Personal note

To Jo and Fritz — Thank you for your guidance and for embodying what it means to be a true scientist: ever-curious, unwaveringly persistent, and driven by an insatiable thirst for knowledge.

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Ik hou van je!

Short curriculum vitae

2009 – 2016: Master of Science in Medicine
Ghent University, Belgium

2016 – 2022: Master of Science in Internal Medicine
Ghent University, Belgium

2022 – 2024: Specialization in Clinical Hematology
Ghent University Hospital, Belgium

2020– present: Doctoral fellow
OncoRNALab, Ghent University, Belgium

2023: Research stay
German Cancer Research Center (DKFZ)
Heidelberg, Germany

2024 – present: Clinical Hematologist
Ghent University Hospital, Belgium

Examination committee

Prof. dr. ir. Katleen De Preter (chairman)
Department of Biomolecular Medicine, VIB-UGent Center for Medical Biotechnology (Ghent, Belgium)

Prof. dr. Peter Vandenberghe
Department of Hematology, University Hospitals Leuven (Leuven, Belgium)

Prof. dr. J.H.M. van den Berg
Department of Pathology and Medical biology, University Medical Centre Groningen (Groningen, The Netherlands)

Prof. dr. Bram De Wilde
Department of Internal Medicine and Pediatrics, Ghent University Hospital (Ghent, Belgium)

Prof. dr. Tom Van Maerken
Department of Laboratory Medicine, AZ Groeninge (Kortrijk, Belgium)

Dr. Mattias Hofmans
Department of Laboratory Medicine, Ghent University Hospital (Ghent, Belgium)

Contact details

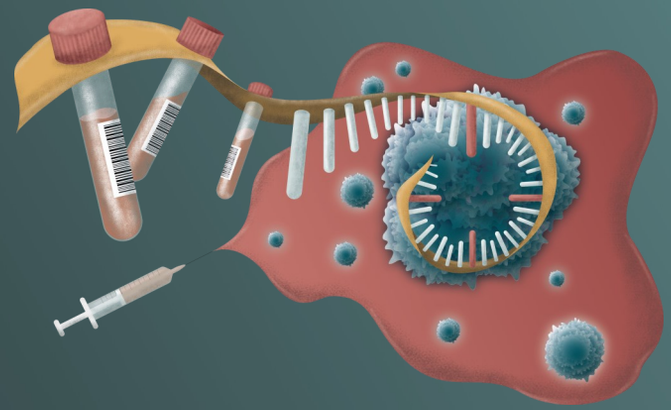
philippe.decruyenaere@uzgent.be

linkedin.com/in/philippe-decruyenaere

PhD thesis

Exploring the Cell-Free Transcriptome as a Biomarker Source in Non-Hodgkin Lymphoma

Philippe Decruyenaere



Promotor
Prof. dr. ir. Jo Vandesompele

Co-promotor
Prof. dr. Fritz Offner

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FACULTY OF MEDICINE AND HEALTH SCIENCES

Achtergrond

Ons lichaam bestaat uit miljarden cellen die voortdurend communiceren en informatie uitwisselen. Hierbij komen kleine stukjes genetisch materiaal in het bloed terecht, zoals celvrij RNA (cfRNA). Bij kanker – waaronder verschillende vormen van lymfeklierkanker – kunnen ook tumorcellen cfRNA afgeven. Door dit cfRNA op te vangen via een eenvoudige bloedafname, kunnen we waardevolle informatie verzamelen over de tumor, zonder dat er een ingrijpende biopsie nodig is. Dit wordt een vloeibare biopsie genoemd.

cfRNA geeft bovendien niet alleen een momentopname van wat er in de tumor gebeurt, maar toont ook hoe het immuunsysteem reageert. Zo kunnen artsen beter begrijpen welk type lymfoom iemand heeft, hoe agressief het is, en of een behandeling zal aanslaan. Dit kan het mogelijk maken om sneller en preciezer in te grijpen, afgestemd op de unieke situatie van de patiënt. Onderzoek naar cfRNA biedt dus een veelbelovende stap richting meer gepersonaliseerde, minder belastende en effectievere kankerzorg in de toekomst.

