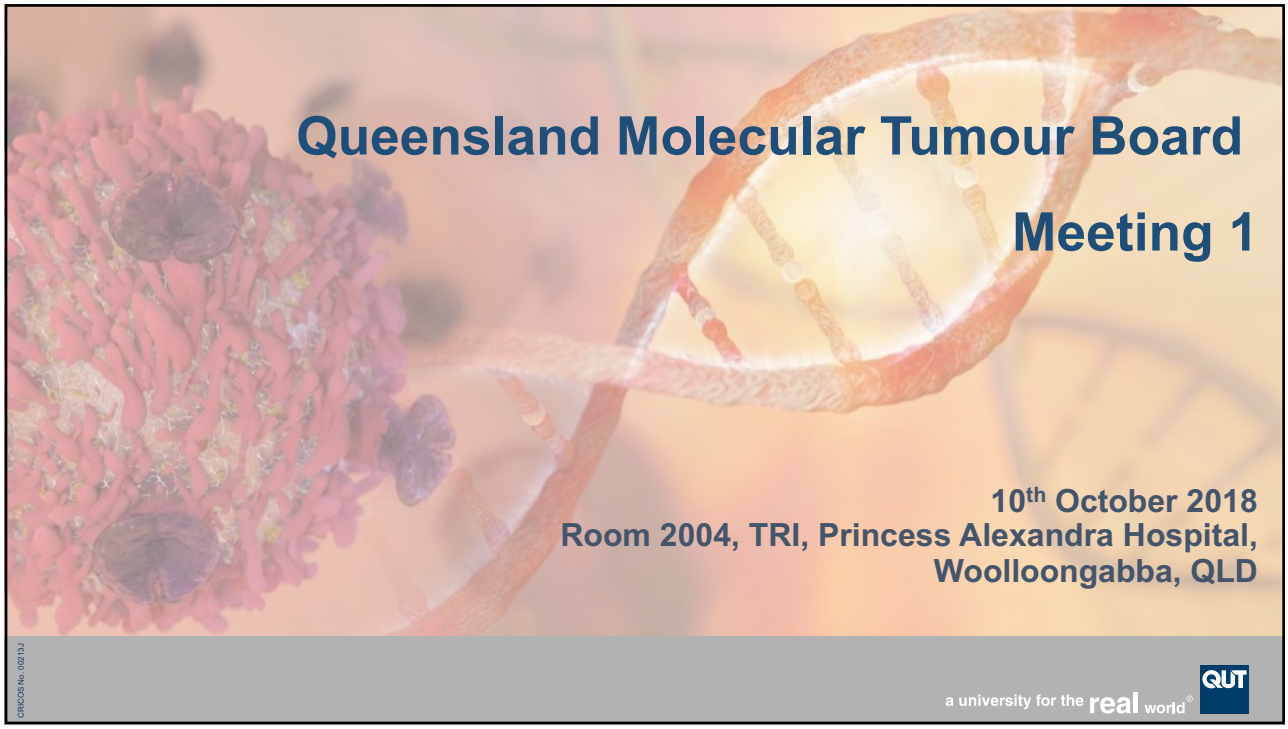

Queensland Molecular Tumour Board

Meeting 1

10th October 2018
Room 2004, TRI, Princess Alexandra Hospital,
Woolloongabba, QLD

Overview

- MoST clinical trial coming in 2019
- Breast Cancer Patient (Wen Wu and Maree Colosimo)
- NSCLC Patient (Ken O'Byrne)
- Urachal Adenocarcinoma (Ken O'Byrne)

