



CD Maps – Antigen Density Measurements of on Human Leukocytes

HCDM and CD nomenclature



Human Cell Differentiation Molecule:

- Independent, academic organisation

which runs HLDA

(Human Leucocyte Differentiation Antigen) Workshops and names and characterizes CD molecules.

- Nomenclature committee of the International Union of Immunological Societies (IUIS)

Presentation overview:

HCDM and expression data

Aims and scope

Approach

Expression Map - Selection of results

HLDA Workshops I-X (1982-2014)

	Workshop	CDs assigned
I.	Paris 1982	CD1-CDw15
II.	Boston 1984	CD16-CDw26
III.	Oxford 1987	CD27-CD45
IV.	Vienna 1989	CD46-CDw78
V.	Boston 1993	CD79-CDw109
VI.	Kobe 1996	CD110-CD166
VII.	Harrogate 2000	CD167-CD247
VIII.	Adelaide 2004	CD248-CD339
IX.	Barcelona 2009	CD340-CD364
X.	Wollongong 2014	CD365-CD371

Over the past 30 y, the data generated by the 10 Human Leukocyte Differentiation Antigens (HLDA) Workshops have led to the characterization and formal designation of more than 400 molecules. Engel P et al. J Immunol. 2015;195(10):4555-63

Why should companies be interested in the results of CDMaps

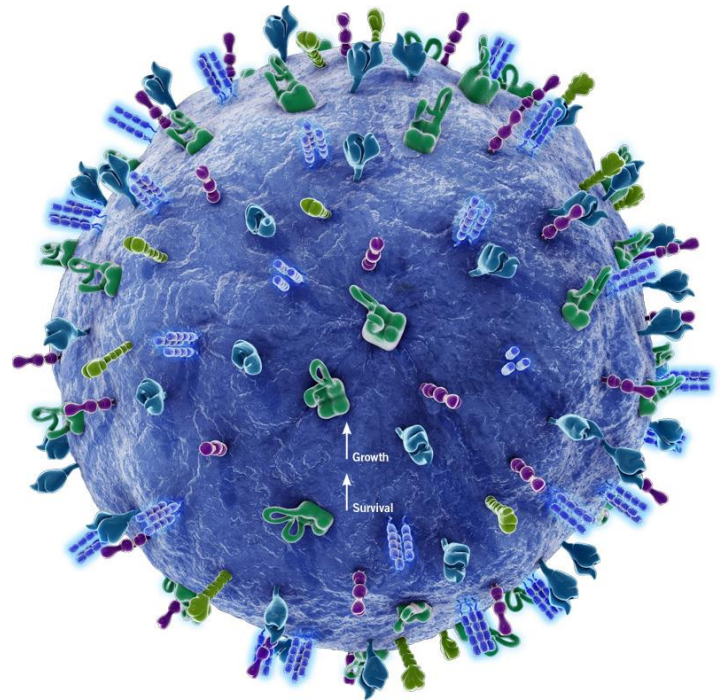
- Benchmarking reagents for expression profile (QC and catalog data)
- CD Mapping – clustering reagents to the benchmark reagents (validation by HCMD)
- Panel design – quantitative expression knowledge is crucial
- Assay conditions setup and optimization – is my staining as expected? (QC and validation)
- Analysis of the expression patterns with state of the art technology (catalog data)
- Complete information on the expression on rare subsets such as pDC (catalog data)

Aims and scope

Antigen expression Density Measurements of CD1-CD100 on Human Leukocytes

- Pilot: CD1-CD100
- Defined cells and subsets
- Quantify # molecules/cell

- Generate an interactive data base



<http://www.biooncology.com/>

Approach

4 laboratories

BCN → BD Fortessa

PRG → BD LSR II

ROT → BD Cantoll

SAL → BD Cantoll

Standardized flow
cytometer settings



Kalina, *Leukemia*, 2012

8-color flow cytometry

CD1-100 as PE

BD QuantiBRITE

Blood

B- and T-cell tube

Innate leukocyte tube

Tonsil

B-cell maturation tube

Thymus

T-cell differentiation
tube

FlowJo
(Excel)

