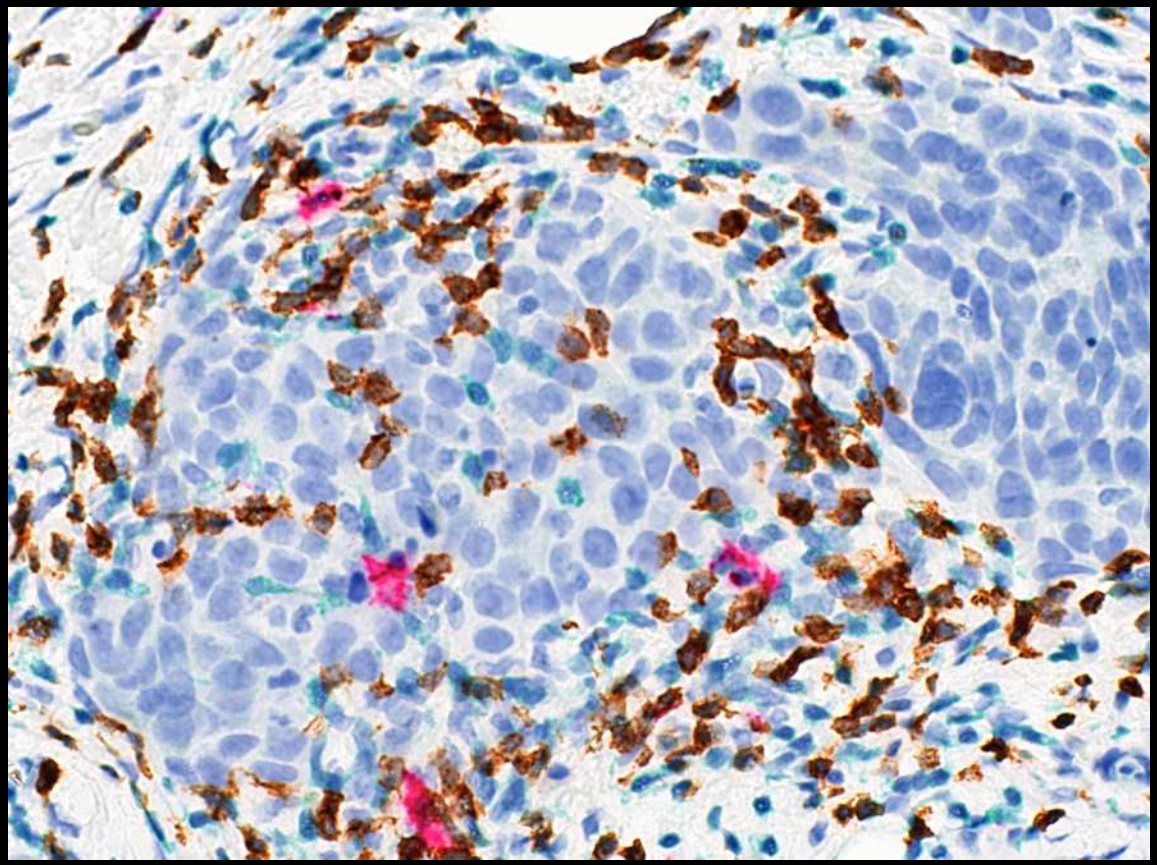
Toward personalized immunotherapy for lymphoma: Identification of common driver mutations recognized by patient T cells

