FUNCTIONAL ROLE OF OVEREXPRESSED GENES IN iAMP21+ B-CELL PRECURSOR ACUTE LYMPHOBLASTIC LEUKEMIA

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INTRODUCTION

AMP21 has been described in 2% of B-cell Precursor Acute Lymphoblastic Leukemia (BCP-ALL) and is associated with unfavorable prognosis due to high relapse risk (Fig. 1).

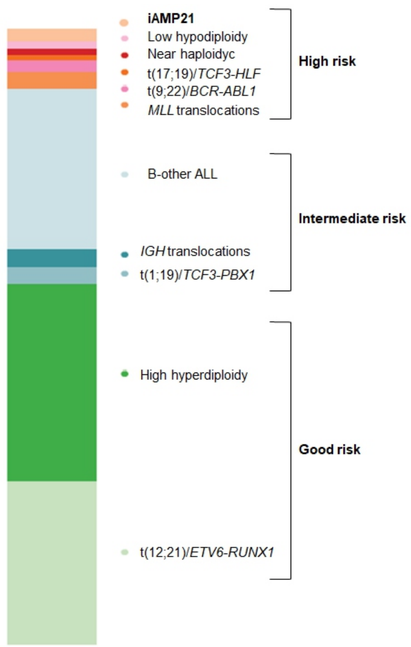

Figure 1. A. Commom Cytogenetic Abnormalites and Their Prognostic Revelence in Children and Adults With B-Cell Acute Lymphoblastic Leukemia – Modified of Moorman. Haematologica. 2016 iAMP21 = intrachromosomal amplification of chromosome 21.

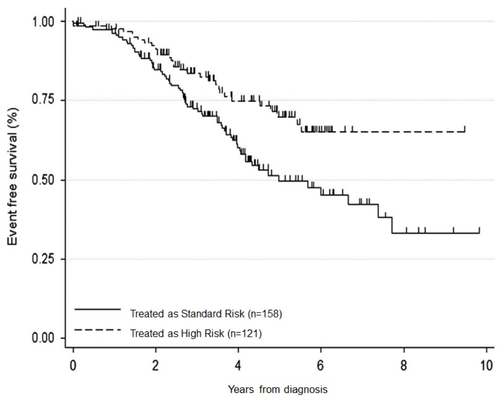

Figure 1. B. Kaplan-Meier Survival Graph of iAMP21 patients according to treatment (Modified of HARRISON CJ et al., 2014)

A striking feature is that all patients exhibit a common region of amplification at chromosome 21 (*ERG* and *ETS2* genes), as well as overexpression of *LGMN* gene (Fig. 2).

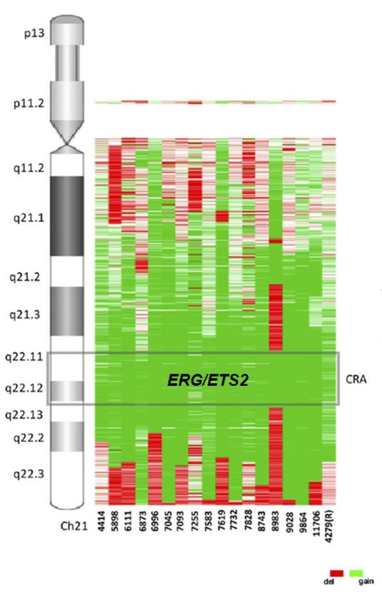

Figure 2.A. Summary of abnormalities in iAMP21 BCP-ALL. Heat map of chromosome 21 abnormalities detailing the regions of deletion (red), gain (green), and normal copy number (white) relative to genomic location. (Modified of Rand V, et al., 2011).

