ZNF429: a new tumor suppressor?

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INTRODUCTION

A recent study has shown *ZNF429,* a poorly described zinc-finger protein with associated Krüppel-box domain, to be a candidate for favorable prognostic marker in ovarian cancer. Zinc-finger proteins form the largest family of sequence-specific DNA-binding proteins encoded by the human genome and are commonly associated with other functional domains that can control the protein function. Many zinc-fingers have already been detected in cancer with different impact at outcome, which poses the question of tissue-specificity for their functionality. Given its predicted function as a transcription factor and its prognostic value, we hypothesized that *ZNF429* may regulate important cancer pathways.

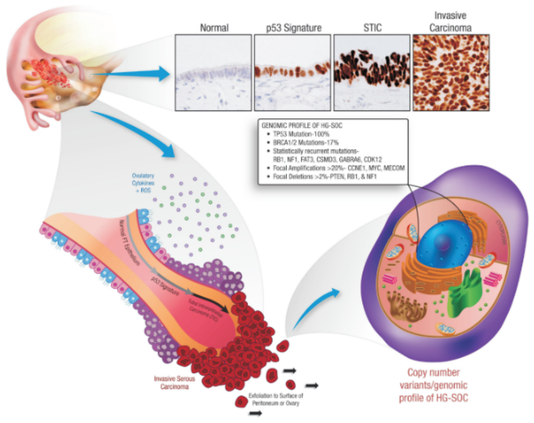


Figure 1: Ovarian cancer originates of mutated cells from fallopian tube epithelium. Ref: Jones and Drapkin, *Front. Oncol*, 2013

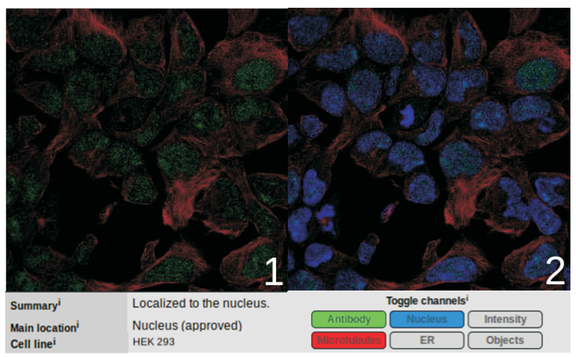
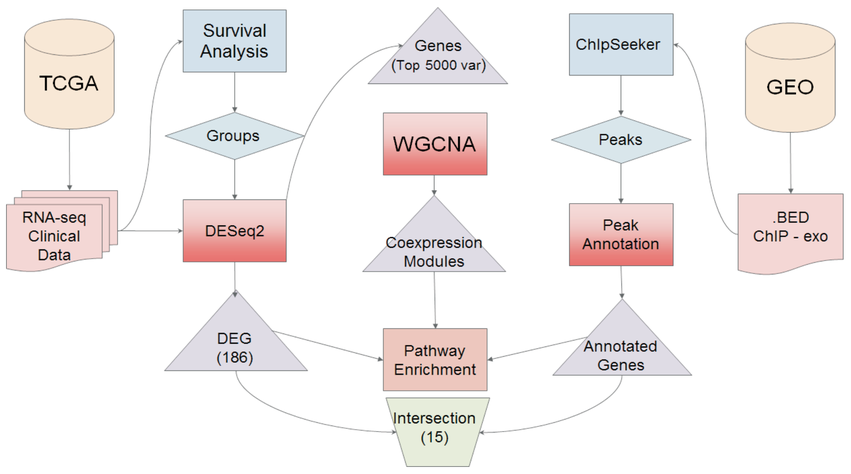


Figure 2: Immunofluorescence showing *ZNF429* localization in the cell. Ref : https://www.proteinatlas.org/ENSG00000197013-ZNF429/tissue

MATERIAL AND METHODS

In this study we analyzed 182 samples from the TCGA-OV project RNA-seq data. We evaluated the impact of *ZNF429* expression levels on patient survival and inspected differentially expressed genes (DEG) related with *ZNF429* expression. We also analyzed *ZNF429* ChIP-exo data and compared with DEG.



