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## EMU016021: Smad4 (adenomatosis polyposis coli)

Validation using qRT-PCR:	
Knock-down rate:	85,94 ± 0,09
Cell line used:	Mouse ES cells (R1/E)
qRT-PCR primer used:	for: GGGTCAGGTGCCTTAGTGAC
	rev: ACACTGCCGCAGATCAAAGA

Average of 2 biological replicates

**RLUC:** Mouse ES cells (R1/E) transfected with Renilla Luciferase esiRNA (RLUC, Cat.No. EHURLUC) as negative control. **Smad4**: Mouse ES cells (R1/E) transfected with Smad4 esiRNA (Cat.No. EMU016021).

**qRT-PCR:** At 24h post-transfection, total RNA was isolated with an RNeasy Mini Kit (Qiagen) and reverse transcribed with SuperScript III reverse transcriptase (Invitrogen). Quantitative PCR reactions were carried out with the SYBR Green qPCR kit (Abgene) on a CFX96 qPCR machine (Bio-Rad). Relative mRNA levels were calculated according to the  $\Delta\Delta$ Ct method and normalized to the RLUC control; GAPDH was used as housekeeping reference gene.