# 생명정보개론 제3강

Week03 - 차세대 염기서열 해독법

2018.01.26

### O. Nic Volker 0 0 7

172번의 수술 800일 병원생활 치료의 효과는 닉에게 일시적일 뿐 다시 증상악화

The First Child Saved By DNA Sequencing



Matthew Herper, FORBES STAFF &

Since he was a toddler, six-year-old Nicholas

sequenced his DNA.

I cover science and medicine, and believe this is biology's century. FULL BIO



- 면역 결핍이 있음

확인이 불가한 심각한 질병임

기존치료법에 장기적 위험성이 있음

Volker's intestine had been dangerously inflamed, necessitating a hundred surgeries including the removal of his colon. No one knew the cause, but it seemed certain 치료의 성공은 근본적인 mechanism의 파악에 달려있음 that the boy was dying. In a desperate attempt to figure out what was wrong, doctors at the Medical College of Wisconsin did something desperate and unproven: they

적절한 면역 재건 치료가 논의되었지만 매우 위험

# 0. Nic Volker 이야기 Nic Volker논문 Introduction

Single gene testing대신

Exome sequencing을 통해

원인이 되는 돌연변이 동정이 필요

Exon의 총 집합체 (-ome) Proteome, Genome (Funding of will

Genetics in Madeire v. Volume 11, November 1, March 2011

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### MATERIALS AND METHODS

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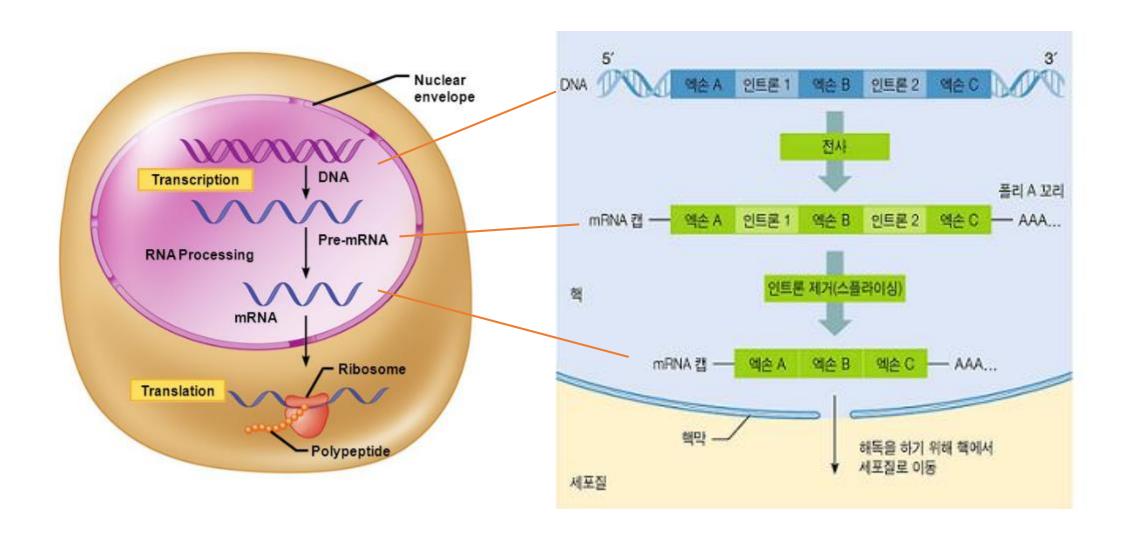
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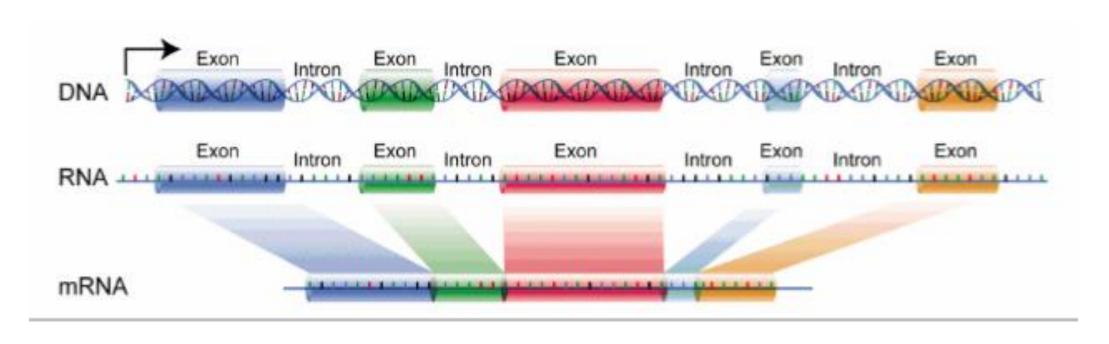
## 1. 엑솜시퀀싱의 개념어가기전기초설명



