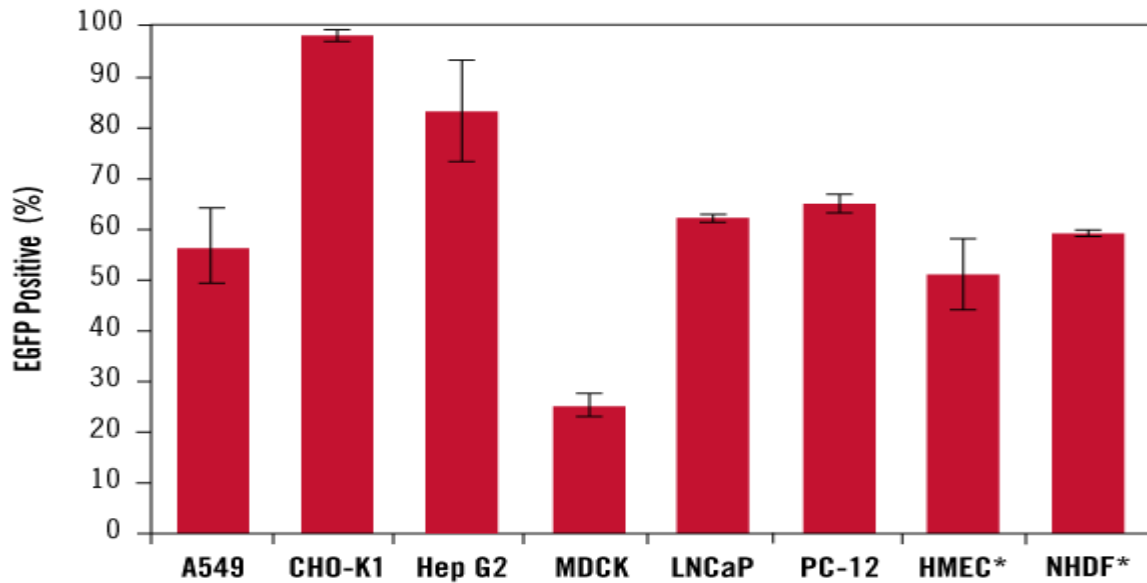
An innovative, polymeric system for efficient delivery of plasmid DNA and siRNA/miRNA

**Figure 1. Visualization of High GFP Expression using *TransIT-X2*<sup>™</sup> Dynamic Delivery System.** *TransIT-X2* Dynamic Delivery System was used to transfect plasmid DNA encoding EGFP into A549, CHO-K1, HepG2, LNCaP, MDCK, PC12, primary human mammary epithelial cells (HMEC) and normal human dermal fibroblasts (NHDF). Transfections were performed in 35 mm MatTek dishes using 4-8  $\mu$ l of *TransIT-X2* to deliver 2  $\mu$ g of DNA. Images (32X) were captured at 48 hours post-transfection using a Zeiss Axiovert S100 inverted fluorescence microscope.

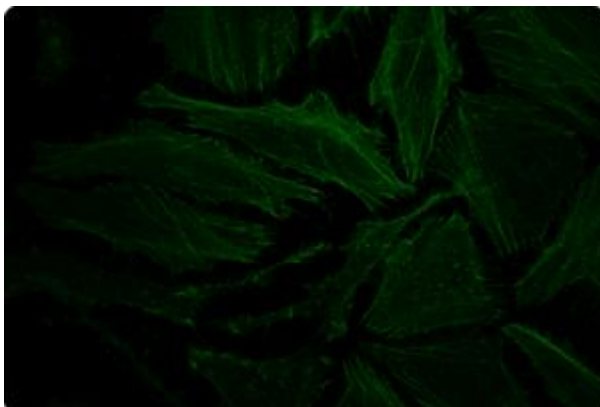


**Figure 2. High GFP Transfection Efficiency in Multiple Cell Lines and Primary Cells using *TransIT-X2*<sup>™</sup> Dynamic Delivery System.** *TransIT-X2* Dynamic Delivery System was used to transfect plasmid DNA encoding EGFP into A549, CHO-K1, Hep G2, MDCK, LNCaP, PC-12, primary human mammary epithelial cells (HMEC) and normal human dermal fibroblasts (NHDF). Transfections were performed in 96-well plates using 0.2-0.4  $\mu$ l of *TransIT-X2* to deliver 0.1  $\mu$ g of DNA (2:1, 3:1 or 4:1 reagent: DNA ratio). Triplicate wells were assayed 48 hours post-transfection using guava easyCyte<sup>™</sup> 5HT Flow Cytometer.