Julie Nielsen, PhD
BC Cancer Agency
Victoria, Canada

Presenter Disclosure Information

Nothing to disclose

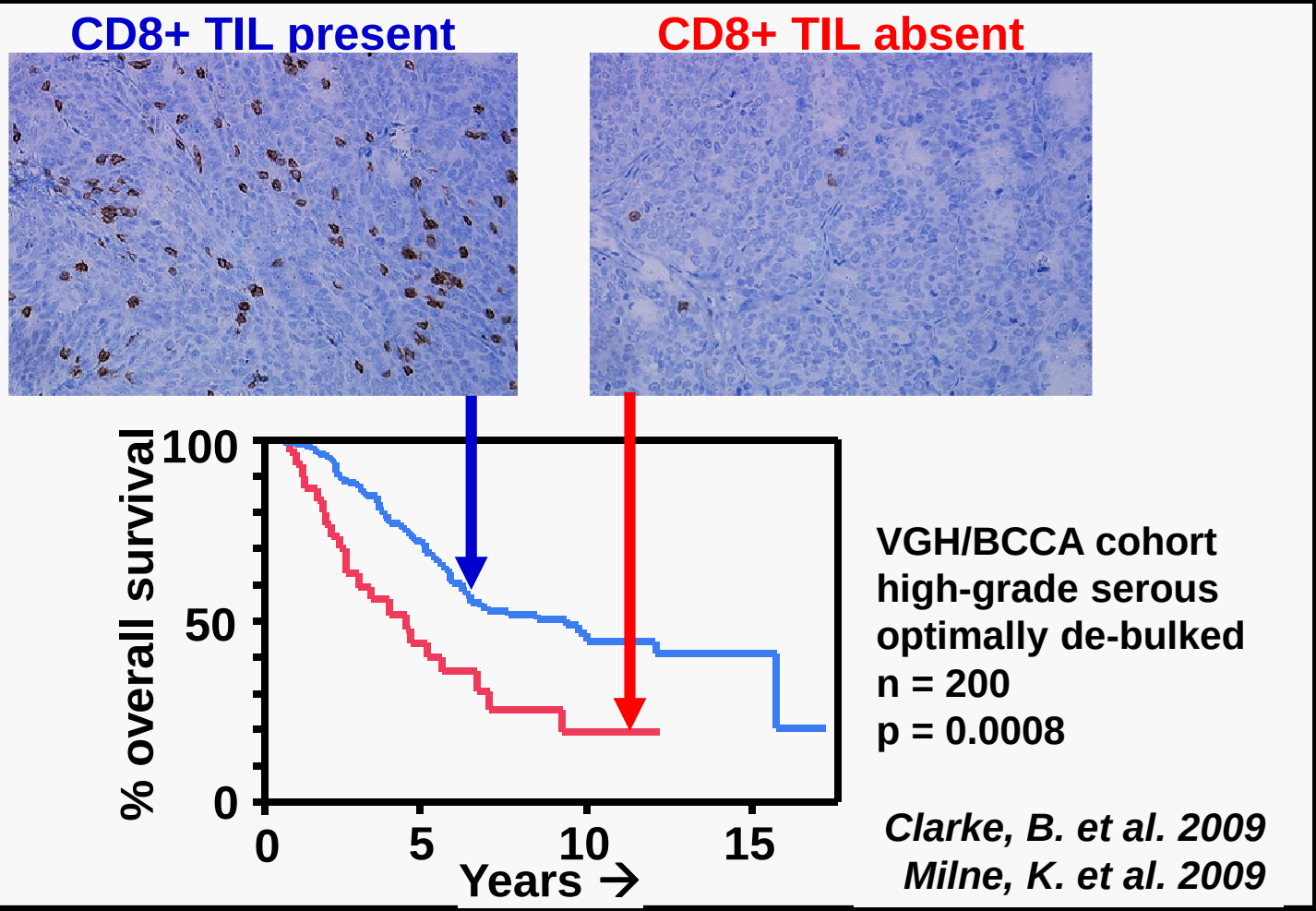
The immune system actively responds to cancer



CD8+ T cells
CD3+ (CD8-) T cells
CD20+ B cells
Tumour cells

Image courtesy of Katy Milne

Tumour-infiltrating T cells are associated with patient survival



Immune response to lymphoma

- Follicular lymphoma is the most common type of indolent non-Hodgkin lymphoma
- B cell-derived
- Incurable with current therapies
- Many recent studies describe correlations between tumour-infiltrating T cells & prognosis

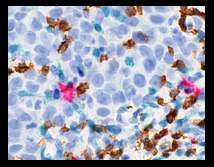
Immunotherapy for lymphoma

- Question: What to target in lymphoma patients?
- There are a collection of frequently mutated genes in lymphoma (tumour-specific)
- The resulting mutations are typically drivers (essential for tumour survival / progression)
- Hypothesis: Tumour-specific mutant proteins derived from driver mutations are effective targets for T cell-based therapy in lymphoma
- Strategy: Stimulate patient T cells from peripheral blood to identify both naïve and pre-existing responses to mutations

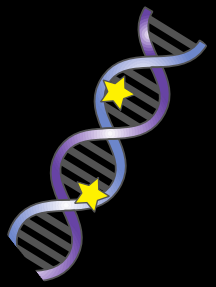
Patient cohort

- Enrolled 53 follicular lymphoma patients
- Project requirements:
  - Tumour samples for identification of somatic mutations
  - Matched blood samples: 2-4 annual blood draws/patient for T cell assays (200ml/draw)

