

Cell-free Synthetic Biology

EE-490J

07.11.2023

Objectives

- How to purify ribosomes from bacterial cells?
 - Grow the cells – Culturing
 - Extract and purify ribosomes from cell lysate

- Concentrate PURE proteins from last week

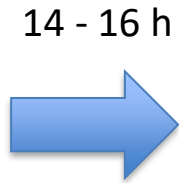
- SDS-PAGE analysis of PURE proteins

Culturing the cells for ribosome purification

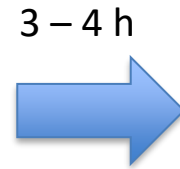
- Prepare the overnight culture (mother culture) for cells
 - E.coli Rb1 strain



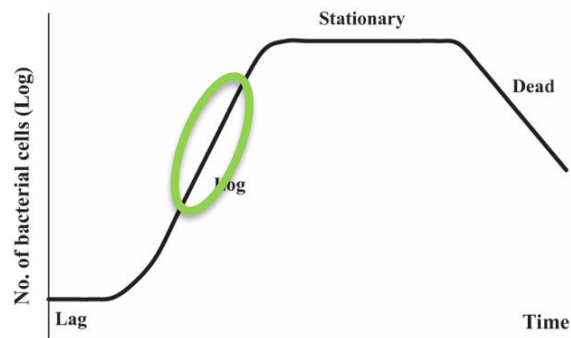
Glycerol stock



Overnight culture

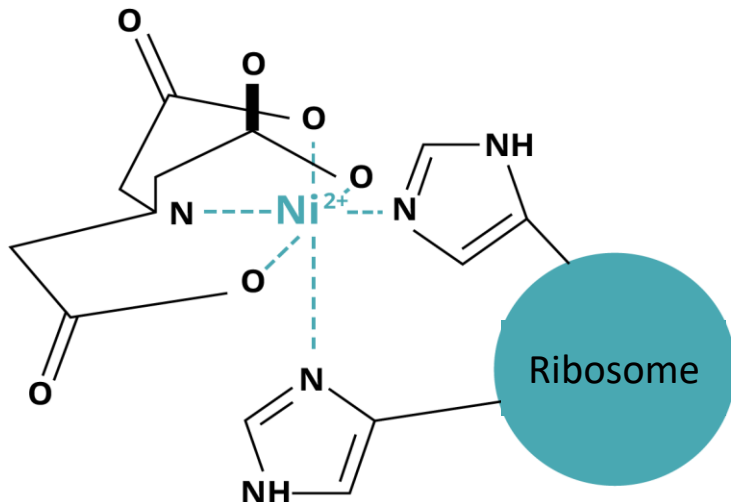
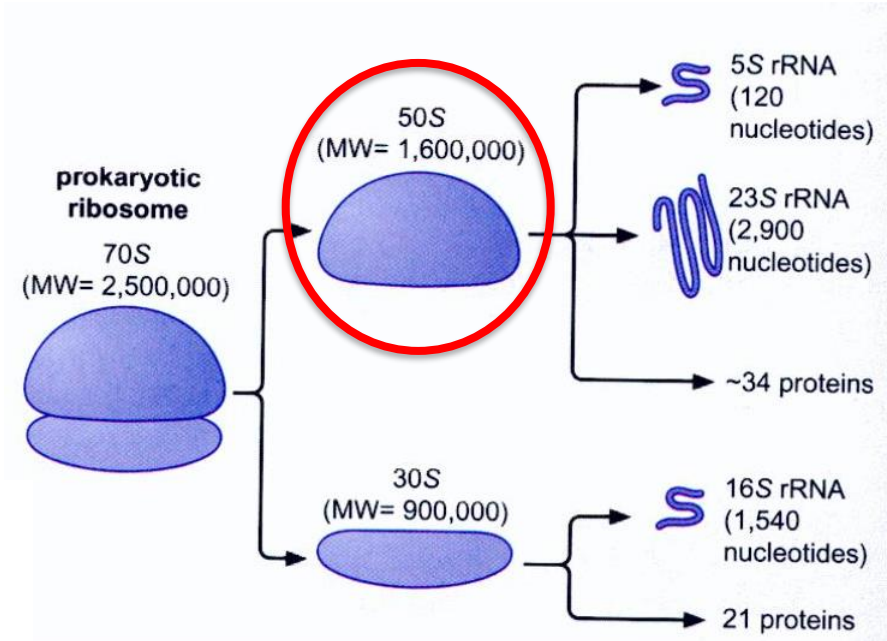


