

# Remodelling of the bacterial ribosome during transition into stationary growth phase

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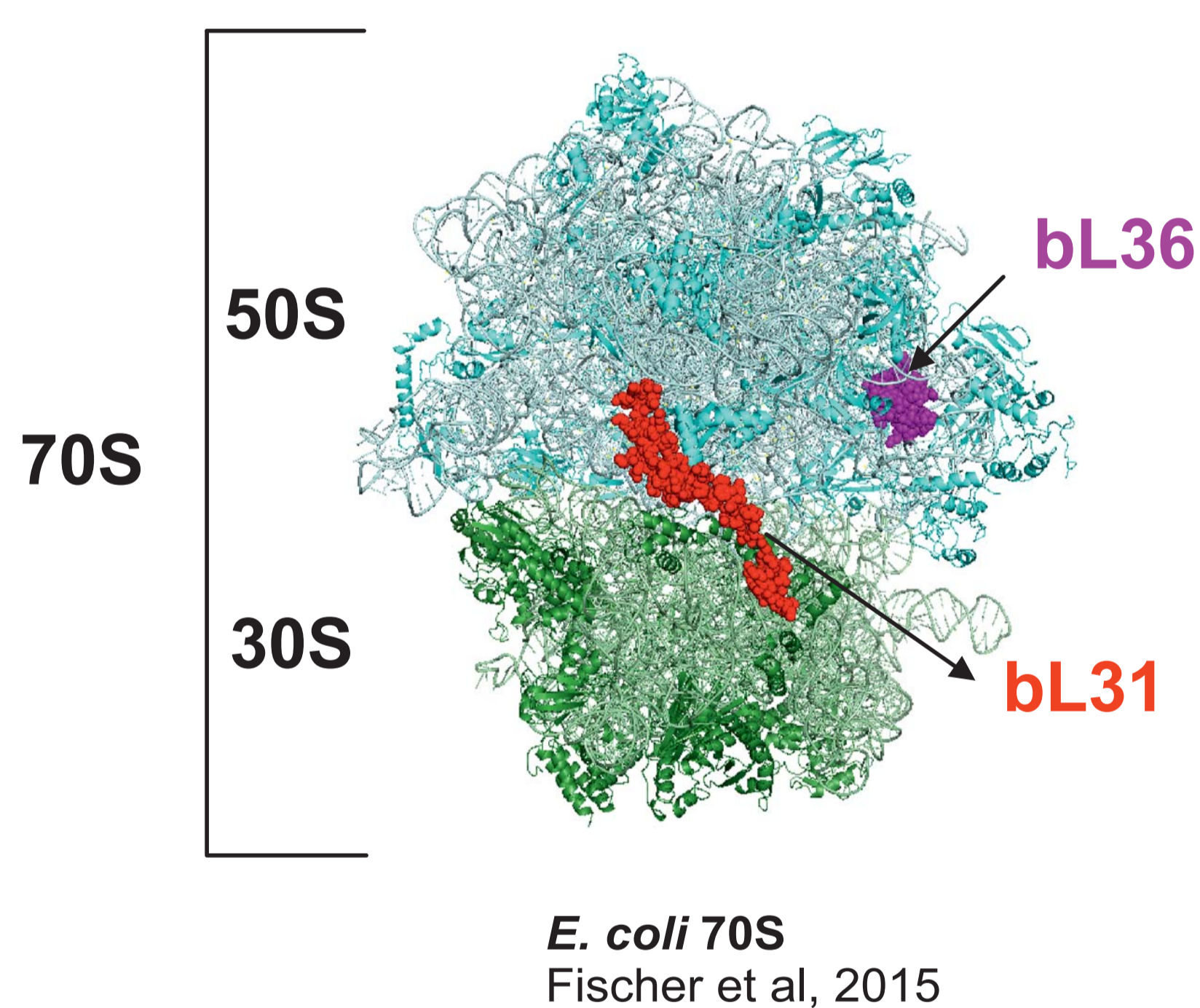
## INTRODUCTION

