Gender Innovation in Clinical Genetic Tests

2015. 08. 27

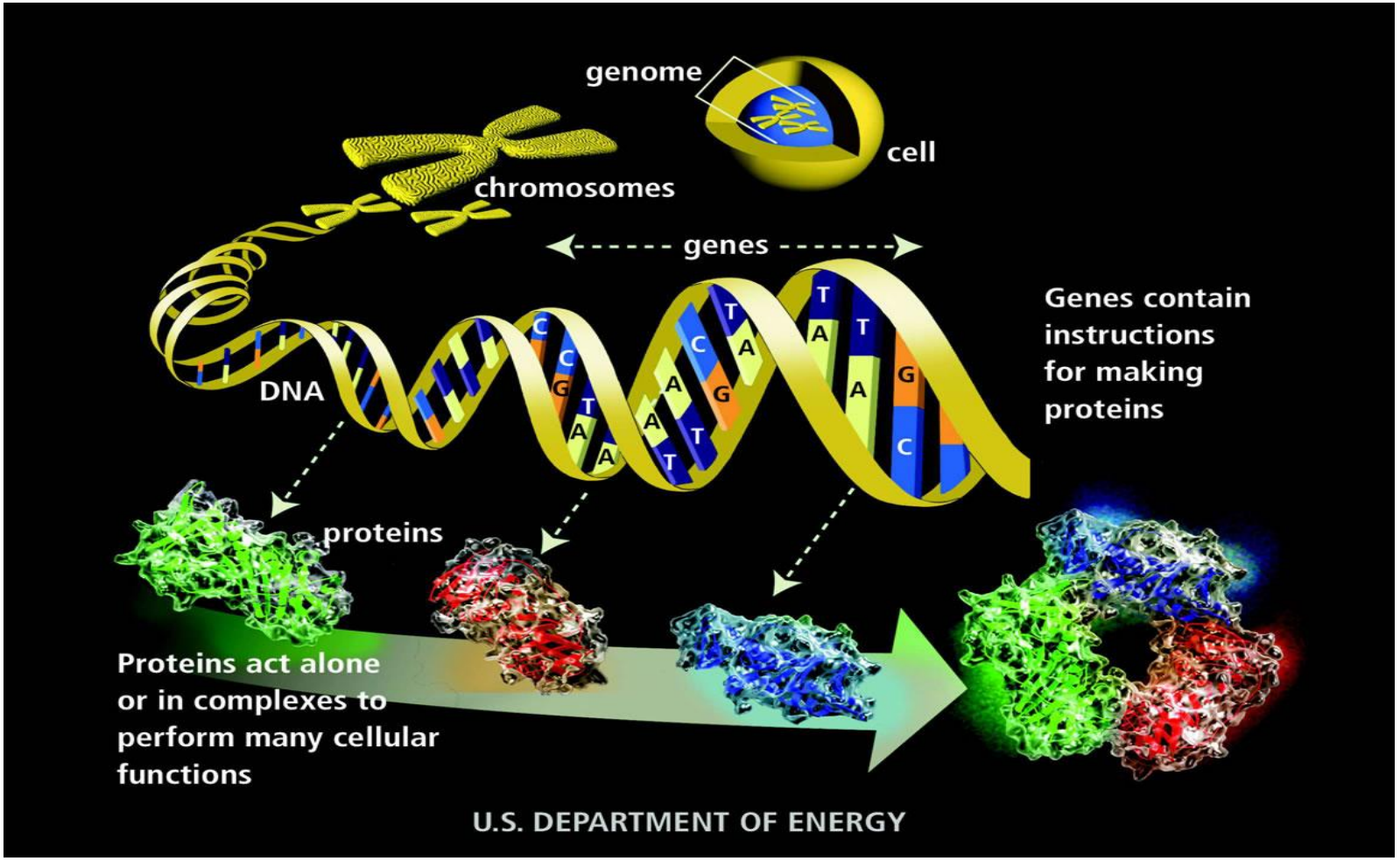
Chang Won Park

Macrogen

Contents

01 NGS Technologies & Instruments

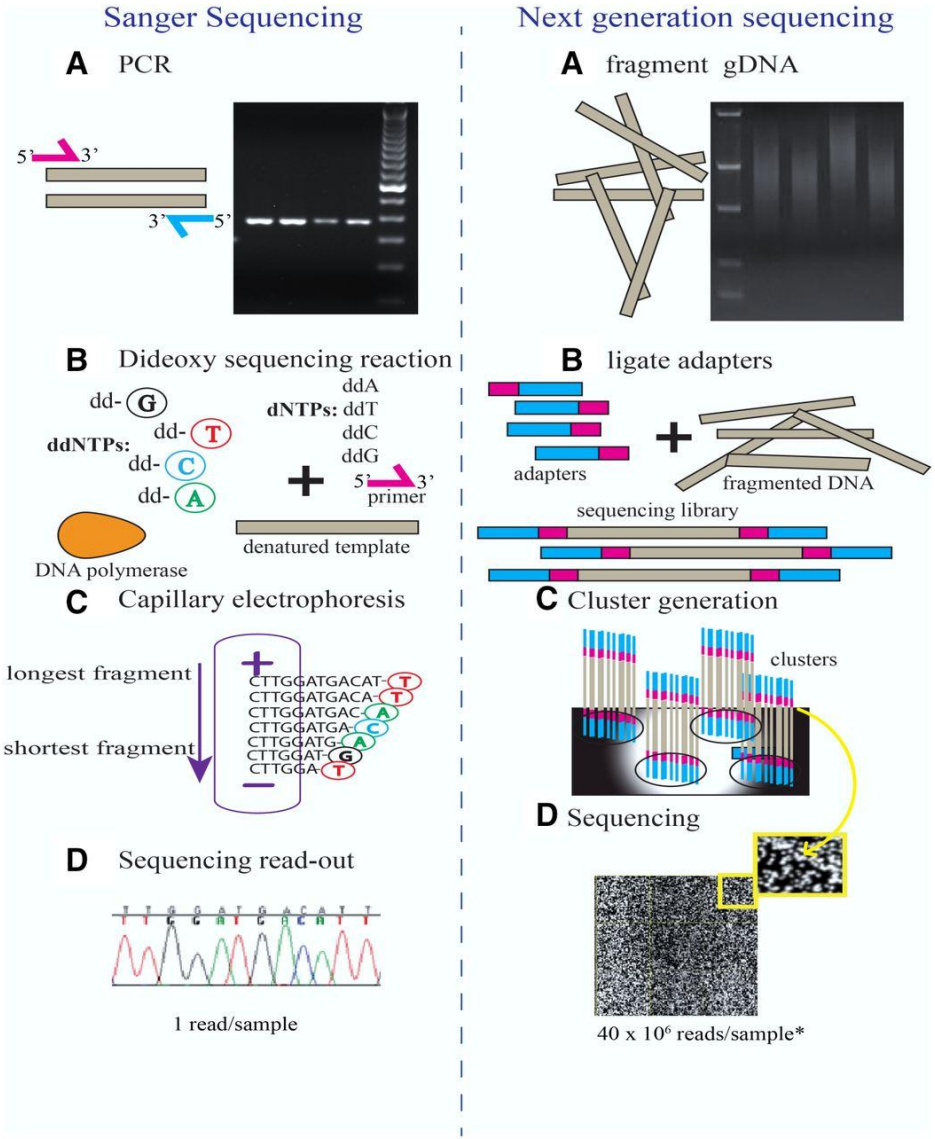
Chromosomes, Genes, DNA sequences



Note 1) A (Adenine), **T** (Thymine), **C** (Cytosine), **G** (Guanine)

