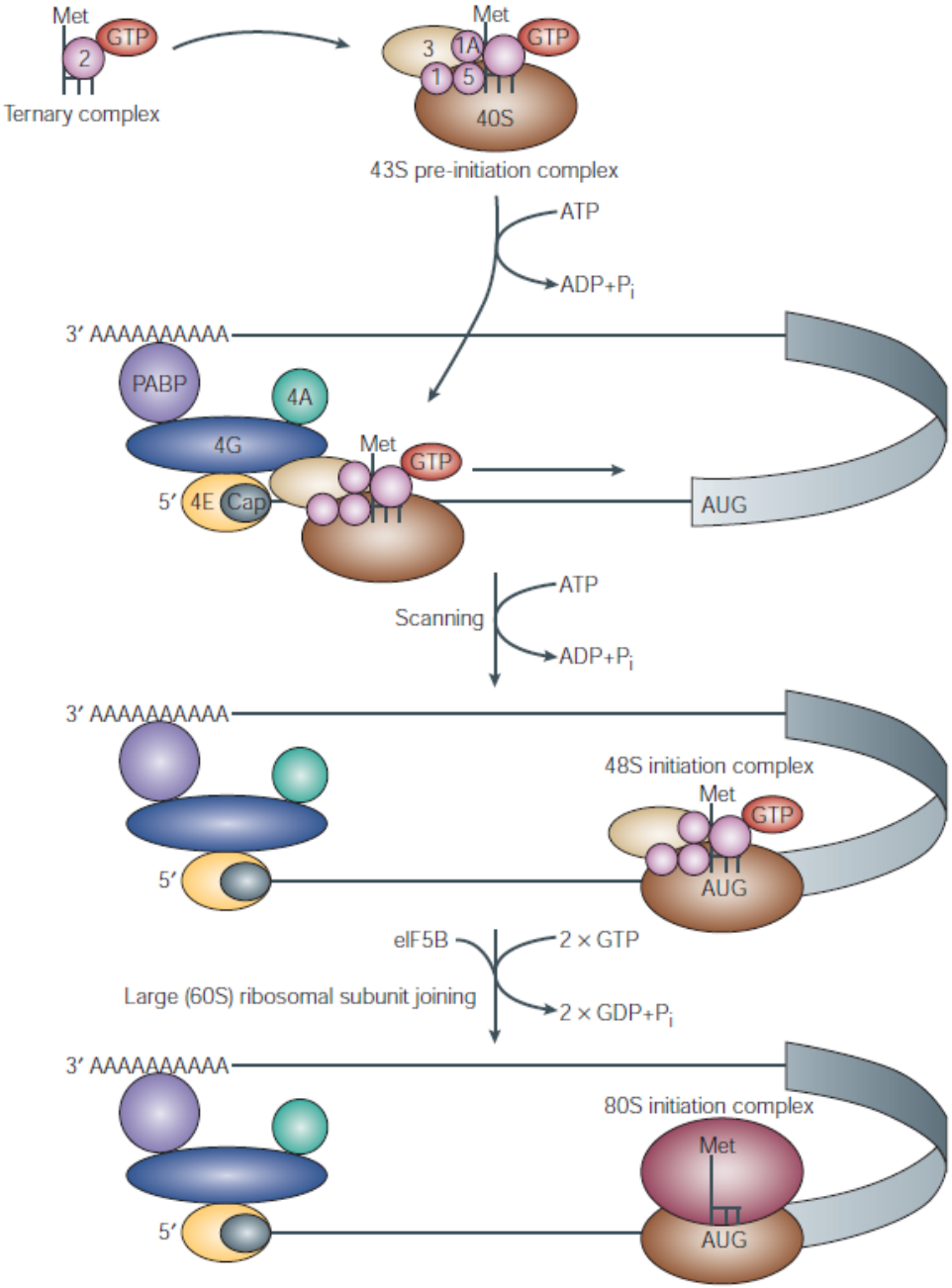
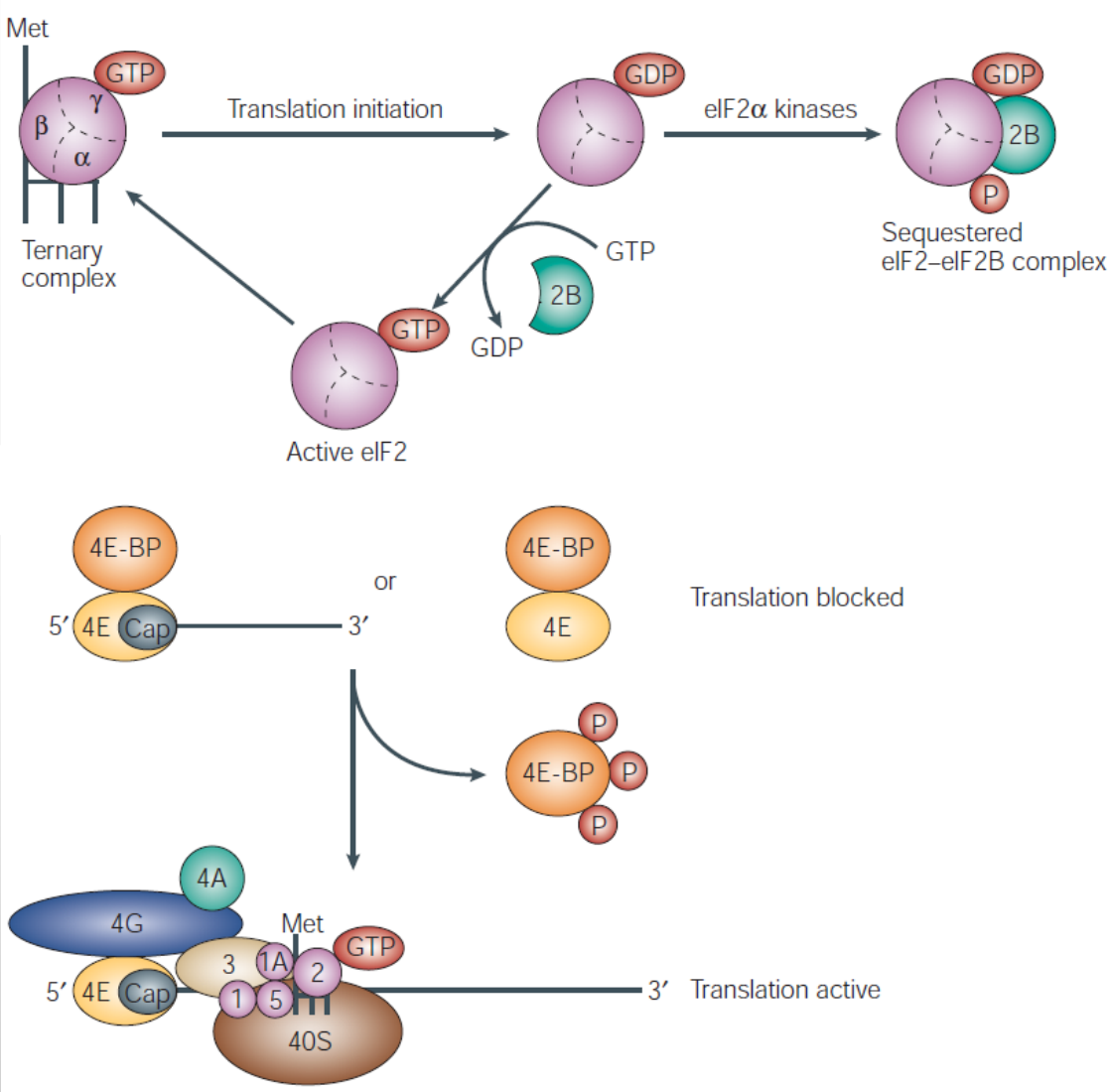
Quantitative profiling of initiating ribosomes in vivo