Patient Review Format

1. Clinical History
2. Molecular Profile
3. Discussion

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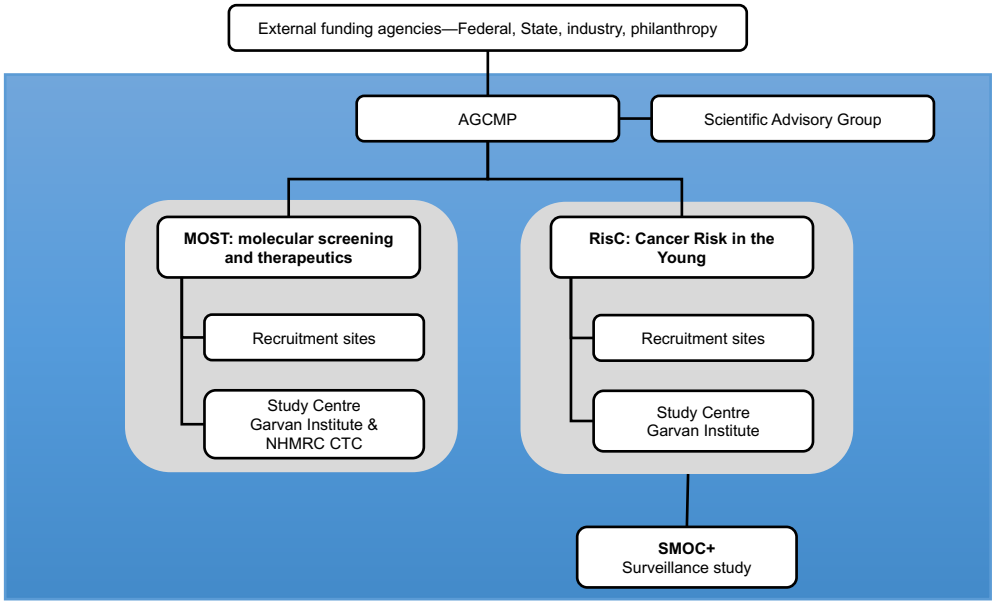
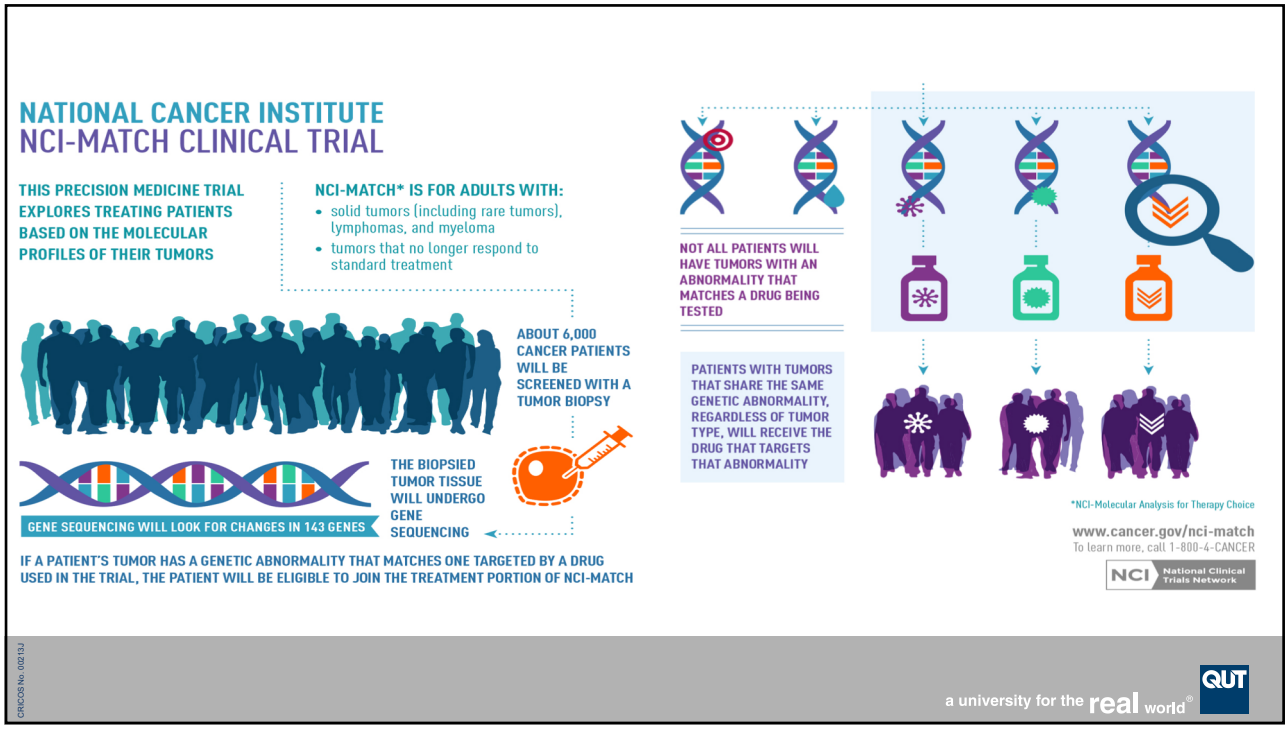
Australian Genomic Cancer Medicine Program



Lead sites in Queensland

- Princess Alexandra Hospital/MetroSouth HSS
  - Node lead Prof Kenneth O'Byrne
- Pathology Queensland
  - Lead clinician Prof Sunil Lakhani
- **Aim** to expand the program into all main hospitals in Brisbane and Queensland including
  - Lady Cilento Children's Hospital
  - Royal Brisbane and Women's Hospital
- **Will look for further support from QH and QGHA**



The MoST program

GARVAN INSTITUTE OF MEDICAL RESEARCH

Aims of the Molecular Screening and Therapeutics (MoST) program:

- Facilitate and expedite clinical testing of novel drugs using an innovative signal-seeking clinical trial design
- Provide access to molecular screening and novel therapies to patients with advanced cancer and unmet clinical need; focus: rare/neglected cancers

Modular signal-seeking trial design:
overarching 'framework protocol' for molecular screening platform and multiple, parallel biomarker-driven signal-seeking clinical substudies

➡ expedited addition of new therapeutic substudies

The Molecular Screening and Therapeutics study (MoST)

