

ISEV WORKSHOP – Blood EVs

Educational Day

Wednesday 31.8.2022

“Getting blood-derived EVs into the clinics”

Moderator Rienk Nieuwland, Academic Medical Centre Amsterdam & Pia Siljander, University of Helsinki

9:00-9:20 General welcome to the Educational day & Blood EV workshop (Saara Laitinen, Finnish Red Cross Blood Service & Pia Siljander, University of Helsinki)

9:20-9:50 Introduction to blood EVs (Rienk Nieuwland, Academic Medical Centre Amsterdam)

9:50-10:20 Blood as a source of EVs (Edit Buzas, Semmelweis University)

10:20- 10:50 Detection methods for EVs in blood (Ken Witwer, John Hopkins University)

10:50-11:20 **BREAK**

11:20-11:50 Pre-analytical considerations for blood collection and processing in translational and clinical EV research (Fabrice Lucien-Matteoni, Mayo Clinic)

11:50- 12:20 A clinician’s perspective on blood EV biomarkers (Aleksandra Gasecka, Medical University of Warsaw & Academic Medical Centre Amsterdam)

12:20-13:05 Company presentations (Izon, ParticleMetrix, TimeGate Instruments, BioNavis, Carmine Therapeutics)

LUNCH 13-14:00

14.00-14:30 Reference values of blood EVs (Metka Lenassi, University of Ljubljana)

14:30-15:00 Approaching clinical feasibility with EV flow cytometry (Edwin van der Pol, Academic Medical Centre Amsterdam)

15:00- 15:20 Company presentations (Luminex, Sartorius)

15:20-15:50 **BREAK**

15:50-16:20 Heterogeneous platelet-EVs: theranostic opportunities? (Pia Siljander, University of Helsinki)

16:20-16:50 Post-isolation modifications of EVs for therapeutic purposes (Minh Le, National University of Singapore)

16:50-17:20 Summary and discussion (Rienk Nieuwland, Academic Medical Centre Amsterdam)

17:20-18:00 Results of the attendant survey & the blood EV analyses by participating technologies

19.00 Welcome reception at the Botanic gardens

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ISEV WORKSHOP ON BLOOD EVs
DAY 1 Program Blood EVs – matrix prerequisites & technologies & biomarkers

Thursday 1.9.2022

Moderators Pia Siljander (University of Helsinki) & Edit Buzas (Sемmelweis University) & Rienk Nieuwland (Academic Medical Centre Amsterdam) & Lorraine O’Driscoll (Trinity Collage)

9:00-9:10 Introduction to working in the workshops, Pia Siljander

9:10-9:25 Introduction to the topic: Blood as a source of EVs Part I (Edit Buzas & Rienk Nieuwland)

- 9:25 Comparison of collection tubes and processing intervals for extracellular vesicle analysis in blood (An Hendrix, Ghent University)
- 9:35 Influence of isolation and anticoagulant on analysis of plasma-derived EVs by imaging flow cytometry (Tobias Tertel, University Hospital Essen)
- 9:45 Characterization of platelet-derived extracellular vesicle subpopulations (Johanna Puutio, University of Helsinki)
- 9:55 Plasma EV ID: Fluorescence-based phenotyping of blood-borne extracellular vesicles (Danilo Mladenovic, Tallin University)
- 10:05-10:20 questions

5 minutes break

- 10:25 Label-free detection and characterization of single sub-micrometer particles in plasma of prostate cancer patients (Agustin Enciso-Martinez, Leiden University Medical Center)
- 10:35 Stratification of Breast cancer risk by Raman Spectroscopy characterization of Extracellular Vesicles (Lorena Signati, University of Milan)
- 10:45 Optimization of mutational analysis workflow using next-generation sequencing on plasma derived extracellular vesicles (Rebekka van Hoof, Flemish Institute for Technological Research)
- 10:55-11:10 questions

11:10-11:15 Break and organization into three roundtables

11:15-12:30 Roundtable discussions (60’+ 15’ wrap-up in writing by the moderators)

Topics will raise organically from the participants/ electronic platform or by the chairs, for example

- What do we need to consider regarding blood EV preanalytics, isolation, analytics, applications?
- What do we not understand yet?

