Identificación de lncRNAs y circRNAs en las neoplasias linfoblásticas de células T para mejorar el pronóstico y fortalecer el desarrollo de una oncología de precisión

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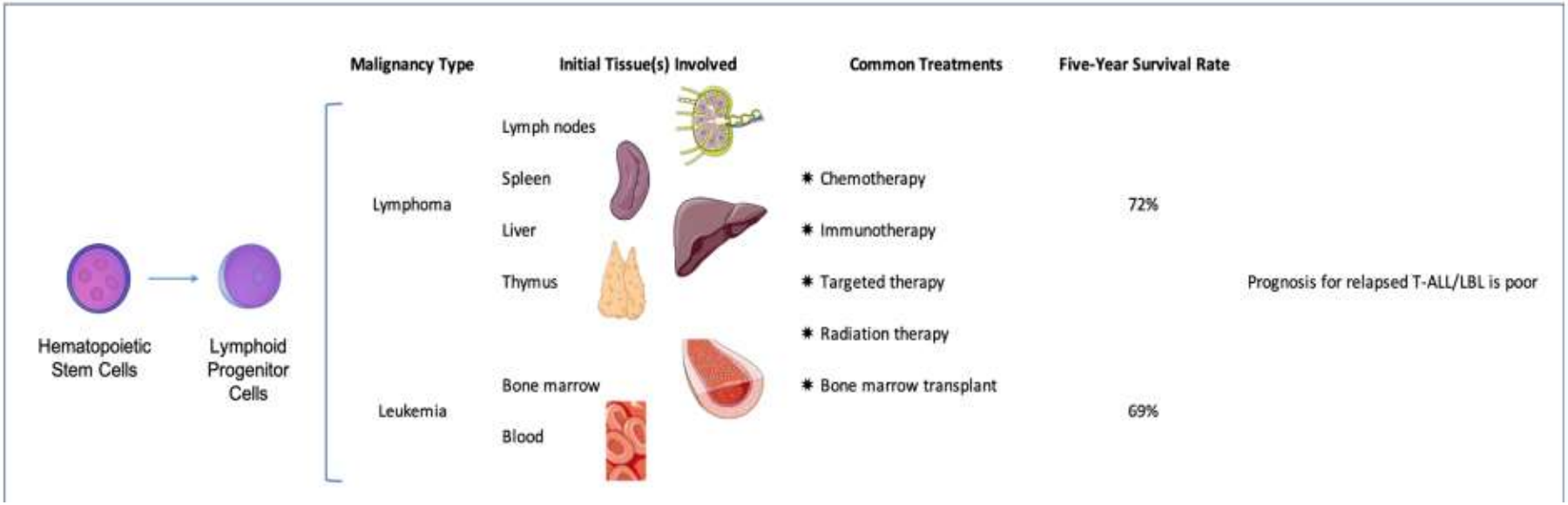
T-cell lymphoblastic neoplasia



T-cell lymphoblastic neoplasia is a type of aggressive haematological malignancy that arises from the transformation of immature precursor T cells mostly affecting children and adolescents[1]. It represents the most common type of malignancy arising from thymocytes and is mainly characterized by massive infiltration of immature T cells in the mediastinum and other lymphoid organs (T-LBL) or with significant involvement of peripheral blood (PB), bone marrow (BM), and cerebral spinal fluid compartments (T-ALL) [2].

LncRNAs are key regulators of gene expression

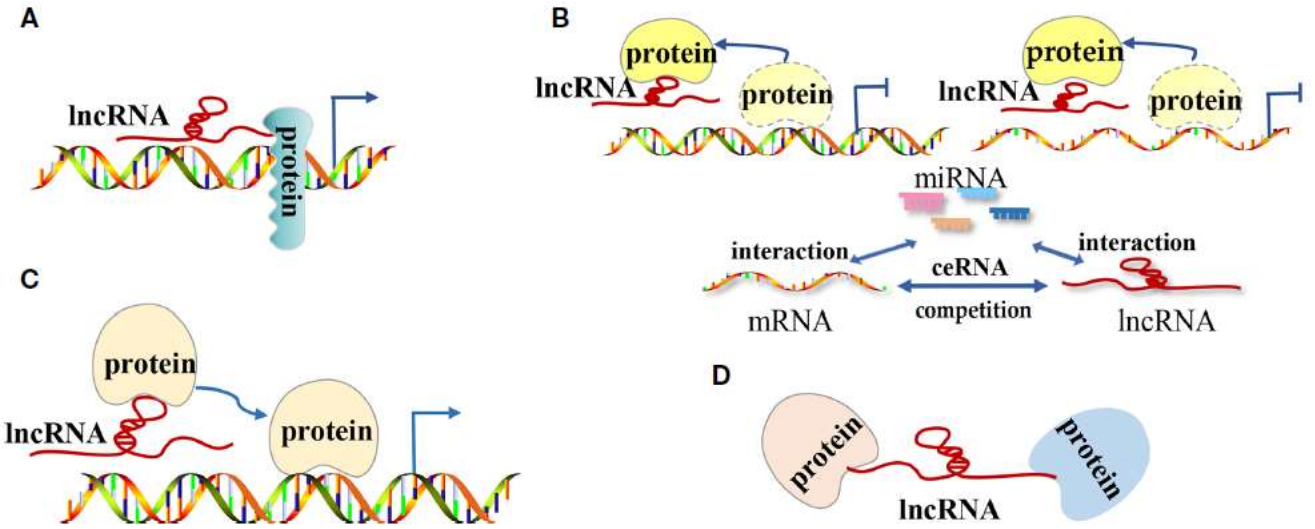


FIGURE 1 | The modes of action of long non-coding RNAs (lncRNAs) in tumors. **(A)** LncRNAs as signal molecules can be used alone or combined with some proteins(such as transcription factors) to mediate the transcription of downstream genes; **(B)** LncRNAs as decoy molecules bind to some functional protein molecules to block the protein molecules from regulating DNA and mRNA molecules or bind to miRNA molecules competitively with mRNA molecules to block the inhibitory effect of miRNA on mRNA molecules; **(C)** LncRNAs as guide molecules carries some functional protein molecules and locates them in the target area to perform functions; **(D)** LncRNAs as a scaffold molecule guide related different types of macromolecular complexes to assemble in the target area to work together.