### Ribosome heterogeneity

- the coexistence of structurally different ribosomes in an organism
- demonstrated at rRNA and r-protein level in prokaryotic and eukaryotic organisms
- at r-protein level: r-protein paralogs, posttranslational modifications, stoichiometric differences

### Bacterial ribosome

- 2.3 MDa 2 subunit enzyme (50S, 30S)
- RNA/ protein ratio 2:1  
3 rRNAs + 54 r-proteins in *E. coli*
- highly abundant in fast growing cells: appr. 1/3 of cell dry mass and 70 000 copies per *E. coli* cell



<b>bL31 paralogs (A and B)</b>	<b>bL36 paralogs (A and B)</b>
both < 10 kDa, sequence ID 35,6%	both < 6 kDa, sequence ID 34,8%
large contact surface with both subunits, forms B1b intersubunit bridge	contacts only large subunit rRNA
widely conserved bacteria-specific large subunit proteins	
A paralogs bind one Zn <sup>2+</sup> ion, B paralogs do not bind Zn <sup>2+</sup>	
duplicated genes in several completely sequenced bacterial genomes: in <i>E. coli</i> B paralogs encoded by the same operon, A paralogs by different operons	

### Open questions

What is the biological role of bacterial ribosome heterogeneity?

Does ribosome structural heterogeneity confer functional heterogeneity?