Parrinello Natalia
TJC
10.03.2015

Translatic **ıd highly**
regulated pro **ɔtein factors**



Global control of protein synthesis



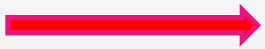
Ribosome profiling: new views of translation, from single codons to genome scale

Nicholas T. Ingolia

Profiles of ribosome occupancy across mRNAs →

global and gene-specific features of translational speed

- A caveat, however, exists because the average ribosome density on mRNAs is negatively influenced by the elongation speed;

- Dynamic changes of initiation rates are masked by the varied elongation speed under different growth conditions 

A method capable of **mapping start-codon selection and quantifying the rate of 80S ribosome assembly at individual translation initiation sites (TISs)**

NATURE METHODS | VOL.12 NO.2 | FEBRUARY 2015 | 147

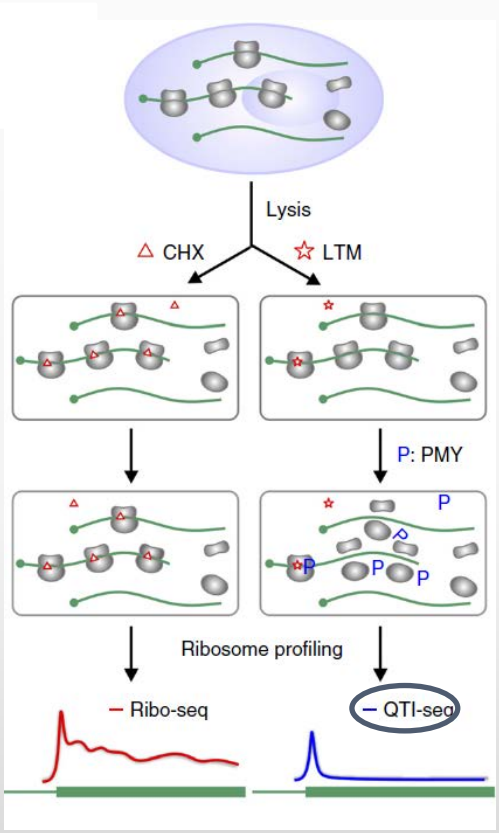
Quantitative profiling of initiating ribosomes *in vivo*

Xiangwei Gao[1,5,6], Ji Wan[1,6], Botao Liu[2], Ming Ma[3], Ben Shen[3,4] & Shu-Bing Qian[1,2]

- **quantitative translation initiation sequencing (QTI-seq), with which the initiating ribosomes can be profiled in real time at single-nucleotide resolution**

