

SBM 0/Monday Sep 29: Introduction

SBM -1/Thursday Oct 2: iGEM (Chris French)

SBM -2/Monday Oct 6: random graphs 1

SBM -3/Thursday Oct 9: random graphs 2

SBM -4/Monday Oct 13: network rewiring (after Wendell Lim)

Thursday Oct 16: no course!

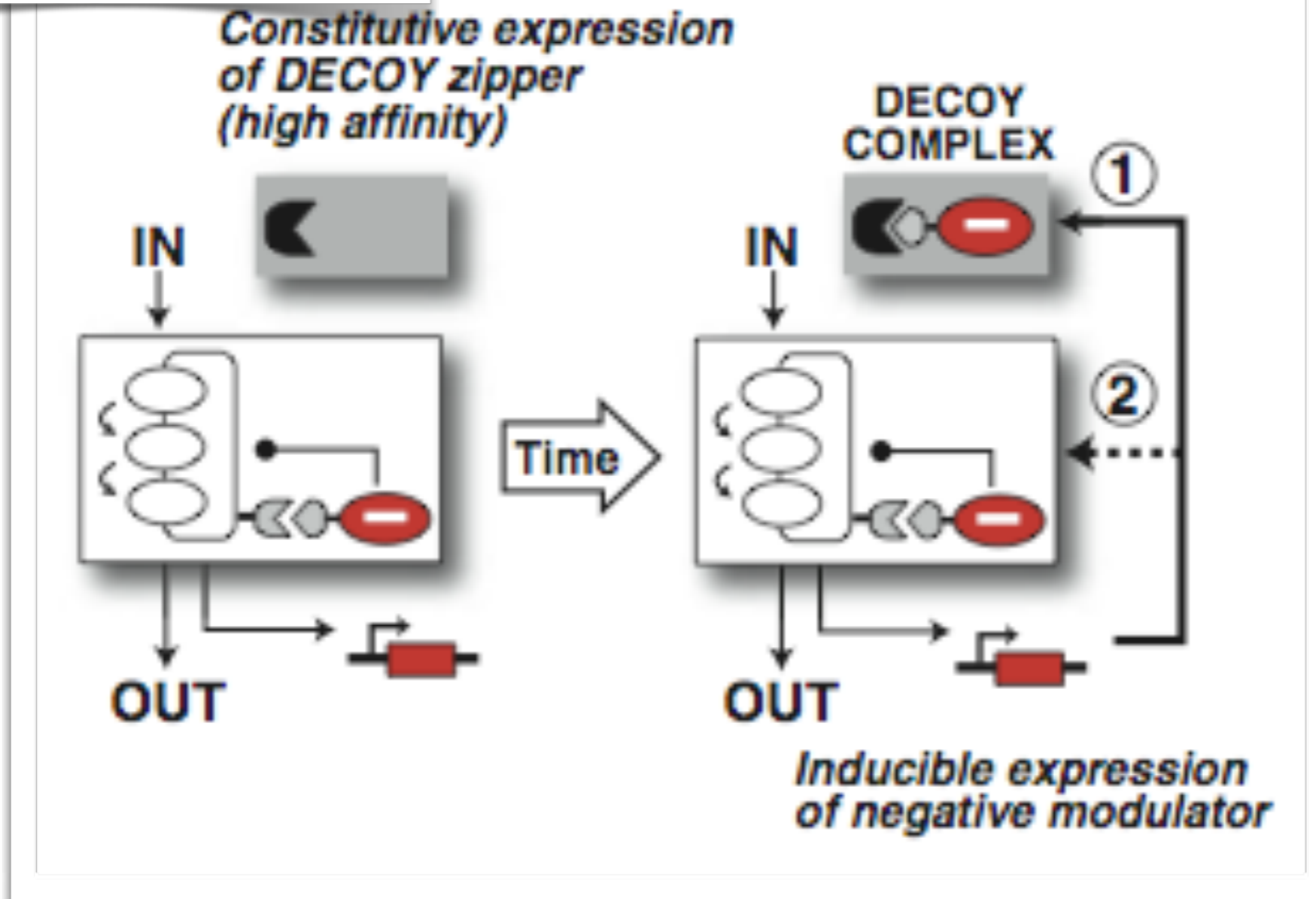
SBM -5/Monday Oct 20: pseudo prey-predator ecosystem

SBM -6/Thursday Oct 23: compilation to Bio-Bricks (Michael Pedersen)

SBM -7/Monday Oct 27: modular proteins (Maria-L. Guerriero)