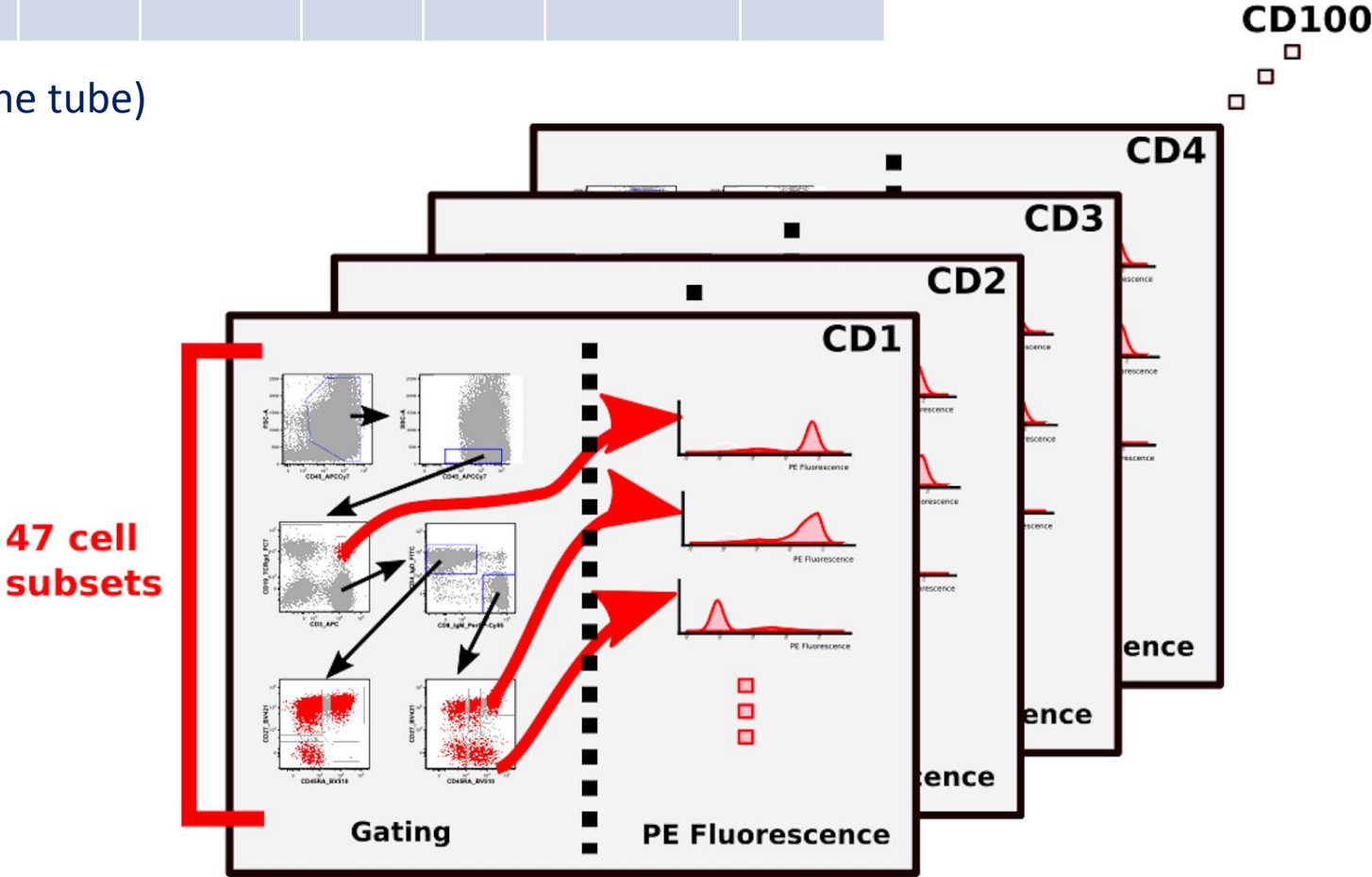
R – project

Shiny on
CESNET server

Blood: B- and T-cell tube

1	2	3	4	5	6	7	8
PE marker	CD45	CD3	TCRgd CD19	CD4 IgM	CD8 IgD	CD45RA	CD27

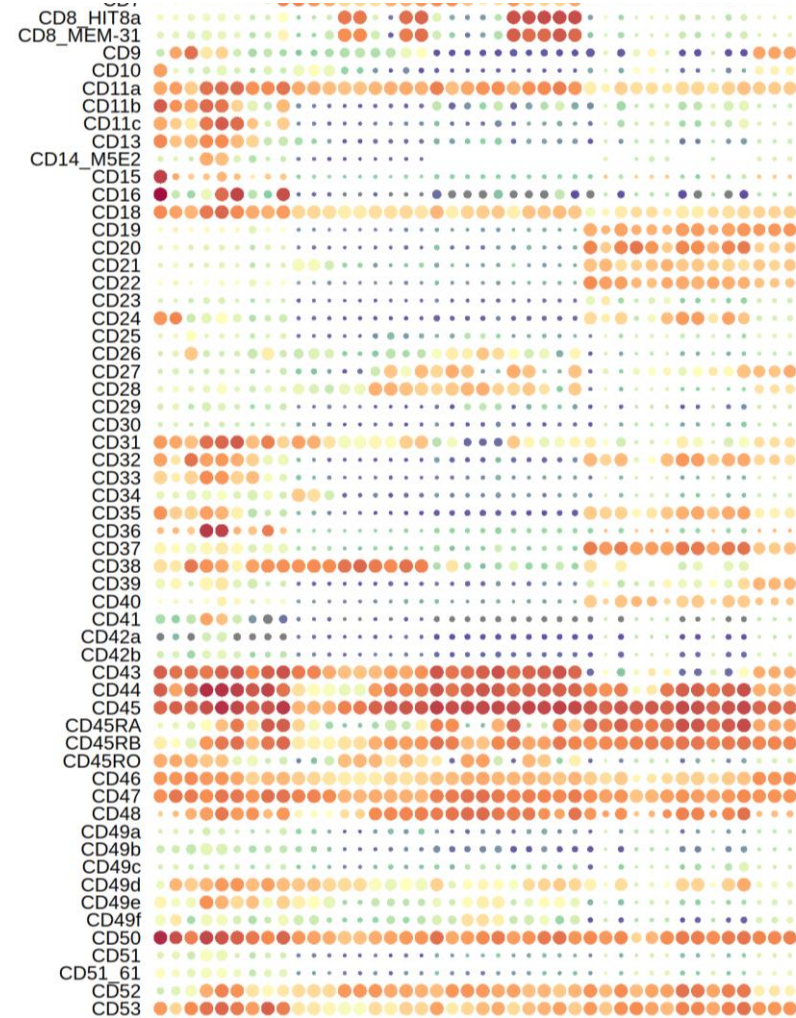
(example of one tube)





Snapshot map

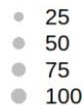
Web interface to the CDMaps dataset



Fluorescence
[log₁₀(Median ABC)]



Frequency of PE positive cells [%]



USER SELECTIONS:

select Ab panel

one of 11 measured variables

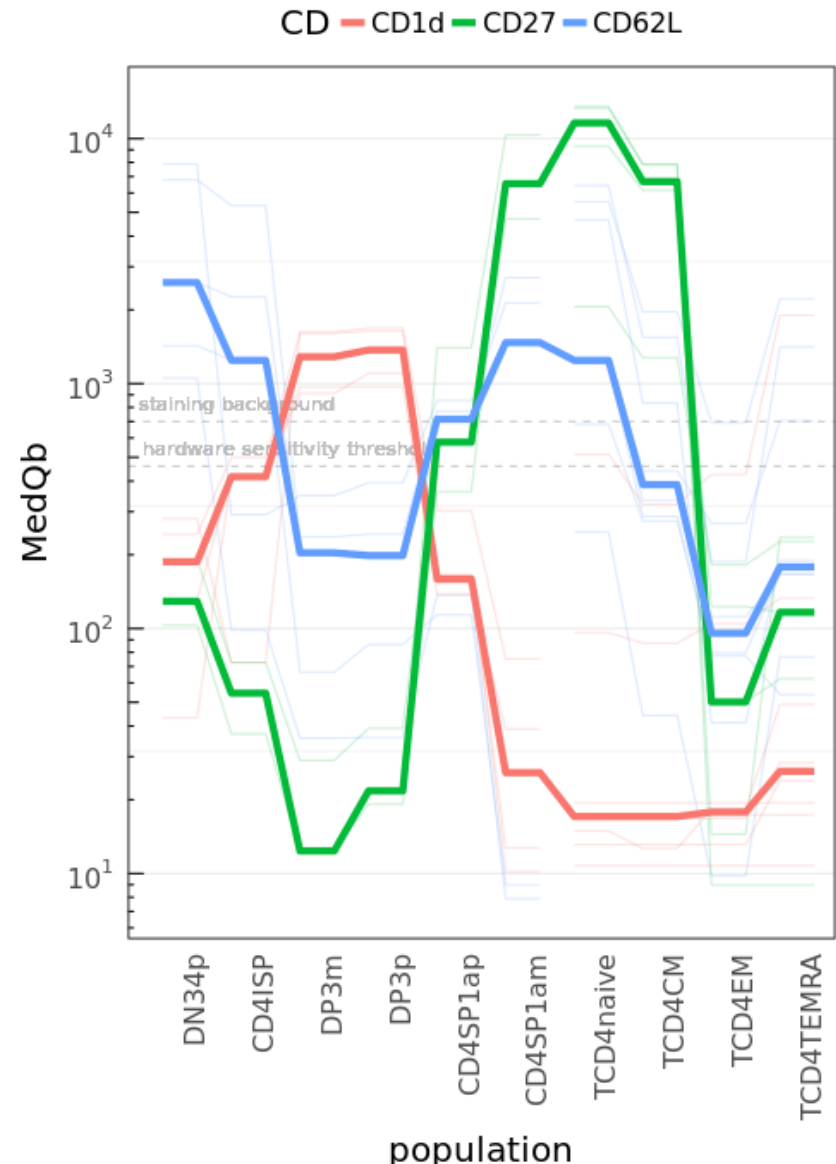
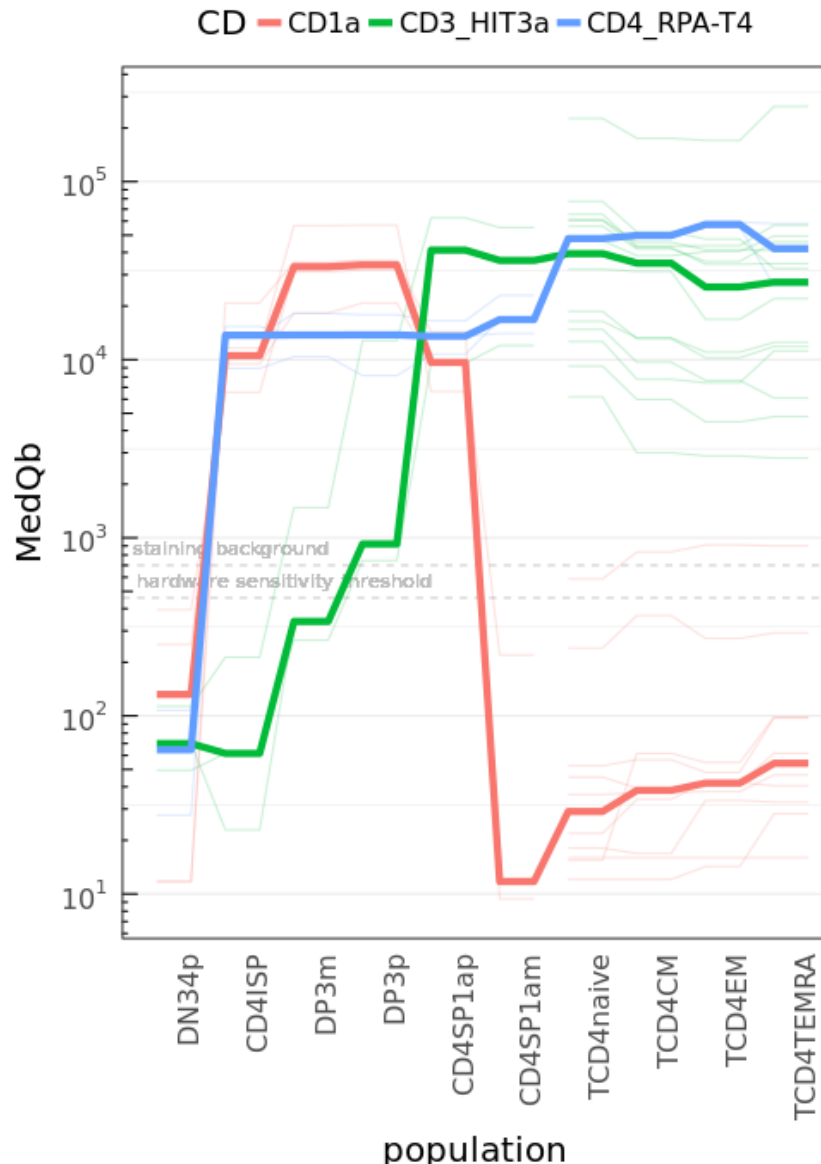
CD markers