Molecular Screening

Actionable mutation: >20%

No actionable mutation: <80%

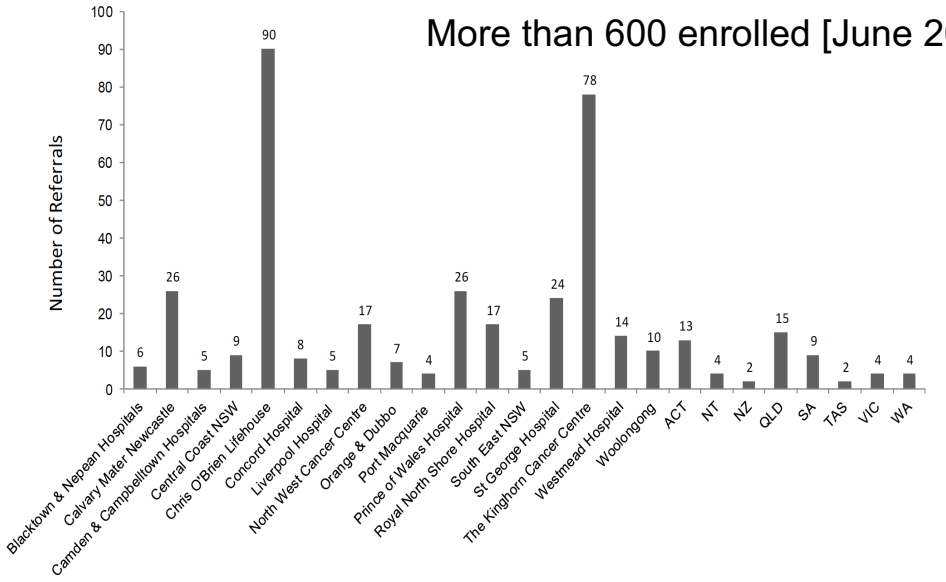
Targeted therapy

Immunotherapy

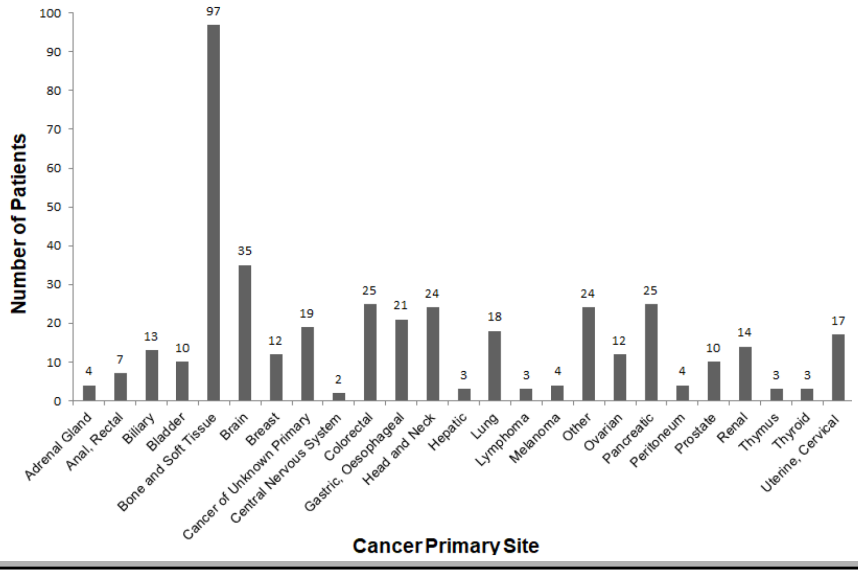
CDK4/6 inhibitor	PARP inhibitor	Other	PD-L1+CTLA-4 inhibitors
Rb pathway: CCND1/2/3, CDK4, CDKN2A mut/ampl/del **Palbociclib** 16 patients	HR DNA repair: BRCA1/2 + BRCA complex mut/del **Olaparib** + Durvalumab 48 patients (3 subgroups)		No actionable alterations **Durvalumab + Tremelimumab** 64 patients (4 subgroups)
Closed	Opened Nov 2017		Closed

MoST progress to date

More than 600 enrolled [June 2018]



MoST progress to date



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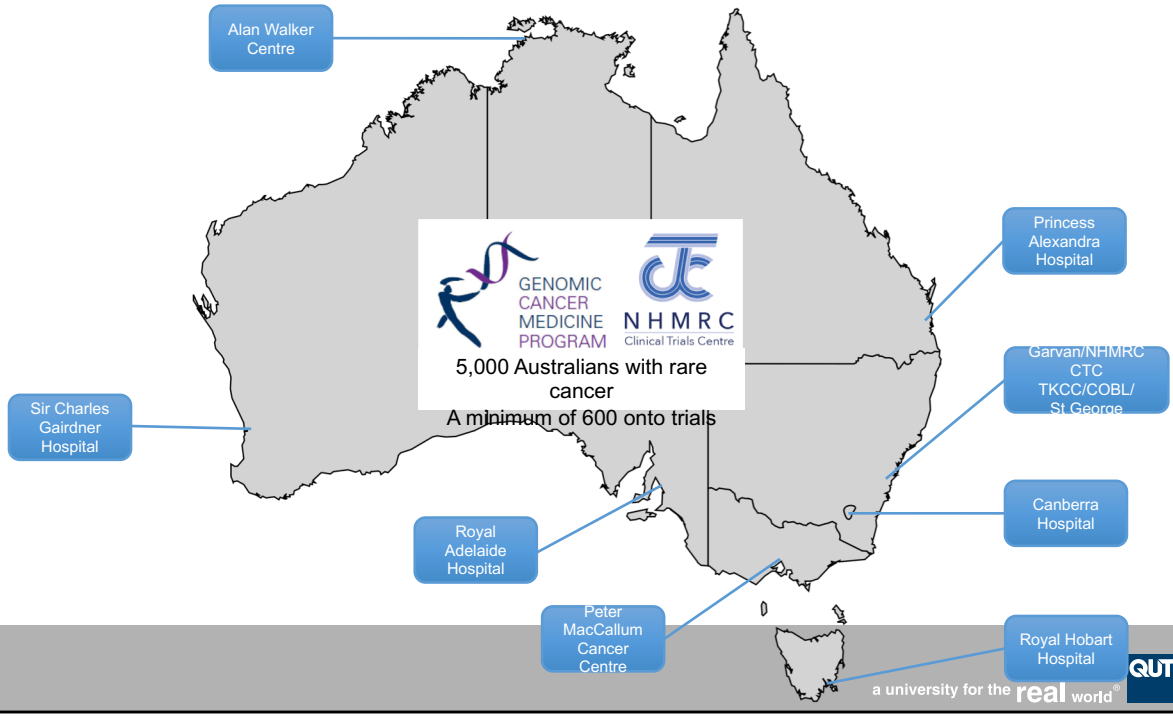