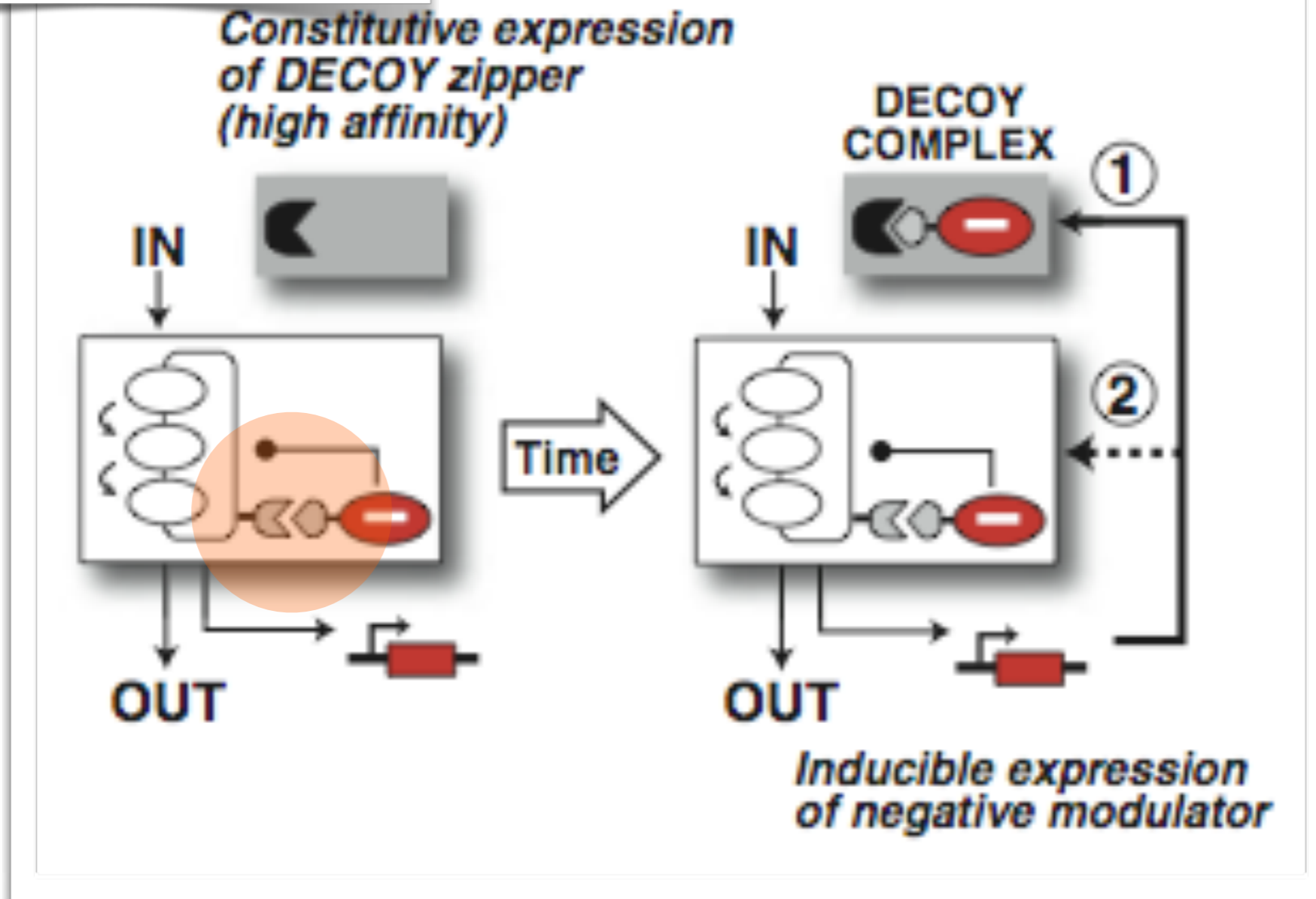
ppi - trailer

a buffered negative FB



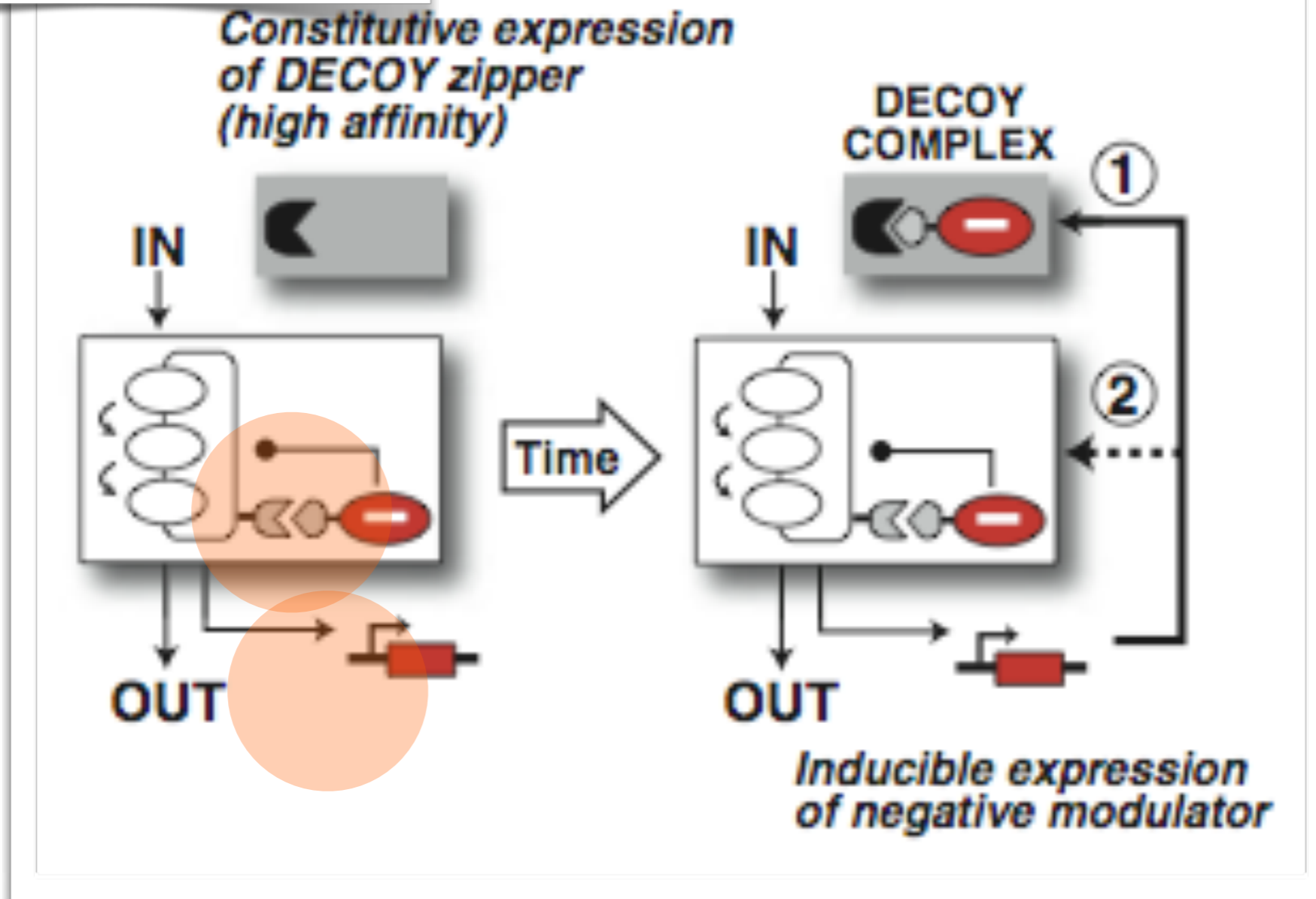
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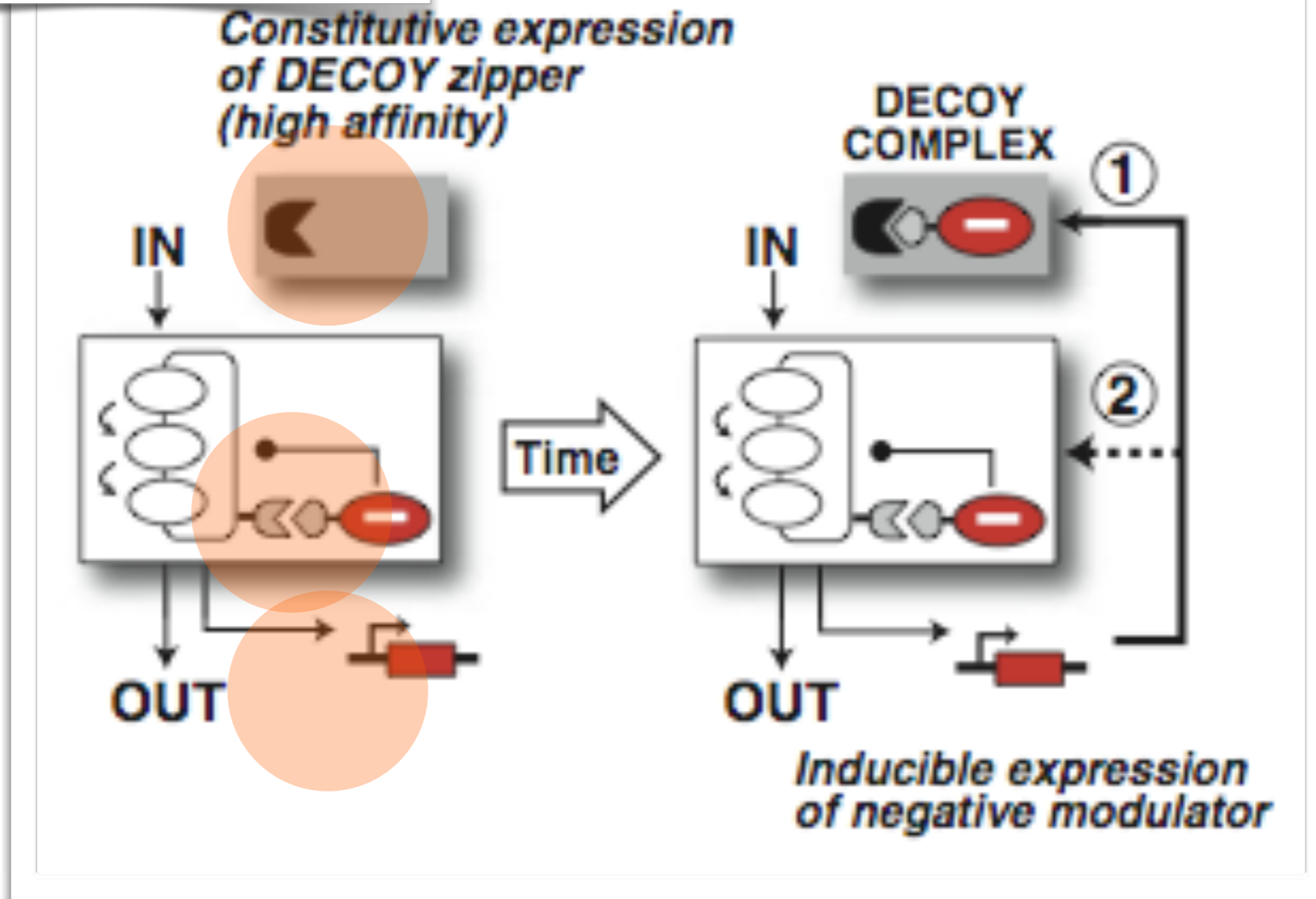
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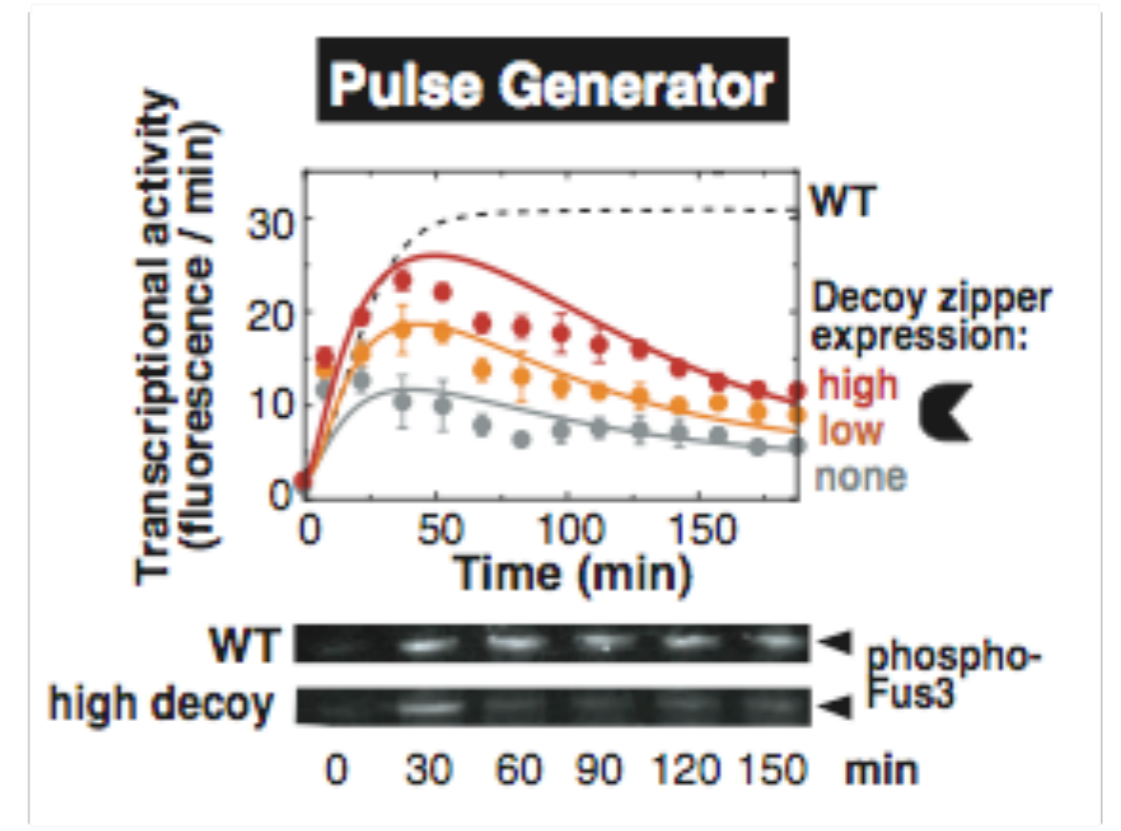
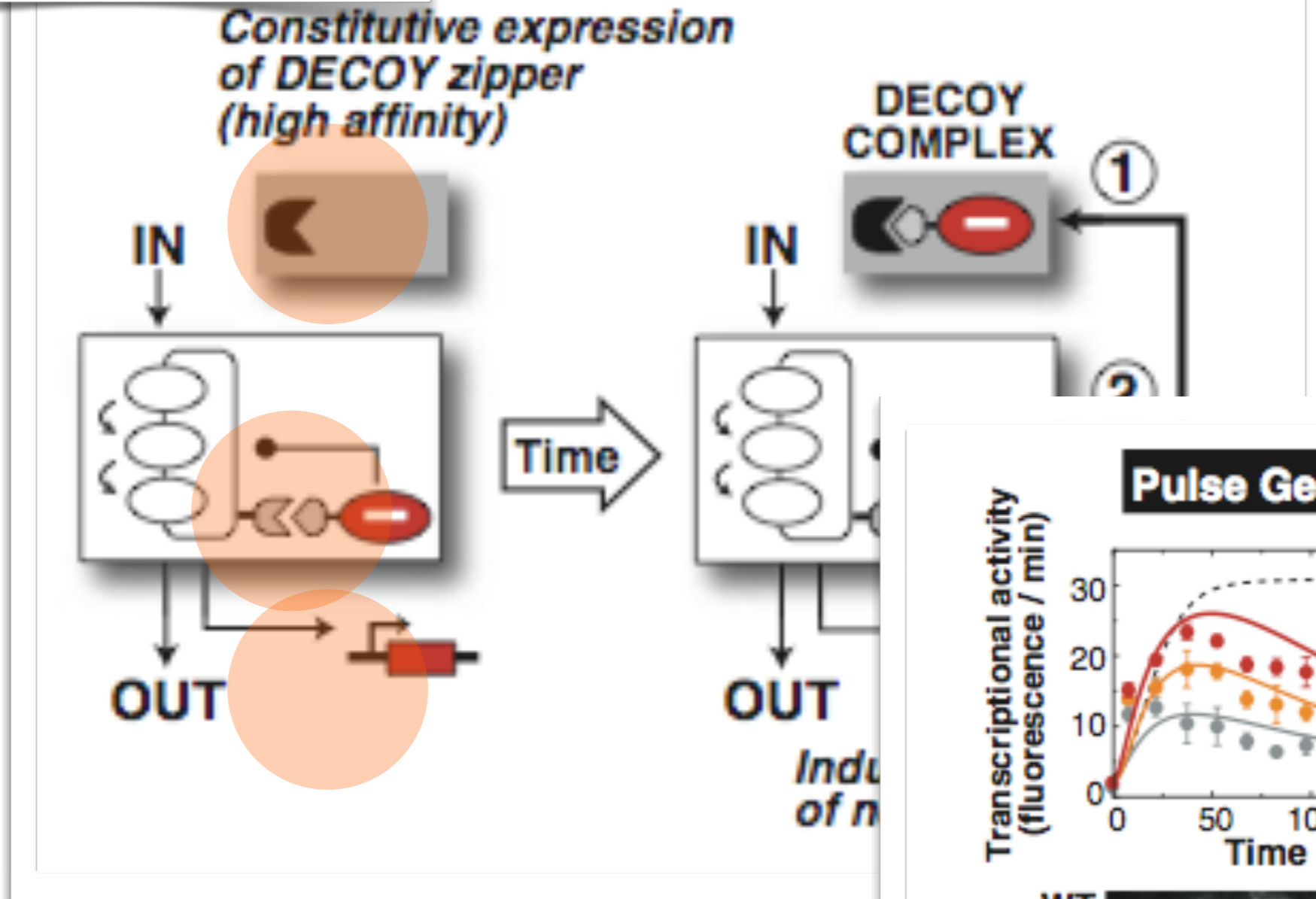
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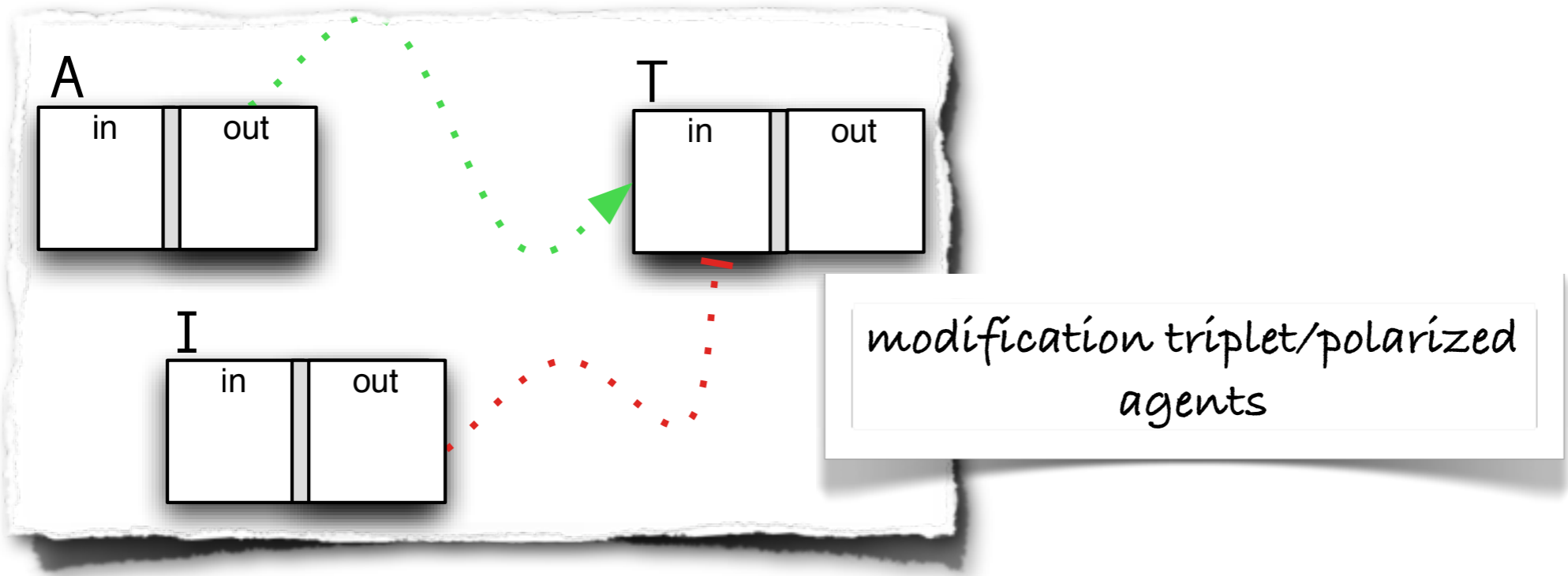
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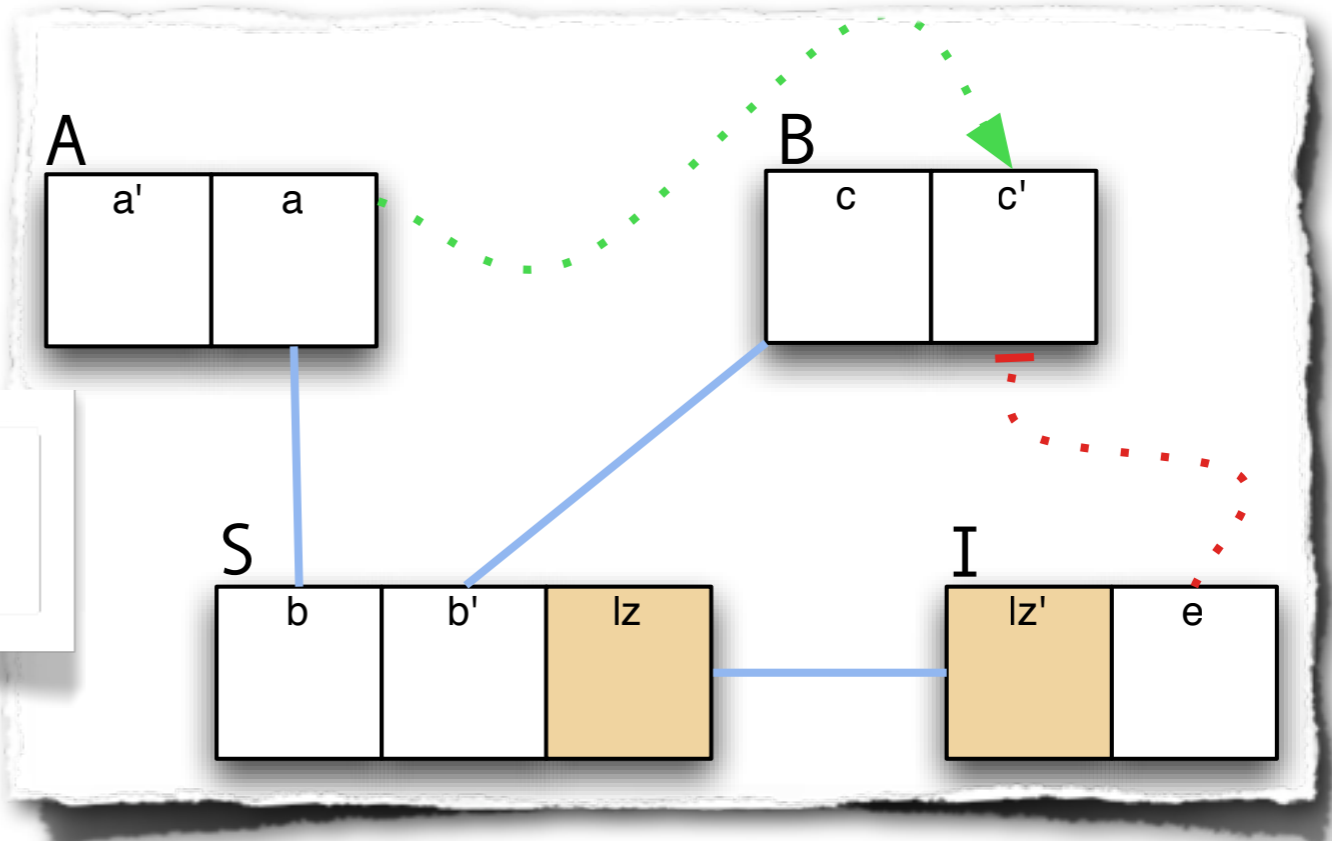


SBM -5

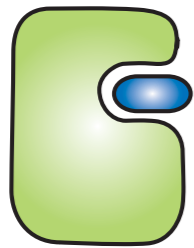
using engineered scaffold interactions to reshape MAP kinase pathway
signaling dynamics
(2008) Lim et al Science 319



scaffolds/beyond the "simple" modification triplet



Input:
upstream
regulators



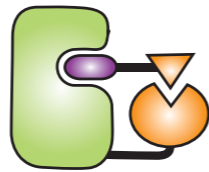
Output:
substrate/
effector



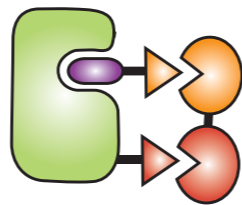
Active site



Docking interactions



Modular domains



Scaffolds/adapters

*modular proteins
(María-Luís)*

us: abstract jargon of **solid/liquid DD/PP/PD wires**
to discuss at the end

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jargon

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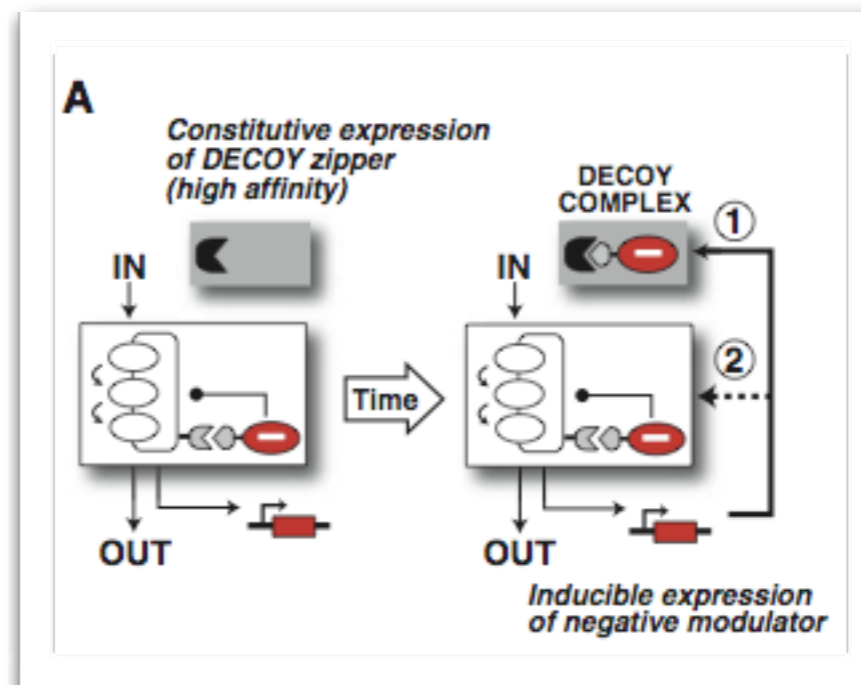
jargon

