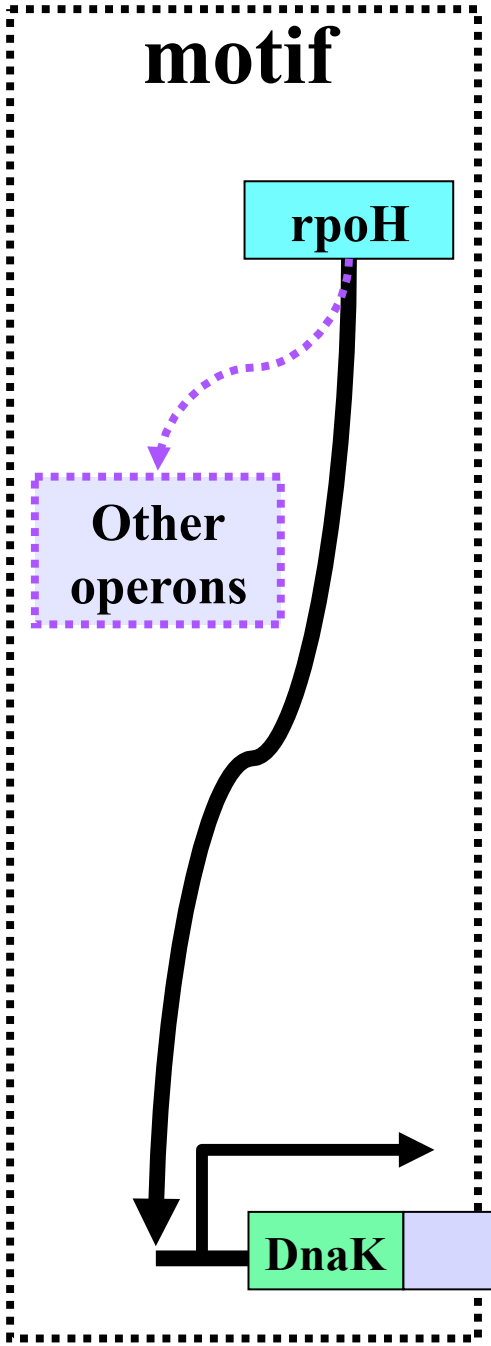
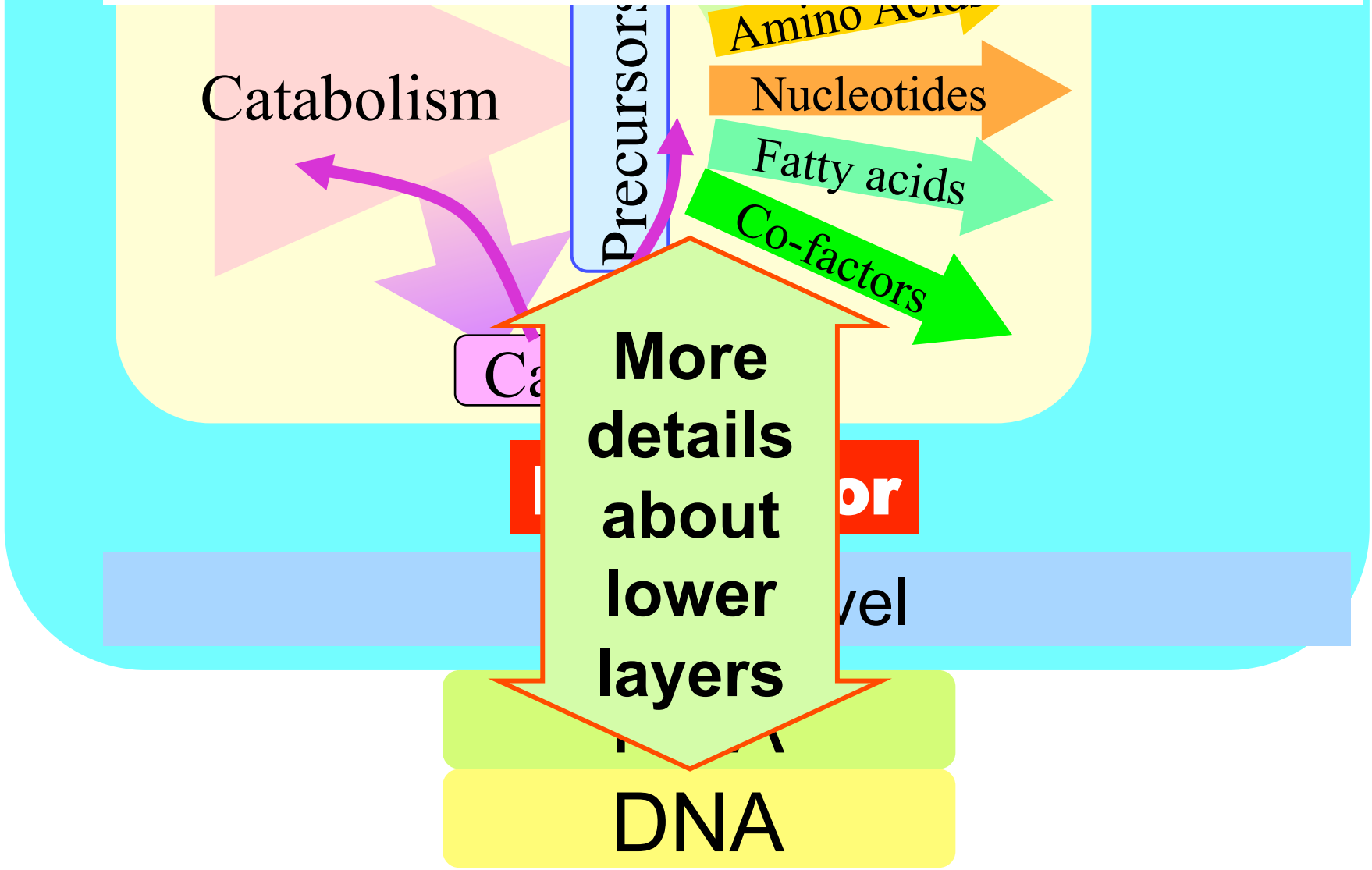


Note: we are focusing on

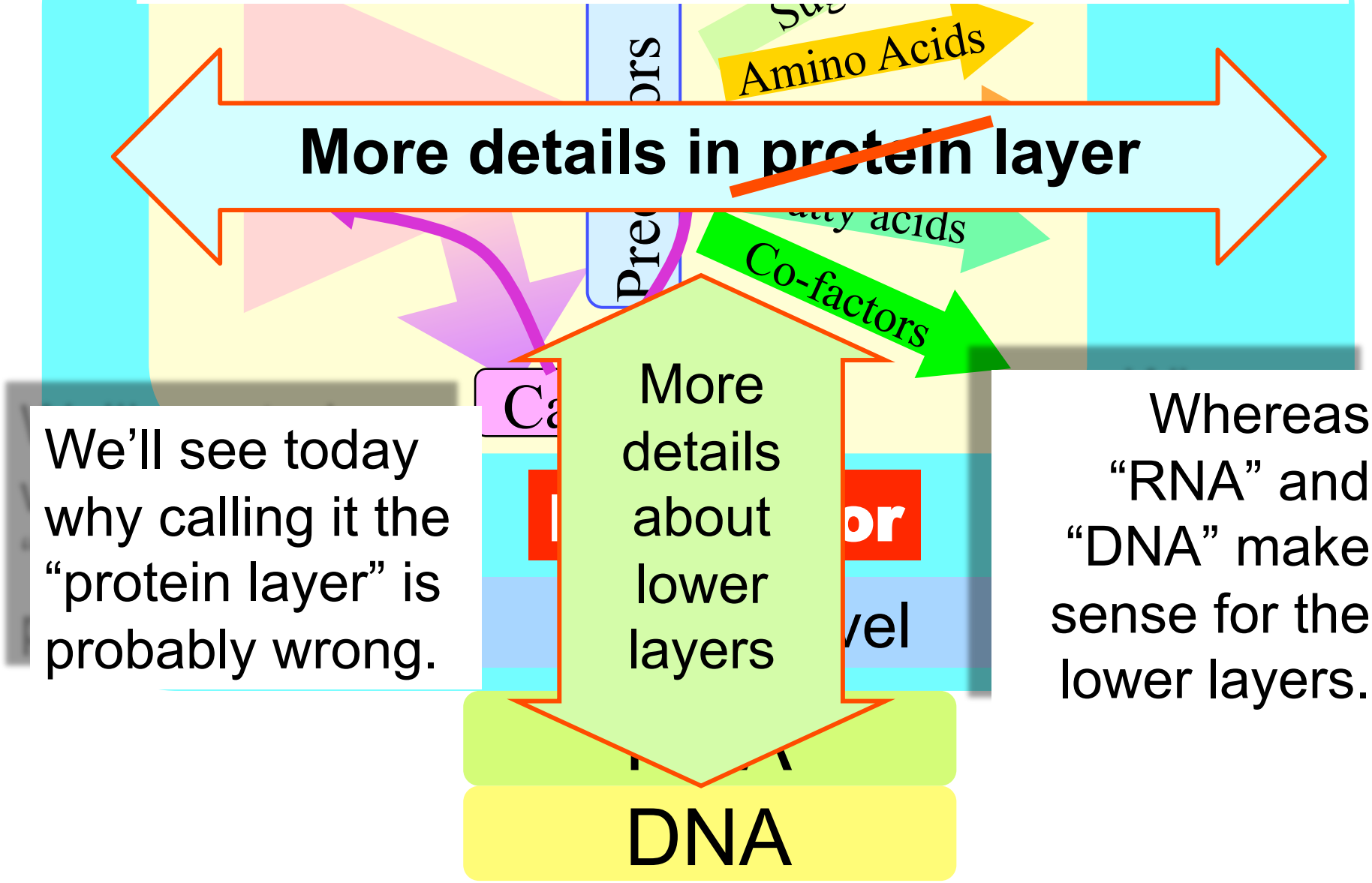
- using well-known case studies to illuminate general principles
- deliberate orthogonality to the standard stories
- which are mostly about circuits, less about networks, and almost nothing about architecture
- not a substitute but a complement
- also not yet right but hopefully heading in the right direction

You have to read the *PNAS* paper on heat shock for details

Heat shock control will emphasize the lower layers. It is one thin slice but illuminates general principles.



We will come back to other case studies in the “application layer”.



We’ll see today why calling it the “protein layer” is probably wrong.

Whereas “RNA” and “DNA” make sense for the lower layers.

Network motifs in the transcriptional regulation network of *Escherichia coli*

Shai S. Shen-Orr[1], Ron Milo[2], Shmoolik Mangan[1] & Uri Alon[1,2]

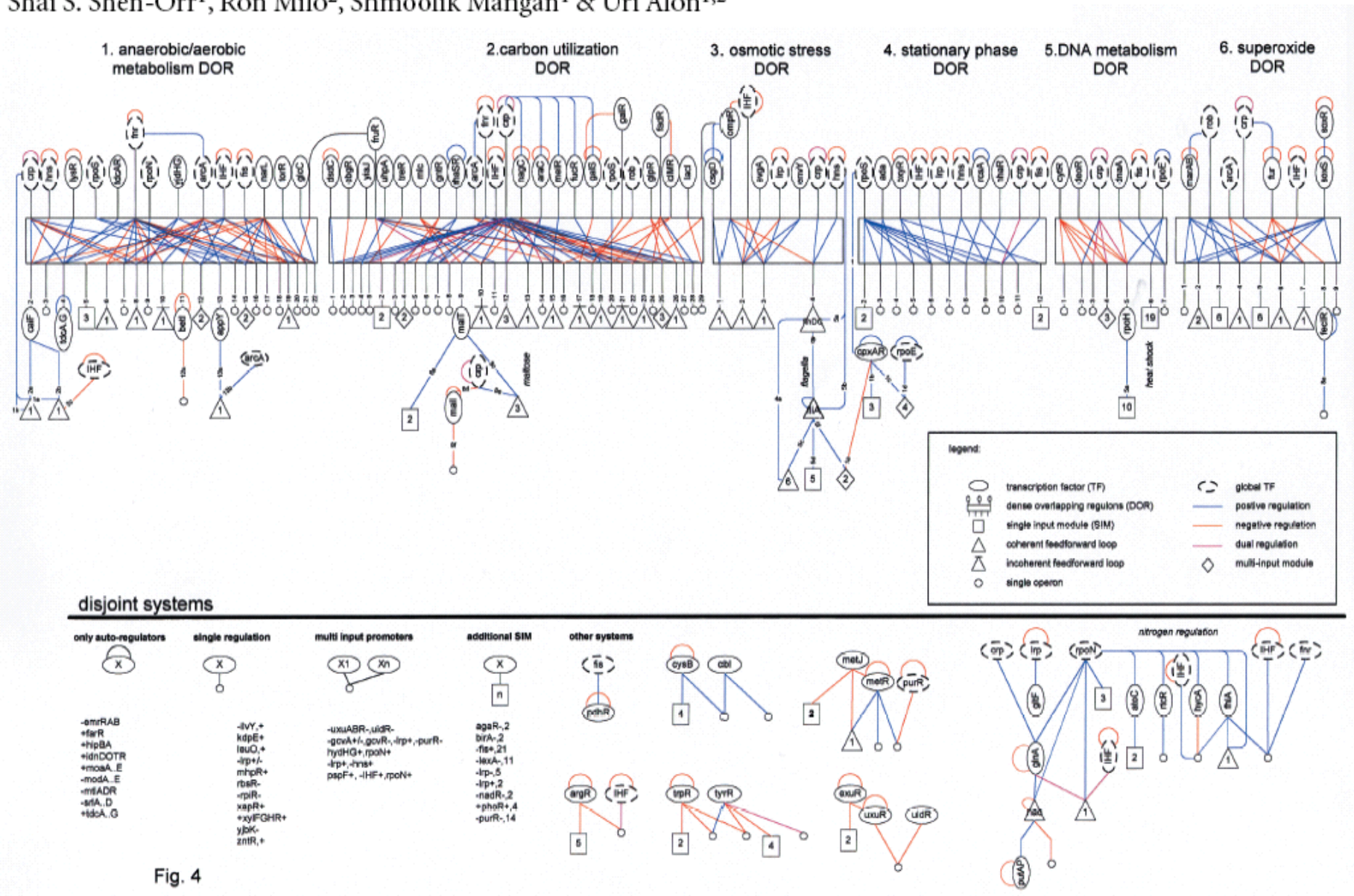
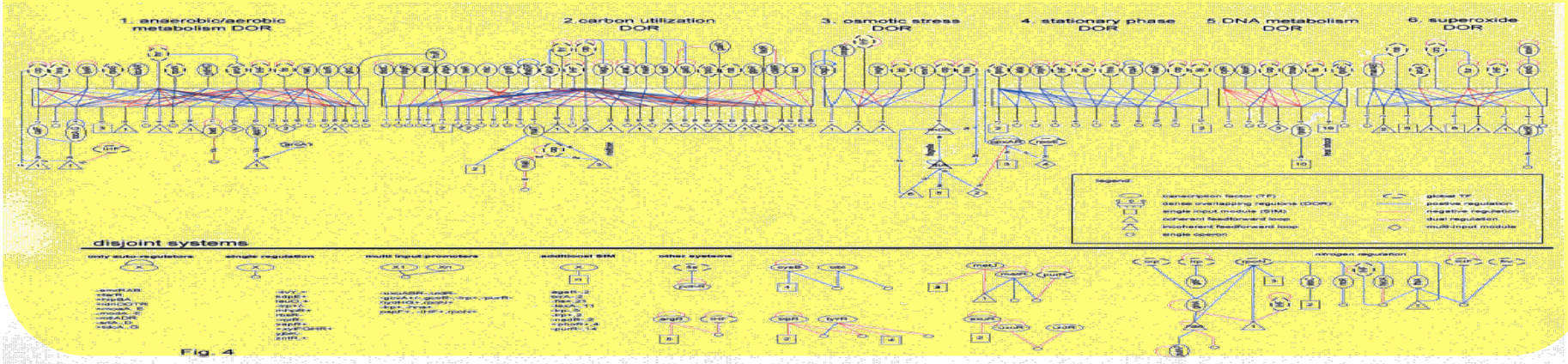


Fig. 4

All within the DNA layer



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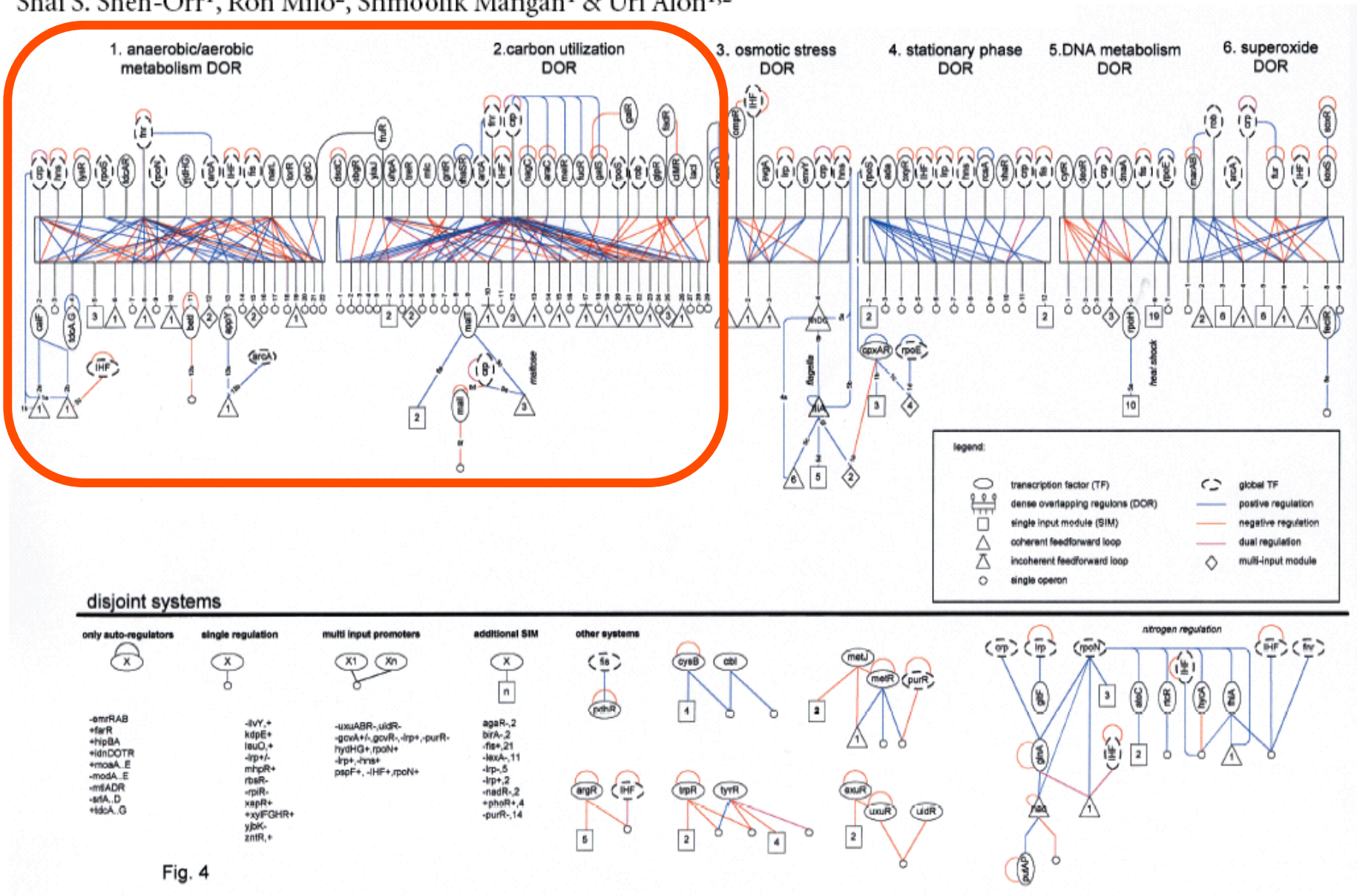
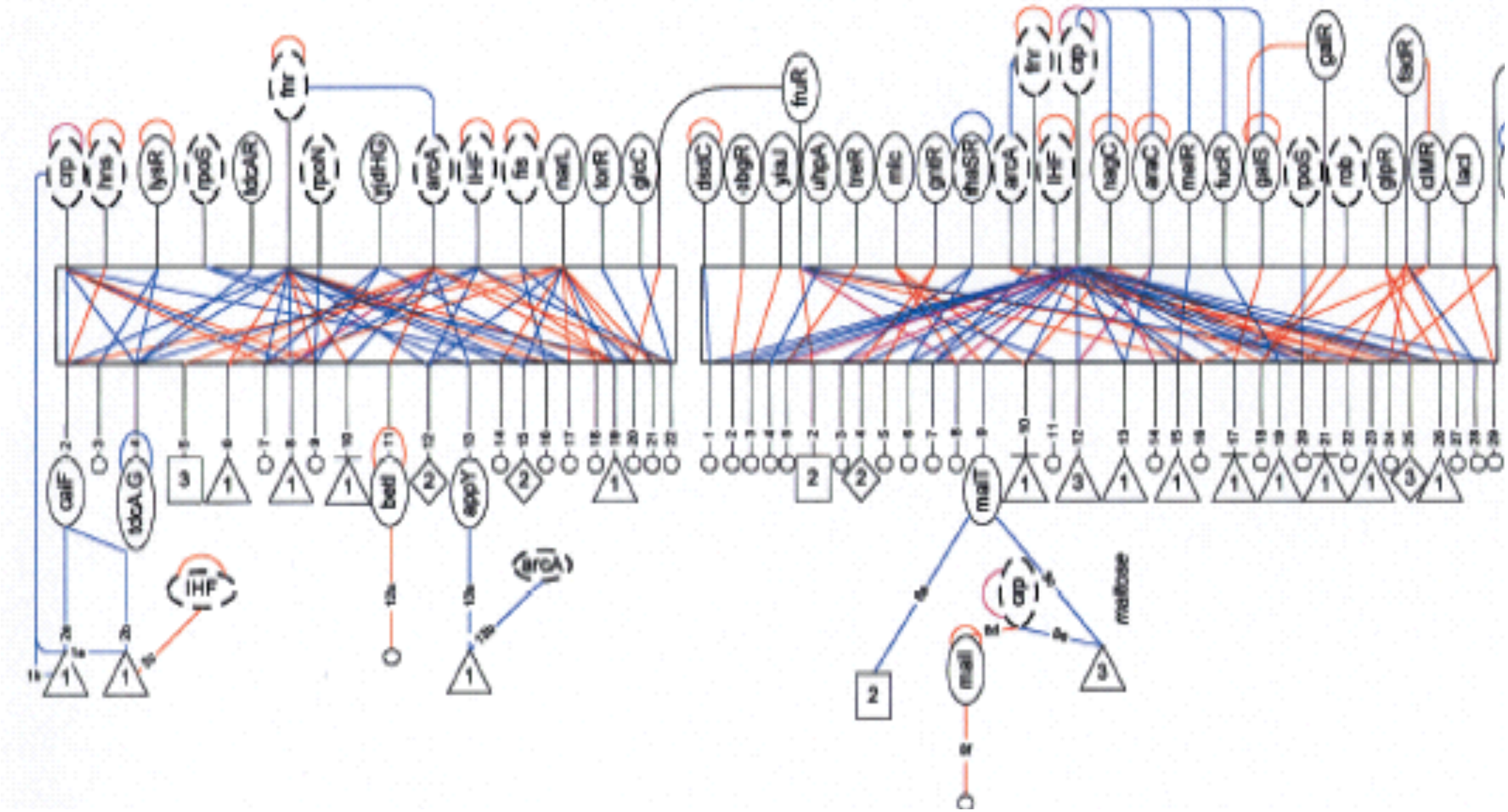
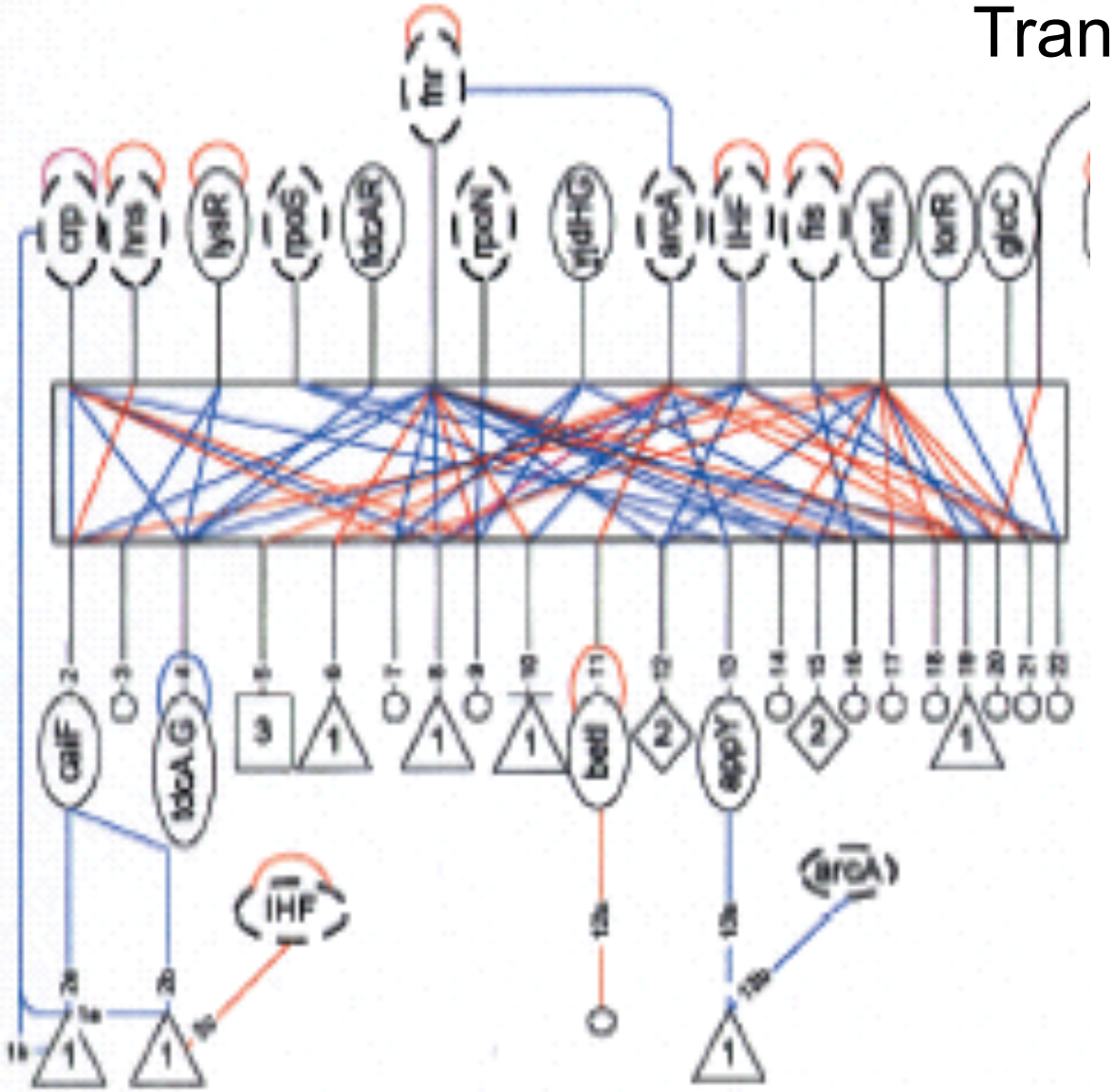


