

## First need a blast database

- n The sequence(s) you are going to blast to.
- n Create this using the command:  

```
makeblastdb -in <fasta file> -dbtype <nucl>
```
- q types: `nucl` or `prot`
- n If you include the “.fasta” file extension when the command, it will become part of the db name:  
q -in reference.fasta versus -in reference

## Then blast your seqs to your db

- n blastn Search a **nucleotide** database using a **nucleotide** query
- n blastp Search **protein** database using a **protein** query
- n blastx Search **protein** database using a **translated nucleotide** query
- n tblastn Search **translated nucleotide** database using a **protein** query
- n tblastx Search **translated nucleotide** database using a **translated nucleotide** query

## Then blast your seqs to your db

- n 

```
blastn -query <input.fasta> -db <dbname> -out <output file name> -outfmt 6
```

  - n `blastn`, `blastp`, `blastx`, `tblastn`, `tblastx`
- n Various output formats are available
  - q `-outfmt 6` provides tab-delimited text
    - n useful for batch processing of output
  - q you can also specify which variables are reported
    - n `-outfmt "6 qseqid sseqid pident length mismatch gapopen qstart qend sstart send evaluate bitscore btop"`
    - n `btop` produces a cigar-like field (details on the alignment)

## Can specify blast parameters

- n `-evalue 0.001` (only output alignments with values  $\leq$  this value)
  - q `-e 1e10-4`
  - q `-G` Cost to open gap [Integer]: default = 5 for nucleotides/ 11 for proteins
  - q `-E` Cost to extend gap [Integer]: default = 2 for nucleotides/ 1 for proteins
  - q `-q` Penalty for nucleotide mismatch [Integer]: default = -3
  - q `-r` reward for nucleotide match [Integer]: default = 1