caveat

GOAL: hook +ve and -ve feedback (FB) on a scaffold =: **solid PP-wire**

NB: the FB is via transcription -
ie a **solid DD-wire**
unlike say ERK's on SOS

Example: a buffered negative FB

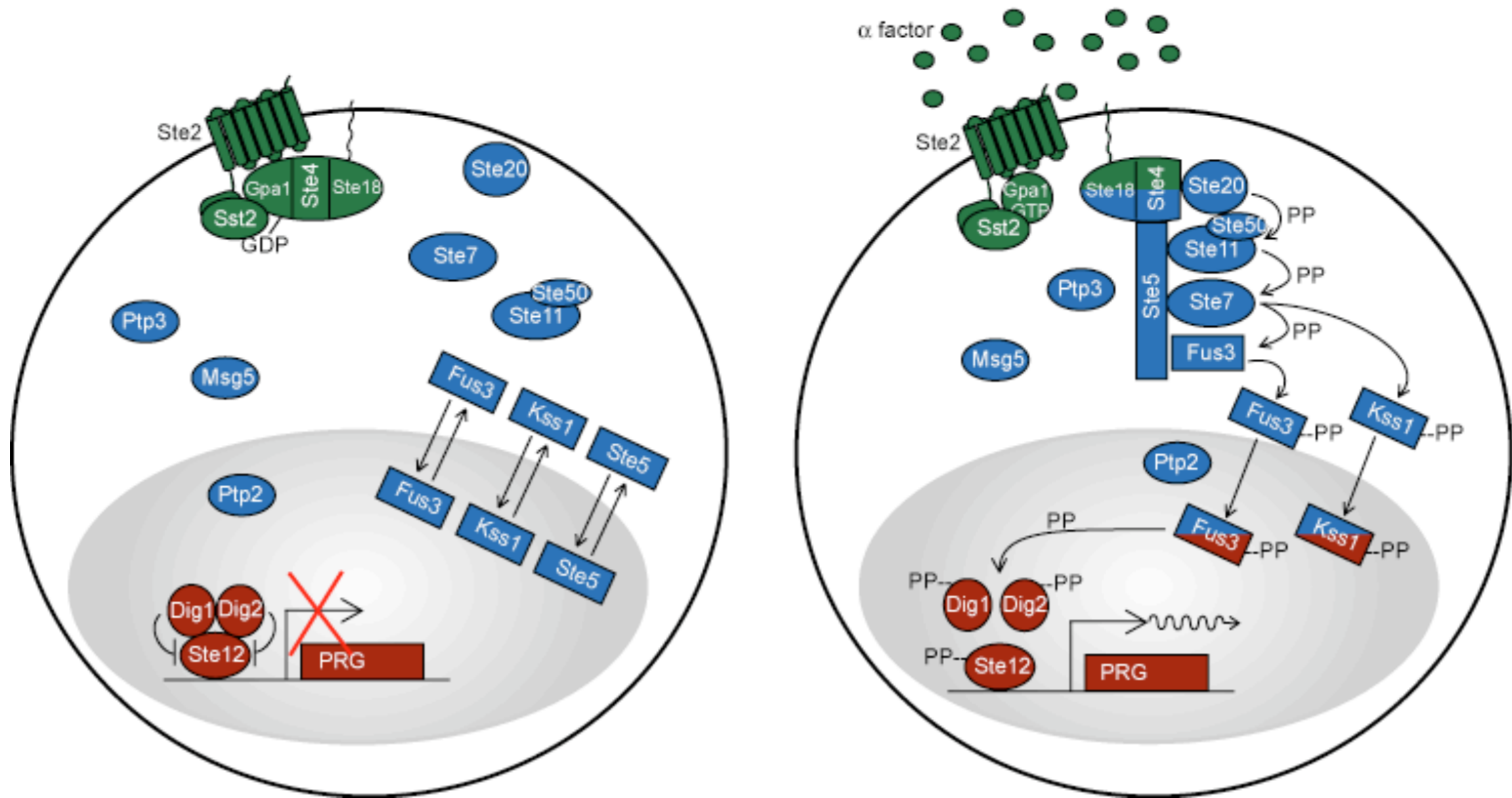


BELIEF/perspective:

1) the manipulations to come -because they are very successful, simple as they are- establish the flexibility (aka reprogrammability, versatility, plasticity, evolvability) of signalling networks; 2) they mirror what variation/selection does

