Co-Design in Synthetic Biology

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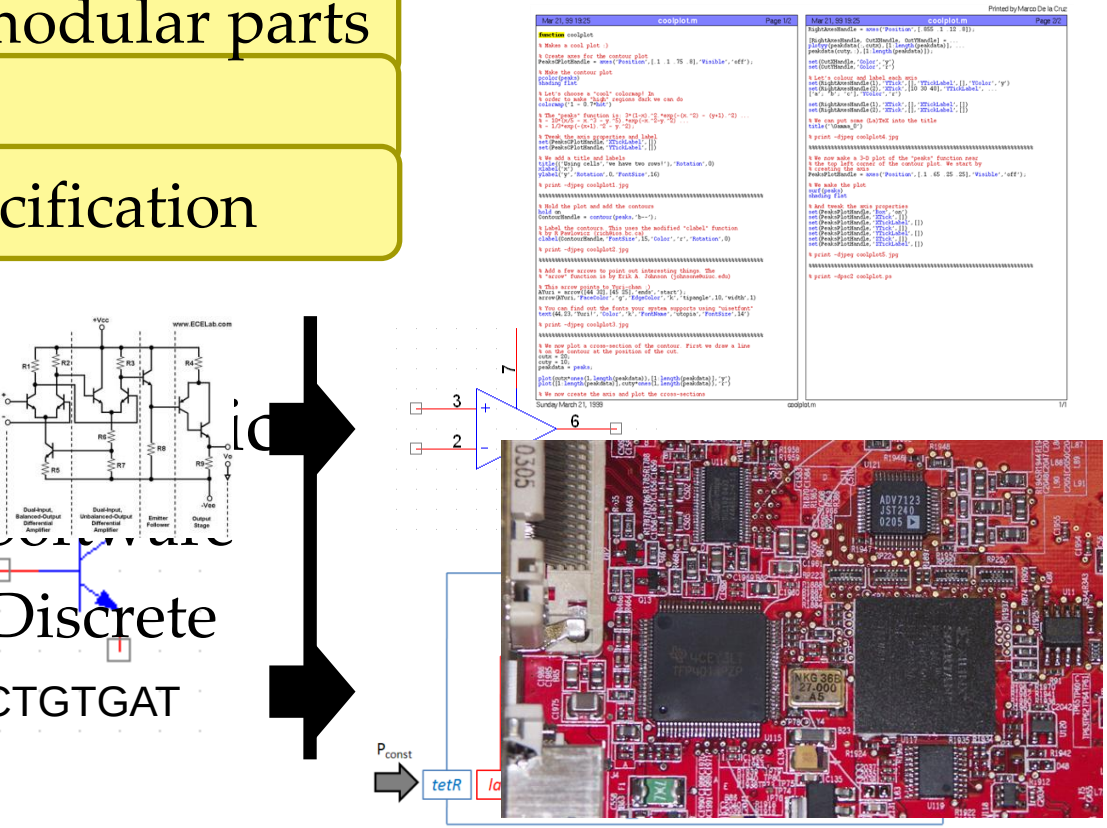
Engineering Biology to Specification

