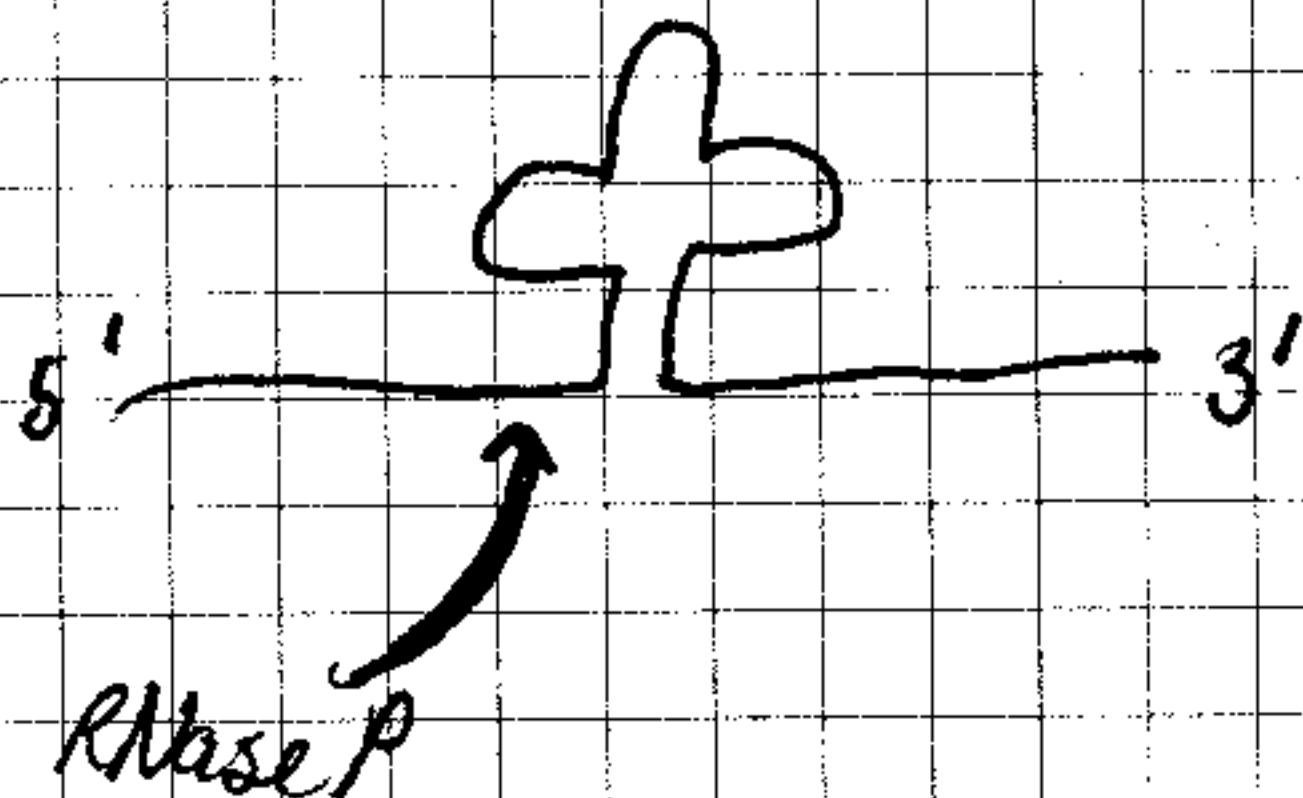


Diversity can be invisible.

- ① align homologous sequences
- ② count differences in each pair of sequences
- ③ calculate the evolutionary distance between the sequences
- ④ define the tree that best fits ALL distances

## Structural Analysis



- RNase P cuts 5' end of tRNA
  - v. similar in *E. coli* & *B. subtilis*
  - RNA and protein
  - RNA is catalytic unit

- what is structure of RNA subunit of RNase P

2°  
3°

- difficulties
  - ① short binding regions (~5 BP) yet only 4 diff. bases so very many possibilities

②

## 3 methods of working out 2<sup>o</sup> structure

①

② minimum energy

③ phylogeny

- compare E. coli & B. subtilis  
since NOT v. similar can  
pick a closer relative  
(based on RNA phylogeny)

- see where similar complements  
occur in closely related  
organisms.

After get general 2<sup>o</sup> structure

① deletion experiments

- moved "gaps" across molecule  
and activity remained.