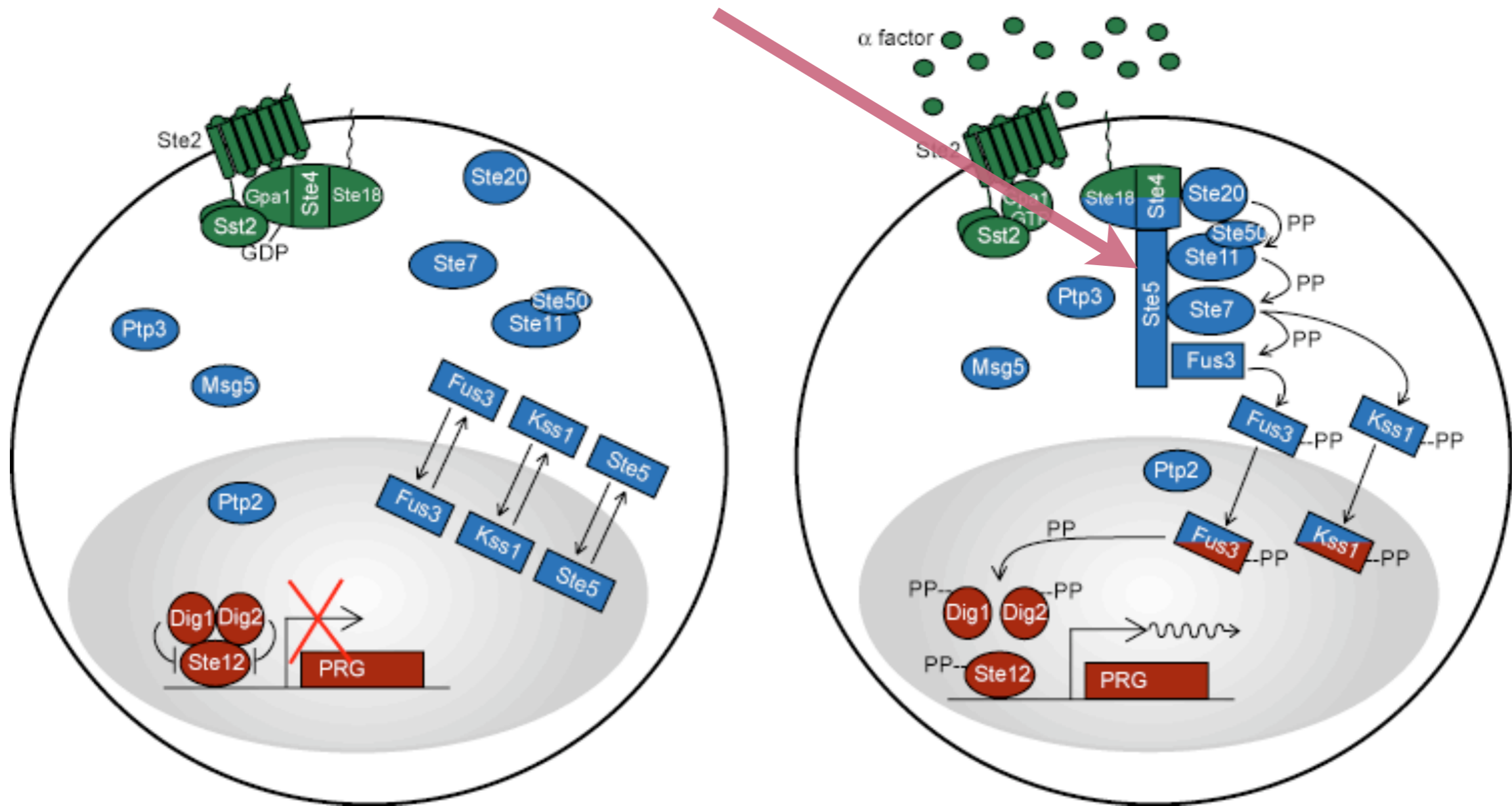
the patient

<http://yeastpheromonemodel.org>



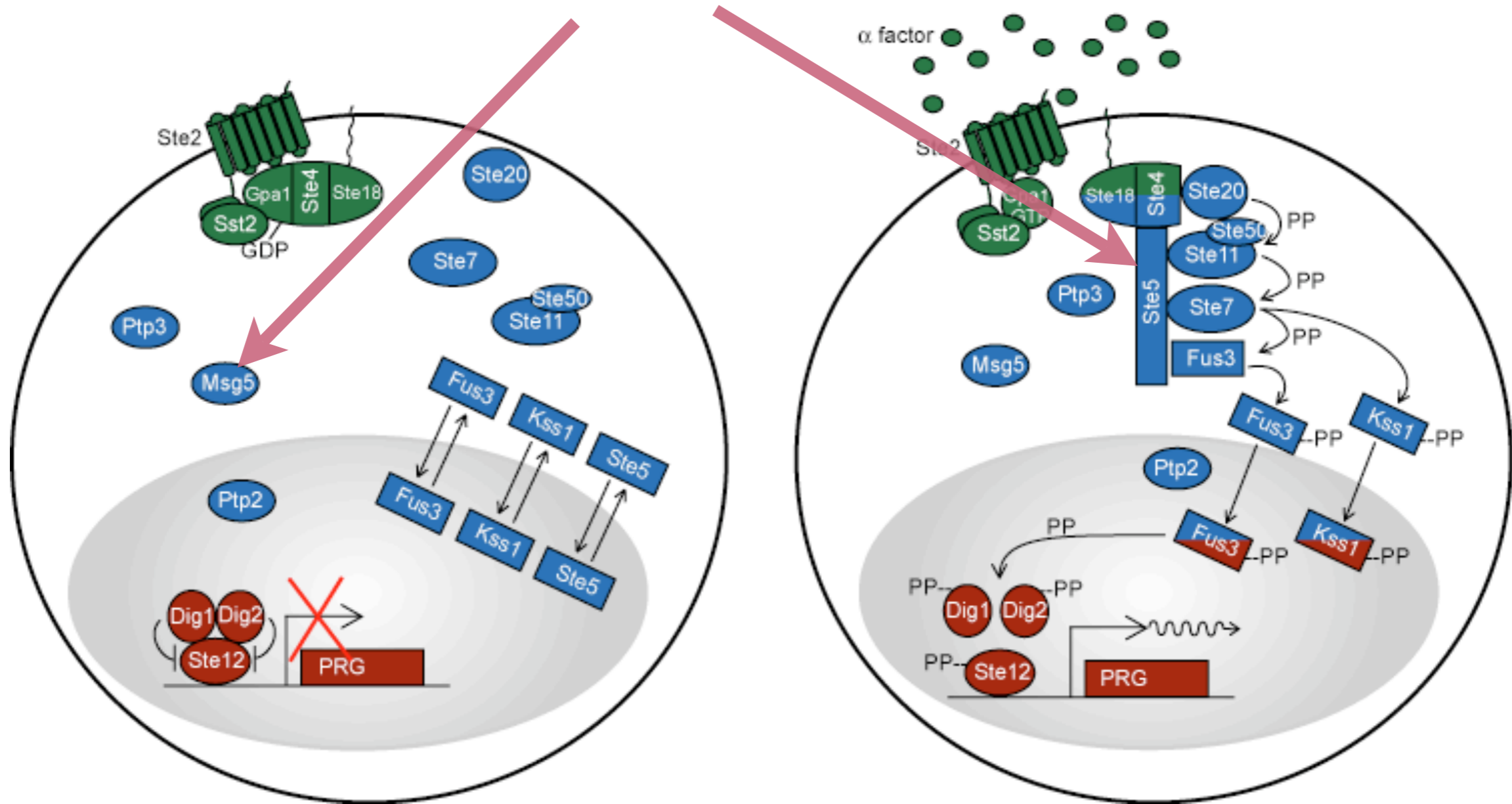
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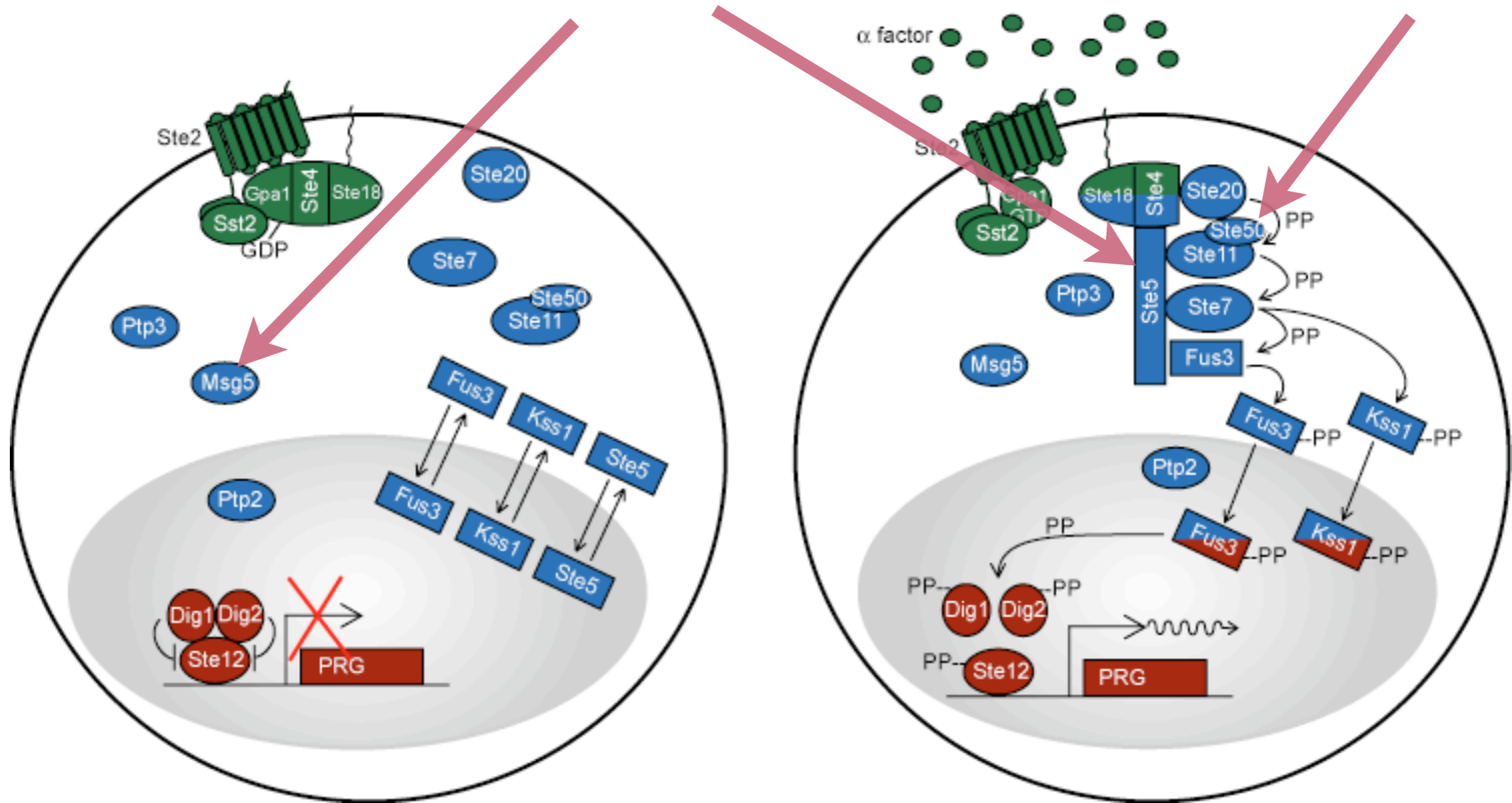
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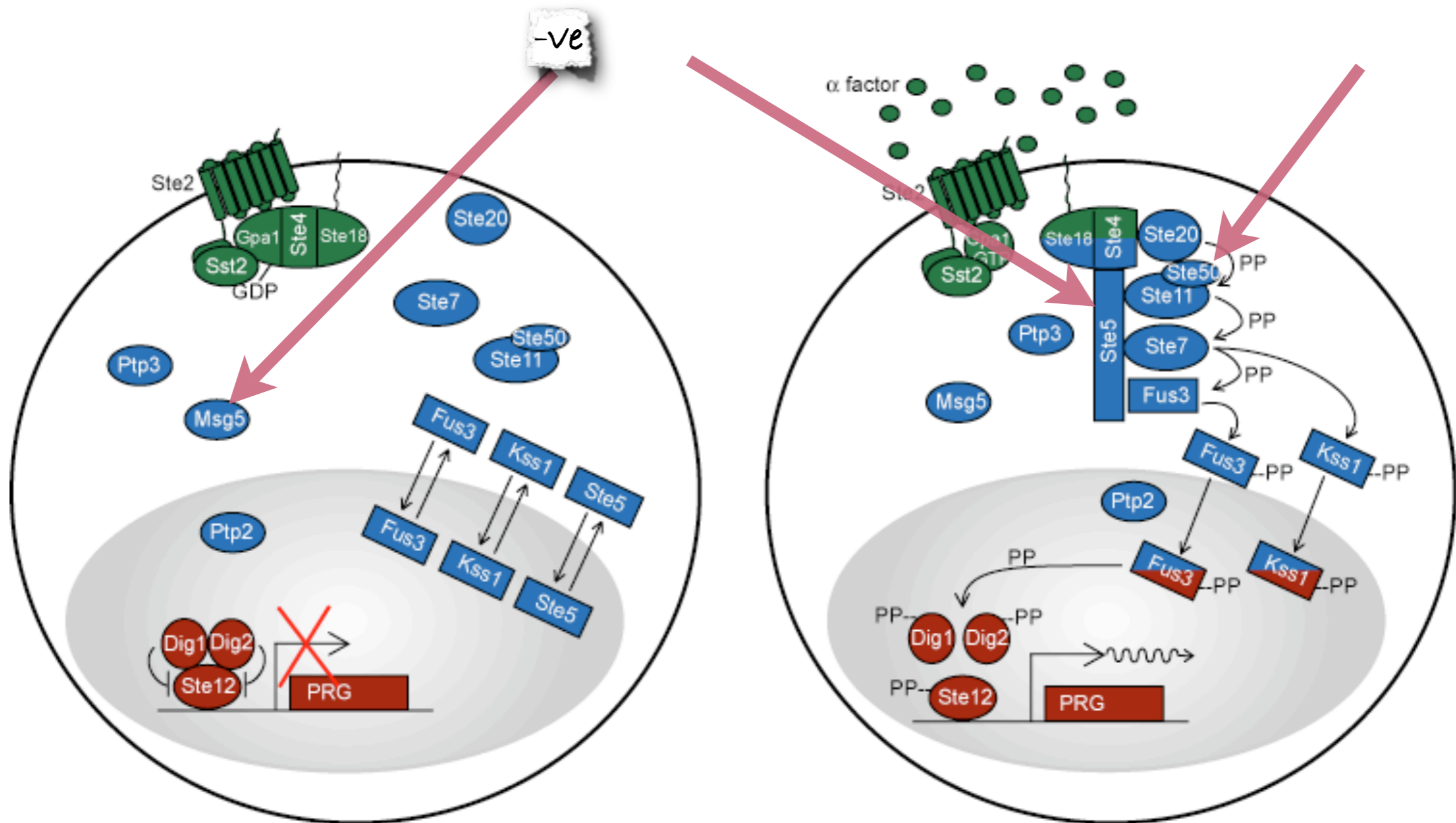
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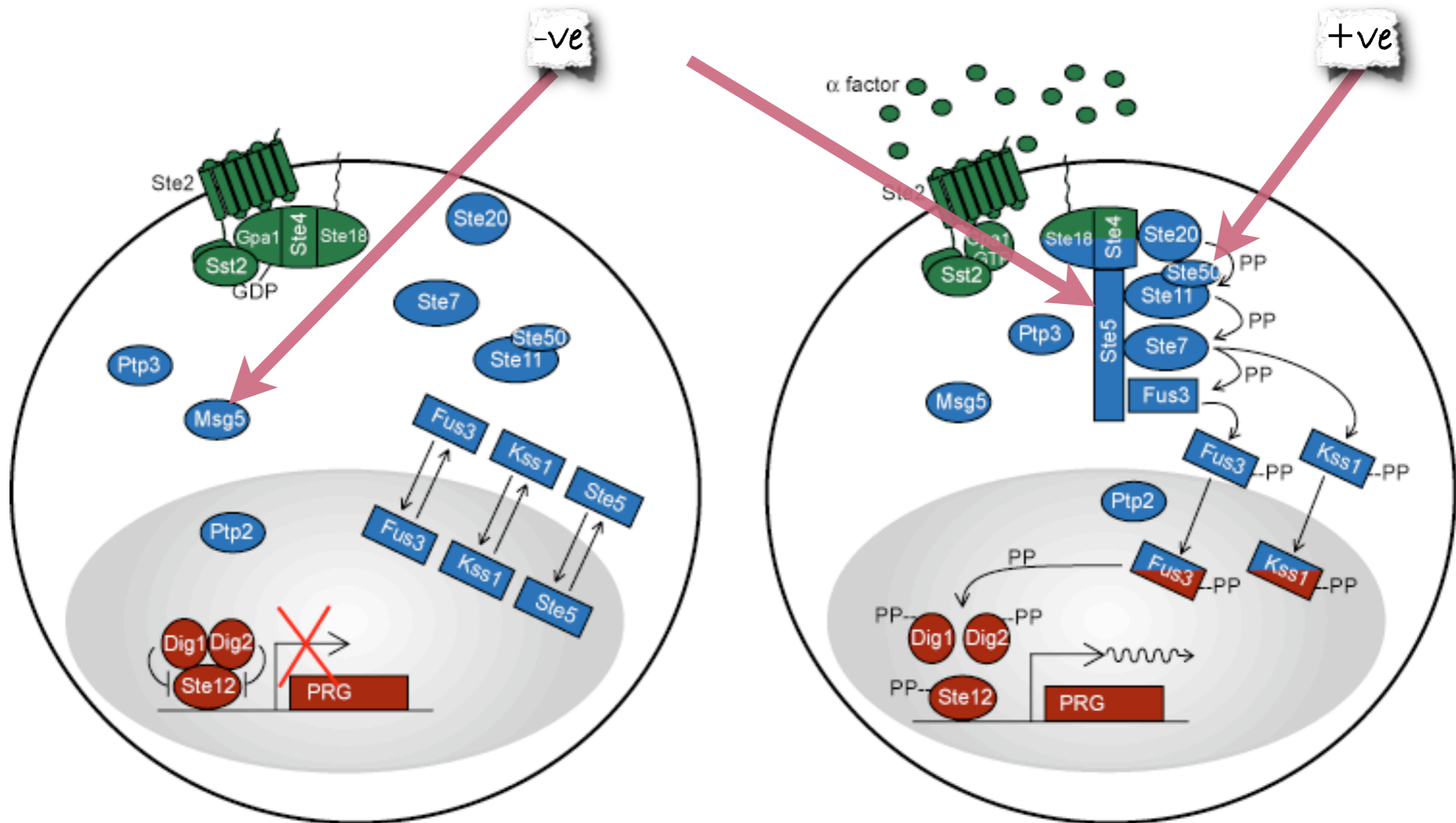
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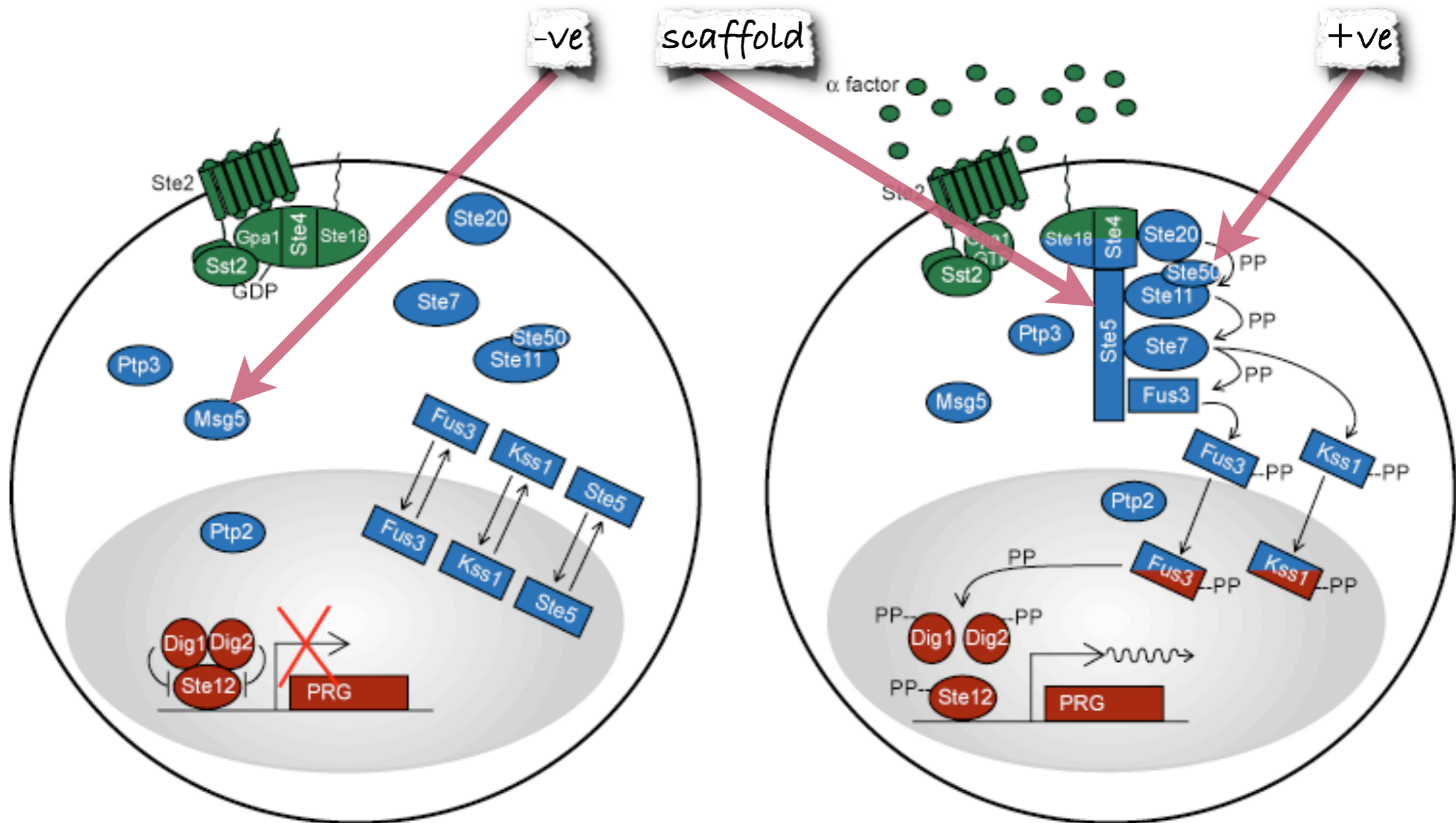
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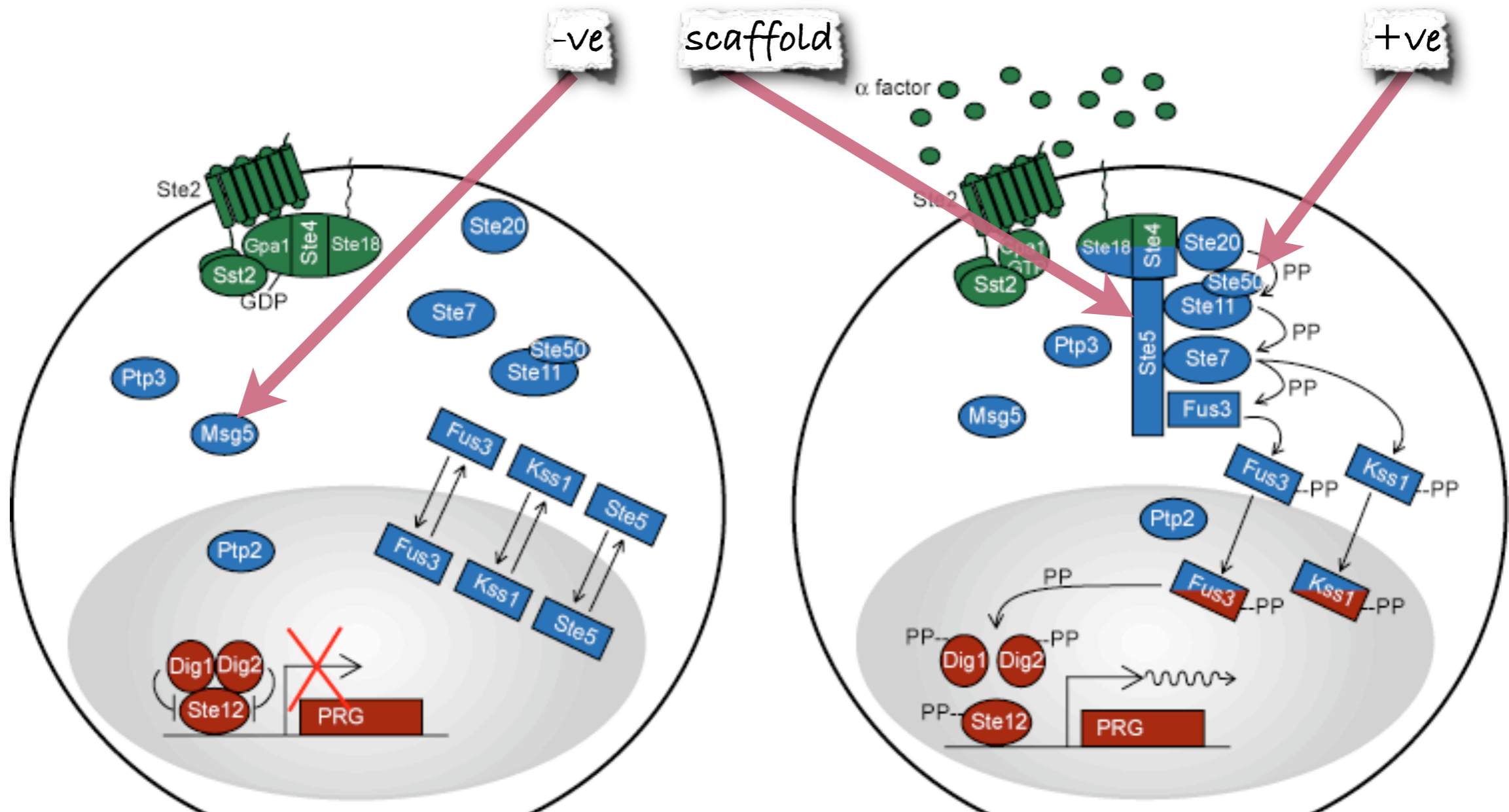
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hooking Ste5 the "mating" scaffold to its 2 modulators -via a tri-FB

concatenations and wires

uses 2 bio-engineering "wires"

1. heterodimerisation PP liquid wire

pairs of leucine zippers (available in different K_d 's -over a range of 10^3)

2. transcriptional DD solid wire

mating-responsive promoters Pmr (available in different strengths pFIG1 or pPRM2)

This gives the following concatenations (aka fusions):

- proteins: Ste5:zip, Msg5:zip', Ste20:zip'

(the last two known to have respectively -ve, +ve influence on response)

- genes: Pmr::Msg5:zip', Pmr::Ste20:zip', and Pmr::GST:zip

(neutral -used as a decoy)

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concatenations and wires (cont')