## RESULTS

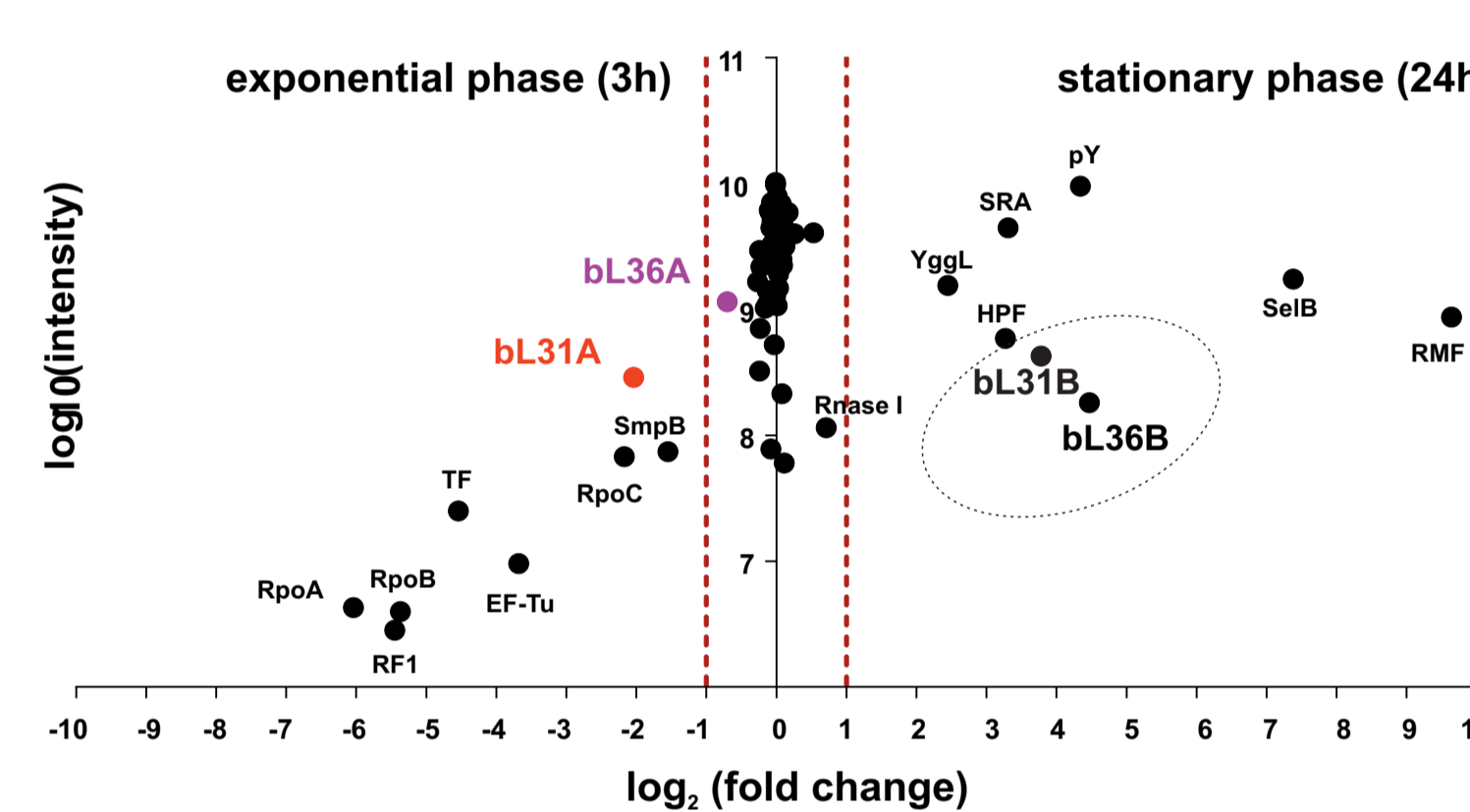
### Hypothesis

Bacterial ribosomal protein composition changes during growth.

### Conclusions

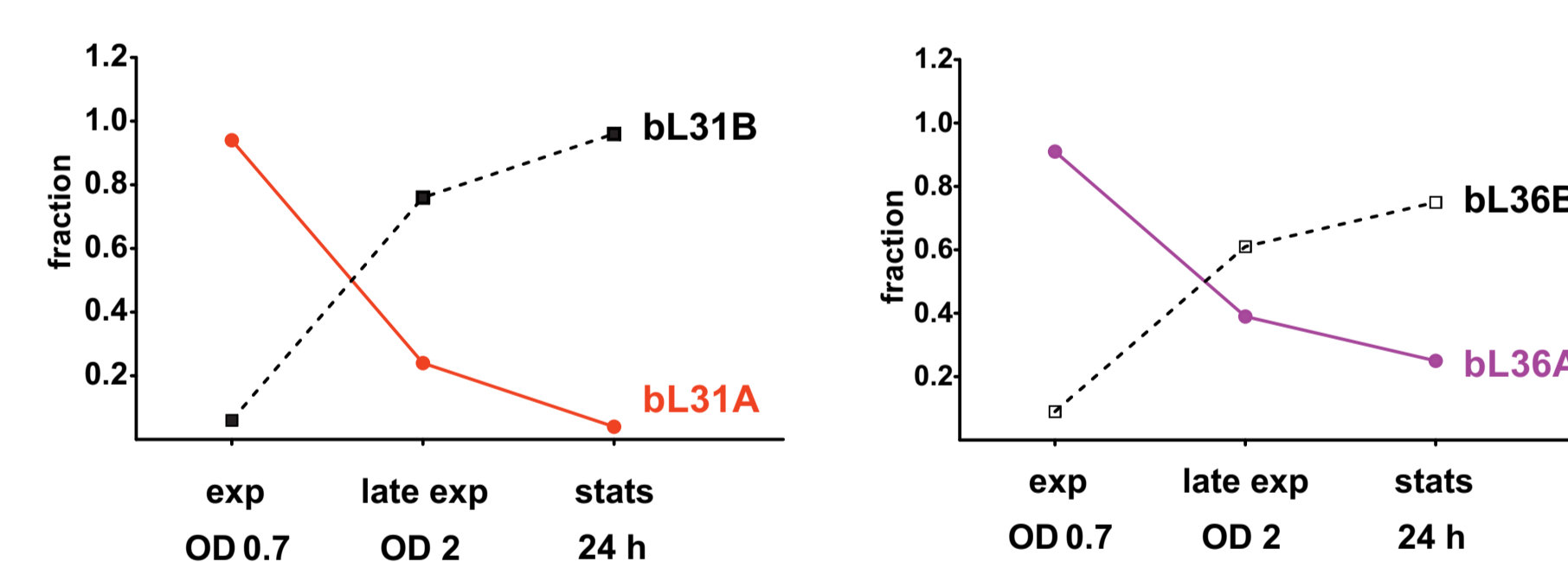
- E. coli* ribosomes are heterogeneous with respect to bL31 and bL36 paralogs.
- bL31 and bL36 paralogs are the only r-proteins whose fraction changes during growth.
- bL31B is able to replace bL31A in mature ribosomes via protein exchange.
- In growth competition of bL31 paralogs bL31A supports cyclic growth more effectively than bL31B.