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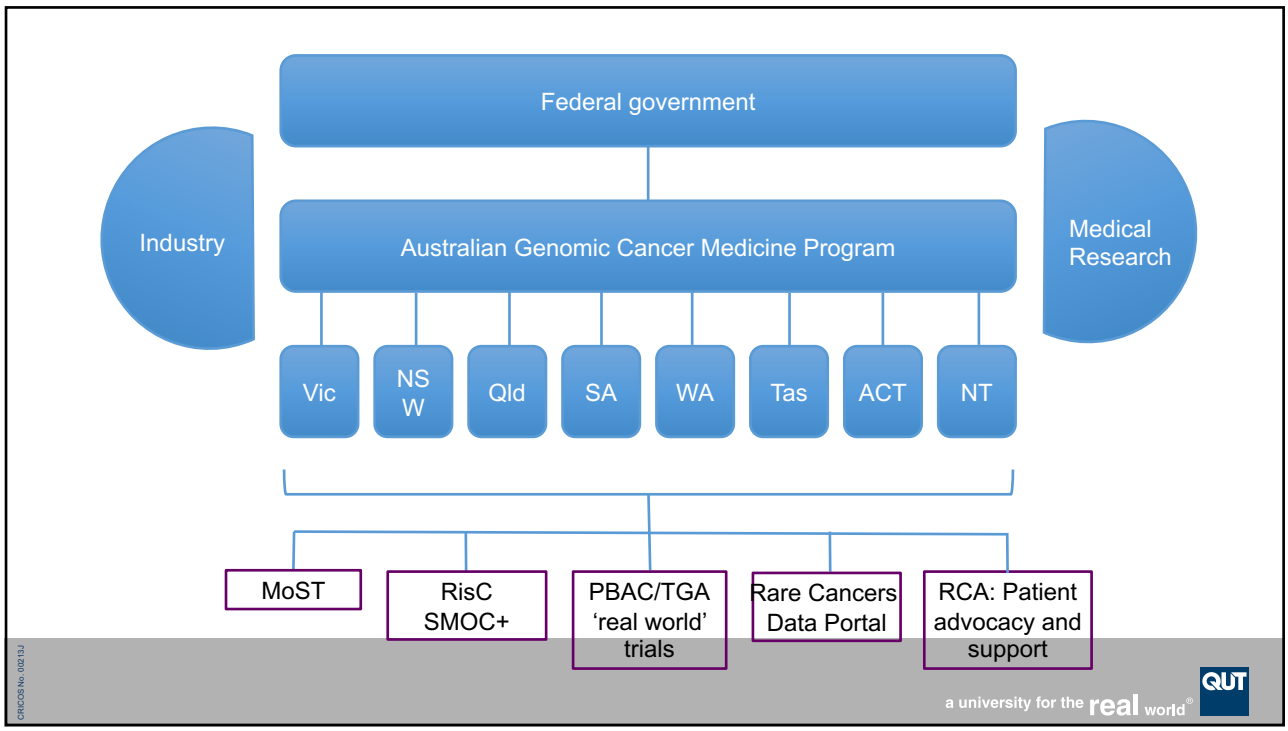
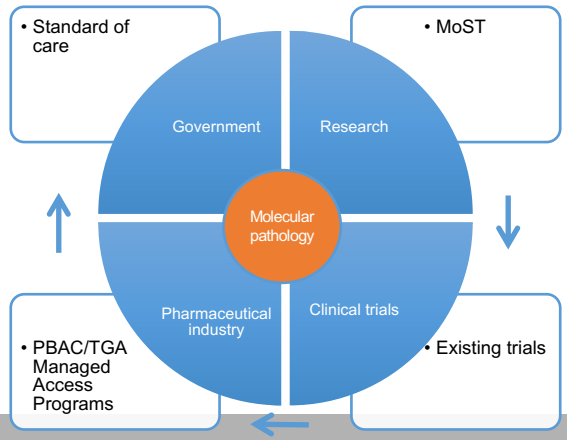
a university for the real world® QUT



An ecosystem that brings together pharma, research, clinical care and government



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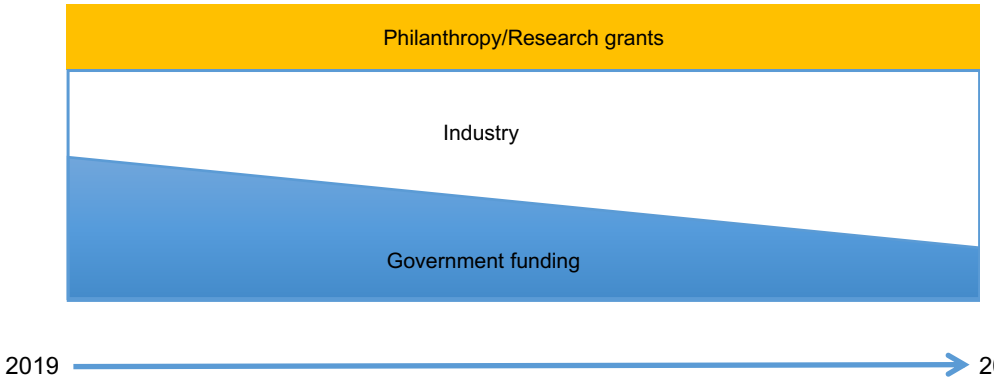
A sustainable business plan