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LncRNAs are key regulators of gene expression

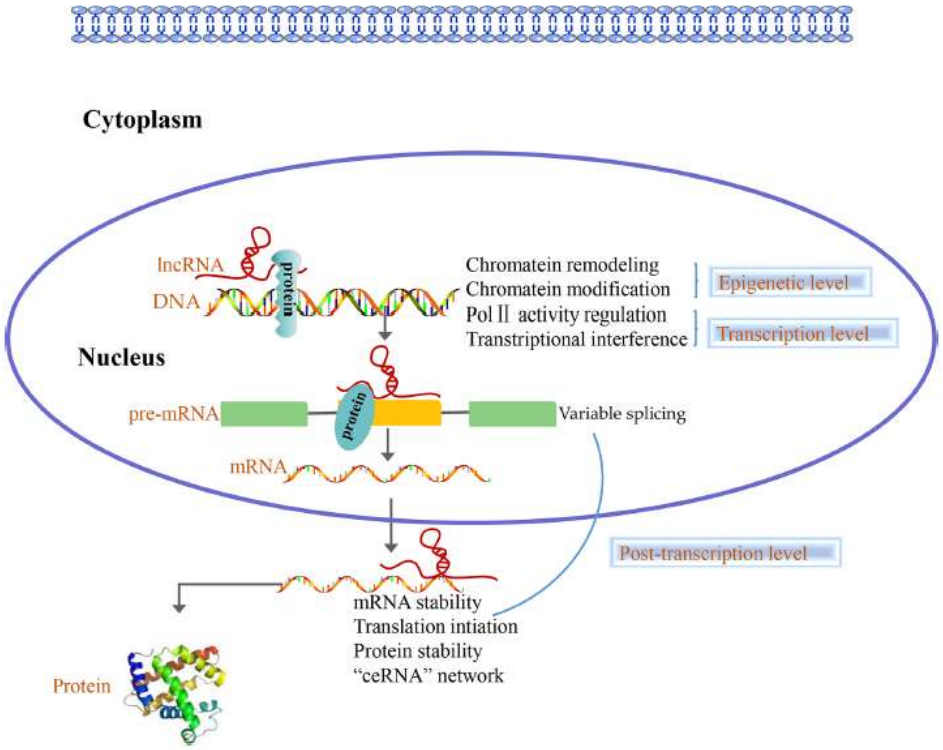

FIGURE 2 | Long non-coding RNAs (LncRNAs) function at three levels. In the nucleus, lncRNAs control the epigenetic state of genes, participate in transcriptional regulation and are involved in the process of pre-mRNA alternative splicing. In the cytoplasm, lncRNAs are involved in post-transcriptional regulation such as the mRNAs stability, mRNAs translation, protein stability and "ceRNA" network.

