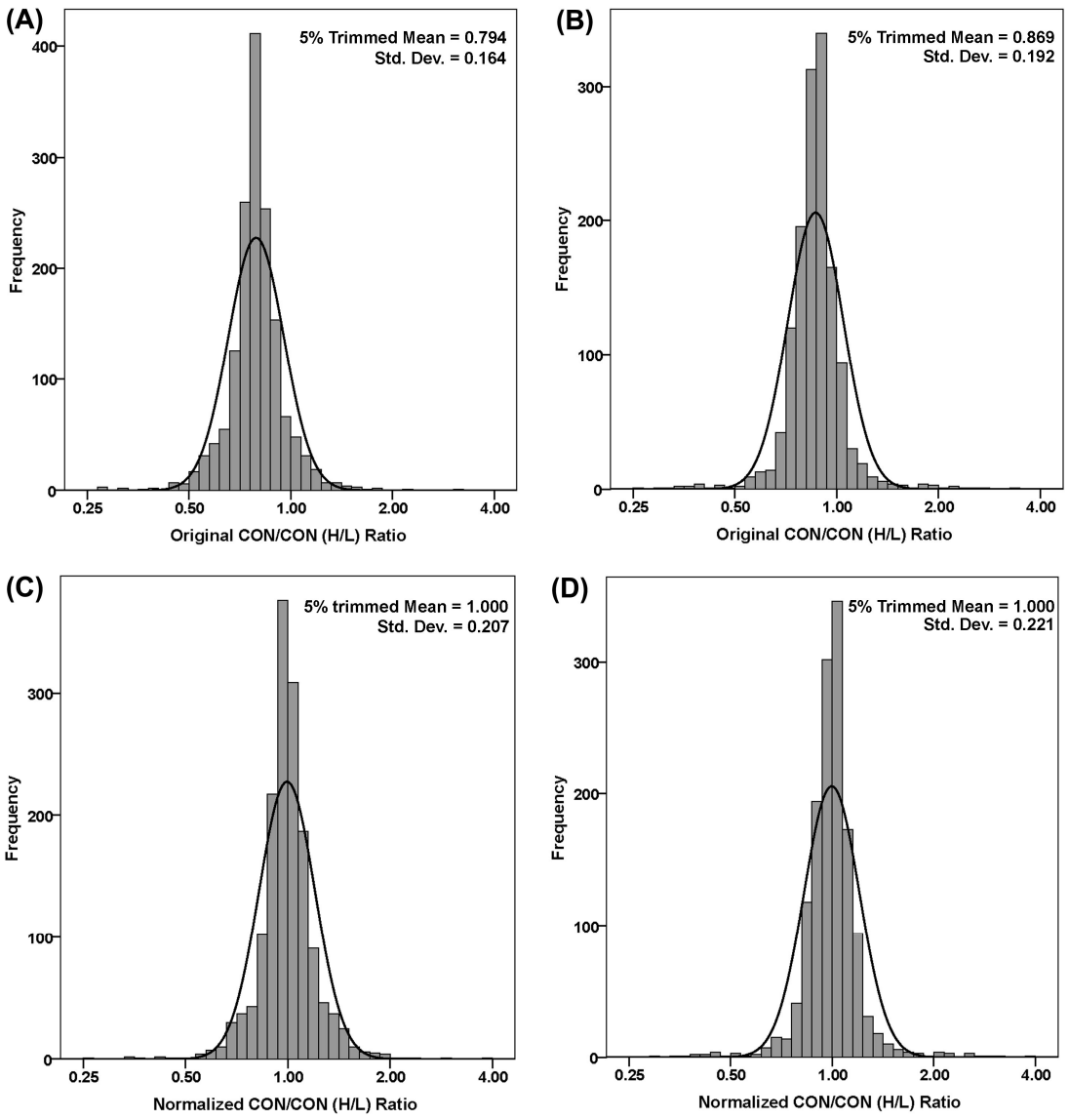

Figure S2. SILAC ratio distributions before and after multi-point normalization. The original CON/CON (H/L) ratios for both biological replicates (A & B) were normalized against their respective 5% trimmed means so that the ratios were centered around the theoretical value 1.00 (C & D).

