FREQUENCY OF *JAK2* AND *JAK3* MUTATIONS AND THEIR PROGNOSTIC ROLE IN ACUTE LYMPHOBLASTIC LEUKAEMIA

Karolyne Wolch[1], Thais Ferraz Aguiar[1,2], Lavínia Lustosa Bergier[3], Thais Alcantara Bonilha[2], Eduardo Cerello Chapchap[4], Elaine Sobral Costa[5], Jane Almeida Dobbin[6], Maura Rosane Valerio Ikoma[7], Marcelo Land[5], Mecneide Mendes Lins[8], Marcia Trindade Schramm[3,6], Mariana Emerenciano[1] and Marcela B. Mansur[1]

[1]Molecular Cancer Study group, Division of Clinical Research, Research Centre, Instituto Nacional de Câncer - INCA, Rio de Janeiro, RJ, Brazil; [2]HEMORIO, RJ, Brazil; [3]Prontobaby Hospital da Criança, RJ, Brazil; [4]Hospital Albert Einstein, SP, Brazil; 5IPPMG - UFRJ, RJ, Brazil; [6]Hospital do Câncer I – INCA, RJ, Brazil; [7]Hospital Amaral Carvalho, SP, Brazil; [8]IMIP, PE, Brazil

Introduction

- Acute lymphoblastic leukaemia (ALL) is the most common haematological malignancy in paediatric patients, but it also occurs in adults.
- ALL has a heterogeneous molecular profile characterized by sequential acquisition of genomic abnormalities that contribute to the leukaemogenic process (Figure 1).

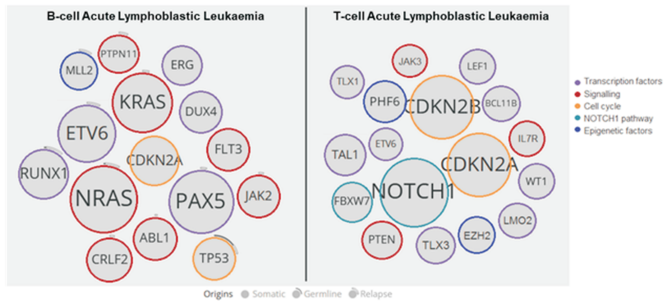


Figure 1. Genomic profile of acute lymphoblastic leukaemia (ALL). The schemes show the most frequently observed alterations in B- and T-cell ALL, respectively. Alterations are coloured according to gene function, i.e. transcription factors, signalling, cell cycle, NOTCH1 pathway and epigenetic factors. The size of the circles represents the frequency of alterations affecting those genes. Both figures were adapted from 2015-2018 St. Jude Children's Research Hospital – Pediatric Cancer Data Portal (PeCan).

- Mutations in genes of the JAK family, e.g. *JAK2* and *JAK3*, are potential biomarkers for ALL risk stratification and therapeutic decisions.
- *JAK2* and *JAK3* mutations result in constitutive activation of JAK-STAT, PI3K/AKT and MAPK signalling pathways contributing to the malignant transformation of haematopoietic precursors (Figure 2).

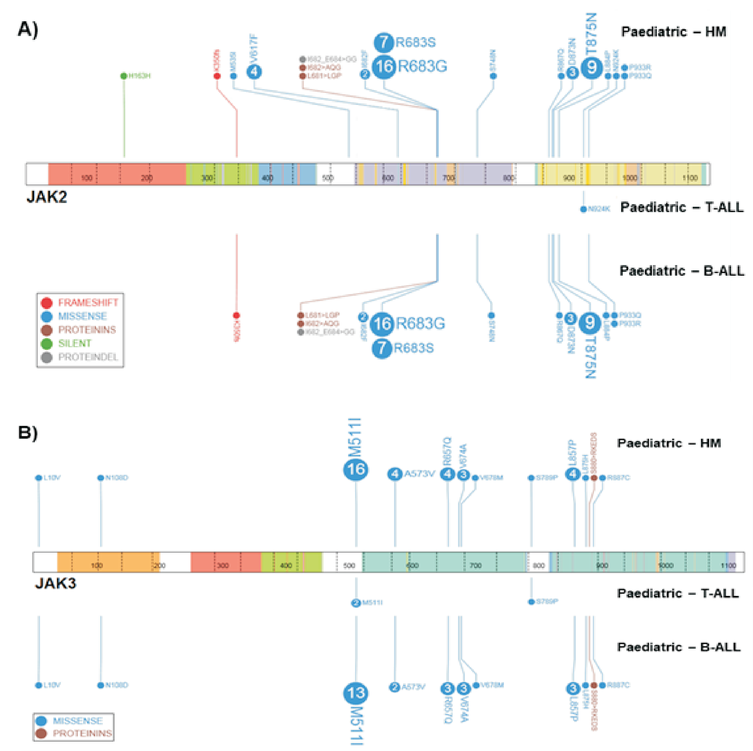
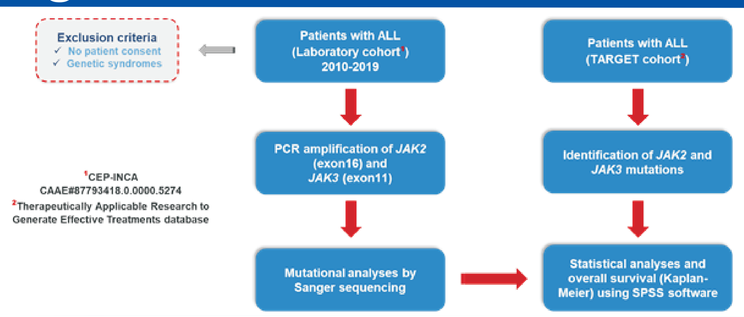


