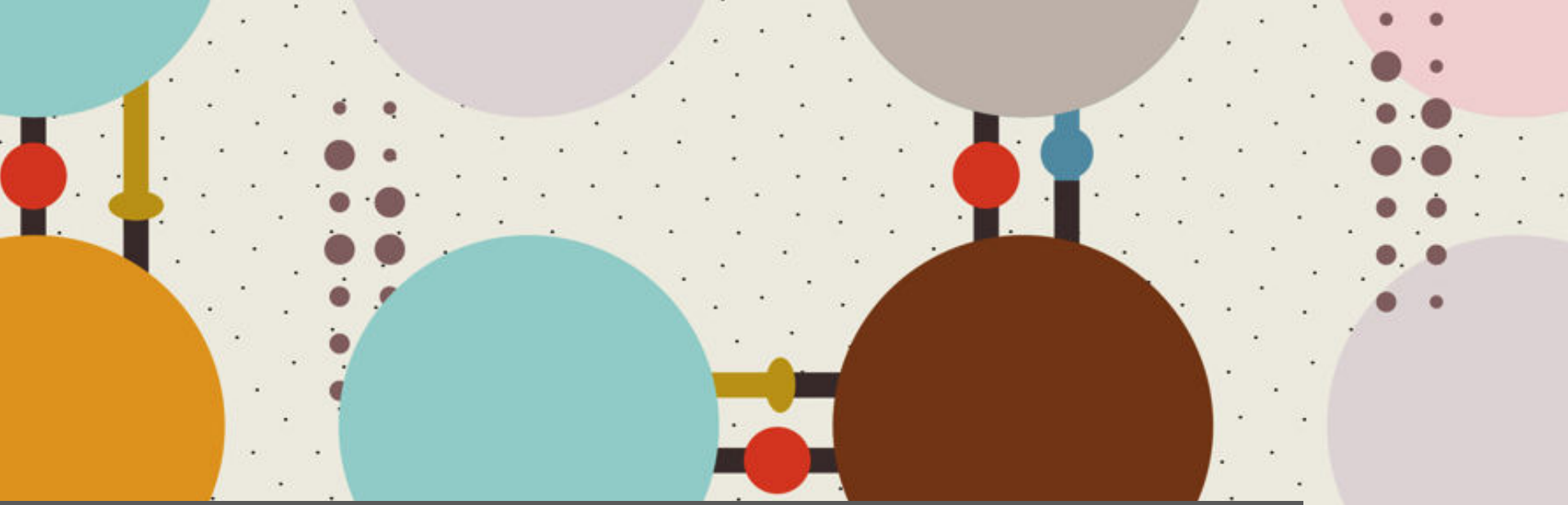


IO17 | Large Scale Bioinformatics for Immuno-Oncology

Prediction of tumor neoantigens

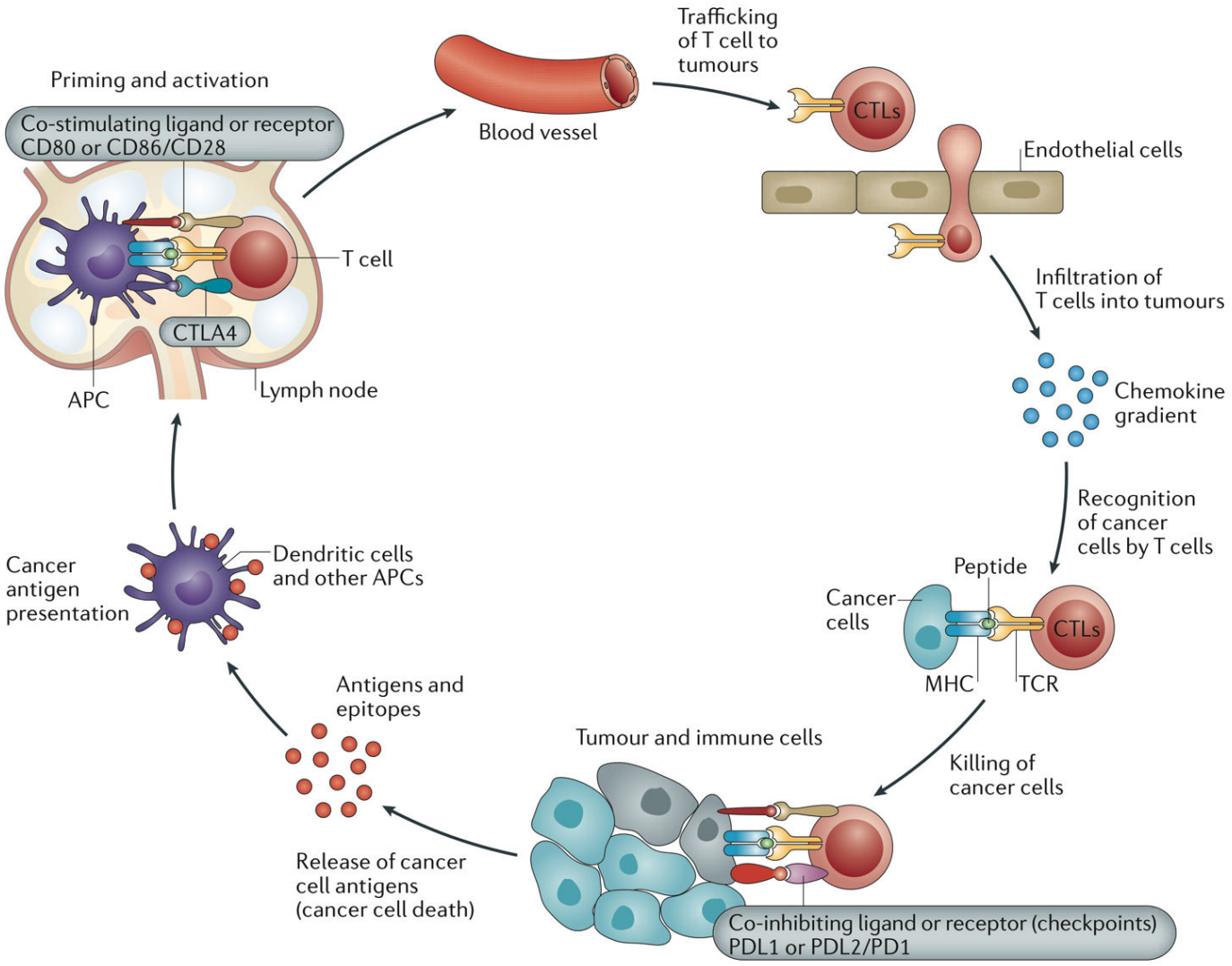
Francesca Finotello, Federica Eduati, and Pedro L. Fernandes