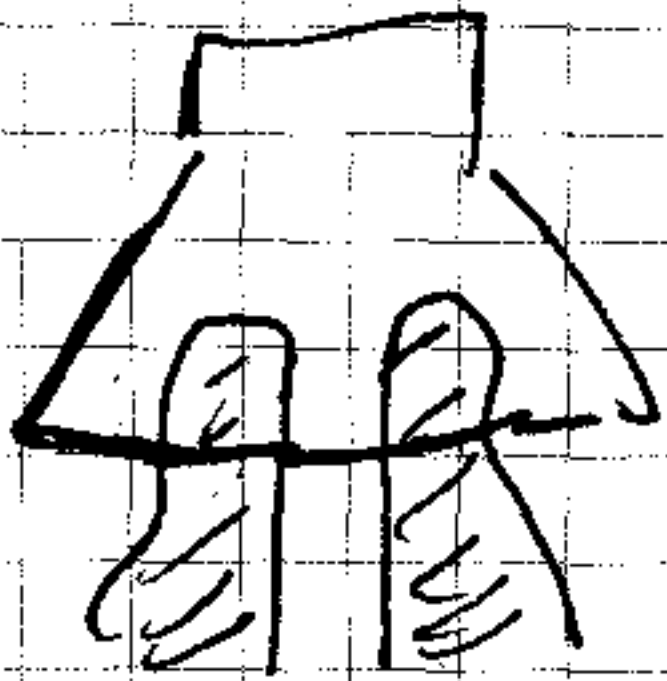
② see what deletions  
lead to damage

③ see what deletions  
do nothing

w/ conserved structure that should be  
the region of conserved function  
So can remove other sequences

# Upper Temperature Limits of life

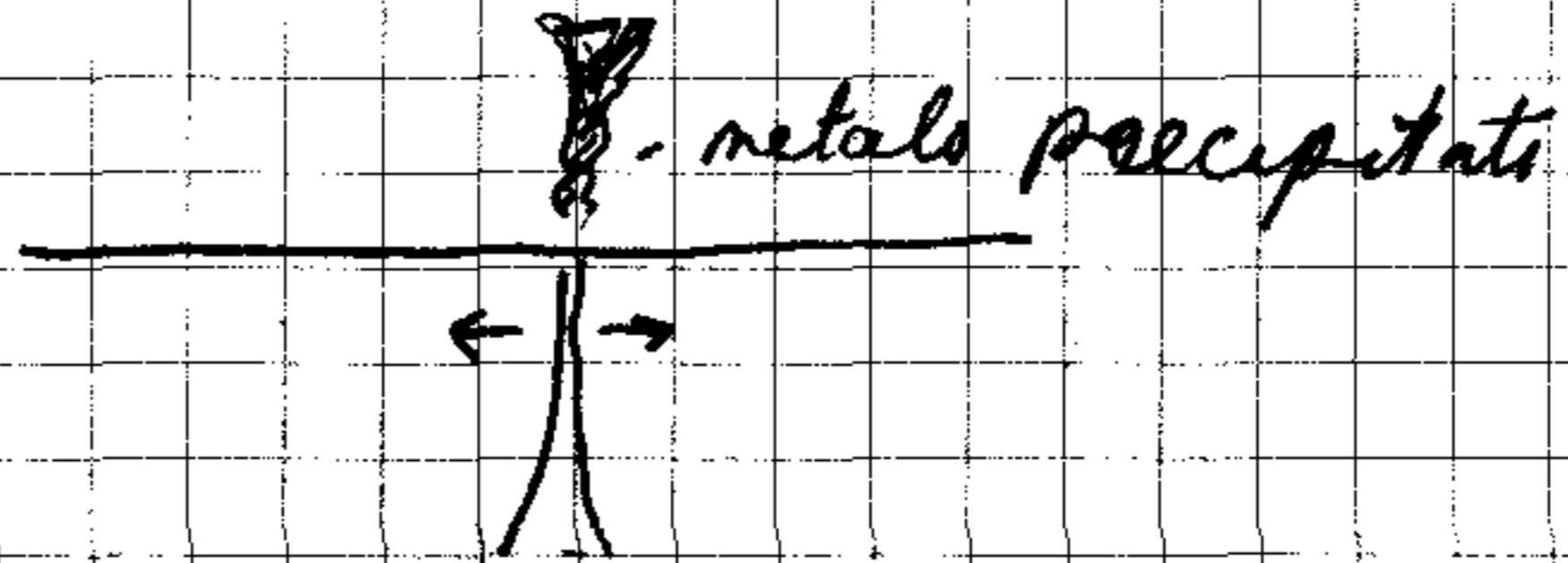
- ① put contact slides down
- ② vent caps



May be able to do phylogeny on  
single cells through oligonucleotide  
probes.