Project overview



Obtain
tumour samples



Identify patient-specific driver mutations

Assess
T cell responses

Develop personalized
T cell-based therapies

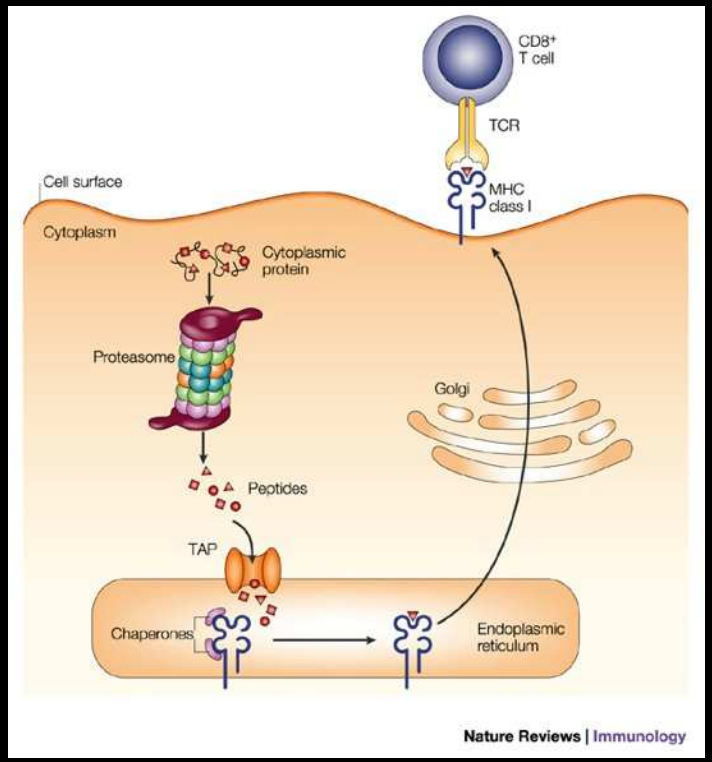
- Every mutation has the potential to create a new T cell target

Using T cells to target mutations

- T cells can
  - Detect small changes in proteins (even single amino acid substitutions)
  - Eliminate tumour cells expressing mutant proteins, regardless of protein function
  - Recognize proteins that reside in any part of the cell

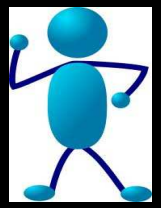
Criteria for immunogenicity

- Protein must be processed to generate the peptide of interest
- Peptides must bind patient's MHC molecules
- T cells with the appropriate T cell receptor must exist in the naïve repertoire