Gro

Industry

Community organisations

Collaborators

Case 1

Case 1

- A 42 year old woman with an unremarkable medical history presented with a R breast lump
- Mammogram and ultrasound found this to be innocent but identified a suspicious lesion deeper in breast tissue.
- Needle biopsy confirmed it was a Grade 2 ER+ cancer which measured <1cm.
- Nodes were negative
- Lumpectomy and radiotherapy were performed.
- Tamoxifen was prescribed but not well tolerated and discontinued after 9 months

Medical History continued

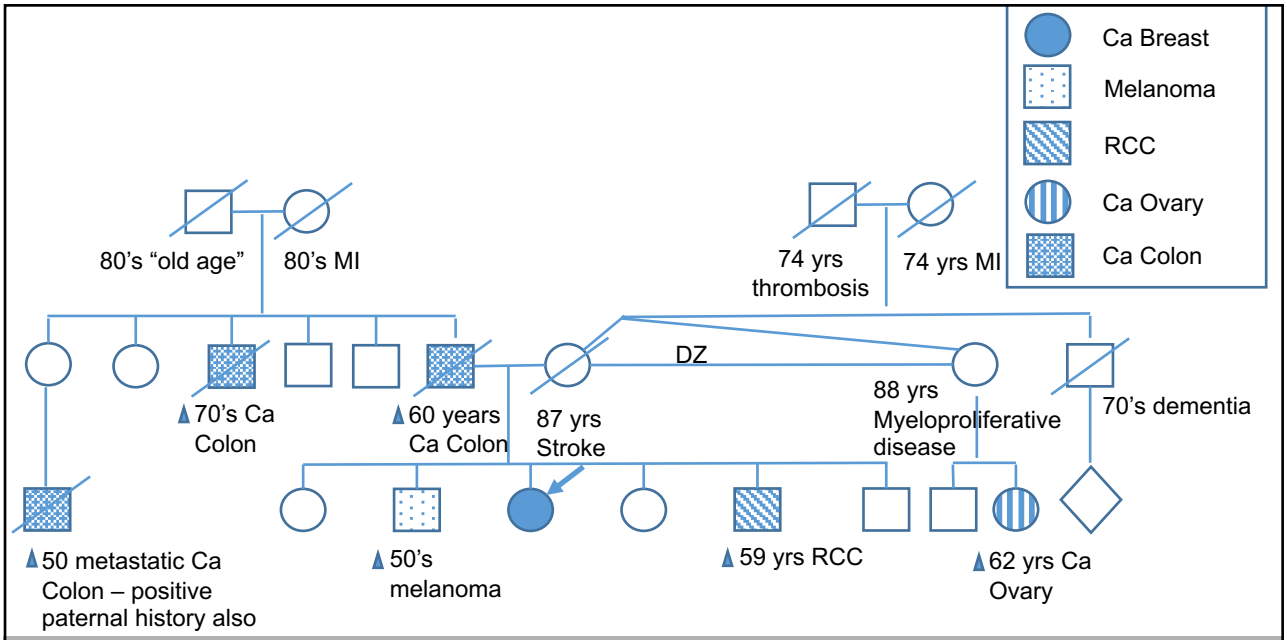
- At 56 years of age, routine mammogram and ultrasound was performed
- Suspicious lesion detected on ultrasound, also in R breast
- Grade 2, <1cm, ER+
- Believed to be a second primary
- Unilateral mastectomy
- Sentinel lymph node confirmed that it hadn't migrated to apex axilla
- Tamoxifen – for five years
- No chemotherapy

Medical History cont….

- In Feb 2018, at 61 years routine screening identified enlarged lymph nodes in L axilla.
- Not clinically palpable.
- Diagnosed with axillary tail breast cancer, ER+ 95%, PR+ 5%, HER -ve
- Second mastectomy and axillary dissection – believed to be 3rd primary
- PET scan showed bone and liver metastases with two positive pancreatic nodes
- Femara (aromatase inhibitor)(2.5mg once daily)
- Treatment
  - Kisquali 600mg daily for three weeks a month
  - XGEVA (denosumab) – monthly injection (bone metastases)
  - Tolerating medication well, though does have Kisquali associated neutropenia and macrocytosis (treated with folate)

Medical History cont…

- In May, presented with a post-menopausal bleed.
- U/S diagnosed polyp which was removed surgically
- Prophylactic oophorectomy performed at same time.
- In July, repeat PET scan showed reduction in bony metastasis, reduction in size of liver metastases and the two pancreatic nodes were no longer visible.



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Germline Testing

- QML March 2018
  - *ATM, BRCA1, BRCA2, CDH1, CHEK2, PALB2, PTEN, STK11,TP53*
  - Negative for point mutations and CNVs
- COLOR July 2018
  - *APC, ATM, BAP1, BARD1, BMPR1A, BRCA1, BRCA2, BRIP1, CDH1, CDK4, CDKN2A, CHEK2, EPCAM, GREM1, MITF, MLH1, MSH2, MSH6, MUTYH, NBN, PALB2, MSH2, POLD1, POLE, PTEN, RAD51C, RAD51D, SMAD4, STK11, TP53*
  - Negative for point mutations in all

Germline Testing

- QML March 2018
  - *ATM, **BRCA1, BRCA2, CDH1**, CHEK2, PALB2, **PTEN, STK11,TP53***
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  - Negative for point mutations in all

Highly penetrant breast cancer genes which account for 25% of familial cases

Germline Testing

- QML March 2018
  - *ATM, BRCA1, BRCA2, CDH1, CHEK2, PALB2, PTEN, **STK11**,TP53*
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  - Negative for point mutations in all

Colorectal cancer susceptibility genes – account for 5-7% of all cases

Patient has had regular colonoscopies since the age of 40 years and has only had two benign polyps removed in her mid fifties.