RESULTS

High expression of *ZNF429* shows better survival for ovarian cancer patient

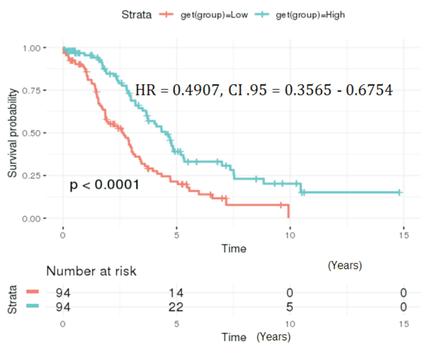
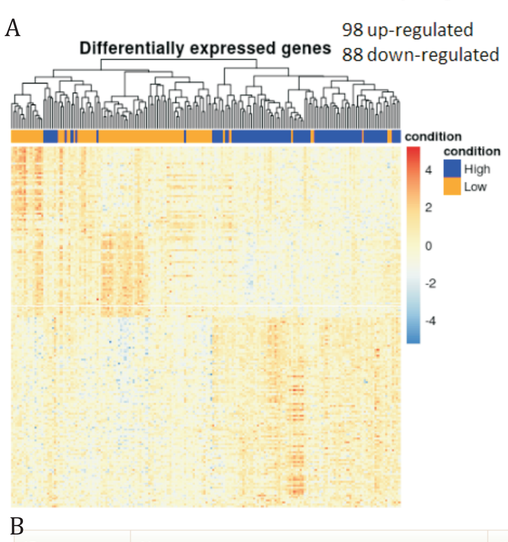
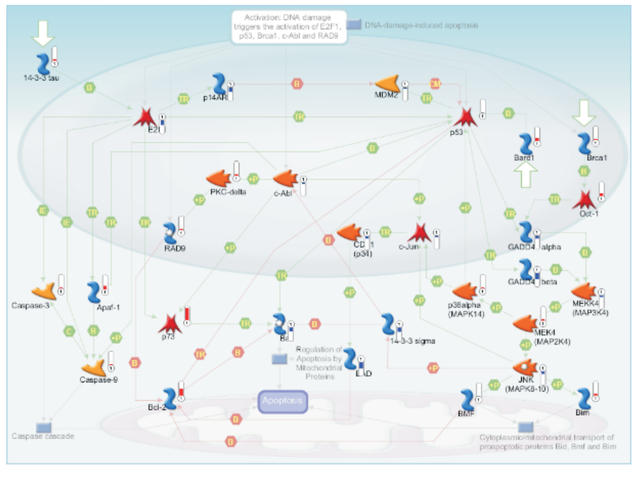


Figure 3: Impact of ZNF429 on patient's survival. Kaplan Meyer survival showing overall survival differences of groups of patients with high(blue) and low(red) expression of *ZNF429*. The table below shows the number of living patients at every 5 years. The hazard ratio (HR) for death in the High group is shown with 95% confidence interval (CI).

ZNF429 may regulate tumor suppressor gene expression





B

ID	Name	#Gene	FDR
R-HSA-212436	Generic Transcription Pathway	20	1.66e-03
R-HSA-168249	Innate Immune System	4	1.02e-01
R-HSA-168256	Immune System	8	3.58e-01
R-HSA-556833	Metabolism of lipids and lipoproteins	4	4.48e-01
R-HSA-1643685	Disease	3	6.05e-01
R-HSA-2262752	Cellular responses to stress	9	1.83e-03
R-HSA-2559583	Cellular Senescence	8	2.7e-03

Figure 4: Genes and pathways potentially regulated by ZNF429 A) Normalized gene expression level (z-score) of differentially expressed genes (DEG) between High and Low ZNF429 samples were used to generate heatmaps (p-adj < 0,001 and log Fold-Change (FC) > 0,7). Patients were hierarchical clustered based on pearson correlations of DEGs;

B) Gene-set enrichment analysis of DE genes was performed using the Reactome database . Red pathways are positively related with high expression of ZNF429 and blue pathways are positively related to low expression. C) p53 dependent apoptosis pathway - thermometers are present for genes that are up (red) and down (blue) regulated on the group of High expression of *ZNf429*.

