FROM LIQUID BIOPSY TO THE PATIENT: THERAPY MONITORING…

… *IN PROGRESS OF LYMPHOMA*

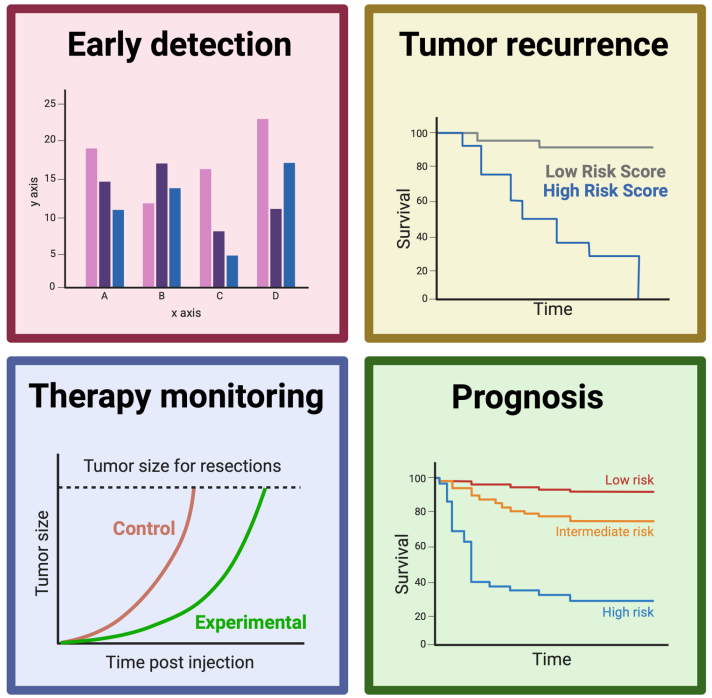
Filippo Maltoni

IRCCS Azienda Ospedaliero-Universitaria di Bologna
Department of Medical and Surgical Sciences
Institute of Hematology "L. and A. Seràgnoli"
University of Bologna
Bologna, Italy

Disclosures of Filippo Maltoni

Company name	Research support	Employee	Consultant	Stockholder	Speakers bureau	Advisory board	Other
None							

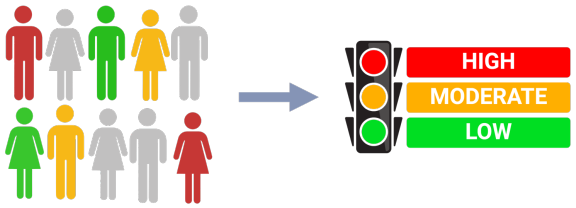
Liquid biopsy has emerged as a promising diagnostic and monitoring approach for the detection and characterization of cancers using bodily biofluids, such as blood



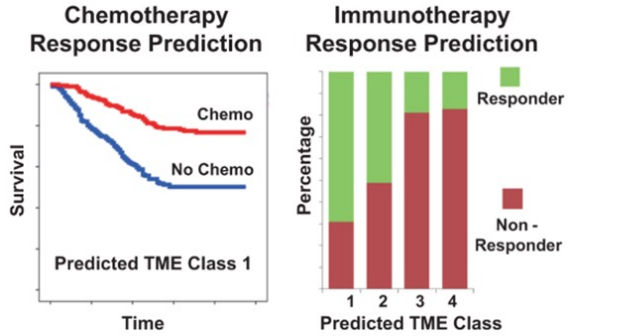
	Tissue biopsy	Liquid biopsy	Imaging/PET-CT
Accessibility	Invasive	Minimally invasive	Non-invasive
Sampling risk	Non-minimal, biopsy site dependent	Minimal	Minimal
Data analysis	Days to weeks	Weeks to months	Hours to days
Relative cost	Sampling: moderate Analysus: low	Sampling: low Analysis: high	Moderate
Applications	Diagnosis and relapse detection	Diagnosis, response assesment, MRD monitoring and clonal evolution	Principal method for lymphoma staging and menagement, robust NPV for treatment response
Limitations	Tumor heterogeneity, accessibility issues, low-quality material for molecular analyses	Lack of standardization, costly analytical methodologies	Weak PPV for clinical utility, radiation risk, not suitable for MRD, high false.positive rate (risk of overtreatment)

THE NECESSITY OF DISEASE MONITORING IN LYMPHOMA

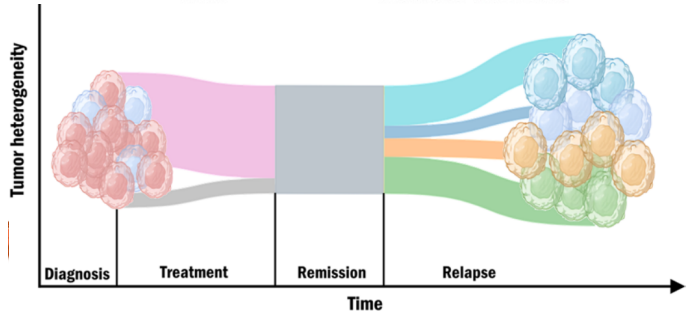
Can we identify high-risk patients at presentation or during initial treatment?