Fig. 4

1. anaerobic/aerobic metabolism DOR

2.carbon utilization DOR



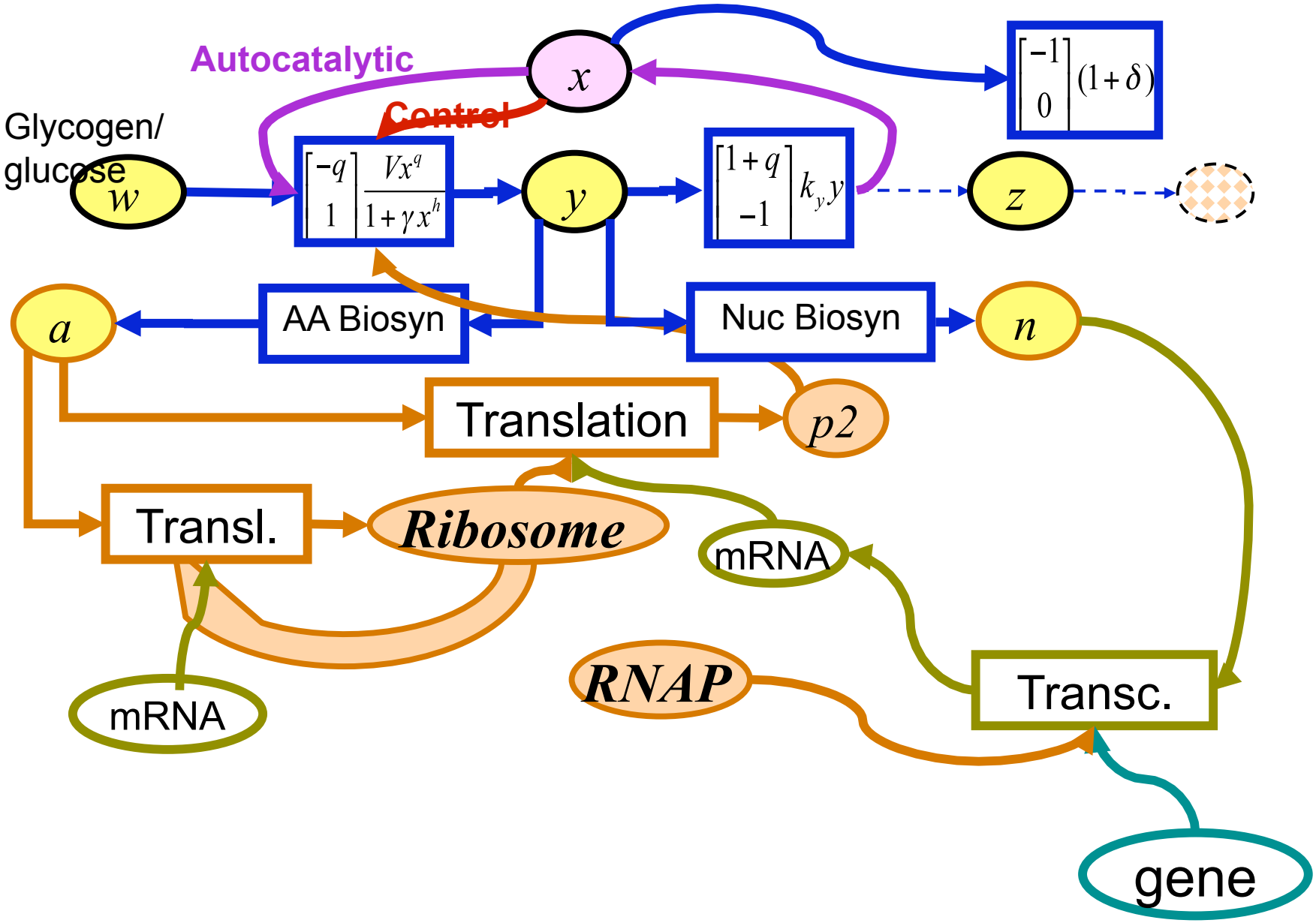


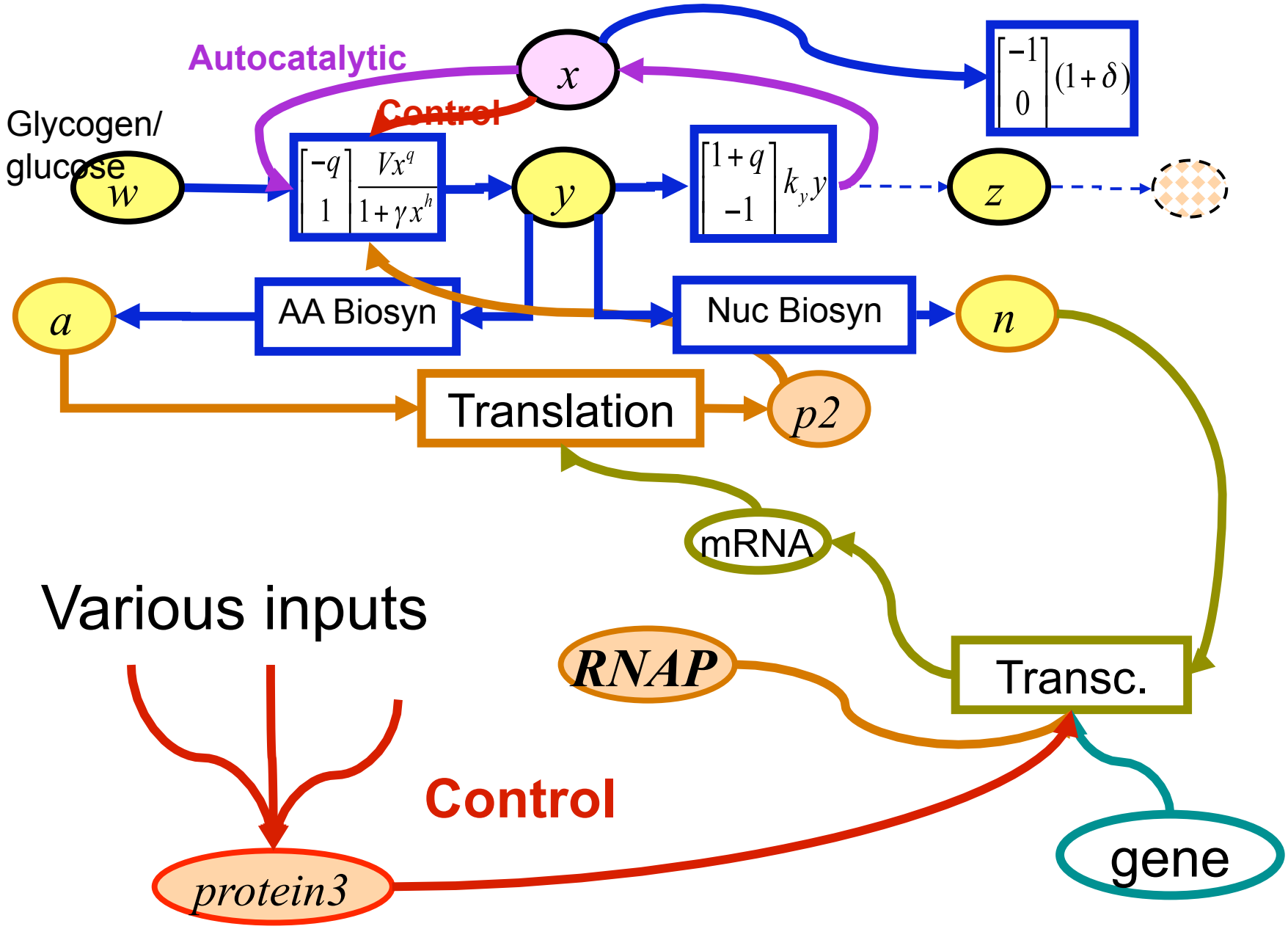
Transcription factors

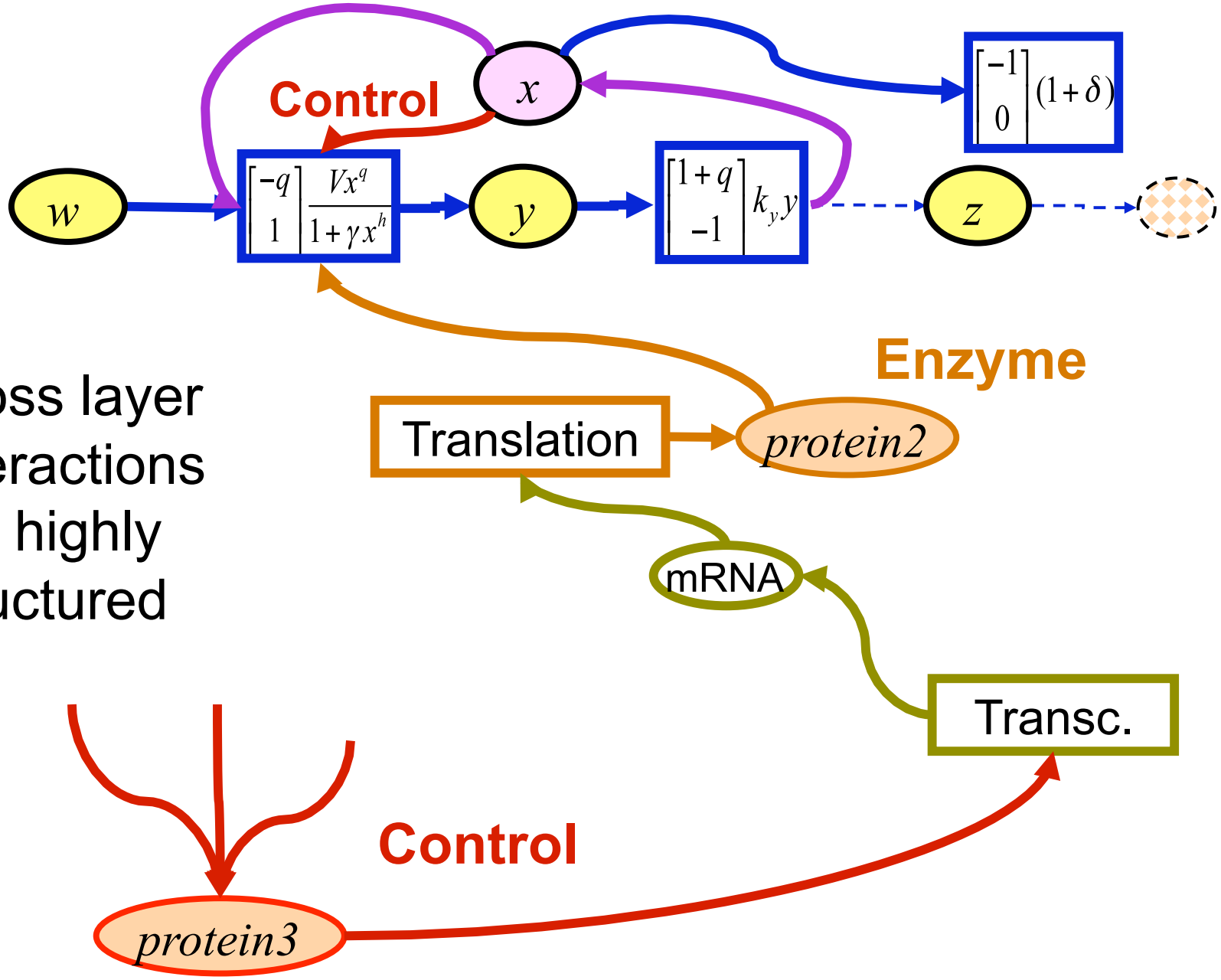
Special purpose proteins that control gene expression

Operons

Small groups of co-regulated genes

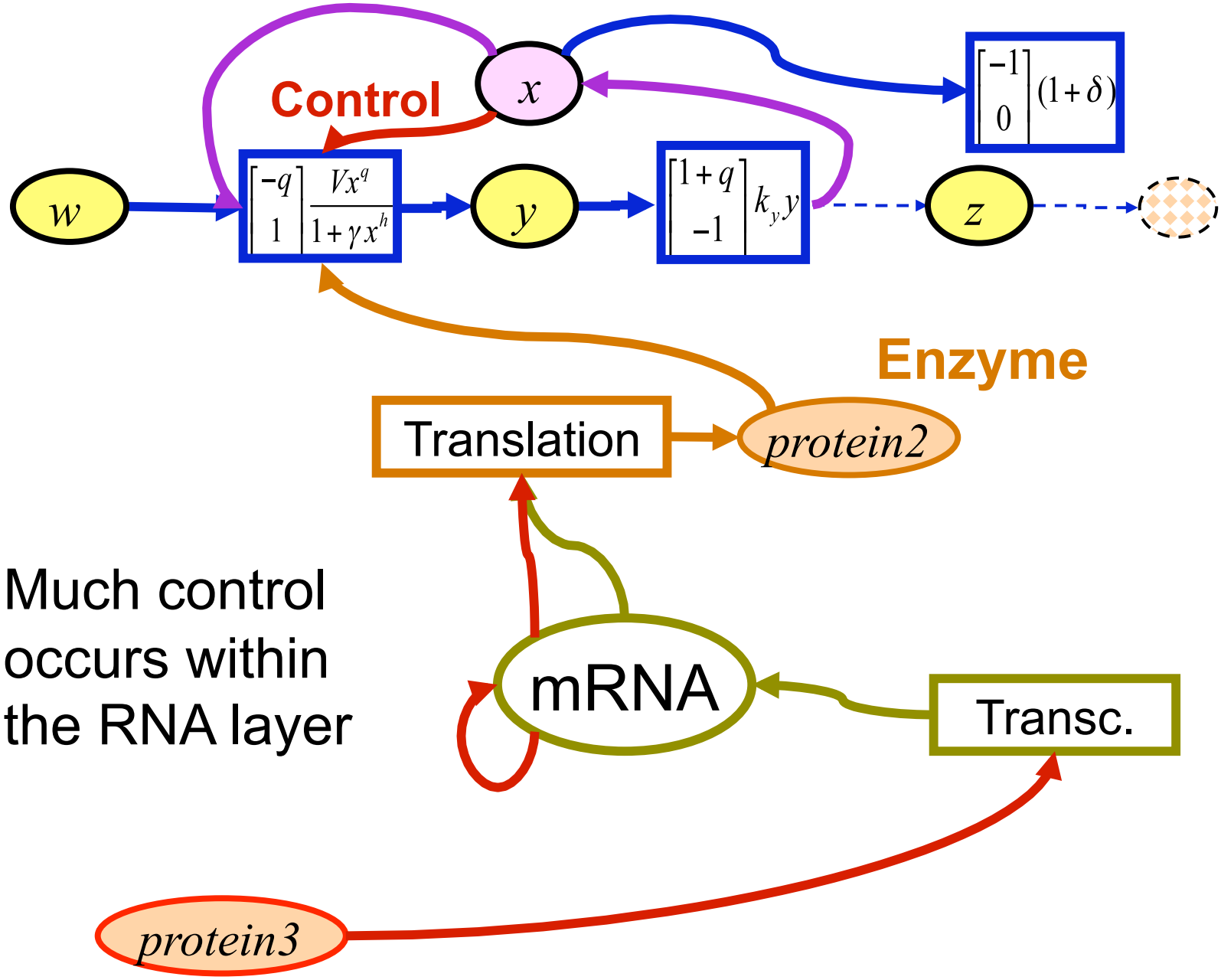

Autocatalytic
Control
Glycogen/
glucose
w
x
y
z
$\begin{bmatrix} -q \\ 1 \end{bmatrix} \frac{Vx^q}{1+\gamma x^h}$
$\begin{bmatrix} 1+q \\ -1 \end{bmatrix} k_y y$
$\begin{bmatrix} -1 \\ 0 \end{bmatrix} (1+\delta)$
a
AA Biosyn
Nuc Biosyn
n
Translation
p2
Transl.
Ribosome
mRNA
mRNA
RNAP
Transc.
gene


Autocatalytic
Control
Glycogen/
glucose
w
x
y
z
$\begin{bmatrix} -q \\ 1 \end{bmatrix} \frac{Vx^q}{1+\gamma x^h}$
$\begin{bmatrix} 1+q \\ -1 \end{bmatrix} k_y y$
$\begin{bmatrix} -1 \\ 0 \end{bmatrix} (1+\delta)$
a
AA Biosyn
Nuc Biosyn
n
Translation
p2
mRNA
Various inputs
RNAP
Transc.
Control
protein3
gene
Transcription factor



Cross layer interactions are highly structured

Transcription factor


Control
x
w
y
z
$\begin{bmatrix} -q \\ 1 \end{bmatrix} \frac{Vx^q}{1+\gamma x^h}$
$\begin{bmatrix} 1+q \\ -1 \end{bmatrix} k_y y$
$\begin{bmatrix} -1 \\ 0 \end{bmatrix} (1+\delta)$
Enzyme
Translation
protein2
Much control
occurs within
the RNA layer
mRNA
Transc.
protein3
Transcription factor

Network motifs in the transcriptional regulation network of *Escherichia coli*

Shai S. Shen-Orr[1], Ron Milo[2], Shmoolik Mangan[1] & Uri Alon[1,2]

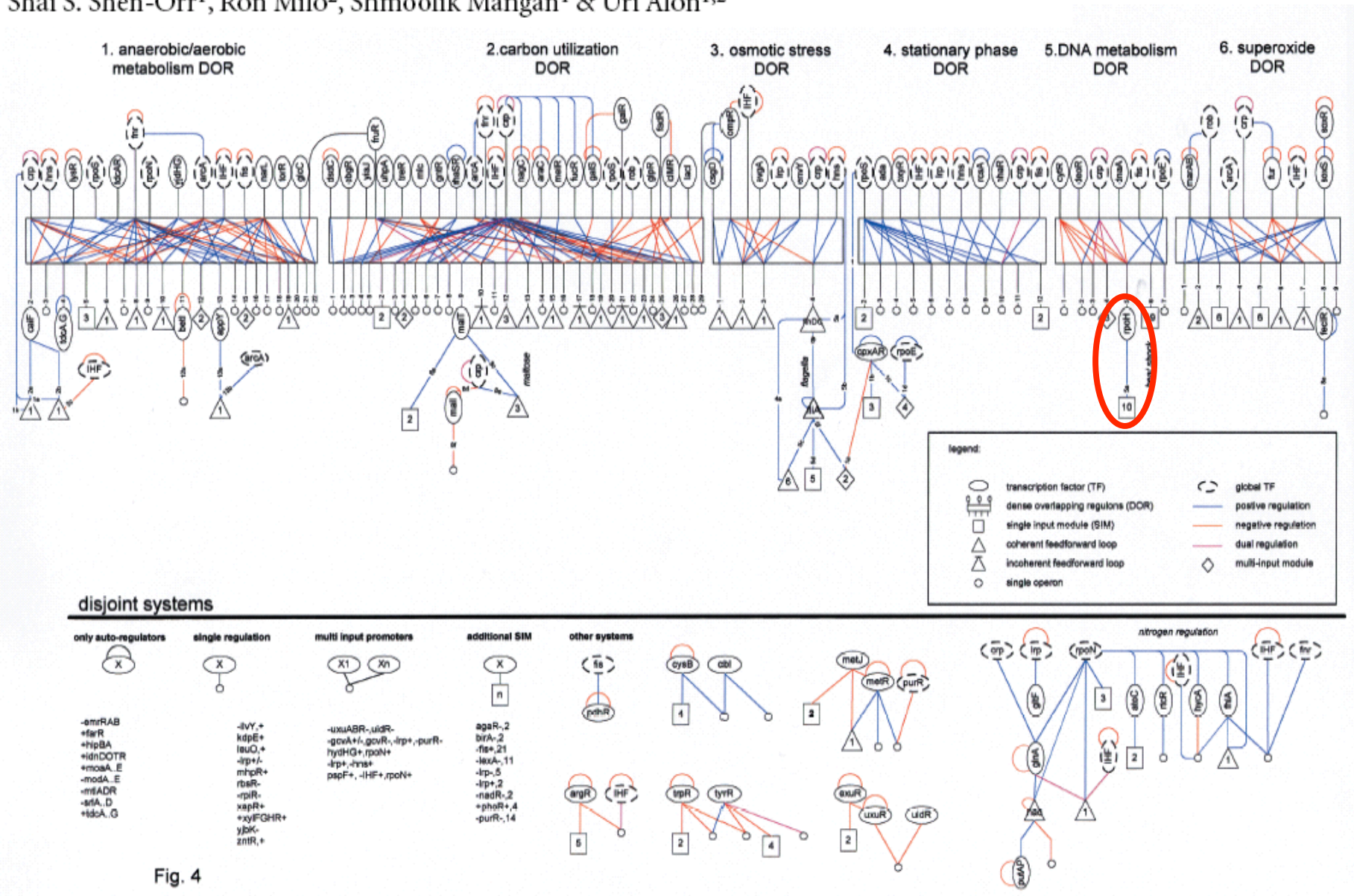
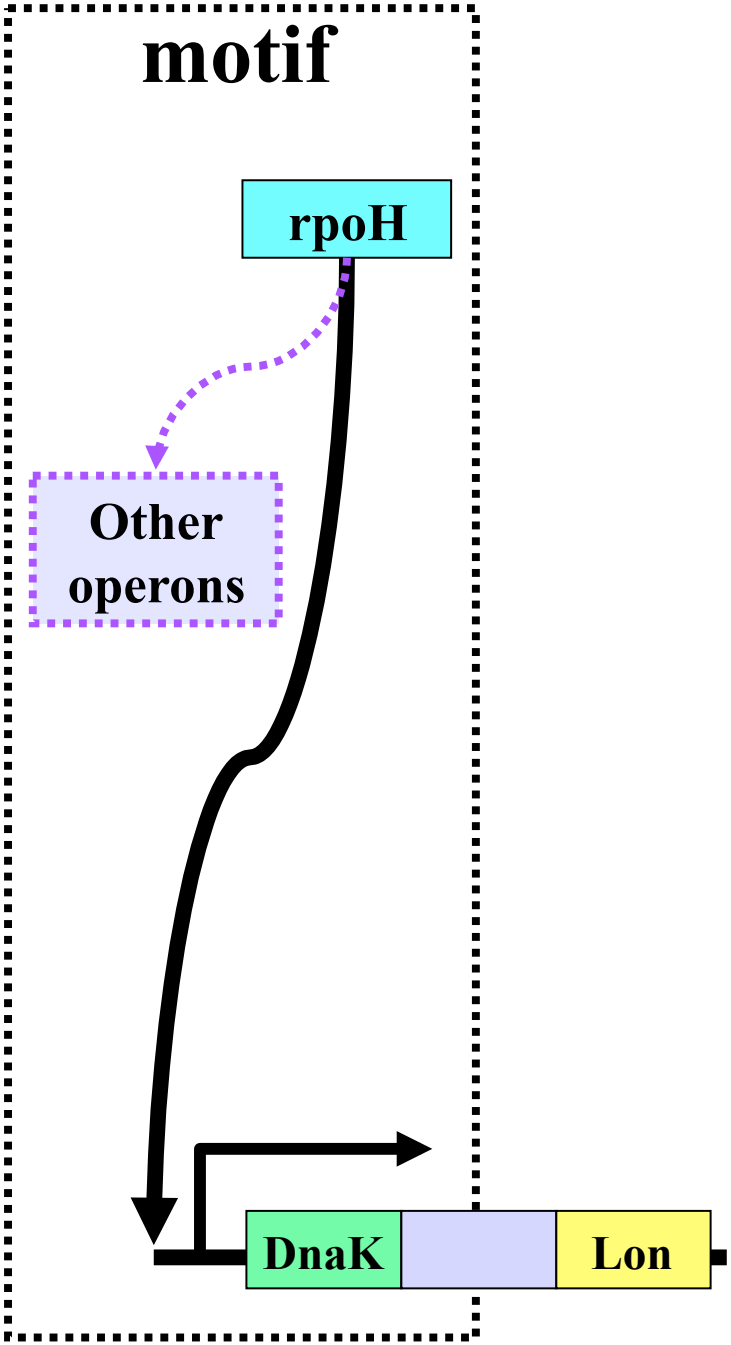
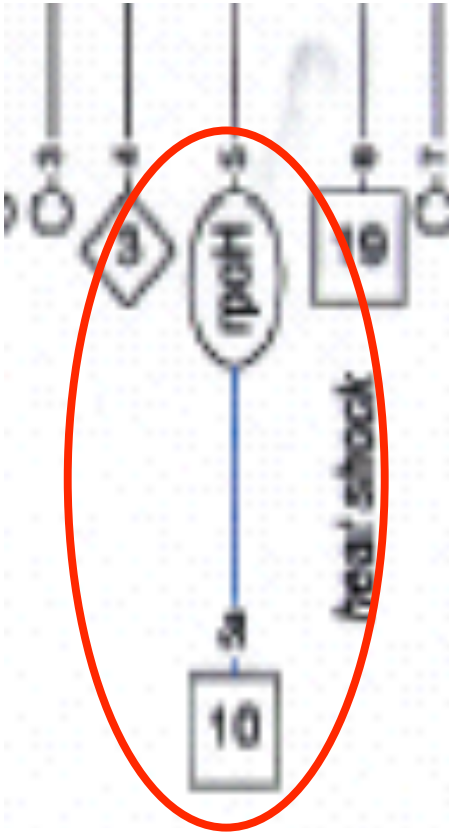