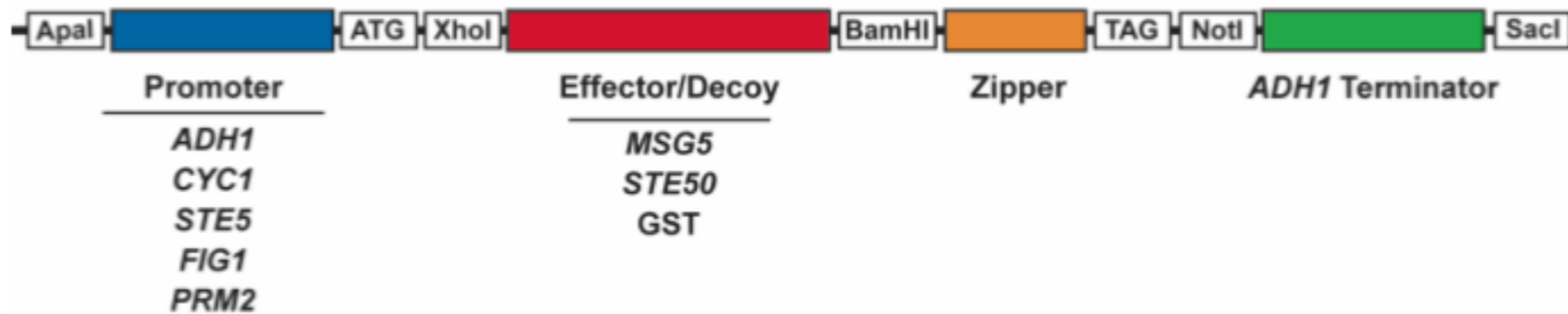
Synthetic Scaffolds

heterodimerisation (protein/protein) liquid wire



Recruited Effectors

transcriptional (DNA/DNA) solid wire



concatenations and wires (cont')

Leucine zipper pair affinities



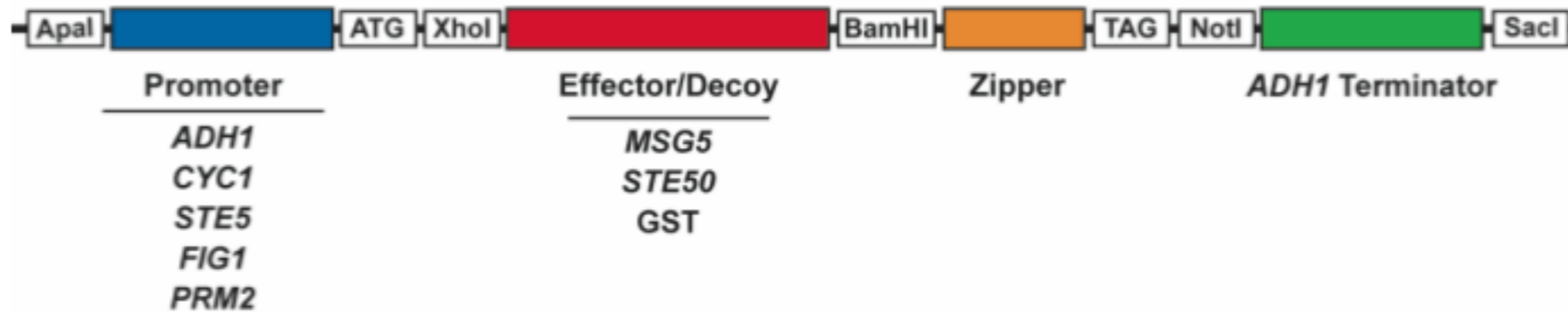
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concatenations and wires (cont')

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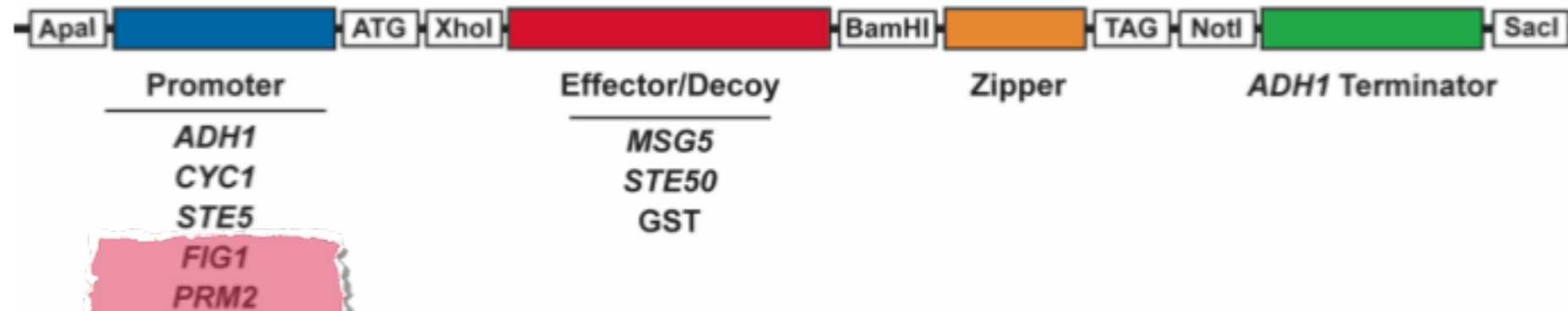
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The constructs/5 steps

1. separate compilation:

constitutive *Msg5:lzip*, *Ste20:lzip* \Leftarrow we verify that they are indeed modulators

2. simple -ve and +ve FB with induced -ve or +ve modulators \Leftarrow loops

3. tuning the above with various *lzip* and promoter strengths

4a. constitutive -ve and induced +ve, and symmetrically \Leftarrow introducing conflict