Table 1 Moderators: Rienk Nieuwland/Pia Siljander

Table 2 Moderators: John Nolan/Edwin van der Pol

Table 3 Moderators: An Hendrix/Metka Lenassi

LUNCH 12:30-13:30

13:30-14:00 Highlights from the morning discussions by roundtable moderators (3 x 5 minutes & audience)

14:00-14:15 Introduction to the topic: Biomarker applications of the blood EV surface Part I (Lorraine O'Driscoll, Trinity College)

- 14:10 Blood EV protein corona: The opening of a new biomarker discovery landscape? (Annalisa Radeghieri, University of Brescia/CSGI, Florence)
- 14:20 Tumour-derived DNA is enriched as short fragments associated with extracellular vesicles (Daniel Hagey, Karolinska Institutet)
- 14:30 The HLA-I immunopeptidome of blood EV -detection of clinically relevant peptides (Simon Powis, University of St. Andrews)
- 14:40-14:55 Questions

14:55-15:25 COFFEE

Biomarker applications of the blood EV surface Part II

15:25 Methods comparison for plasma-derived EVs isolation to perform proteomics analysis (Sandrine Reymond, University of Geneva)

15:35 Identification of B-cell signatures in plasma EVs from pediatric BCP-ALL patients (Marit Inngjerdingen, Oslo University Hospital)

15:45 Circulating extracellular vesicles provide valuable protein biomarkers in metastatic breast cancer (Lorena Martin-Jaular, Institut Curie)

15.45-16:00 questions

10 minutes break & moving to roundtables

16:10-17:25 Roundtable discussions (60'+ 15' wrap-up in writing by the moderators)

Topics will raise organically from the participants/ electronic platform or by the chairs

Table 1 Moderators Lorraine O'Driscoll/Natasha Zavroni

Table 2 Moderators Annalisa Radeghieri/Mari Palviainen

Table3 Moderators Fabrice Lucien-Matteoni/ Lorena Martin-Jaular

5 minutes break & moving to main hall

17:30-18:00 Highlights shared from the afternoon discussions (3 x 5 min & audience)

ISEV WORKSHOP on BLOOD EVs
DAY 2 Program Blood EVs as Therapeutics
Friday 2.9.2022

Moderators Minh Le & Saara Laitinen

9.00-9.30 Introduction to topics: Blood as a therapeutic EV source and in delivery of EV-therapy

Standing on the shoulders of giants, decades of transfusion & blood cell therapy (Saara Laitinen, Finnish Red Cross Blood Service)

Potential therapeutic applications of blood cell derived EVs (Minh Le, National University of Singapore)

- 9:30 Using plasma derived EVs as drug delivery vehicles (Maria Chiara Ferri, University of Genova)
- 9:40 Delivery of therapeutic mRNA via extracellular vesicles to express soluble proteins (Muhammed Nawaz, University of Gothenburg)
- 9:50 Development of designed targeting peptides for EV (Sujaan Das, Crane Biosciences)
- 10:00 Questions & discussion
- 10:15 Developing drug delivery RBCEVs produced from preloaded RBC (Diana Piedrahita, Erytech Pharma)
- 10:25 Characterising safety and efficacy of RBC-EV as therapeutic delivery vehicles for retinal degenerations (Rakshanya Sekar, Australia National University)
- 10:35 Using RBC-EVs as drug delivery particles (Don Haut, Carmine Therapeutics)
- 10:45- 11:00 questions

11.00-11.15 Break and organization into three roundtables

Suggestions for roundtable discussion topics in addition to those which are raised organically

- Standing on the shoulders of giants, decades of transfusion knowledge
- Why and what we should consider?
- Benefits and disadvantages of using RBC-EVs as therapeutics
- Technologies for loading and targeting of RBC EVs
- Blood group and tissue type: challenge or opportunity?
- The effect of blood components on delivering EV
- Blood - friendly highway or hostile environment?
- The effect of processing and storage on EVs

11.15-13.00 Roundtable discussions

Roundtable 1 Red blood cell -derived nanobiotechnology

Moderators Minh Le & Kai Härkönen

Roundtable 2 Different blood cell derived EVs for therapy development

Moderators Saara Laitinen & Ulla Impola

Roundtable 3 Lessons learned from blood EVs: is there advantage?

Moderators Edith Buzas & Pia Siljander

LUNCH 13.00 – 14.00

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14.00-15.00 Wrap-up the Roundtables (Big hall)

15.00 Coffee

15.30-17:00 Summary of the Workshop Days 1 & 2

Moderators: Saara Laitinen & Pia Siljander & Rienk Nieuwland

Wrap up of the three days into deliverables

- Impact and considerations of technological choices on blood EV isolation, detection and analyses (data from blood EVs and sponsor analytics, survey, roundtable outcomes)
- Therapeutic use of blood cell -derived EVs

What should we have discussed and what next?