Fig. 4



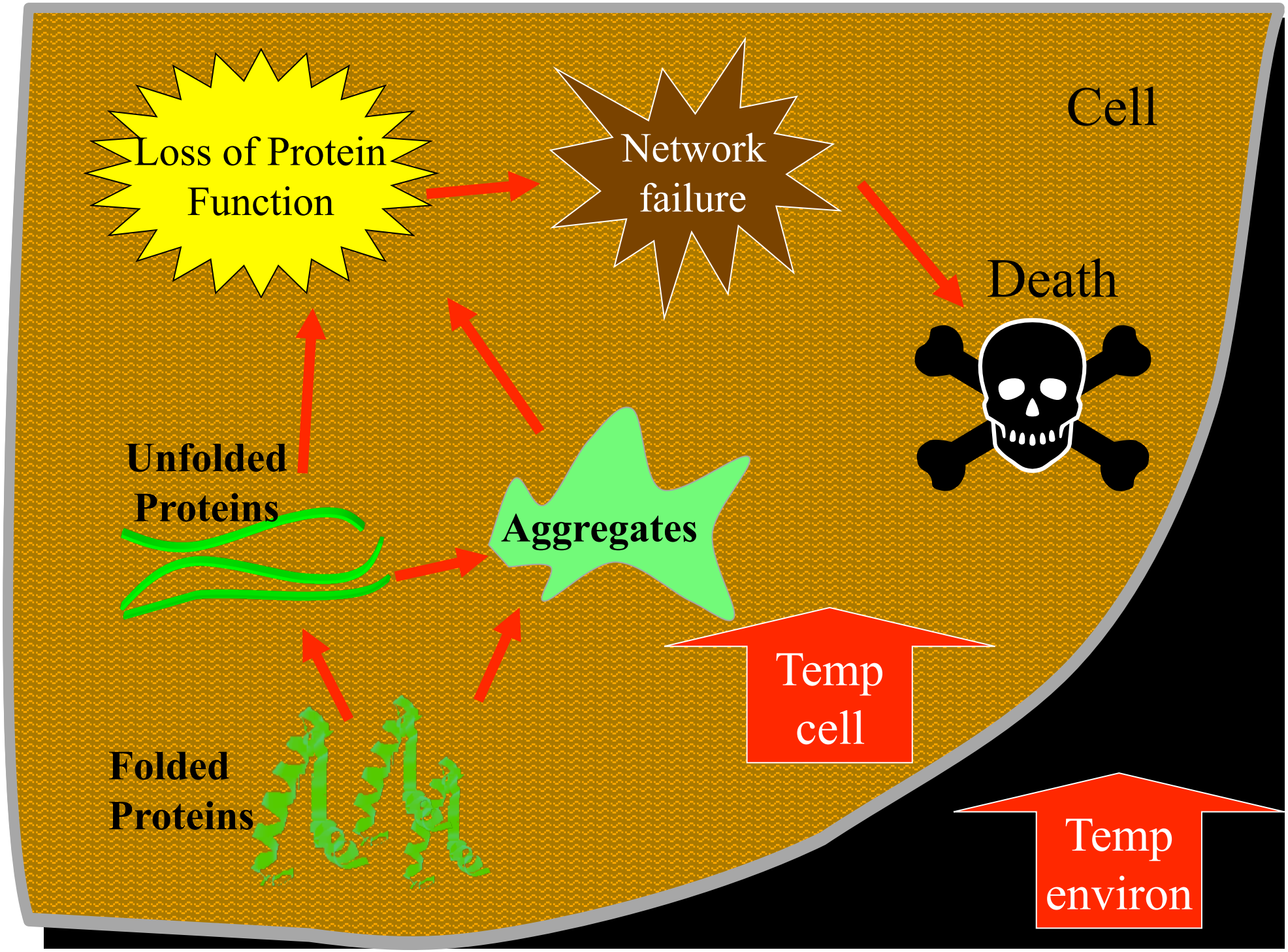


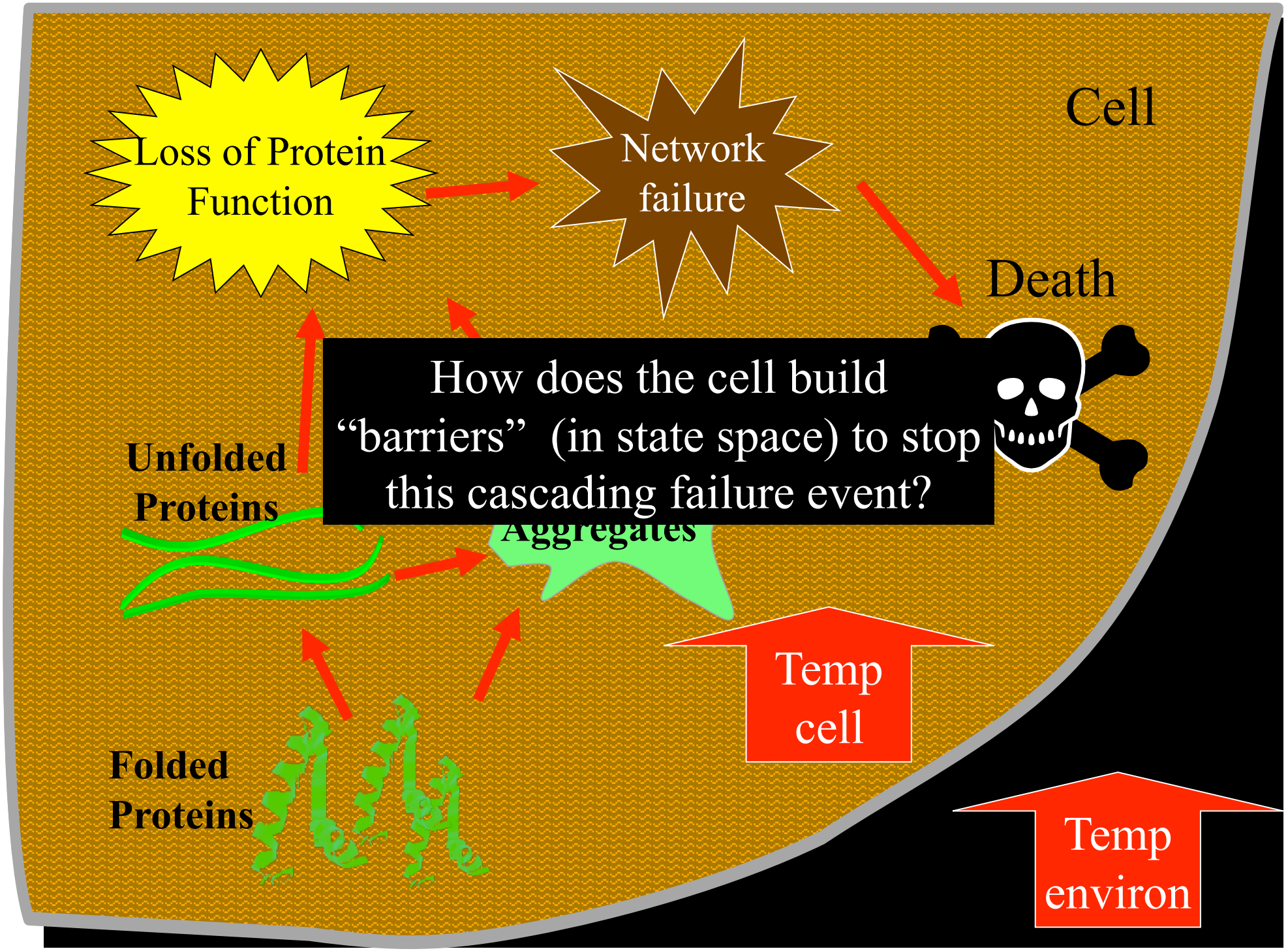
See El-Samad, Kurata, et al…
PNAS, PLOS CompBio

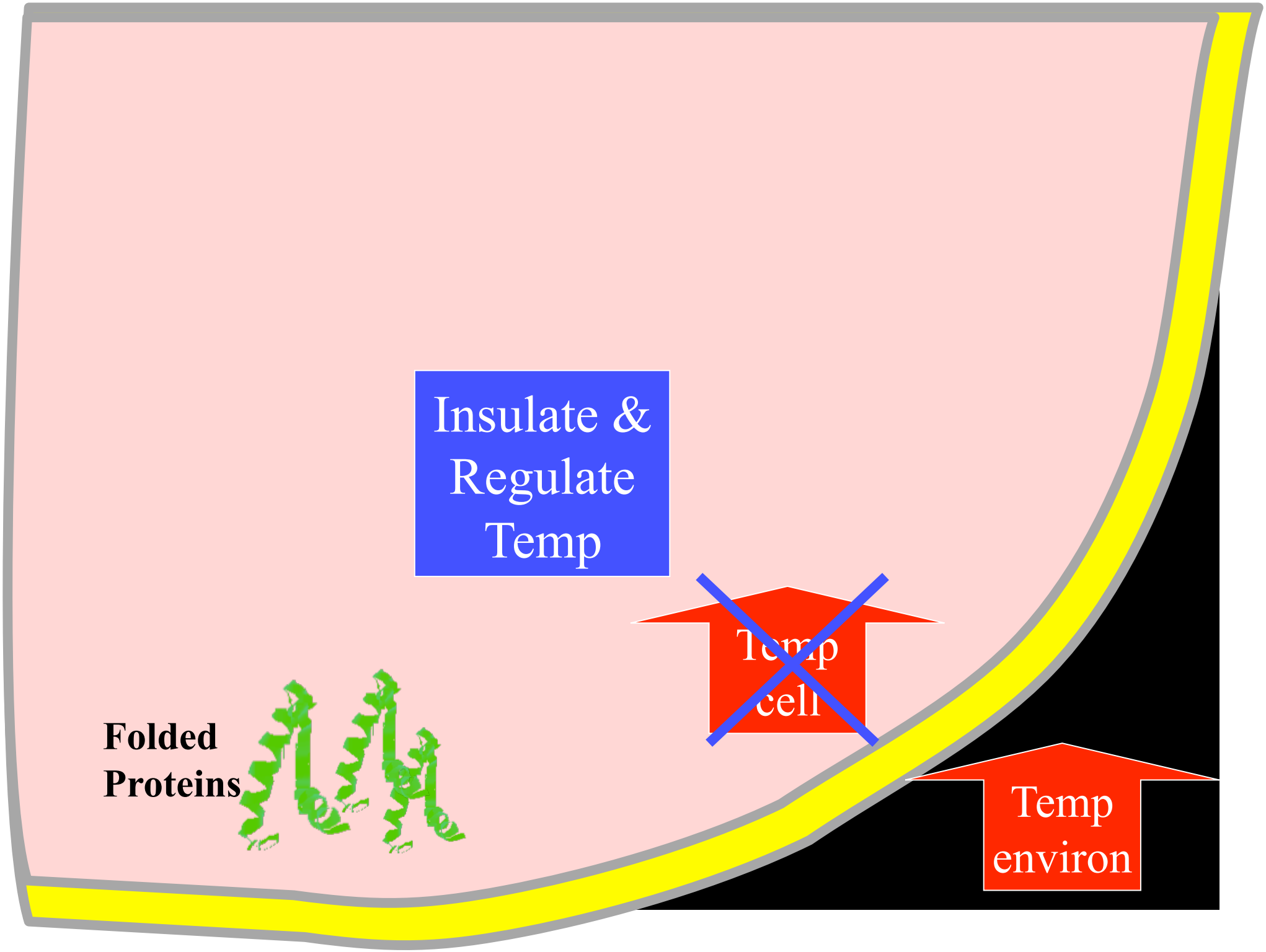
E. Coli cytoplasmic

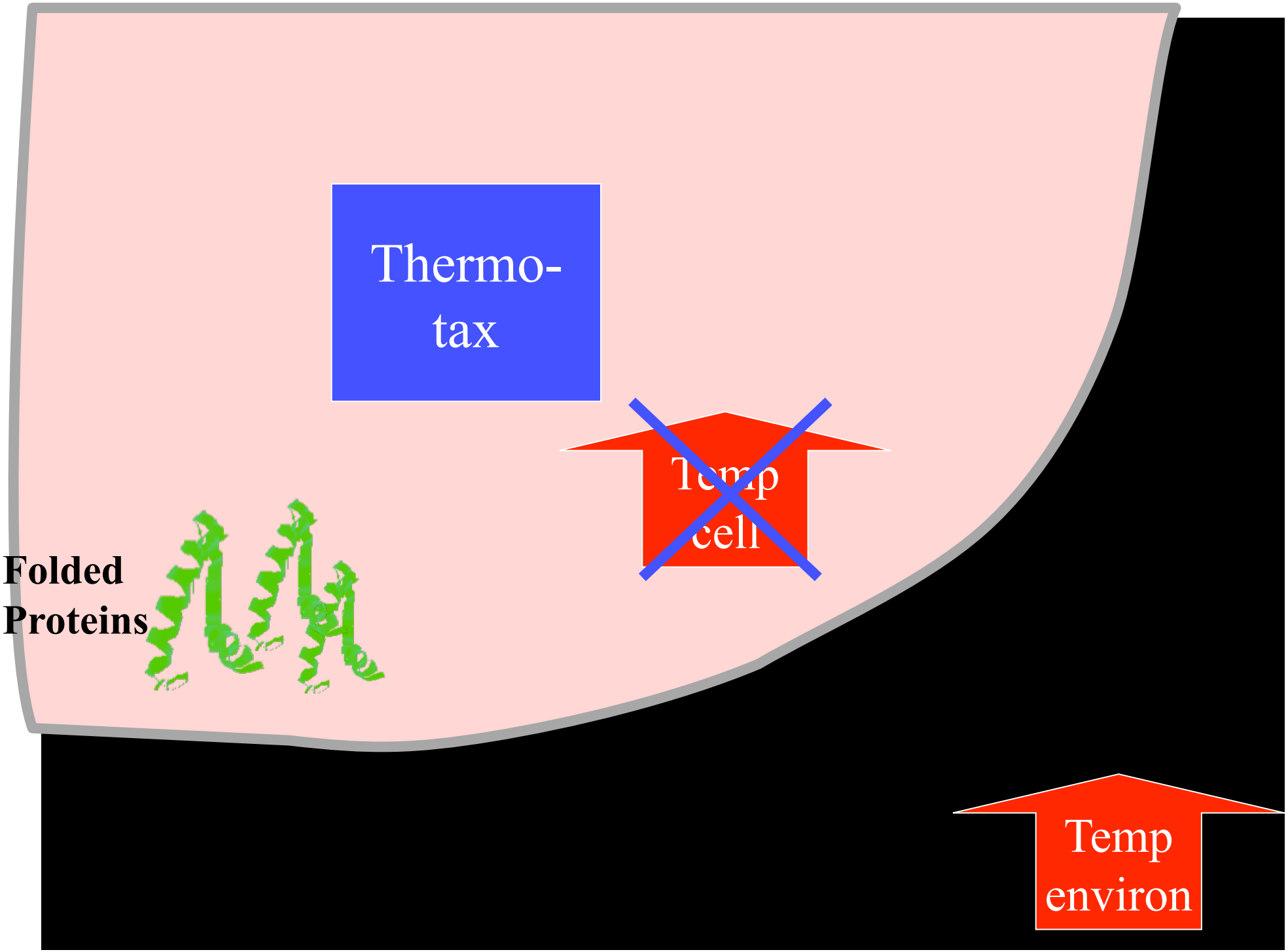
heat shock

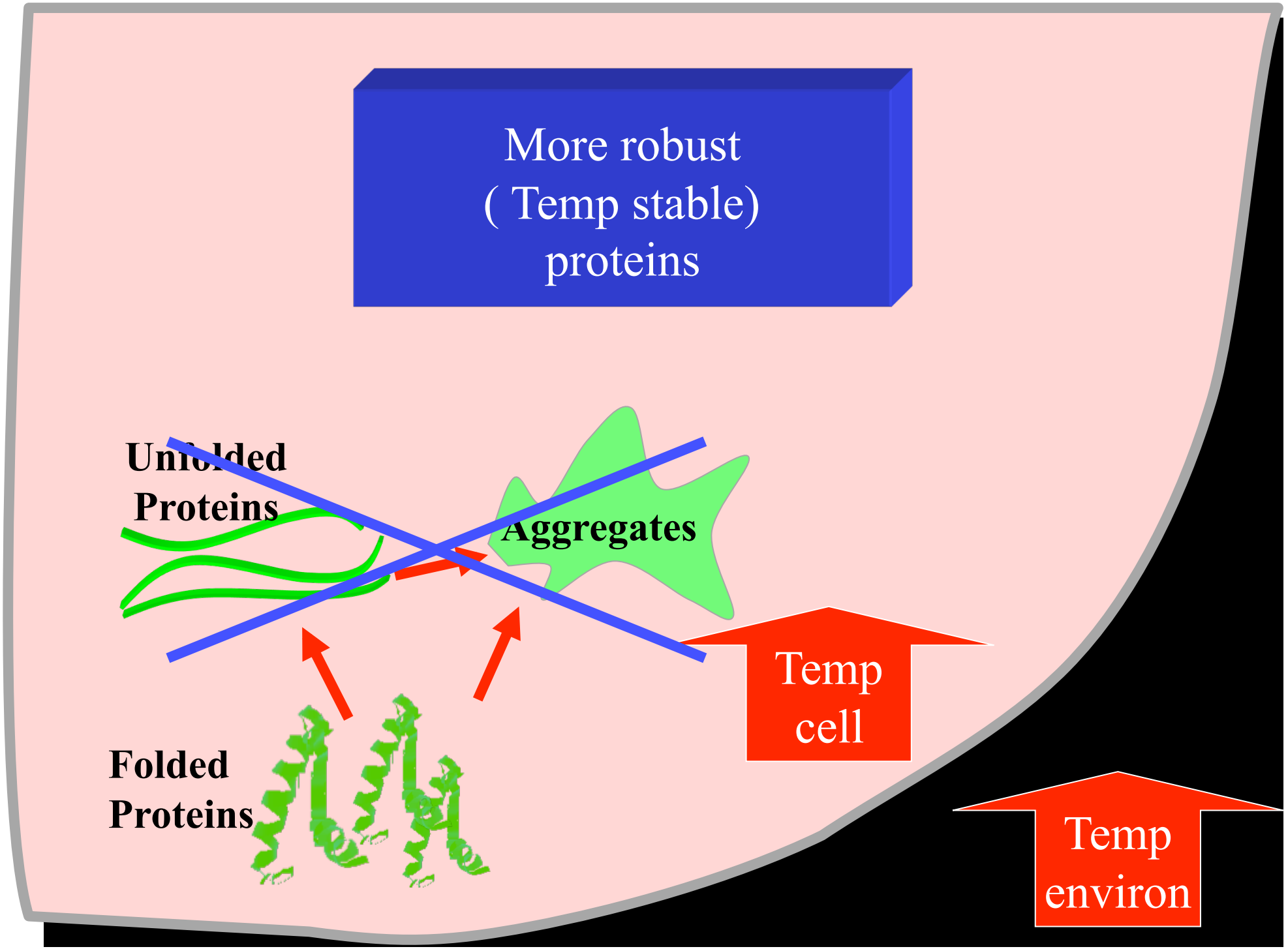
A control system in many layers


Cell
Loss of Protein Function
Network failure
Death
Unfolded Proteins
Aggregates
Folded Proteins
Temp cell
Temp environ

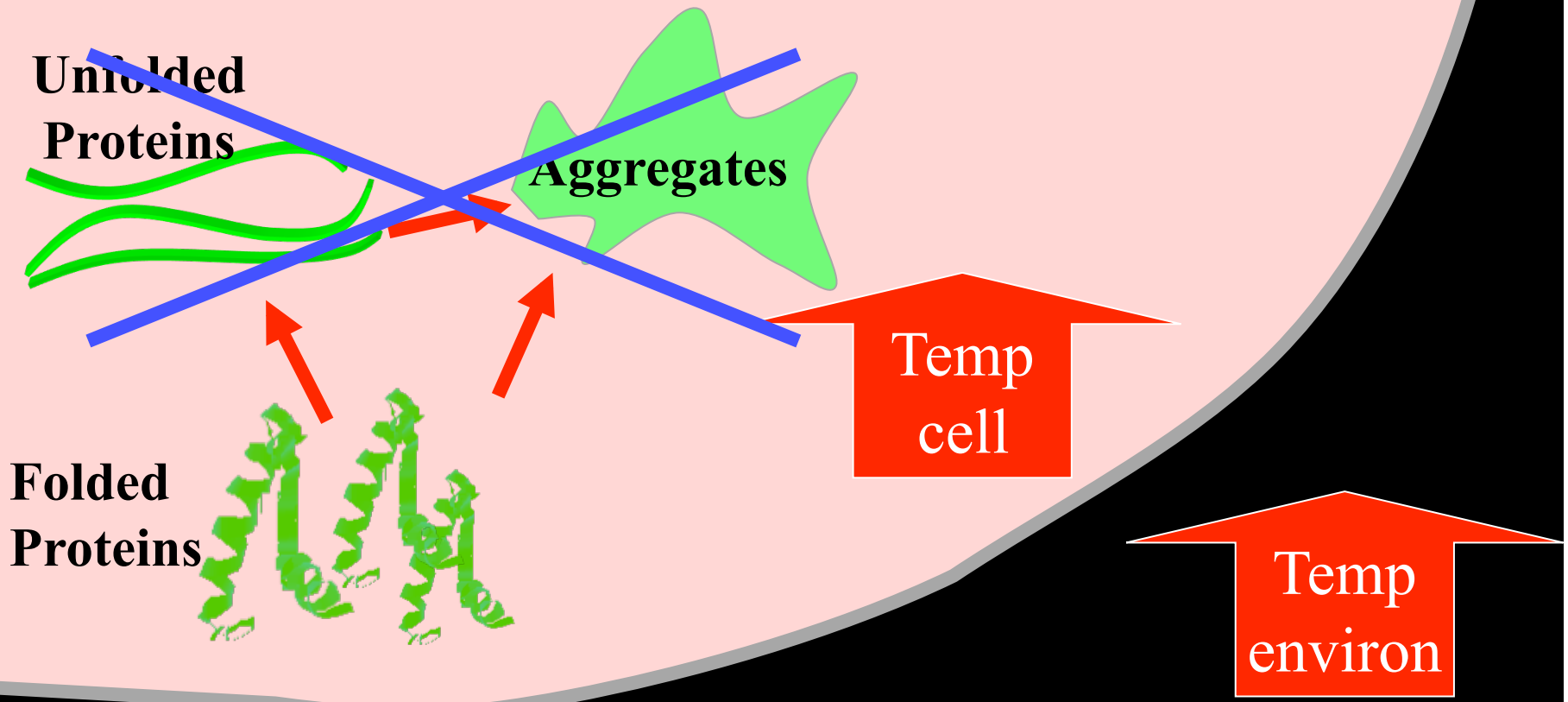

Cell
Loss of Protein Function
Network failure
Death
How does the cell build "barriers" (in state space) to stop this cascading failure event?
Unfolded Proteins
Aggregates
Folded Proteins
Temp cell
Temp environ


