Integrative Analysis of Long non-coding RNA/pseudogene-mediated ceRNA Crosstalk in Endometrial Cancer

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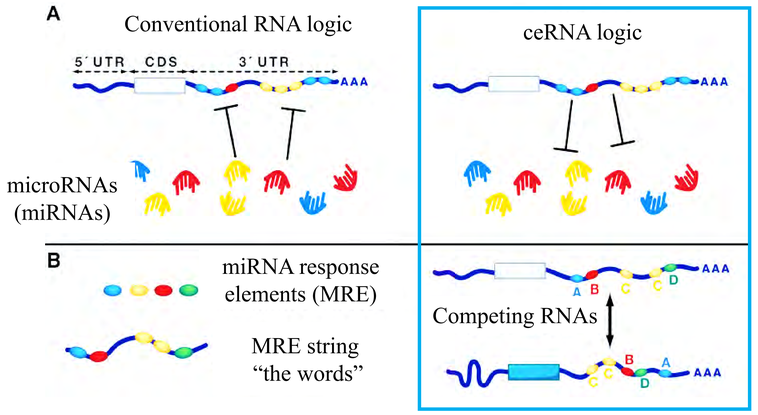
Introduction

Competing endogenous RNA (ceRNA) networks have significant roles in endometrial cancer (EC) pathogenesis by altering the expression of key tumorigenic or tumor-suppressive genes [1].



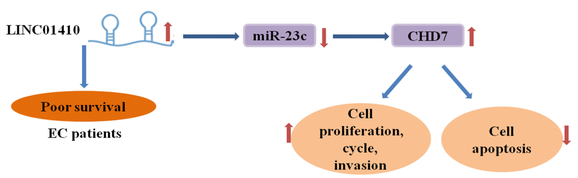


Figure 1. Example of experimentally validated ceRNA association in EC, **LINC01410/miR-23c/CHD7** [2].

Objectives

- Identify novel ceRNA networks in EC using context-specific analytical methods.
- Identify functional relevance of differentially expressed (DE) genes in found EC-associated ceRNA networks.
- Identify genes, long non-coding (lncRNAs), pseudogenes and microRNAs (miRNAs) that are significantly correlated with overall survival of EC patients.

Materials and Methods

1. Collecting and processing RNA expression data of the TCGA-UCEC study [3] (545 cases and 35 controls)
2. Performing differential expression analysis using limma R package [4]
3. Conducting ceRNA network analysis using GDCRNATools R package [5]
4. Performing Cox-regression univariate survival analysis using survival R package [6]

Results and Discussion

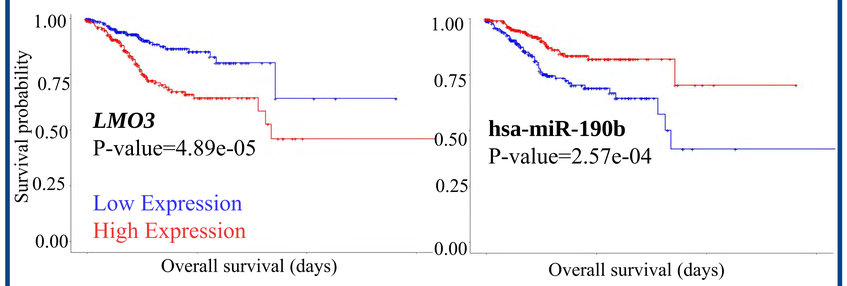
ceRNA network analysis

lncRNA (146)	mRNA (1266)	miRNA (186)	Pseudogene (60)	mRNA (654)	miRNA (209)
2160 ceRNA associations			784 ceRNA associations		

Genes		miRNAs (starts with hsa-miR-)		
RIPOR2	SYNE1	-490-3p	-217	-455-5p
ZBTB38	TNS1	-137	-129-5p	-142-3p
RUSC2	EPB41L3	-184	-375	-125a-5p
MORC3		-139-5p	-126-3p	