Figure 2. JAK2 and JAK3 protein schemes showing the locations/domains affected by recurrent mutations. **A)** R683_ is the most common change caused by a point mutation in the pseudokinase (JH2) domain of *JAK2*, particularly in B-ALL. **B)** In *JAK3*, SH2-pseudokinase (JH2) linker sequence is were the most common mutation (M511I) occurs, particularly in T-ALL cases. Figures were modified from St. Jude PeCan Data Portal (https://pecan.stjude.cloud/proteinpaint/JAK2 and https://pecan.stjude.cloud/proteinpaint/JAK3, respectively).

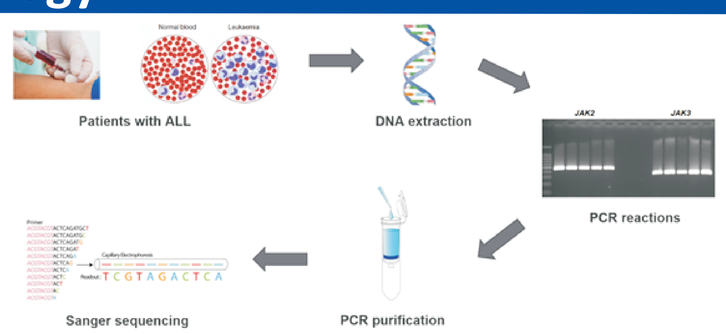
Aims

- The present proposal aims to determine the frequency of *JAK2*/*JAK3* mutations in a series of Brazilian ALL cases and to investigate the potential prognostic impact of these markers on patient's overall survival (OS).

Study-design



Methodology



Results

Table 1. Frequency according to clinical-demographic characteristics of laboratory case series and TARGET cohort.

Variables	n (%)	n TARGET (%)
Age (years)[a]		
<10	30 (39.0)	147 (55.7)
10-21	25 (32.5)	113 (42.8)
>21	20 (26.0)	4 (1.5)
Gender		
Male	50 (64.9)	202 (76.5)
Female	27 (35.1)	62 (23.5)
WBC (x10⁹/L)[b]		
<50	46 (59.7)	91 (34.5)
≥50	28 (36.4)	173 (65.5)
ALL type		
B-ALL	67 (87.0)	-
T-ALL	10 (13.0)	264 (100)
***JAK2* status[c]**		
WT	60 (96.8)	263 (99.62)
Mutated	2 (3.2)	1 (0.38)
***JAK3* status[c]**		
WT[d]	64 (98.5)	244 (92.42)
Mutated	1 (1.5)	20 (7.57)
Total	**77 (100)**	**264 (100)**

a, for 2 cases the age was not informed; b, 3 cases did not have white blood cells (WBC) information; c, 15 cases could not be evaluated for *JAK2* and 12 for *JAK3* due to insufficient biological material (DNA). d, among these cases, 3 SNPs were found (rs55883965, rs144203232 e rs565294516).

Table 2. Molecular characteristics of TARGET patients categorized according to the presence of mutations in *JAK3*.