GTPB | The Gulbenkian Training Programme in Bioinformatics
Instituto Gulbenkian de Ciência, Oeiras, Portugal | Sept 19th-22nd, 2017

INSTITUTO GULBENKIAN DE CIÊNCIA

The anticancer immune response

Effective anticancer immune responses require a series of stepwise events:



H Hackl*, P Charoentong*, F Finotello* et al., Nature Reviews Genetics, 2016

Tumor antigens

Tumor antigens are small peptides bound to the MHC molecules of tumor cells that can be recognized as “non-self” by the immune system

Only two classes of tumor antigens elicit immune responses that are strictly tumor specific:

Neoantigens

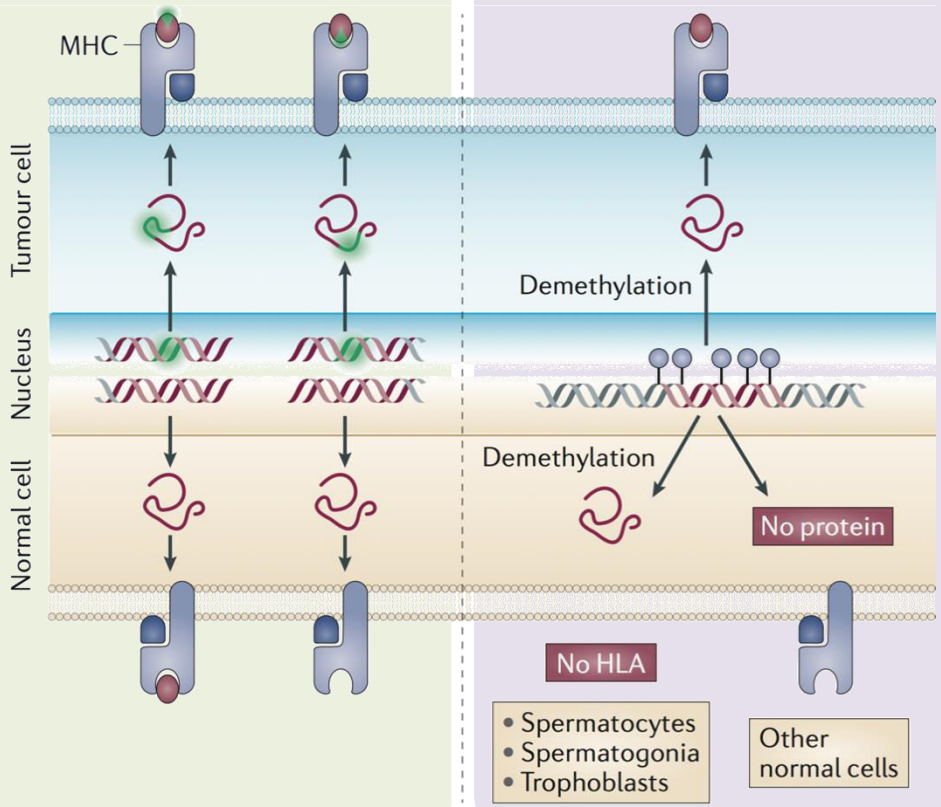
Modified peptides originating from mutations or DNA re-arrangements (or viral seq.)

