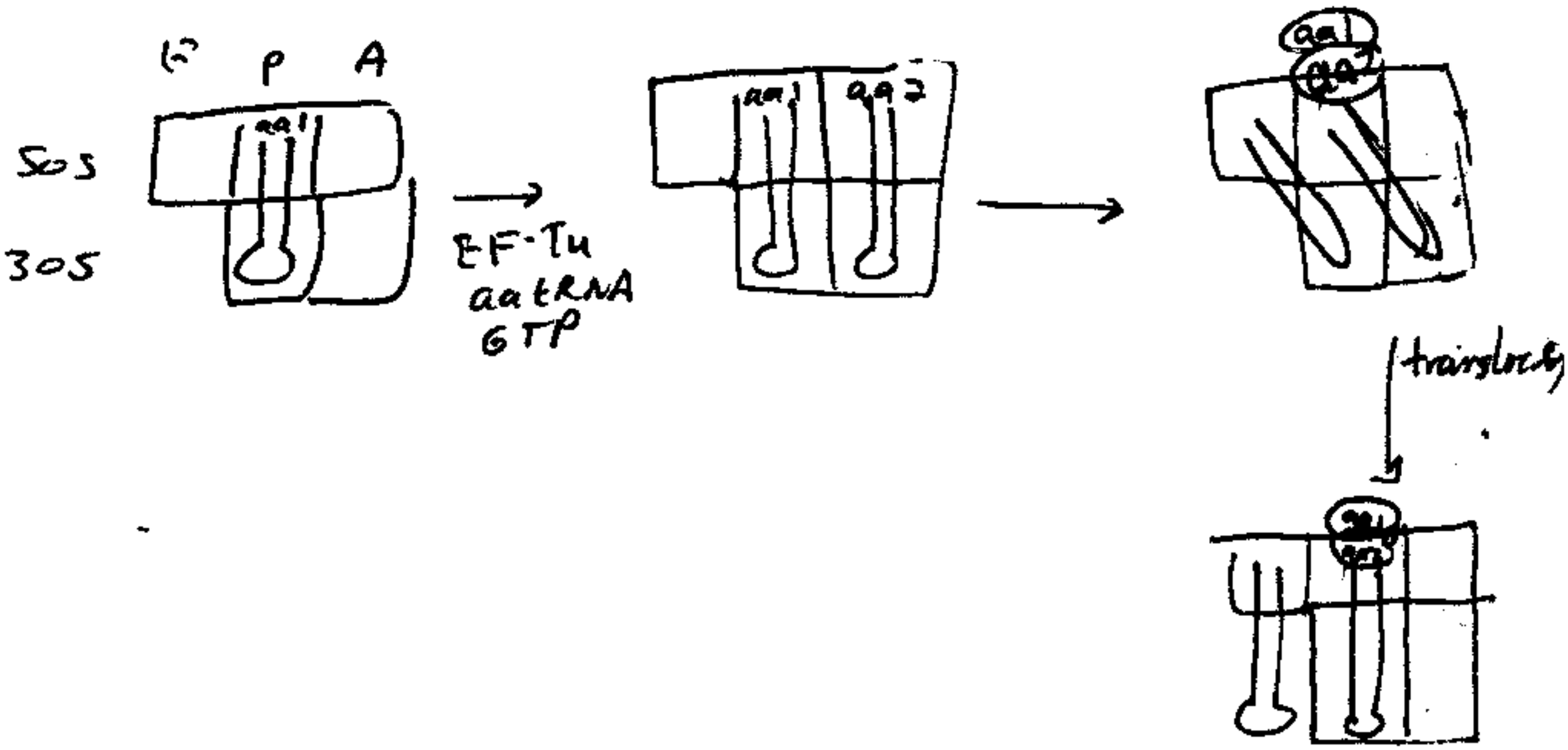
Rachel Green - Peptidyl Transferase 3·24·97

Ecoli - 70S ribosome

30S
16S rRNA (1542 nts)
21 proteins

50S
5S rRNA (~120 nts)
23S rRNA (2904) } peptidyl transferase activity here
34 proteins



Fragment Reaction (Monro 1967)

∴ 50S catalytic

50S + CCAcca-Fmet + puromycin

↓ Mg^{++}, K^+, Methanol

F-met-puromycin

1. Protein omission

Only L2, L3, L4 + 23S rRNA essential

2. Phenol + proteinase

Tag ribosomes → prot K, phenol

23S rRNA + 3 proteins function as minimal particle

Why focus on rRNA?

① Extreme conservation 7°, 2°

② Kethoxal inactivates 30S → due to modifying 16s rRNA

③ tRNA's bound to rRNA protect universally conserved nt.

④ Antibiotics bind to rRNA

⑤ Shine-Delgarno base pairs w/ rRNA

⑥ 23S binds to tRNA

23S

- six domains

- domains IV + V appear important

50S reconstitution

- in vitro tx. 23S rRNA doesn't work well even though natural rRNA works

why?

① natural RNA retains some memory of state

② post-tx modifications important

So.. Iterative natural tx + shell get some activity? ∴ some modification important

In vitro complementation?

——————— natural

~~~~~~~~~~ in-vitro

——— ~~~ Mapped important

~~~ —— in-vivo modification

——— ~~~ to region V.

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