Note 2) Source: https://www.ornl.gov/ornlhome/

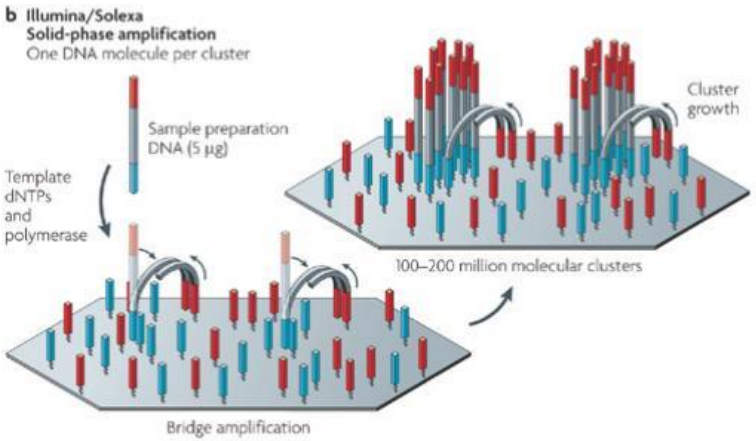
Sanger sequencing vs. NGS

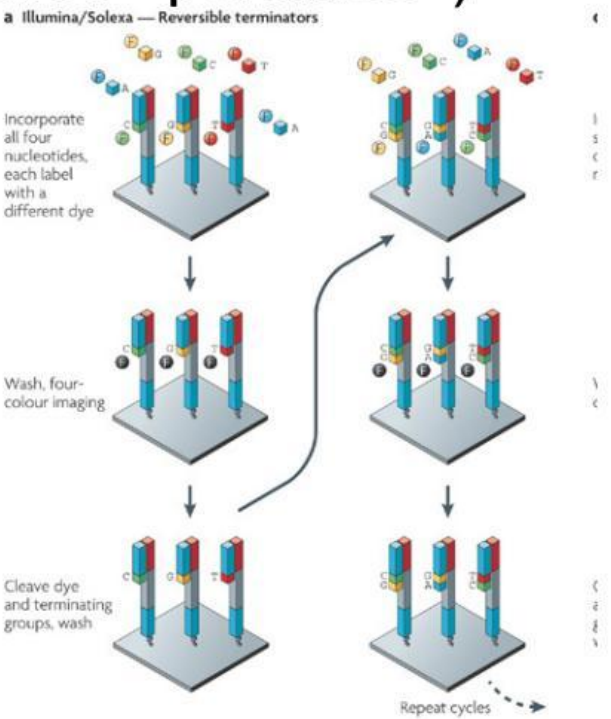


Sanger Dideoxy Sequencing Next Generation Sequencing

Next Generation Sequencing (NGS)

Illumina Colonies (called “polonies”)

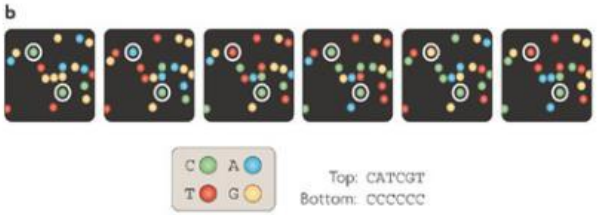




Each nucleotide has a dye with a different color

4-color fluorescent image of chip gathered after each chemical flows through

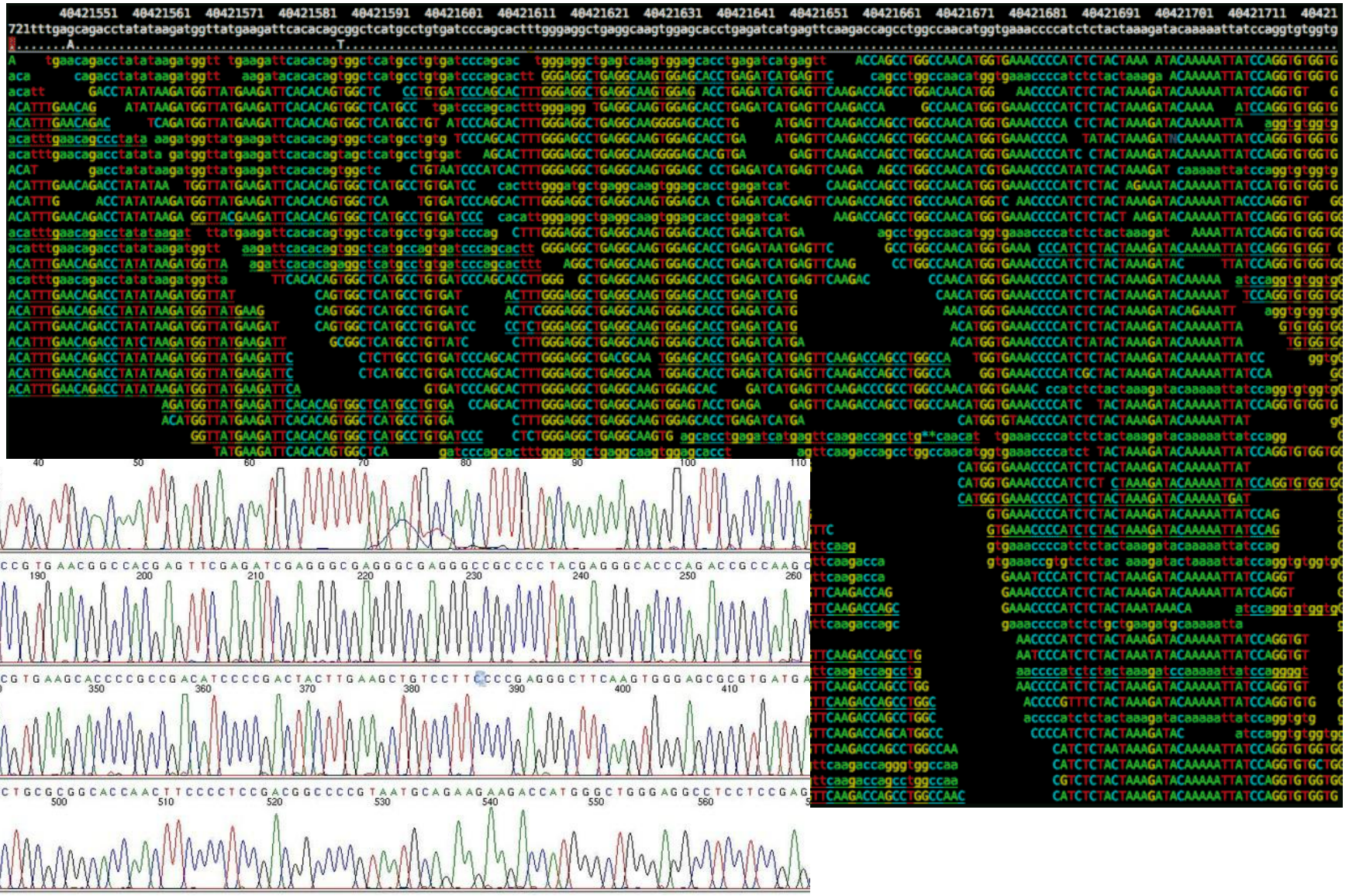
Register each image and follow color change of each colony to determine sequence



