Next-Generation Sequencing: Genomic Profiling Leads to the Advancement of Personalized Cancer Care

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Agenda

1. Formal abstract
2. Topic question & thesis
3. NGS background
4. Illumina sequencing
5. Importance for cancer patients
6. Pharmaceutical benefits
7. Conclusion
Formal Abstract

Next generation sequencing (NGS) technology has revolutionized genomic analysis for disease research thus yielding many groundbreaking clinical applications. Quick insight into the genome has allowed pharmaceutical advancements that have transformed cancer care. By identifying the unique oncogenes and biomarkers that are specifically driving a patient’s cancer, a personalized treatment plan can be developed. Each cancer is unique and by administering targeted therapies, that explicitly target oncogenic drivers within tumor cells, rather than general treatment methods such as radiation and chemotherapy, more successful patient outcomes are yielded. Targeted therapies are in no way the be all end all of cancer care, a problem often arises when resistance mutations develop, but this is combatted by creating second-line inhibitors and developing combination therapy treatment plans. Overall, by understanding oncogenes and biomarkers through genomic profiling conducted by next generation sequencing, pharmaceutical companies are able to create better treatment options for patients, such as targeted therapies. The goal of the following presentation is to demonstrate how next generation sequencing has become the greatest tool in the fight against cancer.
Topic question & thesis statement

• Question: By understanding oncogenes are we better equipped to develop cancer treatments?

• Thesis: Next-generation sequencing has become our greatest tool in the fight against cancer. Through genomic profiling, better treatment options have become available to patients.
What is Next-Generation Sequencing?

• Sequencing is the determination of amino acid order of a DNA or RNA fragment
  • Adenine, Guanine, Cytosine and Thymine

• Next-generation sequencing is a high-throughput methodology that enables rapid sequencing of DNA and RNA samples
  • DNA strands to be sequenced in parallel
  • Accelerating research compared to low-throughput methods

• Sanger sequencing took a decade, Illumina takes 2 days

• Most common use of NGS:
  • Cancer research
What does NGS tell us?

- NGS is used to analyze cancer specimens for the four main classes of genomic alterations
  - Base substitutions
  - Insertions and deletions
  - Copy number alterations
  - Rearrangements
Clinical significance

- A single comprehensive genomic profiling test can reveal alterations in DNA that drive cancer growth

- Example: Foundation Medicine Inc. 342 gene panel

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**Current Gene List**

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Source: Foundation Medicine
NGS & biomarkers

• Biomarkers are substances or processes that indicate the presence of cancer in the body
  • Biomarkers are often molecule’s secreted by a tumor or a specific response of the body to the presence of cancer

• Biomarker examples
  • Programmed death-ligand 1 (PD-L1)
  • Tumor mutational burden (TMB)
  • Microsatellite instability (MSI) status, which indicates deficient mismatch repair (dMMR)
NGS Workflow

1. Oncologist or pharmaceutical company sends patient sample
   • Blood, bone marrow or tumor biopsy
2. Sample reviewed by histologist & pathologist
3. DNA extraction
4. Library construction
   • Addition of adapters
5. Hybrid capture
   • Addition of bait set assay
6. Sequencing
7. Computational biology analysis
8. Medical data report generated
Illumina Sequencing Technologies
Illumina Sequencing

- Illumina produced 90% of all sequencing data
  - Reading of DNA has become more advanced
  - Created new tools to progress NGS field...
    - Kits
    - Reagents
    - Sequencers
Illumina Technologies: Library Construction

- Library construction is the preparation of samples
- Index added to allow for more than one patient sample to be tested at once
- Complementary regions are added to anchor DNA to flowcell

Source: Illumina
Illumina Technologies: Hybrid Capture

• Addition of bait set assay

• Target regions of genome that are relevant for companion diagnostics

• Delivers high discovery power and mutation resolution

Illumina Technologies: Cluster Generation

- Patient DNA travels down 8 lanes of flow cell
- DNA hybridizes to oligo
- Polymerase creates complementary synthetic DNA
- Natural strand washed away

Source: Illumina
Illumina Technologies: Bridge Amplification

- DNA strand folds over
- Adapter region binds to oligo
- Polymerase forms complimentary strand creating double stranded bridge
- Bridge denatures created 2 single strands
- DNA folds over starting process over until amplifies millions of times

Source: Illumina
Illumina Technologies: Sequencing

- Extension of first primer
- Fluorescently tagged nucleotides bind to form complimentary strand
- Clusters excited by light source
- Fluorescent signal emitted

Source: Illumina

https://digitalcommons.sacredheart.edu/acadfest/2020/all/80
Illumina Technologies: Reading Emissions & Data Analysis