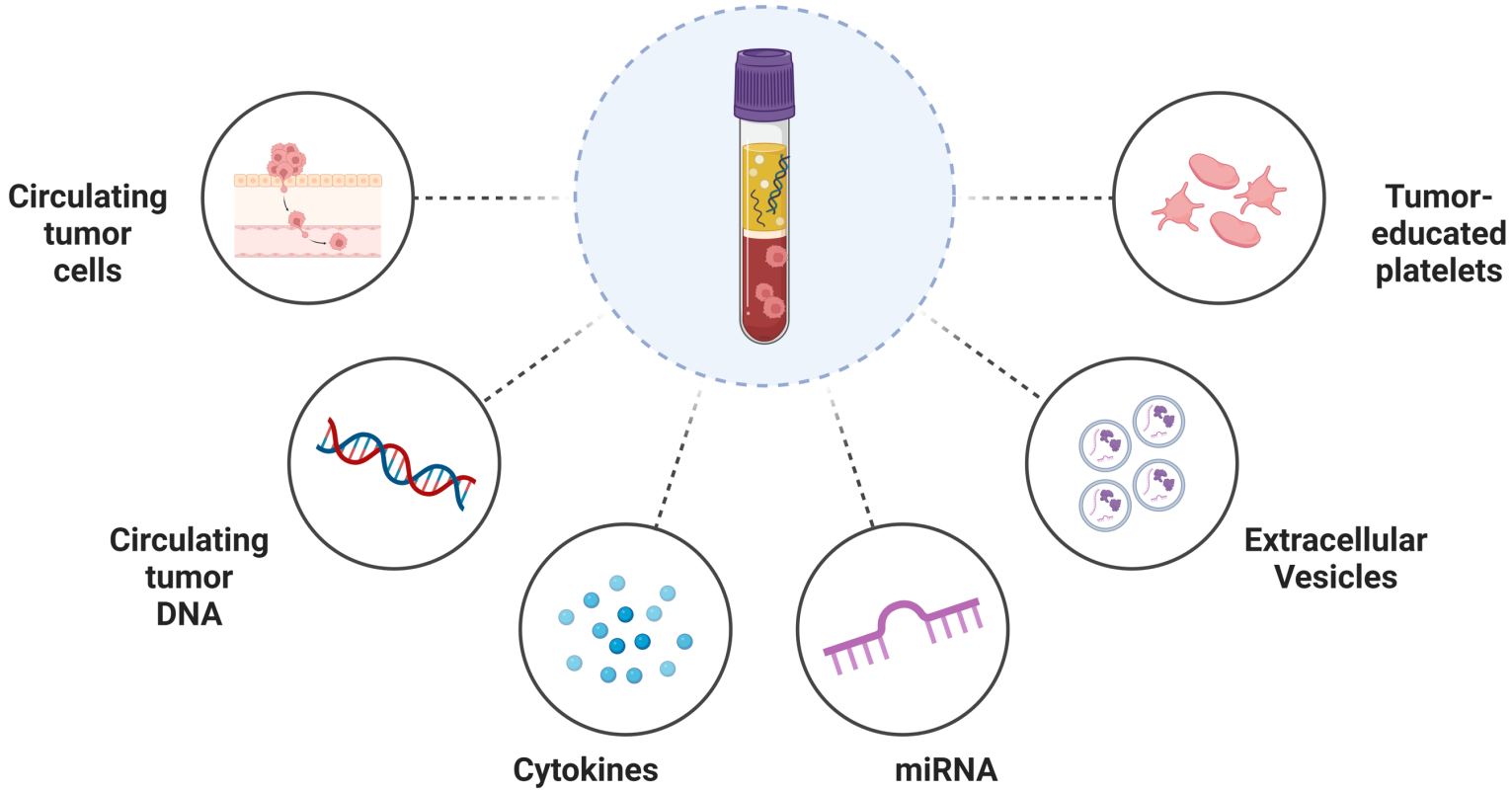
Can we improve patient selection and predict who will respond and who will not to a defined therapy?



Can we monitor the dynamics of genomic alterations in lymphoma during therapy in order to predict therapy failure?



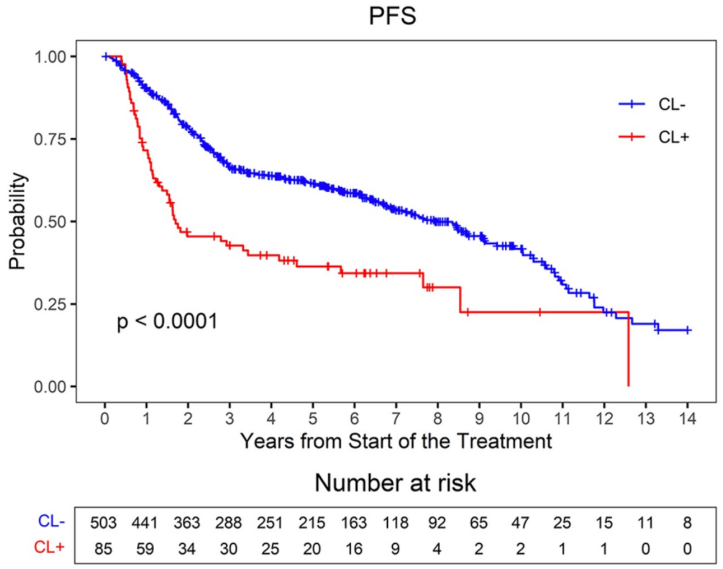
The "**tumor circulome**", defined as the subset of circulating components, is derived from cancer tissue and can be directly or indirectly used as a source of cancer biomarkers in liquid biopsies

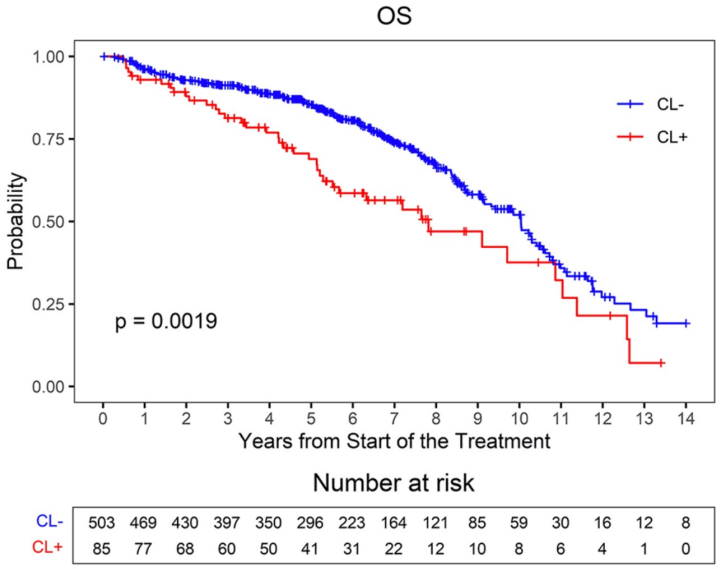


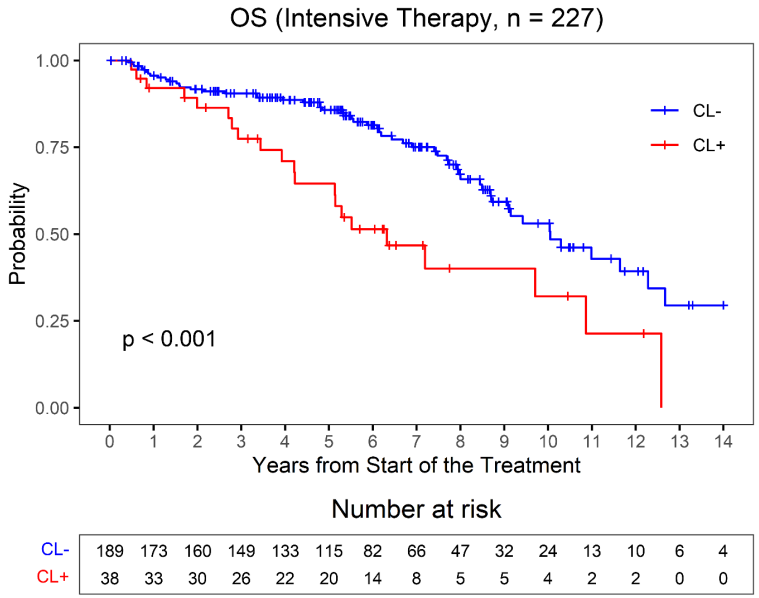
Journal of Hematology & Oncology (2025) 18:4