ECE/BioE 416
Lecture 24

16

Next Generation Sequencing



Next Generation Sequencing

Sequencing DNA:

Previously –

- To figure out the genome sequence itself
- Structure and function of genes
- To elucidate the difference between individuals
- To associate the phenotype and the genotype

Currently –

- Still for the goals listed above
- But shifting toward the clinical purposes – detection of pathogenic variants
- Prediction or diagnosis of diseases
- Prognosis or companion diagnostics
- Needs for analyzing multiple genes or genomic regions simultaneously – NGS technology required

NGS Instruments

Illumina HiSeq2000/2500

- Sequencing by synthesis using reversible flurorescent dye terminators using clonal single molecule array;
- 50 to 100 base reads length
- ~ 800 million high-quality reads per flowcell (~100 million/lane)
- up to 600 Gb output per run

Illumina MiSeq

- Small capacity system. PE 2x150cycles in 27hours.
- 18-20Gb by 2x300bp
- PE 2 x 300bp – error rate for read1 about 2%; read 2 about 4%.
- In preparation – Longer insert size possible 1.5kb

Ion Torrent PGM

- Three types of semiconductor chips:
  - 314 – 10Mb
  - 316 - 100Mb
  - 318 – 1Gb
- Read length depends on base composition 100-400bp
- The fastest sequencing system on the market.

NGS Instruments

HiSeq X Ten, HiSeq X Five

- Factory-Scale WGS System
- sequence 16 whole human genomes at standard 30x coverage
- $800 per genome
- generate 1.8 TB of sequence in under 3 days
- No multiplexing

HiSeq 3000, HiSeq 4000

- Daily throughput 200 ~ >400 GB

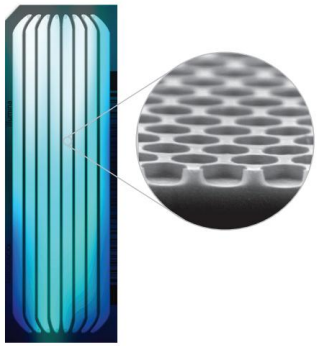
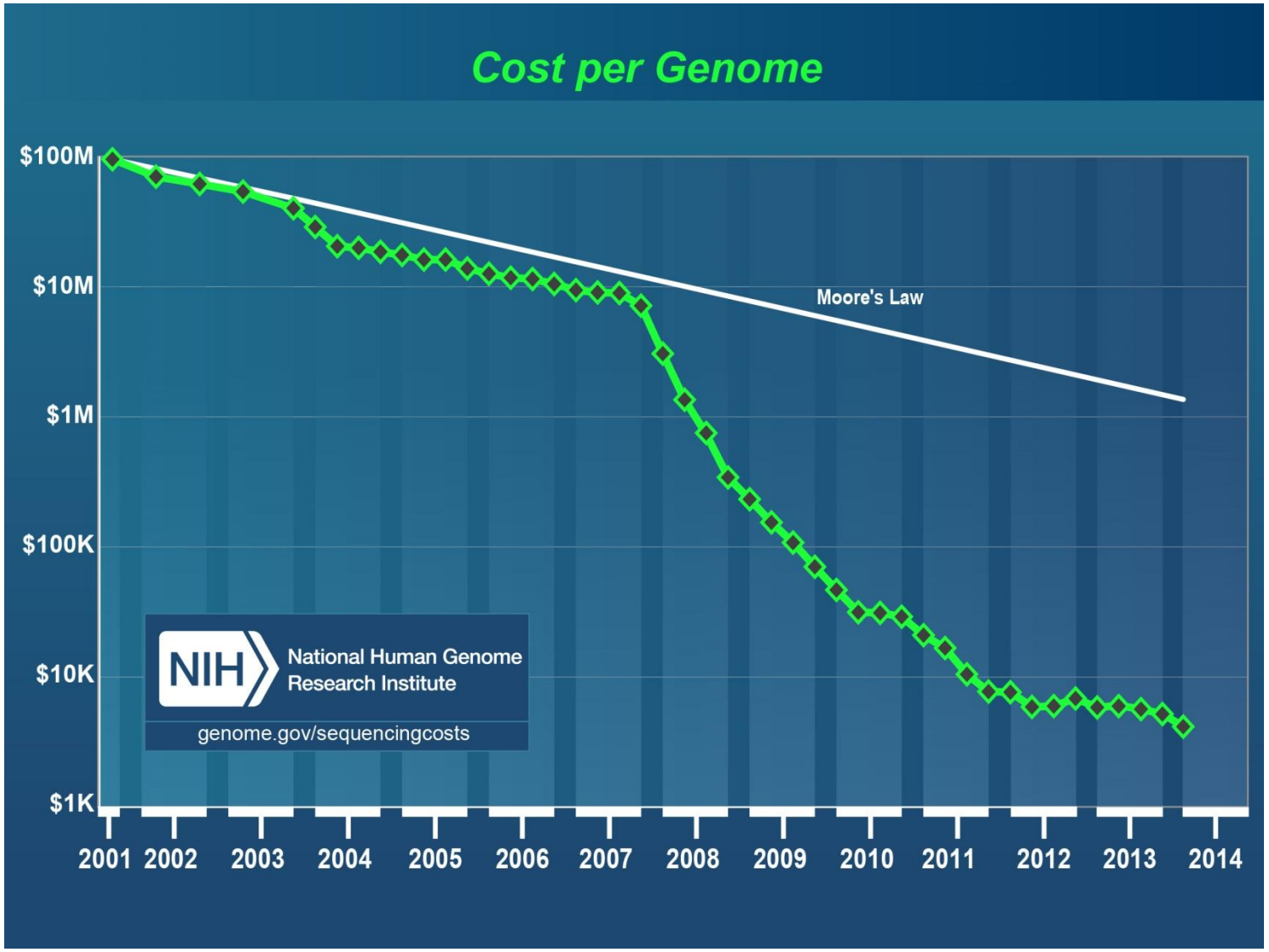


Figure 2: Advanced Patterned Flow Cell Design Enables Ultra-High Throughput. Patterned flow cells contain billions of nanowells at fixed locations, providing even cluster spacing and uniform feature size to deliver extremely high cluster density.