Seen for the first time by the immune system → strong immune responses

Cancer-germline antigens (CGAs)

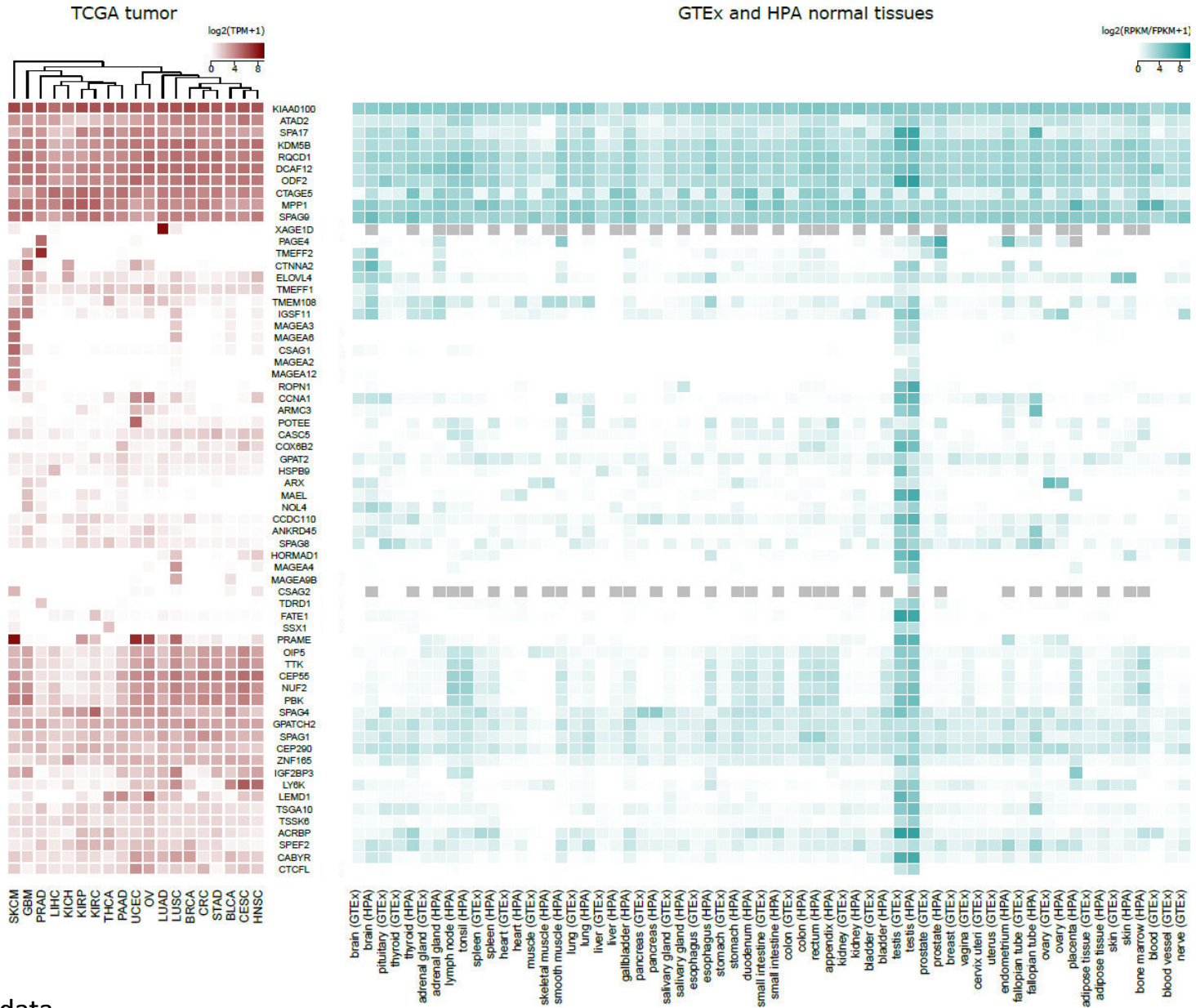
Genes selectively expressed only by germ-line cells (no MHC)