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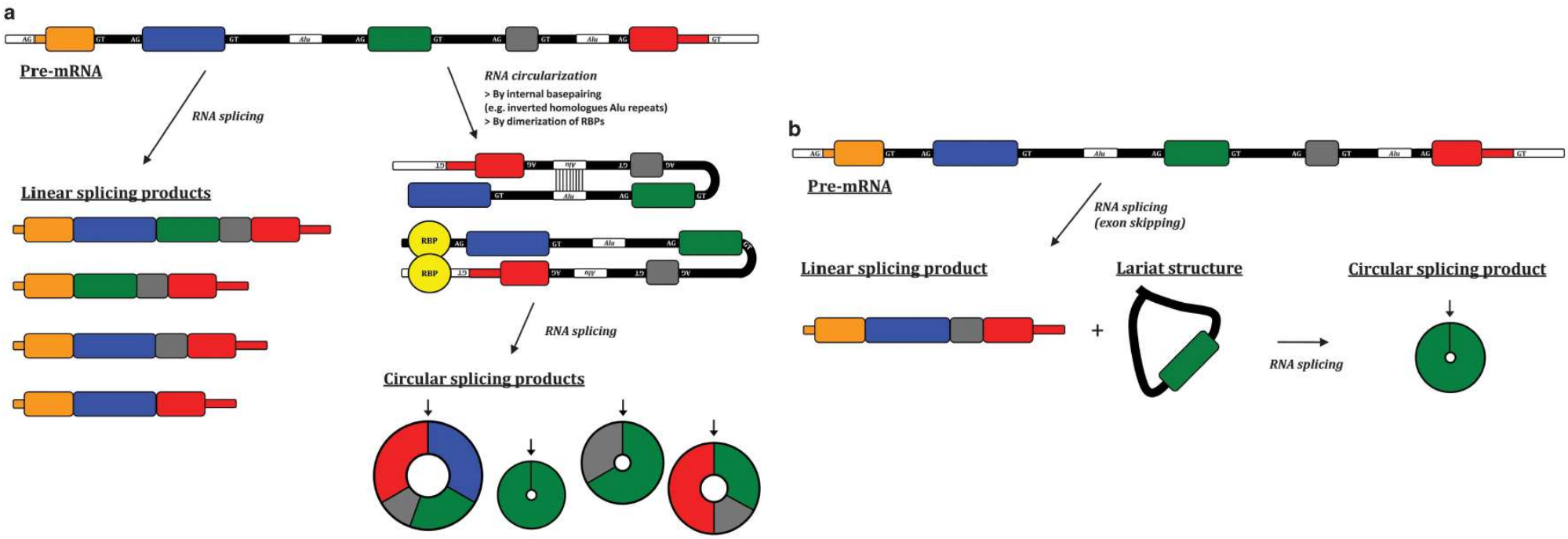
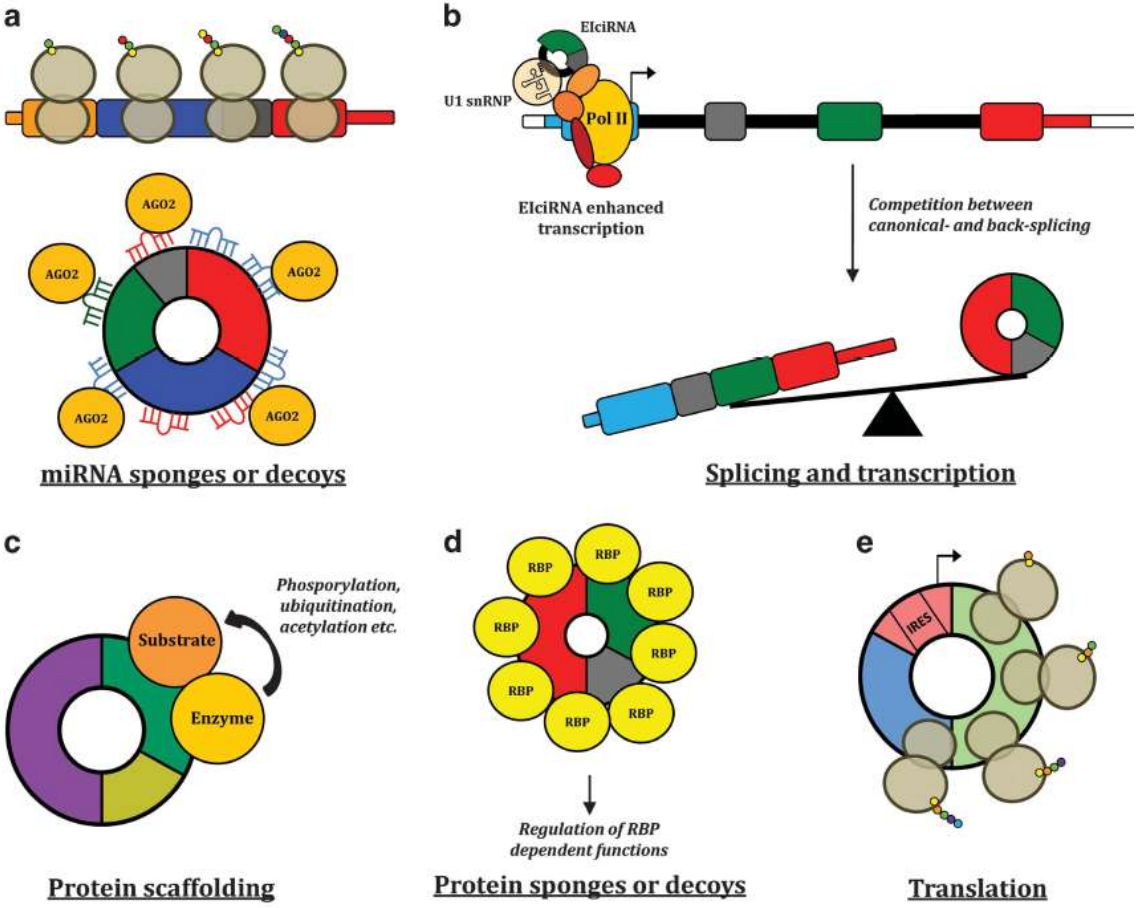
circRNAs are usually generated from genes that also produce linear isoforms



Kristensen et al. 2018

circRNAs novel regulators of gene expression



Kristensen et al. 2018

Patterns of differential expressed circRNAs in human thymocytes

non-coding RNA

MDPI

Article

Patterns of Differentially Expressed circRNAs in Human Thymocytes

Pilar López-Nieva [1,2,*,†], Pablo Fernández-Navarro [3,4,*,†], María Ángeles Cobos-Fernández [1,2], Iria González-Vasconcellos [1,2], Raúl Sánchez Pérez [5], Ángel Aroca [5], José Fernández-Piqueras [1,2] and Javier Santos [1,2]

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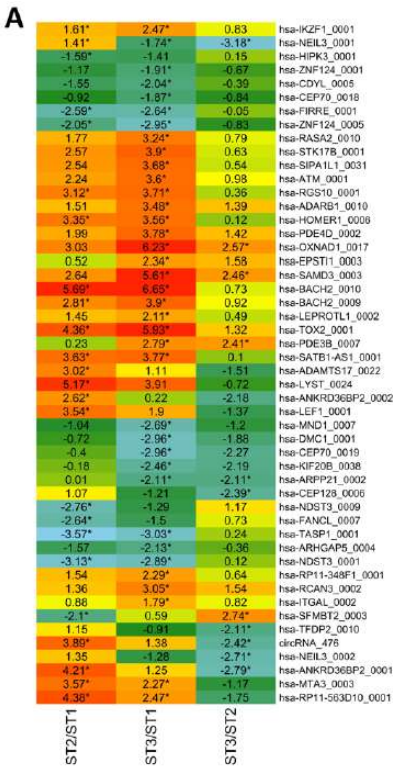
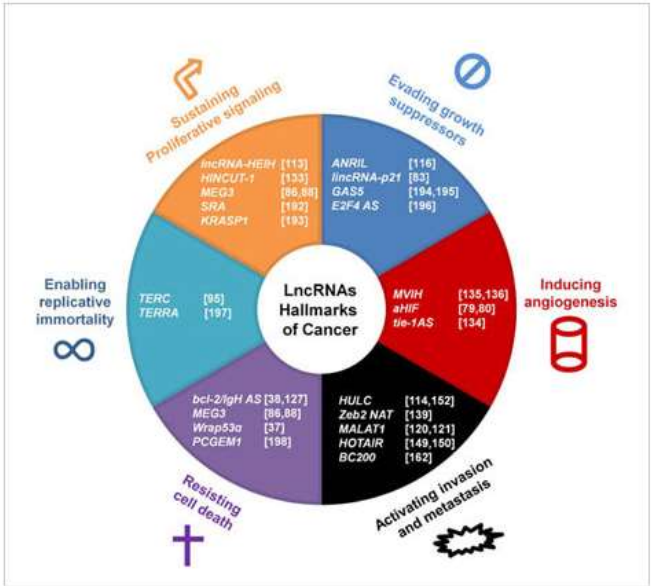
† These authors contributed equally to this work.