### Ribosomal proteome is different during exponential and stationary growth.



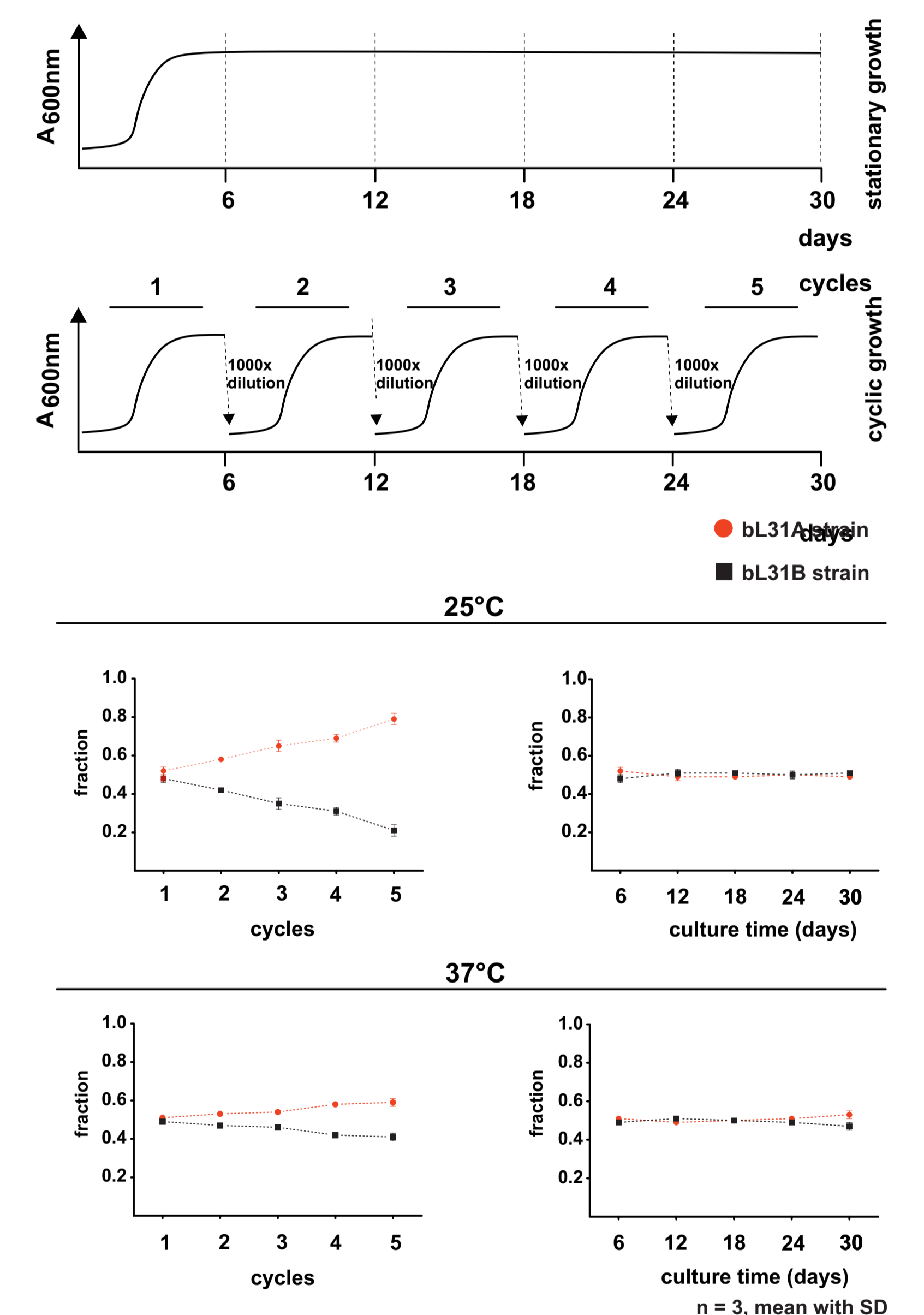
bL31 and bL36 paralogs are the only ribosomal core proteins whose abundance is different in exponential and stationary growth phase as detected by quantitative MS (pSILAC).