Survival analysis

Genes (top-13 among 34)	*LMO3, HIF3A, B4GALNT3, SCGB2A1, CRELD2, KCNK6, GRB7, SSC4D, PGR, TRPM4, CDKN2A, KCNJ12 & EPHB1*
lncRNAs (11)	LINC00908, AL133243.2, NRAV, AL391422.3, AC025154.2, LBX2-AS1, BOLA3-AS1, AC003102.1, LINC01224, AC006329.1, LINC02381
miRNAs (16) (starts with "hsa-miR-" prefix)	-1301-3p, -195-3p, -940, -106b-5p ,-106b-3p ,-3170,-497-5p,-3614-5p,-142-3p,-146a-5p,-3614-3p,-1269a ,-142-5p ,-4668-3p,-4728-3p, & -190b
Pseudogenes (3)	GOLGA2P5, PPP1R14BP3, & AC005077.4



The Cox regression survival analysis resulted 34, 11, 16, and 3 genes, lncRNAs, miRNAs, and pseudogenes, respectively. The survival plots show that patients with low-expressed *LMO3* and high-expressed hsa-miR-190b has better survival.

Functional enrichment analysis

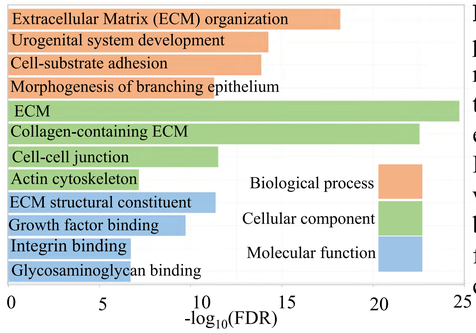


Figure 2. KEGG pathway analysis results have shown that differentially-expressed genes in EC are associated with ECM related biological process, functions and components.

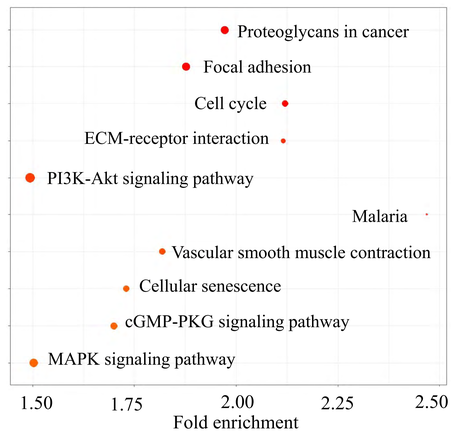


Figure 3. DE genes in EC ceRNA networks are associated with popular cancer-related pathways such as proteoglycans in cancer, PI3K-Akt signaling pathway, and MAPK signaling pathway in Gene ontology (GO) pathway.

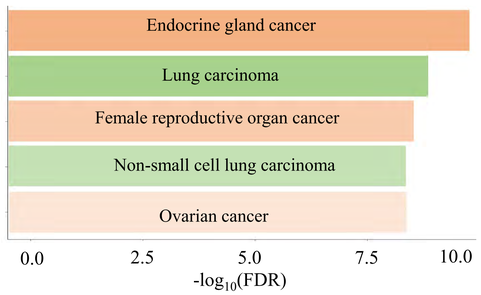


Figure 4. The top five components of disease ontology (DO) pathway results can be recognised as hormone-dependent cancer types.

Conclusion

CeRNA hypothesises bi-directional miRNA regulation mechanism in gene networks. These ceRNA networks can identify lncRNAs and pseudogenes that are competing for MRES. Functional enrichment analysis and survival analysis of differentially expressed genes in ceRNA network will recognise their functional importance and involvement in EC prognosis, respectively. The significant genes and miRNAs involved in ceRNA networks will contribute for future diagnosis and therapeutic experiments in EC.

References

1. Salmena et al., 2011. Cell. 146 (3). 353-358
2. Lu et al., 2020. Molecular and Cellular Biochemistry. 469. 9-19
3. Grossman et al., 2016. New England Journal of Medicine. 375 (12). 1109-1112
4. Ritchie et al., 2015. Nucleic Acids Research. 43 (7). e47
5. Li et al, 2020. Bioinformatics. 34 (14), 2515-1517
6. Therneau & Grambsch, 2020. Springer.

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