Somatic Testing

Genes Reported

ABCB1, ABCC3, AGR2, AKT1, AKT2, AKT3, ALK, AR, BIRC5, BRCA1, BRCA2, CCND1, CCNE1, CD274, CDK4, CDK6,CDKN1B, CDKN2A, CYP2D6, ERBB2, ERBB4, ESR1, FCGR2A, FCGR2B, FCGR3A, FGF3, FGFR1, FGFR2, FOXP3, GNAS,KLLN, MKI67, MMP2, MMP9, MTAP, MTOR, MYD88, NCOA3, NF2, NTRK3, PALB2, PGR, PIK3CA, PIK3R1, PTEN,RAD51D, RB1, RET, RSF1, SF3B1, SGK1, TFF3, TIMP1, TLK2, TOP2A, TP53, TUBB3

Somatic Findings

- No SNPs or InDels of clinical relevance:

- Tumour Purity estimate 70%
- Tumour Burden 2.5 (Muts/Mb)
- Clinically Relevant CNVs in Breast Cancer
  - *RSF1* Gain 21-55 x
  - *PTEN* Homozygous Loss (0 copies)
- Other CNVs (heterozygous loss)
  FANCC, GSTP1, MRE11, STK11, SMARCB1, NF2, COX10, CYP2D6

RSF1 amplification (remodeling and spacing factor 1)

EID	DESC	DIS	DRUGS	EL	ET	ED	CS	VO	TR
857	In retrospective study, 413 pat...	Breast Cancer	Tamoxifen	B					2★

A retrospective study, (413 pre-menopausal high risk patients were assessed for *RSF1* amplification.

28 patients with *RSF1* high amplification did not show a benefit from adjuvant tamoxifen compared to placebo (12 treated vs 16 placebo). HR=1.11, p=0.09

381 patients without amplification did show a benefit compared to placebo (187 treated vs 194 placebo).

Keilty et al., 2013, et al :PLoS One. 2013 Dec 19;8(12):e81740.

PTEN loss information from clinical databases

Biomarker	Drug	Effect	Evidence	Source	Curator	Tumor type
PTEN						BRCA
PTEN biallelic inactivat...	BYL719 (PIK3CA inhibitor)	Resistant	Case report	PMID:25409150	DTamborero	BRCA
PTEN deletion	PI3K pathway inhibitors	Responsive	Pre-clinical	PMID:212892...	RDientsmann	TH, G, L, OV, BR...
PTEN oncogenic mutati...	ATM inhibitors	Responsive	Pre-clinical	PMID:27397505	RDientsmann	BRCA
PTEN oncogenic mutati...	PI3K pathway inhibitors	Responsive	Pre-clinical	PMID:212892...	RDientsmann	TH, G, L, OV, BR...
PTEN oncogenic mutati...	Everolimus + Trastuzumab + Chemot...	Responsive	Late trials	PMID:27091708	RDientsmann	BRCA

EID	DESC	DIS	DRUGS	EL	ET	ED	CS	VO	TR
		Breast							
1297	Archival tumor samples from ...	Her2-receptor Positive **Breast** ...	Everolimus	B					4★
1385	A subgroup of HER2-positive ...	Her2-receptor Positive **Breast** ...	Trastuzumab	B					3★
1386	While some reports indicate P...	Her2-receptor Positive **Breast** ...	Trastuzumab	B					3★
645	PTEN loss is associated with I...	**Breast** Cancer	Trastuzumab	B					1★

PTEN LOSS evidence 1297 summary:

- BOLERO-1 and -3 trials analyzed for PI3K pathway mutations next-generation sequencing, immunohistochemistry (IHC) and Sanger sequencing.
- Patients with advanced breast cancer were treated with trastuzumab, chemotherapy and randomized to receive either everolimus or placebo.
- In the pooled analysis, patients with PTEN loss (had a longer progression-free survival with everolimus compared to placebo (HR 0.54 (95%CI 0.31 to 0.96, P=0.04)
- Patients with PTEN normal status had a hazard ration of 1.00 (95%CI 0.8 to 1.26, P=0.97).
- Patients with PI3K hyperactive pathway (PTEN loss and AKT E17K, PI3K) had a hazard ratio of 0.67 (95%CI 0.48 to 0.93, P=0.02) compared to HR 1.19 (95%CI 0.87 to 1.62, P=0.28) in patients with PI3K normal pathway.
- These trends were independently confirmed in both trials but only pooled results showed consistent statistical significance.

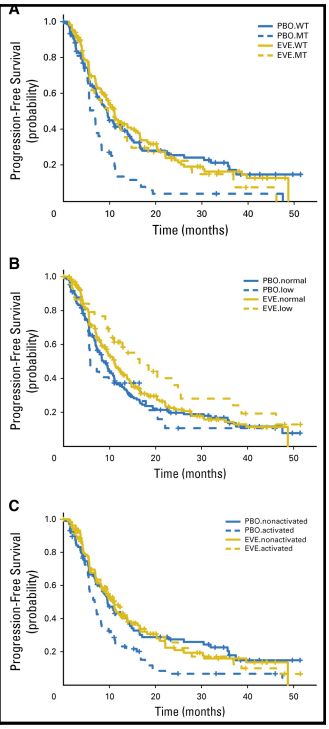
Andre' et al

- From Figure 4 Fig 4.