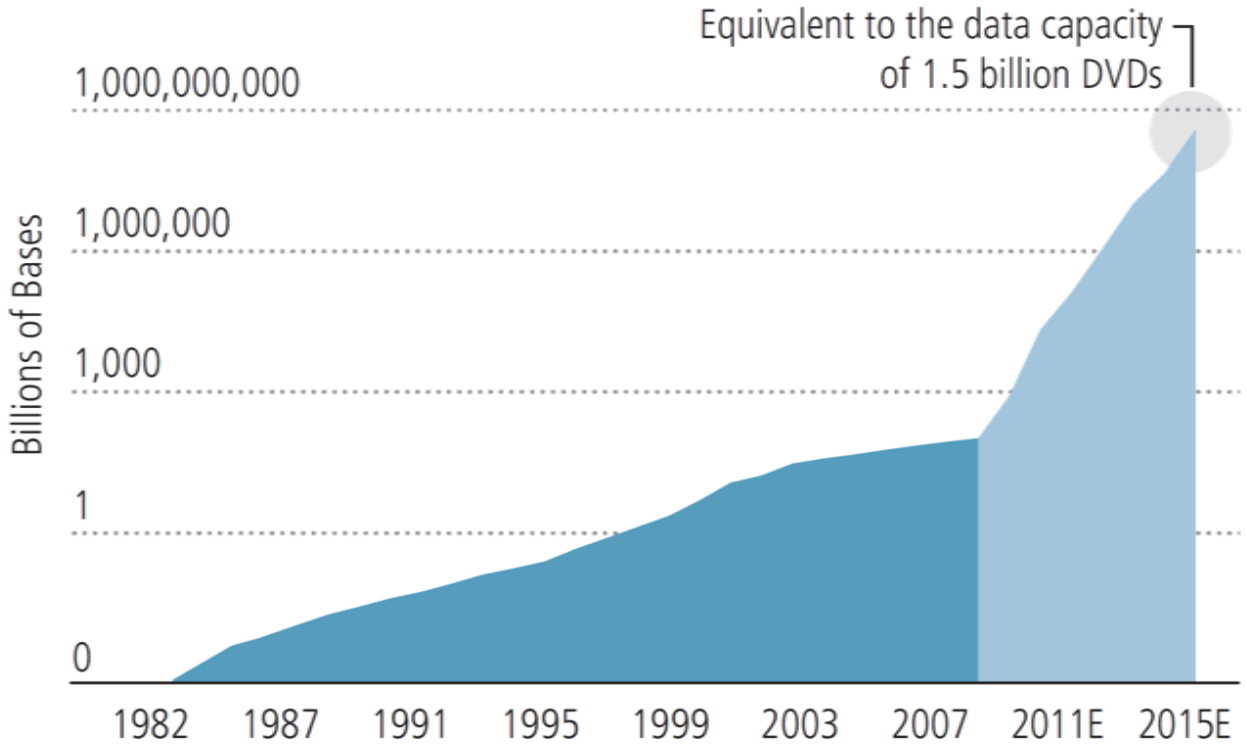
02 Clinical Genomics & Personalized Medicine

WGS with USD $1000



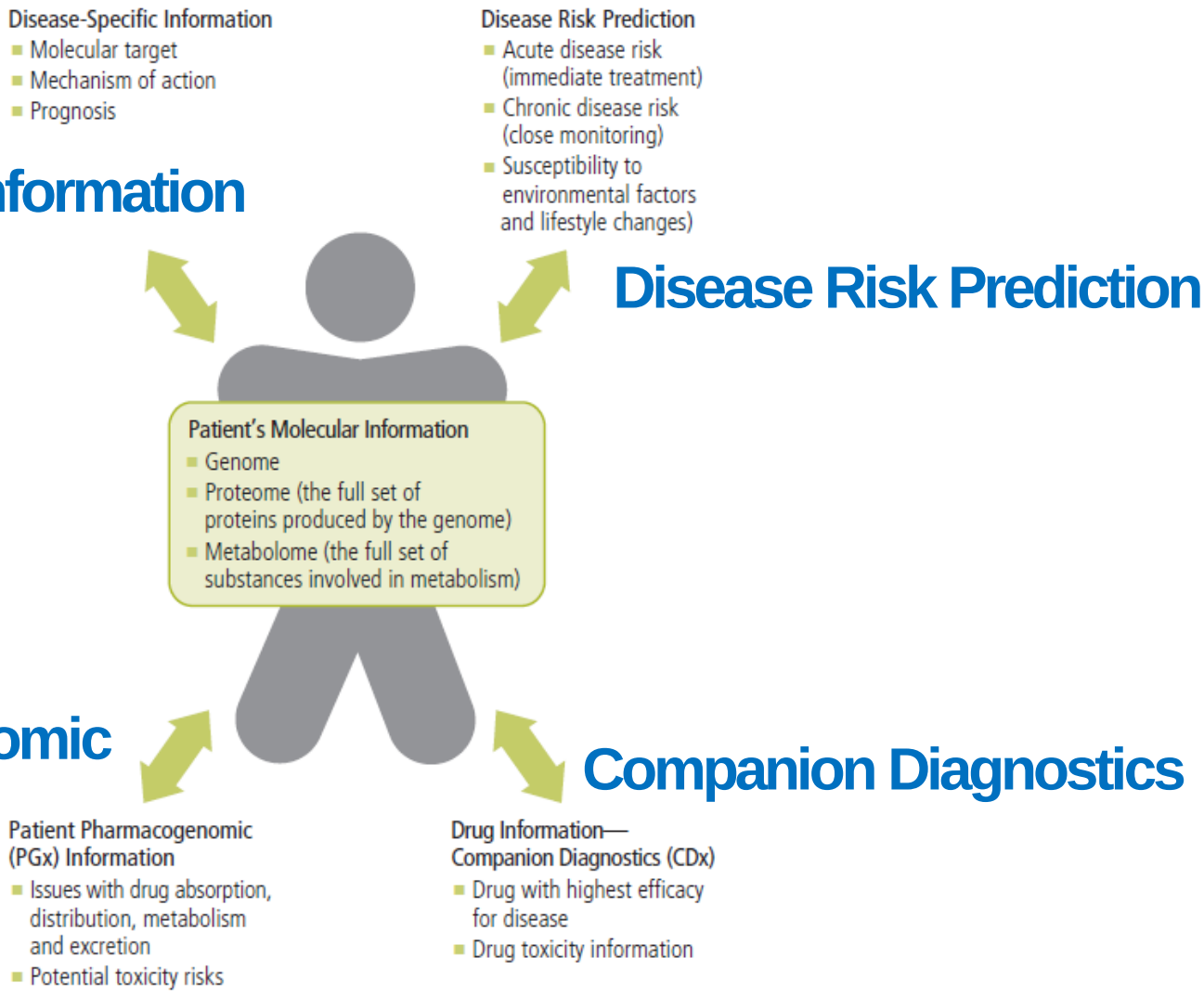
Big Big Data from DNA Sequencing

- DNA sequence data between 2007 and 2010 equivalent to DNA sequence accumulated for the past 24 years