Experimental design

Obtain tumour & matched blood



Sequence candidate genes

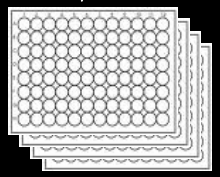
Driver mutations

Epitope predictions

Mutant peptides

T cells

APC



10 days

APC + peptides

14 days

Polyclonal stimulation

14 days

IFN-γ ELISPOT

Minimal peptide

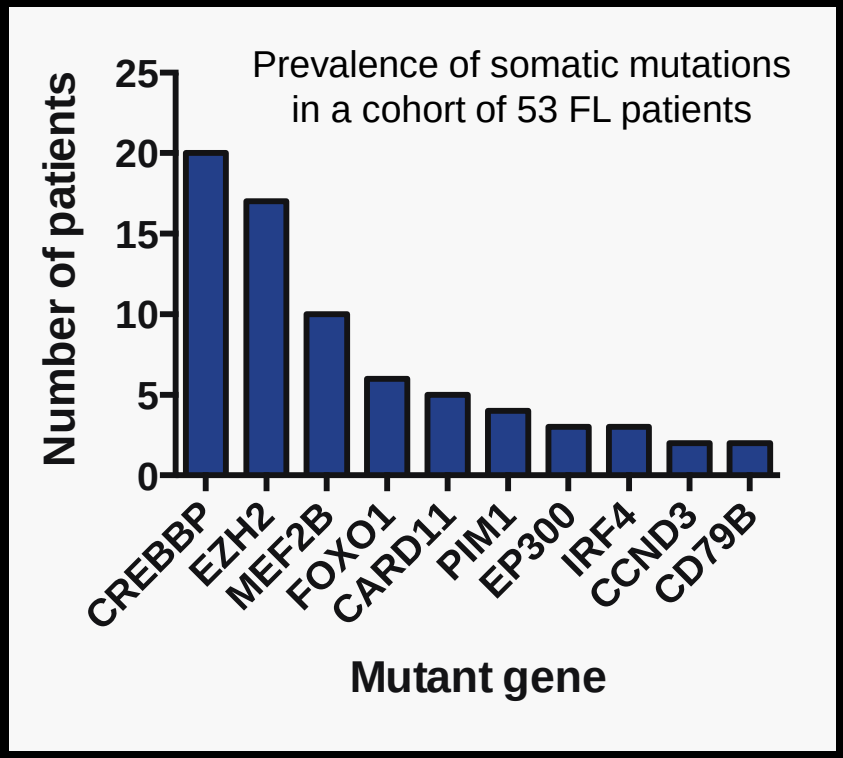
HLA restriction

Cross-reactivity

Protein recognition

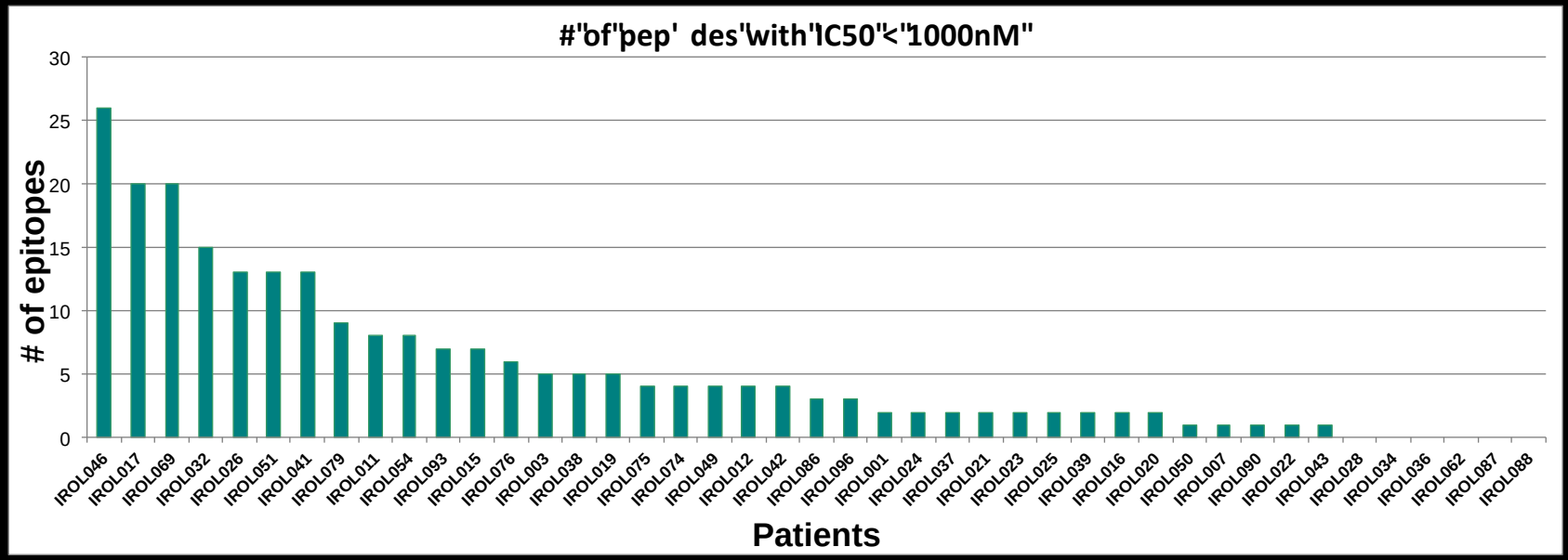
Mutational profiling

- Isolated DNA from tumours & matched normal blood
- Sequenced 10 genes known to be mutated in lymphomas
- Identified mutations in samples from 81% of patients



Epitope predictions

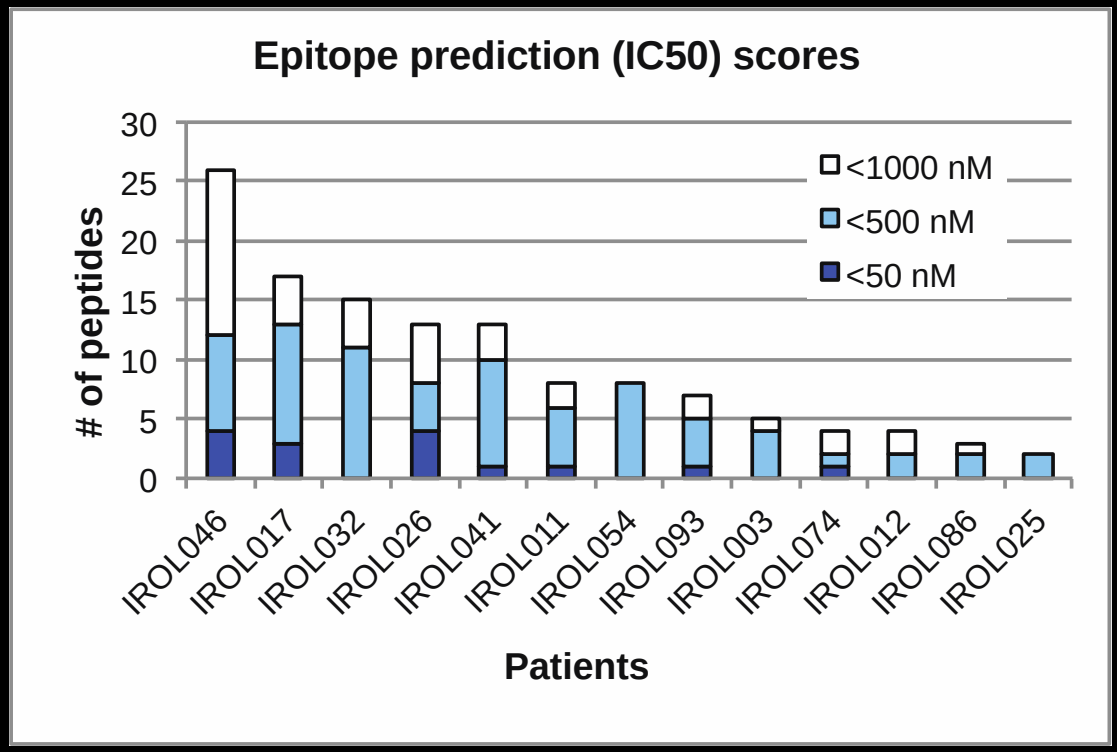
- Sequence-based MHC class I typing of all samples
- Used algorithms to predict which patient-specific mutant peptides would bind patient's MHC molecules