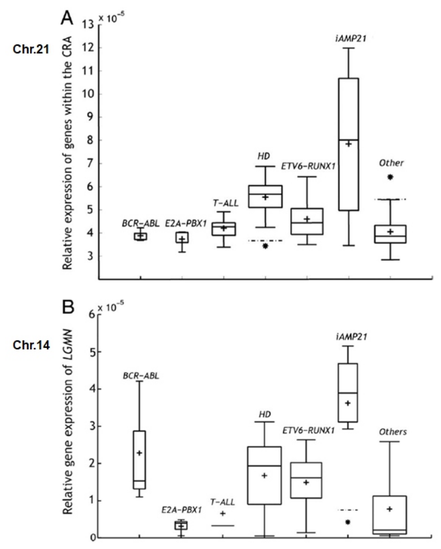

Figure 2.B. Box plot diagrams illustrating expression of those genes within the CRA [A] and *LGMN* expression [B]. (Strefford, JC et al. 2006)

Additionaly, it has been recently shown that *OPN* is differentially expressed in BCP cells at the time of relapse (Fig. 3).

Figure 3: OPN perform distinct roles in HMs. In the bone marrow (BM) niche, upregulated OPN secreted by malignant clones, as well as by additional cells present in the BM microenvironment (e.g. osteoblasts) can promote differentiation of hematopoietic progenitors and stem cells. Overexpressed OPN can then induce several steps elated to HMs progression, such as cell adhesion, invasion, tumor growth, cell survival, dormancy, angiogenesis and osteoclatogenesis. Each of these tumor features mostly occur depending on each type of HM. In response to OPN overexpression, tumor cells can also invade or infiltrate other tissues, such as extramedullary sites and central nervous system (CNS) (Bastos et al., 2017).

It has been described that *ERG, ETS2* and *LGMN* play key roles in cell differentiation.
Besides, deregulated expression of these genes in other tumor types has been associated with poor prognosis and/or resistance to chemotherapy.

HYPOTHESIS

Deregulated expression of *ERG, ETS2, LGMN* and *OPN* is correlated with chemotherapeutic resistance (CR) in BCP-ALL.

OBJECTIVE

To evaluate the role of E*RG, ETS2, LGMN* and *OPN* in ALL CR.