\*indicates primary cell types

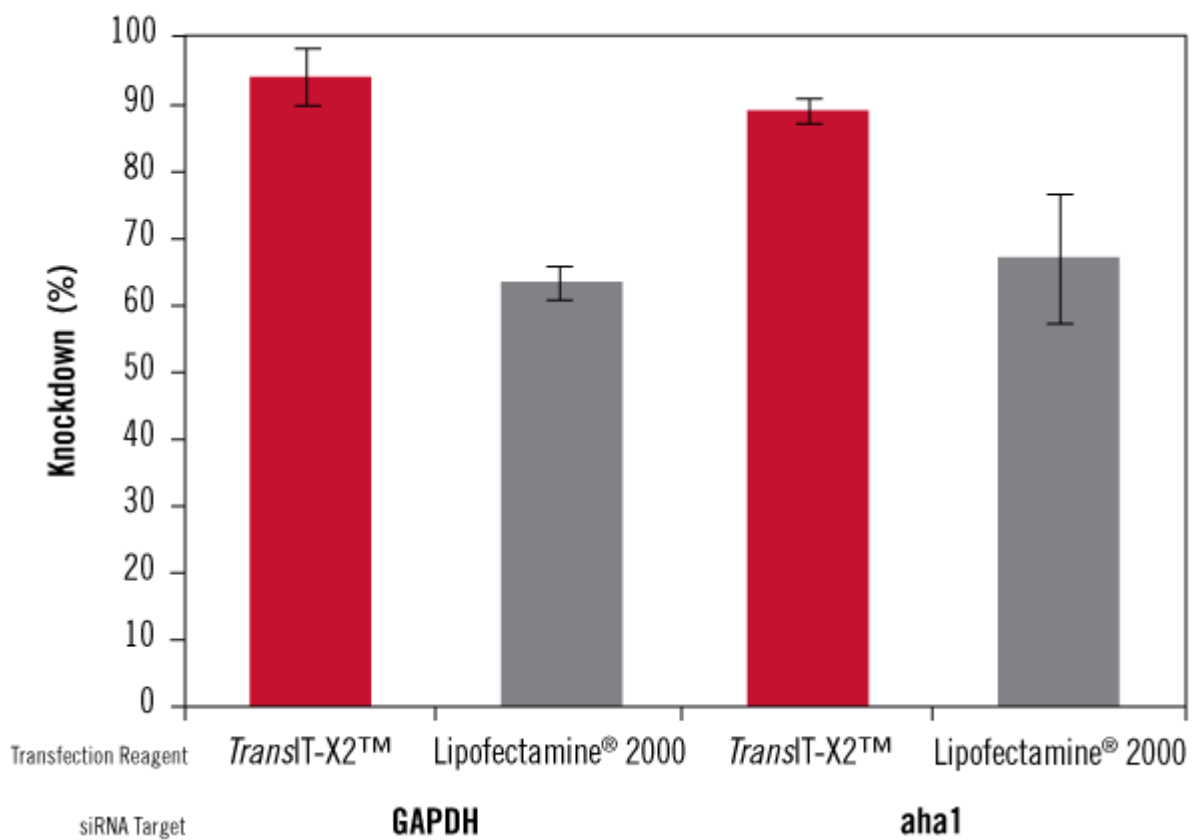


**Figure 3. Functional Co-delivery of Plasmid DNA and siRNA using *TransIT-X2*<sup>™</sup> Dynamic Delivery System.** *TransIT-X2* Dynamic Delivery System was used to transfect plasmid Cy<sup>™</sup>5 labeled DNA encoding nuclear YFP and Cy<sup>™</sup>3 labeled siRNA into HeLa cells. Transfection was performed in a 6-well plates with Poly-L-Lysine (PLL) coated coverslips using 4  $\mu$ l of *TransIT-X2* to deliver 2  $\mu$ g of DNA and 25 nM siRNA (2:1 reagent:DNA ratio). Actin cytoskeleton was stained using Alexa Fluor<sup>®</sup> 350 Phalloidin. Image (63X) were captured at 24 hours post-transfection using a Nikon A1R confocal microscope. Image key: yellow (nuclear YFP), blue (Cy5 labeled DNA), red (Cy3 labeled siRNA), green (actin cytoskeleton)



**Figure 4. *TransIT-X2*<sup>TM</sup> Dynamic Delivery System Achieves Higher Knockdown than Lipofectamine<sup>®</sup> 2000.**

*TransIT-X2* Dynamic Delivery System and Lipofectamine 2000 Transfection Reagent were used to transfect siRNA targeting endogenous proteins - GAPDH, aha1 or non-targeting control in normal human dermal fibroblasts (NHDF). Cells were transfected in a 6-well plate using 4  $\mu$ l of *TransIT-X2* or 6  $\mu$ l of Lipofectamine 2000 and 25 nM siRNA according to each manufacturer's protocol. The amount of GAPDH or aha1 mRNA was measured relative to 18s rRNA levels using qRT-PCR and then normalized to the mRNA levels of the non-targeting control, 48 hours post-transfection. Error bars represent the standard deviation of triplicate wells.



**Figure 5. Effective miRNA Delivery using *TransIT-X2*<sup>TM</sup> Dynamic Delivery System Yields Decreased Levels of PTK9 mRNA.**

*TransIT-X2* Dynamic Delivery System and Lipofectamine<sup>®</sup> 2000 Transfection Reagent were used to transfect Pre-miR<sup>TM</sup> hsa-miR-1 miRNA Precursor or *mirVana*<sup>TM</sup> miRNA mimic, miR-1, both known to decrease PTK9 mRNA levels. A Pre-miR negative control was transfected to assess baseline mRNA levels. T47D cells were transfected in a 12-well plate using 3  $\mu$ l of *TransIT-X2* or Lipofectamine 2000 and 50 nM miRNA according to each manufacturer's protocol. The amount of PTK9 mRNA was measured relative to 18s rRNA levels using qRT-PCR and then normalized to the mRNA levels of the negative control, 48 hours post-transfection. Error bars represent the standard deviation of triplicate wells.