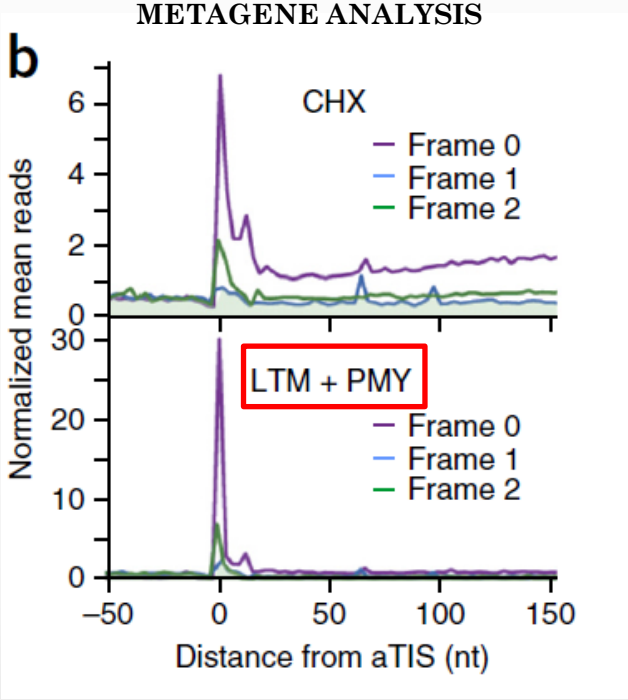
- **resultant initiation maps not only delineated variations of start-codon selection but also highlighted a dynamic range of initiation rates in response to nutrient starvation**

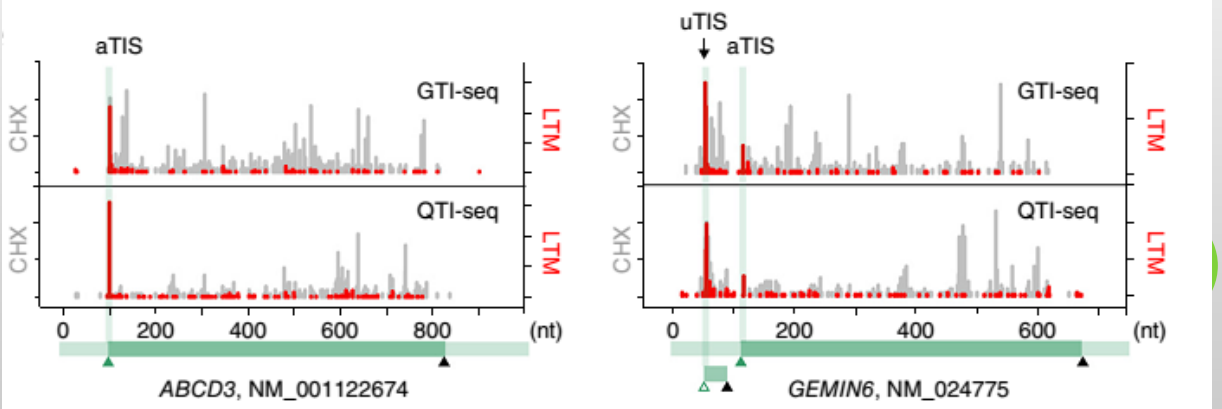
QTI-seq captures real-time translation initiation events