Impact of circulating lymphoma cells at diagnosis on outcomes in patients with newly diagnosed *de novo* diffuse large B-cell lymphoma

Sayan Mullick Chowdhury[1†], Subodh Bhatta[1†], Timothy J. Voorhees[1], Kaitlin Annunzio[1], David A. Bond[1], Yazeed Sawalha[1], Audrey Sigmund[1], Walter Hanel[1], Lalit Sehgal[1], Lapo Alinari[1], Robert Baiocchi[1], Kami Maddocks[1], Beth Christian[1], Dan Jones[2] and Narendranath Epperla[1,3,4*]







The presence of circulating lymphoma cells (CL) at diagnosis was associated with inferior response rates and survival compared to those without CL

Median OS was significantly inferior among patients who received first-line intensive-induction chemotherapies in the CL+ vs CL- group

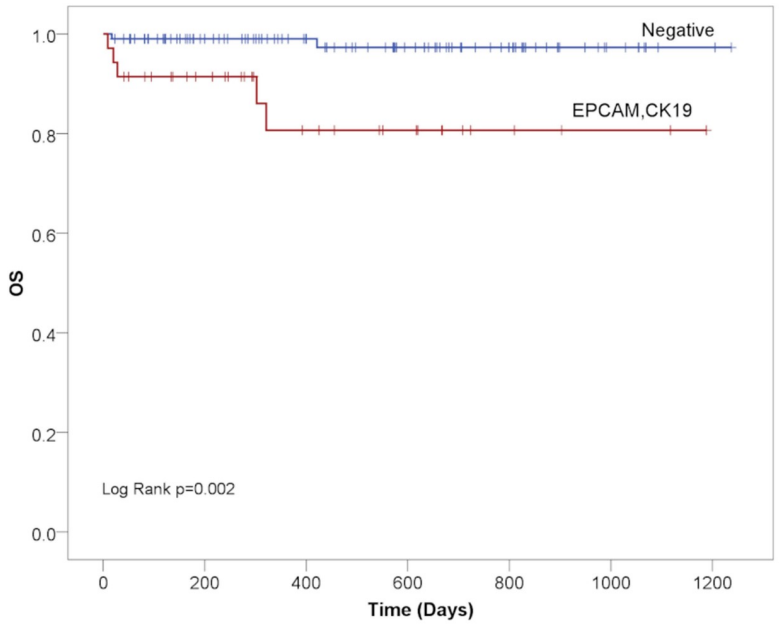
OncoTargets and Therapy 2022:15

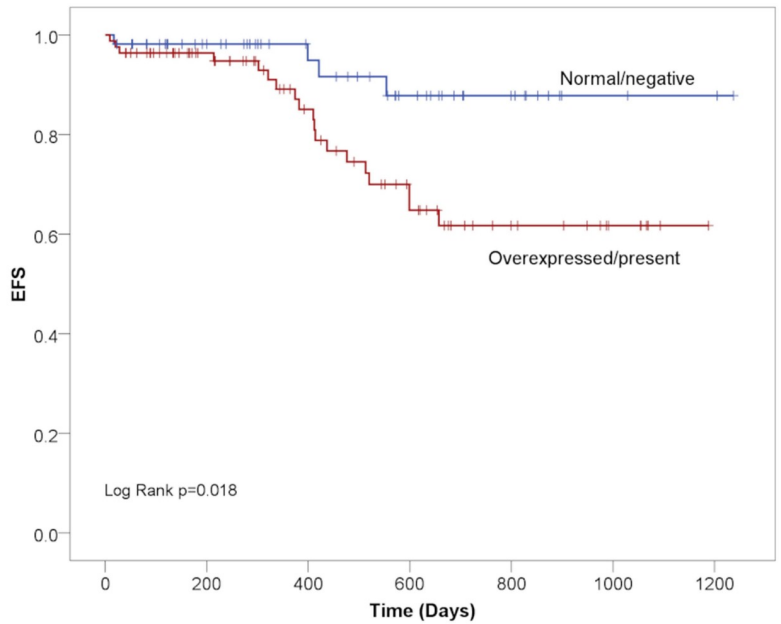
Overexpression of *BCL2*, *BCL6*, *VEGFR1* and *TWIST1* in Circulating Tumor Cells Derived from Patients with DLBCL Decreases Event-Free Survival

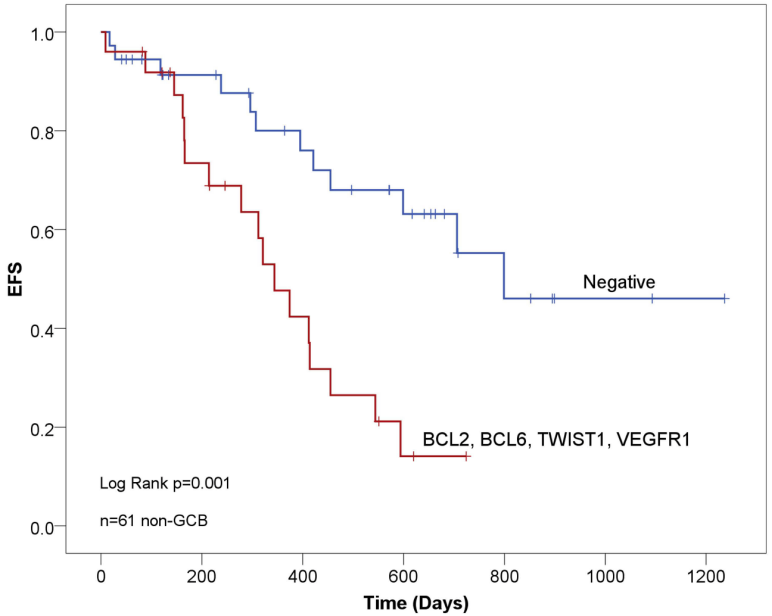
Rafael Cerón[1,2], Adolfo Martínez[2], Christian Ramos[3], Adrián De la Cruz[2], Anel García[2], Iveth Mendoza[2], Goujon Palmeros[2], Efreen Horacio Montaño Figueroa[3], Juan Navarrete[4], Silvia Jiménez-Morales[5], Carlos Martinez-Murillo[3], Irma Olarte[2]