4b. constitutive decoy and induced -ve, and symmetrically \Leftarrow 3rd party

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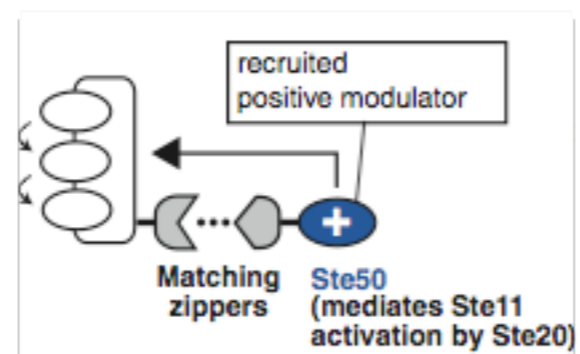
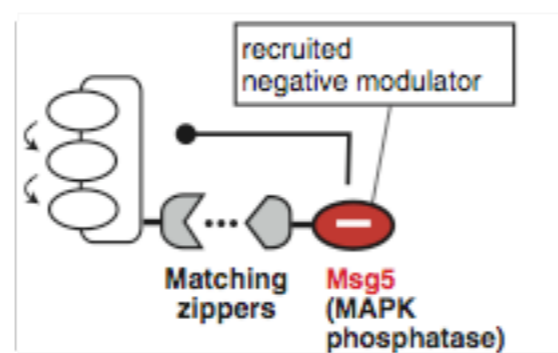
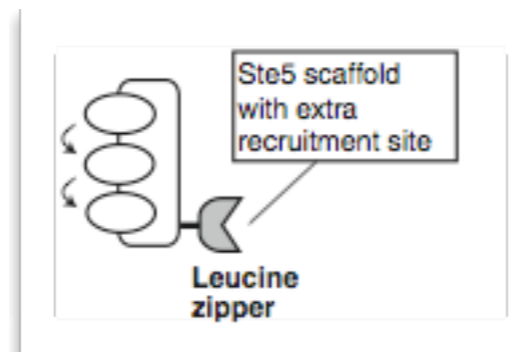
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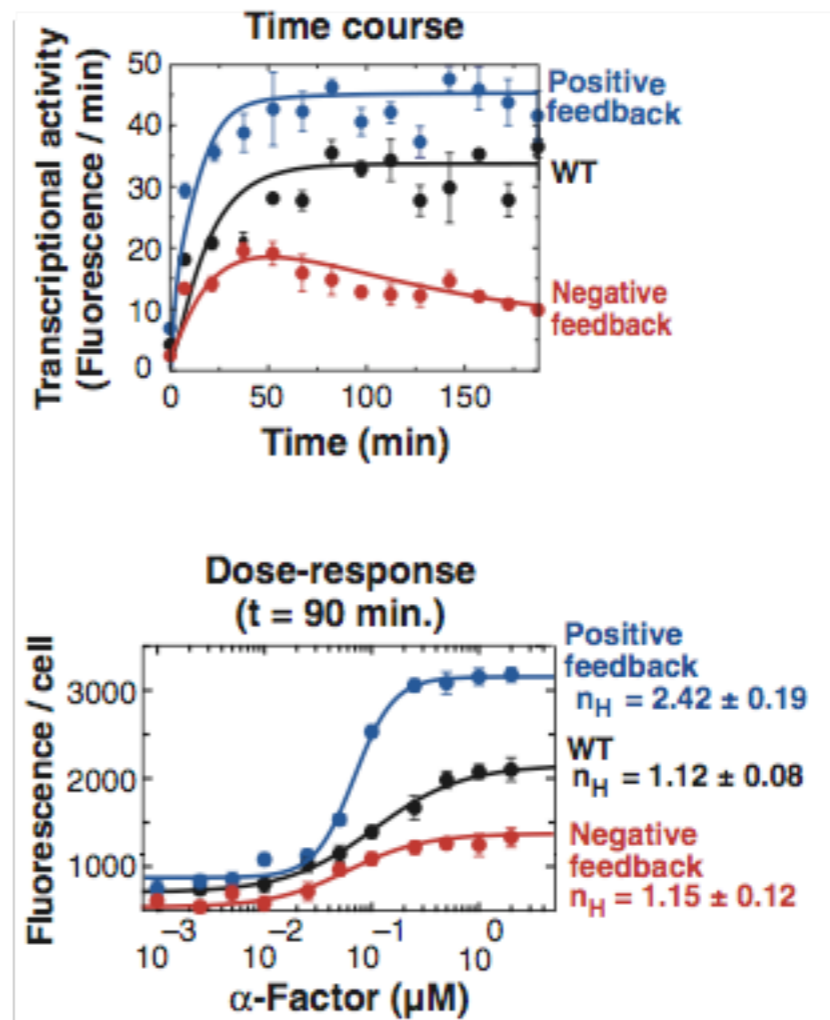
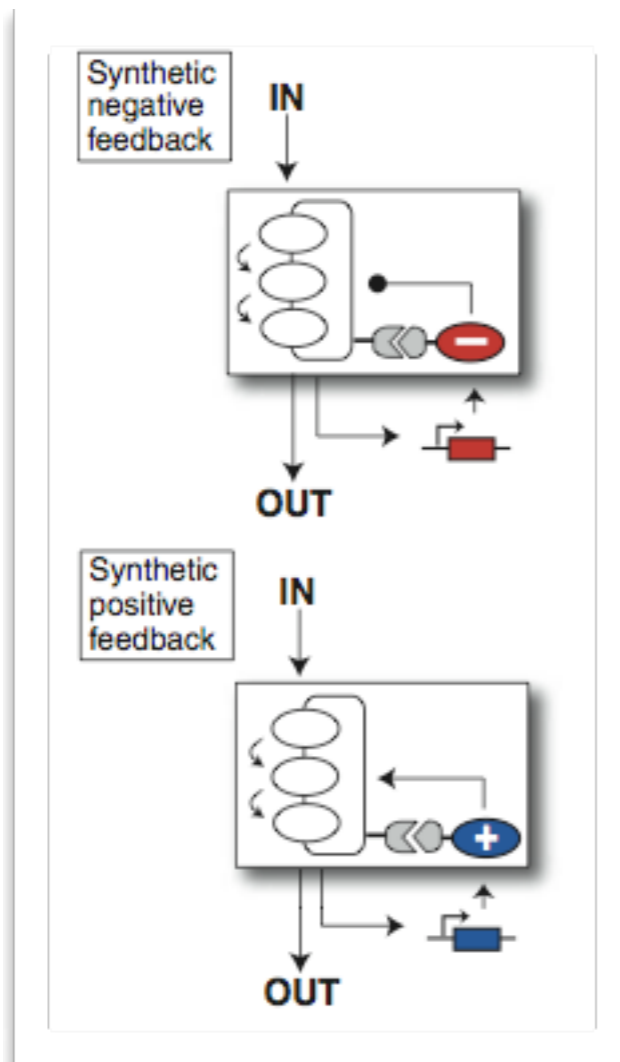
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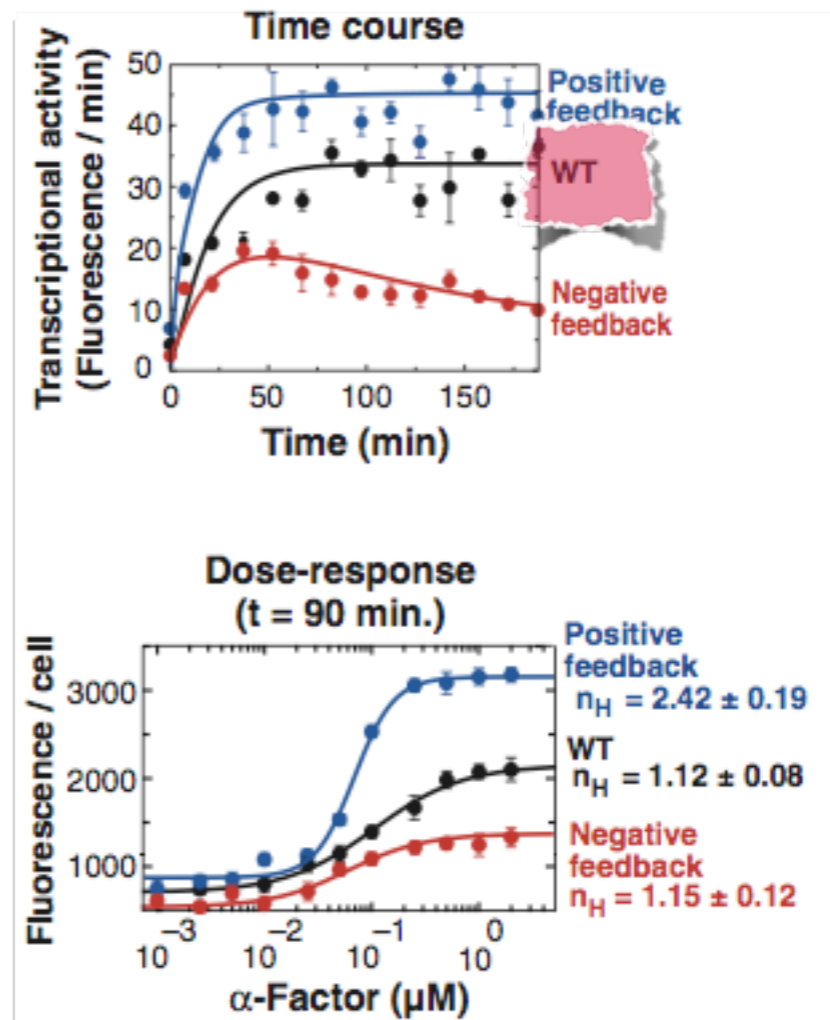
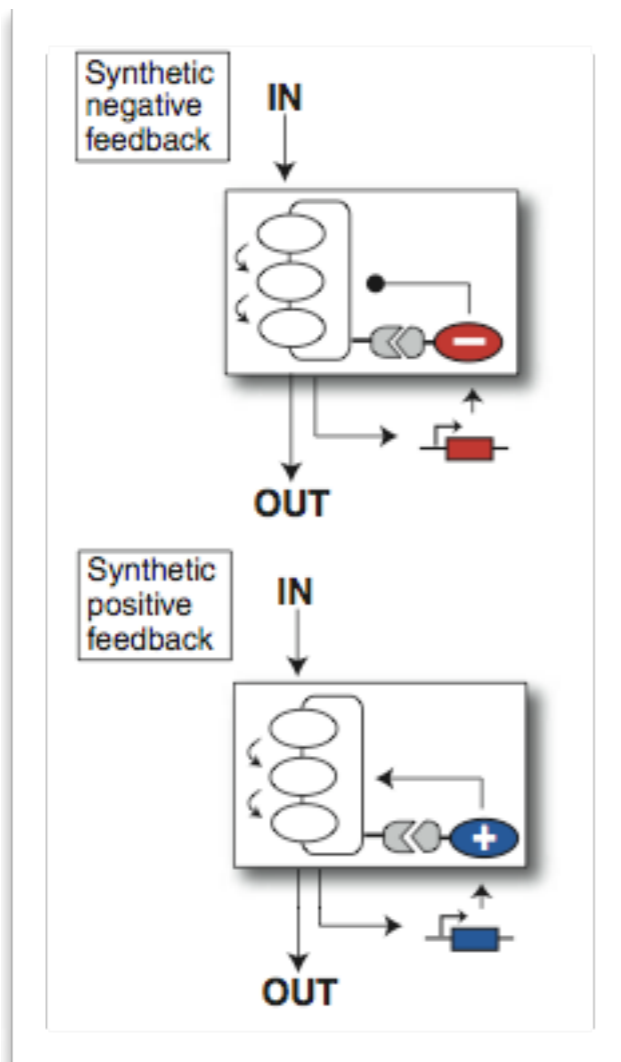
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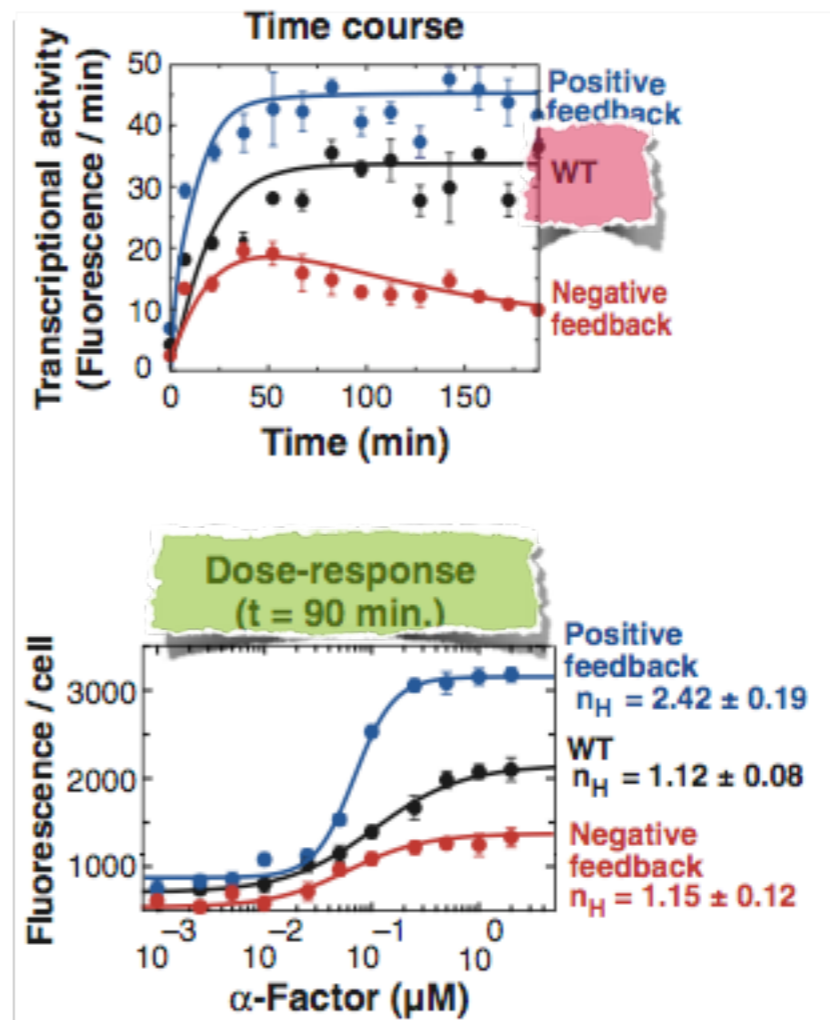
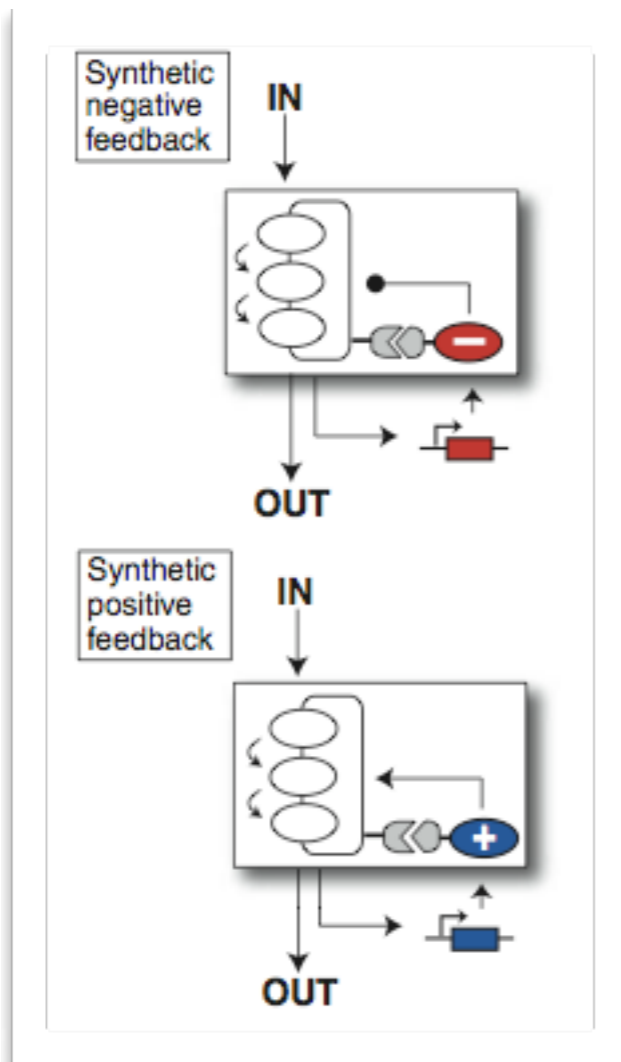
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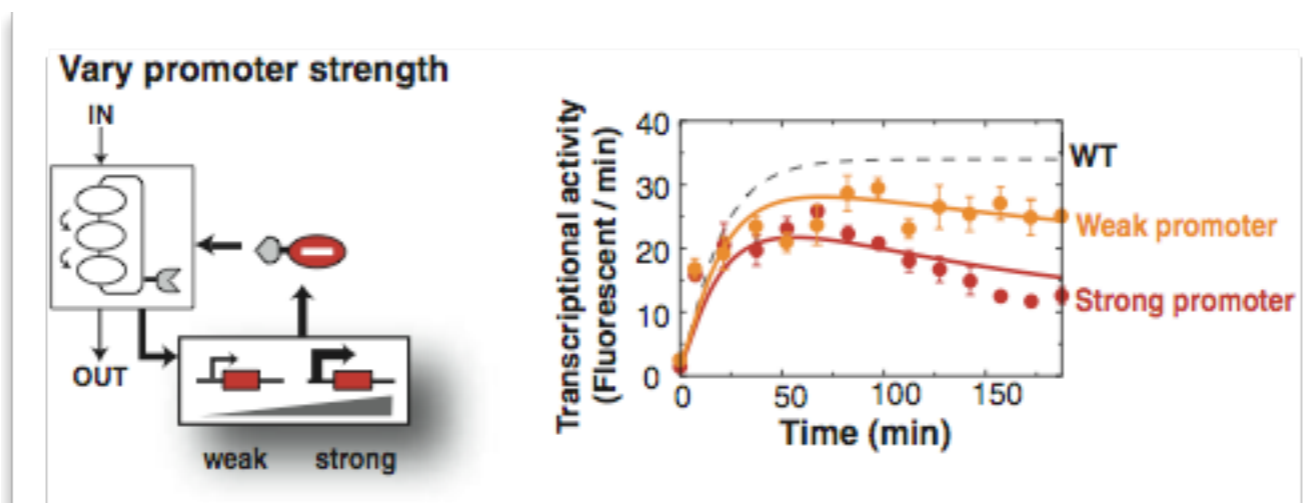
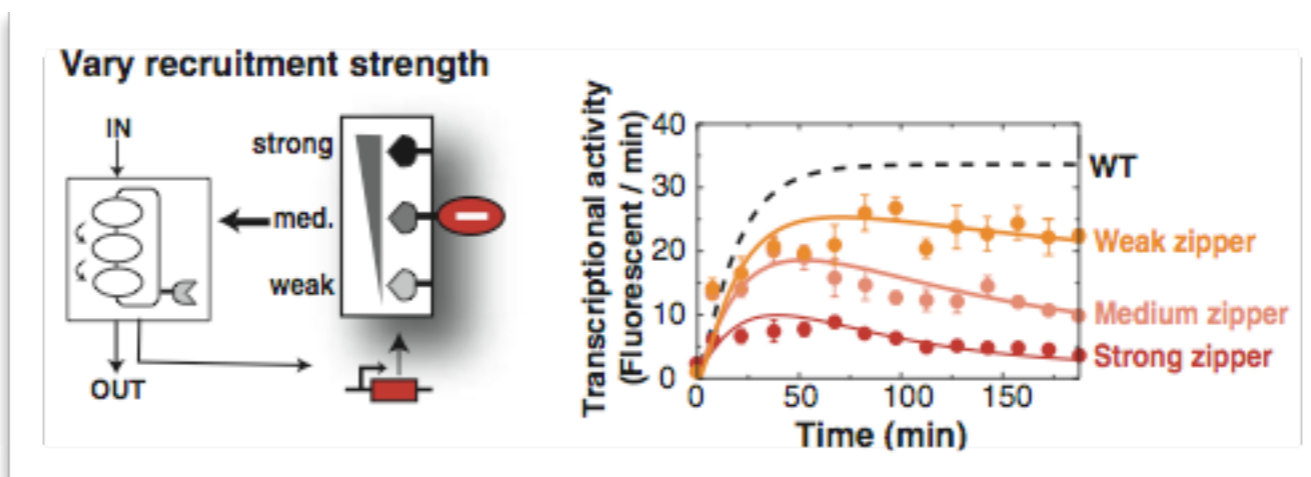
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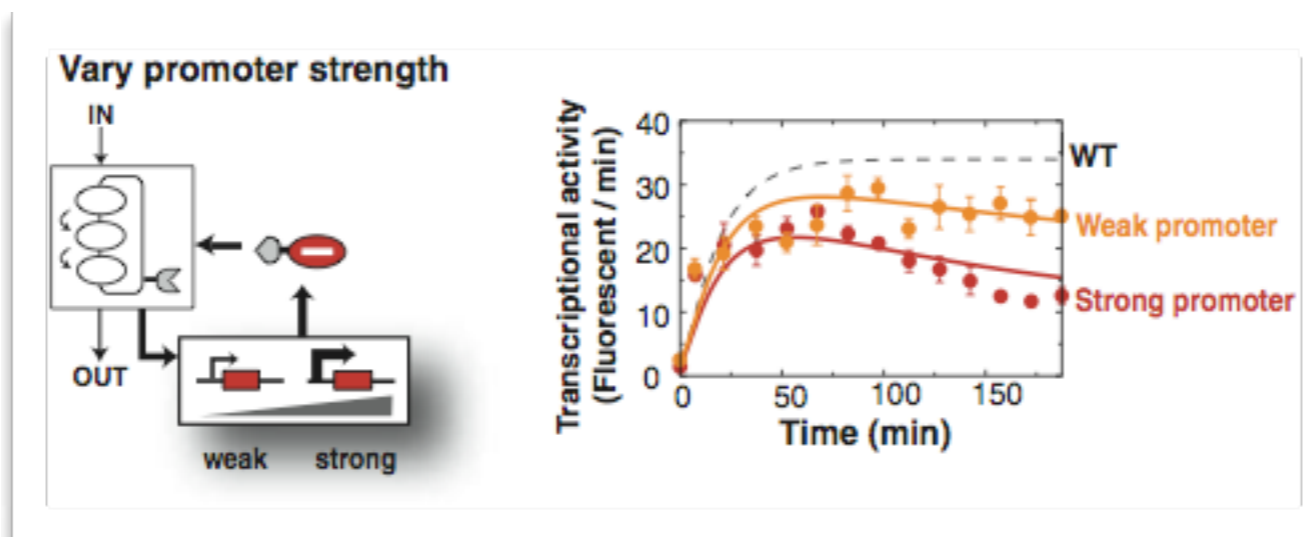
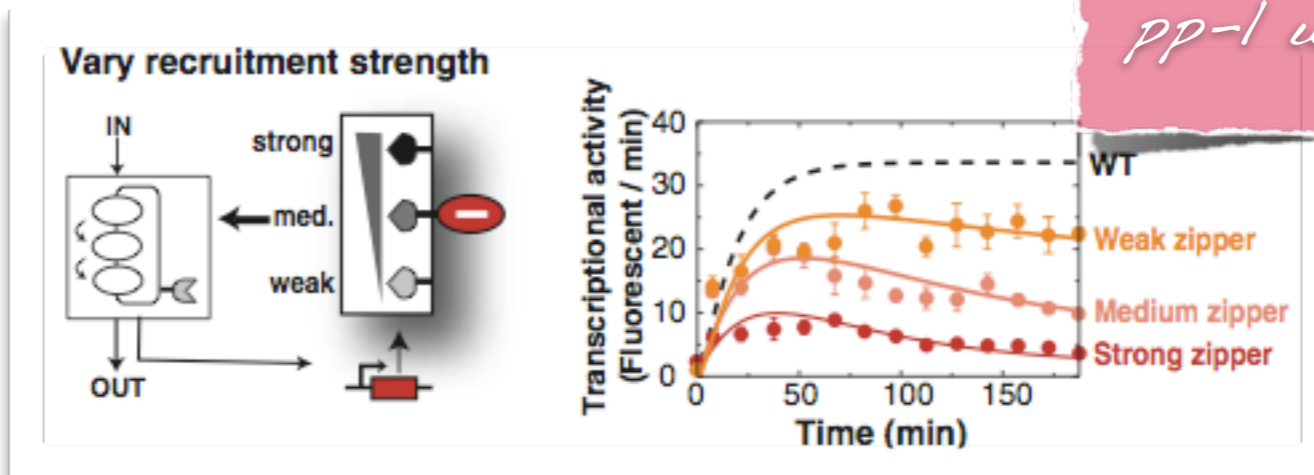