The differential expresión pattern of 50 specicic circRNAs serves to discriminate between the three human thymocyte populations (ST1: DN; ST2 DP; ST3: SP for CD4 and CD8)



LncRNAs and circRNAs are frequently dysregulated in cancer



Di Gesualdo F, Capaccioli S, Lulli M. (2014) **A pathophysiological view of the long non-coding RNA world**. *Oncotarget*

Figure 3. Circular RNAs involved in the hallmarks of cancer. Tumor-suppressor circRNAs are indicated in green and circRNAs with oncogenic properties are indicated in black.

Kristensen et al. 2018 *Oncogene*

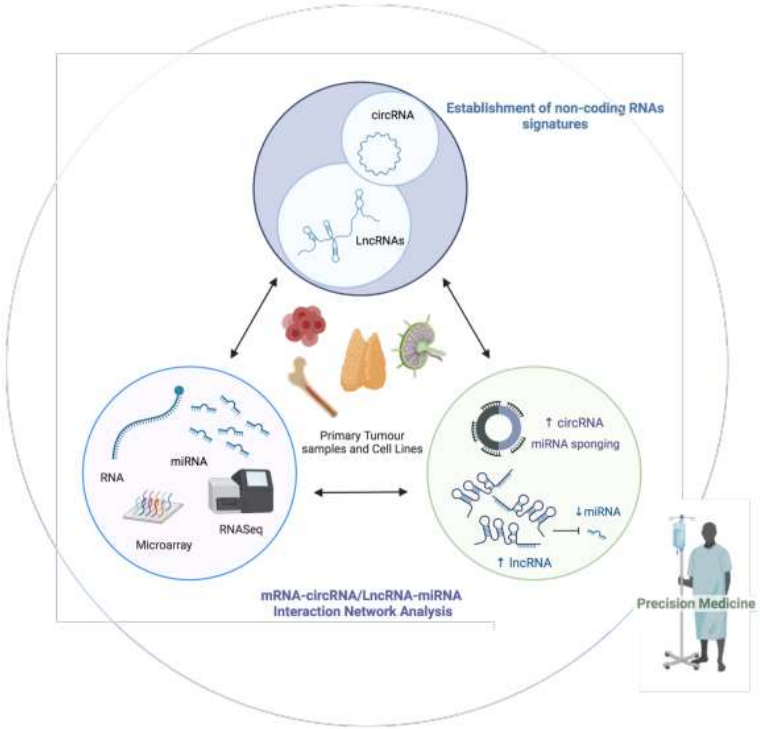
Overexpression of lncRNA **H19/poor prognosis** in gastric cancer
Downregulation of lncRNA **MALAT1/ poor prognosis** in breast cancer
Downregulation of lncRNA **CHRM3-AS2/poor prognosis** in ovarian cancer
Downregulation of lncRNA **XIST/poor prognosis** in breast cancer

Overexpression of **circ_0005909/poor prognosis** in lung cancer
Downregulation of **circ_0005986/poor prognosuis** in liver cancer
Downregulation of **cir_CNTNAP3/poor prognosis** in esophageal cancer

New patient-based molecular data focused on differentially expressed lncRNA and circRNAs

WHAT THE PURPOSE OF THIS RESEARCH IS

Extracting the most precise ncRNA signatures from primary tumours (or relapsed samples) and integrating those specific signatures in new interactions between the coding mRNAs and non-coding genome can help in subdividing T-cell neoplasia into specific set of patients with unique ncRNA signatures matched to their clinical outcomes leading to enhance personalized medicine in T-cell neoplasia in the near future



WHAT THIS STUDY ADDS

- To identify specific non-coding signatures for T-cell lymphoblastic neoplasia
- To discover networks by integrating data from mRNA-miRNA-lncRNA-circRNA with a role in driving tumorigenesis of T-cell lymphoblastic malignancies
- To propose differentially expressed non-coding RNAs as new biomarkers to improve prognosis and current treatments in the context of a personalized medicine
- To contribute to develop better individualized therapies gathering information for a future prognostic specific panel in T-cell lymphoblastic neoplasia

New patient-based molecular data focused on differentially expressed lncRNA and circRNAs

OBJECTIVES

1. **Initial analysis on a discovery cohort to identify differentially expressed lncRNAs and circRNAs**
2. **Validate those candidates in an extended cohort**
3. ***In silico* networks predictions. Those validated lncRNAs and circRNAs will be integrated into networks involving mRNAs and microRNAs**
4. **Experimentally validated those predicted networks *in vitro* by gain and loss function studies.**