Patients with the presence of EpCAM+ or CK19+ CTCs presented worse OS and EFS

The overexpression of the BCL2, BCL6, TWIST1 and VEGFR1 genes confers a poor EFS



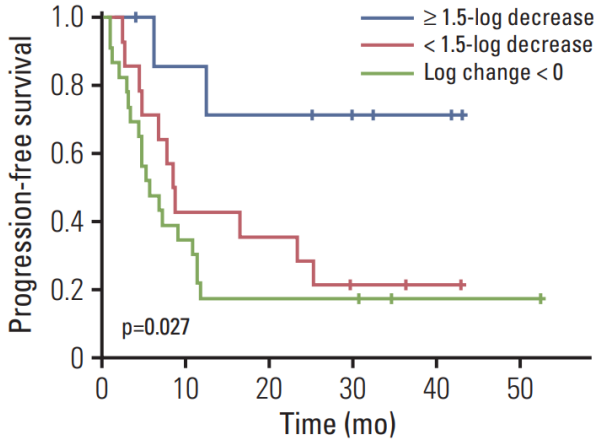


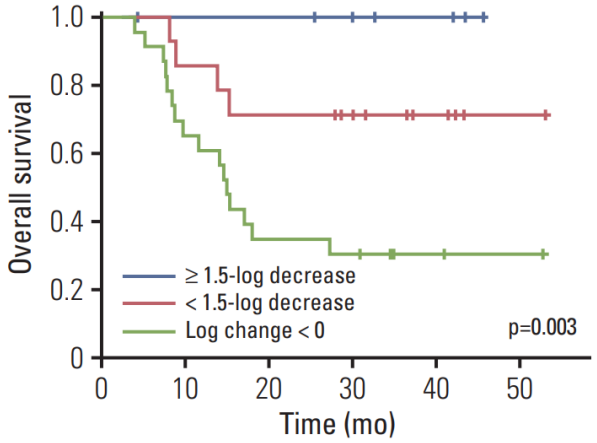


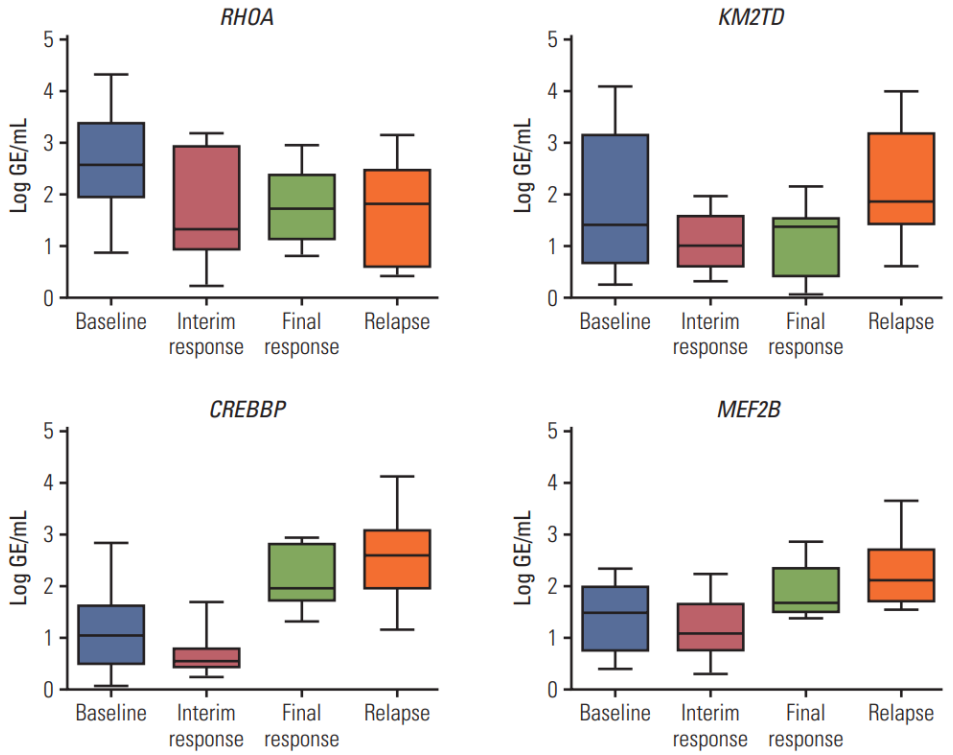
Cancer Res Treat. 2023;55(1):291-303

Circulating Tumor DNA–Based Genotyping and Monitoring for Predicting Disease Relapses of Patients with Peripheral T-Cell Lymphomas

Seok Jin Kim[1,2], Yeon Jeong Kim[3], Sang Eun Yoon[1], Kyung Ju Ryu[2], Bon Park[2], Donghyun Park[4], Duck Cho[5], Hyun-Young Kim[5], Junhun Cho[6], Young Hyeh Ko[6], Woong-Yang Park[2,3], Won Seog Kim[1,2]







Patients with a decrease in mutation volume at the end of treatment showed better progression-free and overall survival

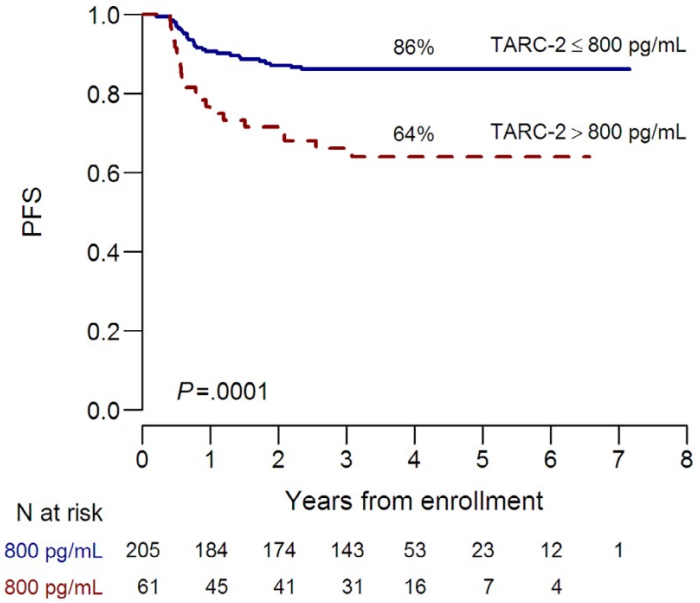
ctDNA mutation profiles showed increased mutation volumes in KMT2D, CREBBP, and MEF2B at the time of relapse.