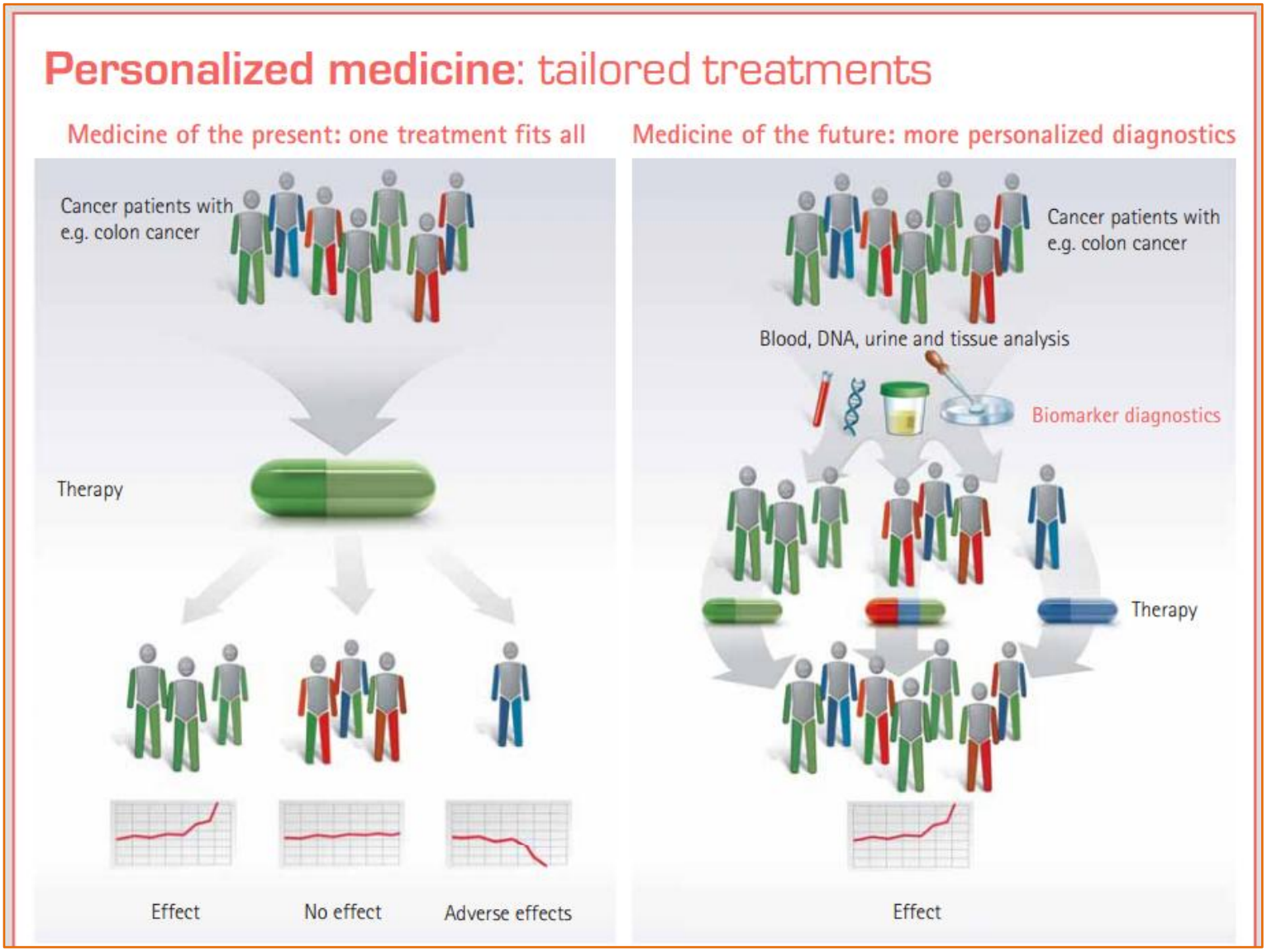


As of February 8, 2011; estimates are subject to change.
Source: Cisco Systems, GenBank and AllianceBernstein

Clinical Genomics in a New Paradigm



Clinical Genomics in a New Paradigm



Clinical Genomics in a New Paradigm

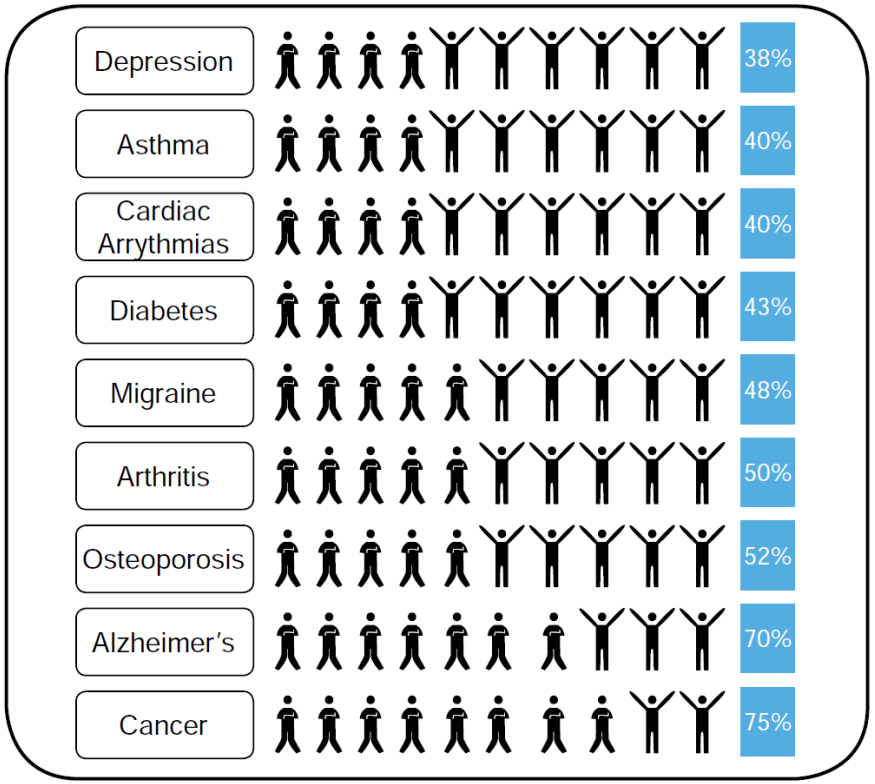
MAYO CLINIC
Mayo Medical Laboratories

CYP2D6

- CYP2D6 involved primarily or substantially in the metabolism of 25% of drugs
  - Important for metabolism of TCAs, SSRIs, and antipsychotics
- Highly polymorphic gene (*CYP2D6*)
- Phenotypes
  - Poor metabolizer (PM)
  - Intermediate metabolizer (IM)
  - Extensive metabolizer (EM)
  - Ultrarapid metabolizer (UM)
- Phenotype prevalence:
  - Caucasian: 5%-10% PM, 35% IM, 7% UM
  - African American and Asian: 1%-3% PM