Culture for purification

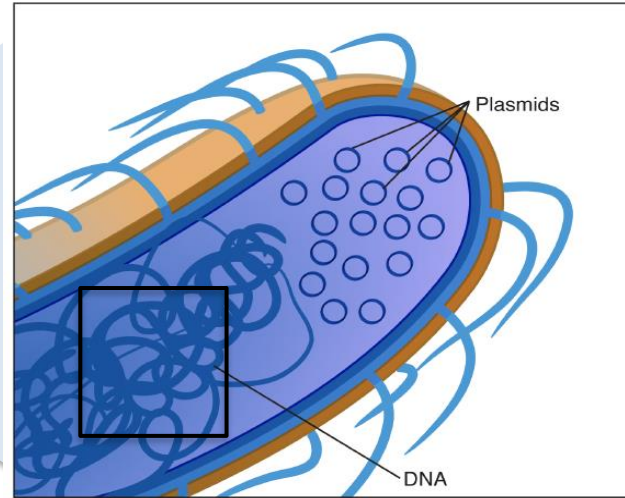
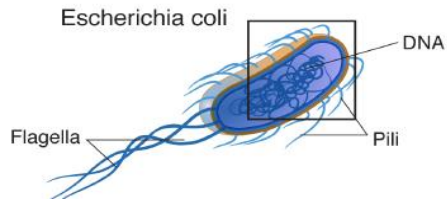


Culturing the cells for ribosome purification

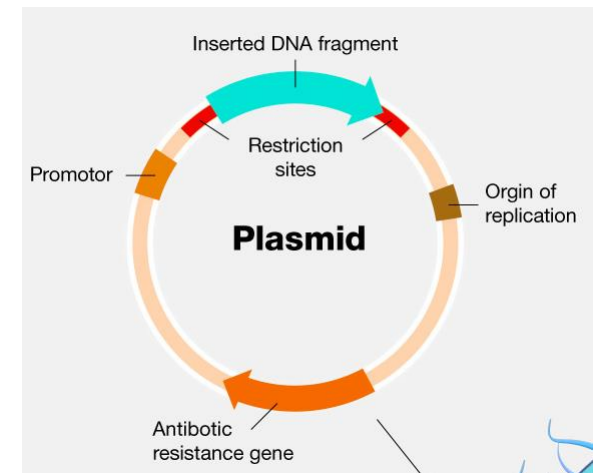
- Prepare the overnight culture (mother culture) for cells
 - E.coli Rb1 strain
- His-tagged ribosome (6X)
- Genomic insertion of tag at 50S subunit
- Constitutively expressed by cells