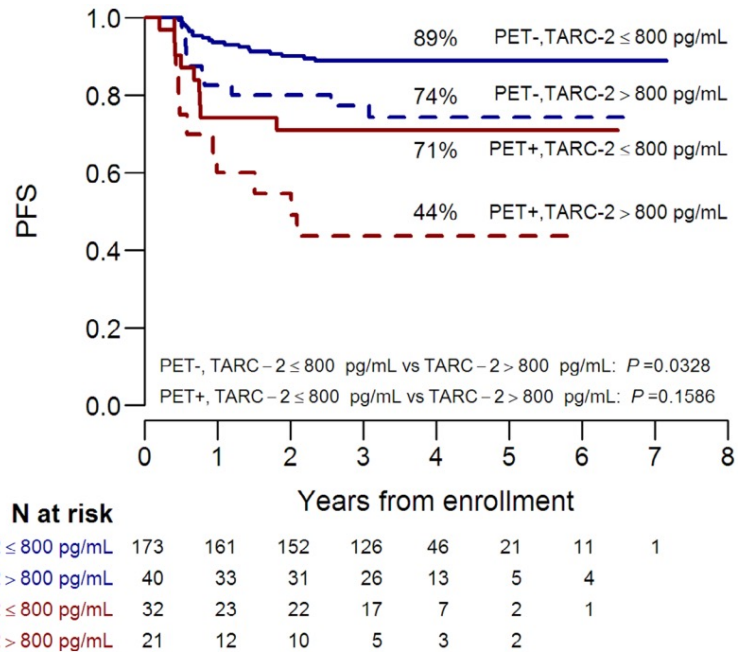
Hematological Oncology. 2020;38:501–508.

Early serum TARC reduction predicts prognosis in advanced-stage Hodgkin lymphoma patients treated with a PET-adapted strategy

Simonetta Viviani[1] | Arabella Mazzocchi[2] | Chiara Pavoni[3] | Francesca Taverna[2] | Andrea Rossi[3] | Caterina Patti[4] | Alessandra Romano[5] | Livio Trentin[6] | Roberto Sorasio[7] | Anna Guidetti[1] | Daniela Gottardi[8] | Corrado Tarella[9,10] | Michele Cimminiello[11] | Roberta Zanotti[12] | Lucia Farina[1] | Andrés José Maria Ferreri[13] | Marina Galbiati[2] | Paolo Corradini[1] | Alessandro Massimo Gianni[1] | Andrea Gallamini[7] | Alessandro Rambaldi[3,14]

TARC serum levels above the cut-off value of 800 pg/mL after completion of the second ABVD cycle (TARC-2) were associated with a high probability of treatment failure, even within the subgroup of patients with negative PET-2 results

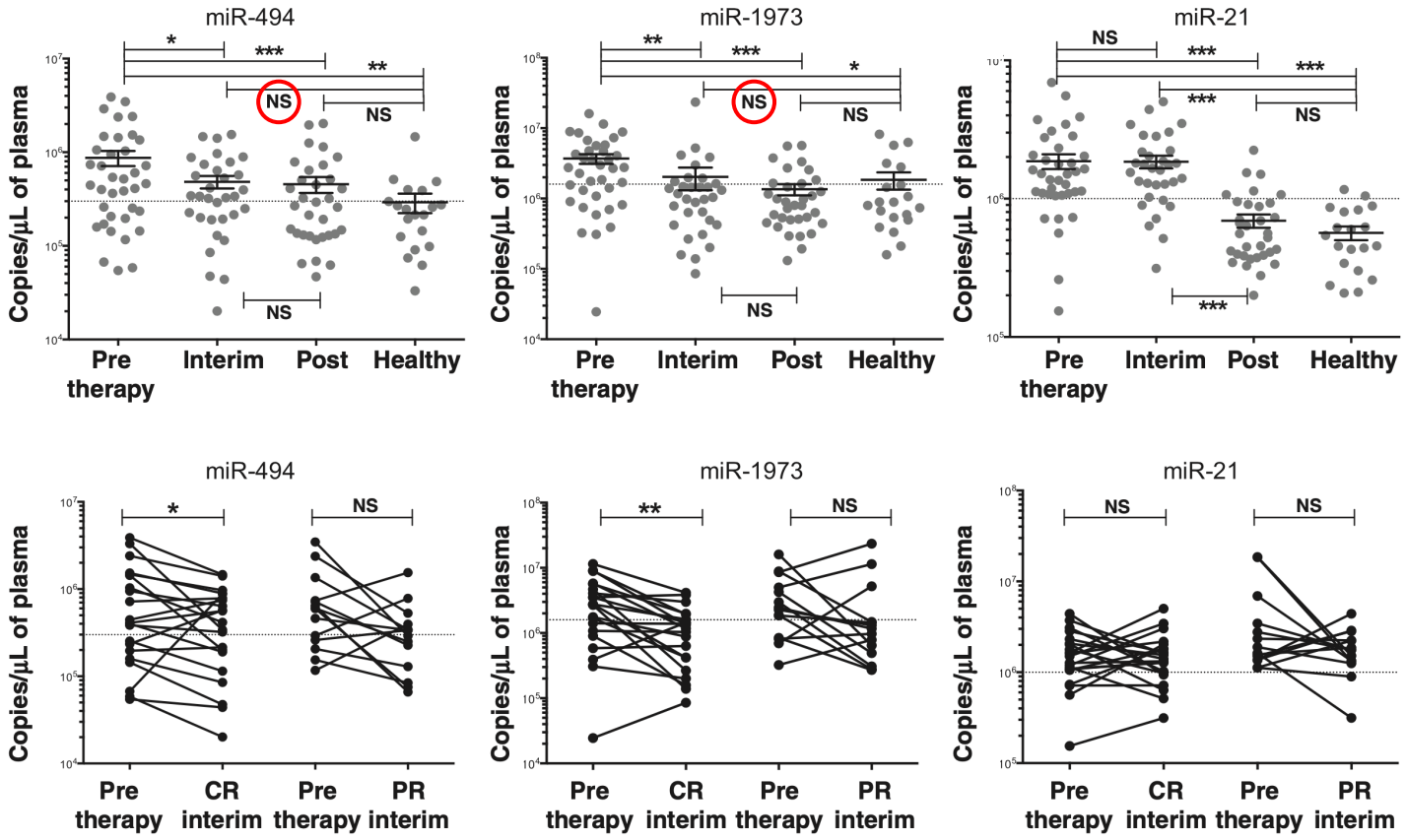




Clin Cancer Res; 20(1) January 1, 2014

Plasma MicroRNA Are Disease Response Biomarkers in Classical Hodgkin Lymphoma

Kimberley Jones[1,2], Jamie P. Nourse[1], Colm Keane[1,3,4], Atul Bhatnagar[1], and Maher K. Gandhi[1,2,3]



In patients who achieved complete remission by 6 months post-therapy, both miR494 and miR-1973 drop to levels equivalent with healthy controls by the interim time point.