Pharmacogenetics

- **"One size does not fit all."**



Companion Diagnostics – New Standard

Drug	Original Approval Date	Therapeutic Area	Biomarker
Arsenic Trioxide	2000	Oncology	PML/RARα
Tretinoin	1995		
Brentuximab Vedotin	2011	Oncology	CD30
Capecitabine	1998	Oncology	DPD
Fluorouracil	1998		
Cetuximab	2004	Oncology	EGFR; KRAS
Panitumumab	2006		
Crizotinib	2011	Oncology	ALK
Denileukin Diftitox	1999	Oncology	CD25/IL2
Exemestane	1999	Oncology	ER/PR
Fulvestrant	2002		
Letrozole	1997		
Imatinib	2003	Oncology	C-Kit, PDGFR, FIP1L1
Lapatinib	2007	Oncology	HER2
Pertuzumab	2012		
Trastuzumab	1998		
Everolimus	2009		
Nilotinib	2007	Oncology	Ph Chromosome
Dasatanib	2006		
Imatanib	2003		

Erbitux® EGFR Colorectal/Lung

Iressa® Gefitinib KRAS Colorectal/Lung

Tarceva® Erlotinib KRAS Colorectal/Lung

03 Gender Factors & BRCA1/2 test by Macrogen

Personalized Medicine & Gender Factors

- Personalized Medicine –
  - Age
  - Ethnicity
  - **Gender – Gender-specific disease susceptibility – Breast Cancer, Ovarian cancer**
  - Current physiological phenotypes
  - Life style
  - Working environment

BRCA1, BRCA2 Genes and Breast/Ovarian Cancer

- 3~5% of breast cancer caused by dominantly inherited mutations
- BRCA1/2 identified by linkage analysis (two decades ago), unstable, Alu (Large genes 81kb regions for BRCA1, 85kb for BRCA2, ave 10-15kb)
- Risk of breast cancer for a variant carrier : BRCA1 (65%) BRCA2 (45%) by age 70 (50-80% lifetime risk)
- Risk of ovarian cancer for a variant carrier: BRCA1 (39%) BRCA2 (11%) by age 70 (20-40% lifetime risk)
- Male BRCA1/2 carriers: 5.8-6.9% lifetime risk of breast cancer (still lower than ave risk of breast cancer in female)
- Male BRCA1/2 carriers: prostate cancer relative risk increased RR 3.4-7.3
- 50% chance to inherit the mutation to female progenies
- Gender difference

BRCA1/2 genetic test

BRCA1, BRCA2 Genes and Breast/Ovarian Cancer

- >3000 different variants in DB (public, www.umd.be/BRCA1(2))

- Preventive intervention
- Surgery – bilateral prophylactic mastectomy, bilateral prophylactic salpingo oophorectomy
- Enhanced screening – mammography, ultrasound
- Chemoprevention – vitamins, tamoxifen

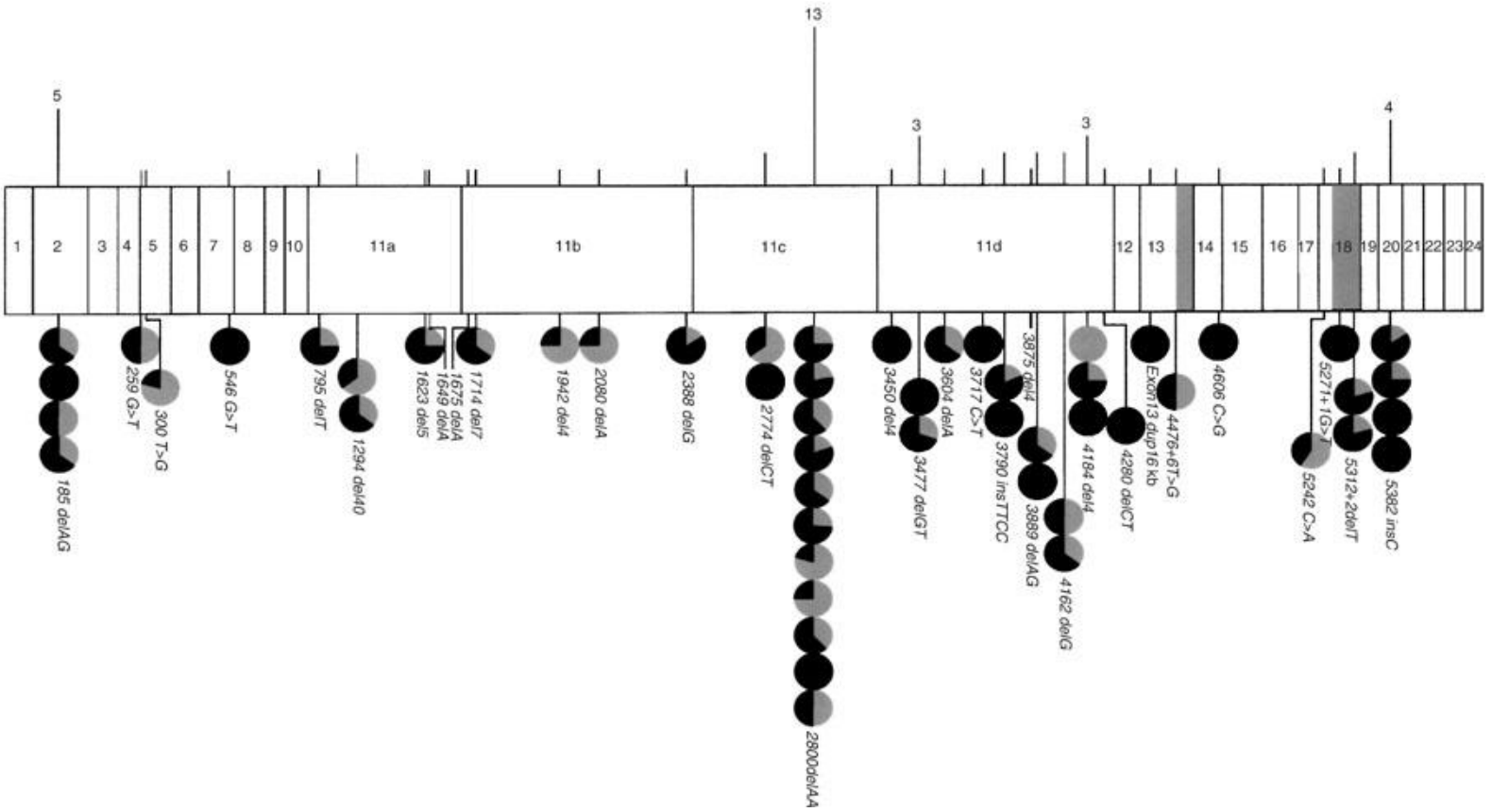
- Current price range of the test – USD 300-5000