METHODS

Experimental design

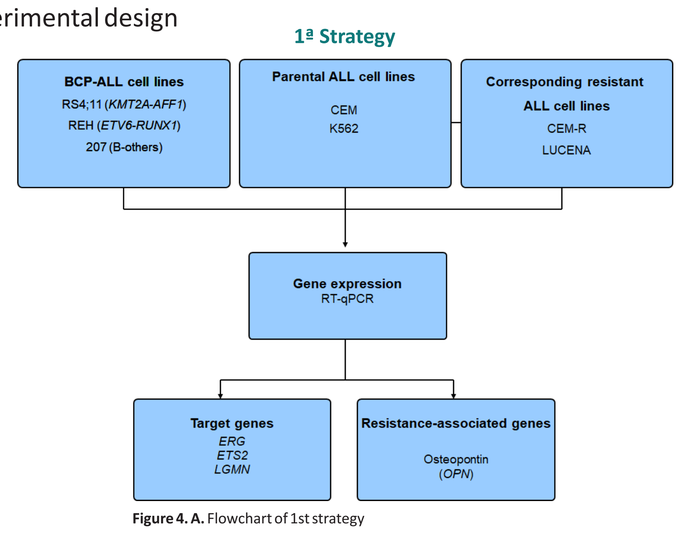

Figure 4. A. Flowchart of 1st strategy

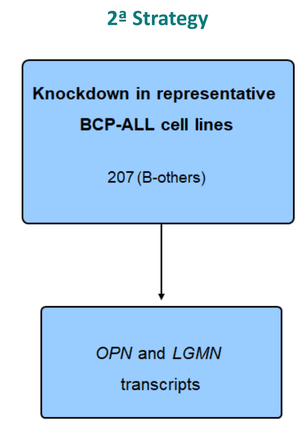

Figure 4. B. Flowchart of 2nd strategy

RESULTS

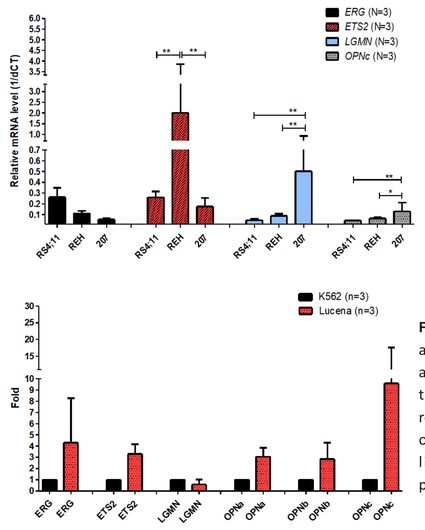

Figure 5. mRNA levels in BCP-ALL cell lines. Higher levels mRNA of *ERG* and *ETS2* were observed in RS4;11 and REH cell lines, respectively. Higher levels of *LGMN* and *OPNc* were found in 207 cells. *P < 0.05; **P < 0.01 (Student's t-test).

Figure 6. mRNA levels in resistant and parental ALL cell lines. *ERG*, *ETS2* and the three OPN splice variants transcripts are overexpressed in both resistant cell lines and *LGMN* is only overexpressed in CEM resistant cell line, when compared to their parental cells.

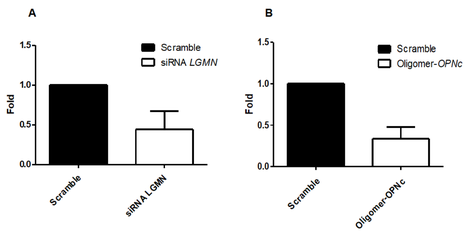

Figure 7. Illustrative figure of the *LGMN* and *OPNc* knockout. **A**. *LGMN* knockdown (207 cell line); **B**. OPNc knockdown (207 cell line).

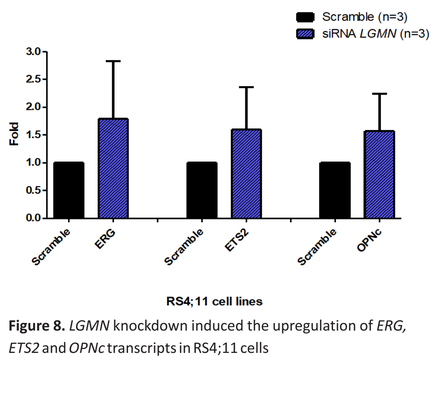

Figure 8. *LGMN* knockdown induced the upregulation of *ERG, ETS2* and *OPNc* transcripts in RS4;11 cells

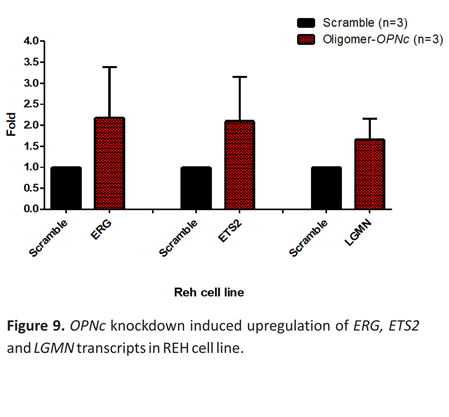

Figure 9. *OPNc* knockdown induced upregulation of *ERG, ETS2* and *LGMN* transcripts in REH cell line.

However, *ERG, ETS2* and *LGMN* transcripts were downregulated when *OPNc* was knocked-down in BCP-ALL cell lines.

CONCLUSION

Our preliminary data suggest that expression of evaluated genes can be differentially modulated upon *LGMN* or *OPNc* silencing in each leukaemia cell lineage subtype.

PERSPECTIVES

Functional *in vitro* assays are ongoing to evaluate their roles in CR.

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