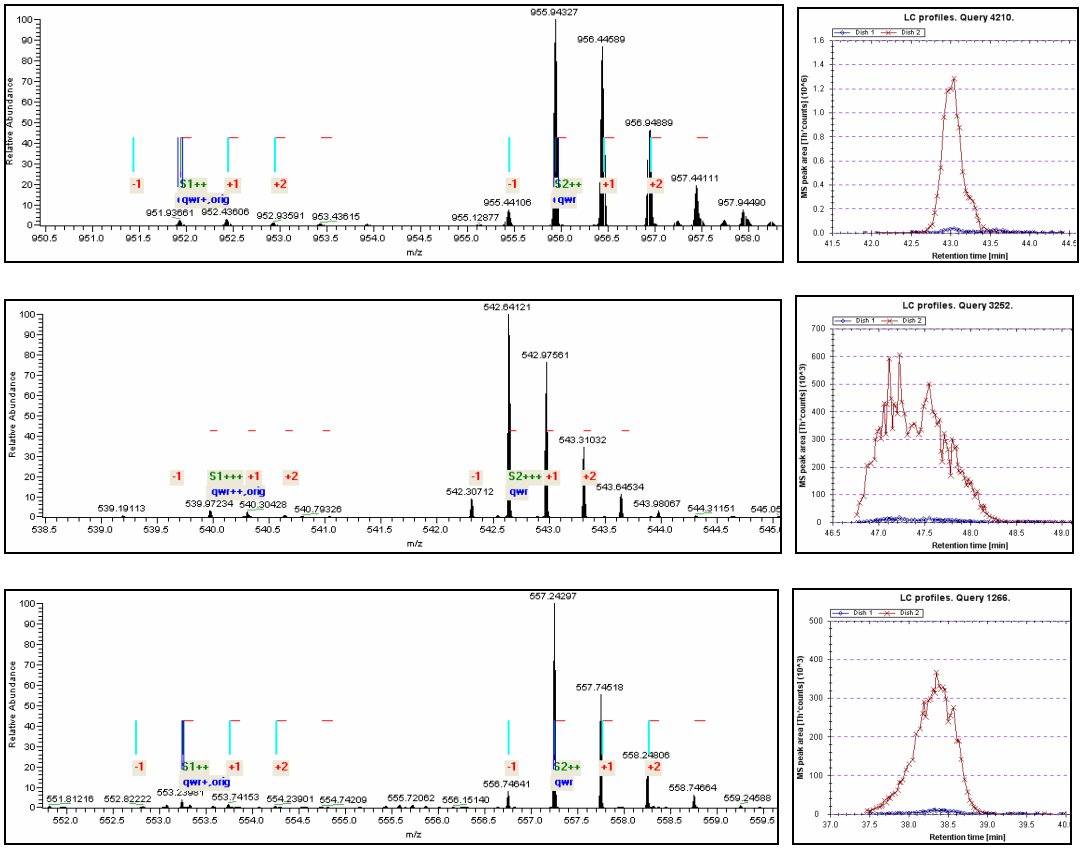

Figure S3. Illustration of the complete incorporation of "heavy" lysine into cellular proteins after eight-day cell culture. Quantification of a doubly charged peptide (*m/z* 951.94) LAEQAERYEDMAAFMK, a triply charged peptide (*m/z* 539.97) NLLSVAYKNVVGGQR, and a doubly charged peptide (*m/z* 553.24) YEDMAAFMK from stratifin (14-3-3σ) showed that the SILAC ratios (mean ± SD) were 35.82 ± 3.28, 38.02 ± 6.76, and 37.09 ± 5.34, respectively.