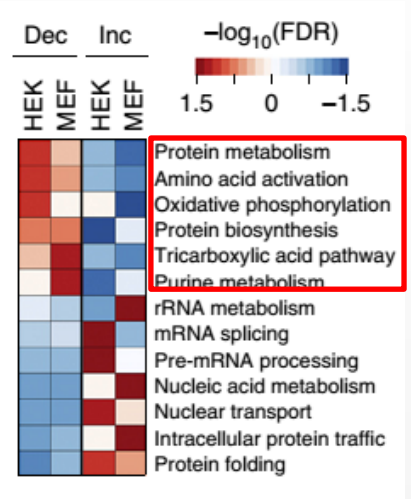
LTM:lactimidomycin

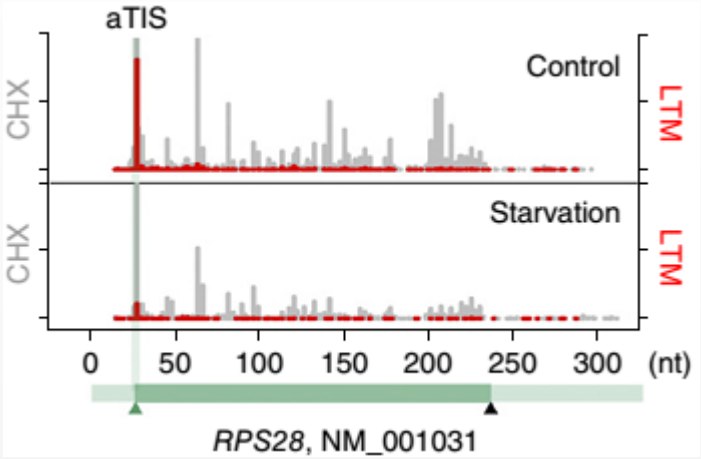
METAGENE ANALYSIS



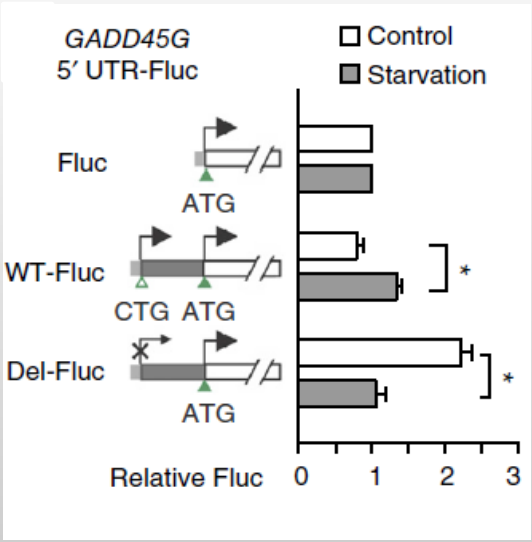


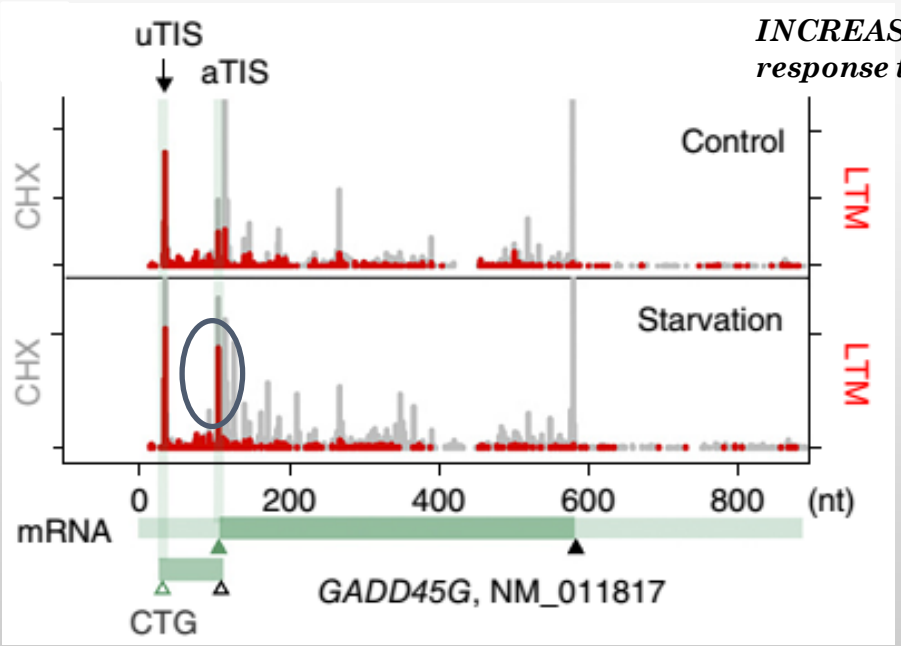
Quantitative TIS profile in response to starvation





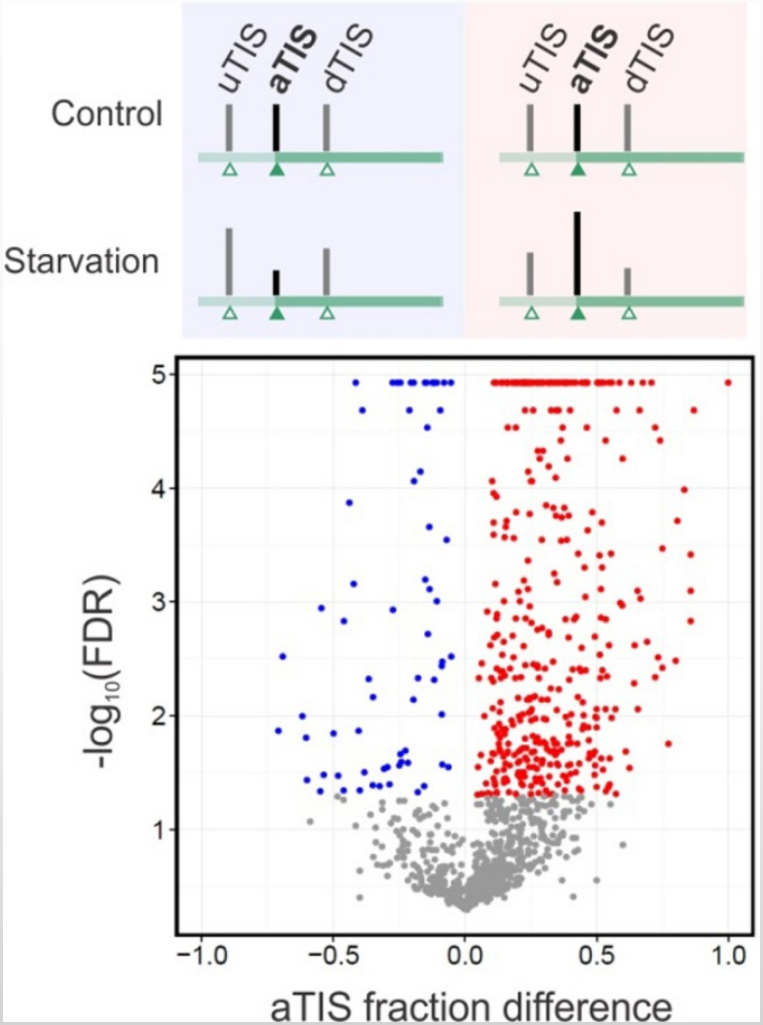
FIVEFOLD decreased aTIS in response to starvation