Aberrant expression in tumor due to demethylation



PG Coulie et al., Nature Reviews Cancer, 2014

Expression of cancer-germline antigens in solid tumors



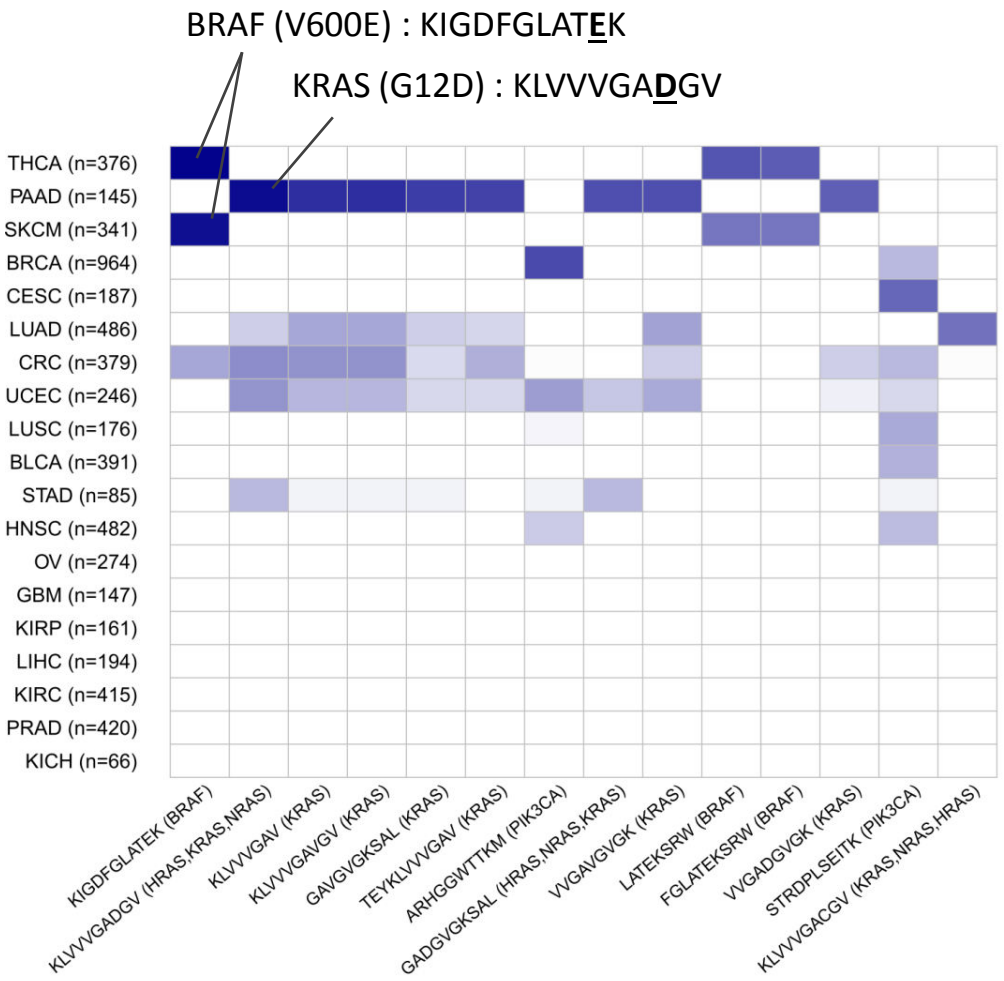
Unpublished data

Neoantigens in solid tumors

~1 petabyte of NGS data from
19 human cancers from TCGA
→ 933,954 neoantigens

7.0-10.6% of neoantigens
from driver mutations

Neoantigens shared in >15%
patients might be good
candidates for vaccination



Neoantigens are **diverse** and not shared because they arise mainly from **passenger** mutations