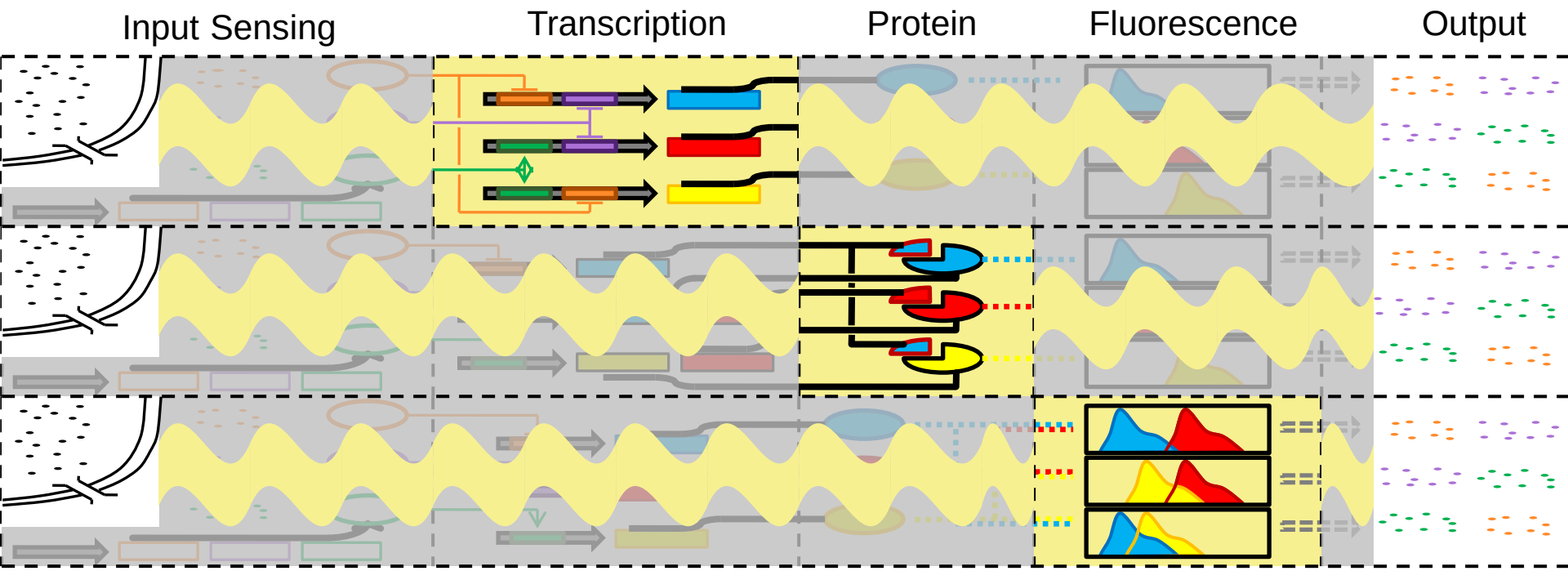
- Goal: Engineer cells like electronics
  - Predictable, modular parts
  - Abstraction
  - Design to specification



- Co-Design in E[illegible]id
  - Hardware vs. Software
  - Integrated vs. Discrete
  - Etc.

Co-Design in Biology

- Implement at different layers of cellular control
  - Transcription, translation, post-translation, detection



Why Co-Design?

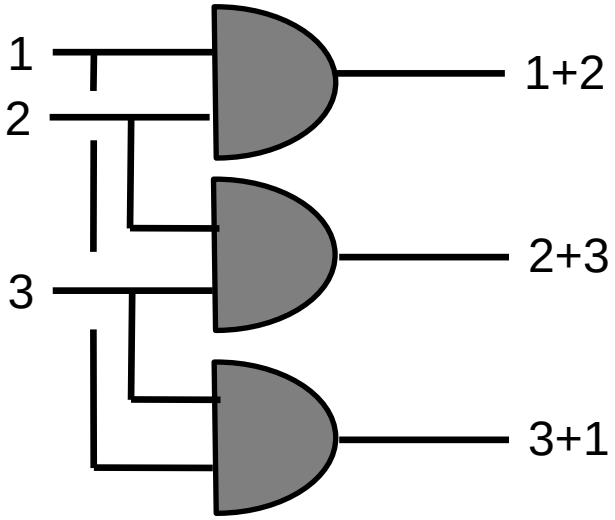
- Compare multiple approaches

- Design in synthetic biology literature typically limited to transcriptional control
  - Some alternative methods, few designs

- Load on cells generally unknown
  - Spread load out
  - Transcriptional control somewhat inefficient