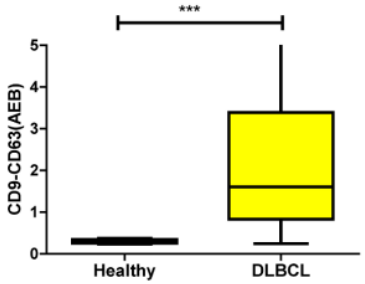
miR-21 interim therapy levels remain equivalent to pre-therapy and elevated compared with healthy controls, dropping to normal levels by 6 months post-therapy

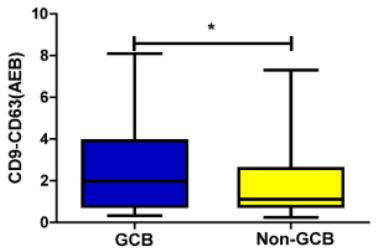
ONCOIMMUNOLOGY
2021, VOL. 10, NO. 1, e1995166

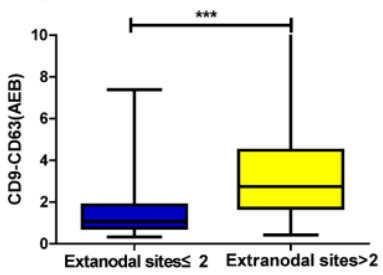
Universal extracellular vesicles and PD-L1+ extracellular vesicles detected by single molecule array technology as circulating biomarkers for diffuse large B cell lymphoma

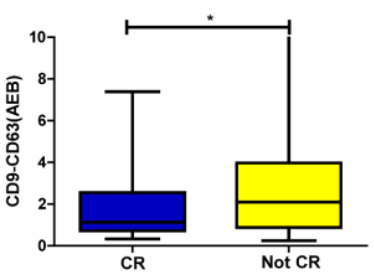
Ji-Wei Li[a,b,c,*], Di Shi[a,b,c,*], Xiao-Chun Wan[a,b,c], Jue Hu[a,b,c], Yi-Fan Su[a,b,c], Yu-Peng Zeng[a,b,c], Zi-Juan Hu[a,b,c], Bao-Hua Yu[a,b,c], Qun-Ling Zhang[b,d], Ping Wei[a,b,c], and Xiao-Yan Zhou[a,b,c]

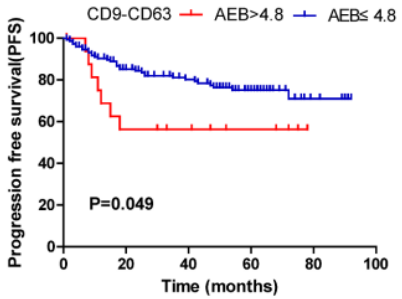
Elevated total and PD-L1+ EVs were
- abundant in the plasma of DLBCL patients
- associated with specific clinical features
- prognostic factors for both progression-free and overall survival

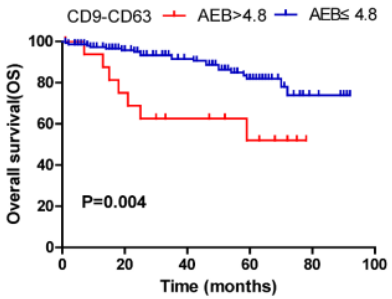


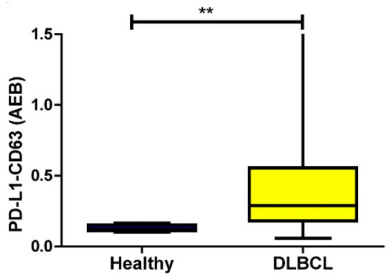


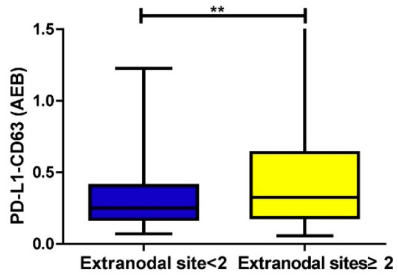


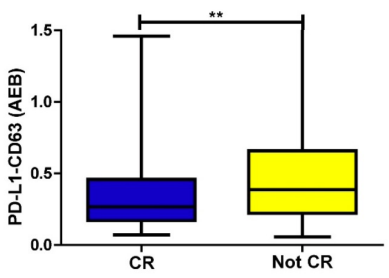


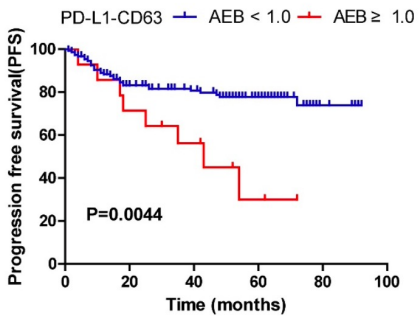


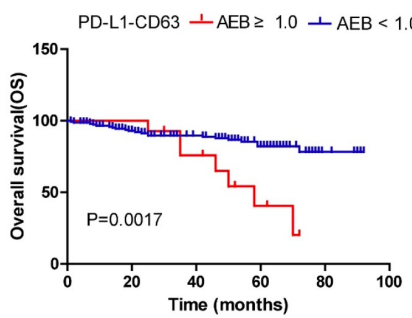








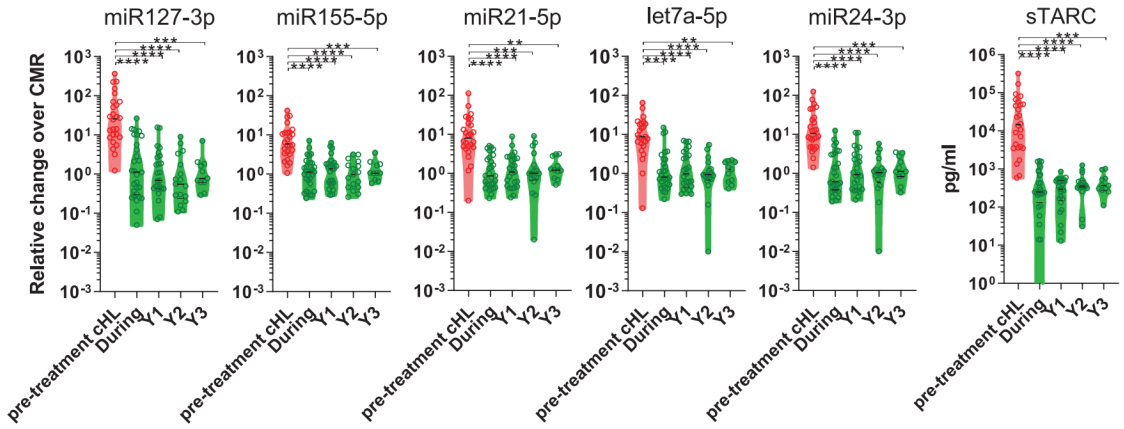


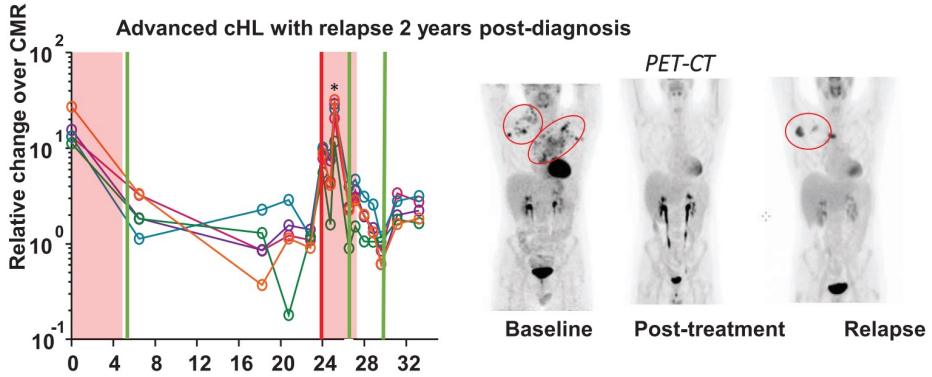


J Extracell Vesicles. 2021;10:e12121.