Key messages

- cfRNA is a promising liquid biopsy tool for non-Hodgkin lymphomas, reflecting functional, real-time changes.
- Standardization is essential for reliable (cf)RNA analysis across labs and clinics.
- We have identified a cfRNA signature that can predict response to first-line treatment in DLBCL.
- Besides tumoral signals, cfRNA profiles reflect healthy tissue and immune signals, with distinct dynamics.
- cfRNA lets us spy on tumors… without needing a scalpel — science meets stealth. 

The details

This PhD thesis investigates the potential of cell-free RNA (cfRNA) as a minimally invasive biomarker for the diagnosis, monitoring, and treatment guidance of non-Hodgkin lymphomas, with a focus on diffuse large B-cell lymphoma (DLBCL), primary mediastinal B-cell lymphoma (PMBCL), and Waldenström's macroglobulinemia (WM). cfRNA reflects real-time molecular changes in both the tumor and the immune system, making it a promising tool for precision medicine.

The research is structured around three key objectives:

1. **Standardization:** The first part addresses the need for standardized protocols in (cf)RNA research. Pre-analytical factors—such as blood collection methods and RNA extraction protocols—were systematically evaluated for their impact on RNA quality and yield. These findings provide guidance for reproducibility and clinical implementation. (**Papers 3** and **5**)
2. **Biomarker Discovery:** The second part involves deep, unbiased profiling of the circulating transcriptome using total RNA sequencing. Distinct cfRNA signatures with diagnostic, prognostic, and predictive value were identified in patients with DLBCL, PMBCL, and WM. These patterns also reveal tumor heterogeneity and track changes during disease progression and treatment. Lastly, cfRNA concentrations by themselves reflect disease status and response. (**Papers 1, 2** and **4**)
3. **Biological Insights**: Lastly, the origin and dynamics of cfRNA were studied in a mouse model bearing human DLBCL tumors. This model enabled detailed tracking of cfRNA fluctuations during tumor growth and therapy, separating tumor-derived from host-derived RNA signals. (**Paper 6**)

Overall, this thesis highlights cfRNA as a powerful liquid biopsy tool, paving the way for its future integration into clinical decision-making and personalized medicine.

Publications included in this thesis

1. **Decruyenaere P** *et al.* 2021. Circulating RNA biomarkers in diffuse large B-cell lymphoma: a systematic review. Experimental Hematology & Oncology.
2. Drandi D*, **Decruyenaere P*** *et al.* 2022. Nucleic Acid Biomarkers in Waldenström Macroglobulinemia and IgM-MGUS: Current Insights and Clinical Relevance. Diagnostics.
3. **Decruyenaere P** *et al.* 2023. RNA Extraction Method Impacts Quality Metrics and Sequencing Results in Formalin-Fixed, Paraffin-Embedded Tissue Samples. Laboratory Investigation.
4. **Decruyenaere P** *et al.* 2023. Exploring the cell-free total RNA transcriptome in diffuse large B-cell lymphoma and primary mediastinal B-cell lymphoma patients as biomarker source in blood plasma liquid biopsies. Frontiers in Oncology.
5. Van der Schueren C*, **Decruyenaere P*** *et al.* 2024. Subpar reporting of pre-analytical variables in RNA-focused blood plasma studies. Molecular Oncology.
6. **Decruyenaere P***, Daneels W* *et al.* 2024. Characterizing the Cell-Free Transcriptome in a Humanized Diffuse Large B-Cell Lymphoma Patient-Derived Tumor Xenograft Model for RNA-Based Liquid Biopsy in a Preclinical Setting. International Journal of Molecular Sciences.

*Contributed equally

(Co)-authored publications not included

1. Morlion A *et al.* 2025. Under revision in Nature Communications.
2. Schoofs K *et al.* 2024. Scientific reports.
3. ExRNAQC consortium. 2024. Nature Communications.
4. Decruyenaere P *et al.* 2022. Acta Clinica Belgica.
5. Decruyenaere P *et al.* 2022. Eurorad.
6. Decruyenaere P *et al.* 2020. Acta Clinica Belgica.