Table 1 Molecular weights and expression yields of purified DARPin C9 and the protamine fusion proteins after expression in *E. coli* and purification using Ni-NTA columns.

Protein*	MW (kDa)	Yield (mg/L culture)	Protein*	MW (kDa)	Yield (mg/L culture)
C9	18.2	48	C9-P	22.6	21
C9D	36.7	36	C9D-P	40.9	17
C9LZ	22.9	39	C9LZ-P	27.2	16

*C9 monomers and dimers without protamine were purified under native conditions. The C9-protamine fusion proteins were lysed under denaturing conditions and refolded on the column by applying a gradient of 8-0 M urea with subsequent elution in native buffer containing 250 mM imidazol. Yields are of a single recombinant expression (250 ml culture) under non-optimized standard conditions.

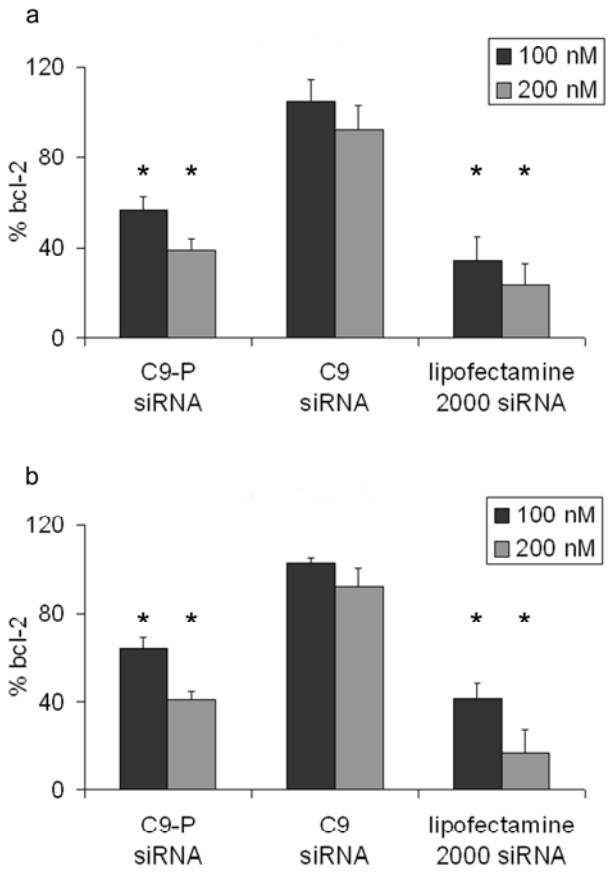

Figure 1. Down-regulation of (**a**) bcl-2 mRNA and (**b**) bcl-2 protein in HT-29 cells upon treatment with bcl-2-targeted siRNA in the presence of C9 or complexed to C9 protamine fusion protein in a ratio of 6:1 or with lipofectamine. Cells were treated for a total of 48 h, then lysed and subjected to mRNA or protein analysis by qPCR or Western blotting, respectively. Values were standardized to ribosomal RNA or actin. Following lysis, bcl-2 protein levels were determined by densitometric quantification of bcl-2 bands and normalization to actin. Error bars are mean + SD, n=3. *: $p<0.01$, compared to untreated control.

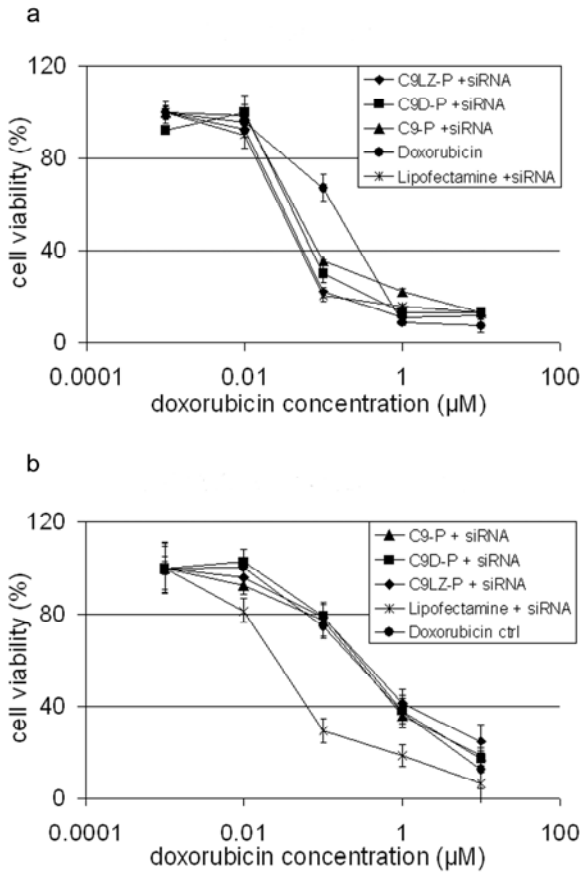

Figure 2. Chemosensitization of (**a**) EpCAM-positive MCF-7 and (**b**) negative HEK293T cells to doxorubicin upon treatment with bcl-2-targeted siRNA. The siRNA was added to cells as indicated in the presence of C9 or as complexes with the various C9 fusion proteins or lipofectamine. 48 h after transfection, cells were treated with the indicated doxorubicin concentrations and another 24 h later the effect on cell viability was determined in MTT assays. Error bars represent the mean ± SD, n=3.