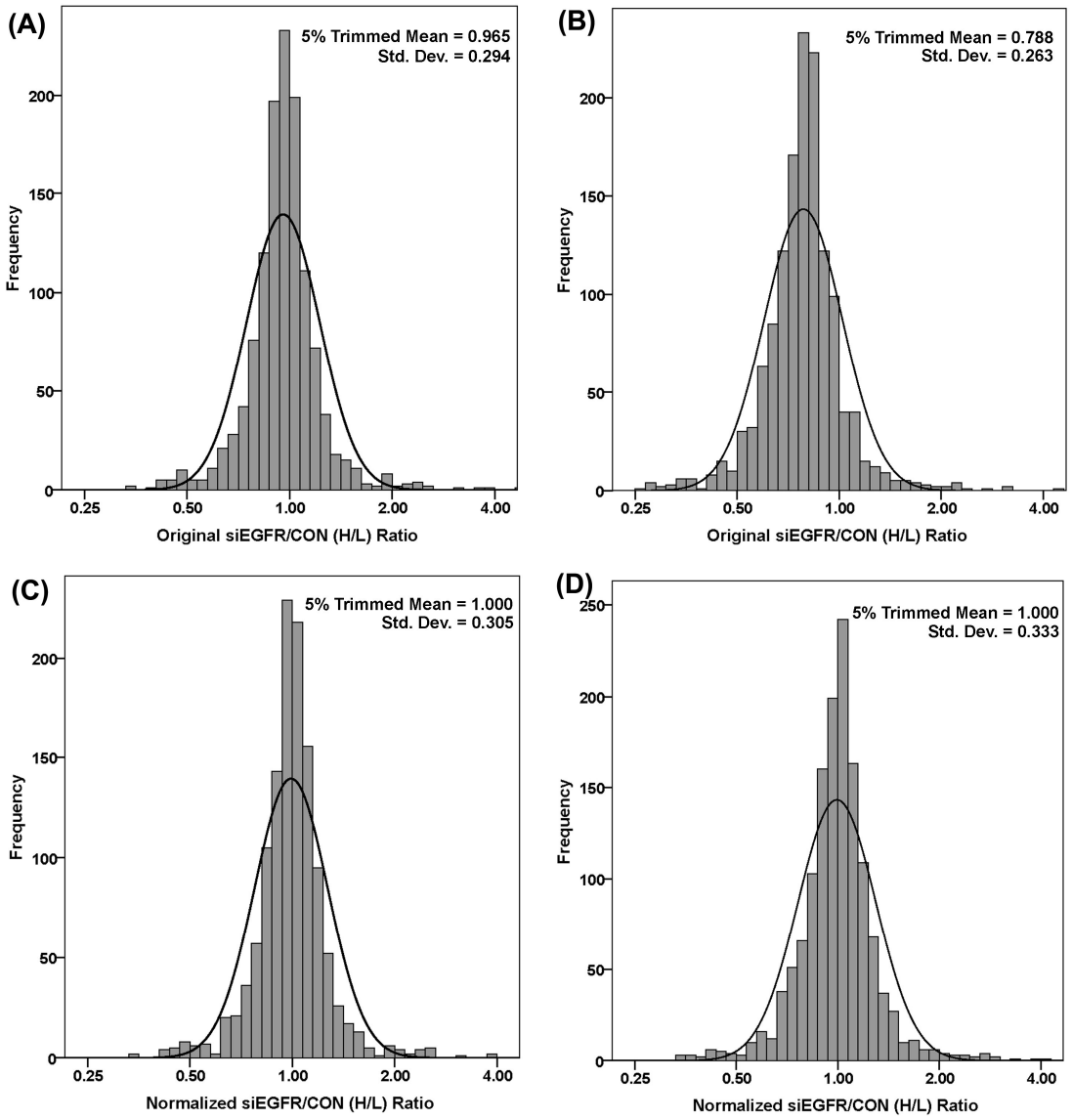

Figure S4. SILAC ratio distributions before and after multi-point normalization. The original siEGFR/CON (H/L) ratios for both biological replicates (A & B) were normalized against their respective 5% trimmed means so that the ratios were centered around the theoretical value 1.00 (C & D).