Insulate &
Regulate
Temp
Temp
cell
Temp
environ
Folded
Proteins

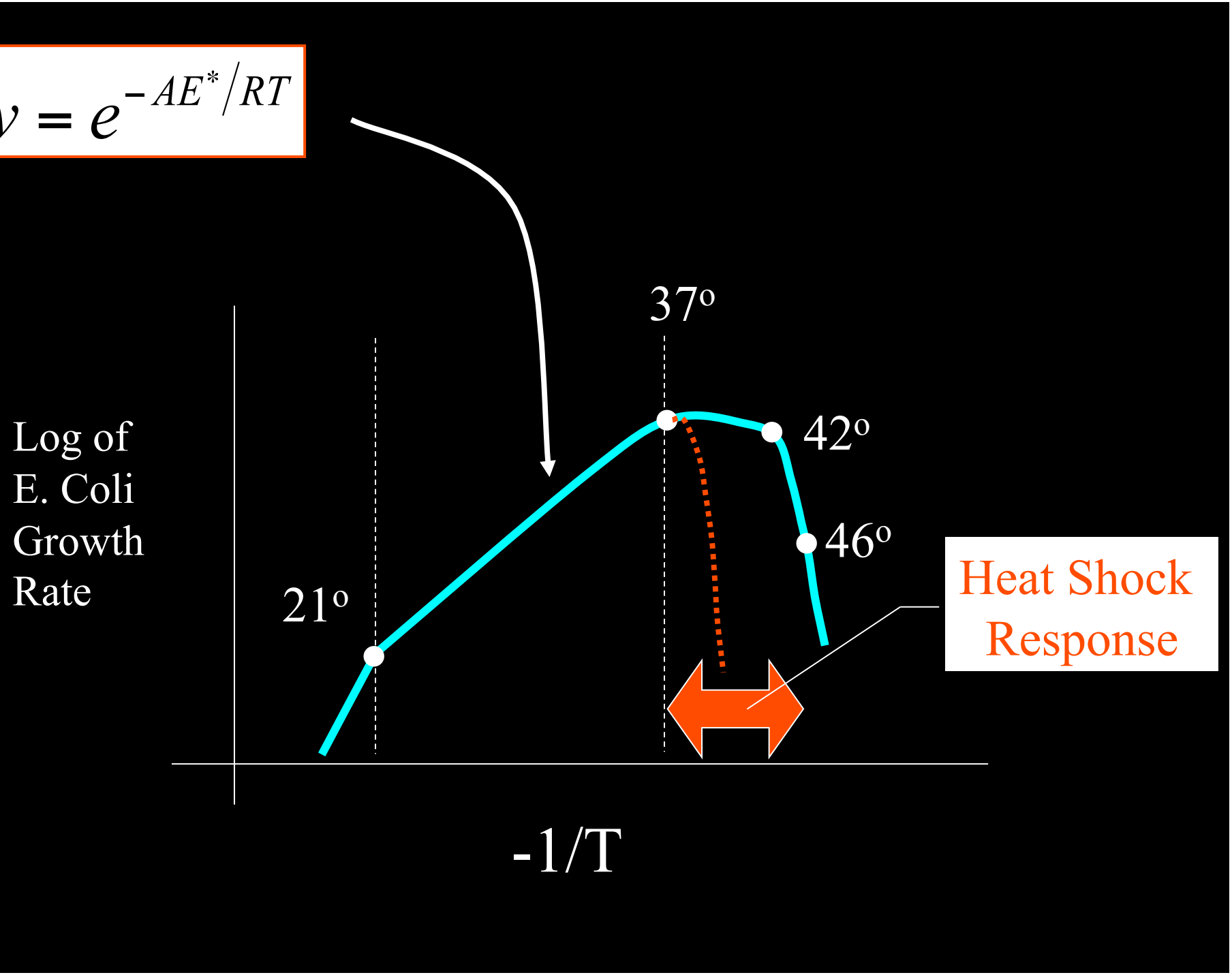

Thermo-
tax
Temp
cell
Folded
Proteins
Temp
environ


More robust
(Temp stable)
proteins
Unfolded
Proteins
Aggregates
Temp
cell
Folded
Proteins
Temp
environ

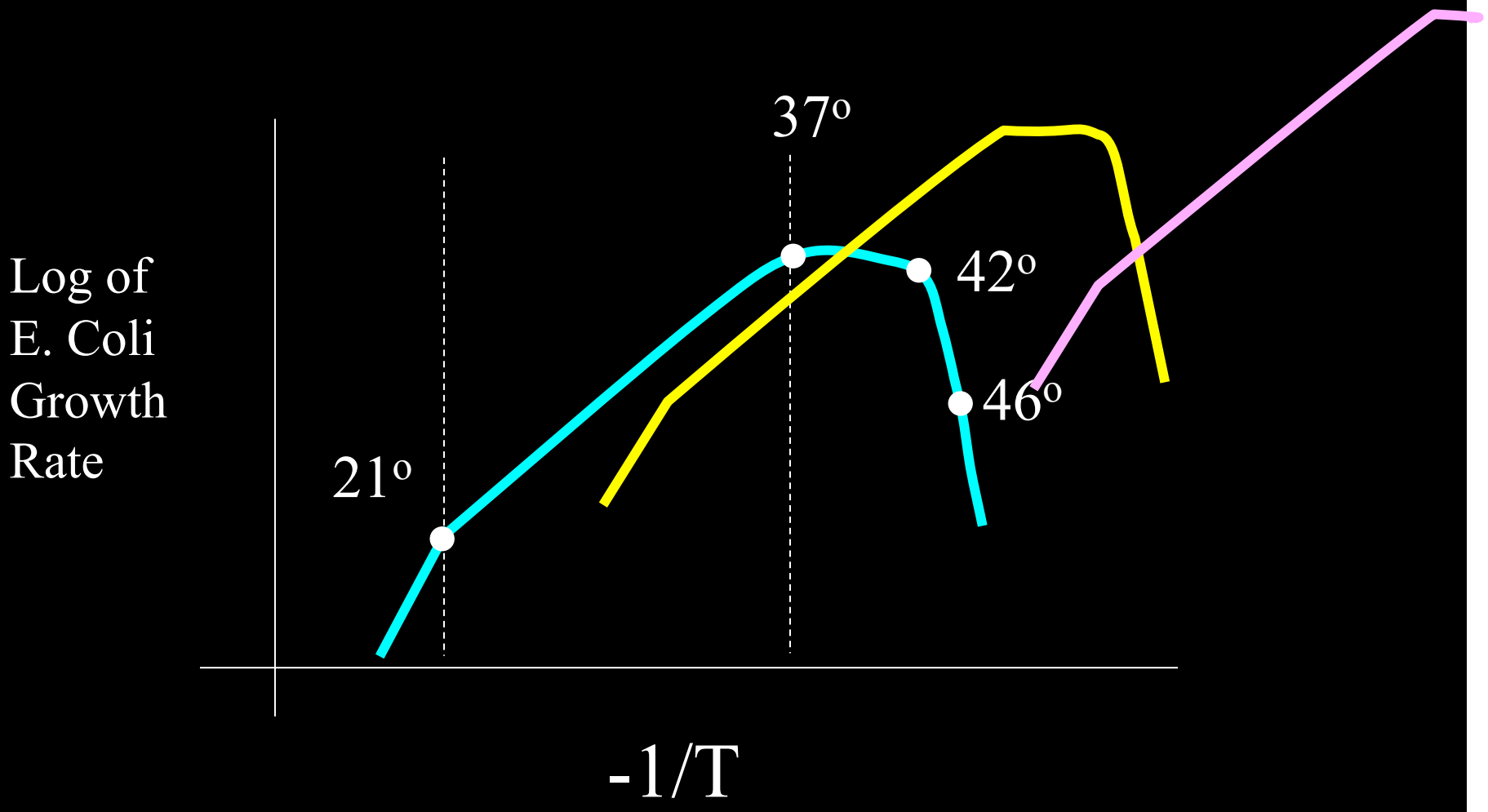
- Key proteins can have multiple (allelic or paralogous) variants
- Allelic variants allow populations to adapt
- Regulated multiple gene loci allow individuals to adapt



$$v = e^{-AE^*/RT}$$


37o
42o
46o
21o
Log of
E. Coli
Growth
Rate
Heat Shock
Response
-1/T

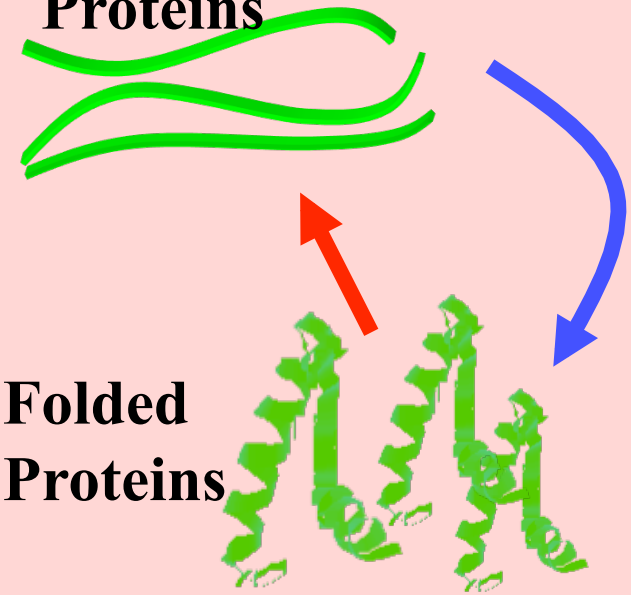
Robustness/performance tradeoff?



Refold denatured proteins

Heat shock response involves complex feedback and feedforward control.

Unfolded Proteins



Folded Proteins

Temp cell

Temp environ

Alternative strategies

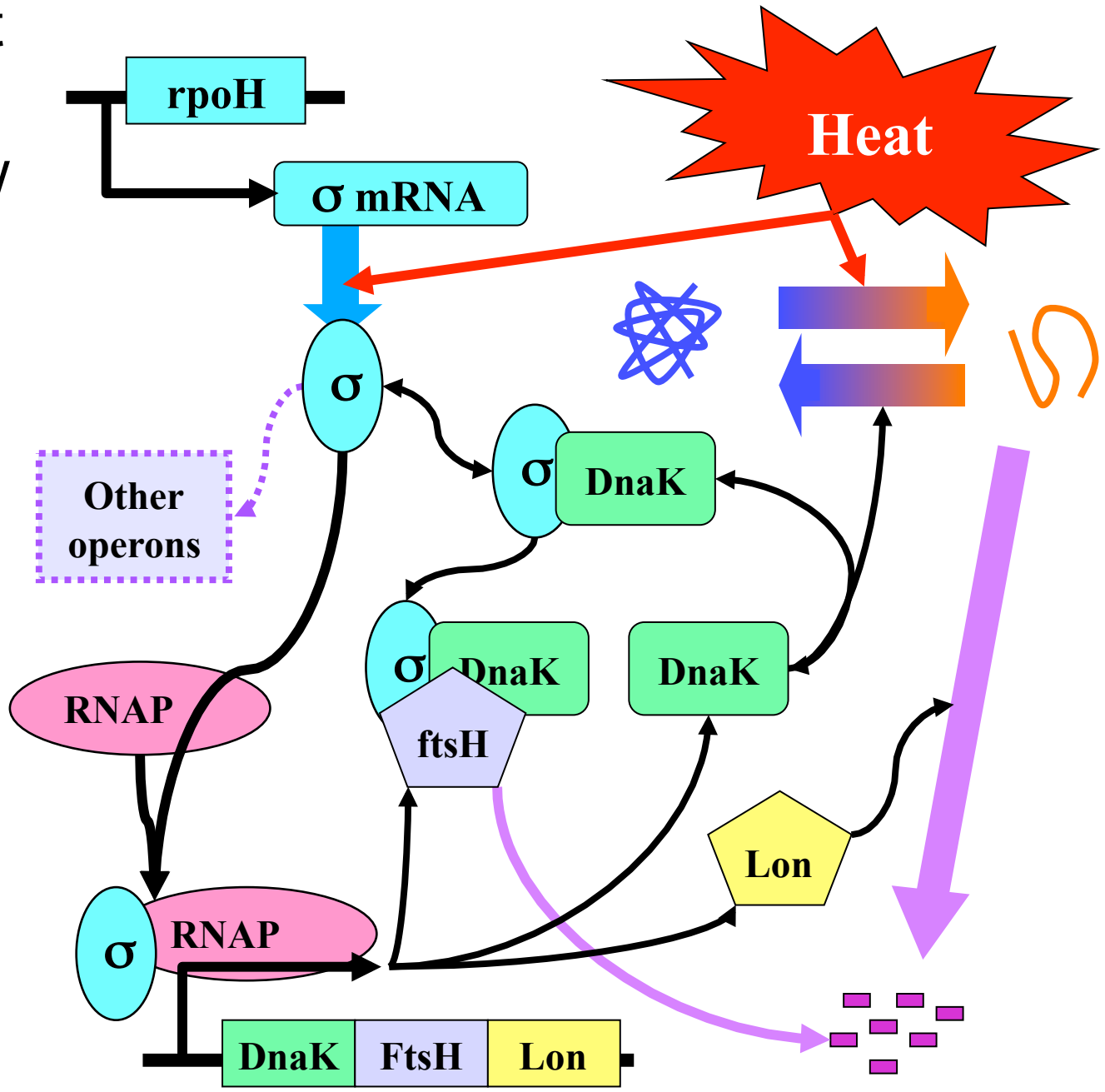
- Robust/redundant proteins
  - Temperature stability
  - Allelic variants
  - Paralogous isozymes
- Regulate temperature
- Thermotax
- Heat shock response
  - Up regulate chaperones and proteases
  - Refold or degraded denatured proteins

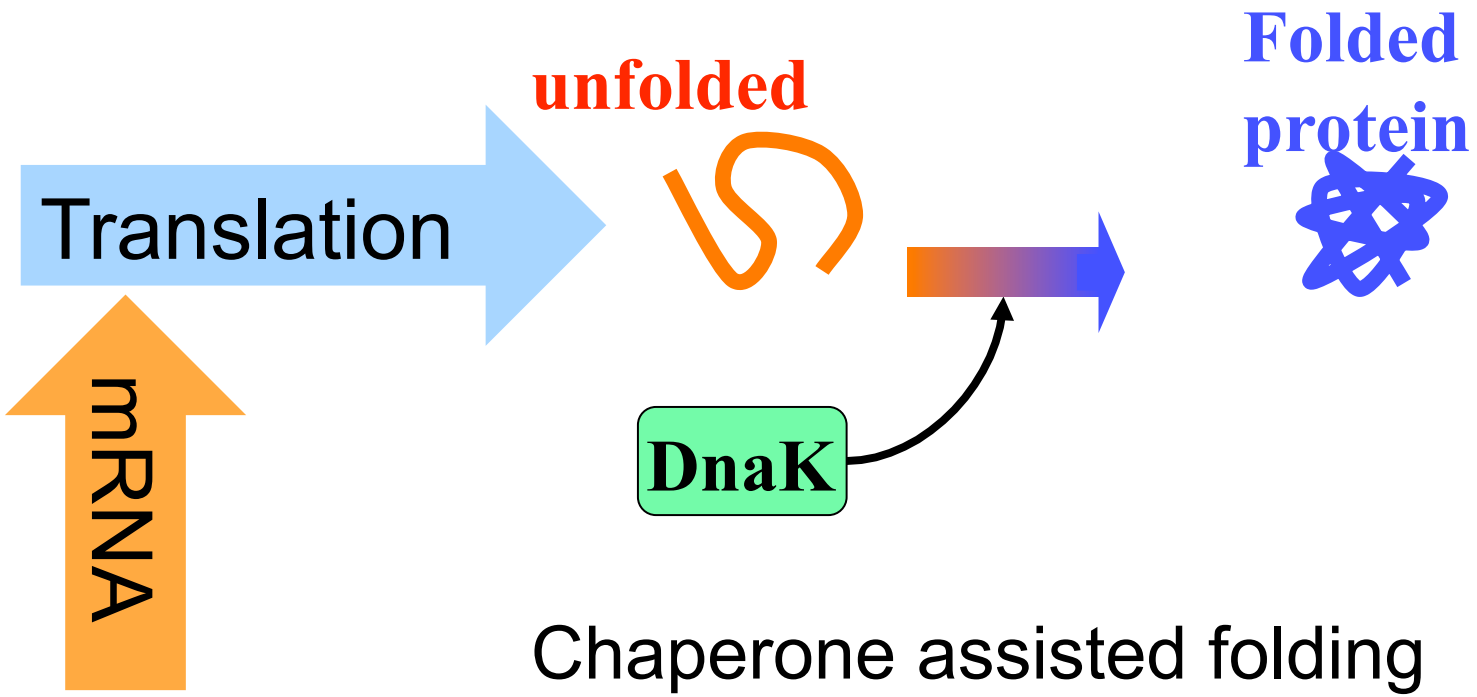
Why does biology (and advanced technology) overwhelmingly opt for the complex control systems instead of just robust components?

This is the sort of cartoon that biologists draw

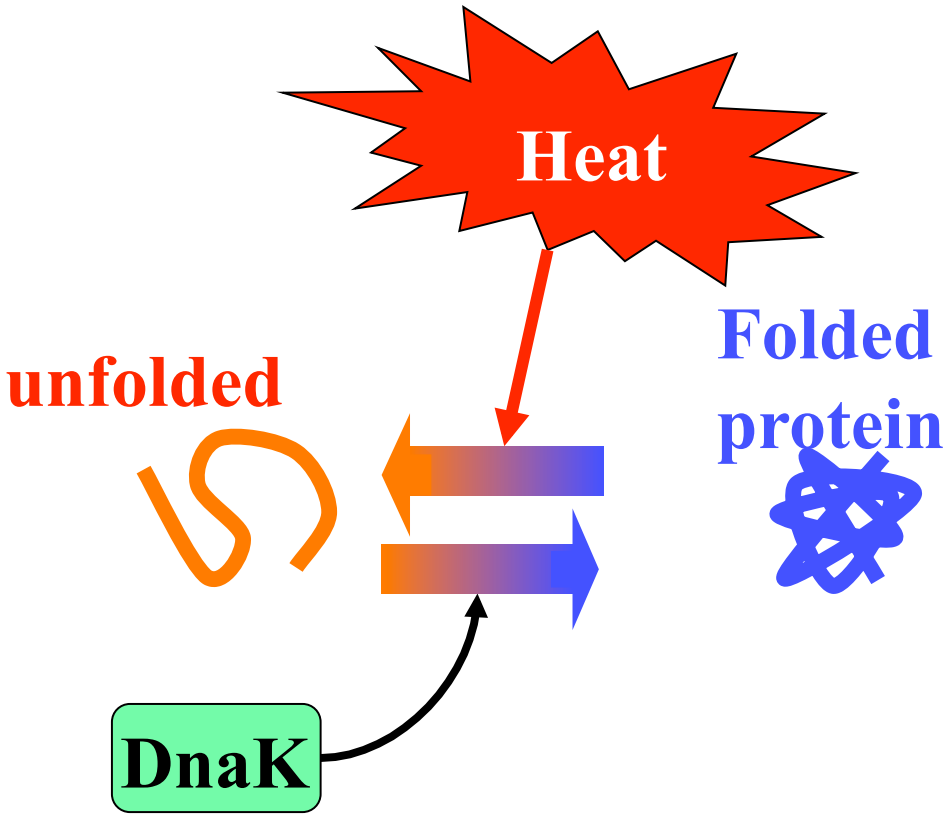
Much to learn

Briefly parse and extract lessons

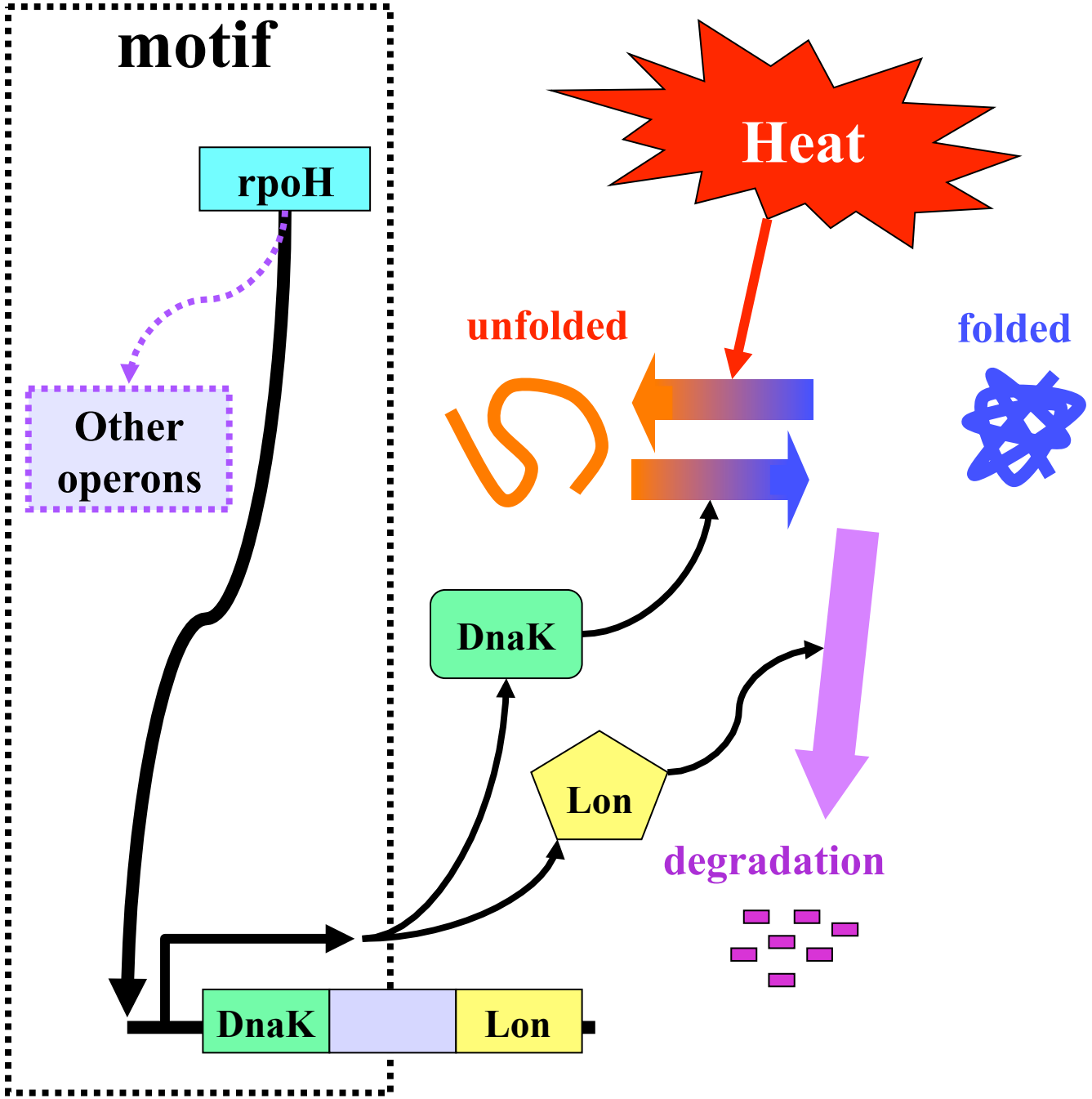


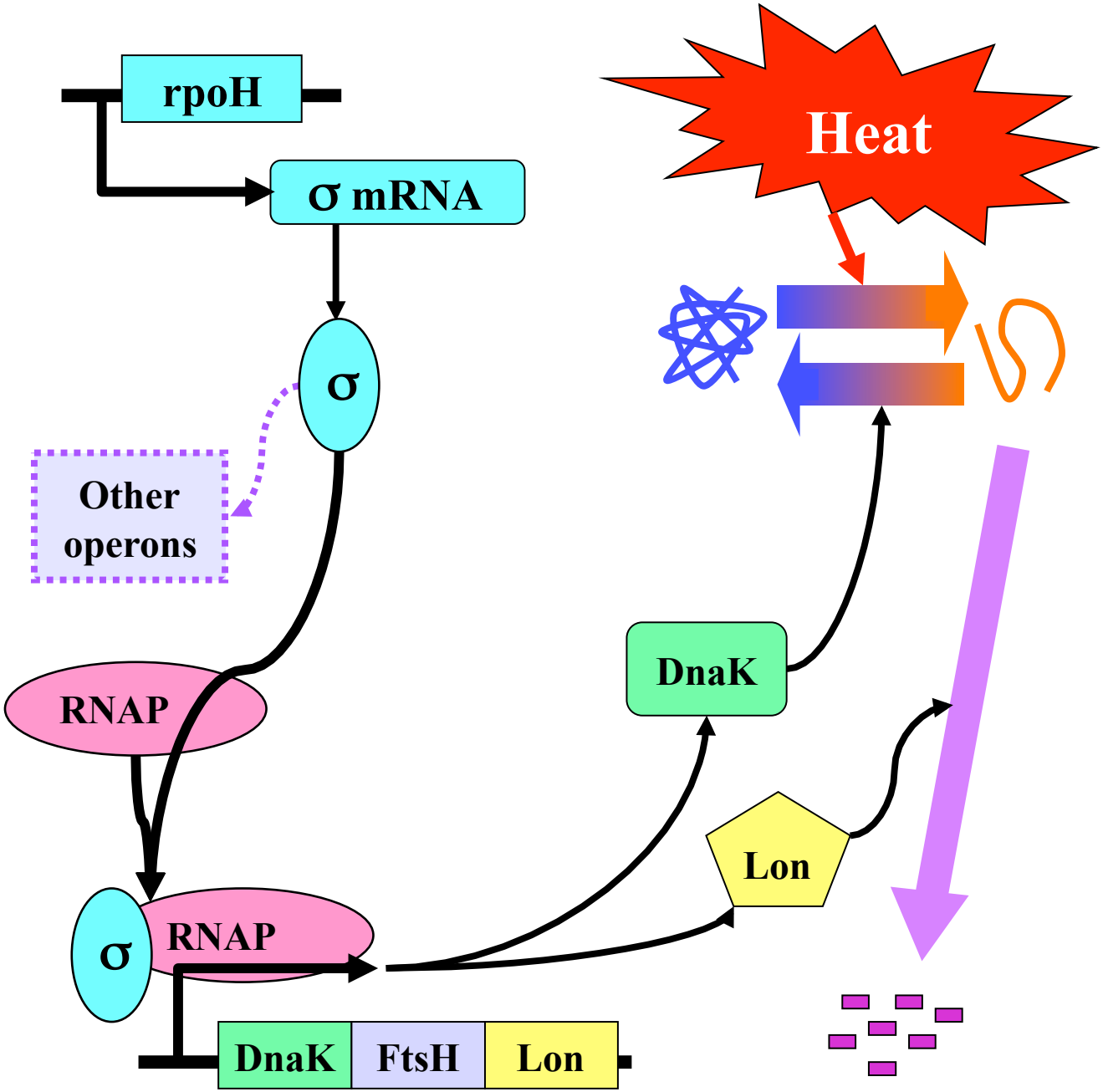


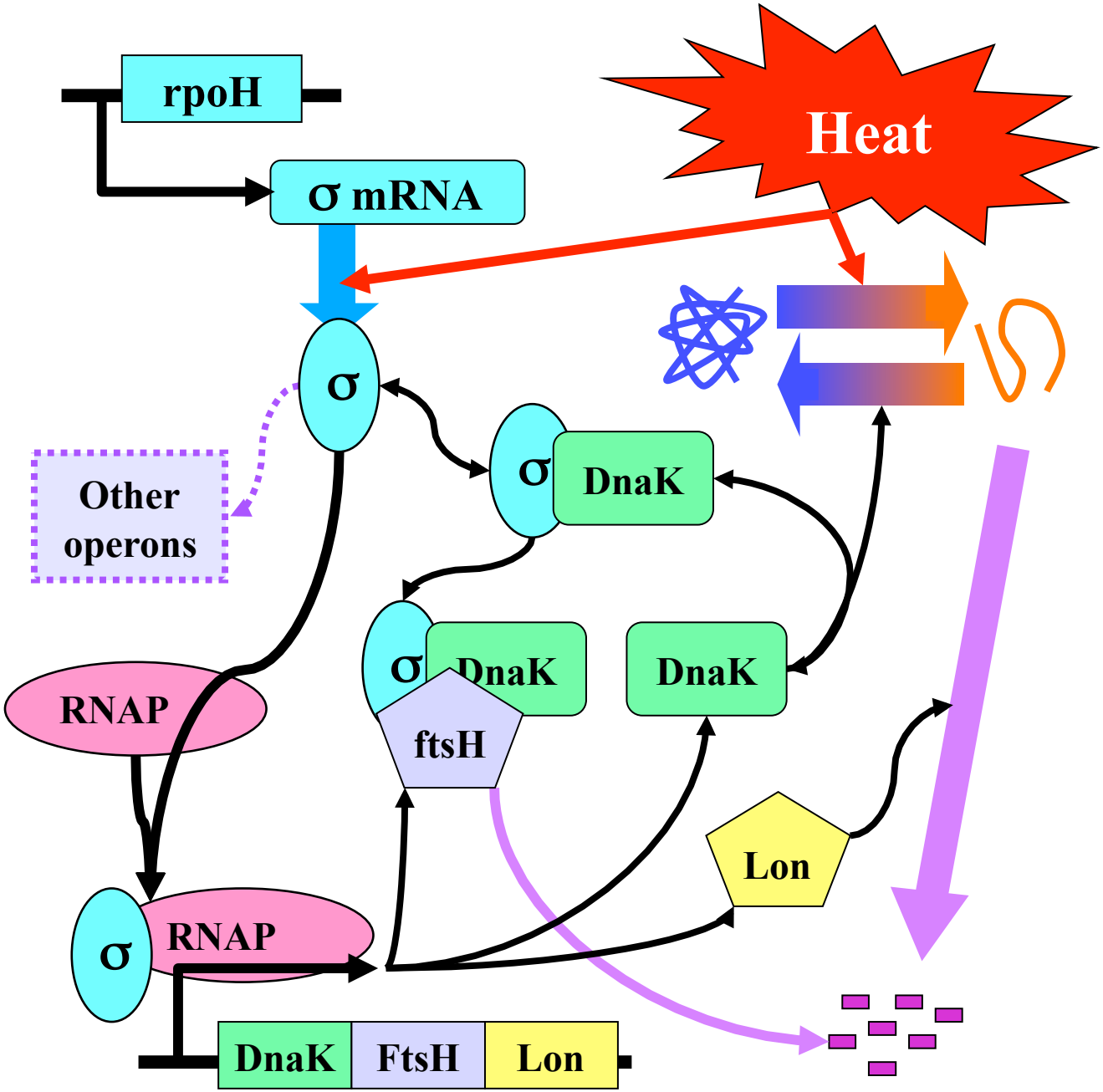
Chaperone assisted folding
(another catalyzed reaction)