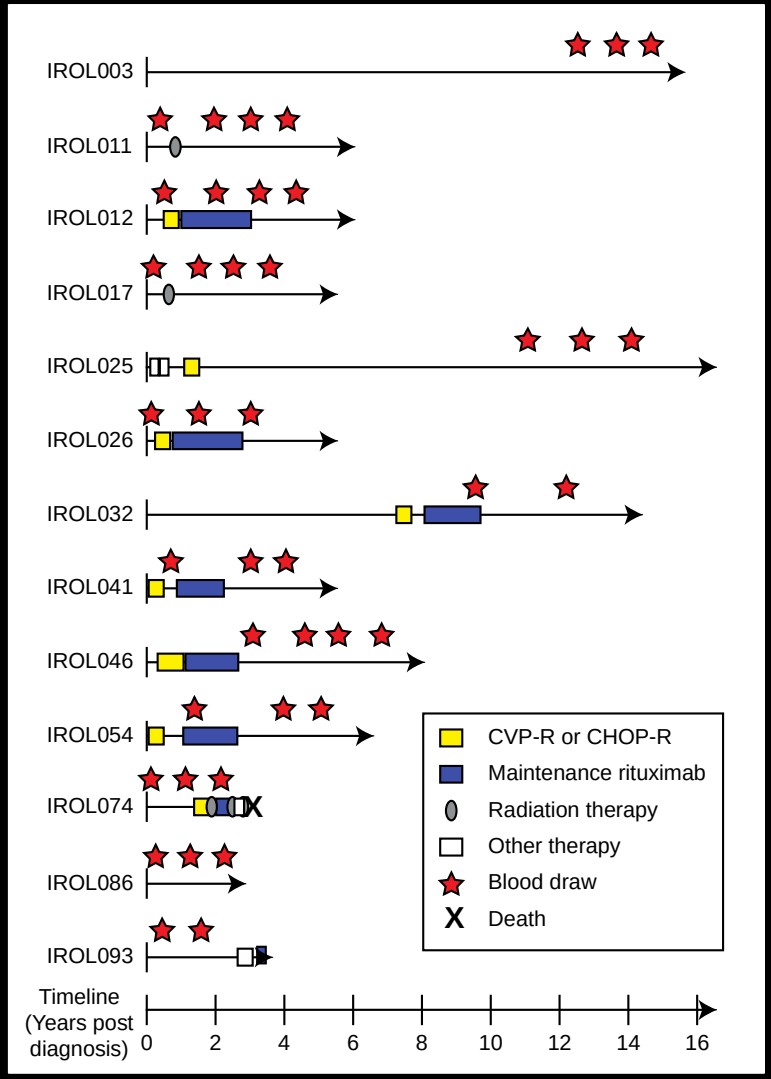
- 86% of patients with mutations had ≥1 predicted epitope

Epitope predictions

- Selected 13 patients with at least one predicted epitope and with sufficient blood available for T cell experiments



