



Fig. S1 - Transient siCHKA efficiently reduced the relevant transcript and protein expression. A. Relative quantification of *CHKA* transcript expression reported as percentage of 2^- Δ CT siCHKA/siNT ratio (*left panel*), asterisks are referred to statistically significant differences; a representative experiment is shown (*right panel*). **B.** Representative Western blotting analysis for ChoKα protein expression in OC cell lines following 72 h transient *CHKA* silencing. β-actin was used as loading control.



Fig. S2 - OC cells' sensitivity to sTRAIL treatment is specifically dependent upon CHKA downmodulation.

A. Representative Western blotting analysis for ChoK α protein expression in OC cell lines following *CHKA* transient silencing and recovery of ChoK α expression. Vinculin was used as loading control. **B.** Relative quantification of *CHKB* transcript expression reported as percentage of 2^- Δ CT siCHKB/siNT ratio, asterisks are referred to statistically significant differences.



Fig. S3 - CHKA silencing affected mitochondrial membrane potential but did not modify expression of common pro/anti apoptosis molecules.

A. Representative flow cytometry analysis of mitochondrial membrane potential (ψ_m) in siCHKA and siNT cell lines as detected by JC-1 labeling. A decrease in the red/green fluorescence intensity ratio indicates mitochondrial membrane depolarization (for each quadrant, the relative cell percentage on total count events is reported). B. Representative Western blotting analysis for Bid, XIAP and Bcl-2 protein expression in OC cell lines following transient CHKA silencing. Vinculin was used as loading control.

SKOV3

siCHKA

A2780

siNT

siCHKA

ChoKα

Vinculin

Bid

XIAP

Bcl-2



Fig. S4 - TRAIL-R2 is the only TRAIL-Rs family member specifically affected by siCHKA.

A. siCHKB does not affect TRAIL-R2 membrane expression as detected by flow cytometry analysis. Fluorescence index are reported **B.** *TNSFRSF10A*, *TNFRSF10C*, *TNSFRSF10D* did not significantly increase in siCHKA cells compared to control. Relative quantification of transcripts expression are reported as percentage of $2^{-}\Delta$ CT siCHKA/siNT ratio. *TNFRSF10B* expression (red bar) already reported in Fig. 4 is shown for comparison. **C, D.** Representative fluorescence indexes detected by flow cytometry summarizing membrane surface expression of TRAIL-Rs in siCHKA and control OC cell lines detected following primary antibodies incubation at 4°C or 37°C, respectively.



Fig. S5 - Assessment of siCHKA efficacy on ex vivo OC cells collected from patients' ascites.

A. Live $20 \times$ microscope image of typical appearance of *ex vivo* OC cells in patient's ascitic fluid. B. Relative quantification of *CHKA* transcript expression in siCHKA OC cells derived from ascites (patient TEM1510), asterisks are referred to statistically significant differences. C. Representative Western blotting analysis for ChoKa protein expression in *ex vivo* OC cells following transient *CHKA* silencing. β -actin was used as loading control.