### Ribosomal protein paralog composition changes during growth



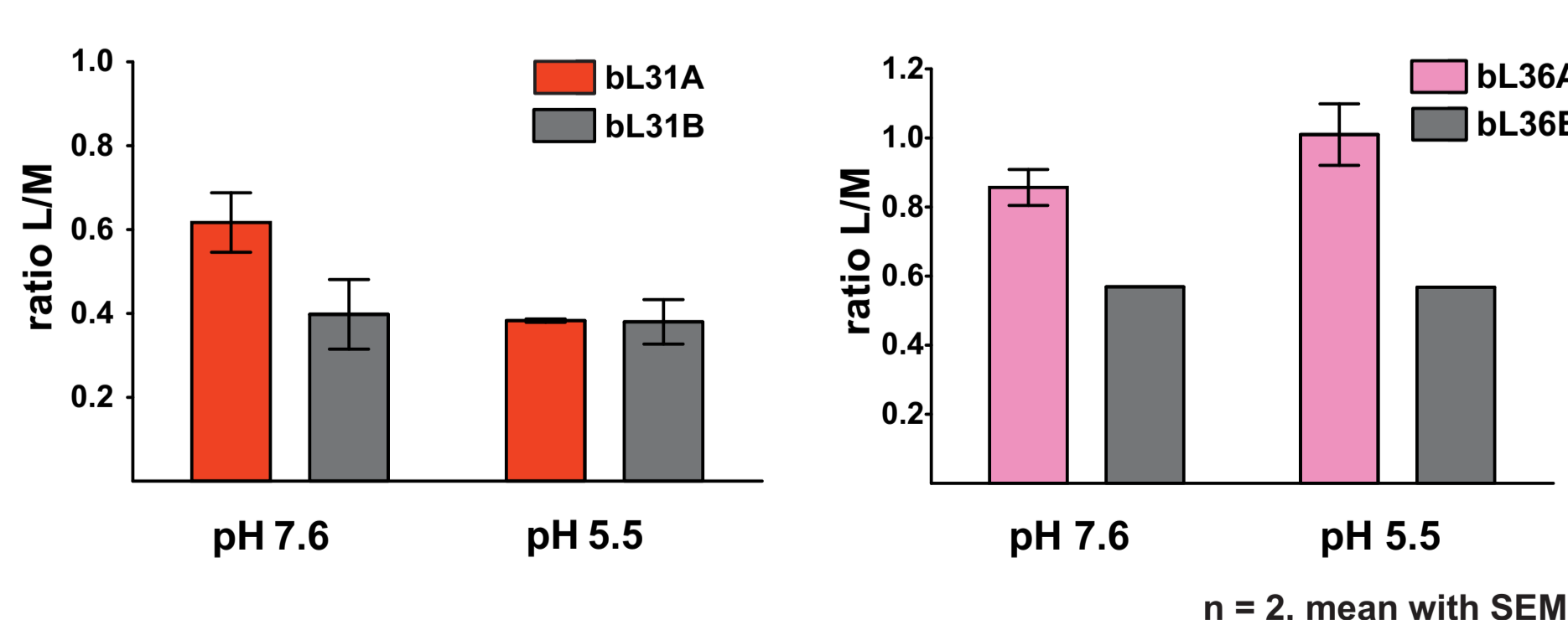
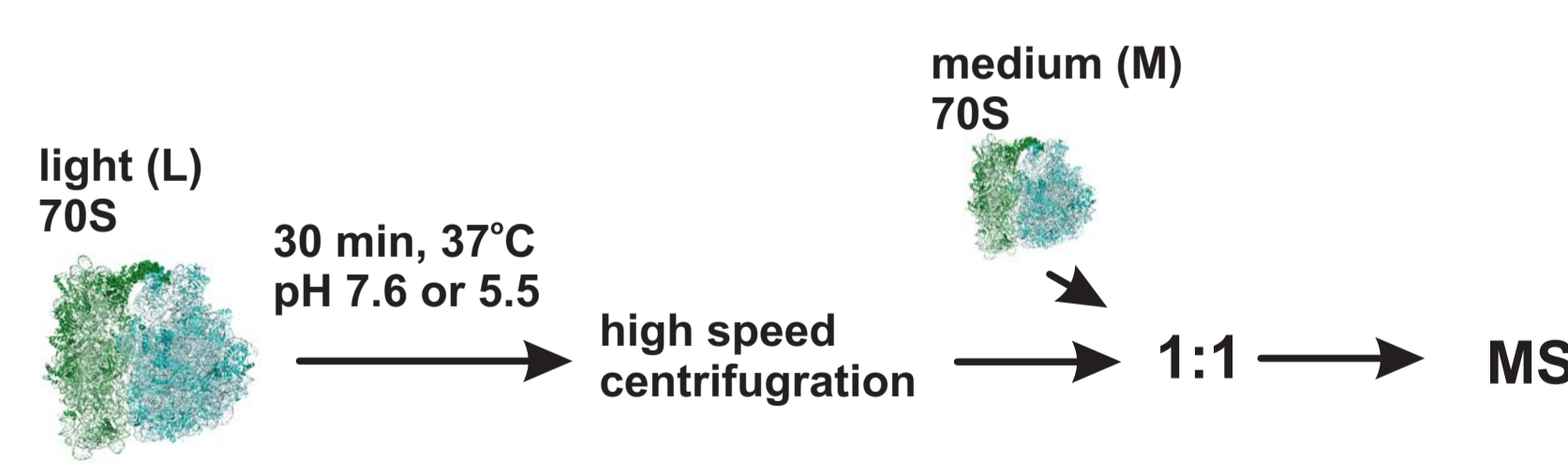
The proportion of bL31A and bL36A in 70S ribosomes decreases and bL31B and bL36B increase during growth phase progression as detected by pSILAC qMS.