Our Specification

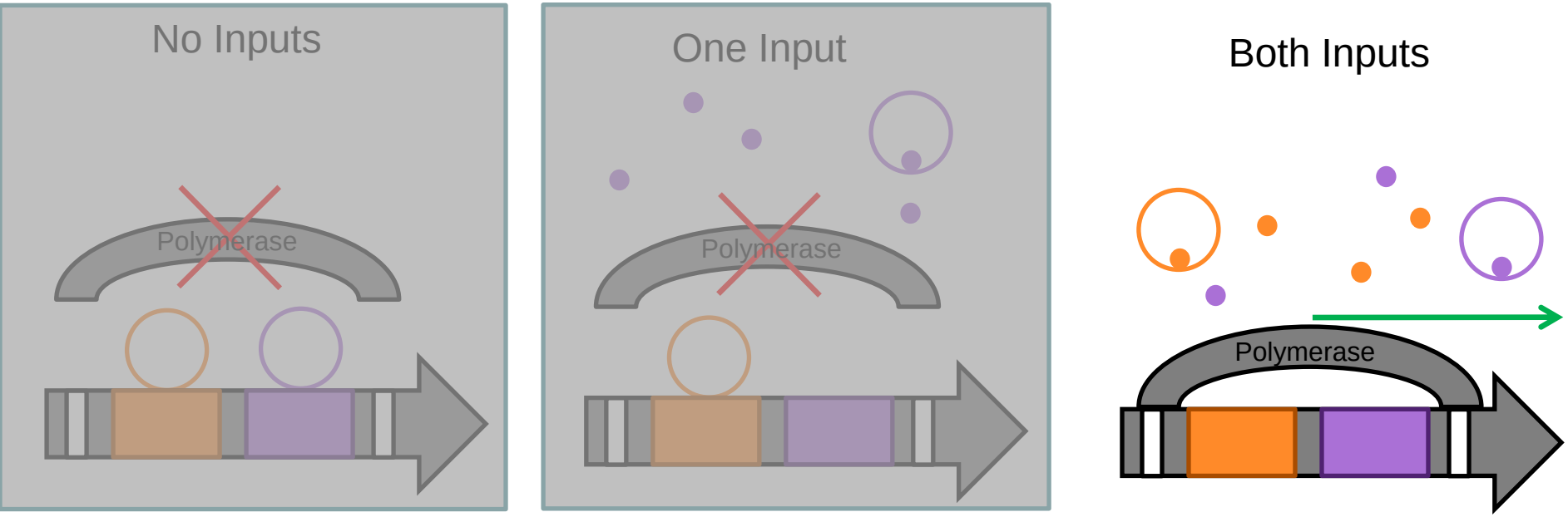
- Collaboration with defense contractor MITRE
- 3 Inputs, unique output for any pair on inputs



Input 1	Input 2	Input 3	Output 1	Output 2	Output 3
-	-	-	-	-	-
-	-	+	-	-	-
-	+	-	-	-	-
-	+	+	-	+	-
+	-	-	-	-	-
+	-	+	-	-	+
+	+	-	+	-	-
+	+	+	+	+	+