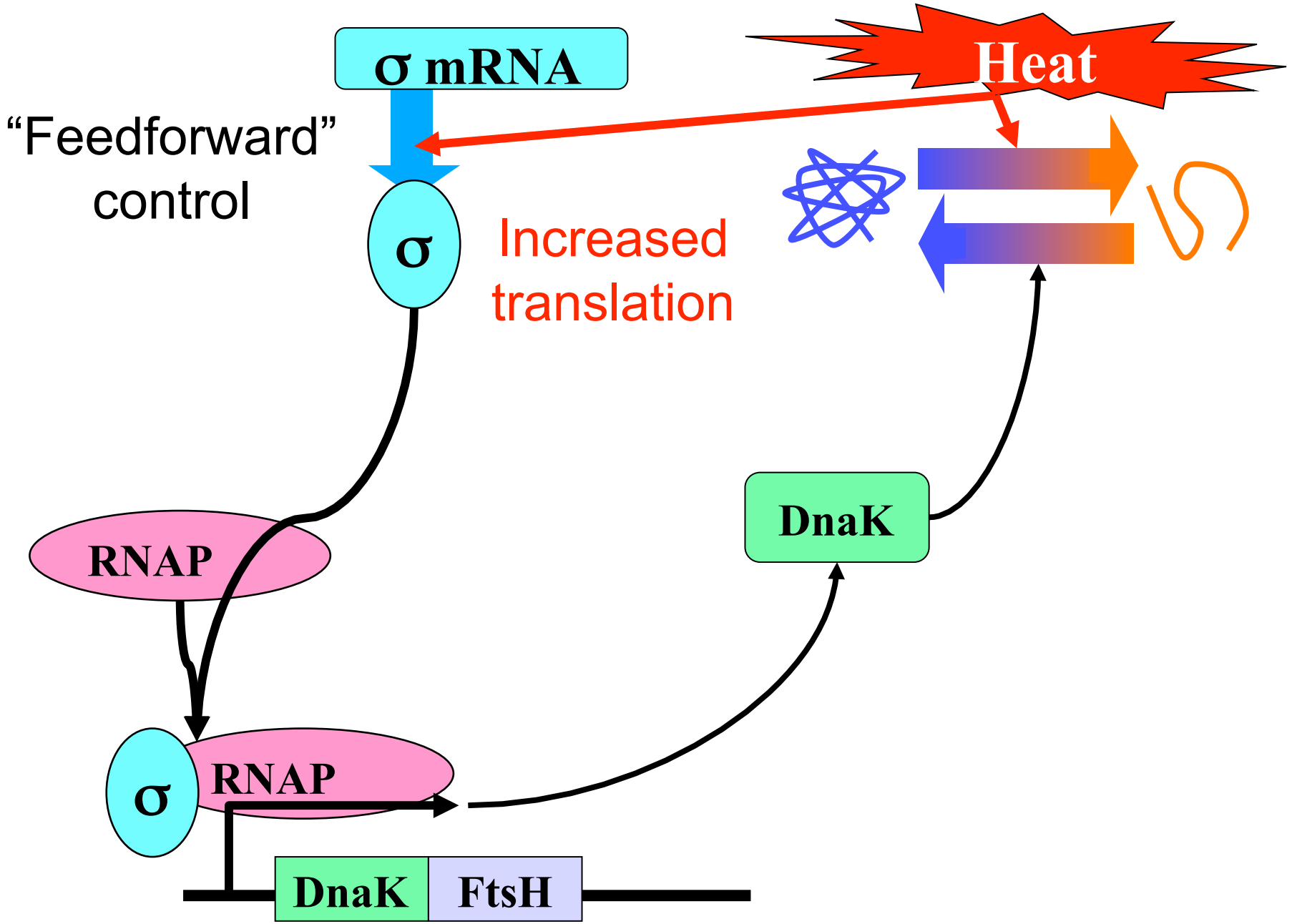


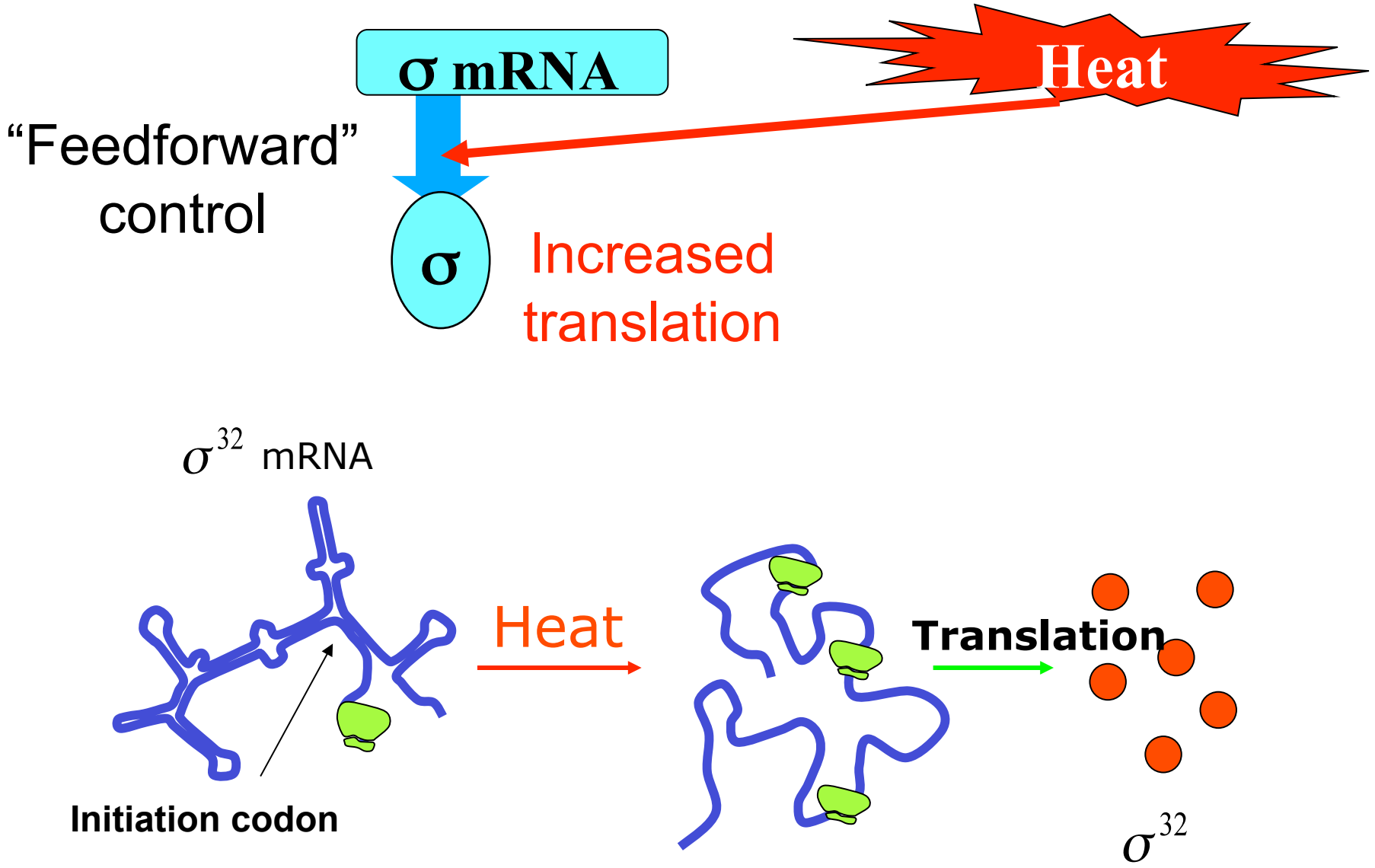
Chaperone assisted refolding
(another catalyzed reaction)


motif
rpoH
Other operons
Heat
unfolded
folded
DnaK
Lon
degradation
DnaK
Lon

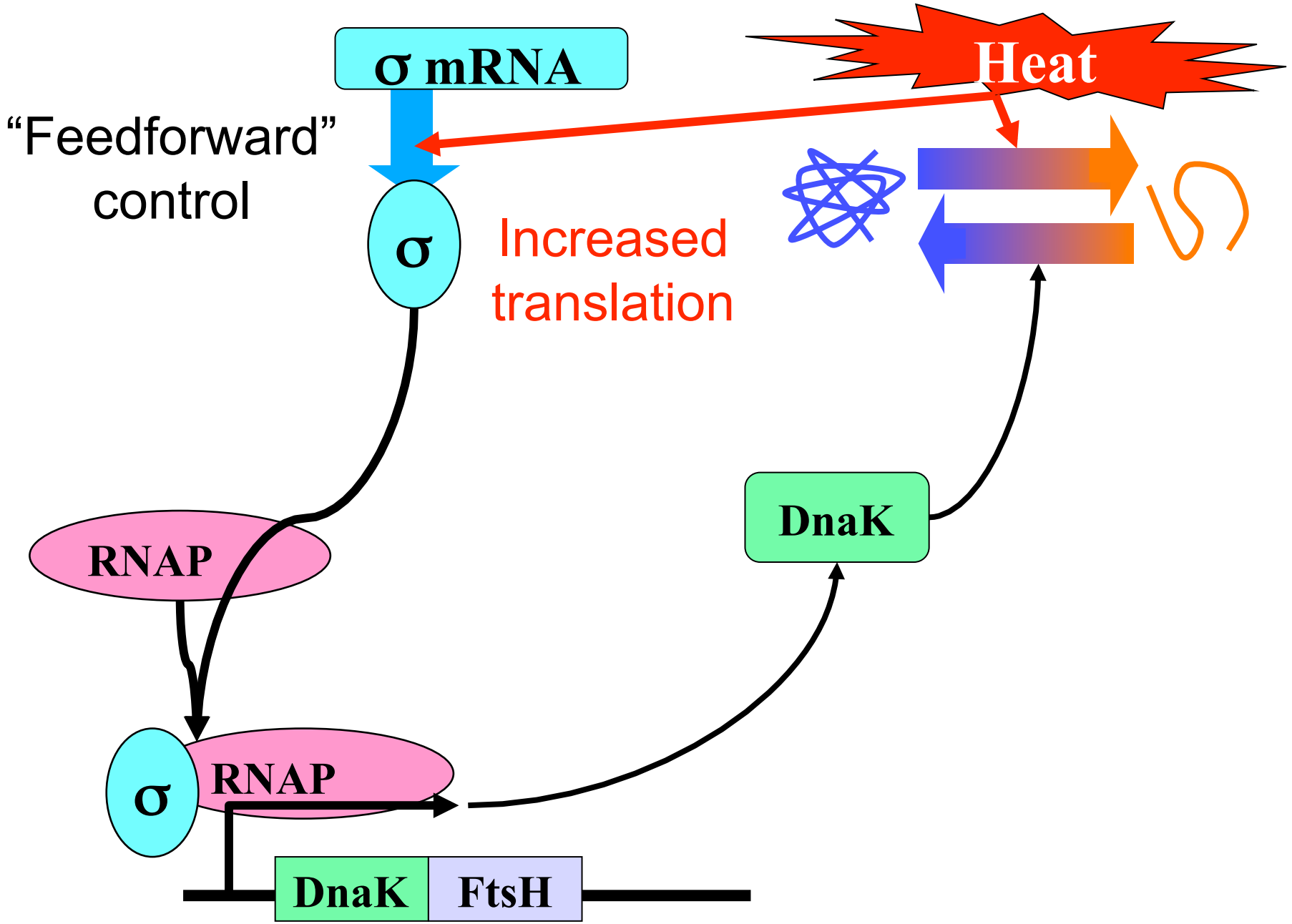

rpoH
σ mRNA
σ
Other operons
RNAP
σ
RNAP
DnaK
FtsH
Lon
Heat
DnaK
Lon

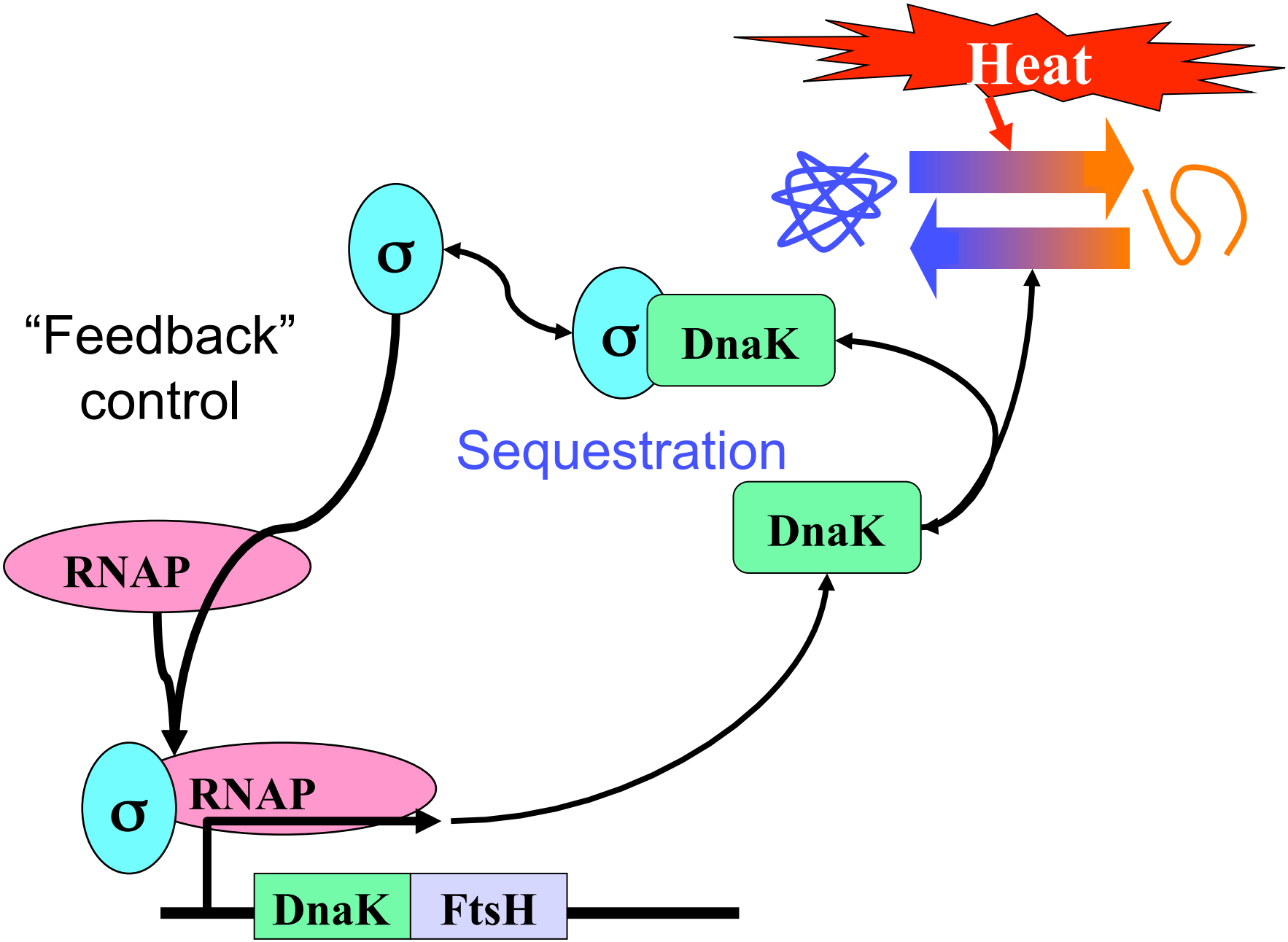

rpoH
σ mRNA
Heat
σ
Other
operons
σ
DnaK
σ
DnaK
DnaK
RNAP
ftsH
Lon
σ
RNAP
DnaK
FtsH
Lon


σ mRNA
Heat
"Feedforward" control
σ
Increased translation
RNAP
DnaK
σ
RNAP
DnaK
FtsH

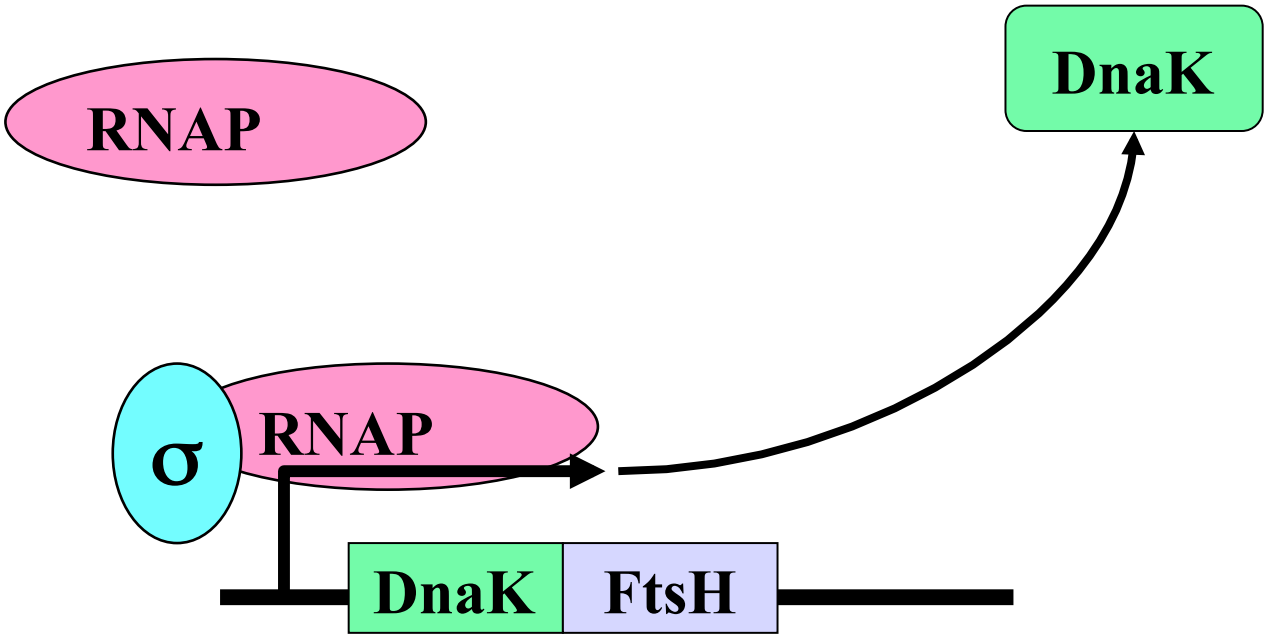


Translational Induction of heat shock transcription factor σ32 : evidence of a built-in thermosensor. *Morita et. al, Genes & Dev. 1999*

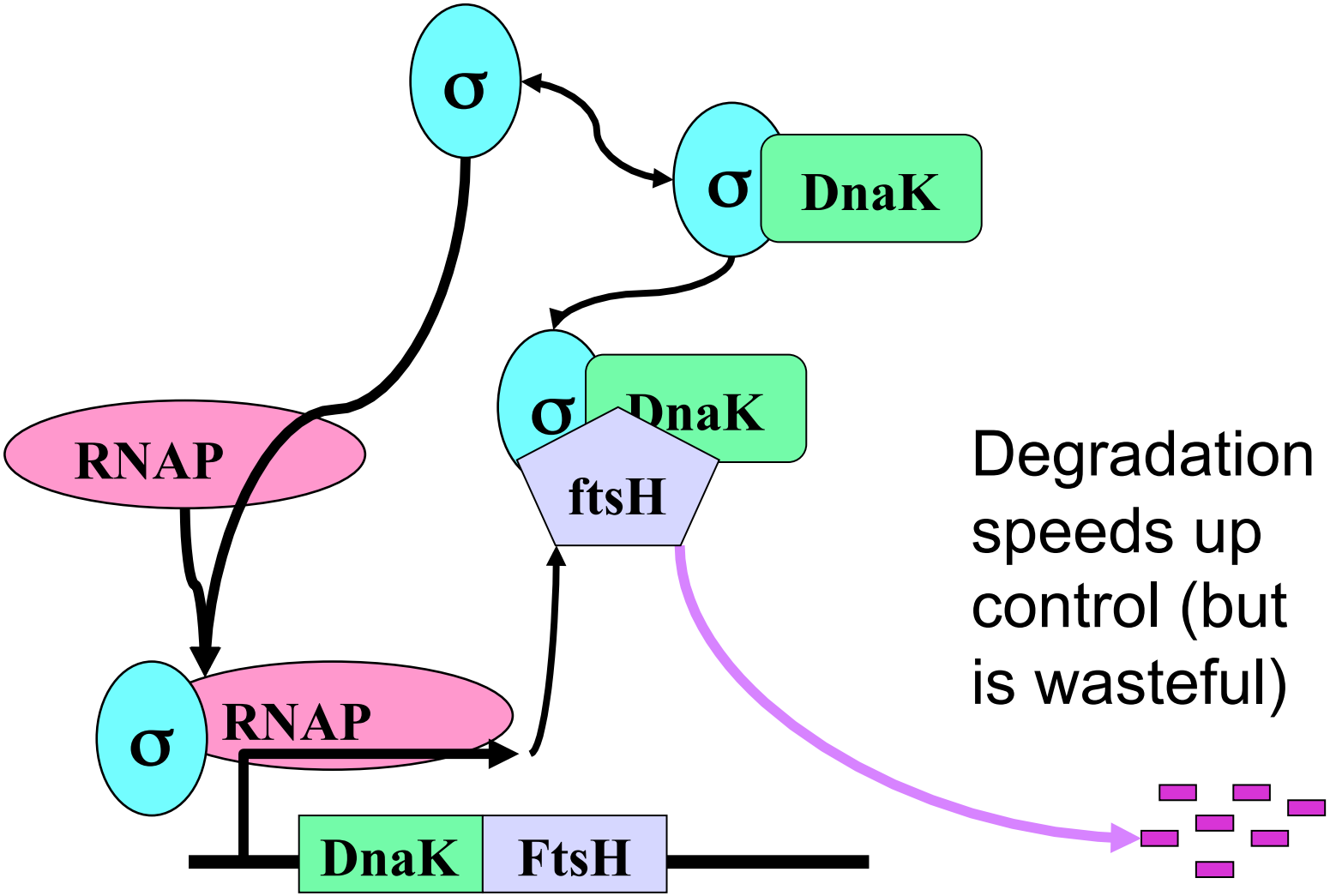

σ mRNA
Heat
"Feedforward" control
σ
Increased translation
RNAP
DnaK
σ
RNAP
DnaK
FtsH