Clinical timeline & blood draws

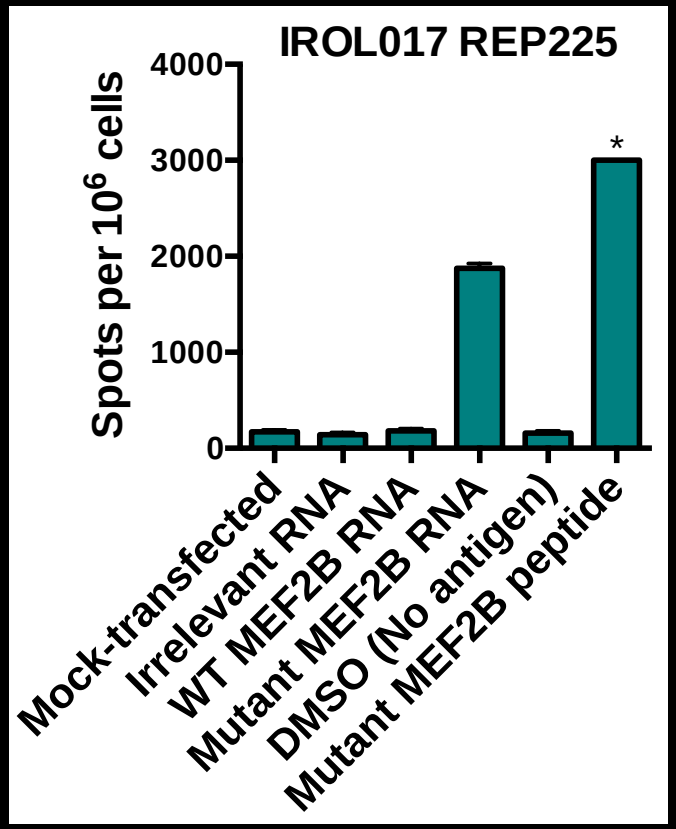


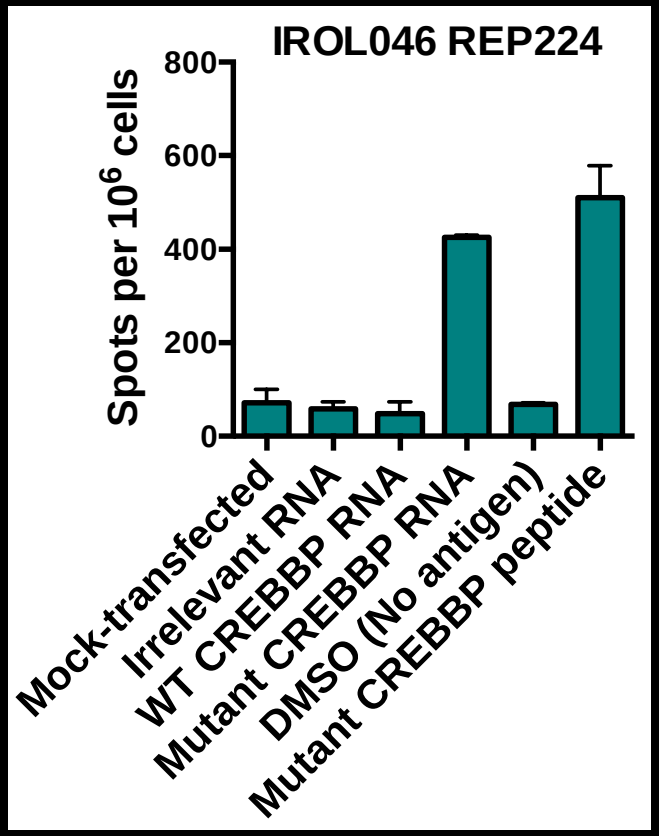
Results of *in vitro* stimulations

Patients	Mutated genes	# of peptides with predicted IC_{50}<1000nM	Peptides eliciting T cell responses
IROL003	EZH2, PIM1	5	None
IROL011	EZH2, FOXO1, MEF2B	8	EZH2
IROL012	CREBBP	4	None
IROL017	CREBBP, MEF2B	17	CREBBP, MEF2B
IROL025	MEF2B	2	None
IROL026	EZH2, MEF2B	12	MEF2B
IROL032	CREBBP, FOXO1	15	None
IROL041	CREBBP, EZH2	13	CREBBP
IROL046	CCND3, CREBBP, PIM1	25	CREBBP
IROL054	CD79B, CREBBP, EZH2, MEF2B	7	MEF2B
IROL074	CCND3, CD79B, PIM1	4	CCND3
IROL086	CREBBP	3	None
IROL093	FOXO1	7	None