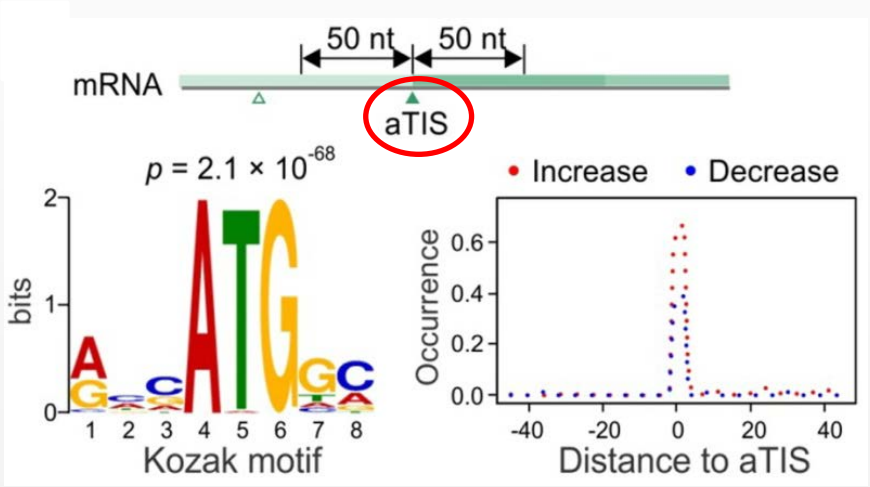


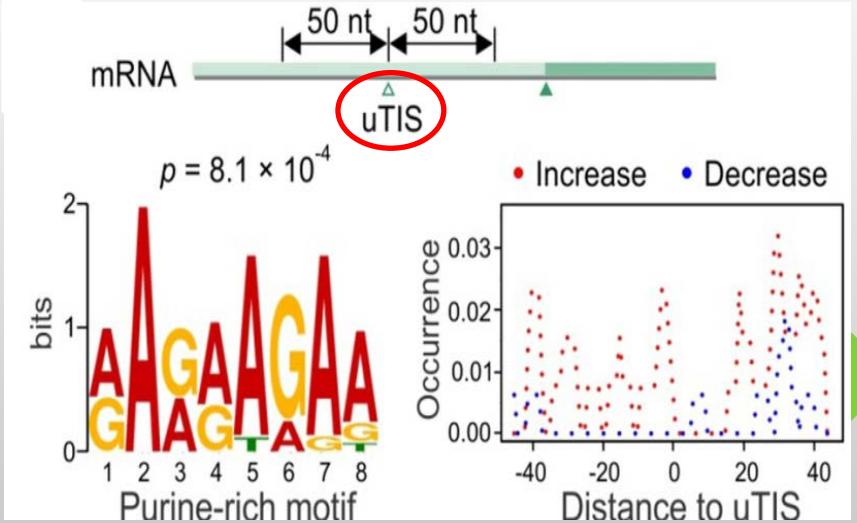
INCREASED aTIS in response to starvation