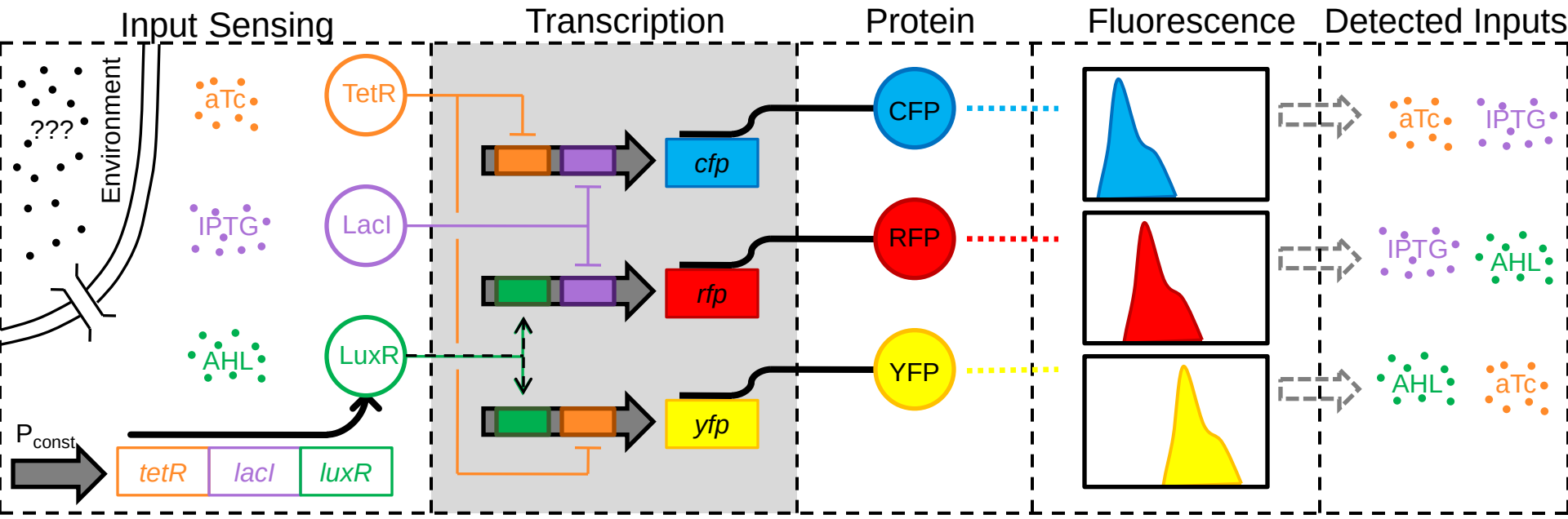
Approach 1: Transcriptional Control

- Sense transcription factor inducers
- Two operators governed by transcription factors



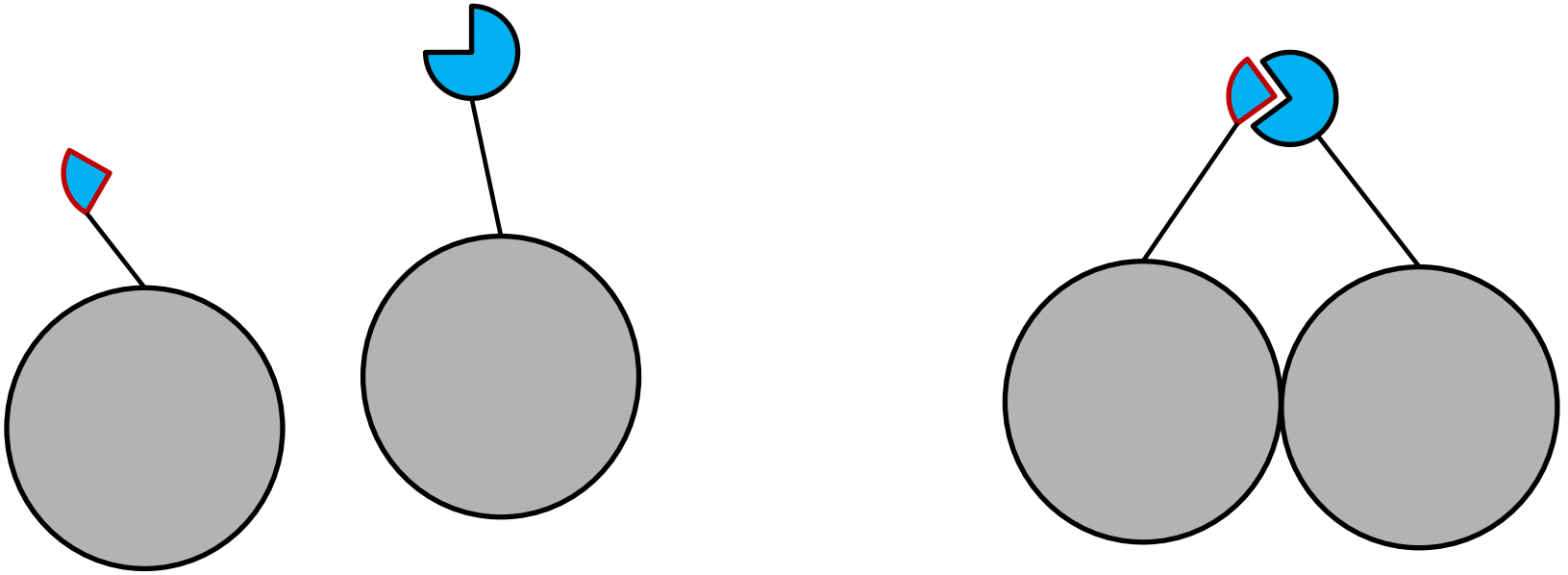
Approach 1: Transcriptional Control

- Used hybrid promoters from literature (in *E. coli)*
  - Two inducers required for transcription of reporter