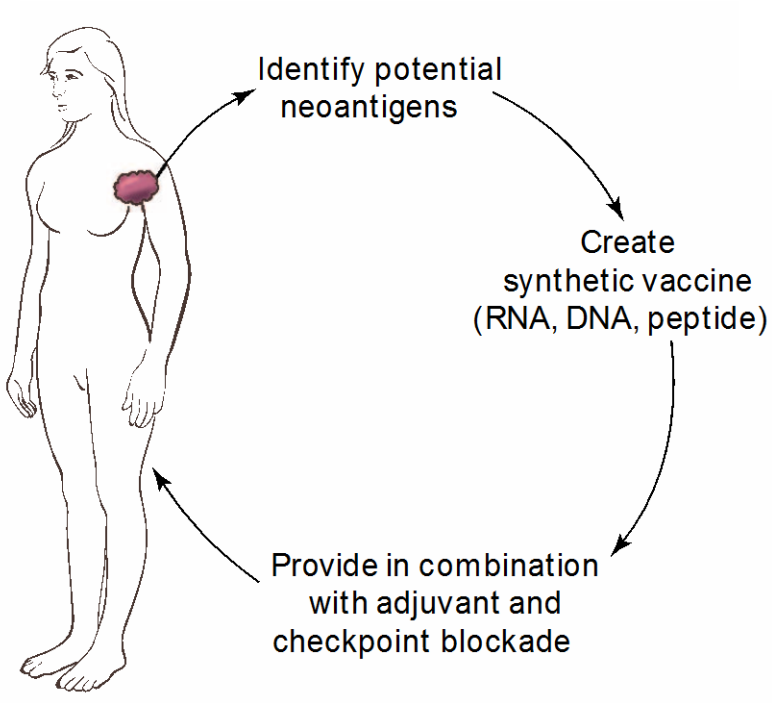
→ personalized therapy

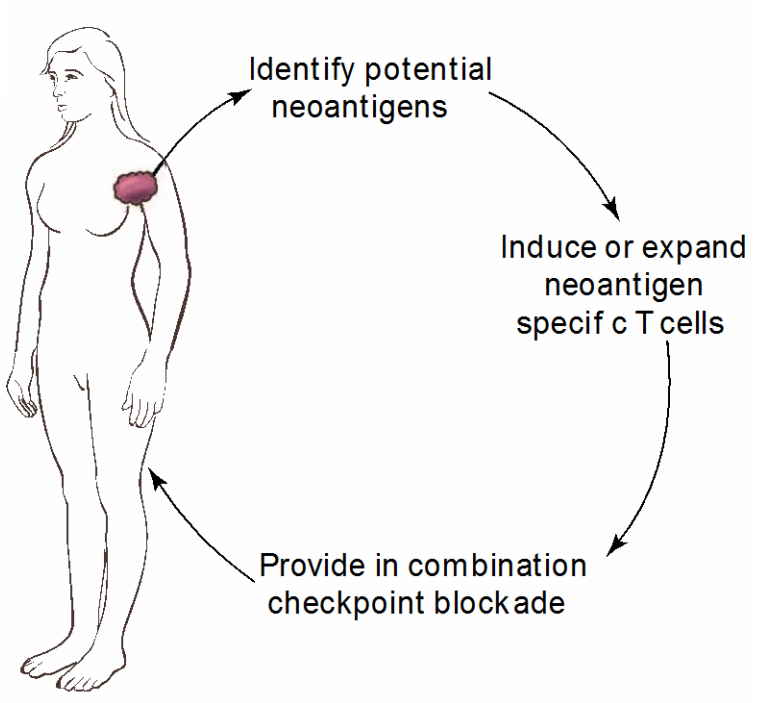
P Charoentong*, F Finotello *, M Angelova* et al., Cell Reports, 2017

Importance of tumor neoantigens for immunotherap

- Immunotherapy has higher success rate in cancer types with high number of non-synonymous mutations (e.g. melanoma or MSI cancers)
- Mutations → neoantigens → recognized as non-self by T cells
- Mutations/neoantigens proposed as biomarker for immunotherapy
- But no clear-cut separation and some cancer types with low mutational load also respond (e.g. clear renal cell carcinoma)

Targeting patient-specific neoantigens





- Synthetic vaccine: DNA minicassette electroporated into patient-derived DCs, RNA vaccine, injected peptide vaccine
- Neoantigen-specific T cells: from patient or healthy donors

TN Schumacher and RD Schreiber, Science,2015

Therapeutic potential of neoantigen-based therapie

LETTER

doi:10.1038/nature22991

An immunogenic personal neoantigen vaccine for patients with melanoma

Patrick A. Ott[1,2,3]*, Zhuting Hu[1]*, Derin B. Keskin[1,3,4], Sachet A. Shukla[1,4], Jing Sun[1], David J. Bozym[1], Wandi Zhang[1], Adrienne Luoma[5], Anita Giobbie-Hurder[6], Lauren Peter[7,8], Christina Chen[1], Oriol Olive[1], Todd A. Carter[4], Shuqiang Li[4], David J. Lieb[4], Thomas Eisenhaure[4], Evisa Gjini[9], Jonathan Stevens[10], William J. Lane[10], Indu Javeri[11], Kaliappanadar Nellaiappan[11], Andres M. Salazar[12], Heather Daley[1], Michael Seaman[7], Elizabeth I. Buchbinder[1,2,3], Charles H. Yoon[3,13], Maegan Harden[4], Niall Lennon[4], Stacey Gabriel[4], Scott J. Rodig[9,10], Dan H. Barouch[3,7,8], Jon C. Aster[3,10], Gad Getz[3,4,14], Kai Wucherpfennig[3,5], Donna Neuberg[6], Jerome Ritz[1,2,3], Eric S. Lander[3,4], Edward F. Fritsch[1,4]†, Nir Hacohen[3,4,15] & Catherine J. Wu[1,2,3,4]