Programmatic TIS regulation in response to starvation

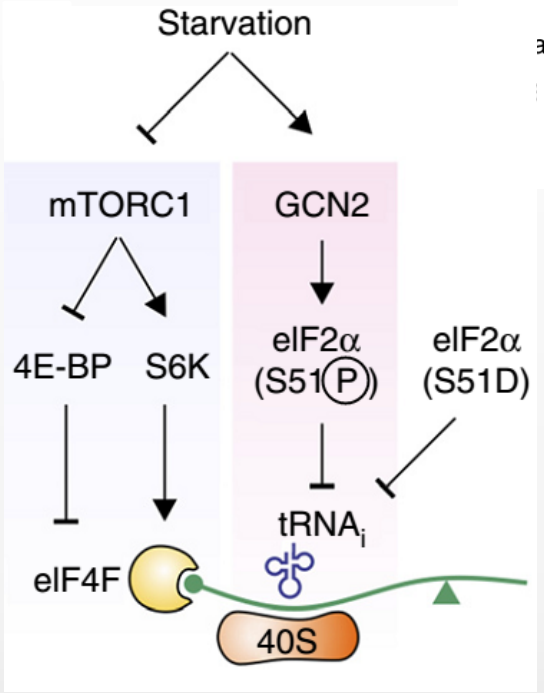


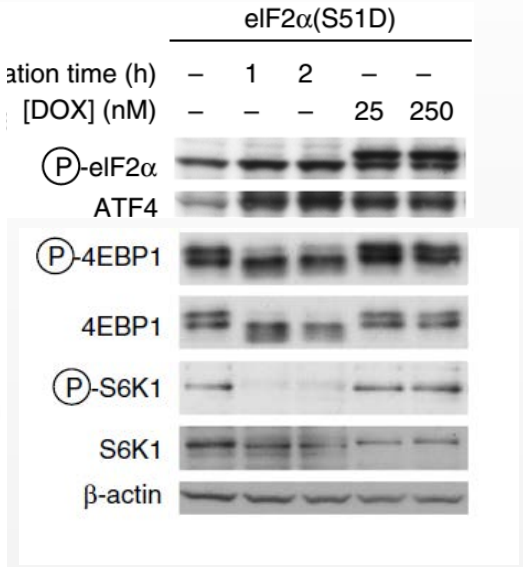
CONSENSUS SEQUENCE MOTIFS

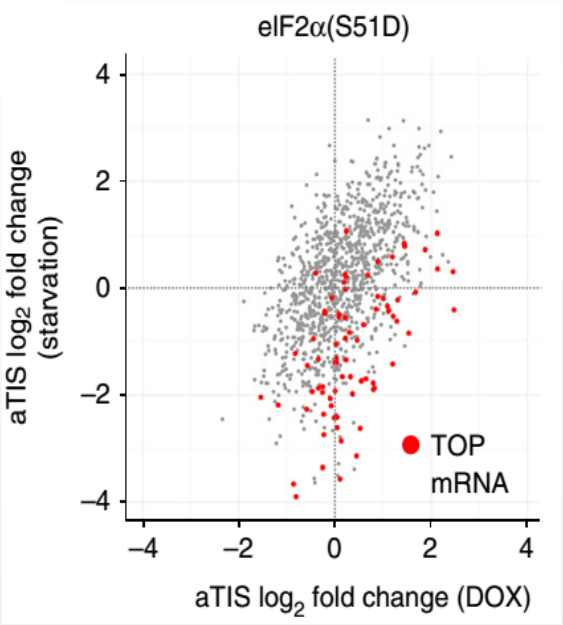




TIS regulation pathway in response to starvation



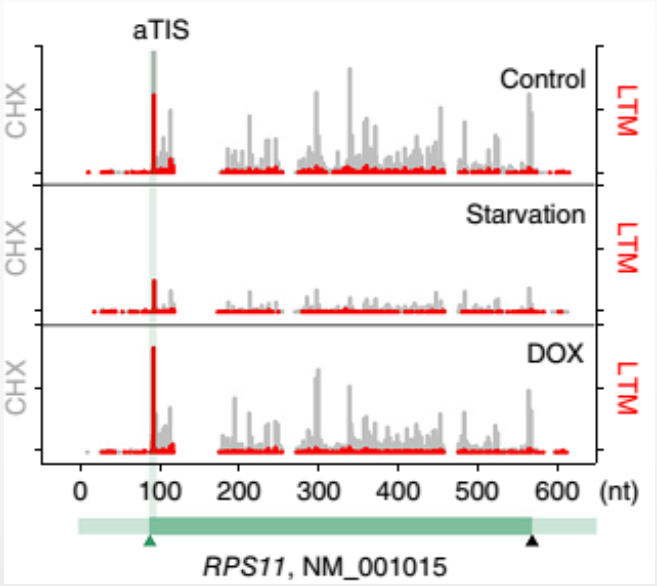


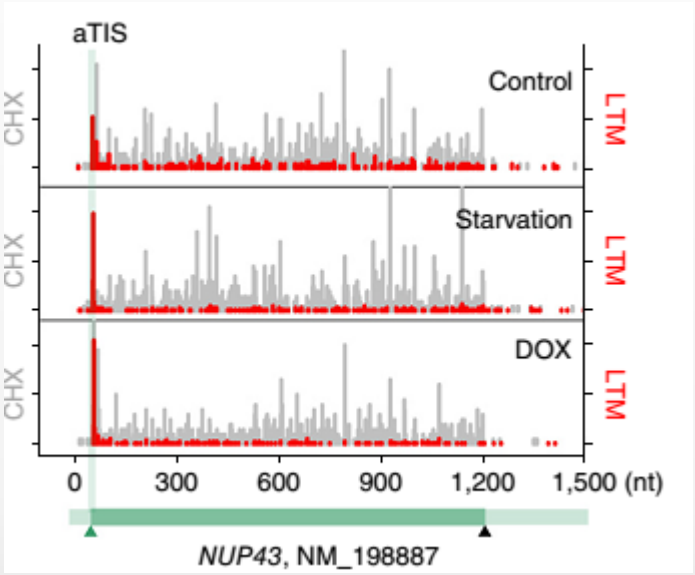


Starvation → ↓mTORC1 signaling → suppression of TOP mRNA translation