METHODLOGY AND WORKPLAN

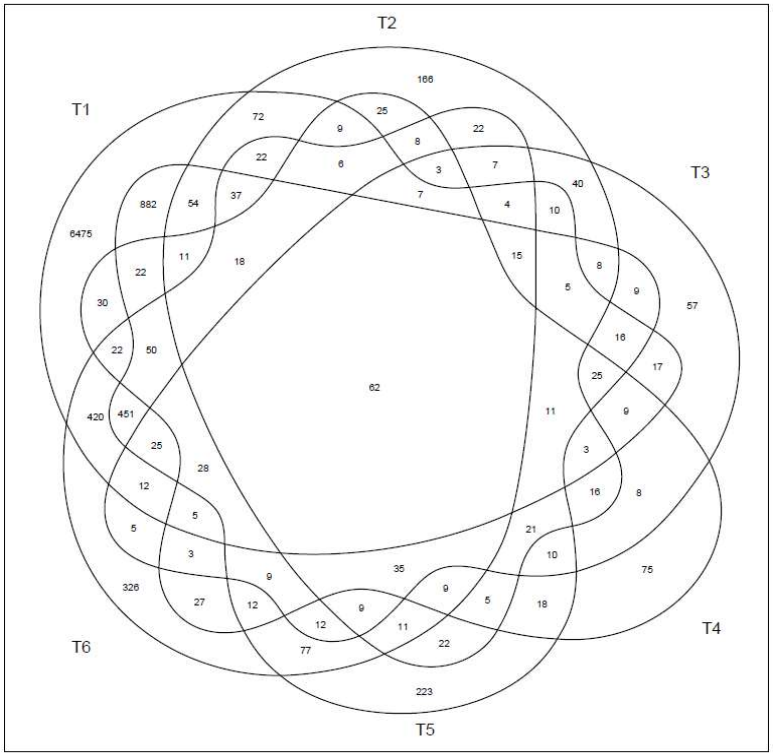
Phase I: Discovery of lncRNAs and circRNAs in T-cell lymphoblastic neoplasia

In phase I, a small cohort will be used to identify a group of candidate biomarkers via profiling assays on a whole transcriptome sequencing (RNA-seq) and lncRNA/circRNA microarrays

Phase II: Validated ncRNAs functional studies in tumour cell lines

In phase II, to understand the potential roles of the newly noted lncRNAs and circRNAs in the development of T-cell neoplasia we will perform via gain and loss of function studies