Kaplan-Meier curves for progression-free survival by biomarker status (pooled data). (A) PIK3CA wild type (WT) versus mutant (MT).

(B) PTEN normal versus low/loss.

(C) PI3K pathway activity normal versus hyperactive (narrow definition). EVE, everolimus; PBO, placebo.



Fabrice André; Sara Hurvitz; Angelica Fasolo; Ling-Ming Tseng; Guy Jerusalem; Sharon Wilks; Ruth O'Regan; Claudine Isaacs; Masakazu Toi; Howard Burris; Wei He; Douglas Robinson; Markus Riester; Tetiana Taran; David Chen; Dennis Slamon; Journal of Clinical Oncology 2016, 34, 2115-2124.
DOI: 10.1200/JCO.2015.63.9161

a

PTEN and *CDK4* pathway

FDA-approval of the CDK4/6 inhibitor palbociclib used with the aromatase inhibitor letrozole for ER+ breast cancer.

Some evidence that supports favourable interaction of *PTEN* loss with *CDK4/6* inhibitors. Though not considered clinically actionable as biomarkers

- Palbociclib has antitumour effects on *PTEN*-deficient endometrial neoplasias [1]
- Cancer-specific mutations such as those affecting receptor tyrosine kinases (RTKs), *RAS, RAF, PI3K* or *PTEN* mutations enhance cyclin D-dependent CDK4/6 activity. [2]

[1] Dosil et al J Pathol 2017; 242: 152–164
[2] Sheer et al Cancer Discov. 2016 Apr; 6(4): 353–367.

Review

- *RSF1* Gain 21-55 x
- *PTEN* Homozygous Loss (0 copies)
- Other CNVs

  FANCC, GSTP1, MRE11, STK11, SMARCB1, NF2, COX10, CYP2D6

PTEN as a biomarker for future treatment?

Significance of *FANCC* and/or *STK11* heterozygous loss?

- ER-positive breast cancer cells became resistant to *CDK4/6* inhibitors, by amplification of the *CCNE1* gene and loss of the *RB1* gene.
- In vitro analysis showed that cells that acquired resistance to *CDK4/6* inhibitors through *CCNE1* gene amplification were sensitive to targeting of *CDK2* whereas those that acquired resistance through *RB1* gene loss were not sensitive

Turner C AARC Abstract 2018

Case 2

Case 2

- A 74 year old woman diagnosed with Urachal Adenocarcinoma

Biopsy1: 05/ Aug /2014

Bladder tumour: High grade papillary and invasive urothelial carcinoma suggested.

Biopsy2: 05/ Sept /2014

Primary urachal adenocarcinoma diagnosed. Carcinoma is clear of the margins.

Biopsy3: 10 / Nov /2016

Subcranial lymph Node “metastatic adenocarcinoma from elsewhere” identified

Biopsy 4: 06/ Feb/ 20018 (WES performed)

Pelvic Mass: Recurrent urachal tumor (fibromuscular tissue Infiltrated by poor differenced adenocarcinoma

Urachal Adenocarcinoma (1% of bladder cancers)

- *KRAS, BRAF, GNAS* and Her2 have been associated with UraC.
- A useful panel for differential diagnostics and clinicopathologic prognostication needs to be developed.
- Study using a gene sequencing assay, shows mutations for adenomatous UraC in Her2 (20%), *KRAS* (20%) and *GNAS* (10%) indicating their usefulness for further evaluation.
- *KRAS* mutations associated with better overall survival.
- *KRAS* and *GNAS* mutations result in the up-regulation of the MAPK signal-transduction pathway, indicating potential for targeted therapy.

From : Behrenht et al. *Genetics and biological markers in urachal cancer* Trans Androl Urol. 2016 Oct; 5(5): 655–661.

Somatic Findings

- No SNPs or InDels or CNV relevant to bladder cancer of clinical relevance:
- Pan-Cancer report provides

- Tumour Purity estimate 70%
- Tumour Burden 6.9 (Muts/Mb)
- Clinically Relevant CNV with Breast Cancer
  BRCA2:p.His1932Asn (missense_variant, VAF = 80.3% Tumour AD = 134,538 Normal AD = 703,1)
- Other CNVs
  FBXW7, SMAD4 loss

Case 3

Case 3

- A 70 year old woman diagnosed NSCLC
- History…

Case 3

- WES sequencing 16th-Feb
- Somatic Reporting on of

PDCD4, BRAF, SMARCA4, ROS1, RRM1, ACTA1, WEE1, PIK3CA, AURKA, ABCB1, KRAS, STK11, ERBB2, NRG1, CDKN2A, RB1,EGFR, AREG, TTF1, MAP2K7, BCL2L11, KIT, TYMS, CBLB, CCND1, GNAS, ALK, FGFR1, NOTCH1, PIM1, CD274, BIRC5, XRCC1,ETS2, TP53, ERCC2

Somatic Findings

- SNPs and Indels

  KRAS Gly12Cys (missense_variant, VAF = 71.2, Tumour AD = [246, 605], Normal AD = [810, 0])

  Pathogenic variant well know in NSCLC

- Tumour Purity estimate 50%
- Tumour Burden 13.2 (Muts/Mb)
- No Clinically Relevant CNV identified but Chr 6 appears intact