eIF2a P → upregulation of a SUBSET of trascripts

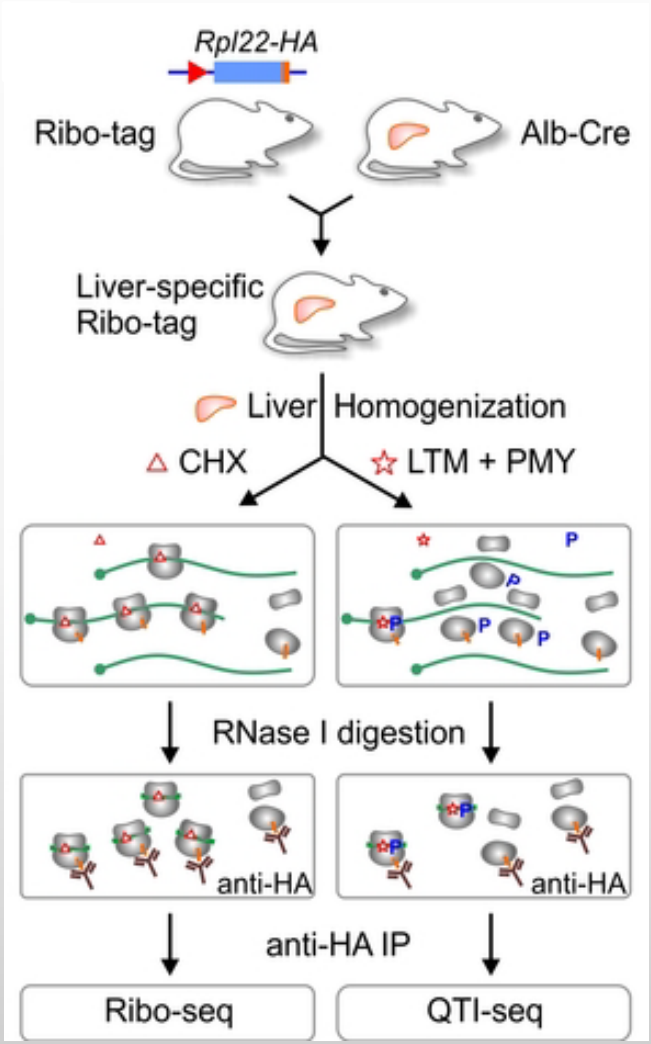
TIS regulation pathway in response to starvation

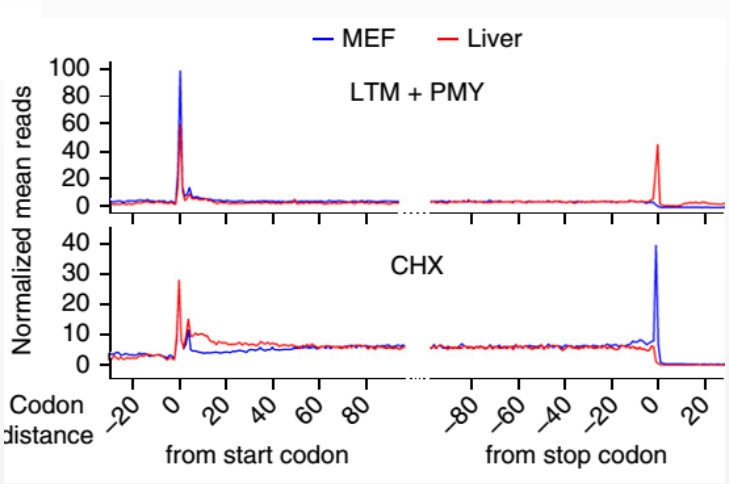




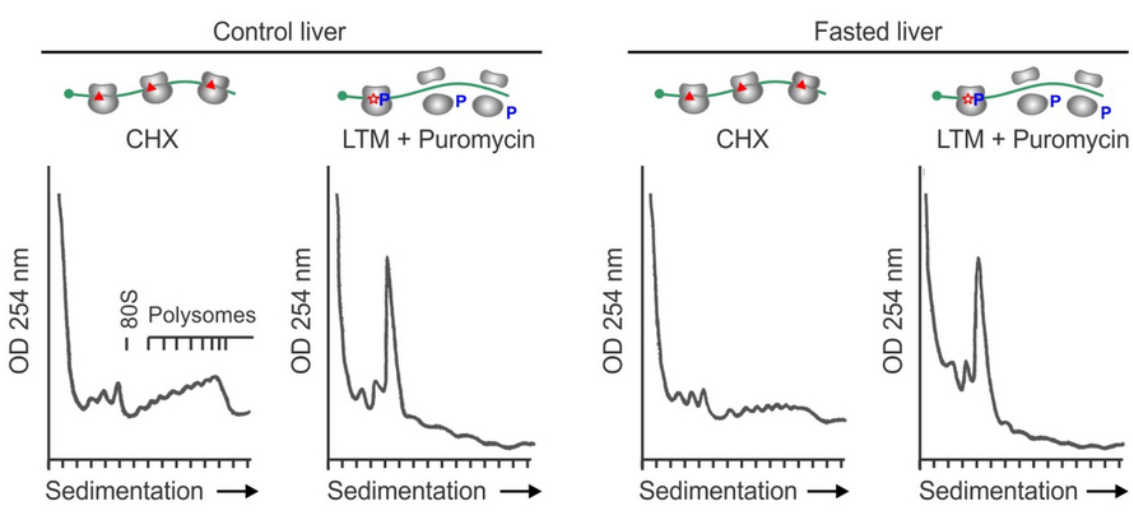
distinct upstream signaling pathways act on different aspects of translation initiation, resulting in a coordinated translational reprogramming to achieve *cellular adaptation*