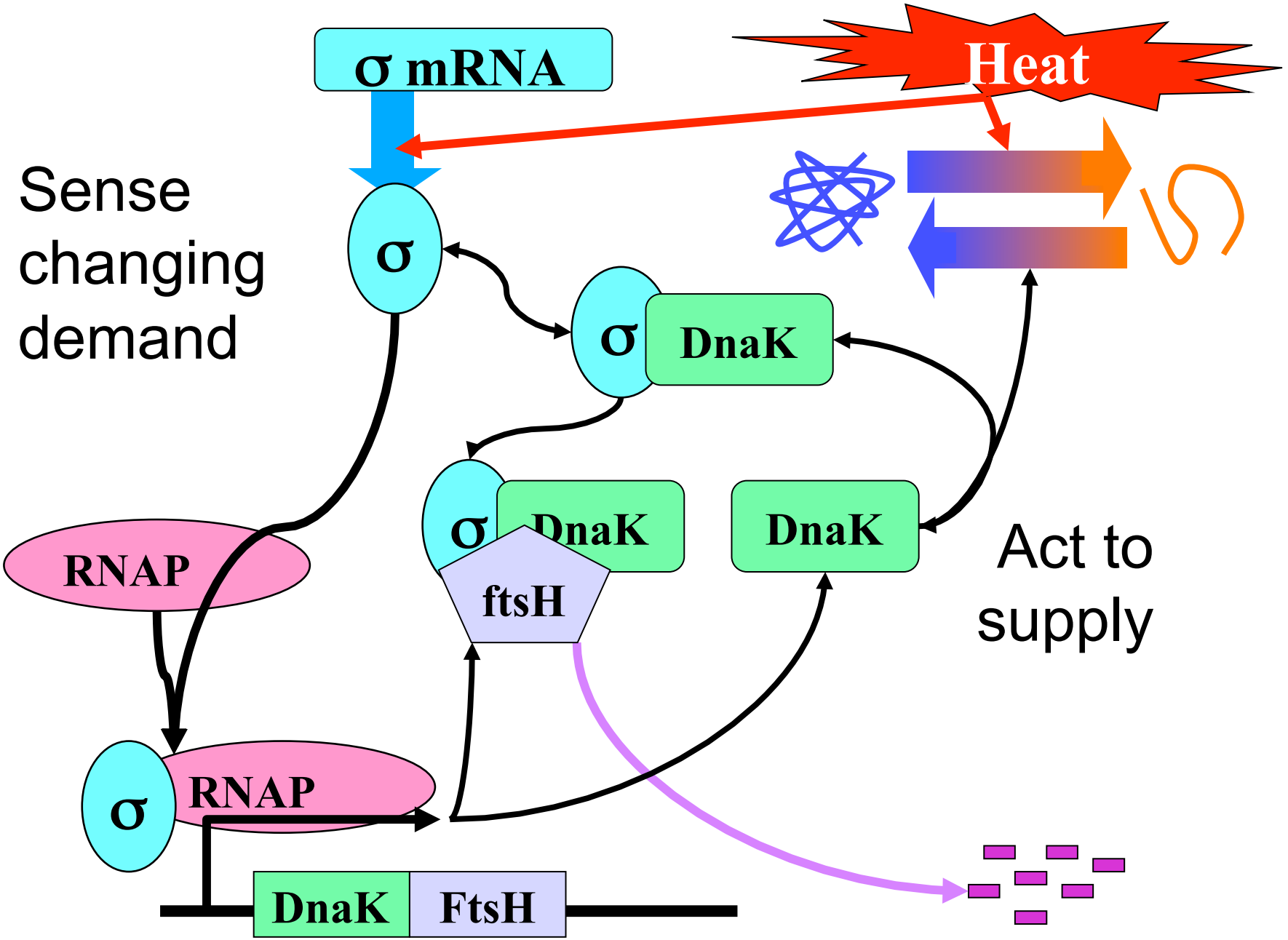

Heat
σ
σ
DnaK
"Feedback"
control
Sequestration
DnaK
RNAP
σ
RNAP
DnaK
FtsH

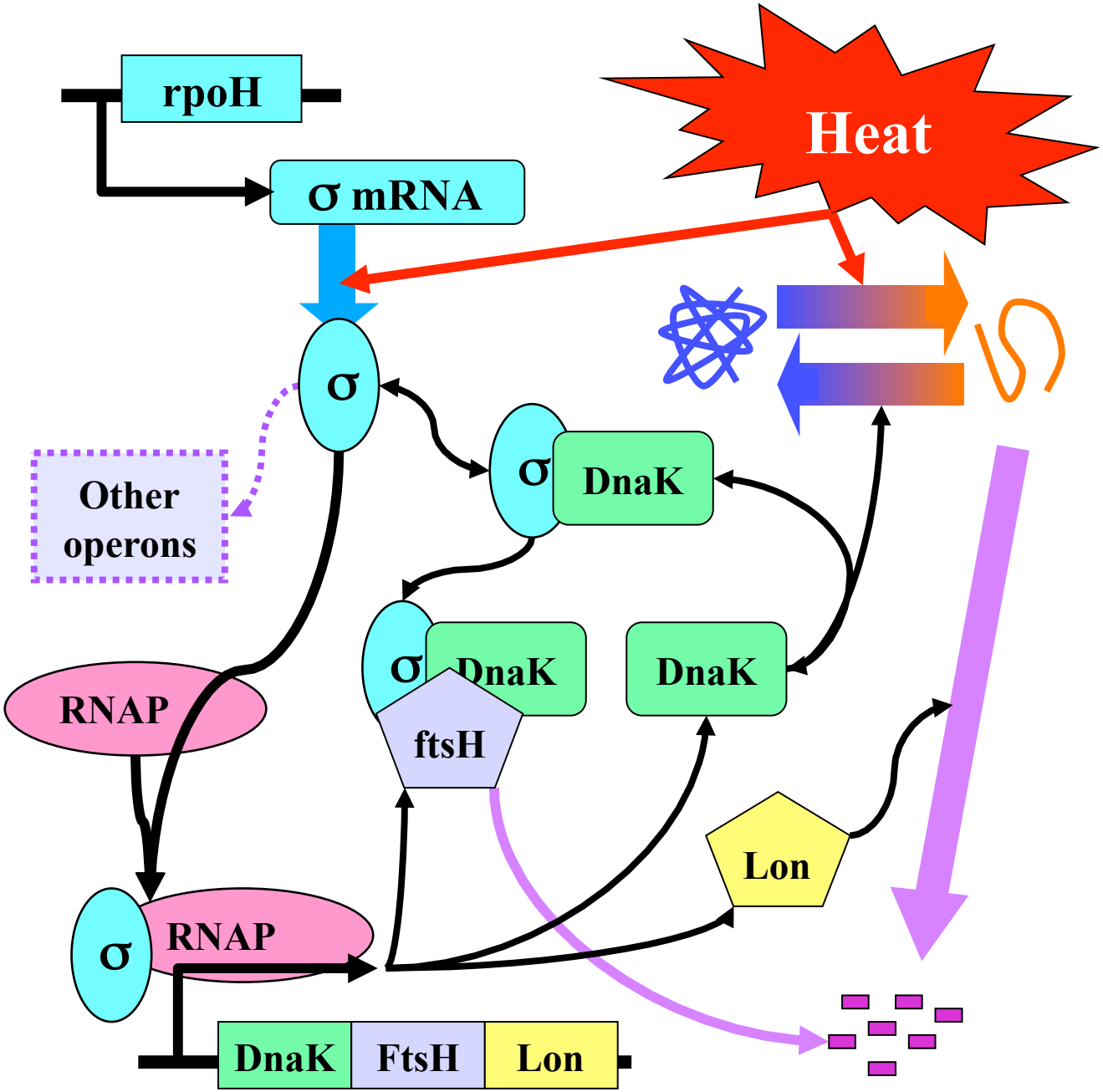
Note: you definitely need to spend an hour or so really reading about what is going on in the promoter and operon and what RNApolymerase does. There is tons online and every text on molecular biology of bacteria has this.



Speed efficiency tradeoff
(see *PNAS* paper)

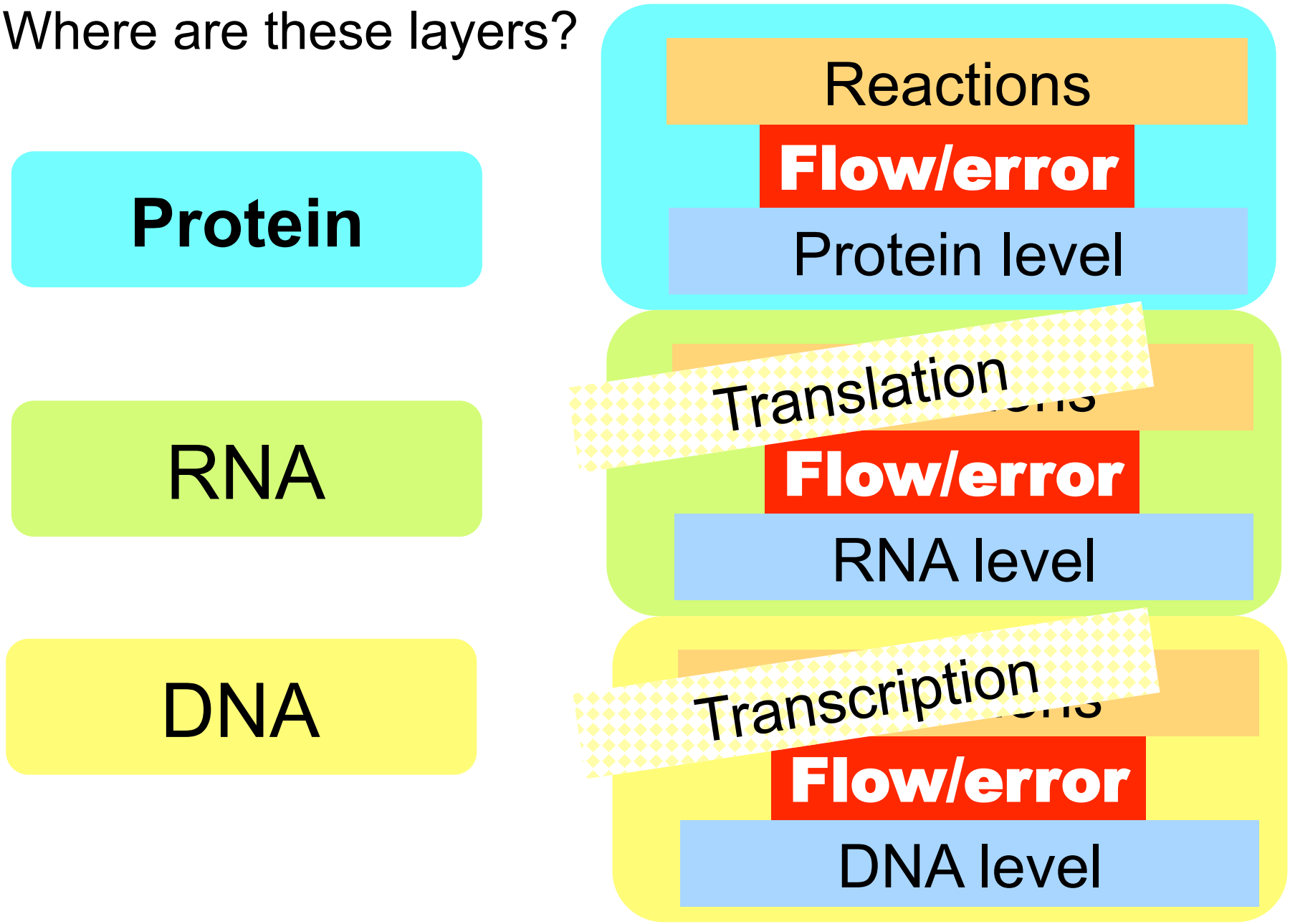


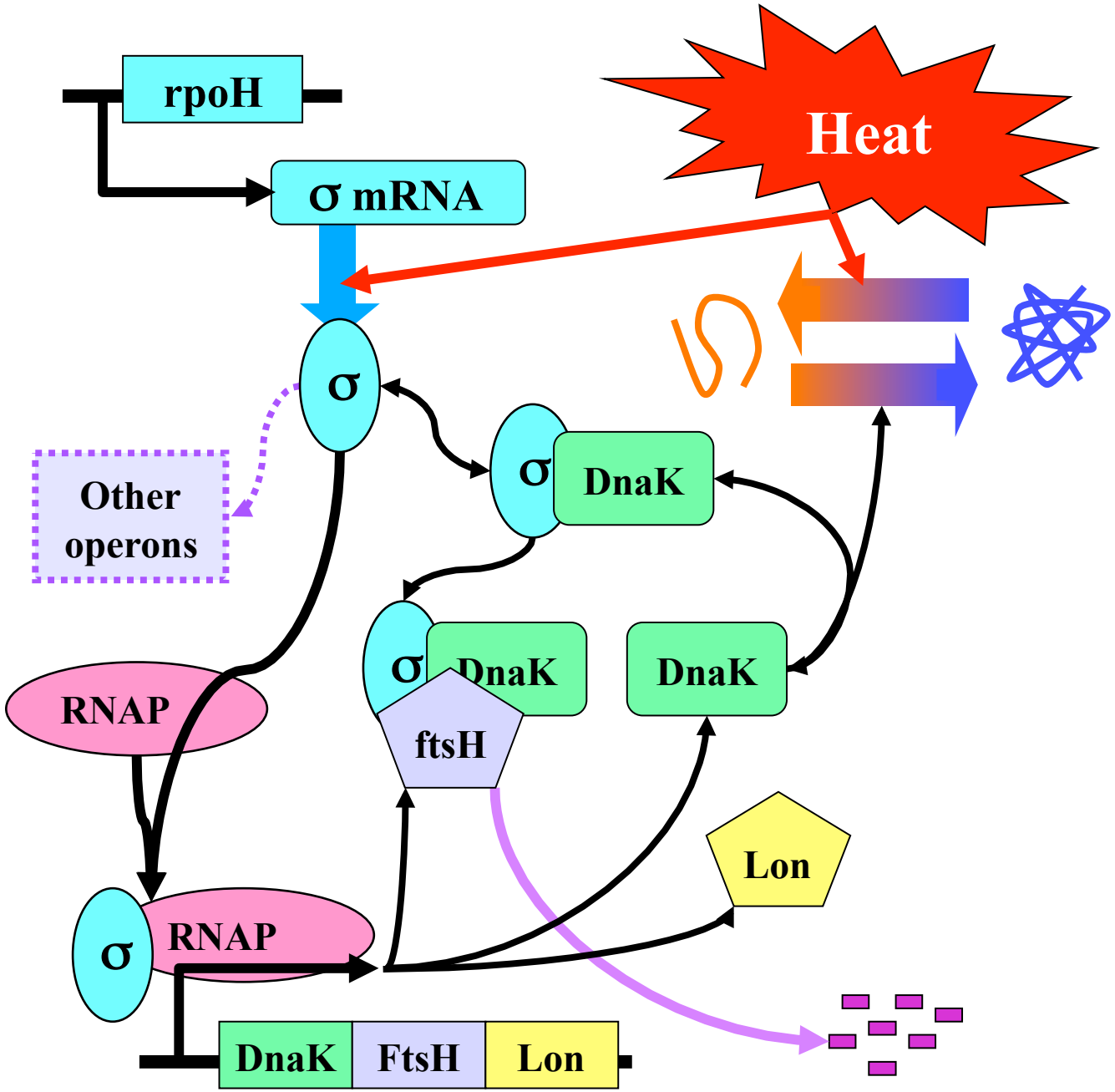

σ mRNA
Heat
Sense changing demand
σ
σ
DnaK
σ
DnaK
DnaK
RNAP
ftsH
Act to supply
σ
RNAP
DnaK
FtsH

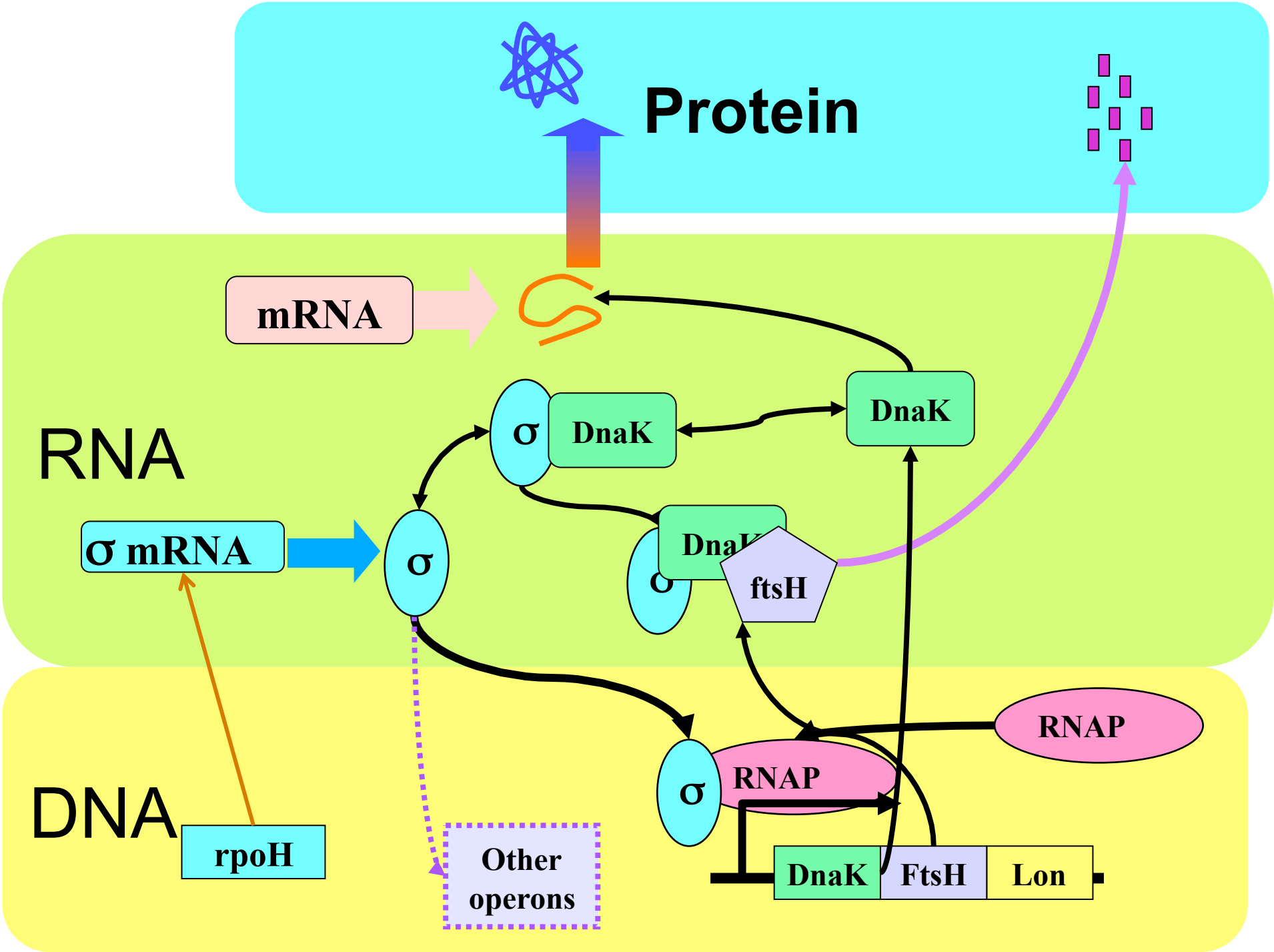


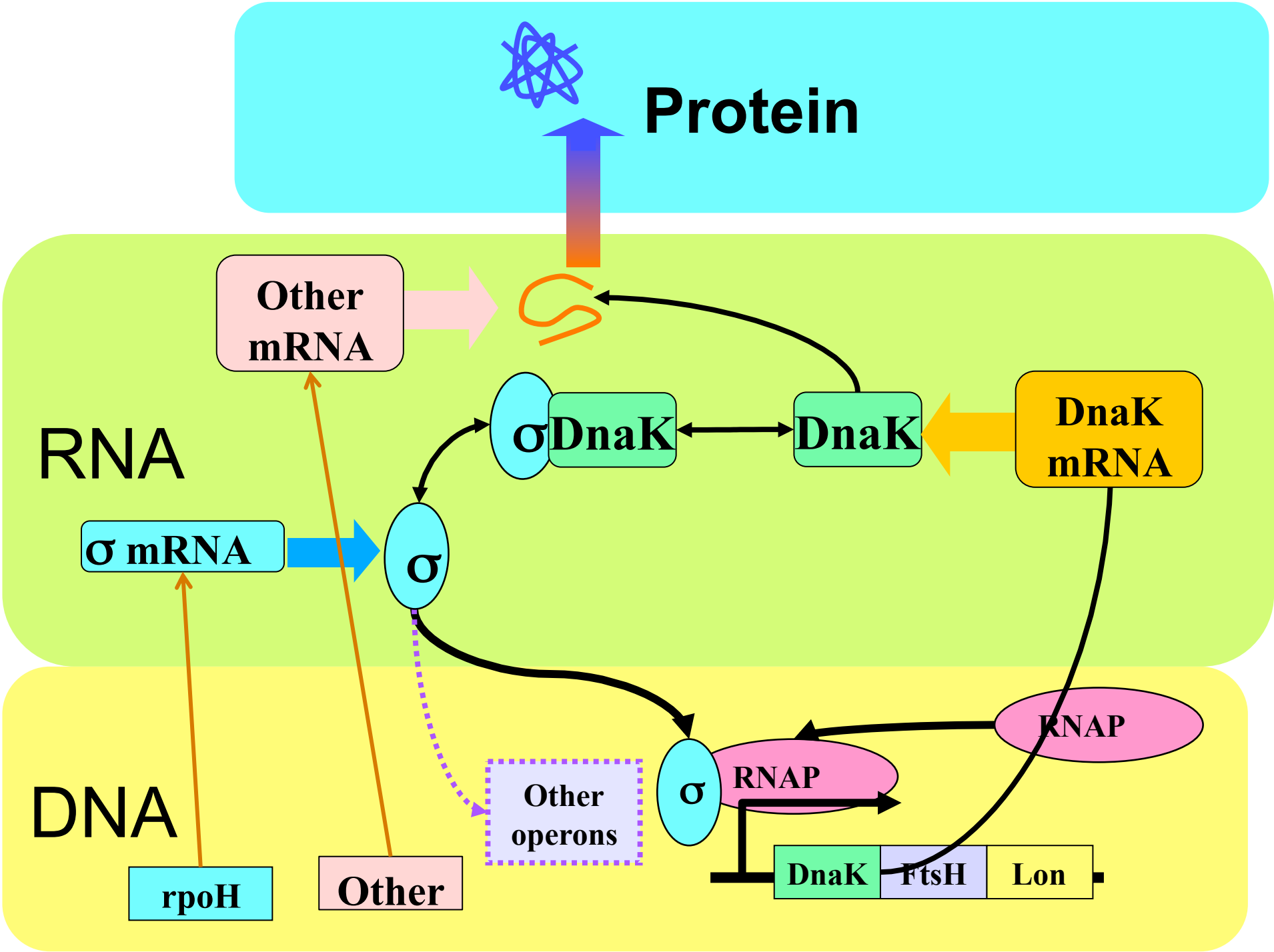
Recycle proteins that can't refold

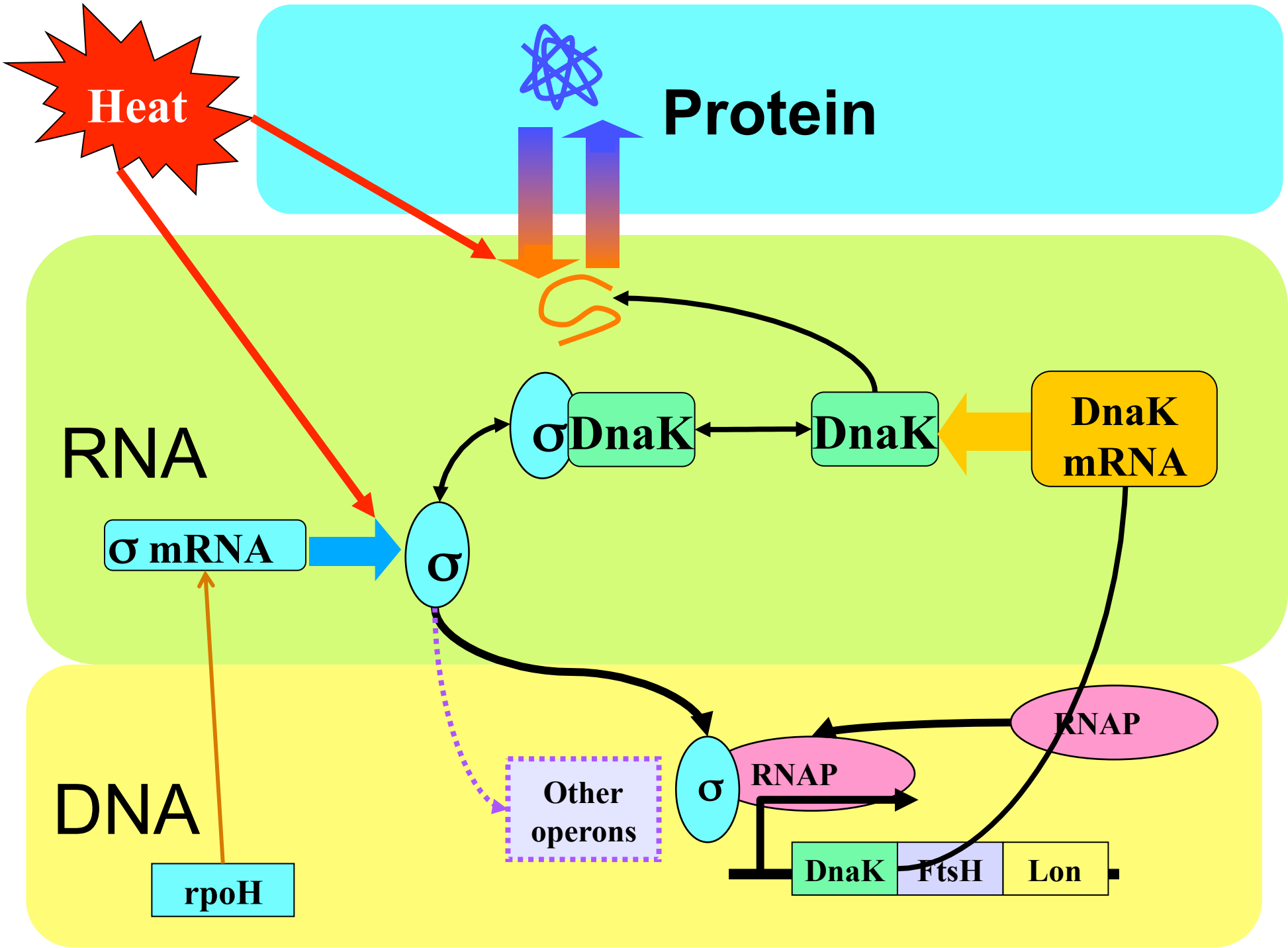
Where are these layers?

