# Chapter 2

Key Concepts:

- **restriction endonucleases** (限制性内切核酸酶): usually called restriction enzymes (限制酶)
  - recognize a specific DNA sequence
  - cut, or restrict, at or near that sequence
- DNA sequencing:
  - **dideoxy sequencing** (双脱氧测序, 1977): first generation, low-throughout and long reads
  - **next generation of sequencing** (NGS, 2005): second-generation, high-throughout and short reads
  - **third-generation NGS systems** (recently): high-throughout and long reads
- **polymerase chain reaction** (PCR, 聚合酶链反应, Kary Mullis 1983):
  - amplicon (扩增子): a single template DNA molecule generates approximately $10^{12}$ amplicons
    - from an invisible target to a clearly visible fluorescent dye–stained (荧光染色) product
  - reverse transcriptase PCR (RT-PCR): allow for RNA templates to be converted to cDNA and then subject to regular PCR
  - applications:
    - identify individuals from residual DNA on crime scene evidence, e.g., a single hair.
    - amplify DNA from wellpreserved samples
- **DNA Microarrays** (微阵列): silicon chip substrates have hundreds of thousands and up to a million or more individual spots in an area about the size of a postage stamp. Two applications:
  - **gene expression profiling** (基因表达谱): measure expression level of mRNA quantitatively.
  - **genotyping** (基因分型): identify **single nucleotide polymorphisms (SNP, 单核苷酸多态性)**, which is a single nucleotide substitutions at a specific genetic locus.
- **Chromatin immunoprecipitation** (ChIP, 染色质免疫沉淀阵列): allows researchers to detect the presence of any protein of interest at a specific DNA sequence in vivo (在体内).
  - ChIP-seq: allow a researcher to obtain a genome-wide footprint of all of the binding sites of the protein of interest (e.g. transcript factor).
- **Transgenic** (转基因): An organism that gains new genetic information from the addition of foreign DNA
  - replace defective genes by functional genes
  - two powerful techniques by homologous recombination (同源重组):
    - delete (knock-out, 敲除)
    - replace (knock-in, 敲入)