2023 – Del8p and TNFRSF10B Loss are associated with Poor Prognosis and Resistance to Fludarabine in Chronic Lymphocytic Leukemia

Ludovic Jondreville[1,2], Lea Deghane[1], Cecile Doualle[1], Luce Smagghe[1], Béatrice Grange[2], Frederic Davi[1,2], Leticia Lerner[1], Delphine Garnier[1], Clotilde Bravetti[1,2], Olivier Tournilhac[3], Damien Roos-Weil[1,2], Marouane Boubaya[4], Elise Chapiro[1,2]*, Santos A Susin[1]*, Florence Nguyen-Khac[1,2]*

1. Centre de Recherche des Cordeliers, INSERM, Sorbonne Université, Paris, France
2. Hematology Department, Pitié-Salpêtrière Hospital, AP-HP, Paris, France
3. Hematology Department, CHU Estaing, Clermont-Ferrand, France
4. Clinical Research Unit, Avicenne Hospital, AP-HP, Bobigny, France

INTRODUCTION

Chronic lymphocytic leukemia (CLL) is a heterogeneous disease, whose prognosis is affected by a variety of **cytogenetic abnormalities**.[1]

The deletion of the short arm of chromosome 8 (**del8p**) is a rare (<5% of cases) but recurrent abnormality in CLL, which has been associated to **poor outcome**, due to higher proliferation rate, concurrent adverse genetics and ibrutinib resistance. A possible role of the ***TNFRSF10A/B* genes**, whose proteins induce apoptosis after their ligand TRAIL binding, has been reported.[2]

OBJECTIVE

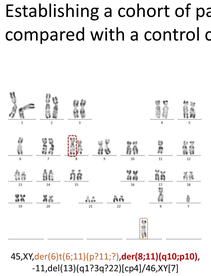
Precise characterization of the del8p in CLL:

. Description of **associated cytogenetic abnormalities**.

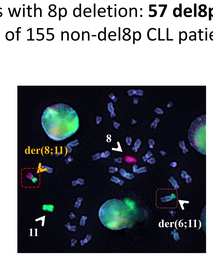
. Identification of **deregulated genes** underlying CLL progression and **drug resistance**.

PATIENTS AND METHODS

Establishing a cohort of patients with 8p deletion: **57 del8p CLL patients identified**, compared with a control cohort of 155 non-del8p CLL patients



Karyotype



Chromosome 8 = red probe;
chromosome 11 = green probe

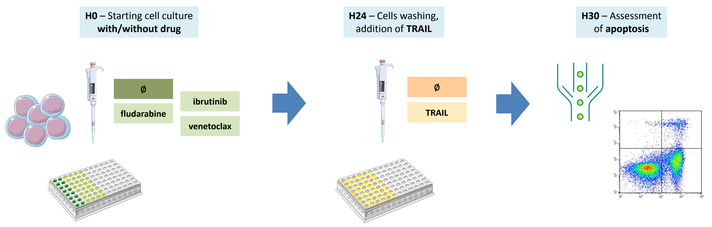
FISH

FISH

Generation of stable ***TNFRSF10A*$^{-/-}$** and/or ***TNFRSF10B*$^{-/-}$ CRISPR/Cas9 edited OSU-CLL cell lines** using the procedure described by Cao et al.[3]

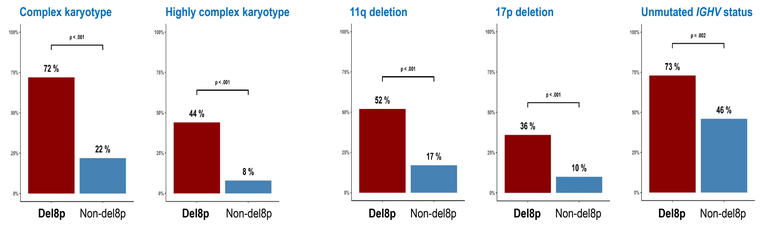
Droplet digital PCR (ddPCR) using Bio-Rad's QX200 ddPCR System; results normalized with *ABL1* gene reference.