3. tuning the above with various zipper and promoter strengths

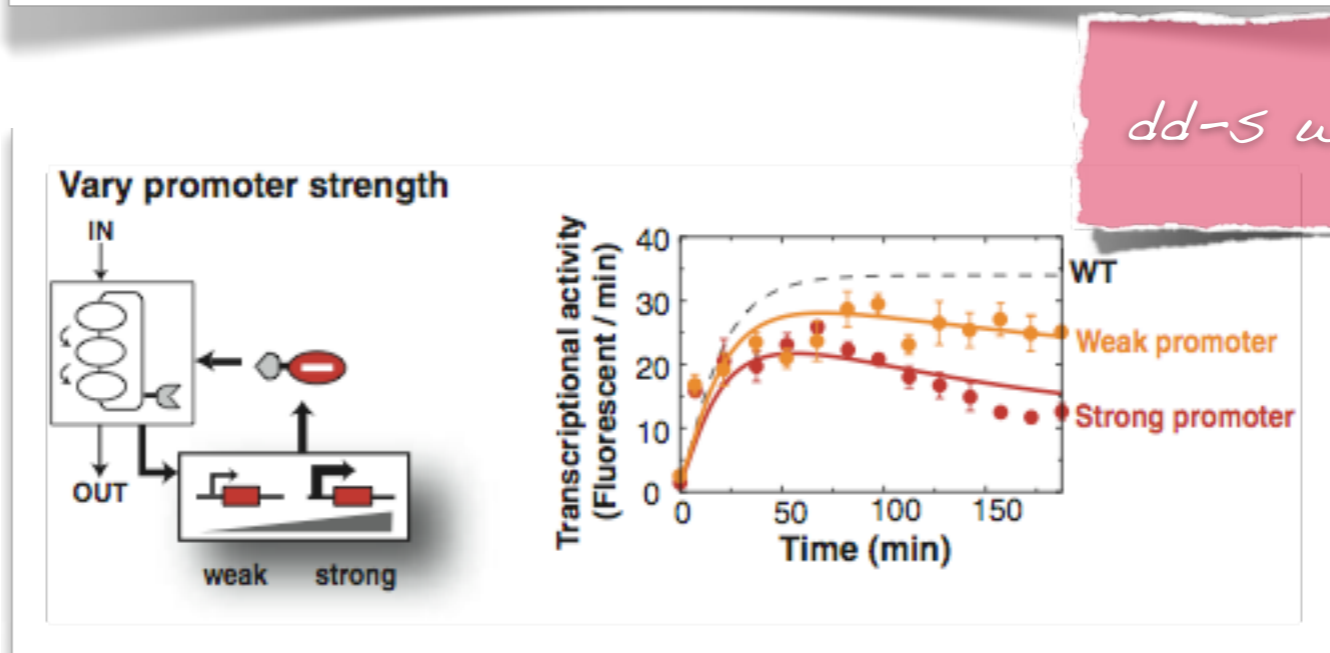
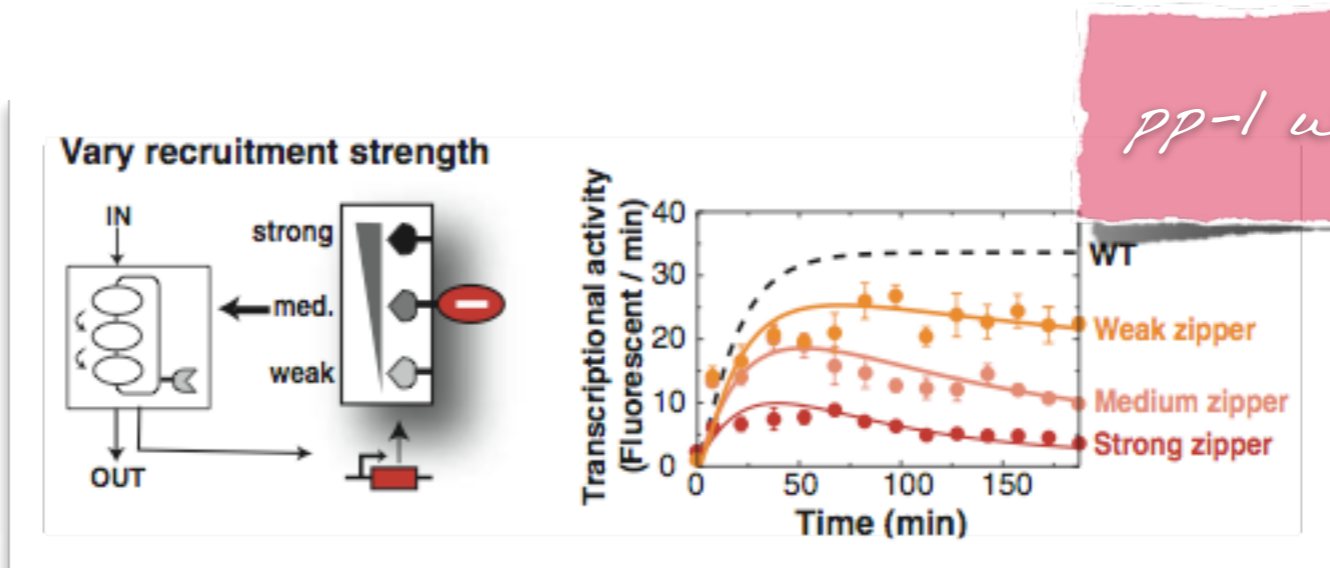


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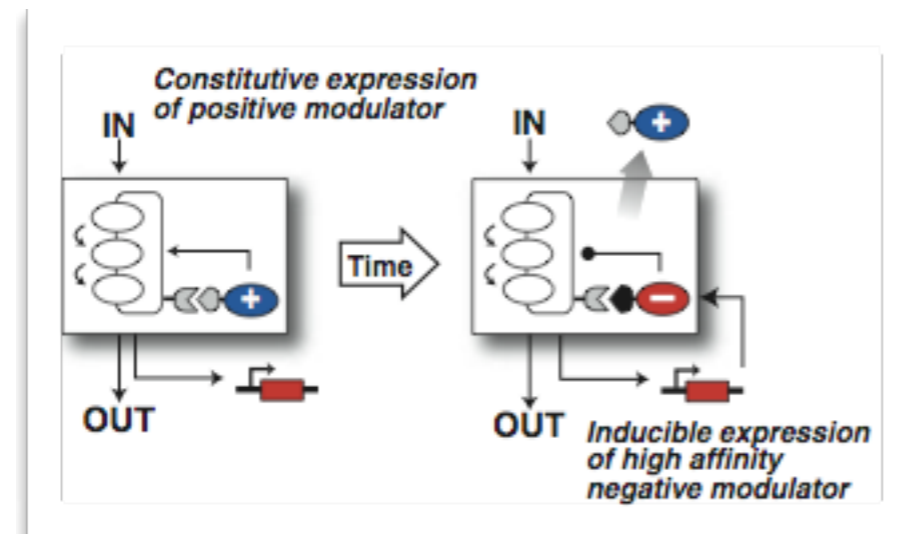
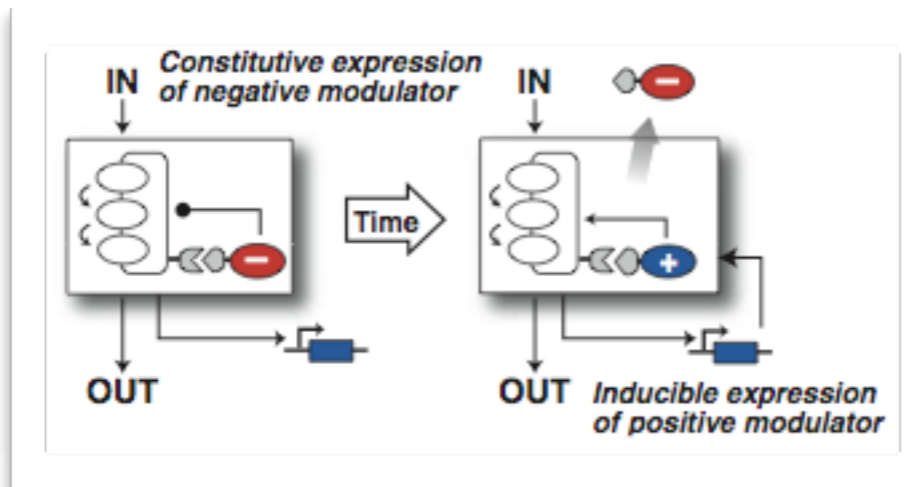
pp-1 wire strength