cell subset

comment, bookmark, save

Ordererd Subsets

Thymus to blood development



What is different between subsets?

Here we compare one subset

Panel (population set)

2. Blood: B- and T-cells ▼

Variable to display

MedQb ▼

Single cell population

TCD8naive ▼

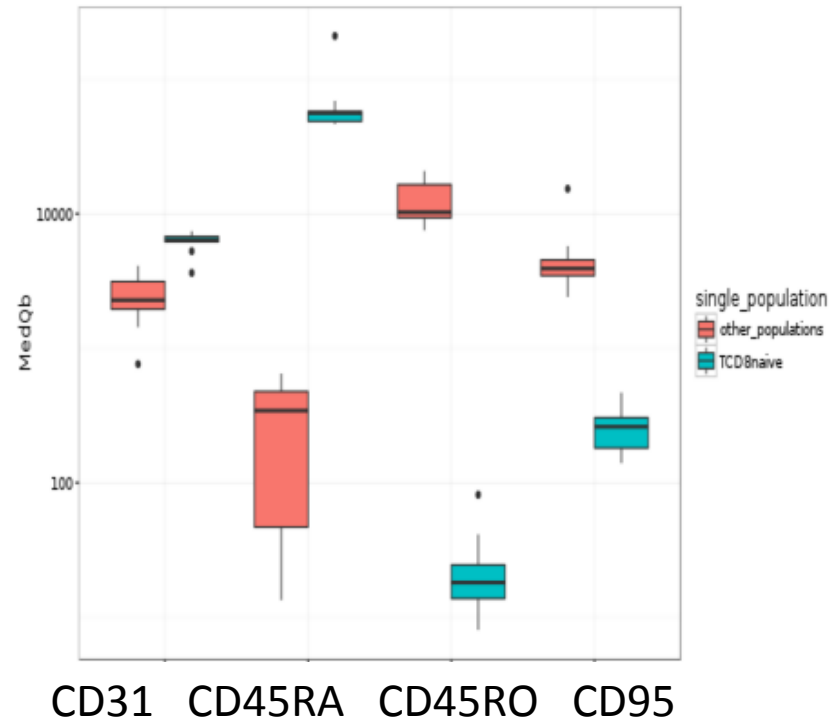
Group of cell populations
(do not use the one selected above)