- T cells from 7/13 patients recognized mutant peptides

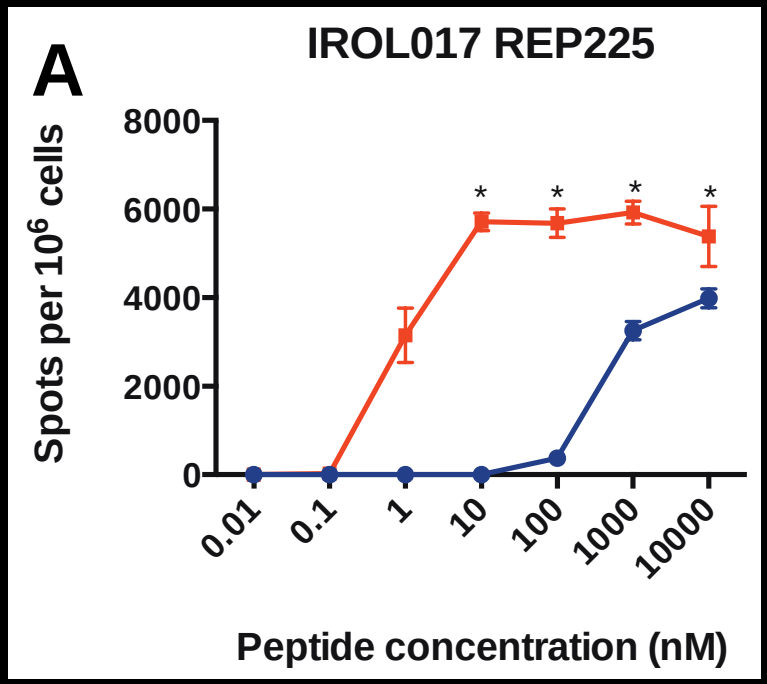
Patient T cells recognize patient-specific mutant proteins (3/7 patients)

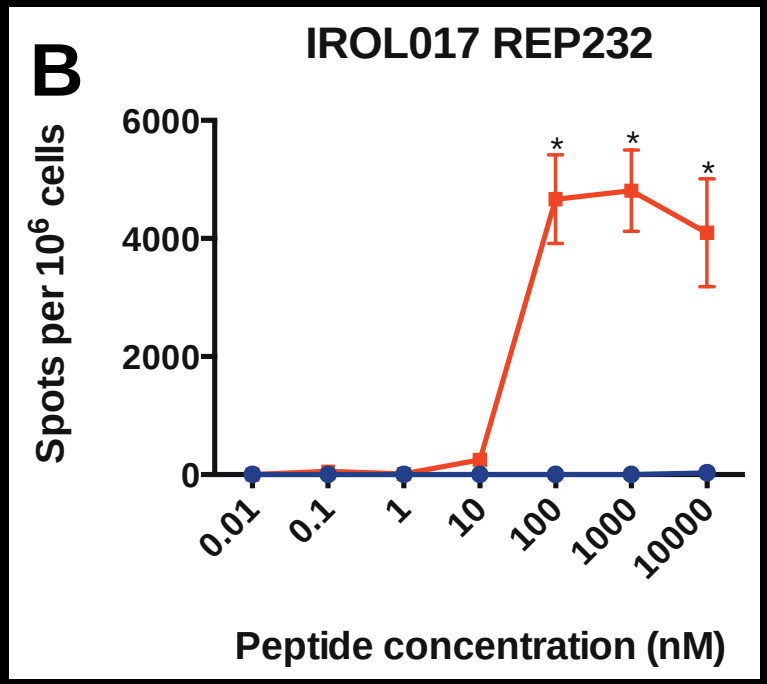




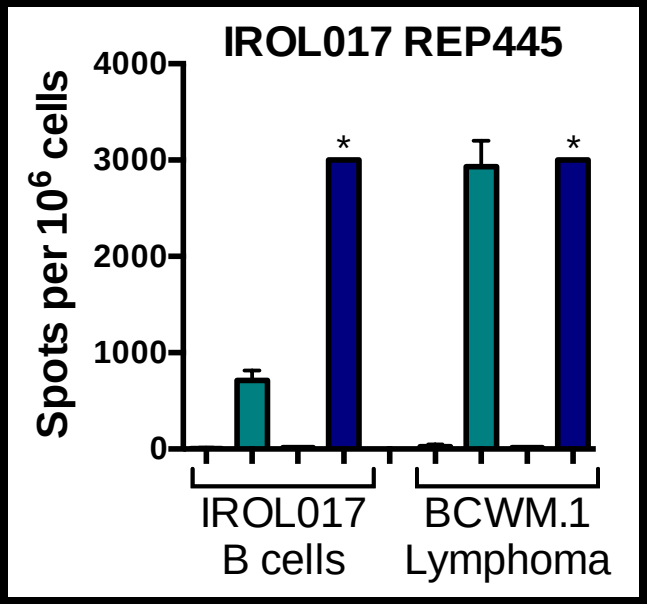
* No autologous tumour available, so transfected autologous B cells with mutant genes