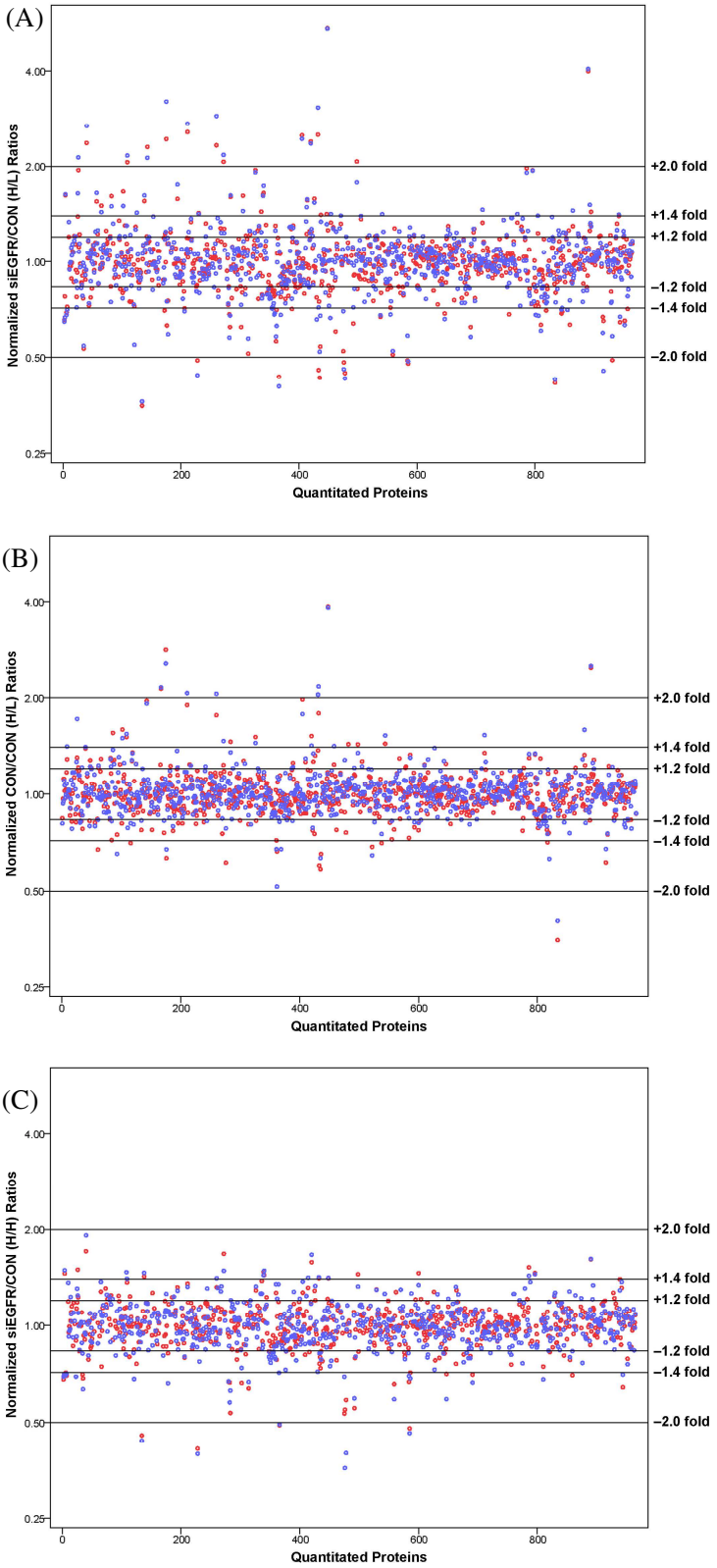

Figure S5. The normalized SILAC ratio distributions. Although some proteins seemed to be regulated 2.0-fold or more by EGFR knockdown (A), many of the changes were actually, at least in part, induced by differential SILAC labeling (B). After normalization, only 35 proteins were found to be significantly (>1.4-fold) regulated and most of the changes were modest (1.4~2.0-fold).