TCD8CM

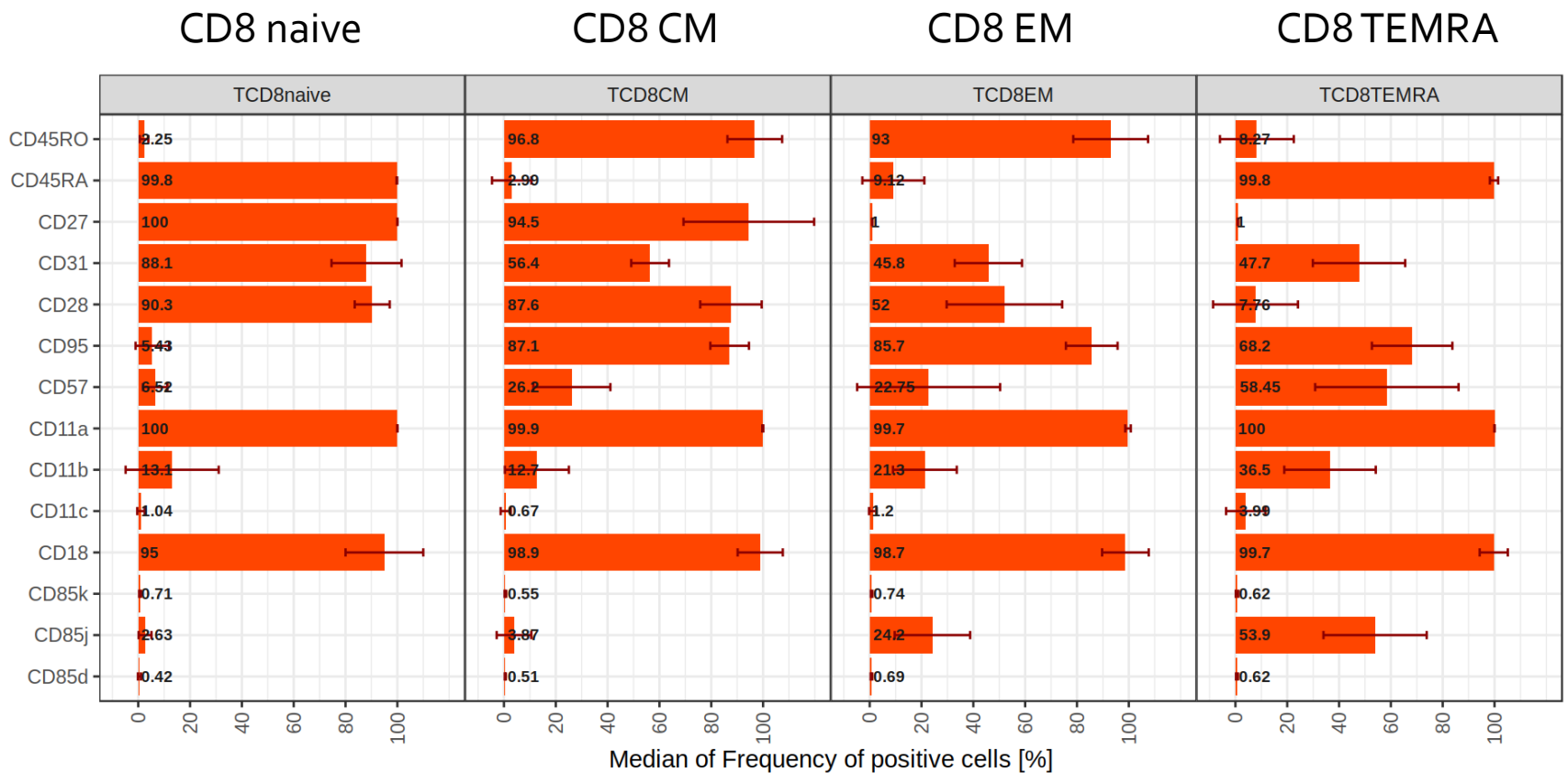
Set p-value threshold

0.001

To one or multiple

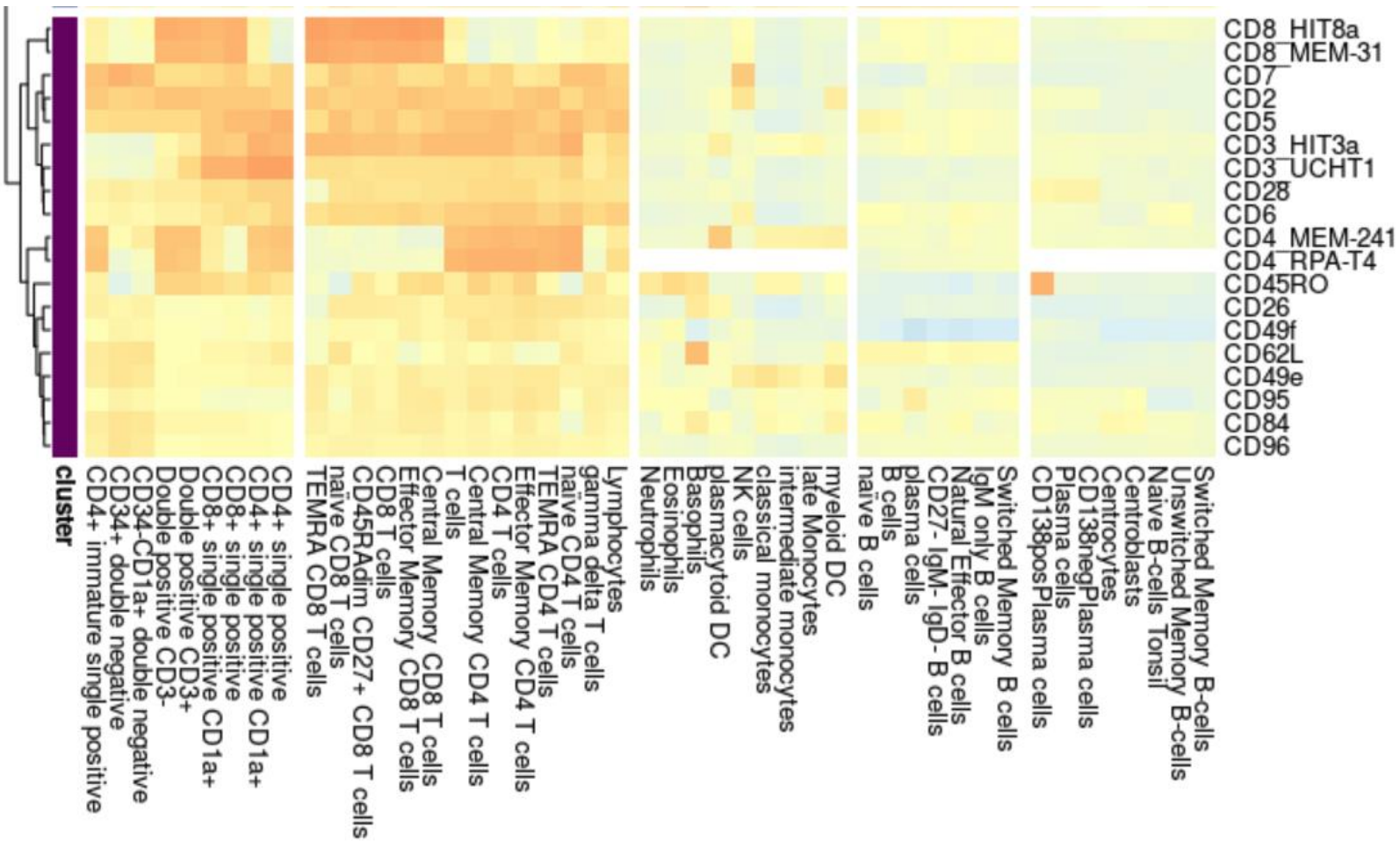


Markers per selected subsets



Clustering

Example – part of the clustering graph



Future goals (second round – 2019-2010)

- Complete the CD list.
- Add new CDs that will come from HLDA11.
- Establish clustering analysis as a validation tool

Volume of work (estimate):

- 12 donors, 550 CDs tested, 12 000 FCS files
- 5 man-month wetlab work,
- 4 man-month bioinformatics+webresource management,
- 5 man-month management, supervision, training, publication preparation

Acknowledgements



Pablo Engel
Marta Cuenca



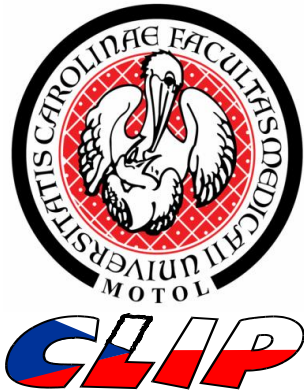
Menno van Zelm
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Bob Balderas



Kelly Lundsten



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Karel Fišer



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Miloslav Suchánek