- ① probes for all life
- ② kingdom level probes
- ③ species " "

Growth rate determines # of ribosomes  
(or is it vice versa)



but

can't cultivate symbionts

- phylogeny may tell a great deal  
about general biochem of symbionts

# Methods of Characterizing Microbial World

## Problems

① cultivation difficult

So what use --

① Phylogeny

- phylogeny can help guide future research (such as was used for RNase P 2<sup>o</sup> structure.)

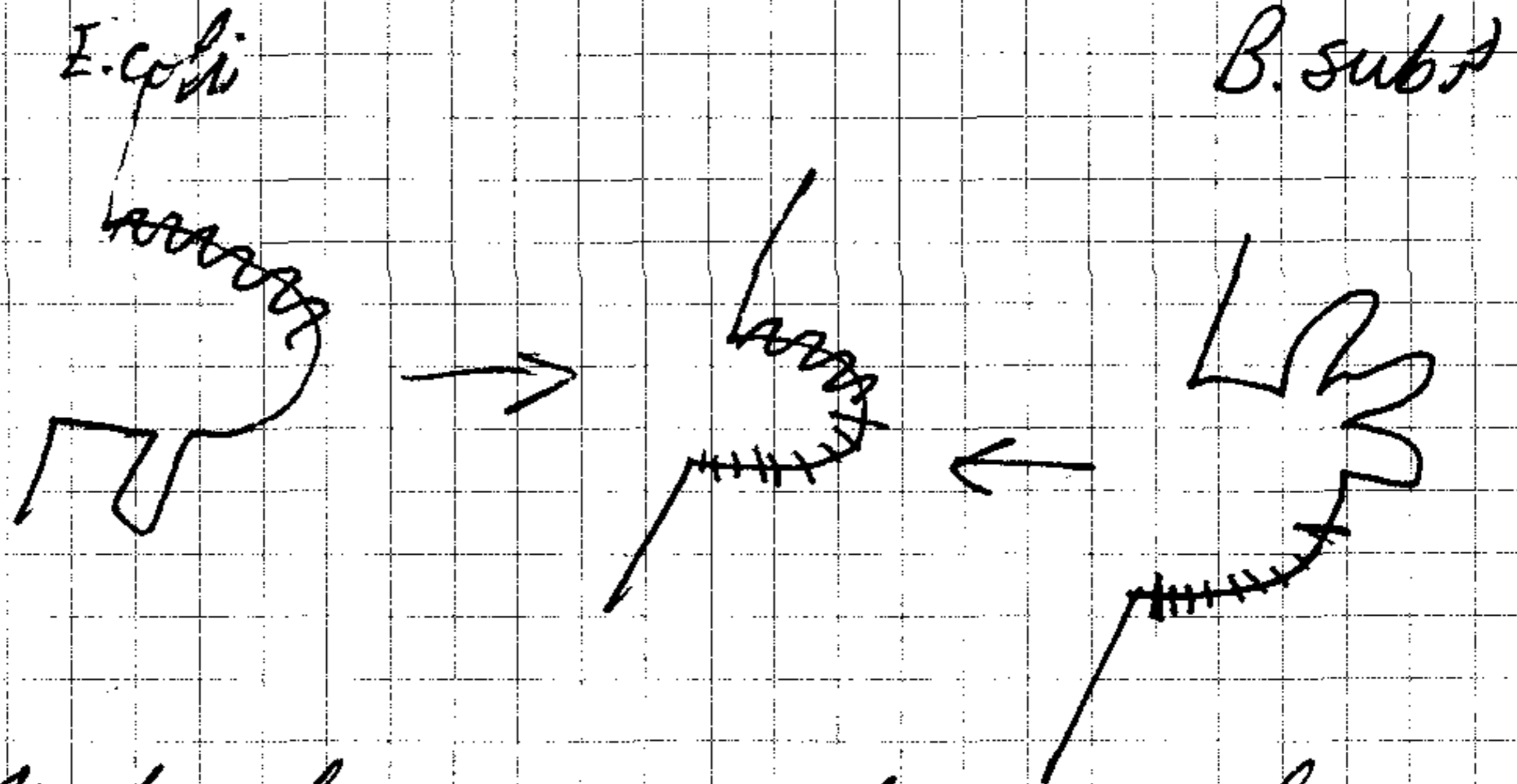
(DNA)

↓  
(Library)

↓  
16S clones

↓  
(Sequences)

but there is a problem because you don't know what to learn



make chimeric - combine simpler regions of two molecules.