Cell apoptosis measured by flow cytometry, using standard **annexin V / PI staining** protocol.
Sequential approach:

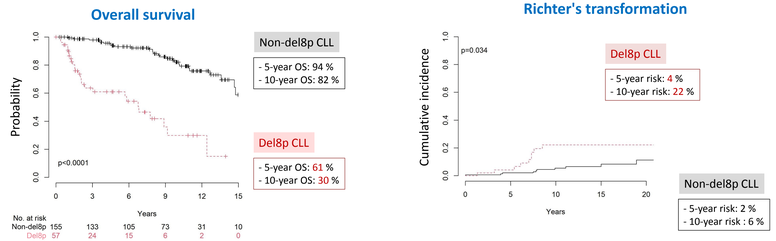


RESULTS

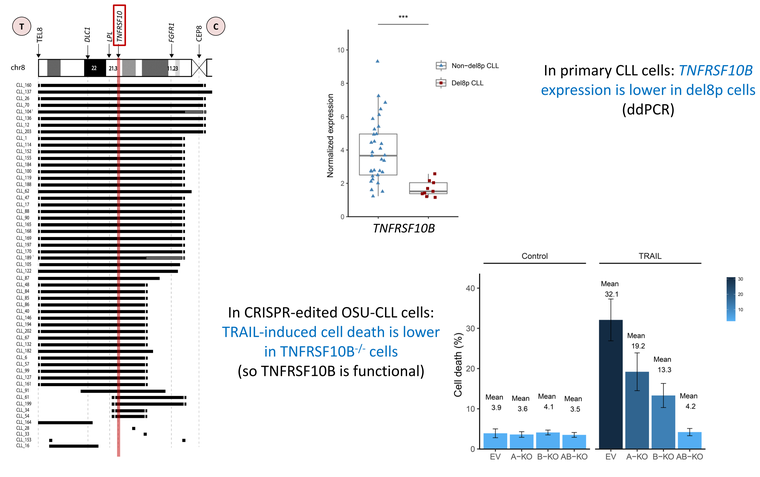
1 Del8p is significantly associated with poor prognostic factors



2 Del8p CLL patients have a poor outcome

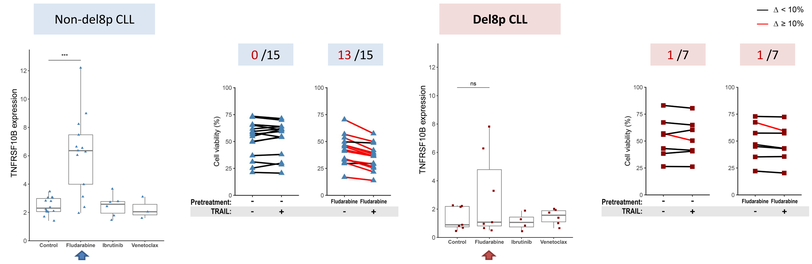


3 One copy of *TNFRSF10B* gene is lost in 91% (51/56) of del8p CLL patients

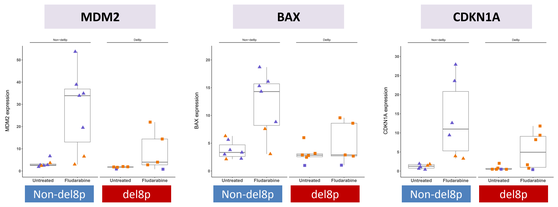


RESULTS

4 Fludarabine significantly increases *TNFRSF10B* expression and therefore TRAIL induced cell mortality in non-del8p CLL samples, but not in del8p CLL samples



5 del8p interferes with the p53-dependent gene expression induced by fludarabin



CONCLUSIONS

Del8p is a rare but recurrent chromosomal abnormality in CLL, associated with **poor prognostic factors** (such as complex and highly complex karyotypes, del11q, del17p, unmutated *IGHV* status), **short TTFT and OS**, and **risk of RT**.

Interestingly, del8p CLL treated with fludarabine-based regimens had a shorter next treatment-free survival (10y: 3.7% vs 21.5% without new treatment, p=.002) and a shorter OS (10y: 45.9% vs. 81.6%, p<.001) compared to non-del8p CLL, including CLL with mutated *IGHV*. We demonstrate that ***TNFRSF10B*** plays an important role in fludarabine resistance in CLL, *in vivo* and *in vitro*.

Our results argue for the assessment of del8p before choosing a fludarabine-based therapy.

ACKNOWLEDGEMENTS

FONDATION ARC POUR LA RECHERCHE SUR LE CANCER
Reconnue d'utilité publique
Inserm
La science pour la santé
From science to health
FRENCH INNOVATIVE LEUKEMIA ORGANIZATION
Gefluc
Les entreprises contre le cancer
Paris - Ile-de-France
INSTITUT NATIONAL DU CANCER

REFERENCES

1. Hallek et al., 2018, Blood
2. Burger et al., 2016, Nat Commun.
3. Cao et al., 2016, Nucleic Acids Res.

CONTACT INFORMATION

ludovic.jondreville@aphp.fr
florence.nguyen-khac@aphp.fr