



Knock-down rate: 76%

Lanes: **1.** HeLa cells transfected with Renilla Luciferase esiRNA (RLUC, Cat.No. EHURLUC) as negative control. **2.** HeLa cells transfected with ATG5 esiRNA (Cat.No. EHU085781).

Quantitative-Immunoblotting: Lysates were separated on SDS-Page and probed with mouse anti-GFP (1:3000) (Roche) and goat anti-GAPDH (1:7000) (Acris) antibodies. The signal intensities of indicated bands were quantified using an Odyssey infrared imaging system (Li-COR) and normalized to the GAPDH signal. At 72h post transfection, a 76% ATG5-GFP protein reduction was measured between the negative control RLUC and the sample treated with ATG5 esiRNA.

HeLa cells: ATG5 was LAP tagged (including GFP) on a bacterial artificial chromosome (BAC) and stably integrated into the genome of HeLa cells (MCB: ky_6472). The BAC preserves the genomic context of the gene, thereby ensuring near physiological expression (Poser I. et al Nat Methods. 2008 May;5(5):409-15).