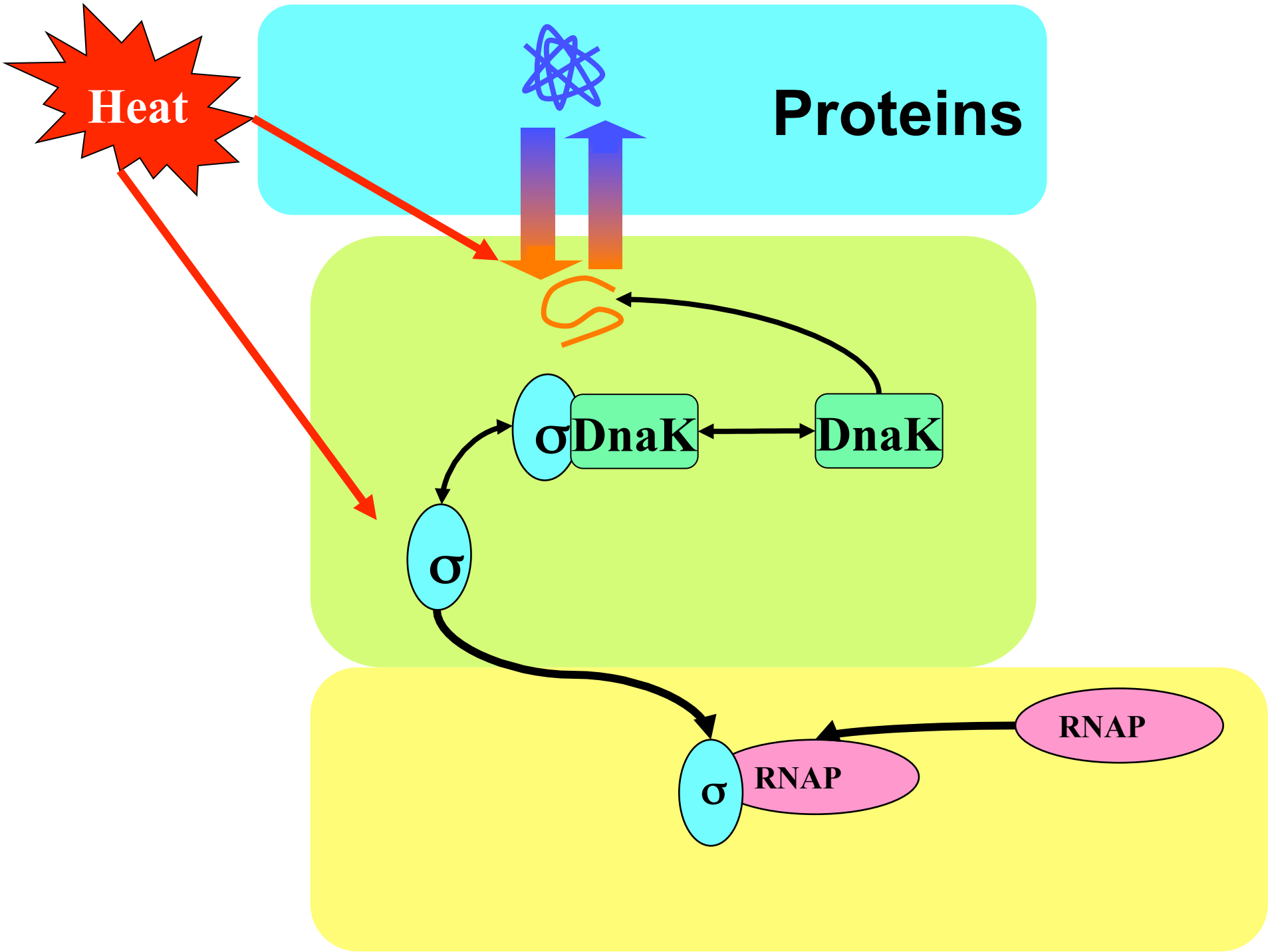


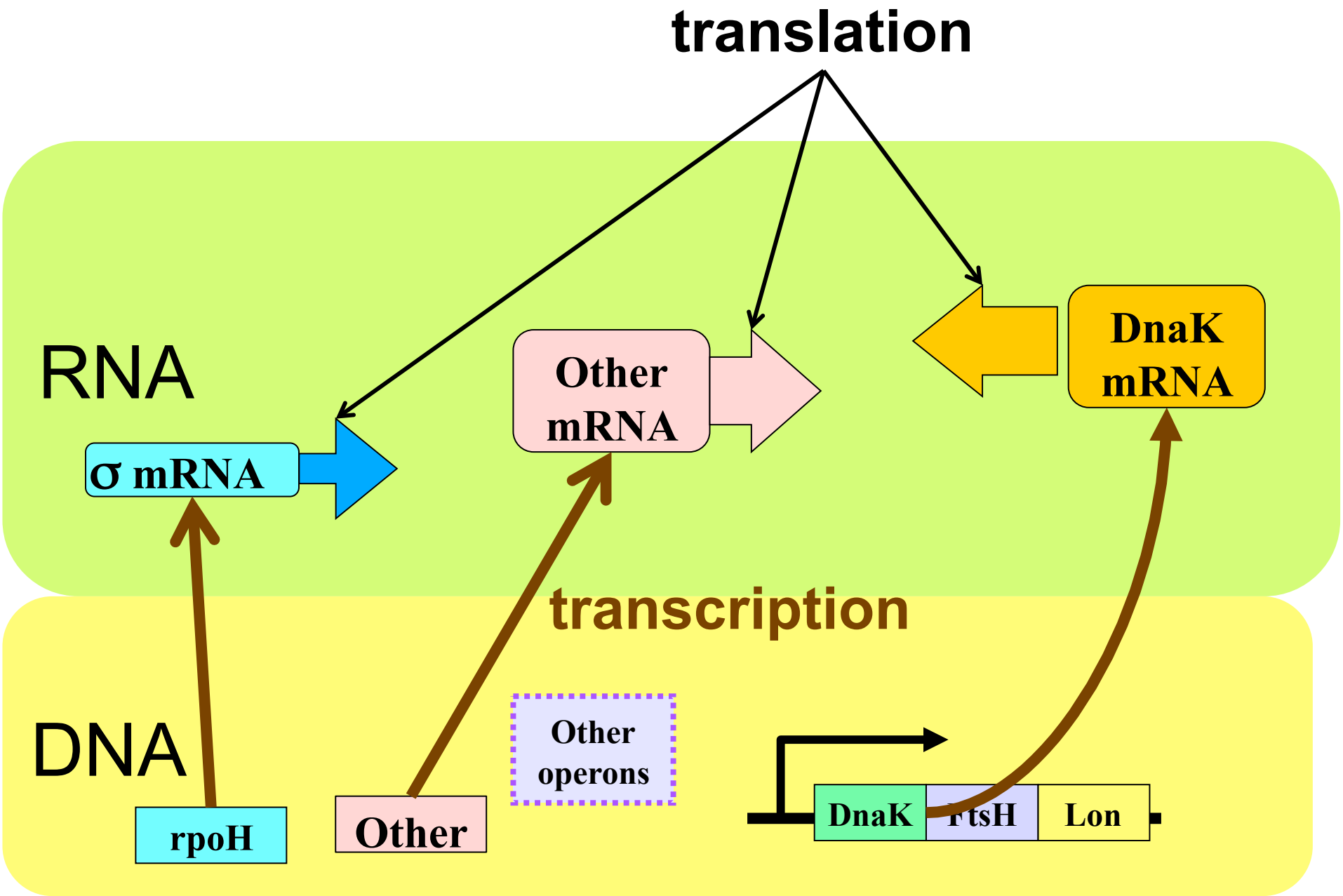

rpoH
σ mRNA
Heat
σ
Other
operons
σ
DnaK
σ
DnaK
DnaK
RNAP
ftsH
Lon
σ
RNAP
DnaK
FtsH
Lon

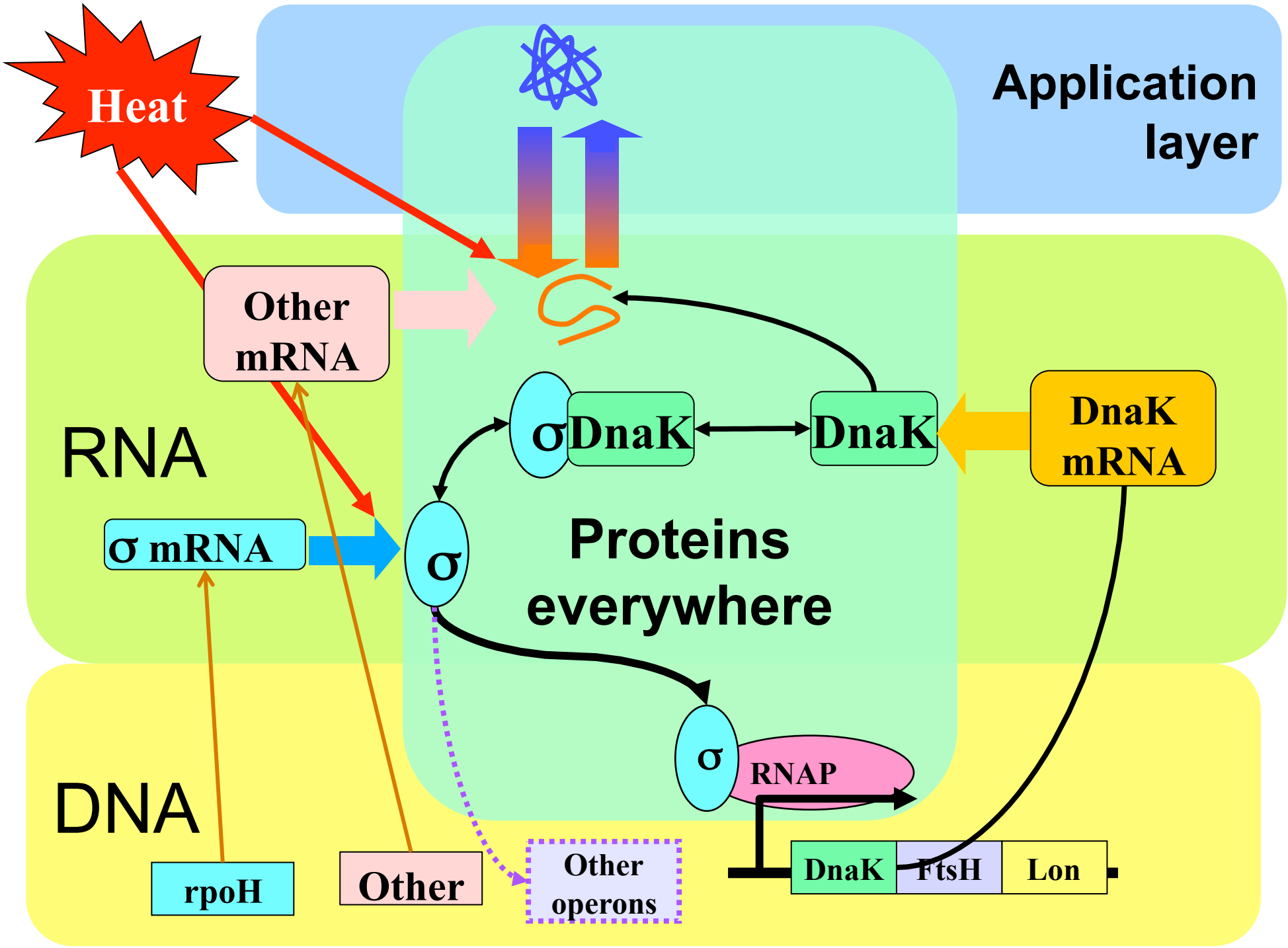

Protein
mRNA
RNA
σ
DnaK
DnaK
σ mRNA
σ
σ
ftsH
RNAP
σ
RNAP
DNA
rpoH
Other
operons
DnaK
FtsH
Lon

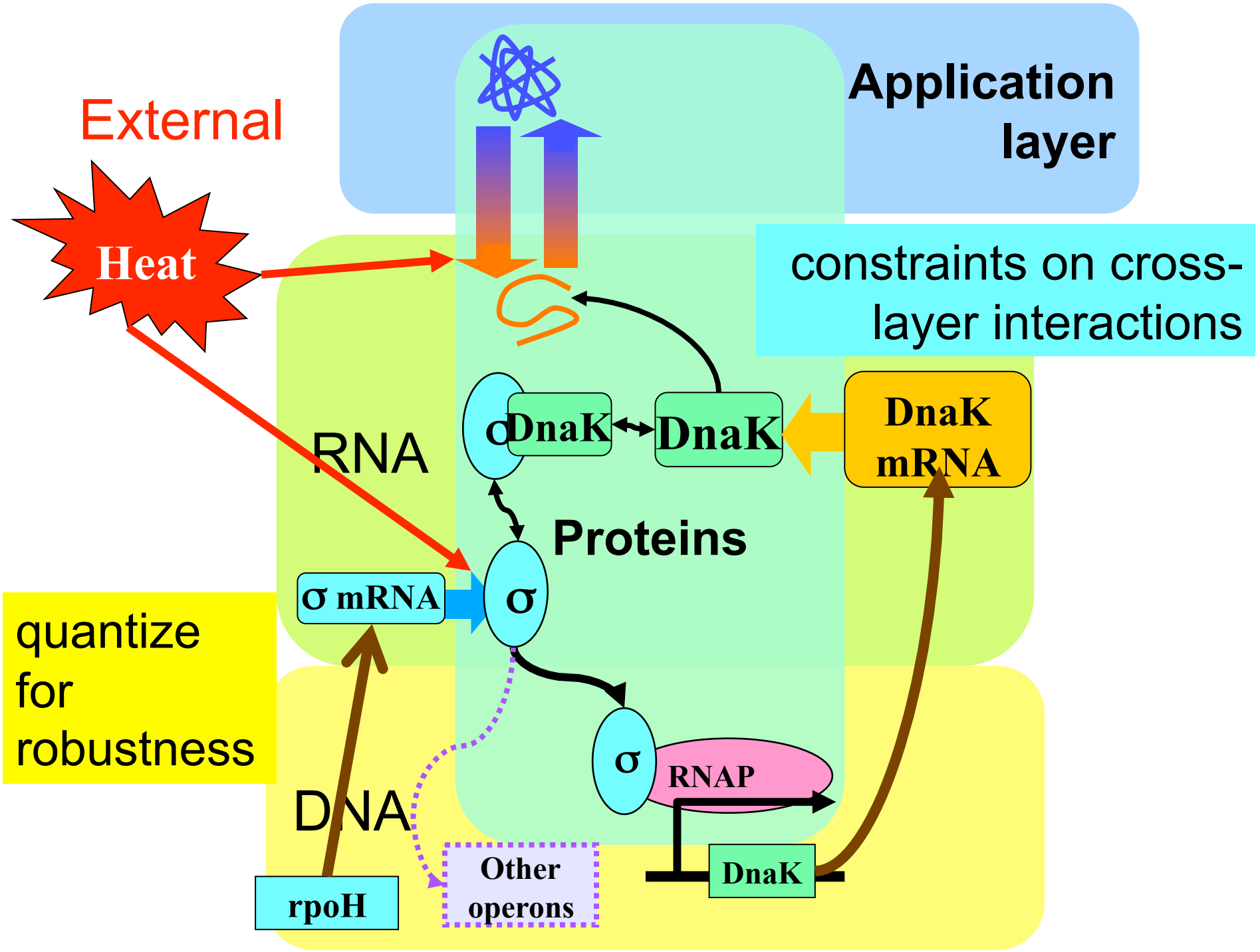

Protein
RNA
Other mRNA
σ DnaK
DnaK
DnaK mRNA
σ mRNA
σ
DNA
rpoH
Other
Other operons
σ
RNAP
RNAP
DnaK
FtsH
Lon

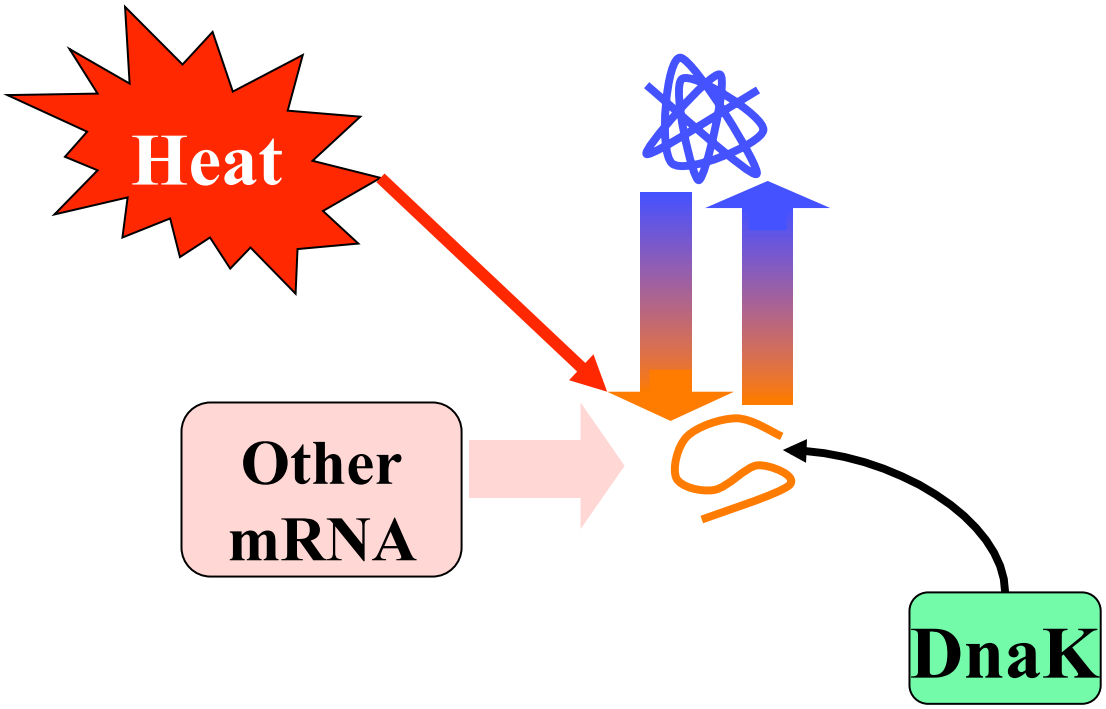

Heat
Protein
RNA
σDnaK
DnaK
DnaK mRNA
σ mRNA
σ
DNA
Other operons
σ
RNAP
RNAP
DnaK
FtsH
Lon
rpoH


Heat
Proteins
σ
σDnaK
DnaK
σ
RNAP
σ
RNAP


translation
RNA
σ mRNA
Other mRNA
DnaK mRNA
transcription
DNA
rpoH
Other
Other operons
DnaK
FtsH
Lon

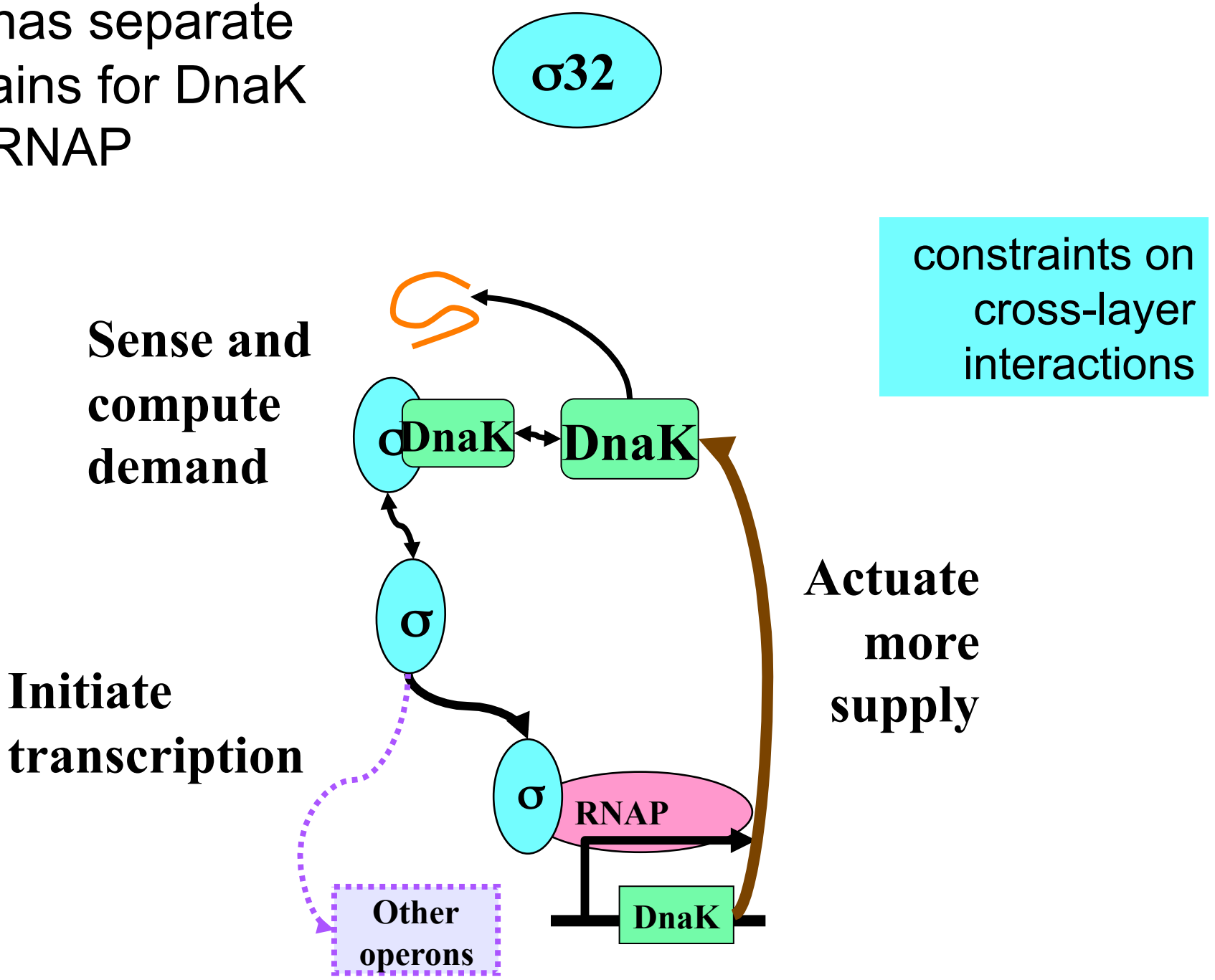

Heat
Application layer
Other mRNA
RNA
σ DnaK
DnaK
DnaK mRNA
σ mRNA
σ
Proteins everywhere
σ
RNAP
DNA
rpoH
Other
Other operons
DnaK
FtsH
Lon

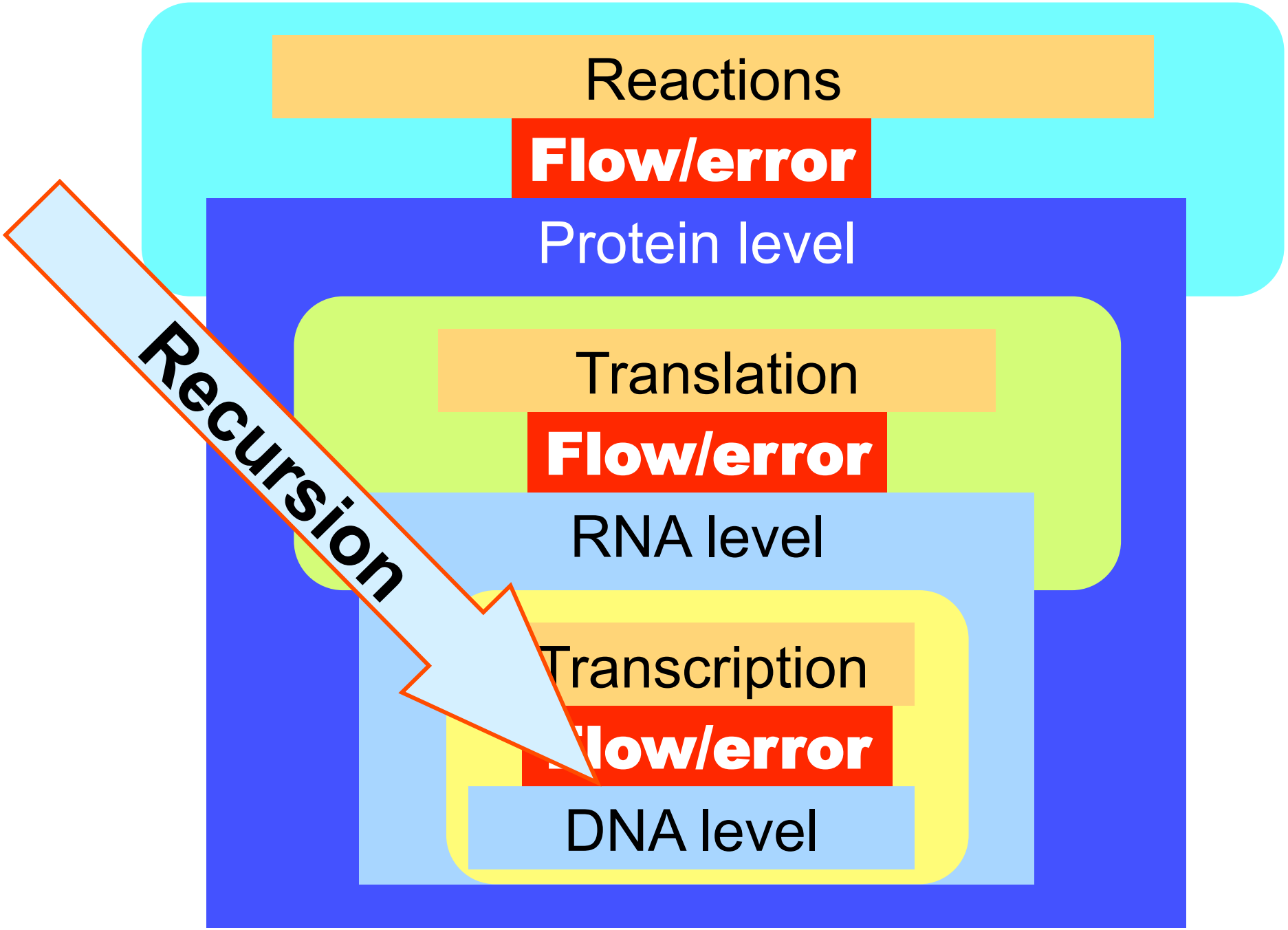

Application layer
External
Heat
constraints on cross-layer interactions
RNA
σ
DnaK
DnaK
DnaK mRNA
Proteins
σ mRNA
σ
quantize for robustness
σ
RNAP
DNA
Other operons
DnaK
rpoH