BRCA1/2 genetic test

- Mother lost to ovarian cancer at the age of 56
- BRCA1 gene mutation
- 87% risk for breast cancer
- 50% risk of ovarian cancer
- Preventive double mastectomy
- Bilateral Salpingo-oophorectomy

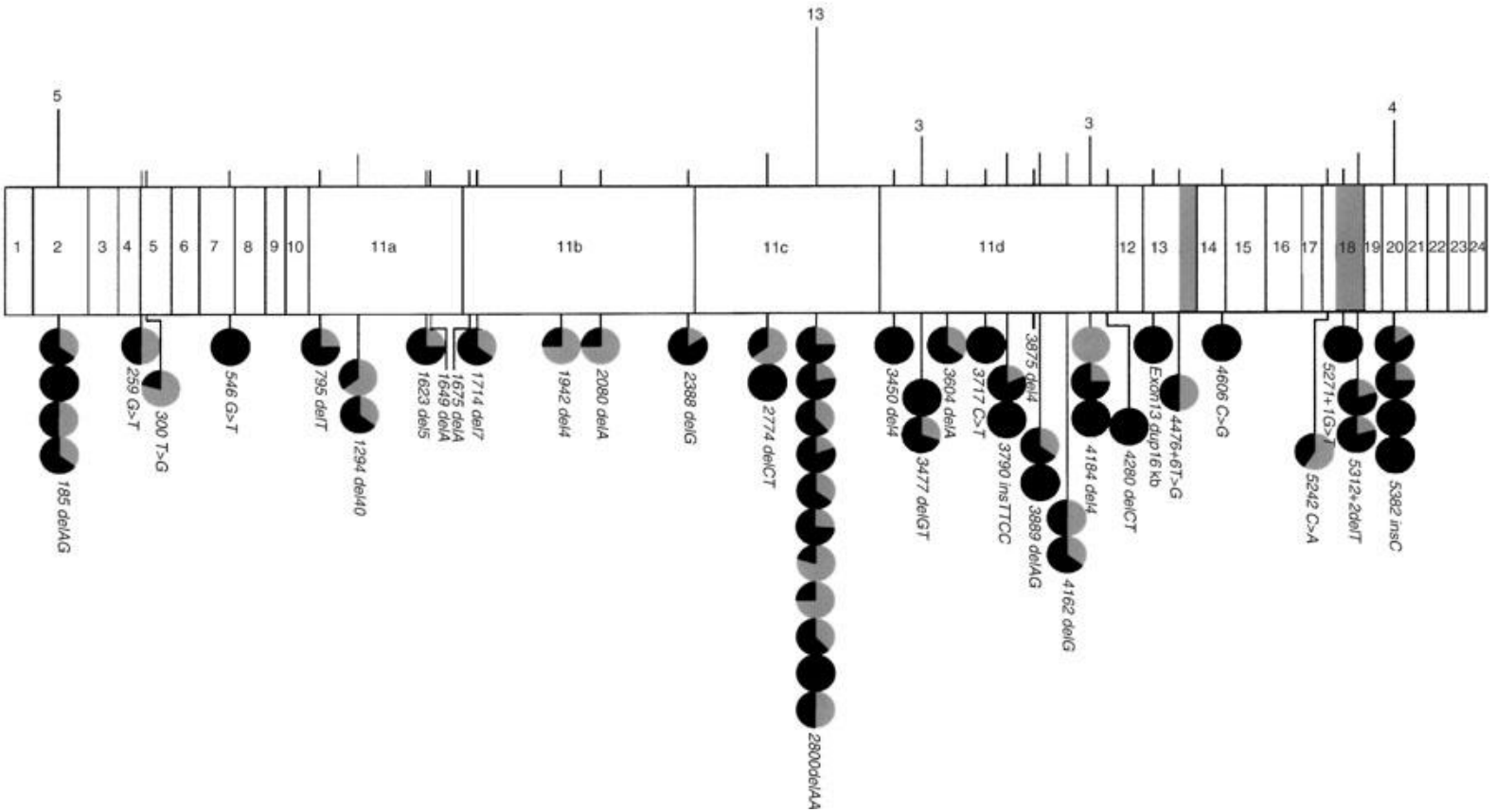
“I made a strong choice that in no way diminishes my femininity.”

BRCA1/2 genetic test - Macrogen



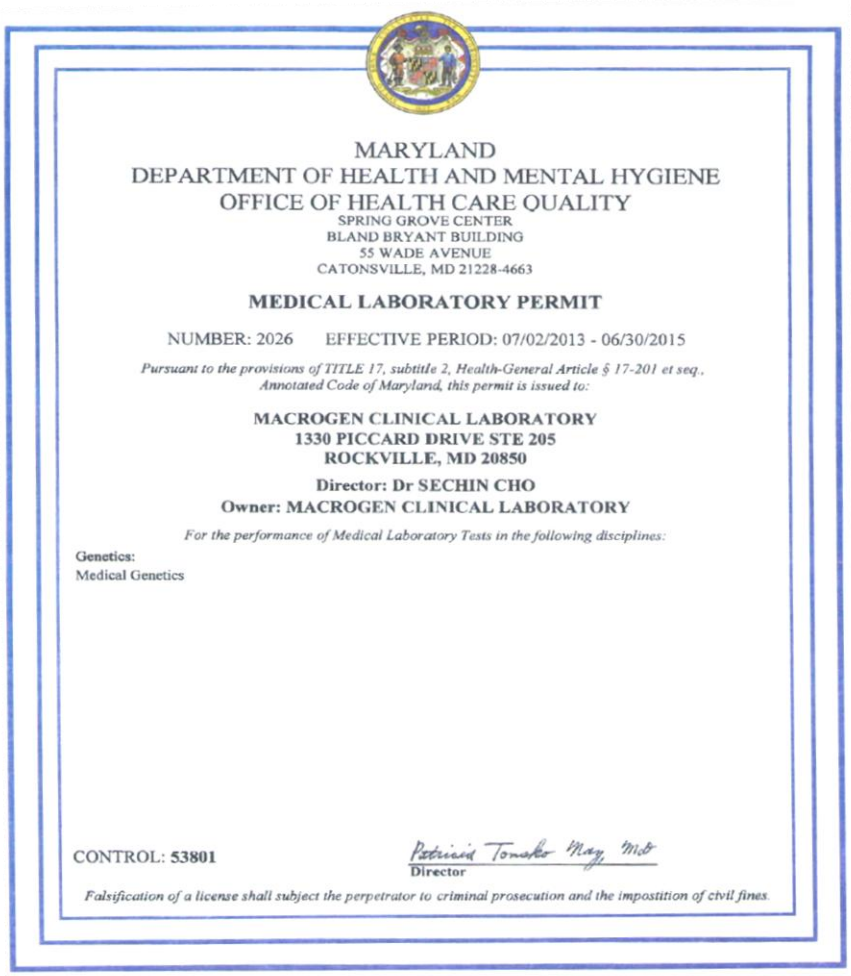
- ►Both traditional (Sanger sequencing) and NGS-panel base
- ►MLPA – CNV analysis complement
- ►R&D to reduce the cost for broad **affordability**