LncRNAs are involved in the development of T-cell lymphoblastic neoplasia

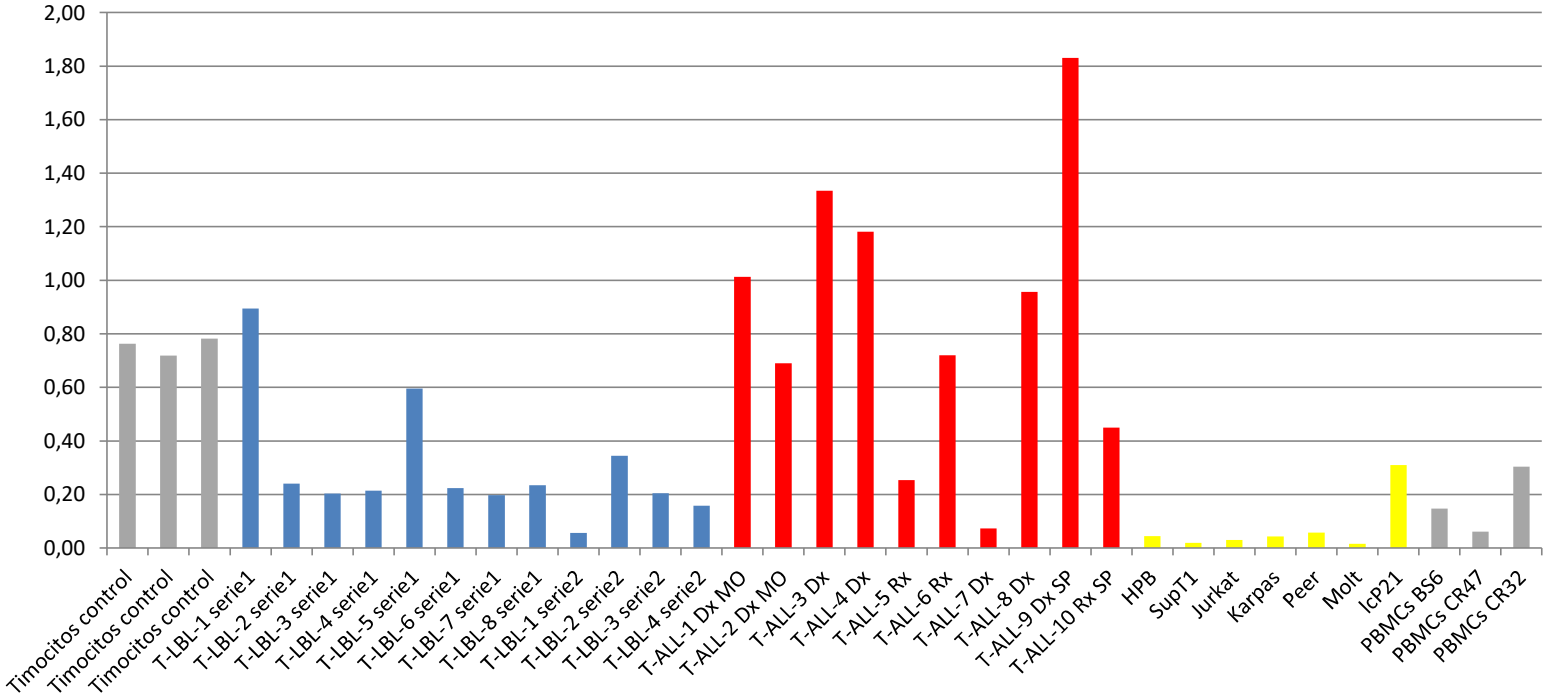


Venn diagram of lncRNA aberrantly expressed in six T-LBLs

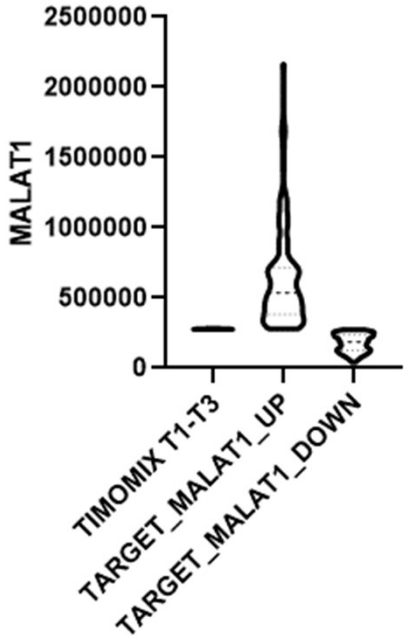
Gene ID	Expr.	Gene ID	Expr.	Gene ID	Expr.	Gene ID	Expr.
AC044849.1	Down	LINC01587	Up	lnc-ESRP2-2	Down	lnc-TBC1D19-2	Down
AL049990	Down	LINC01878	Up	lnc-FAAP20-3	Down	lncTCF7L2-7	Down
AL391422.4	Up	lnc-CXCR4-1	Down	lnc-FAM19A5-5	Down	lnc-THOC2-8	Up
AX746877	Down	lnc-IL6ST-1	Down	lnc-GOLGA4-4	Down	lnc-VPS50-1	Down
BX890604.2	Down	linc-KIF17-1	Down	lnc-H3F3A-7	Down	lnc-ZNF33B-2	Up
CHRM3-AS2	Down	lnc-LRRN3-2	Down	lnc-IL17A-2	Up	MALAT1-215	Down
FARP1-AS1	Down	lnc-PZP-10	Down	lnc-IRX3-80	Down	MALAT1-209	Down
G002089	Down	lnc-RARRES1-2	Up	lnc-IYD-1	Down	PRR34-AS1	Down
G008835	Down	lnc-SOHLH2-4	Up	lnc-LEF1-3	Down	RNF144A-AS1	Down
G017370	Down	lnc-ABHD4-11	Down	lnc-M1AP-6	Down	RP11-277L2.4	Down
G040741	Down	lnc-AMPH-10	Down	lnc-MCUR1-4	Down	SATB1-AS1	Down
G078168	Down	lnc-BACH2-1	Down	lnc-MGAM2-22	Down	SNAP25-AS1	Down
G089363	Down	lnc-CMPK2-16	Down	lnc-MTRNRL3-1	Down	TTTY15	Up
H19	Up	lnc-CXCR4-3	Down	lnc-PPWD1-2	Down	XIST	Down
IRAIN	Down	Lnc-DAD1-2	Up	lnc-SNRPD3-2	Down		
LINC01578	Down	lnc-ECHDC3	Down	lnc-SRY-9	Up		

LncRNA signatures with altered expression in the T-LBL cohort

Heterogeneous expression of lncRNAs in T-cell lymphoblastic neoplasia: MALAT1 as an example

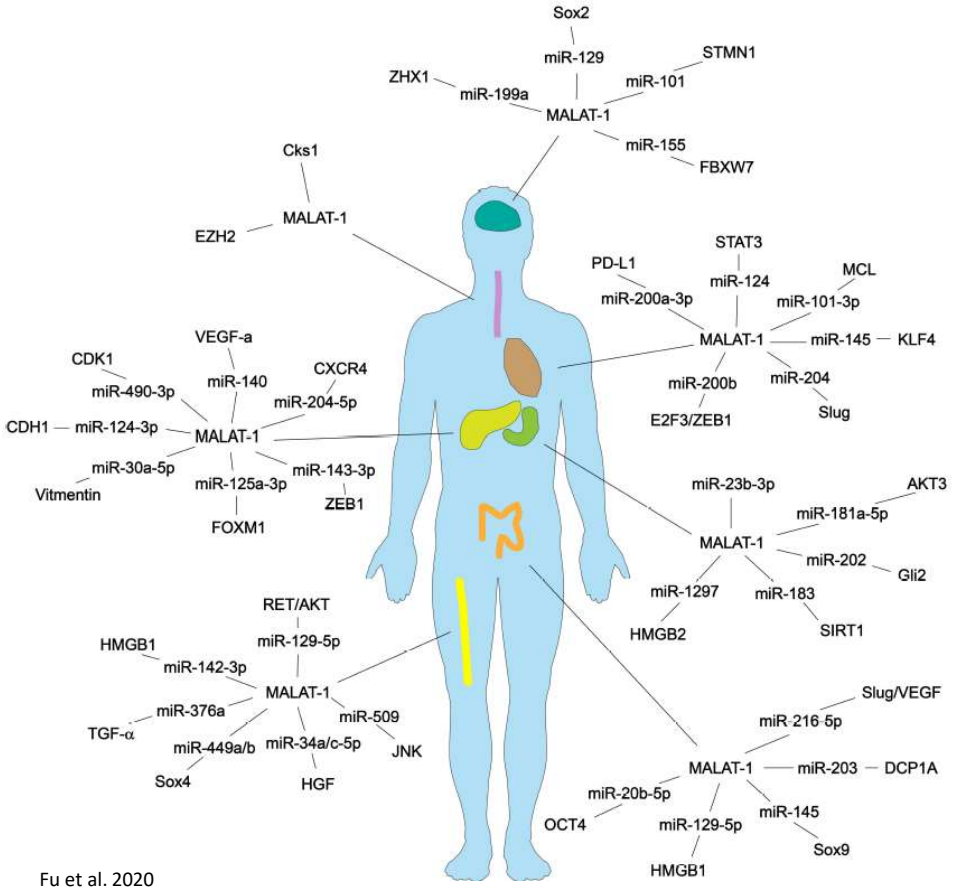


Expression of MALAT1 determined by RT-qPCR in primary T-LBLs and T-ALLs samples, as well as T-LBL/T-ALL-derived cell lines