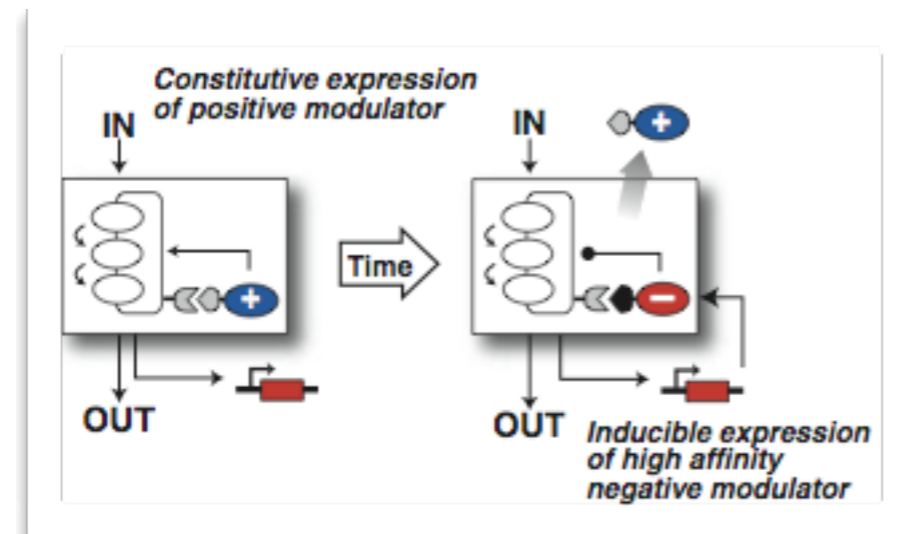
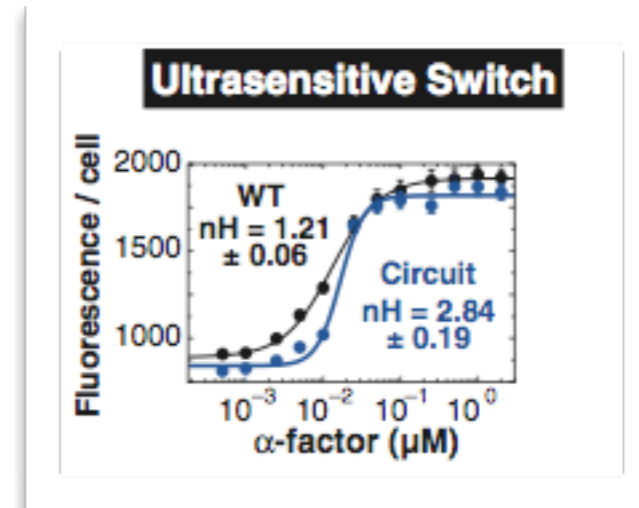
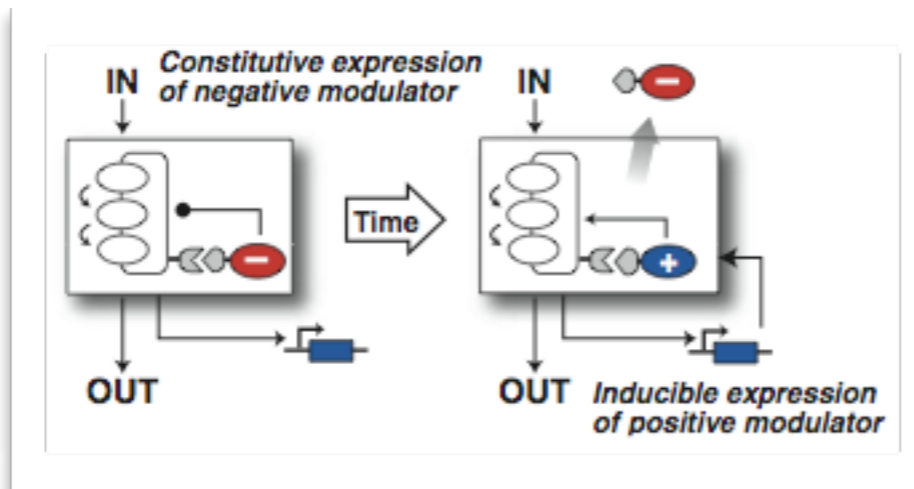
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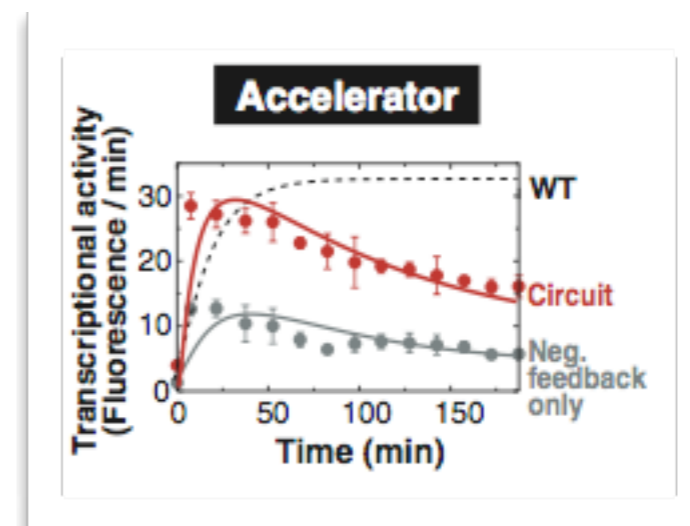
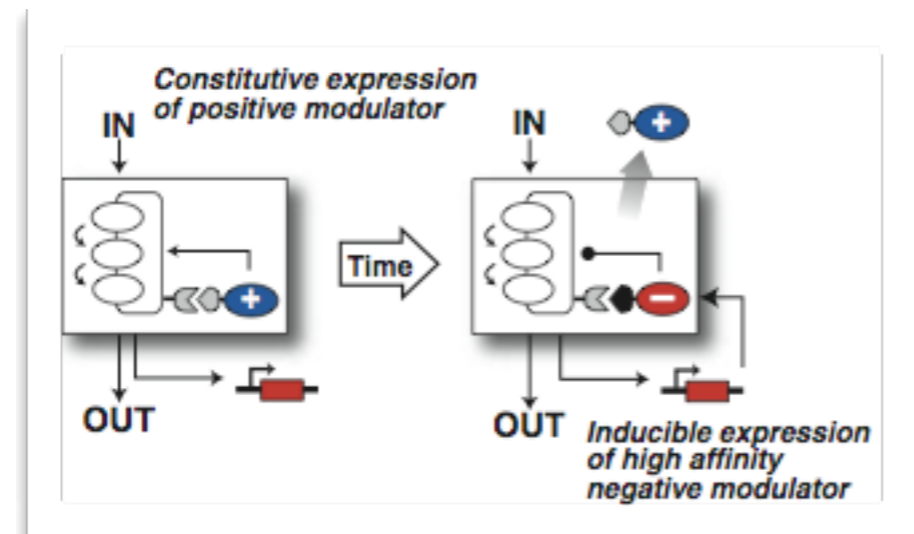
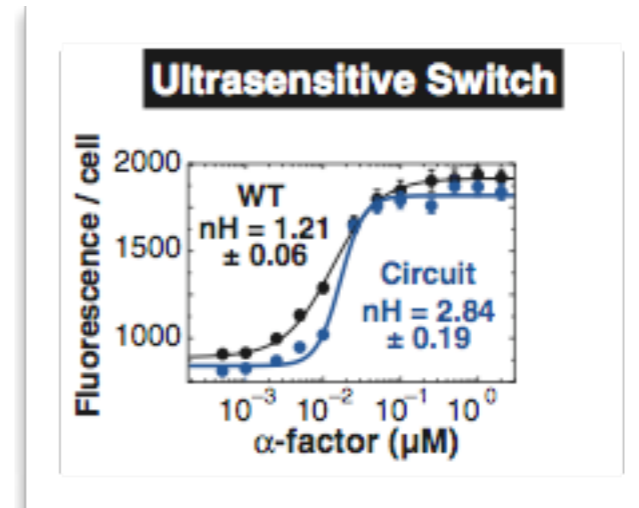
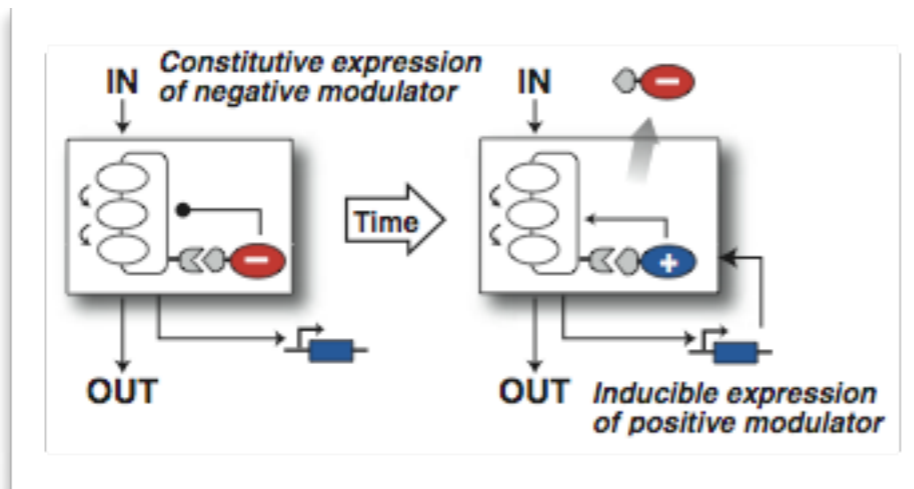
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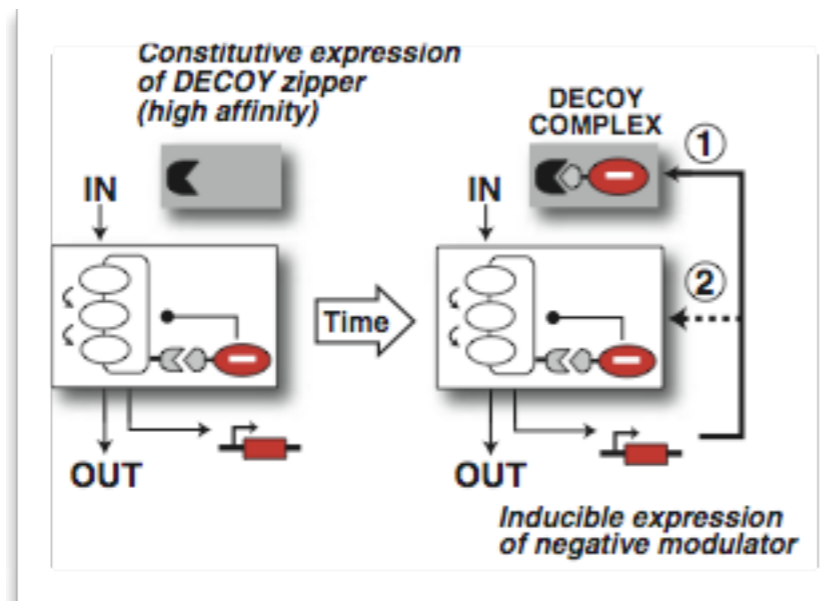
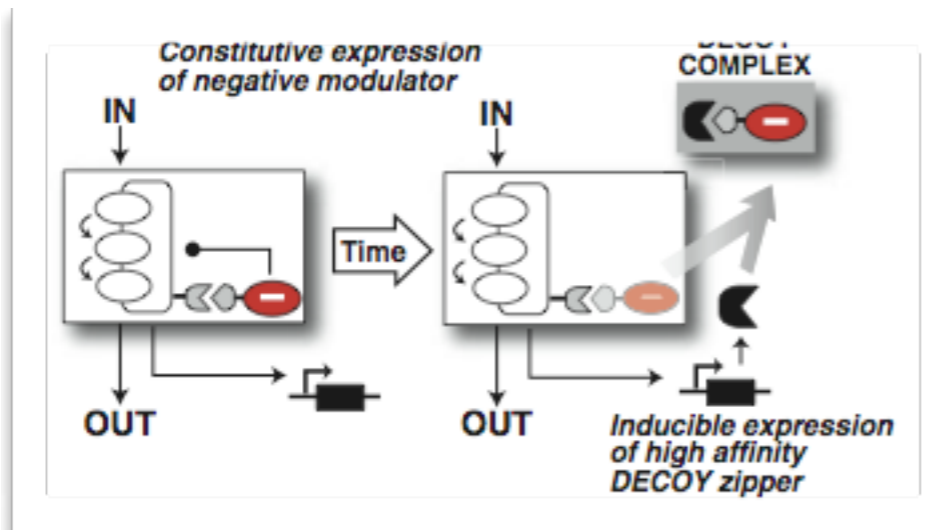
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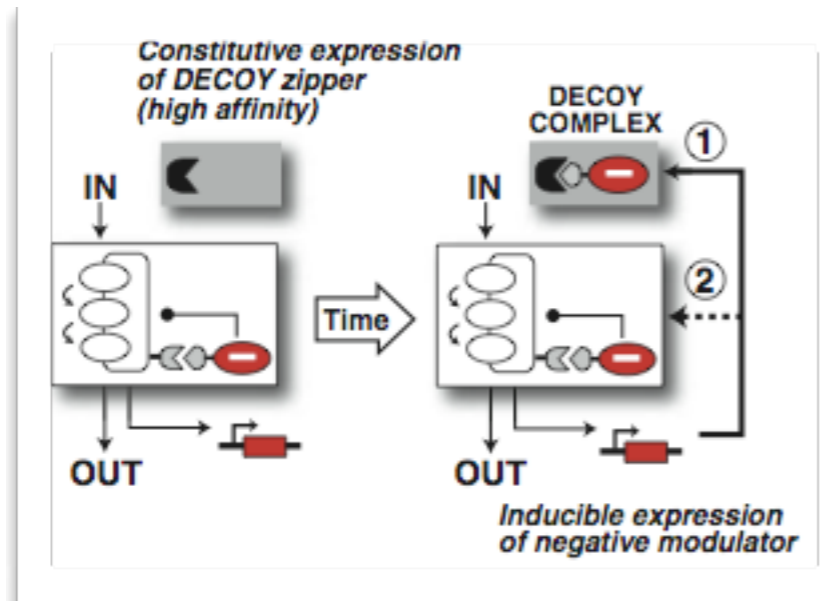
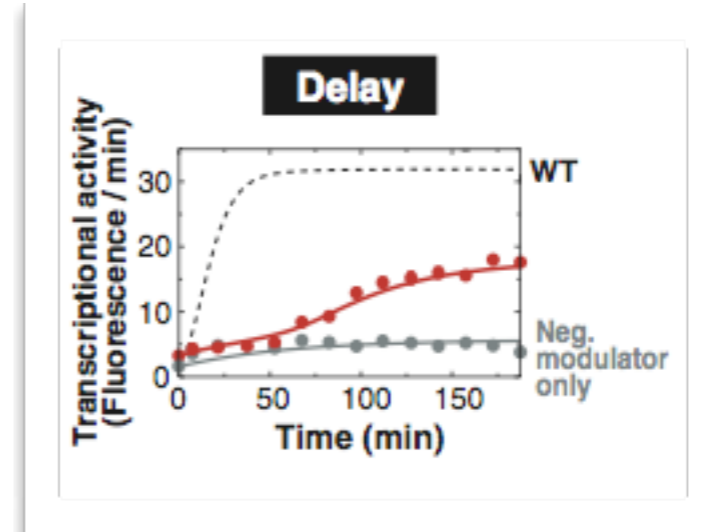
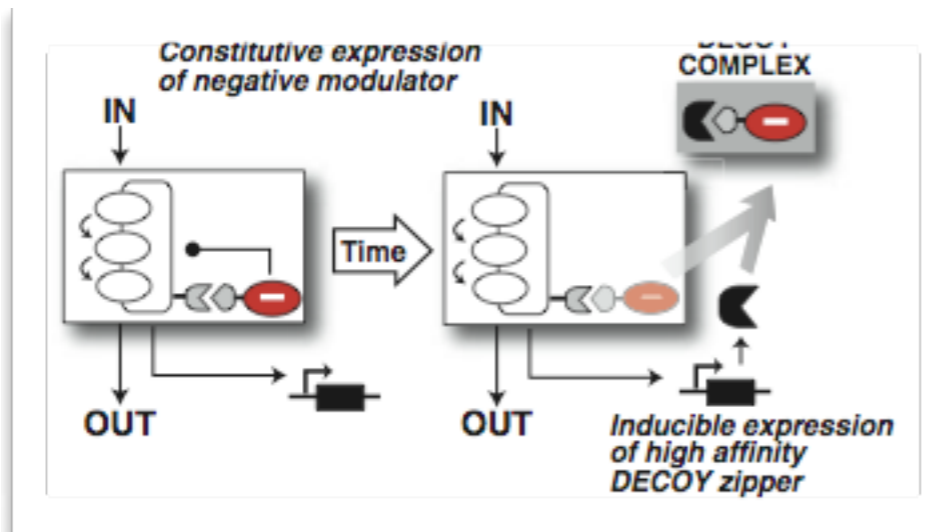
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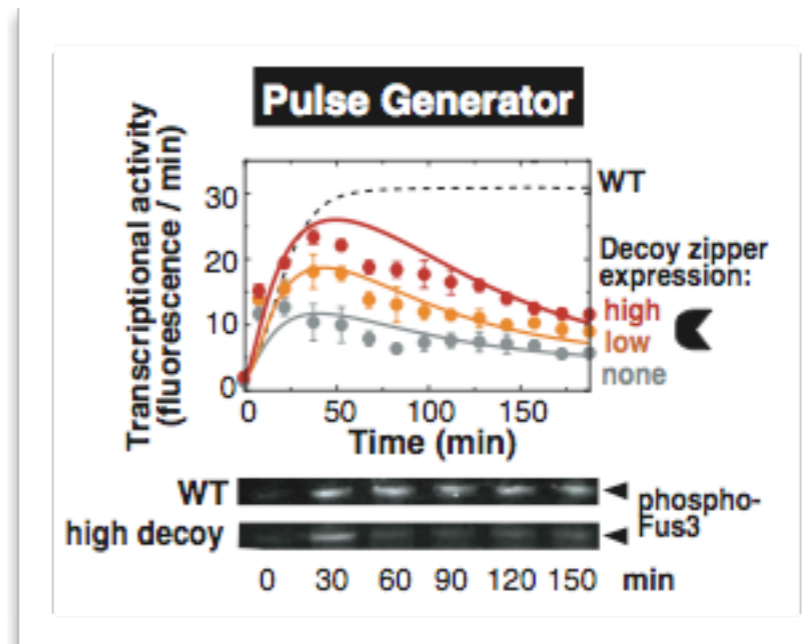
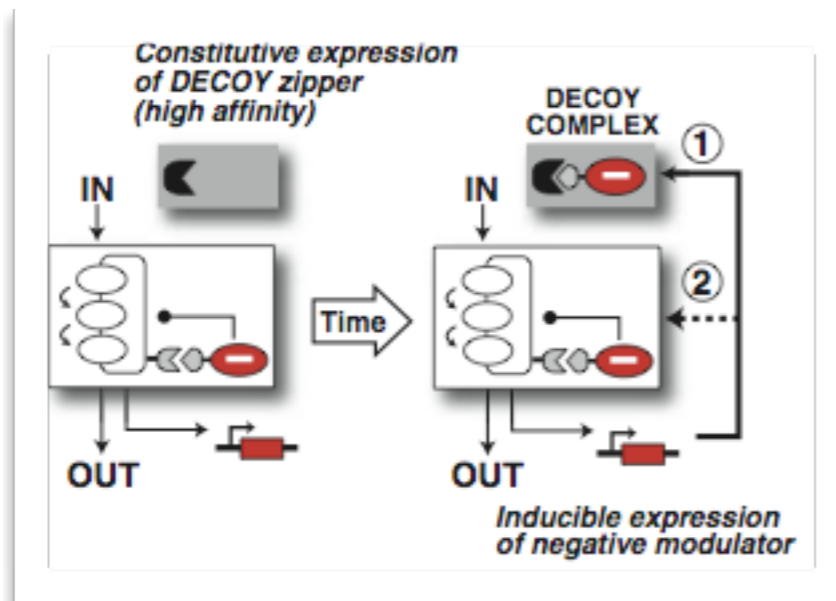
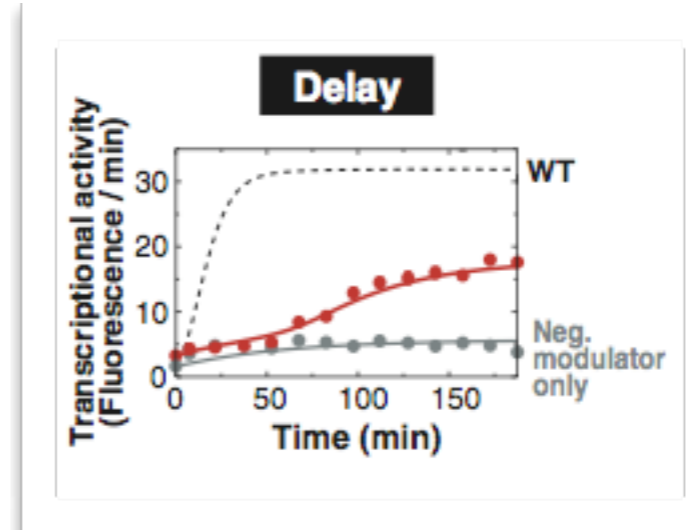
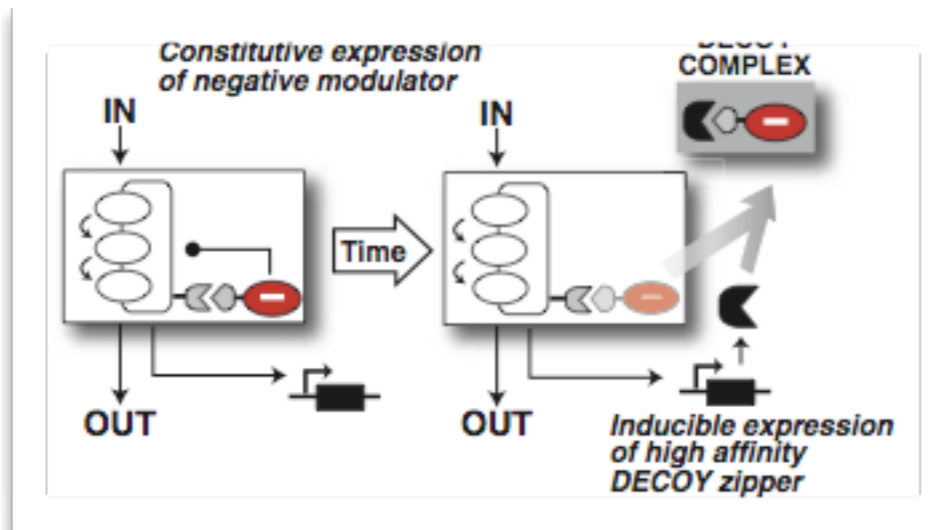
4b. constitutive decoy and induced -ve, and symmetrically



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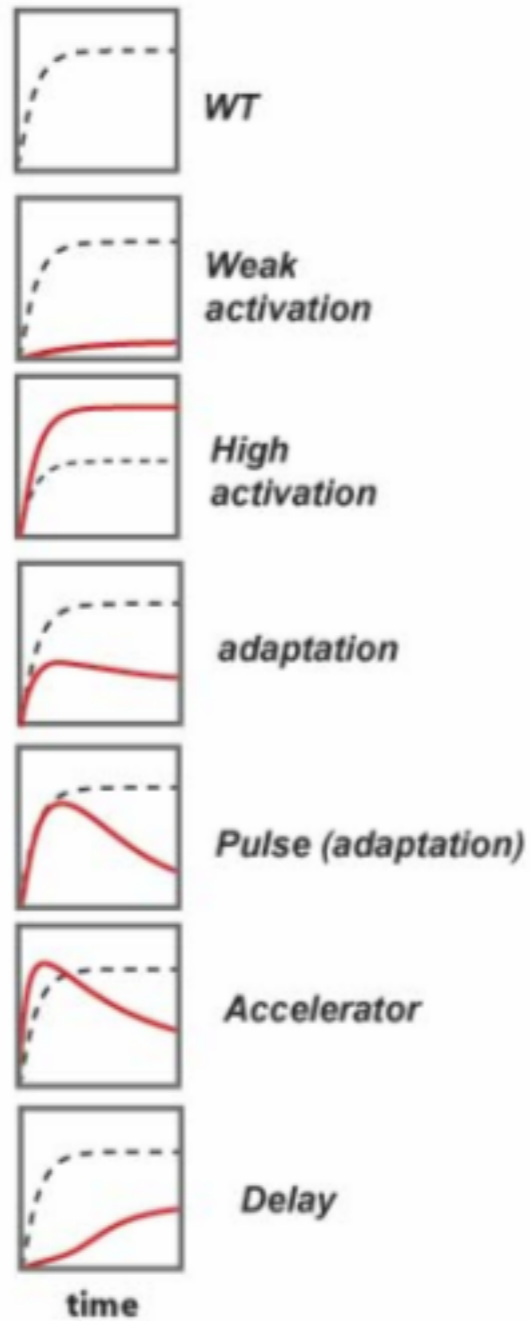
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- ✓ Elo approach where all this is done at once combinatorially?
- ✓ This is all going through a slow transcriptional step
- ✓ is it concentration/gradient or binding??
- ✓ leucine-zipper physically flexible?

aside: the behaviour classifiers (CTL-ish)

Dynamic Behavior Classes



Dose-Response Behavior Classes