Synthetic long peptides representing up to 20 patient-specific neoantigens

LETTER

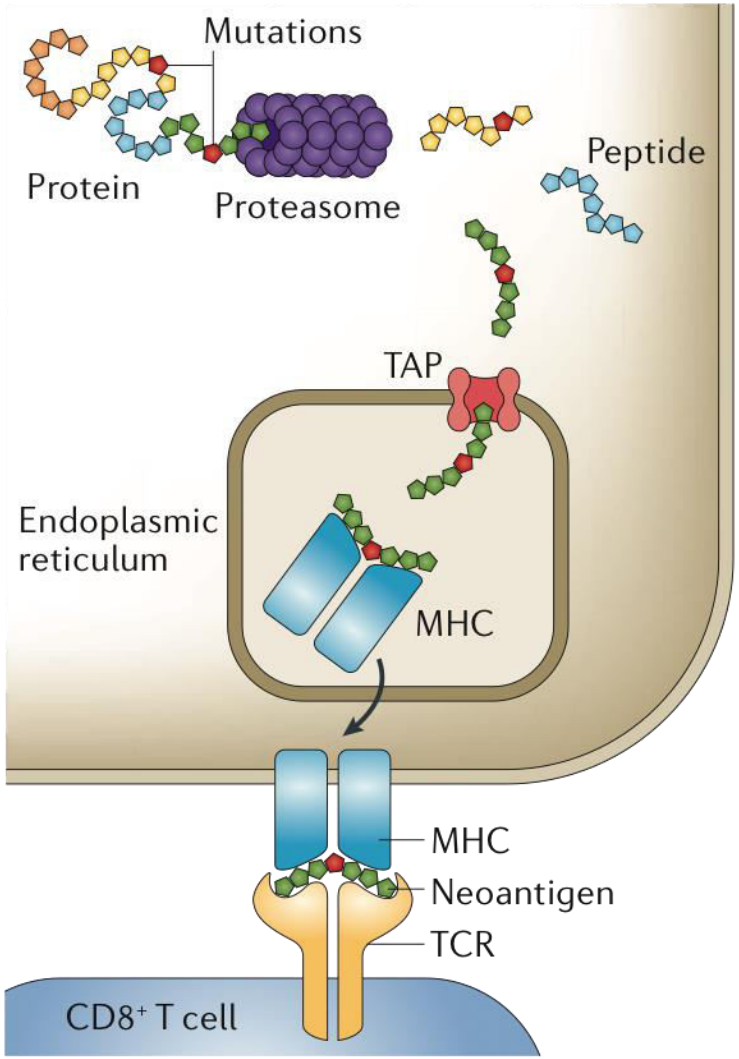
doi:10.1038/nature23003

Personalized RNA mutanome vaccines mobilize poly-specific therapeutic immunity against cancer

Ugur Sahin[1,2,3], Evelyna Derhovanessian[1], Matthias Miller[1], Björn-Philipp Kloke[1], Petra Simon[1], Martin Löwer[2], Valesca Bukur[1,2], Arbel D. Tadmor[2], Ulrich Luxemburger[1], Barbara Schrörs[2], Tana Omokoko[1], Mathias Vormehr[1,3], Christian Albrecht[2], Anna Paruzynski[1], Andreas N. Kuhn[1], Janina Buck[1], Sandra Heesch[1], Katharina H. Schreeb[1], Felicitas Müller[1], Inga Ortseifer[1], Isabel Vogler[1], Eva Godehardt[1], Sebastian Attig[2,3], Richard Rae[2], Andrea Breitkreuz[1], Claudia Tolliver[1], Martin Suchan[2], Goran Martic[2], Alexander Hohberger[3], Patrick Sorn[2], Jan Diekmann[1], Janko Ciesla[4], Olga Waksmann[4], Alexandra-Kemmer Brück[1], Meike Witt[1], Martina Zillgen[1], Andree Rothermel[2], Barbara Kasemann[2], David Langer[1], Stefanie Bolte[1], Mustafa Diken[1,2], Sebastian Kreiter[1,2], Romina Nemecek[5], Christoffer Gebhardt[6,7], Stephan Grabbe[3], Christoph Höller[5], Jochen Utikal[6,7], Christoph Huber[1,2,3], Carmen Loquai[3]* & Özlem Türeci[8]*