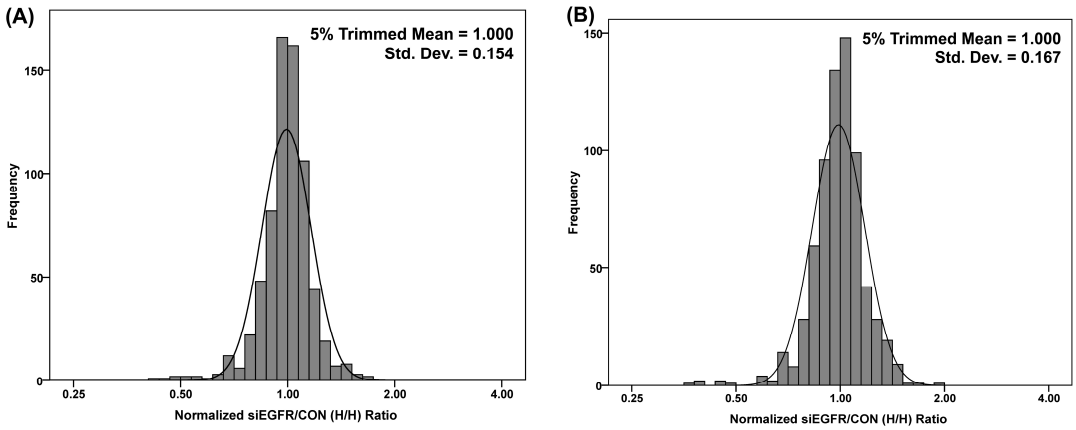

Figure S6. Distibution of siEGFR/CON (H/H) ratios. The normalized siEGFR/CON (H/L) ratios were further normalized against normalized CON/CON (H/L) ratios for both biological replicates to minimize the systematic errors introduced by differential SILAC labeling. The standard deviations for both datasets are about 0.16.

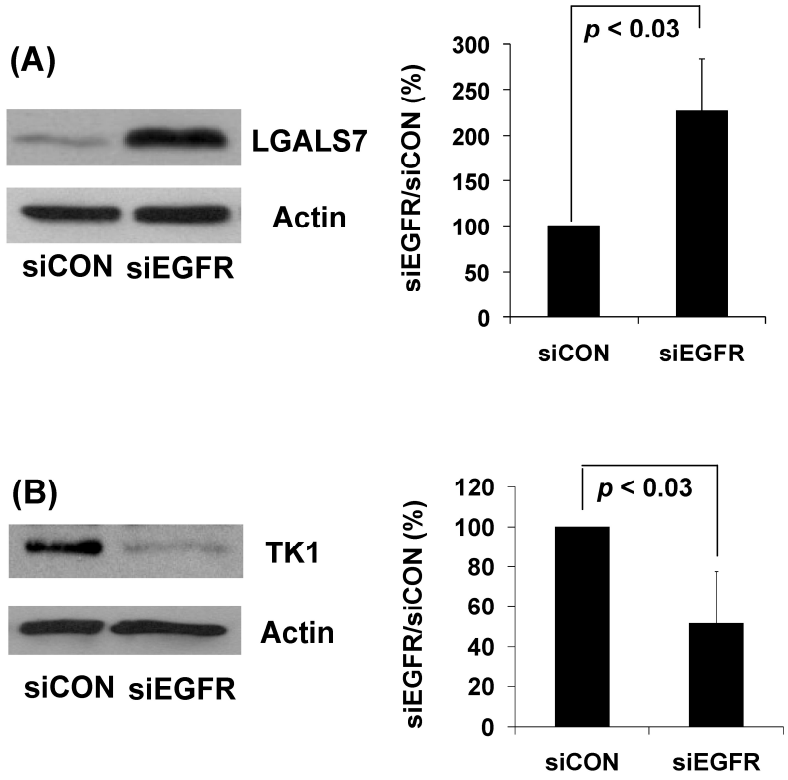

Figure S7. The significantly changed proteins were regulated by EGFR knockdown and not by transfection *per se*. The protein ratios determined by western blotting analysis were consistent with those determined by SILAC quantitation.