Tissue-specific QTI-seq in liver cells

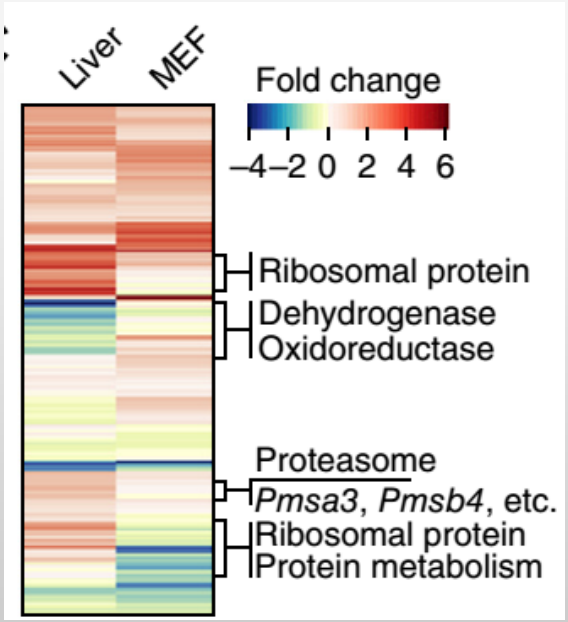




Liver-specific TIS profile in response to fasting

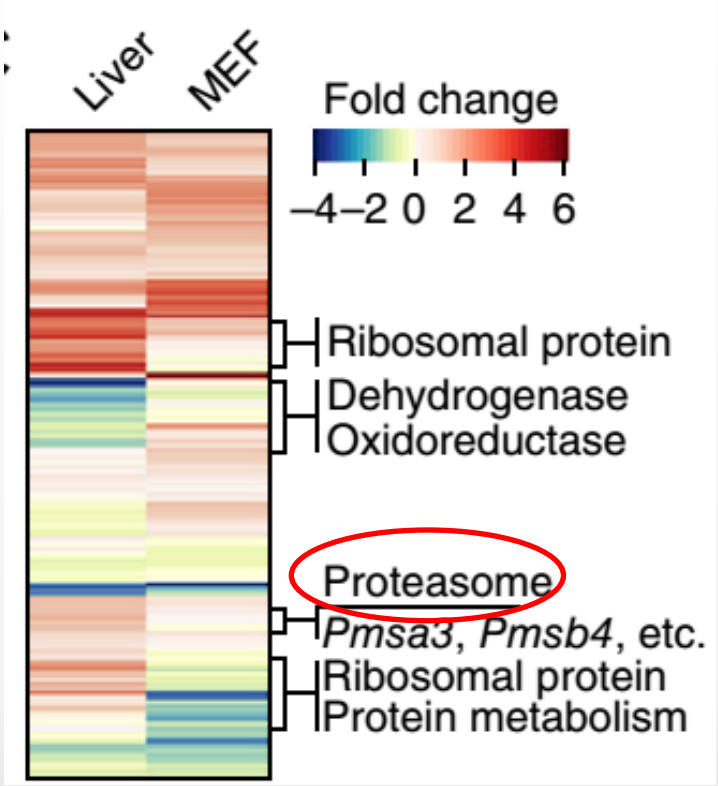


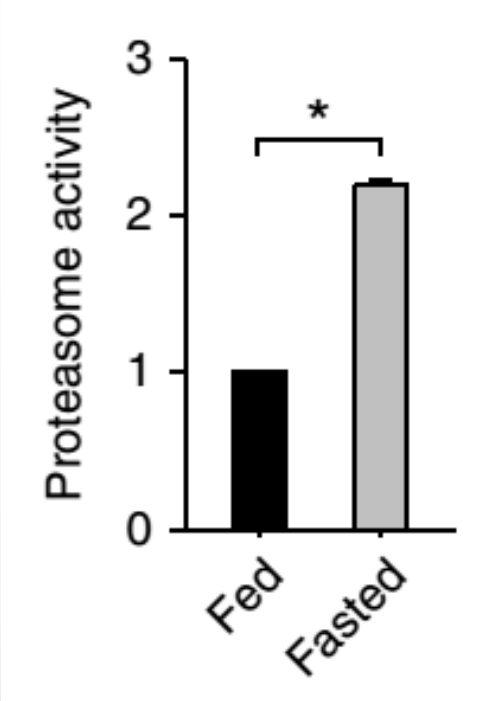
Food deprivation resulted in disassembly of polysomes in liver tissues



Under prolonged fasting conditions, a continuous supply of ribosomal proteins is likely needed for selective protein synthesis

Liver-specific TIS profile in response to fasting





Coordinated regulation provides an elegant mechanism for cells and tissues to achieve metabolic homeostasis under nutrient deprivation

SUMMARY

- Translation initiation is a crucial point of regulation in eukaryotic gene expression

- The significance of translation initiation is substantiated by the existence of alternative translation that utilizes one or more potential TISs in addition to the main start codon

- QTI-seq represents a conceptually distinct approach that permits quantitative profiling of initiating ribosomes in samples from cells in culture and solid tissues

- With RiboTag mice tool, they demonstrated liver cell–specific profiling of initiating ribosomes as a prototype for tissue-specific ribosome profiling

- Notably, the global reduction of TOP mRNA translation was no longer evident, (continuous supply of ribosomal proteins is needed for selective protein synthesis)

- QTI-seq uncovered a potent translational reprogramming for the proteasome system

THANK YOU FOR YOUR ATTENTION!!!