Asian Cancer Genomics: Urothelial Cancer and Fibroepithelial Tumors of Breast

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Disclosure

- I have nothing to disclose
Asian Cancers – A Vast Unmet Clinical Need

- Worldwide 14 million new cancer cases and 8.2 million cancer related deaths in 2012
- New cases expected to rise by 70% over the next 2 decades.
- >60% of new annual cases occur in Africa, Asia and Central and South America, accounting for 70% of the world’s cancer deaths.
- Top cancer killers are lung, liver and gastric cancers

Source: WHO
Asian Cancers and Group I Carcinogens

Gastric Cancer

Helicobacter pylori

Bile Duct Cancer (Cholangiocarcinoma)

Opisthorchis viverrini (Liver Fluke)

Urinary Tract Cancer

Aristolochia Plants (eg Birthwort)
Our Team Science

Urinary Tract Cancers

Gastric Cancer

Bile Duct Cancer

Breast Tumors

Our works result in >25 publications since 2011, including:

4x Nature Genetics
1x Journal of Clinical Oncology
1x Cancer Discovery
2x Science Translational Medicine
2x Gastroenterology
3x Gut ….
SingHealth/Duke-NUS Precision Medicine Institute (PRISM)
Integrating Genomics with Medical Phenotypes (The SPECTRA Database)

- Build high-quality genomic-medical database of Asian patient normality
- **Target 5,000** healthy volunteers
  - Whole genome sequencing
  - Serum Metabolomics and Immunophenotyping
  - Cardiac Imaging and EMR data
  - Prospective follow-up over 10 years
- SPECTRA will function as a reference database for disease studies conducted by specialty centers
Today’s Topics

1) Mutation Signatures Caused by Carcinogens
   - Aristolochic Acid and Urothelial Cancer

2) Disease Genes and Cancer Progression
   - Breast Fibroepithelial Tumors
Aristolochic Acid (AA)

~ The major active components within the *Aristolochia* species (also known as birthworts, pipevines or dutchman’s pipes).

~ The plants were commonly used in traditional herbal preparations for snake bites, cough syrups, slimming, arthritis, gout, inflammation etc.

~ Exposure to AA-containing traditional remedies is associated with high risk of nephrotoxicity and upper urinary tract urothelial cell carcinoma (UTUC), and is classified as Group I carcinogen.

Aristolochic acid associated upper urinary tract urothelial cell carcinoma (AA-UTUC)

- AA reactive intermediates bind to the amino groups of purine bases (A and G) to form DNA adducts.

- Mutations of TP53 have been previously identified and is the only gene known to be associated with AA-UTUC.

? Genome-wide changes in AA-UTUC?

AA-UTUCs Have Massive Mutational Loads
(Collaboration with Chang Gung University, Taiwan)

Whole Genome Sequencing

- Lung (Tobacco)
- Melanoma (UV)
- Urothelial (AA)

Exome Sequencing

  Nikolaev et al. (2012) Nature Genetics
AA-UTUC

AA-UTUC demonstrates a predominance of A:T to T:A transversion on non-transcribed strand.
AA Mutations Cause a Unique Sequence Pattern in the Genome ("Mutation Signature")

A-C/T–A-G-G is a sequence hotspot for AA Mutations

A>T Prevalence

\[ \text{XAX} \]

± 1 base context for A>T mutations

\[ \text{XCAGX} \]

± 2 base context for C[A>T]G

\[ \text{XTAGX} \]

± 2 base context for T[A>T]G
Purified AA is Sufficient to Induce the AA Mutation Signature *In Vitro*

HK-2 Cells
Proximal Tubule

AA Treatment (6 months)

Emergent Clones
AA alone is sufficient to recapitulate the AA-induced mutagenicity and nephrotoxicity
Detecting the AA “Mutation Fingerprint” in Other Asian Cancers

Genome-wide survey of recurrent HBV integration in hepatocellular carcinoma

Wing-Kin Sung1,4,16, Hancheng Zheng5,16, Shuyu Li6,16, Ronghua Chen7,16, Xiao Liu5,16, Yingrui Li5, Nikki P Lee1, Wah H Lee4, Pramila N Ariyaratne4, Chandana Tennakoon2,3, Fabianus H Mulawadi4, Kwong F Wong4,8–10, Angela M Liu1,8–10, Ronnie T Poon1, Sheung Tat Fan1, Kwong L Chan1, Zhuolin Gong5, Yujie Hu5, Zhao Lin5, Guan Wang5, Qinghui Zhang5, Thomas D Barber6, Wen-Chi Chou6, Amit Aggarwal6, Ke Hao7, Wei Zhou7, Chunsheng Zhang7, James Hardwick2,11, Carolyn Buser7, Jiangchun Xu12, Zhengyan Kan12, Hongyue Dai7, Mao Mao11,12, Christoph Reinhard6, Jun Wang5,13,14 & John M Luk1,8-10,15