# 1. 엑솜시퀀싱의 개념어가기전기초설명 DNA to Protein 비디오



### 1. 엑솜시퀀싱의 개불어가기전기초설명



- · Intron이 훨씬 길다 (1000bp: 140bp)
- · Coding gene 약 20,000개 ·
- · Average Gene Exon 개수: 약 10개
- · Exome 개수: 약 200,000개

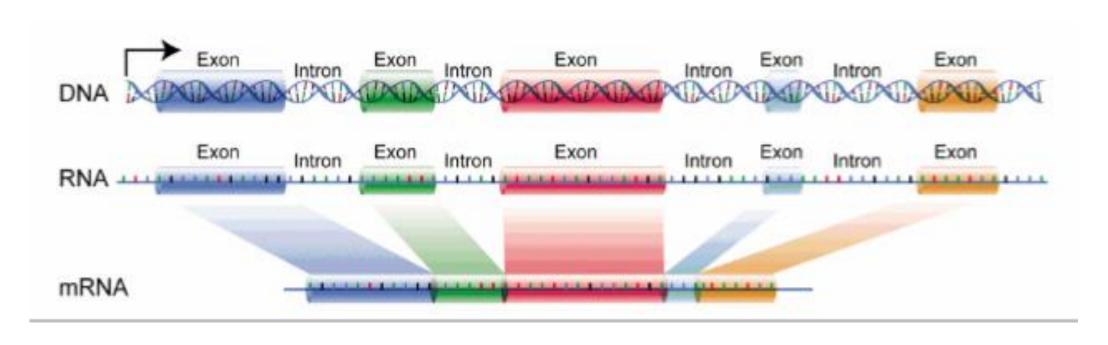
# Whole Genome Sequencing

VS

# Exome Sequencing

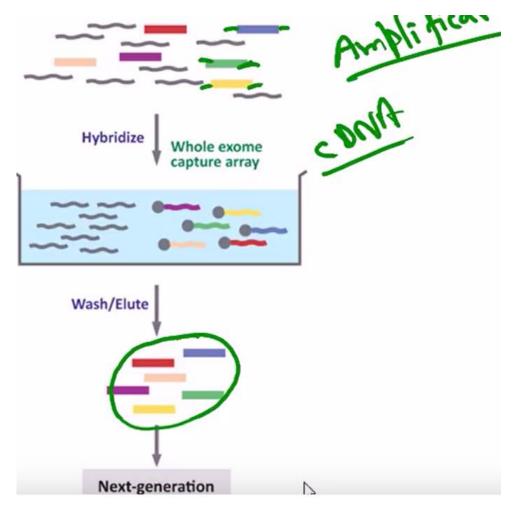
- -> Exon만 모으면 Compact한 데이터량 / 비용절감
- -> 특히 엑솜 영역에서도 아미노산 변화가 단백질 기능에 영향을 미치는 non-synonymous한 변이에 초점을 맞출 수 있음.

### **Exome Capture Process**



1. 어디가 Exon인지 아직 몰라! DNA를 무작위로 듬성듬성 자른다.

### **Exome Capture Process**



- 2) 잘라진 Nucleotide중 Exon만 스크리닝
- 하기 위해 c DNA Library를 이용한다.
- 3) Exon만 Capture 후 붙지 않는 Arrary 들은 씻어 목표하는 Array만 남게 한다.
- 4) NGS로 exome의 서열 밝힘

Next Generation Sequencong Roche 454 GS FLX



Pyrosequencing 원리

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## 2. Nic은 어떤 절차를 밟았을까?

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### MATERIALS AND METHODS

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## 2. Nic은 어떤 절차를 밟았을까?

### Nic Volker논문 Material/Method

- Exome Capture/서열 해독
- 닉의 genomic DNA 5µg서 엑손캡쳐
- 180,000 exons, 700 miRNA exons을 타깃으로
- 탐지가 가능한 양으로 증폭시킨 captured exon 조각을 Roche 454 GS FLX로 서열해독

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## 2. Nic은 어떤 절차를 밟았을까?

### Nic Volker논문 Material/Method

- Roche 454 gsMapper software tool

- Exome analysis
- <u>Mapping</u>: 해독된 DNA read를 human
  - genome reference sequence 에 정렬
- <u>Variant calling</u>: Reference 서열과 환자의 서열

사이에 차이가 발생한 서열 분석

(insertions, deletions and substitutions)

