NEWSLETTER - October 2018

Next Generation Sequencing (NGS) and Droplet Digital PCR mutation testing project for optimal monitoring of your CML and Ph+ ALL patients

Dear colleagues,

*Since the start of our project in 2016, 152 samples from CML and Ph+ ALL patients have been evaluated, these were sent by 26 centers accross Belgium and Luxembourg. We detected **34 clinically relevant** mutations : 19% of CML samples had TKI resistance mutation(s) and 50% of the Ph+ ALL samples. We are looking forward to receiving additional samples for analysis!*

Details of detected mutations

CML Sum	Mutation														Mutated
	M237T	G250V	E255K	V299M	T315I	C330R	E355A	F359I	E459K	Y253H/ T315I	E255K/ V299L	E255K/ T315I	E255K/ E275K/ V299L/ F317L	E255K/ T315I/ M244V/ E355G	
136	1	1	3	1	3	1	1	2	1	3	1	5	1	1	19%

Ph+ALL Sum	Mutation					Mutated
	E255K	T315I	F317L	D455G	F359V / Y253H	
18	4	2	1	1	1	50%

Compound mutations

CLINICAL SIGNIFICANCE OF COMPOUND MUTATIONS

An emerging problem in patients with Philadelphia (Phi)-positive leukaemia is the occurrence of cells with multiple mutations in the BCR-ABL1 tyrosine kinase domain (TKD) associated with high resistance to different tyrosine kinase inhibitors. Rapid and sensitive detection of leukaemic subclones carrying such changes, referred to as compound mutations, is therefore of increasing clinical relevance and supports the optimal therapy choice.

CHANGES IN THE NGS PROJECT !

- We will continue (for free) NGS & digital PCR testing
- NEW: We will include compound mutation analysis

Rationale: Another key component in TKI resistance are **compound mutations.** Distinguishing between Bcr-Abl1 mutations located on the same DNA strand, coming from an identical malignant clone, and those located on 2 or more different DNA strands, coming from 2 or more different malignant clones could be useful to monitor and module the patient's therapeutic strategy and prevent progressions or treatment failures of Phi+ leukaemia. This distinction can be performed by digital PCR phasing. After validation of the analysis tool by using synthetic oligonucleotides mimicking monoclonal and multiclonal resistant leukaemia cells, we apply it to clinical samples.

This project is supported by an educational grant from Incyte Biosciences Benelux BV

SAMPLE PROCEDURE

- preferred: 10 to 20 µl cDNA (at room temperature)
- or: 1 µl RNA (or at least < 1 µl) RNA (at -20°C)
- or: 3 ml of EDTA (at room temperature)

SHIP TO (PREFERABLY WITHIN 48 HOURS)

IPG - Dr. Pascal Vannuffel / Dr. Céline De Rop
NGS project
Avenue George Lemaître 25
6041 Gosselies (Belgium)

PLEASE COMPLETE THIS SECTION WITH NECESSARY DATA

Patient ID: ..... Sample ID: ..... Sample date: .....

Sample source: ☐ peripheral blood ☐ bone marrow

Diagnosis: ☐ CP-CML ☐ BP-CML ☐ AP-CML ☐ Ph+ ALL

% BCR-ABL15 transcript level: .....

If Ph+ ALL, specify isoform of BCR-ABL: ☐ P210 ☐ P190

Prior TKI treatment(s): .....

Confirm that there is NO suspected lack of adherence ☐ Yes

Doctor: .....

E-mail: .....

Institution: .....

City: .....

Additional information if possibly relevant: .....

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IMPORTANT

- If questions, do not hesistate to call or e-mail one of the investigations
- We will also be pleased to explain the project during a Hemato Staff meeting in your hospital
- The samples will be analyzed and results will be reported to you within 2 weeks
- Compound mutations results may need 1 week more for analyzing & reporting
- The analysis of samples via this Project does not subsritute for the analysis of samples performed in your routine clinical practice
- The costs of sample analysis are covered by the project
- This project is supported by an unrestricted educational grant from Incyte Biosciences Benelux

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