RNA-based vaccines

How neoantigens originate



H Hackl*, P Charoentong*, F Finotello* et al., Nature Reviews Genetics, 2016

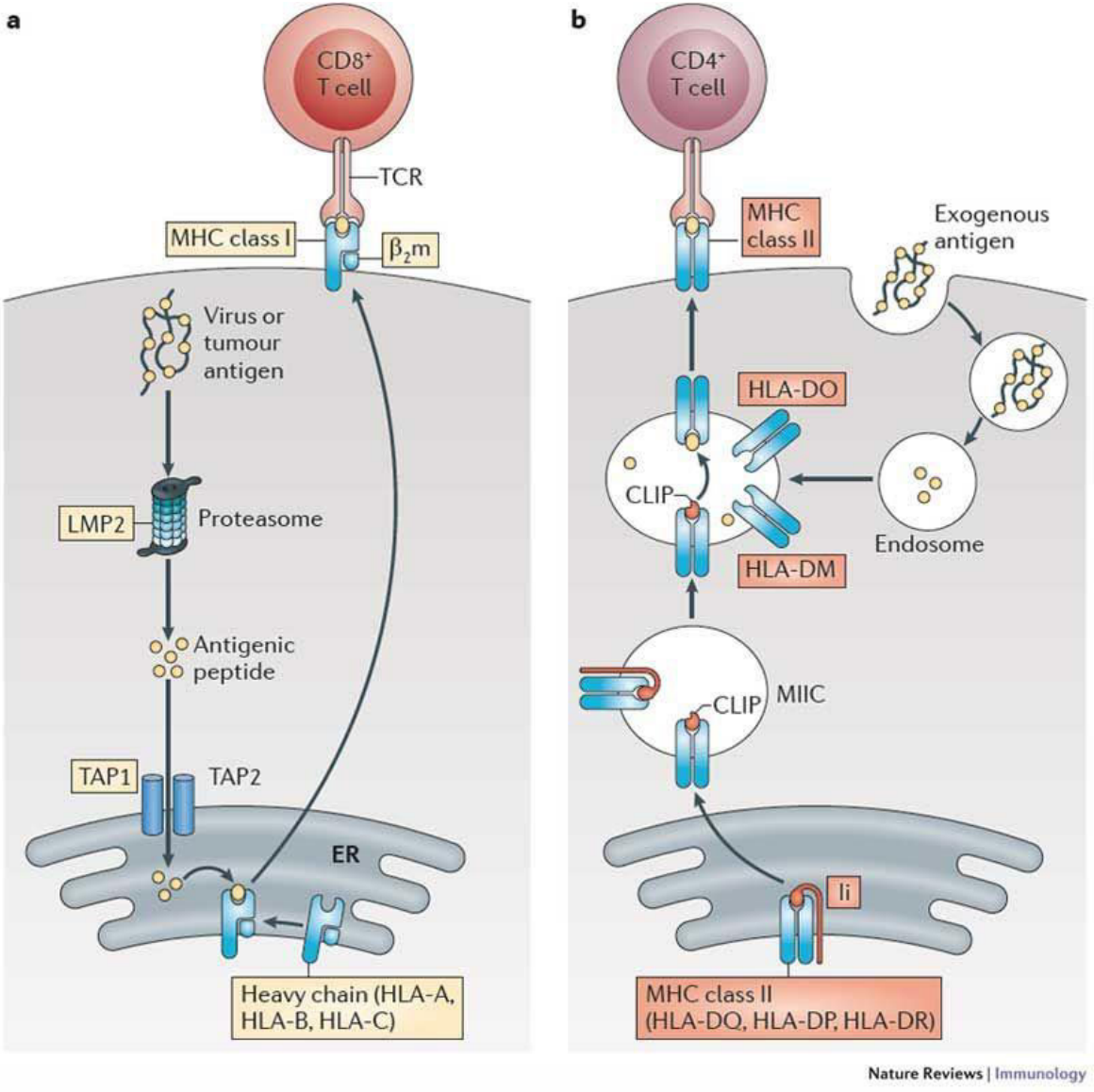
Class-I and class-II antigens

a) **Class-I** MHC molecules

- expressed on all nucleated cells (with some exceptions)
- Present 8-11 amino acid long peptides from intracellular proteins to **CD8+ T cells**