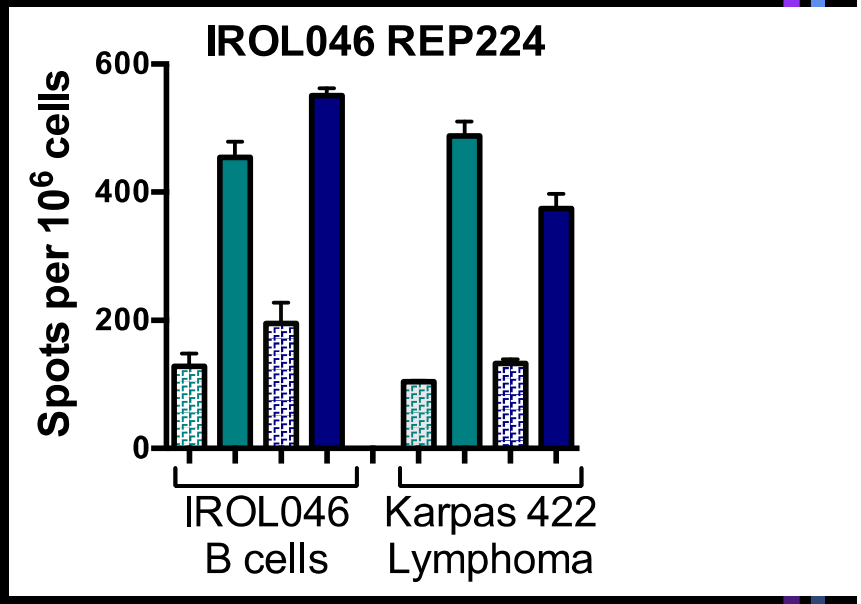
T cells are specific for mutant peptides (compared to wild type)

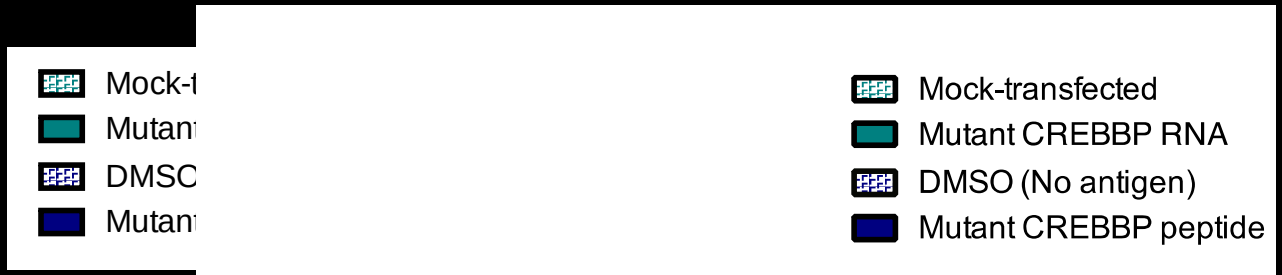




T cells recognize HLA-matched tumour cells expressing mutations







Summary & future directions

- Ideal immunotherapy targets are tumour-specific
- Over 80% of follicular lymphoma patients have mutations in at least 1 of the 10 genes in our panel
- These mutations are typically drivers & are therefore particularly appealing therapeutic targets
- T cells from 3/13 patients recognized processed mutant proteins
- T cell responses were found at low frequencies in peripheral blood & only at a single timepoint in each patient, suggesting that responses may have been elicited from the naïve repertoire
- Additional targets will be identified by assessing responses to more driver mutations & by assessing CD4+ T cell responses

Acknowledgements

- BC Cancer Agency's Deeley Research Centre
  - Brad Nelson, PhD
  - Colin Sedgwick
  - John Webb, PhD
  - Lewis Liu
  - David Kroeger, PhD
- Patients & control donors
- Funding
  - BC Cancer Foundation
  - Canadian Cancer Society Research Institute
  - Lymphoma Foundation Canada
  - Canadian Institutes of Health Research
  - Genome BC
- BCCA's Michael Smith Genome Sciences Centre
  - Marco Marra, PhD
  - Richard Moore, PhD
- Simon Fraser University
  - Ryan Morin, PhD
  - Zabrina Brumme, PhD
  - Aniqa Shahid
  - Zusheng Zong
  - Stephen Yu
  - Natalie Kinloch
- BCCA & Royal Jubilee Hospital
  - Nicol Macpherson, MD PhD
  - Joe Connors, MD
  - Randy Gascoyne, MD
  - Brian Berry, MD
  - Qiwei Xu, MD
  - Adria Devlieger
  - Tim Turcotte
  - Jason Hart, MD
  - Ken Wilson, MD
  - Adrian Yee, MD
- Dana-Farber Cancer Institute
  - Steve Treon, MD PhD