88 Liver Cancer Whole Genomes (Hong Kong & Southern China)

Song et al., Science Transl Med, 2013
AA Signature as Part of the Cancer Signature Canon (International Cancer Genome Consortium, 2015)

Alexandrov et al., (2013) Nature

Aristolochic acid
AA-like Signatures in Multiple Tumor Types

HCC : Poon et al., (2013)  
CCA : Zou et al. (2015) Nat Comms  
UTUC : Poon et al. (2013), Hoang et al. (2013)  
RCC : Scelo et al. (2014) Nat Comms  
AA-UTUC

? Checkpoint Blockade
Immunotherapy
The top fifteen recurrent mutated genes in AA-UCC.

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* Denotes chromatin modifier
Scientists have been studying cancer for decades. Yet the more the disease is studied, the more researchers discover what they don’t know. In recent years, scientists around the globe have joined forces to tackle cancer, steadily making progress in unravelling its complexity.

According to GLOBOCAN 2012, a report by the World Health Organisation’s International Agency for Research on Cancer, there were 14.1 million new cancer cases, 8.2 million cancer deaths and 36.9 million people living with cancer (within five years of diagnosis) in 2012 worldwide.

Unless we understand this disease better, the WHO expects there to be 19.3 million new cancer cases per year by 2025.

One thing almost all cancers do is change our genetic blueprint, or genome. These changes disrupt biological pathways, causing cells to grow uncontrollably. This process is called a genetic change, and scientists have been mapping the genetic changes in each cancer.

To perform this浩瀚的 task researchers around the world are contributing efforts. In 2003, the International Cancer Genome Consortium (ICGC) was formed to deliver genomic data on more than 80 types of cancer in 10 years. With the global reach, the consortium has been one of the most ambitious biomedical research efforts since the Human Genome Project, a mammoth mission by the US Department of Energy and the National Institutes of Health that ran from 1990 to 2003.

Crucially, the ICGC makes its data rapidly and freely available.

China

Taiwan TV and Press

Also Highlighted by ScienceNOW (AAAS), Nature Genetics, Nature Reviews Urology, and The Scientist.
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   - Breast Fibroepithelial Tumors
Fibroepithelial Tumors of the Breast

- Distinct from breast carcinomas, separate category in WHO classification
- Breast tumors comprising epithelial and stromal compartments ("biphasic")

- Ranging from benign breast fibroadenomas to malignant phyllodes tumors
- Phyllodes tumors are rare in Western populations (1-2.5%), more common in Asia (7% in Singapore)
Breast Fibroadenoma ("Breast Lumps")

- Most common **benign** breast tumor in young women
- 1 in 10 women, aka Hundreds of millions worldwide
- 2-3 cm in size, hormone-dependent
- Admixture of epithelial and stromal cells
Breast Fibroadenomas Exhibit Highly Frequent
**MED12 Exon 2 Mutations**

58% of Fibroadenomas Have **MED12 exon 2 stromal mutations**

Lim et al., 2014 Nature Genetics
Fig. 1. MED complex involvement in cell transcriptional regulation mechanism. Biochemical and structural studies reveal that MED tightly binds Pol II enzyme thus starting pre-initiation complex formation. Transcriptional machinery is composed by several components: activator proteins, general transcription factors (TFⅠA, TFⅠB, TFⅠD, TBP, TFⅡE, TFⅡF, and TFⅡH), Pol II and cohesin protein. Altogether these factors are assembled in a DNA loop where the start and the end are represented by enhancer region/core promoter. Generally, when the kinase module binds to the Mediator core, Pol II is released and the transcription mechanism is impeded. The Mediator modules head, middle, tail, and kinase are coloured red, yellow, blue, and green, respectively. In the lower panel a scheme depicting the structure of mammalian chromatin is illustrated.
Next-Gen Sequencing Exposes Frequent MED12 Mutations and Actionable Therapeutic Targets in Phyllodes Tumors

Exome sequencing identifies highly recurrent MED12 somatic mutations in breast fibroadenoma

Mutational analysis of MED12 in fibroadenomas and phyllodes tumors of the breast by means of targeted next-generation sequencing

MED12 exon 2 mutations in phyllodes tumors of the breast

MED12 is frequently mutated in breast phyllodes tumours: a study of 112 cases

Distribution of MED12 Mutations in Fibroadenomas and Phyllodes Tumors of the Breast—Implications for Tumor Biology and Pathological Diagnosis

MED12 somatic mutations in fibroadenomas and phyllodes tumours of the breast

Frequent MED12 mutations in phyllodes tumours of the breast
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