Despite considerable research on K-Ras inhibitors, none had been developed until now. We synthesized nuclease-resistant synthetic miR-143 (miR-143#12), which strongly silenced K-Ras, its effector signal molecules AKT and ERKs, and the K-Ras activator SoS1. We examined the anti-proliferative effect of miR-143#12 and the mechanism in human colon cancer DLD-1 cell (G13D) and other cell types harboring K-Ras mutations. Cell growth was markedly suppressed in a concentration-dependent manner by miR-143#12 (IC50: 1.32 nM) with a decrease in the K-Ras mRNA level. Interestingly, this mRNA level was also down-regulated by either a PI3K/AKT or MEK inhibitor, which finding indicates a positive circuit of K-Ras mRNA expression. MiR-143#12 silenced cytoplasmic K-Ras mRNA expression and impaired the positive circuit by directly targeting AKT and ERK mRNAs. Combination treatment with miR-143#12 and a low-dose EGFR inhibitor induced a synergistic inhibition of growth with a marked inactivation of both PI3K/AKT and MAPK/ERK signaling pathways. However, silencing K-Ras by siR-KRas instead of miR-143#12 did not induce this synergism by the combined treatment with the EGFR inhibitor. Thus, miR-143#12 perturbed the K-Ras expression system and K-Ras activation by silencing SOS1 and resultantly, recovered the efficacy of the EGFR inhibitors. In vivo results also supported those of the in vitro experiments. The extremely potent miR-143#12 enabled us to well understand K-Ras networks and shut them down by combination treatment with this miRNA and EGFR inhibitor in K-Ras-driven colon cancer cells.