ZNF429 is amplified in ovarian tumor samples of High Group

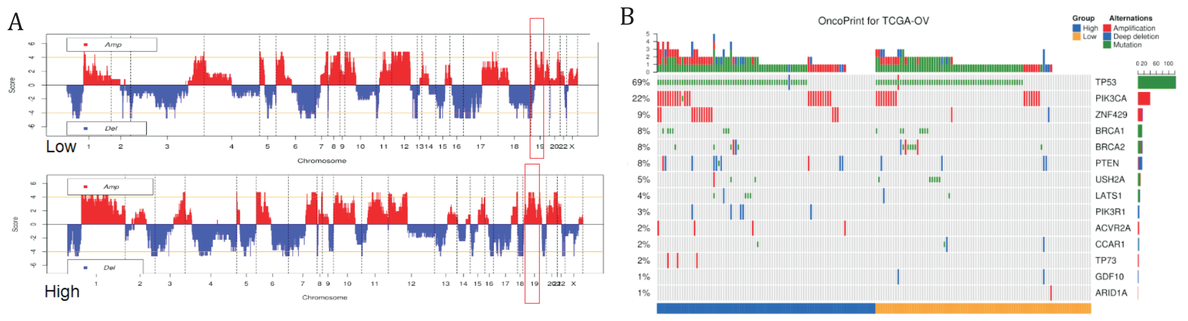


Figure 5: Genomic alterations frequencies in the two groups. A) Copy number variation across chromosomes in the groups with Low and High *ZNF429* expression. The red boxes show the amplified region in chromossome 19 in the High Group. B) The OncoPrint provided an overview of genomic alterations (legend) in particular genes (rows) affecting individual samples (columns). The truncated mutations, deep deletions and amplifications, were shown as green, blue and red color .

ZNF429 is co-expressed with important genes for cancer development

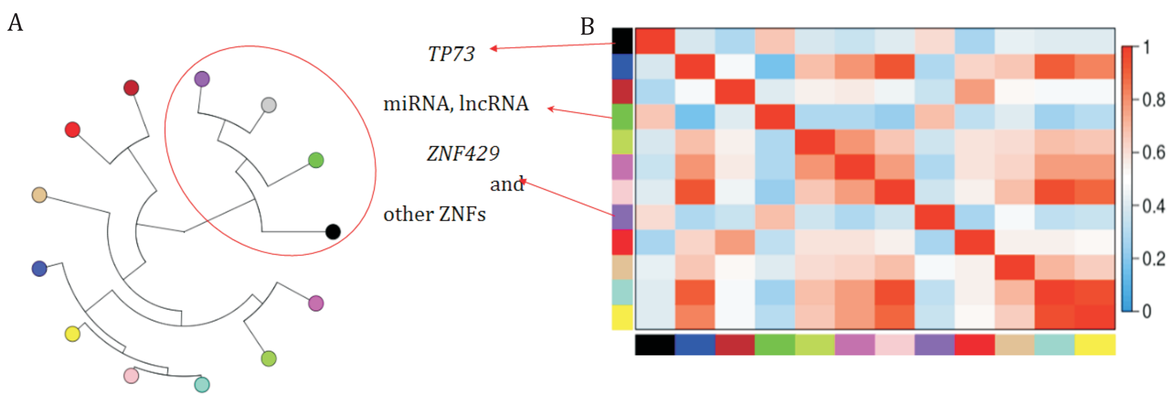


Figure 6 Co-expressed genes modules A) Clustering of modules generated with the weighted correlation network analysis. Red circle highlights the purple module, which is enriched with *ZNF429* and other *ZNFs,* and the highly correlated modules gray (*GDF-10*), lime (miRNAs and lncRNAs) and black (*TP73*); B) Heatmap of module correlation. Genes found in the correlated modules to the *ZNF429* are highlighted.