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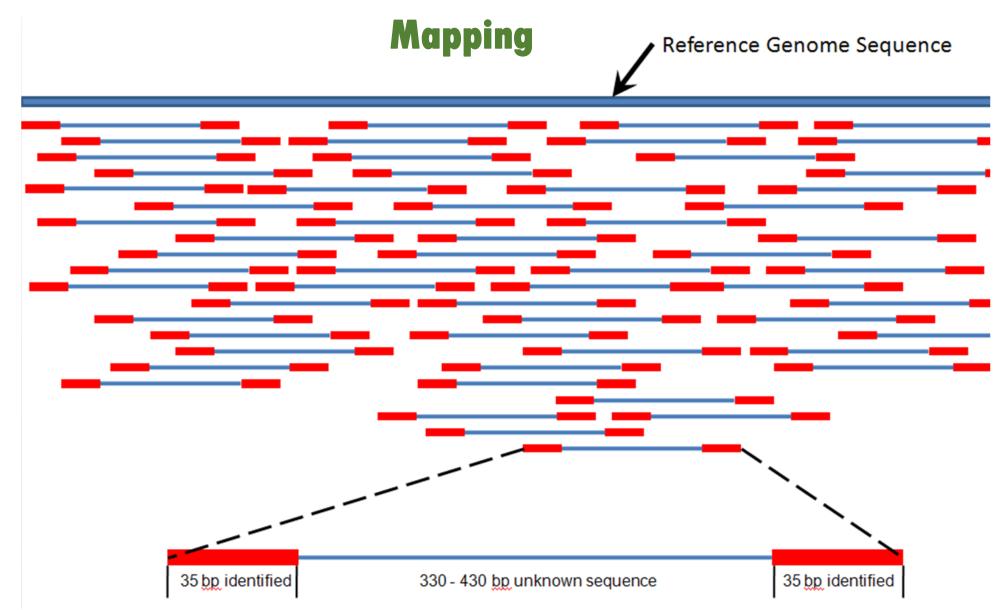
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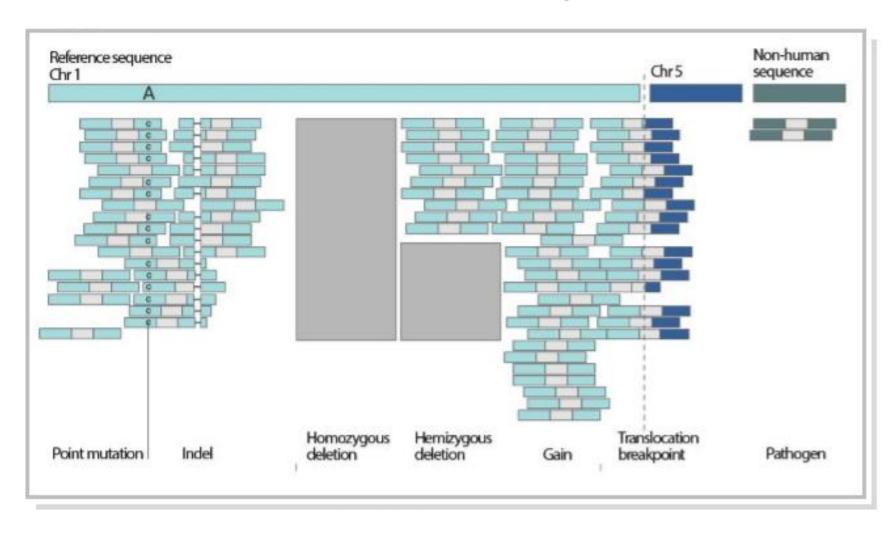
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# 3. 엑솜시퀀싱데이터처리

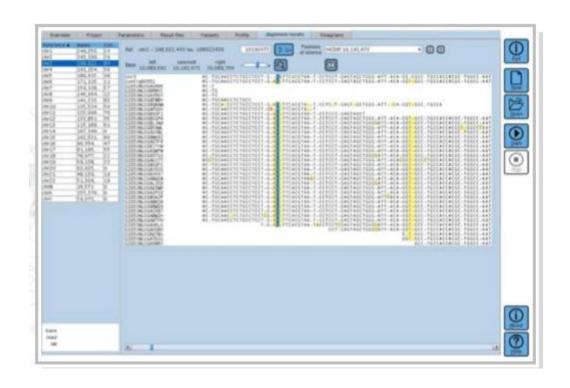


# 3. 엑솜시퀀싱데이터처리

### **Varient Calling**

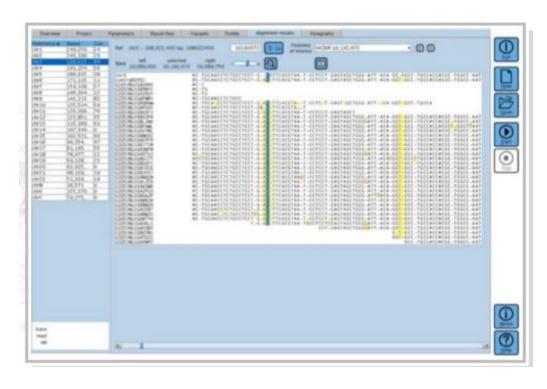


# 3. 엑솜시퀀싱데이터 처리 - gsMapper



· Roche 454 얻은 reads를 Human reference sequence에 매핑해주는 소프트웨어

# 3. 엑솜시퀀싱데이터 처리 - gsMapper



### 시퀀싱 에러

두 개 나와야지 에러? 세 개 나와야지 에러? 많은 데이터를 생산하기 때문에

### 통계적 접근이 필요함

소프트웨어에서는 오류의 Cut off정해서 판단함.

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