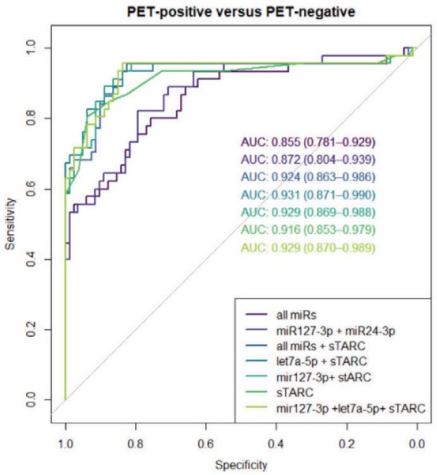
Extracellular vesicle miRNA predict FDG-PET status in patients with classical Hodgkin Lymphoma

Esther E. E. Drees[1] | Margaretha G. M. Roemer[1] | Nils J. Groenewegen[1,2] | Jennifer Perez-Boza[1] | Monique A. J. van Eijndhoven[1] | Leah I. Prins[1] | Sandra A. W. M. Verkuijlen[1] | Xuan-Mai Tran[1] | Julia Driessen[3] | G. J. C. Zwezerijnen[4] | Phylicia Stathi[1] | Kevin Mol[1] | Joey J. J. P. Karregat[1] | Aikaterini Kalantidou[1] | Andrea Vallés-Martí[1] | T. J. Molenaar[1] | Ernesto Aparicio-Puerta[5] | Erik van Dijk[1] | Bauke Ylstra[1] | Catharina G. M. Groothuis-Oudshoorn[6] | Michael Hackenberg[2,5] | Daphne de Jong[1] | Josée M. Zijlstra[7] | D. Michiel Pegtel[1]





Biomarkers included	AUC (CI)	Optimization cut-off	Sens. (%)	Spec. (%)	NPV (%)	PPV (%)
5 miRNAs	0.855 (0.781 – 0.929)	Youden-index Closest top left	80.0 80.0	75.6 75.6	87.3 87.3	64.3 64.3
miR127-3p + miR24-3p	0.872 (0.804-0.939)	Youden-index Closest top left	82.2 82.2	79.3 79.3	89.0 89.0	68.5 68.5
5 miRNAs + sTARC	0.924 (0.863-0.986)	Youden-index Closest top left	95.5 95.5	82.5 82.5	97.1 97.1	75.0 75.0
let7a-5p + sTARC	0.931 (0.871 – 0.990)	Youden-index Closest top left	93.5 87.0	83.8 88.8	95.7 92.2	76.8 81.6
miR127-3p + sTARC	0.929 (0.869 – 0.988)	Youden-index Closest top left	93.5 89.1	85.0 88.8	95.8 93.4	78.2 82.0
sTARC	0.916 (0.853 – 0.979)	Youden-index Closest top left	80.4 84.8	93.8 87.5	89.3 90.9	88.1 79.6
miR127-3p + let7a-5p+ sTARC	0.929 (0.870 – 0.989)	Youden-index Closest top left	95.6 93.5	83.8 85.0	97.1 95.8	77.2 78.2



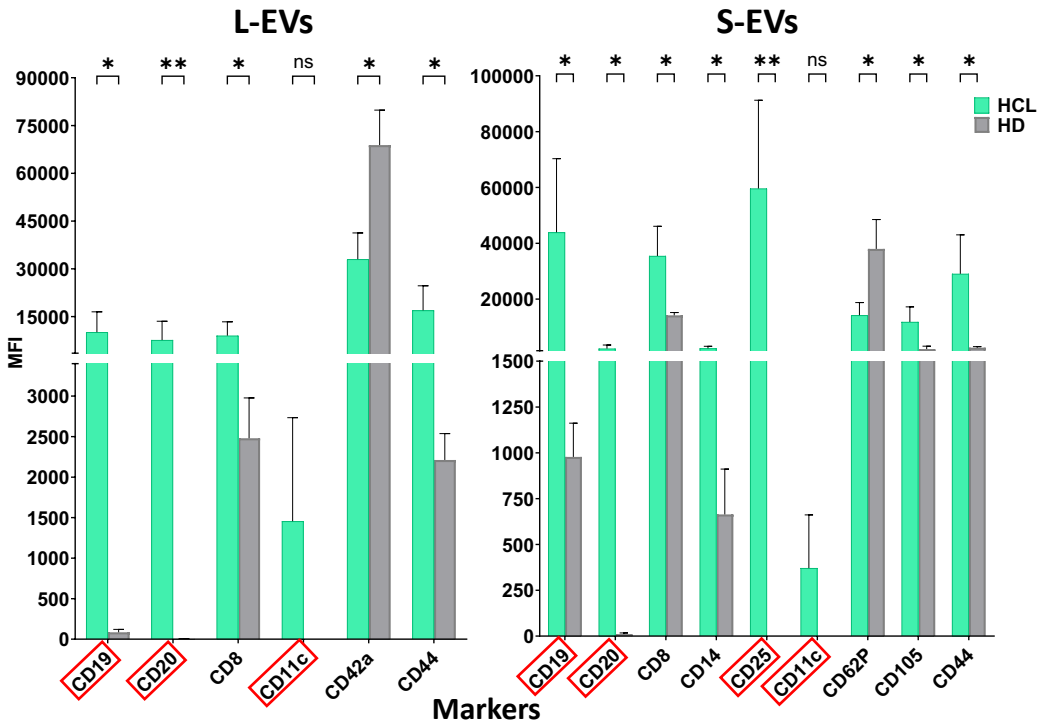
EV-miRNA levels correlate with the presence of PET detectable disease lesions during treatment.

EV-miRNAs of complete responders decrease early during treatment and remain stable in post-treatment follow up unless a relapse occurs.