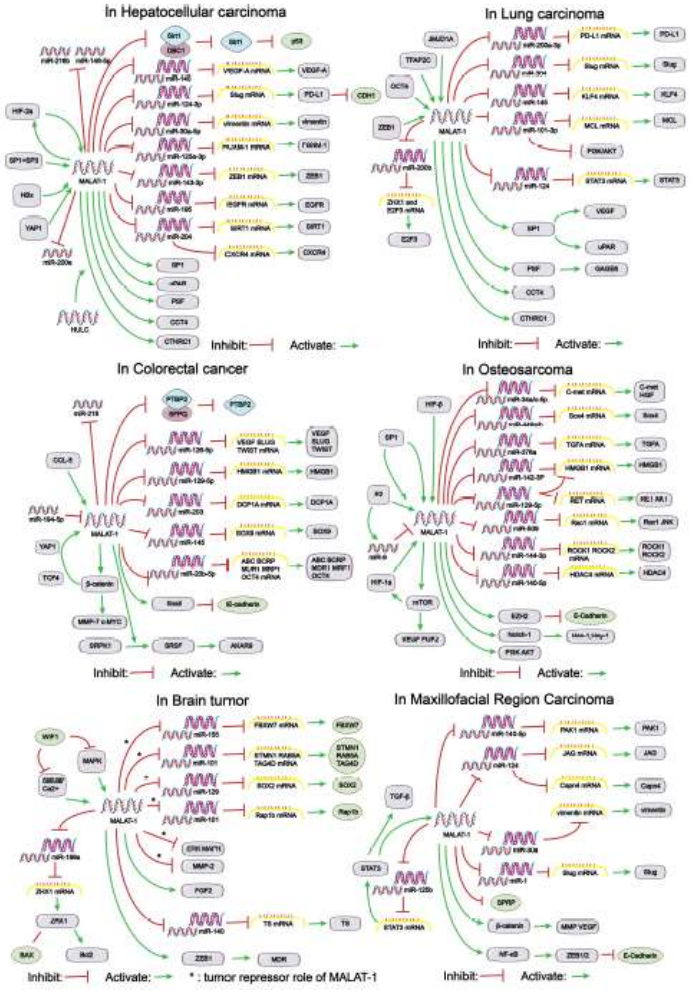


Analysis *in silico* of MALAT1 expression in a cohort of 264 pedriatic T-ALLs (UP, n=138, DOWN, n=126)

Regulatory networks of MALAT1 in T-cell lymphoblastic neoplasia: a pending question



Fu et al. 2020



Dual regulatory role of MALAT1 in the development of T-cell lymphoblastic neoplasia

C3_regulatory targets:
T-ALL_TARGET_MALAT1_DOWN

67 genesets (P-value<0,01: genetic signatures that are transcriptional targets of transcription factors

UPREGULATED

Gene Set Enrichment Analysis (GSEA)

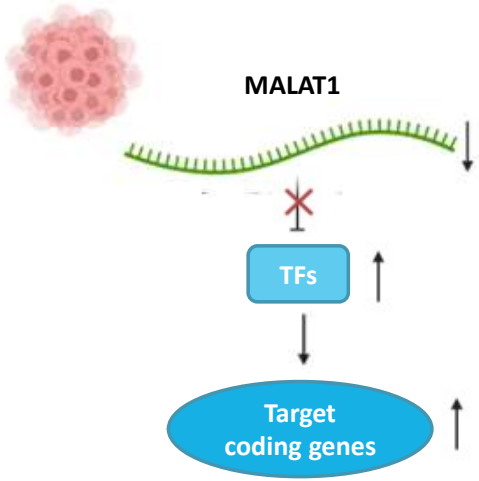
T-ALL TARGET cohort

MALAT1 DOWN
vs
MALAT1 UP

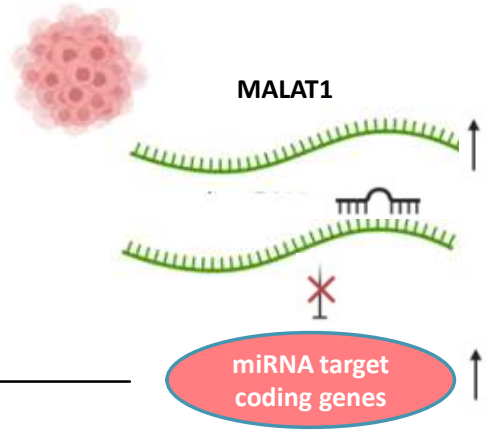
C3_regulatory targets:
T-ALL_TARGET_MALAT1_UP