BRCA1/2 genetic test - Macrogen



- Aiming at below USD 100 for the service price
- For free - low income classes
- Affordability and Availability Worldwide

BRCA1/2 genetic test - Macrogen



MARYLAND
DEPARTMENT OF HEALTH AND MENTAL HYGIENE
OFFICE OF HEALTH CARE QUALITY
SPRING GROVE CENTER
BLAND BRYANT BUILDING
55 WADE AVENUE
CATONSVILLE, MD 21228-4663

MEDICAL LABORATORY PERMIT

NUMBER: 2026 EFFECTIVE PERIOD: 07/02/2013 - 06/30/2015

Pursuant to the provisions of TITLE 17, subtitle 2, Health-General Article § 17-201 et seq., Annotated Code of Maryland, this permit is issued to:

MACROGEN CLINICAL LABORATORY
1330 PICCARD DRIVE STE 205
ROCKVILLE, MD 20850

Director: Dr SECHIN CHO
Owner: MACROGEN CLINICAL LABORATORY

For the performance of Medical Laboratory Tests in the following disciplines:

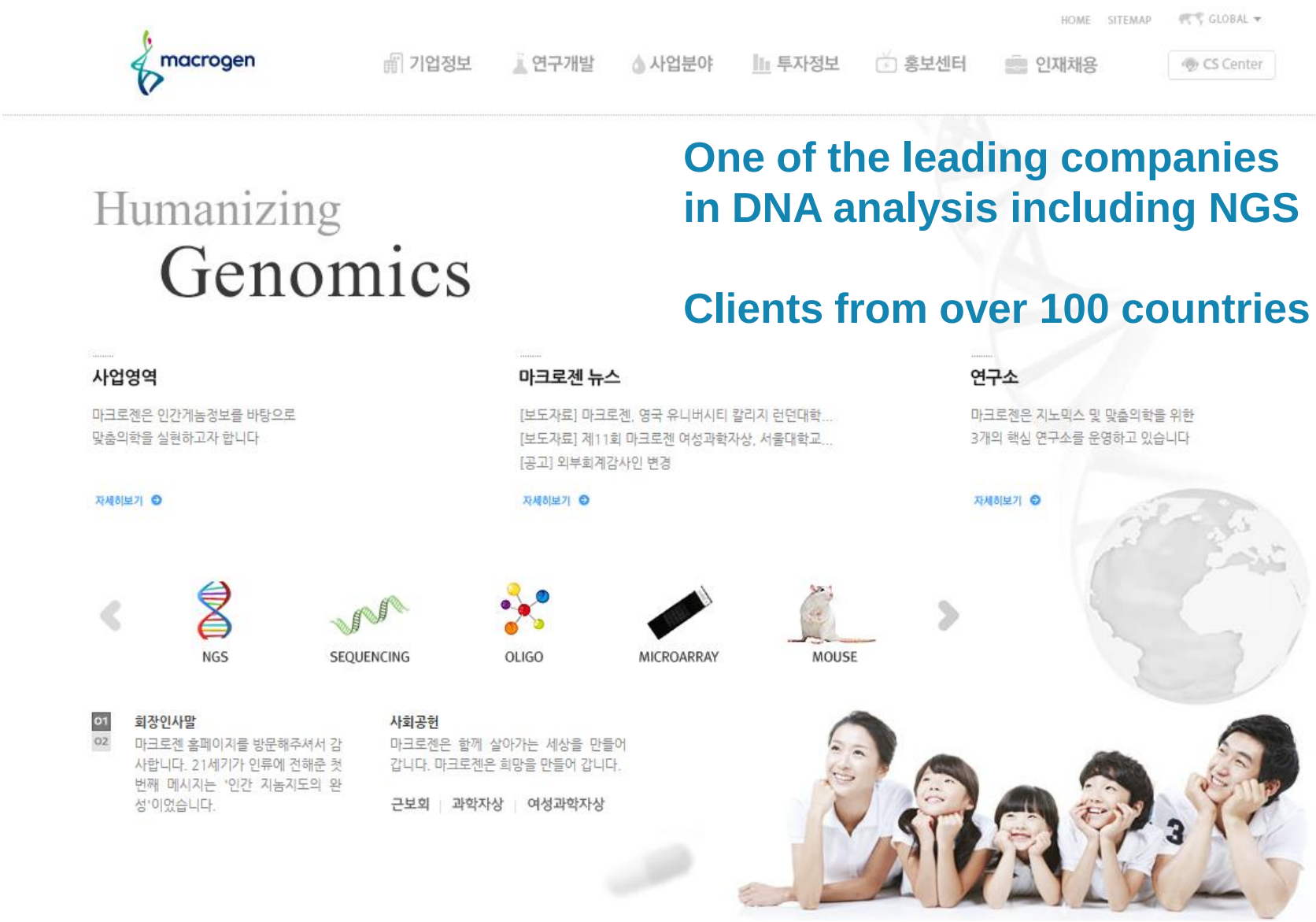
Genetics:
Medical Genetics

CONTROL: **53801**

Patricia Tomaka May, M.D.
Director

Falsification of a license shall subject the perpetrator to criminal prosecution and the imposition of civil fines.

- **CLIA-certified lab in the US**
- **Global service availability**



World Top-Class Sequencing Capacity

Next Generation Sequencing Instruments

over 18,000 human genomes per year at the price of $1000 per genome

Miseq/HiSeq 2500
HiSeq X/HiSeq 4000

IonPGM-Dx
IonProton/IonPGM

PacBio RSII

Over 60 units of the most advanced sequencing platforms

Thank You for Your Attention.

Humanizing Genomics
macrogen

Next Generation Sequencing

- Whole Genome Sequencing
- Exome Sequencing
- Transcriptome Sequencing
- Epigenome Sequencing
- Metagenome Sequencing

Capillary Electrophoresis Sequencing

- Standard Sequencing
- EZ-Seq Service
- Metagenome Sequencing
- Microsatellite Analysis
- Primer Walking

Microarray

- Illumina
- Agilent
- Affymetrix

Oligonucleotide Synthesis

- Oligo DNA
- Modified / Labeled Oligo

Genetically Engineered Mouse

- Trangenic Mouse
- Knock-out Mouse