### Growth competition of bL31A or bL31B expressing strains



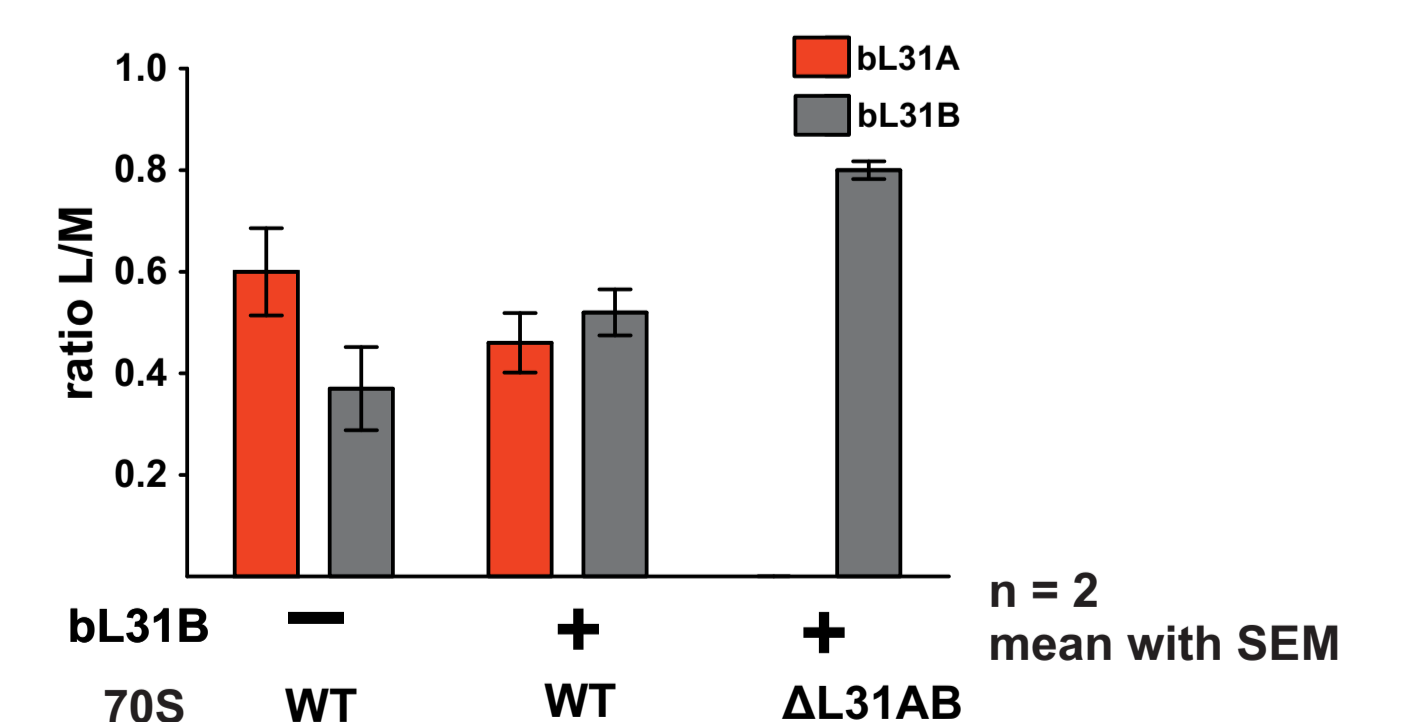
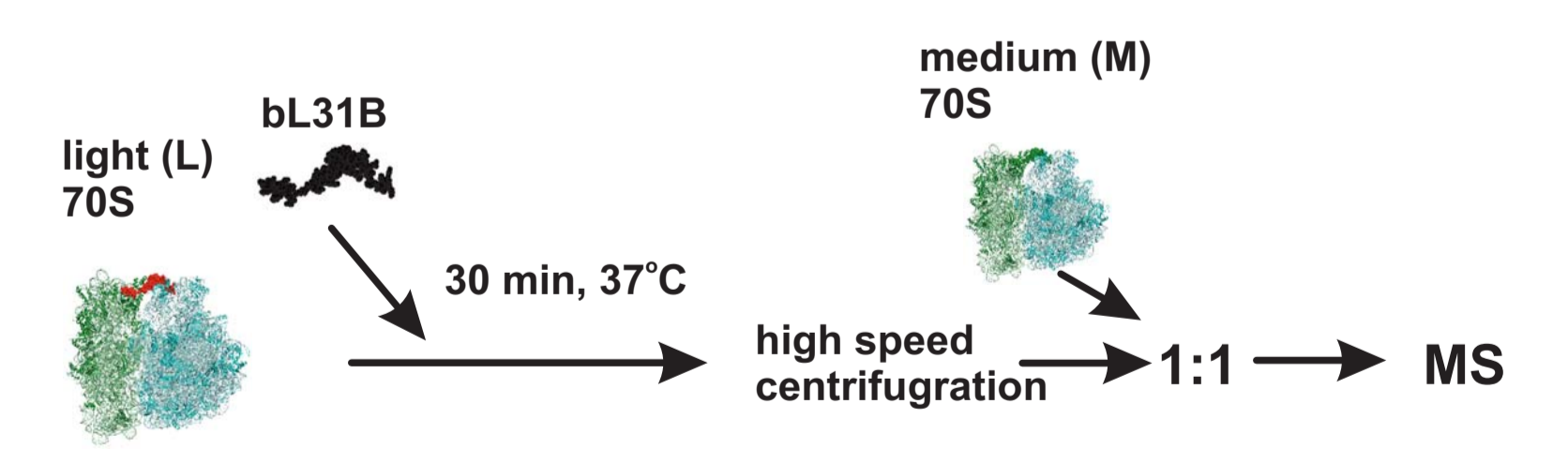
bL31A supports bacterial cyclic growth more effectively than bL31B.

### bL31A dissociates from the ribosome in acidic environment *in vitro*



Under pH 5.5 the fraction of bL31A in 70S ribosomes decreases by more than 30%. No pH-dependent effect on bL31B and both bL36 paralogs' was detected.

### bL31B replaces bL31A in 70S ribosomes *in vitro*



Addition of purified bL31B to 70S ribosomes results in decrease of bL31A fraction and simultaneous increase of bL31B fraction.