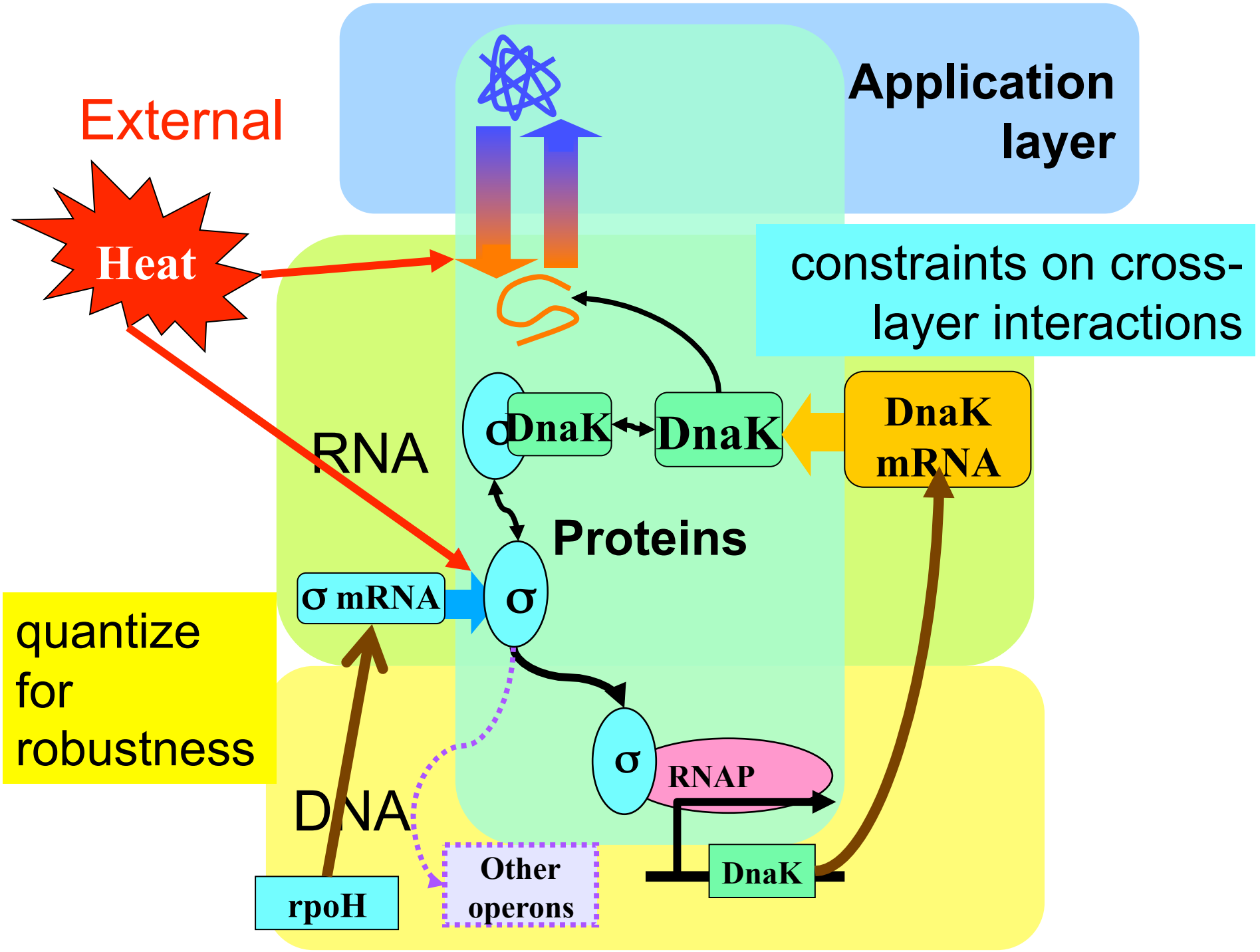


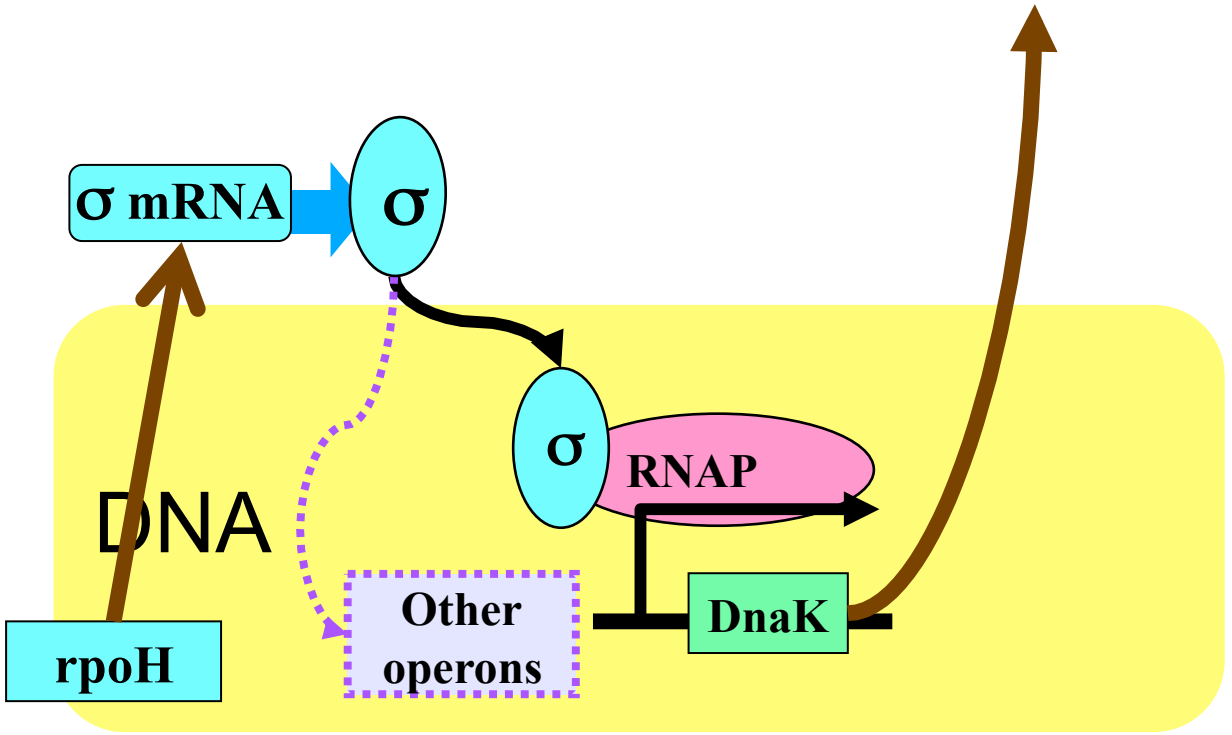
General purpose chaperones for folding proteins

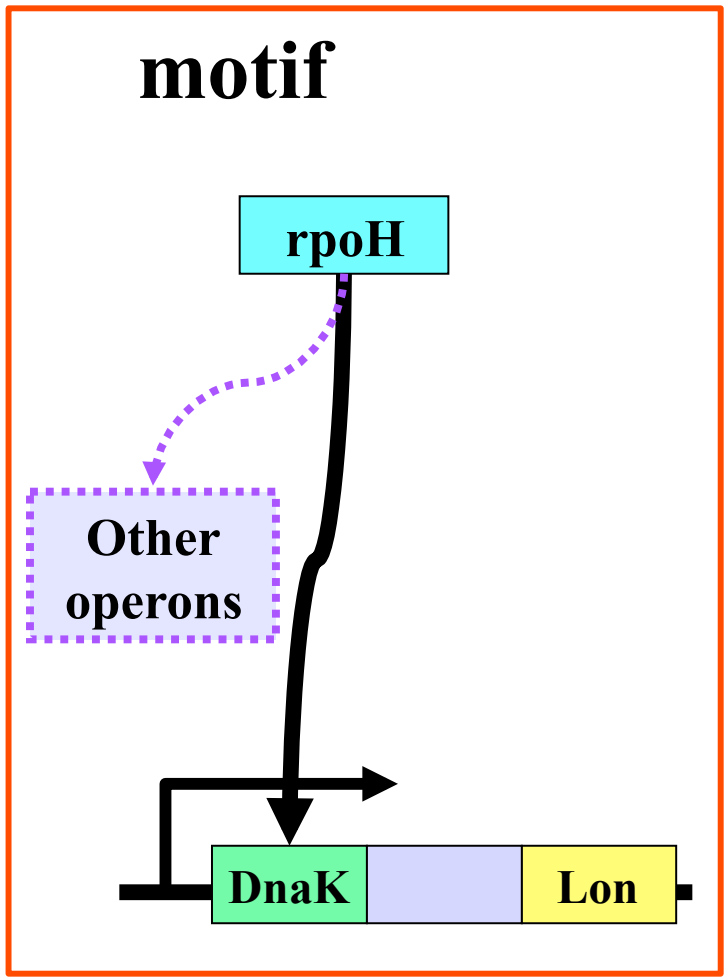
σ32 has separate domains for DnaK and RNAP



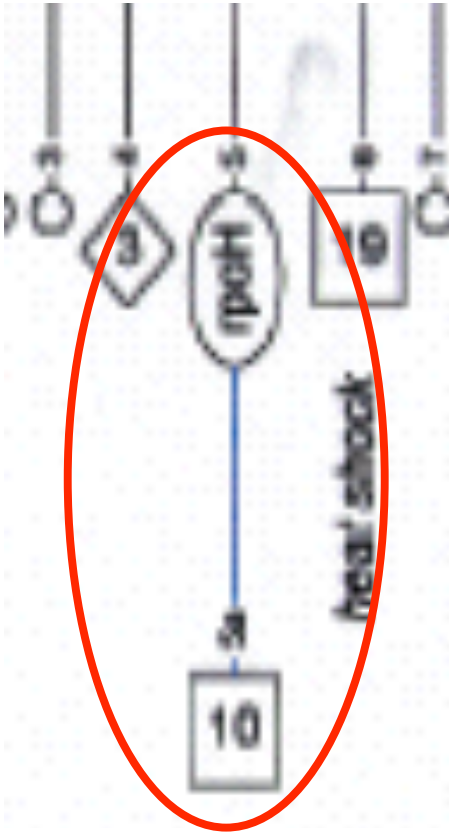

Reactions
Flow/error
Protein level
Translation
Flow/error
RNA level
Transcription
Flow/error
DNA level
Recursion

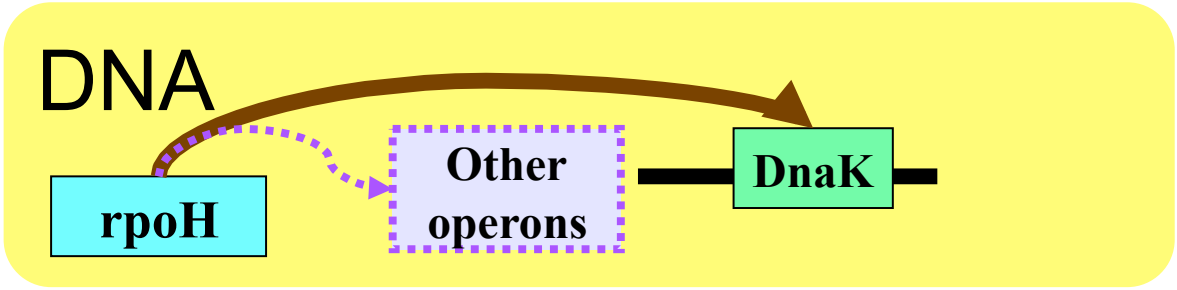

Application layer
External
Heat
constraints on cross-layer interactions
RNA
σ
DnaK
DnaK
DnaK mRNA
Proteins
σ mRNA
σ
quantize for robustness
σ
RNAP
DNA
Other operons
DnaK
rpoH


σ mRNA
σ
σ
RNAP
DNA
Other
operons
DnaK
rpoH



Different cartoons
of the same circuit





All at the DNA layer

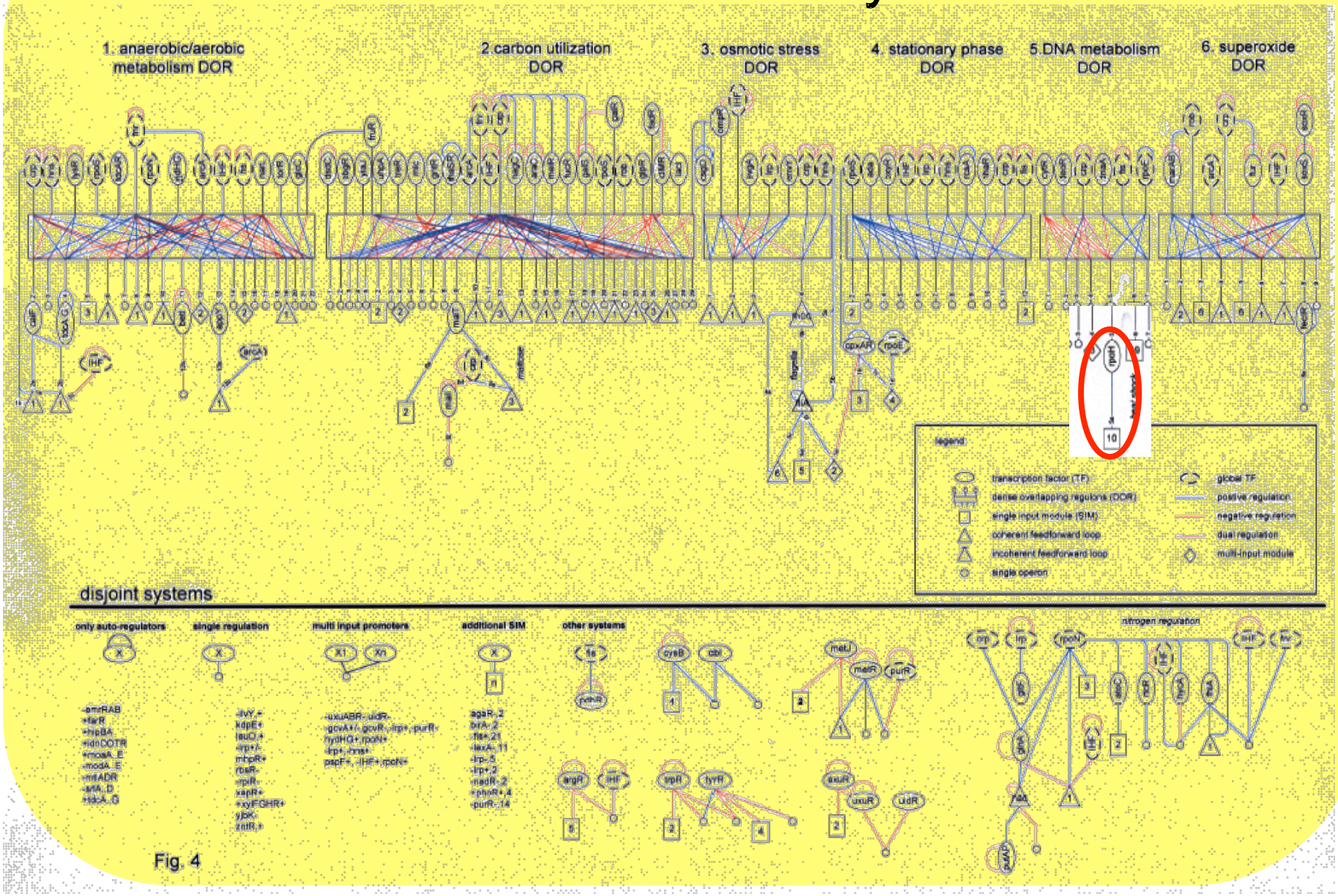


Fig. 4

Hierarchical structure of *E. coli* transc. regulatory network.

All transcriptional regulatory links are downward. Nodes are operons. Global regulators are red. Yellow marked nodes are operons in the longest regulatory pathway related with flagella motility.

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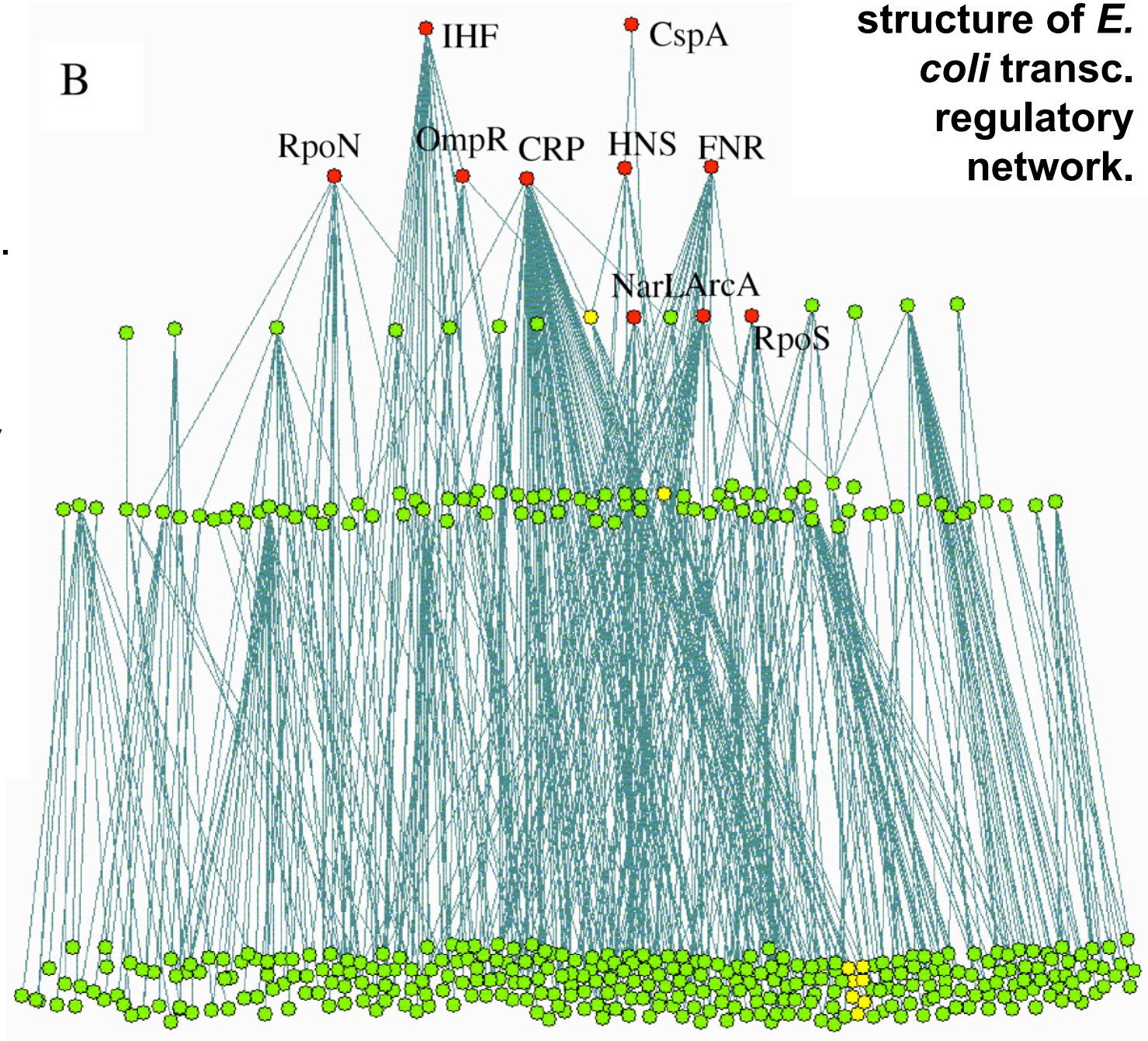
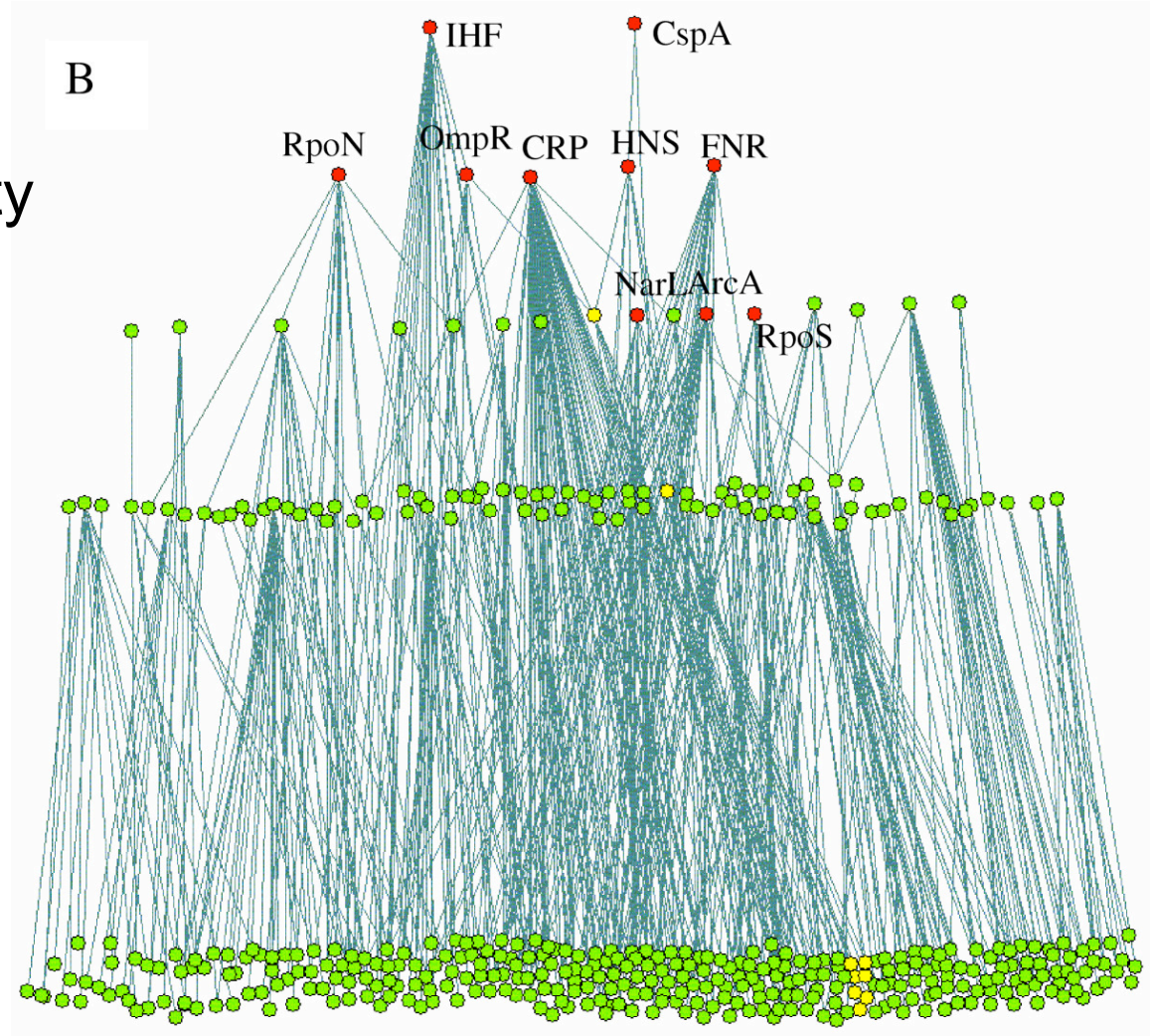


Note: all feedback in this picture has been removed in two ways:

1) There are self-loops where an operon is controlled by one it's own genes
2) All the real complexity is in the protein interactions not shown (see the HS details above)

These are not ***control*** systems, they just initiate manufacturing



Functional modules in the transcriptional regulatory network of *E. coli*. Operons in different modules are shown in different colors. The ten global regulators form the core part of the network. The periphery modules are connected mainly through the global regulators. Depending on the connectivity between the modules and their connectivity to the global regulators, these modules can be further grouped to larger modules at a higher level.

Ma *et al. BMC Bioinformatics* 2004 **5**:199 doi: 10.1186/1471-2105-5-199