- Measures emission from clusters
- Wavelength and intensity used to identify nucleotide base
- Reads are sorted, lined up and analyzed by computational biologists
- Medical data report generated for oncologist

Source: Illumina
Importance of Next-Generation Sequencing for cancer patients

- Each cancer is unique, each tumor varies at the molecular level
- Personalized treatment developed based on a tumor’s unique genomic profile

**Genomic information allows for...**

- **Targeted therapy**
  - Works by targeting the cancer's specific genes, proteins, or the tissue environment that contributes to cancer growth and survival

- **Immunotherapy**
  - Works by activating or suppressing the patient’s immune system

**Rather than...**

- **Radiation therapy**
  - uses beams of intense energy to kill cancer cells

- **Chemotherapy**
  - uses powerful chemicals to kill fast-growing cells in your body.
Targeted & immunotherapies informed by NGS

• Ipatasertib
  • Genentech utilized genomic profiling insights for drug development
  • Resulted in first line treatment of TNBC

• Tecentriq
  • Genentech utilized genomic profiling insights to determine patient eligibility
  • Resulted in major NSCLC treatment breakthrough

• Entrectinib
  • Genomic profiling insights apart of basket-study
  • Resulted in groundbreaking treatment for neuroblastomas
Ipatasertib General Overview

• Currently in phase III clinical trial, filing for FDA approval in 2020

• **First line treatment** for triple negative breast cancer (TNBC)
  • TNCB tumors are negative for estrogen receptors, progesterone receptors, and HER2 proteins

• IPAT samples are rushed in the lab due to 13 week prognosis
What is Ipatasertib?

- Ipatasertib is a small molecule inhibitor taken orally
- Targets the AKT pathway specifically 3 crucial biomarkers
  - PI3K (phosphoinositide 3-kinase)
  - PTEN (phosphatase & tensin homolog protein)
  - AKT (protein kinase B)
Healthy AKT pathway

- AKT pathway supports cell proliferation, growth, survival & metabolism
Cancerous Dysregulated AKT Pathway

- When PI3K, AKT or PTEN are mutated, it causes hyperactivation of PI3K and AKT.
- PTEN loss of function.

Source: Genentech
IPAT Targeting AKT inhibitor

- IPAT binds to the ATP binding site of AKT to inhibit its down stream signaling
- Reduce metastasis and activate apoptotic signaling
Tecentriq General Overview

- Drug created by Genentech
- Immunotherapy treatment
  - Anti-programmed death-ligand 1 (PD-L1) antibody
- FDA approved for Non Small-Cell Lung Cancer (NSCLC) & Bladder cancer
Importance For Patients

- In a clinical trial of 696 people with metastatic NSCLC, patients lived 4.5 months longer when treated with TECENTRIQ + other medicines compared to other treatments.

Median overall survival (OS)

14.7 months on bevacizumab + chemotherapy

19.2 months on TECENTRIQ + other medicines

Source: Genentech
Healthy Immune Response
Tecentriq PD-L1 Inhibitor

- T-cells are inhibited by PD-L1 signaling in cancer cells
- No immune response due to cancer cells appearing healthy
- Cancer cell survives, grows & reproduces

Source: Genentech

https://digitalcommons.sacredheart.edu/acadfest/2020/all/80
PD-L1 Inhibition by Tecentriq Visual

- Tecentriq binds to PD-L1 to inhibit it
Tumor Mutational Burden & immunotherapies

• Tumor mutational burden (TMB) is a biomarker, measuring the number of mutations within a tumor genome

• FMI & collaborators at UCSD examined the relationship between TMB and response to immunotherapy
  • Using retrospective analysis, the study found patients with high TMB had better outcomes compared to patients with lower TMB
  • TMB-high, tumor harbors a high number of mutations

• Significant improvement in overall survival was observed for high TMB patients compared to those with low-to-intermediate TMB
  • 12.8 months without their tumor growing, nearly four times longer than patients with non-high TMB

Source: FMI
Tecentriq & TMB

• Higher tumor mutation burden (TMB) correlates with response to Tecentriq

• TMB-high cells have more neoantigens to trigger more T-cells for immune response

• FMI looks at associations of TMB with Tecentriq response in clinical trials that have FMI TMB data
  • Linking correct patients to Tecentriq
Entrectinib Treatment Breakthrough

Significantly shrank NTRK fusion-positive neuroblastoma in pediatric patient

Source: FDA

https://digitalcommons.sacredheart.edu/acadfest/2020/all/80
Conclusion...

Next-generation sequencing has become our greatest tool in the fight against cancer. Through genomic profiling, better treatment options have become available to patients.
References


Foundation Medicine Inc. [internet]. Foundationmedicine.com