b) **Class-II** MHC molecules

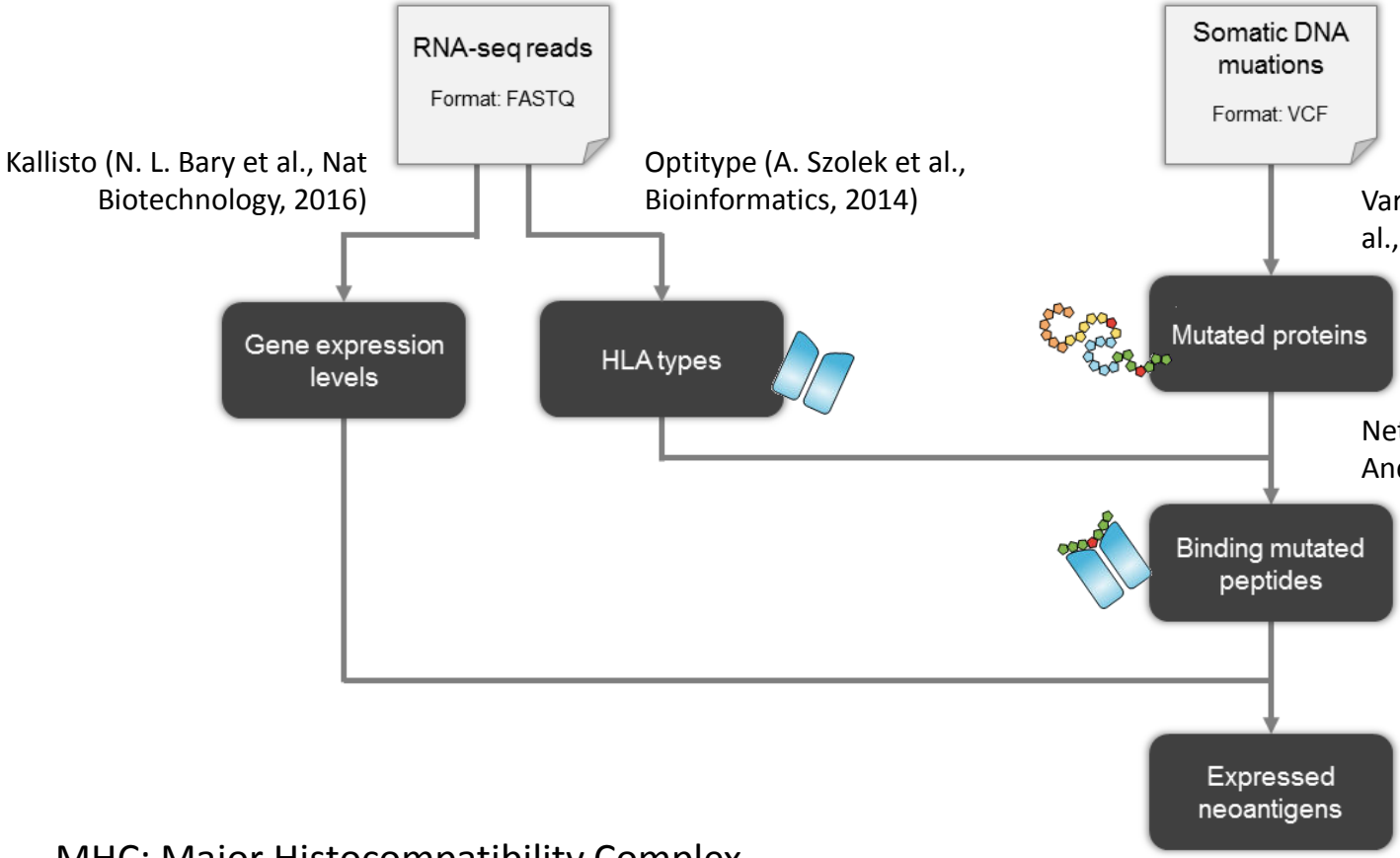
- Expressed on professional antigen presenting cells (APC) like dendritic cells, macrophages, and B cells
- Present 10-30 amino acid long peptides from extracellular proteins to **CD4+ T cells**



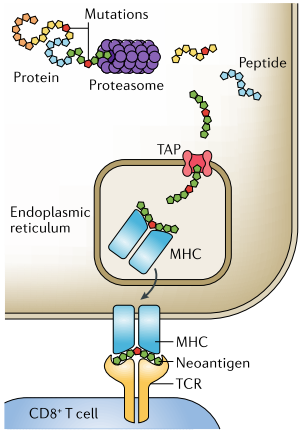
KS Kobayashi and PJ Van Den Elsen, Nature reviews Immunology, 2012

TIminer pipeline for the prediction of class-I neoantigen

Note: algorithms for class-I neoantigen prediction are more accurate than those for class-II



MHC: Major Histocompatibility Complex
HLA: Human Leukocyte Antigen (MHC in humans)



E Tappeiner, F Finotello et al., Bioinformatics, 2017

Running TIminer pipeline

TIminer is available as Docker image (easy installation and usage):

http://www.icbi.at/software/timiner/timiner.shtml

TIminer documentation:

http://www.icbi.at/software/timiner/doc/index.html

The full pipeline can be run with a single Python script: **TIminerPipeline.py**

After the installation, small example data can be analyzed with the full pipeline by executing from the "scripts" directory the command:

```
python TIminerPipeline.py --input ../samples/inputInfo.txt
    --out ../samples/out
```

Running single TIminer modules

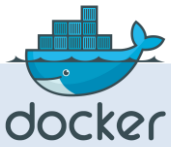
```
from TIminer import TIminerAPI

TIminerAPI.executeKallisto(inputFile1="path/to/input_1.fastq",
                           inputFile2="path/to/input_2.fastq”,
                           outputFile="path/to/norm_expr.txt”,
                           subjectId="Subject_1”,
                           threadCount=2)
```

Code saved in the script **myKallistoScript.py**

The Python script can be executed with:

```
python myKallistoScript.py
```



To use TIminer, Docker must be running on your computer!

Version for multi-sample analysis: `TIminerAPI.executeKallistoDir`