Next generation sequencing applications

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Basic idea:

- 1. Convert molecule to DNA
- 2. Apply 2nd generation sequencing

Exome sequencing

ATGGGAATTCACGAATTCCTAGACCTGCCCCGGAAACCTACCGCCGCG

DNA molecule

ATGGGAATTCACGAAŢTCCTAGACCTGCCCCGGAAACCTACCGCCGCG

Protein coding exon

ACCTGCCCGGAAACCTACC

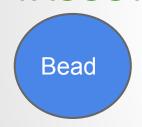
GCCGCG

ATGGGAATTCACGAATTCCTAG

Fragment DNA

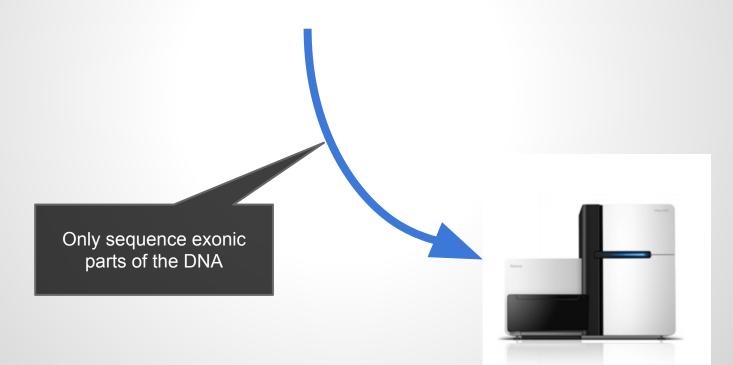
ATGGGAATTCACGAATTCCTAG

TACCCTTAAGTGCTTAAGGATC



Exonic DNA binds to complementary DNA on beads attached to a chip

ATGGGAATTCACGAATTCCTAG



RNA-seq

Fragmented RNA molecule

AUGGGAAUUCACGAAUUCCUAGAAAAAAA

AUGGGAAUUCACGAAUUCCUAGAAAAAAA

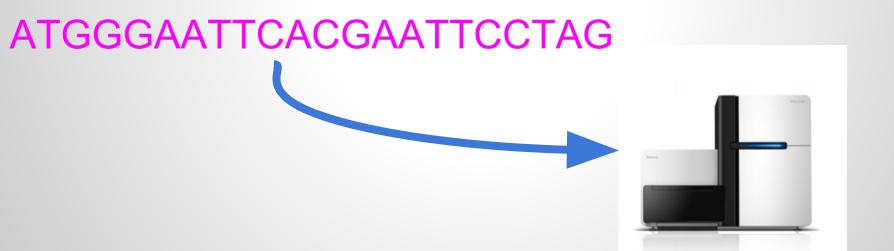
Capture mature RNA by poly(A) tail

AUGGGAAUUCACGAAUUCCUAGAAAAAAA

Reverse transcribe into complementary DNA (cDNA)

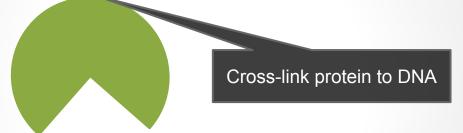
ATGGGAATTCACGAATTCCTAG

AUGGGAAUUCACGAAUUCCUAGAAAAAAA



Chip-Seq

GGAACCATGGGAATTCACGAATTCCTAACCATTA



CATTA GGAA CCATGGGAATTCACGAATTCCTAAC

Fragment DNA

CATTAG GGAA CCATGGGAATTCACGAATTCCTAAC

Antibody pulldown

CCATGGGAATTCACGAATTCCTAAC



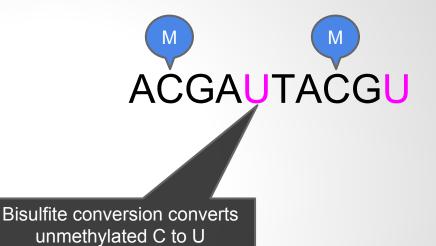
Bisulfite sequencing (methylation)





Split DNA into two aliquots (identical samples)





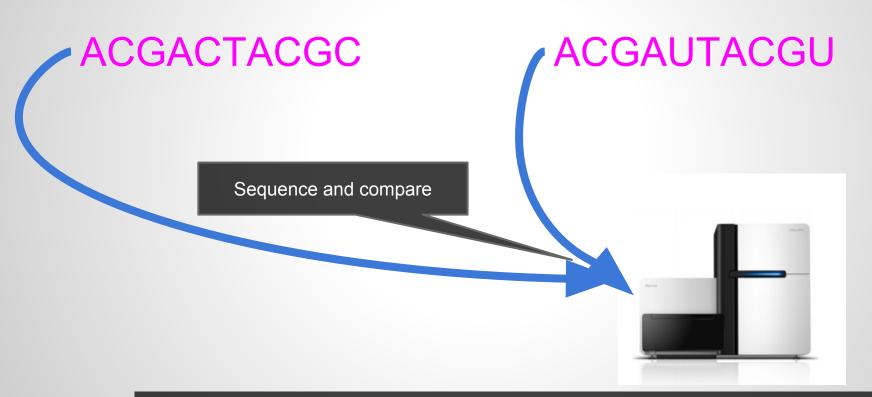


Image adapted from: http://www.atdbio.com/content/20/Sequencing-forensic-analysis-and-genetic-analysis