871 genesets (P-value<0,01: genetic signatures that are targets of microRNAs

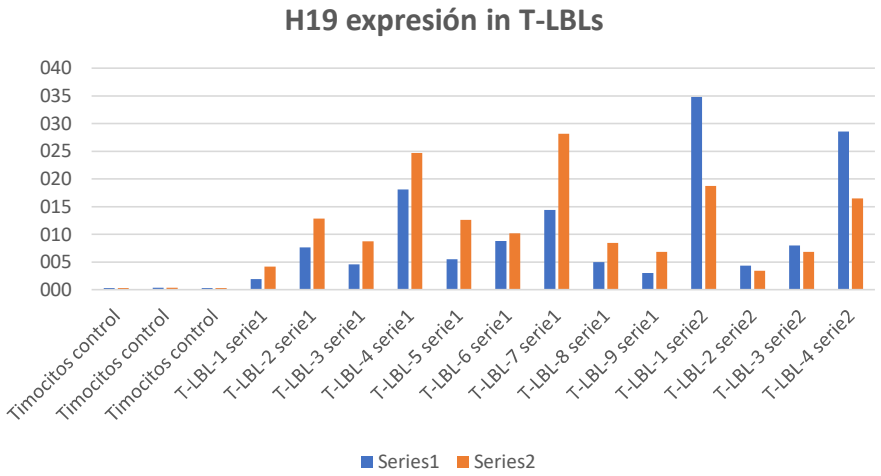
UPREGULATED

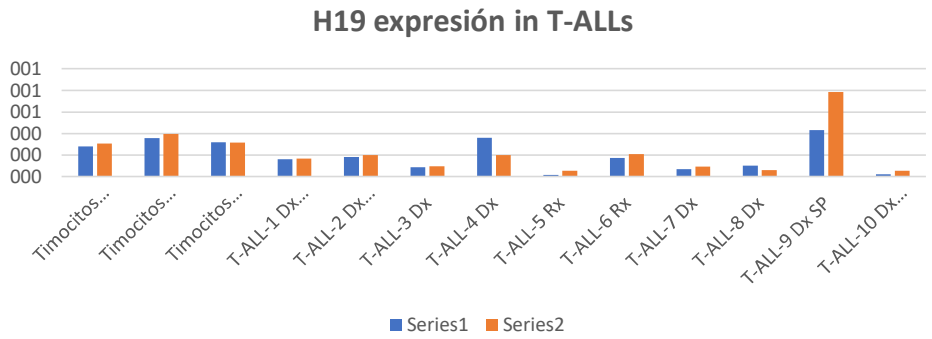


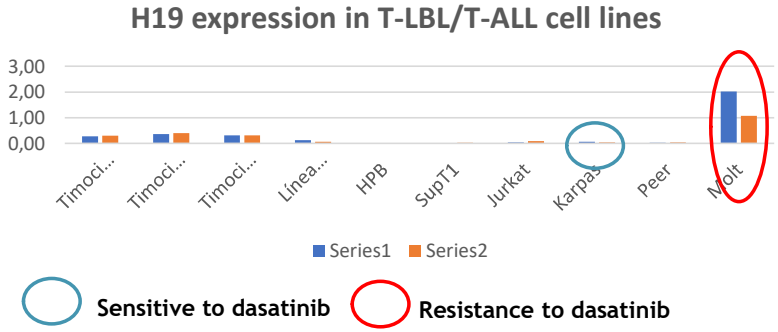
CARCINOGENESIS



The role of H19 in conferring chemoresistance of T- lymphoblastic lymphoma cells







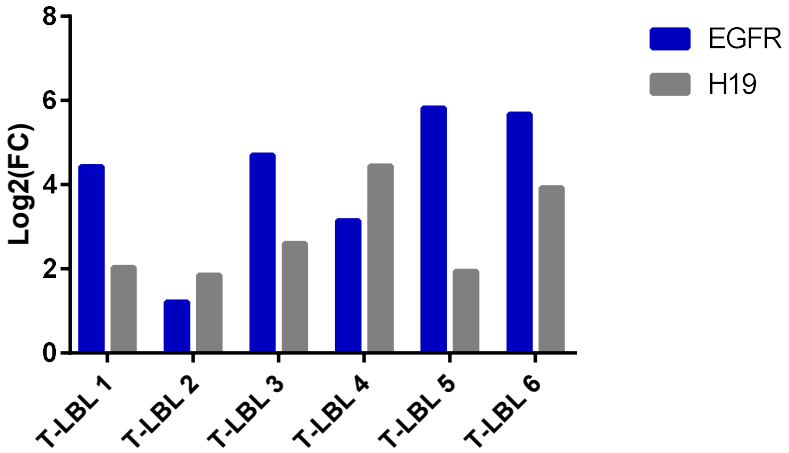
Sensitive to dasatinib

Resistance to dasatinib

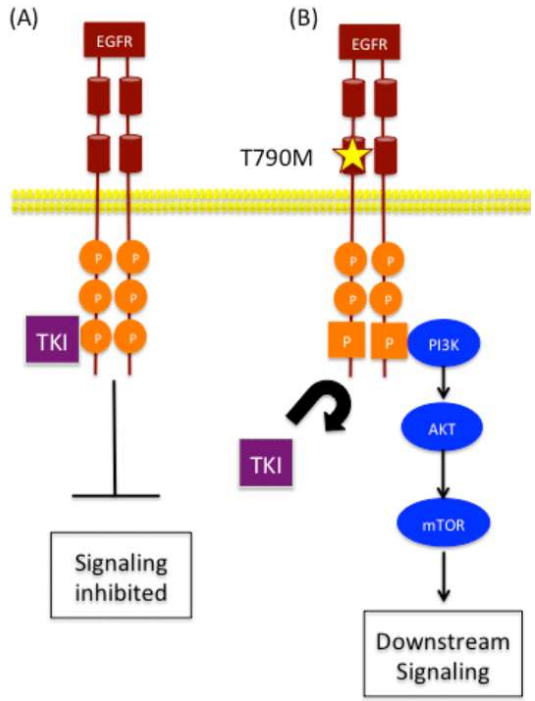
H19: a druggable lncRNA for targeted anti-cancer approaches

LncACTdb3 DownLoad File

Detail	CeRNA1	CeRNA1 type	miRNAs	CeRNA2	Species	Diseases	PubMed ID
	H19	LncRNA	miR-21	EGFR	Homo sapiens (human)	Gastric Cancer	29719612



Concomitant overexpression of *H19* and *EGFR* in T-LBLs



Inhibition of EGFR Signaling by Tyrosine Kinase Inhibitors and Mechanisms of Resistance in Non-small Cell Lung Cancers
(Lam and Levine, 2014)

› Cancer Lett. 2020 Aug 28;486:58-70. doi: 10.1016/j.canlet.2020.05.009. Epub 2020 May 18.

LncRNA H19 downregulation confers erlotinib resistance through upregulation of PKM2 and phosphorylation of AKT in EGFR-mutant lung cancers

Chen Chen [1], Wei-Ran Liu [2], Bin Zhang [1], Lian-Min Zhang [1], Chen-Guang Li [1], Chang Liu [1], Hua Zhang [1], Yan-Song Huo [1], Yu-Chen Ma [1], Peng-Fei Tian [1], Qi Qi [1], Jing-Jing Li [1], Zhe Tang [1], Zhen-Fa Zhang [1], Giuseppe Giaccone [3], Dong-Sheng Yue [4], Chang-Li Wang [5]

H19 overexpression confers erlotinib sensitivity in T-LBL/T-ALL ?????


CBM