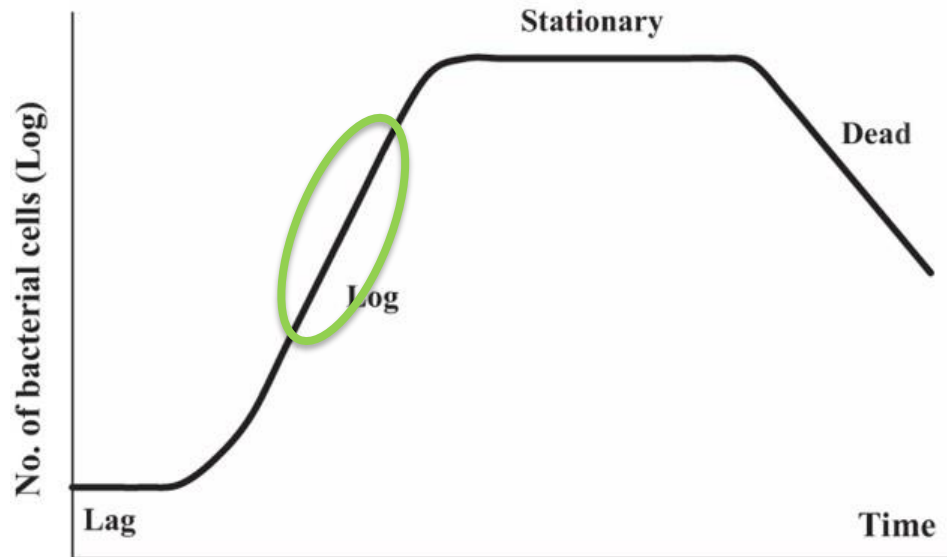
Rb1 his-tagged ribosome



Escherichia coli

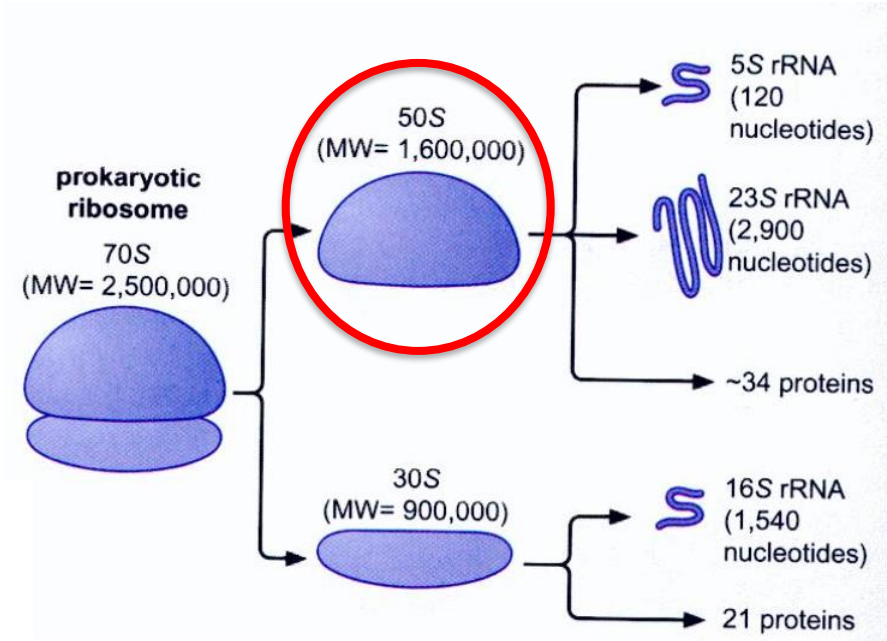
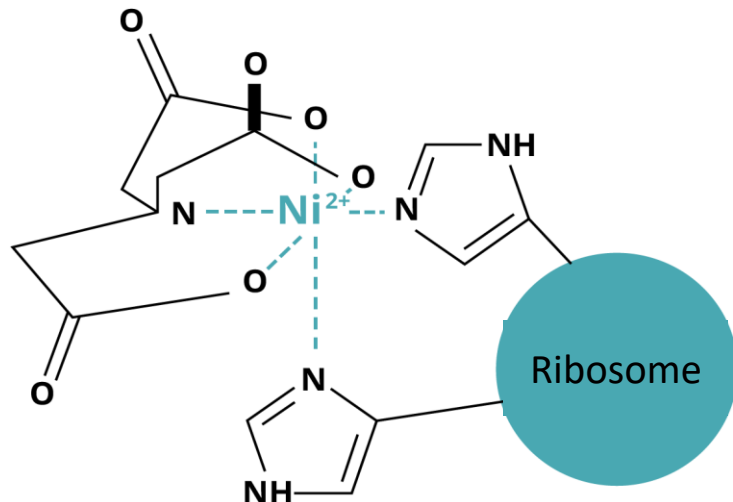


Bacterial growth curve



Culturing the cells for ribosome purification

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His-tag ribosome purification