ZNF429 binds to different regions in the genome

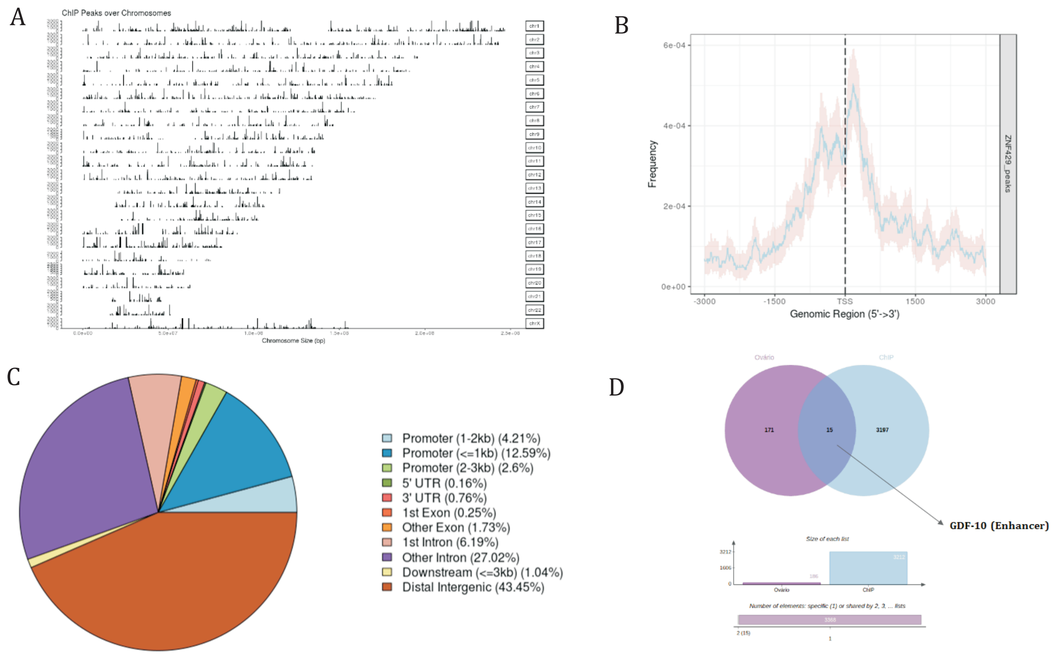


Figure 7: *ZNF429* acts as a transcription factor regulating several genes across the genome. A) ChiP peaks over chromosomes. B) Frequency of peaks in the TSS region; C) Localization of ChIP-exo peaks and percentage of peaks per potential gene expression control region. D) Overlap between DEG and Genes regulated by ZNF429 according annotation of the identified binding sites.

CONCLUSION

- *ZNF429* is amplified in some ovarian cancers and its overexpression has a positive impact on patient survival.
- High levels of *ZNF429* expression is related with the overexpression of proapoptotic genes that seems to be co-expressed in ovarian cancer.
- *ZNF429* regulates the potential tumor-suppressor *GDF-10* expression by ligating to an enhancer region. whose super expression being correlated with cell cycle arrest and promoting DNA damage induced apoptosis through the ATM-p53 axis, increasing cell sensibility to genotoxic agents by peak on an enhancer for one of these genes,
- In physiological conditions, *ZNF429* could be related to aging, hormone biosynthesis and signalling pathways related to cancer progression.
- We hypothesize that *ZNF429* exerts control on important pathways for cancer development control, directly or indirectly, regulating the expression of important genes at these pathways.

REFERENCES

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Cheng CW *et al* (2016) Loss of GDF10/BMP3b as a prognostic marker collaborates with TGFBR3 to enhance chemotherapy resistance and epithelial-mesenchymal transition in oral squamous cell carcinoma. Mol Carcinog, 55(5):499-513.

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