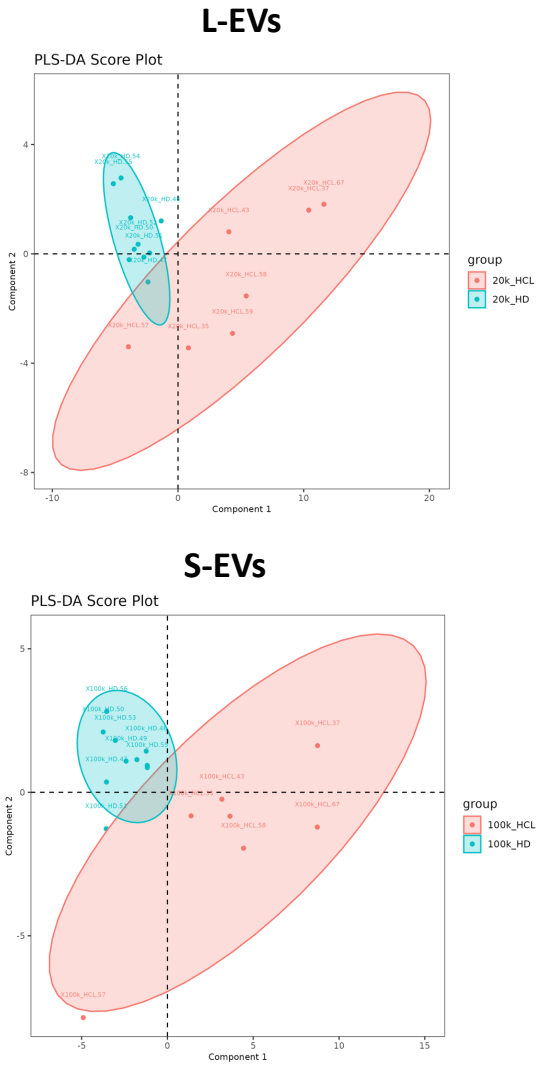
Combining EV-miR-127-3p and/or EV-let7a-5p levels with serum TARC increases the accuracy for predicting PET-status.

EXTRACELLULAR VESICLES IN HAIRY CELL LEUKEMA

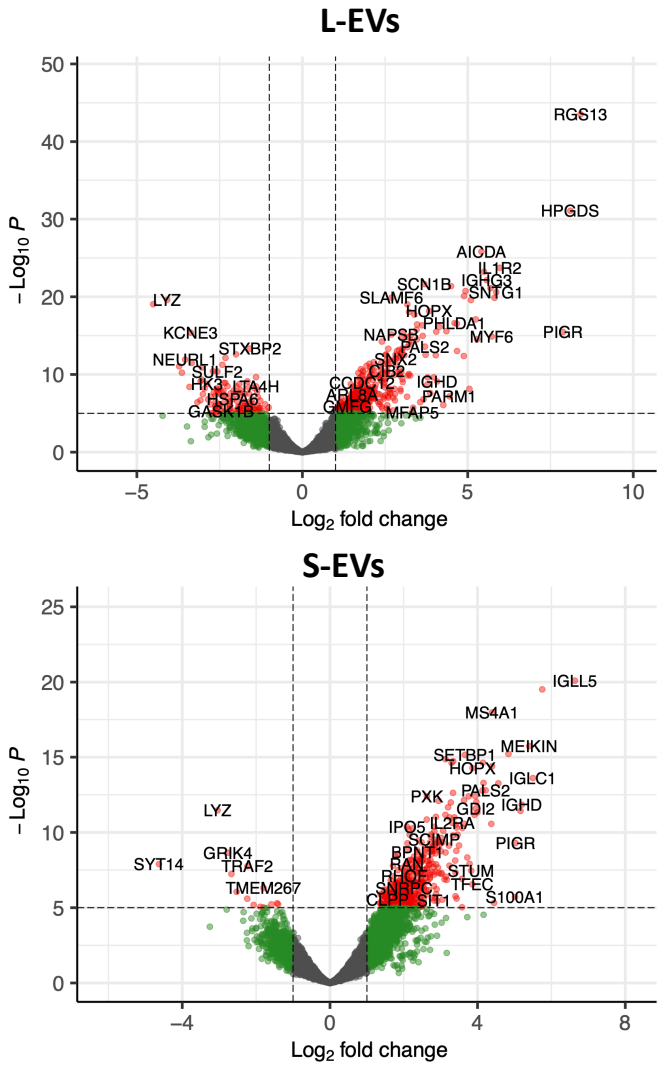
Flow cytometric analysis of EV surface signature



Lipidomic analysis



mRNA sequencing analysis



(Unpublished data)

CLINICAL IMPLICATIONS:

- **Robust validation** through expanded patient cohorts, considering potential variations across lymphoma subtypes and stages.
- The exact contribution and clinical utility of **integrative approaches** (proteomics, transcriptomics, and epigenomics).
- Exploration of **optimal timing, frequency, and specific clinical contexts** for its application.
- Understanding how liquid biopsy data can effectively inform treatment decisions **compared to traditional imaging modalities**.

FUTURE PERSPECTIVES:

- **Tumor-educated platelets**
- **cfDNA fragmentation patterns**: ctDNA fragment lengths of lymphoma patients may vary in each individual with a correlation with disease stage. Moreover, the fragmentation patterns were able to predict outcomes in DLBCL (Meriranta *et al.* Blood 2022).
- **Epigenetic features**: abnormal tumor-specific DNA methylation patterns detected in cfDNA are associated with poor outcomes in DLBCL (Wedge *et al.* Am J Hematol 2017).

CONCLUSIONS

Liquid biopsy techniques have shown significant promise as a **non-invasive alternative** to traditional tissue biopsies for lymphoma **diagnosis, therapy monitoring, relapse detection, and outcome prediction**.

The **real-time monitoring** of treatment and **timely adjustments** to treatment plans based on identifiable characteristics will provide enhanced benefits to patients.

The **combination** of liquid biopsy approaches **with PET/CT scans** at interim timepoints may further improve outcome prediction and treatment tailoring during therapy.

In-depth research examining the tumor circulome will also help clarify the **pathogenesis of lymphomas** and promote the **development of new therapeutic strategies**.

ACKNOLEDGEMENTS

Ghazal Narimanfar
Dorian Forte
Martina Barone
Lucia Catani

Simone Tiberi

Roberto Maria Pellegrino
Carla Emiliani

Giulia Gabrielli
Lisa Argnani
Alessandro Broccoli

Pier Luigi Zinzani

POLICLINICO DI SANT'ORSOLA
SERVIZIO SANITARIO REGIONALE
EMILIA-ROMAGNA
Azienda Ospedaliero - Universitaria di Bologna

AIL Bologna
ASSOCIAZIONE ITALIANA
CONTRO LEUCEMIE
LINFOMI E MIELOMA

ALMA MATER STUDIORUM
UNIVERSITÀ DI BOLOGNA