	***JAK3* status n (%)**		
Variables	**Mutated**	**WT**	***p value***
***NOTCH1* status**			*0.023*
Mutated	19 (95.0)	175 (71.7)	
WT	1 (5.0)	69 (28.3)	
***FBXW7* status**			*0.043*
Mutated	1 (5.0)	61 (25.0)	
WT	19 (95.0)	183 (75.0)	
***NOTCH1/FBXW7* status**			*0.061*
Mutated	19 (95.0)	188 (77.0)	
WT	5 (62.50)	56 (23.0)	
***CRLF2* expression**			*0.272*
High	5 (25.0)	38 (15.6)	
Low	15 (75.0)	206 (84.4)	
TOTAL	**20 (7.6)**	**244 (92.4)**	

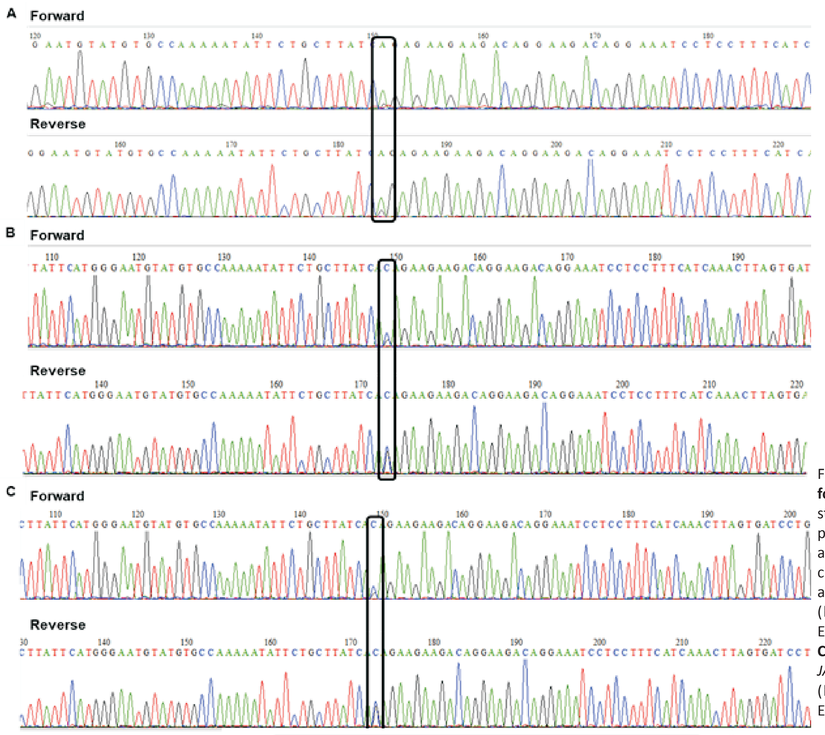


Figure 3. *JAK2*-exon 16 and *JAK3*-exon 11 sequencing for mutated cases. As we can observe in both DNA strands (forward and reverse) there are overlapping peaks (inside the black boxes) proving the occurrence of a point mutation in these positions. **A)** a simple base change A>G at position c.2541 for *JAK2*, **B)** a change G>C at position c.2542 for *JAK2* (https://www.ncbi.nlm.nih.gov/gene/3717 and Ensembl: ENSG00000096968). **C)** We detected the c.1633G>C/p.M511I mutation for *JAK3* in a T-ALLcase (https://www.ncbi.nlm.nih.gov/gene/3718 and Ensembl: ENSG00000105639).

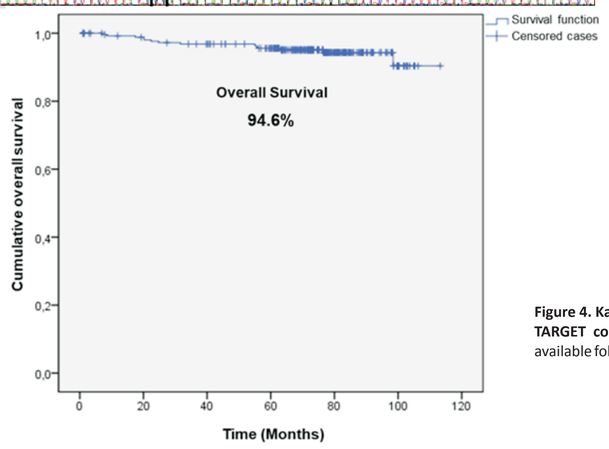


Figure 4. Kaplan-Meier curve of OS for T-ALL patients with in the TARGET cohort. Cumulative OS for the 261/264 patients with available follow-up data was 94.6%.

Conclusion

- All the *JAK2* and *JAK3* mutations detected in our series have already been described in the literature as alterations that result in constitutive activation of these proteins.
- To further explore the role of *NOTCH1/FBXW7*, we intend to analyse which type of mutations are affecting these genes and if different mutational profiles associate with *JAK2/JAK3* status.
- The 94.6% probability of OS observed in the T-ALL TARGET cohort is relatively high when compared to the literature (~75%), but it is important to highlight that TARGET patients were all diagnosed and treated in a reference centre (St. Jude Children's Research Hospital) that offers molecular risk-adapted therapeutic protocols.

Projeto Gráfico: Área de Edição e Produção de Materiais Técnico-Científicos / INCA

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