KRAS: Clinic Database overview

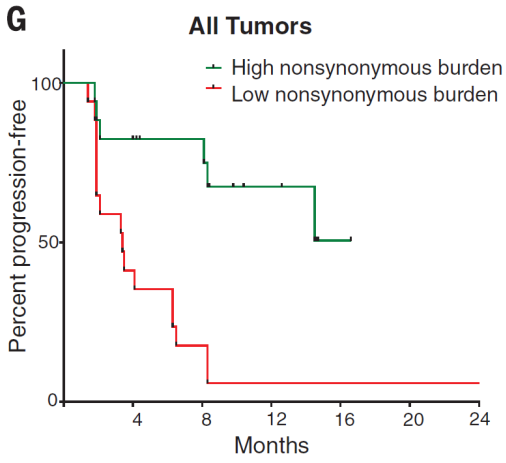
Level	ID	Disease	Type	Direction	Significance	Pubmed ID
B	227	Lung Cancer	Diagnostic	Supports	Positive	23014527
B	1142	Non-small Cell Lung Carcinoma	Predictive	Supports	Sensitivity	26125448
B	1217	Non-small Cell Lung Carcinoma	Prognostic	Supports	Poor Outcome	22247021
B	2257	Lung Cancer	Predictive	Supports	Resistance or Non-Response	17409929

- The prognostic implications for *KRAS* mutations vary between cancer types, but have been shown to be associated with poor outcome in colorectal cancer, non-small cell lung cancer, and others.
- *KRAS* poor outcome, resistance to Selumetinib
- CDK2/4 inhibitors ineffective for *KRAS* mutant alone :
- Results from the phase III JUNIPER trial, abemaciclib (Verzenio) failed to meet its primary endpoint of improving overall survival (OS) versus erlotinib (Tarceva) in patients with *KRAS*-mutated (2017)

Tumour Burden : Nonsynonymous mutation burden associated with clinical benefit of anti–PD-1 therapy

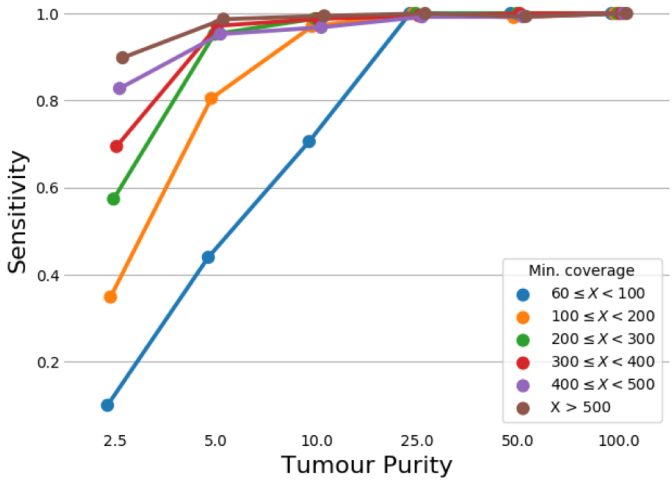
Fig 1: Based on exome sequencing:
(HR 0.19, 95% CI 0.08-0.47,
P = 0.0004)

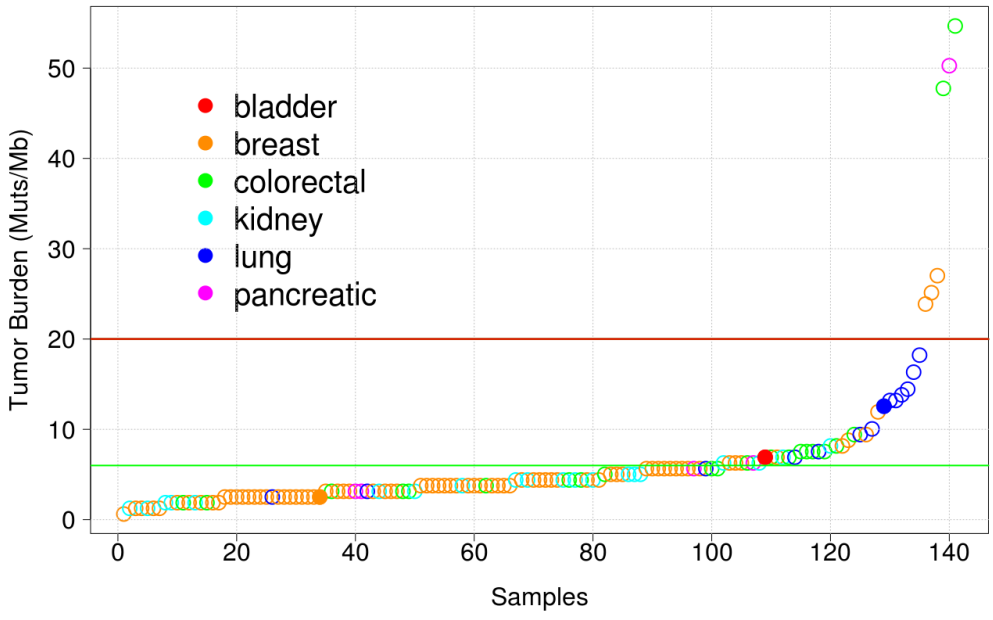
Results similar when use immunogenic burden.



Rizvi et al. Science. Science. 2015 Apr 3; 348(6230): 124–128.

Tumour Burden: Requires consideration for tumour purity and coverage





Review

- *KRAS* positive
- Tumour Burden in intermediate range