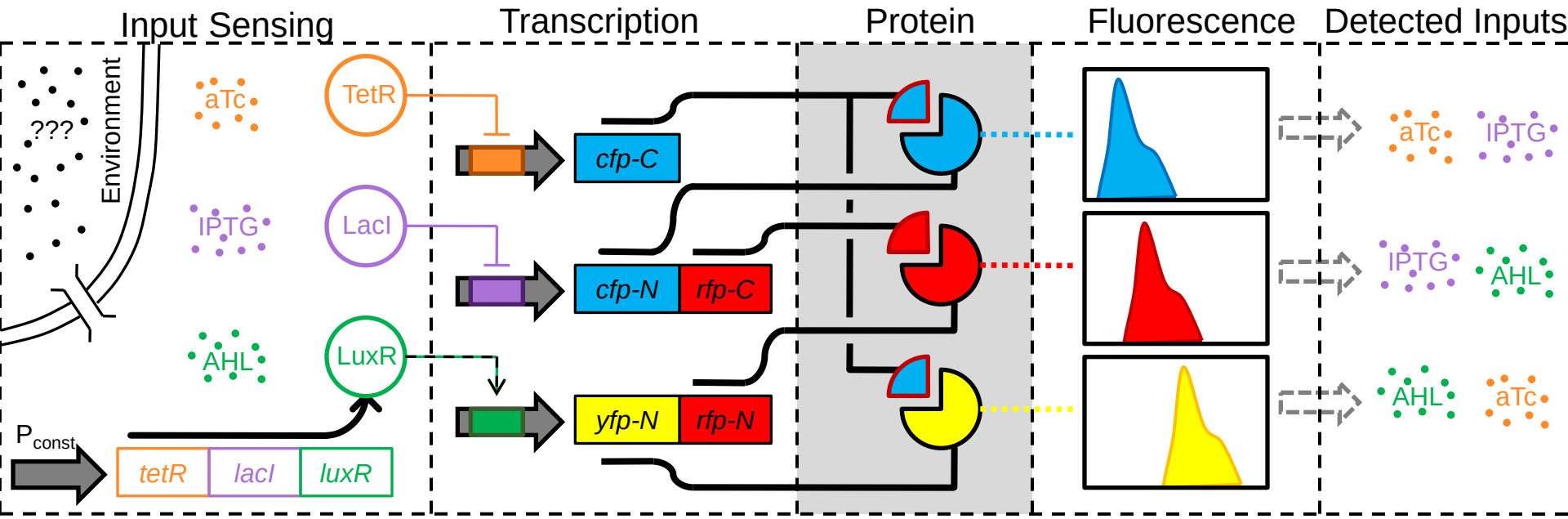
Approach 2: Fluorescence Complementation

- Proteins split into non-fluorescent halves
- Used in protein interaction experiments



Approach 2: Fluorescence Complementation

Fragment	CFP-N	CFP-C	RFP-N	RFP-C	YFP-N
CFP-N	N/C	BLUE	N/C	N/C	N/C
CFP-C	BLUE	N/C	N/C	N/C	YELLOW
RFP-N	N/C	N/C	N/C	RED	N/C
RFP-C	N/C	N/C	RED	N/C	N/C
YFP-N	N/C	YELLOW	N/C	N/C	N/C



Approach 3: Spectral Unmixing

- Fluorescent proteins usually detected using specific filters
- Instead, detect full spectrum:
  - Result must be a linear sum of the components

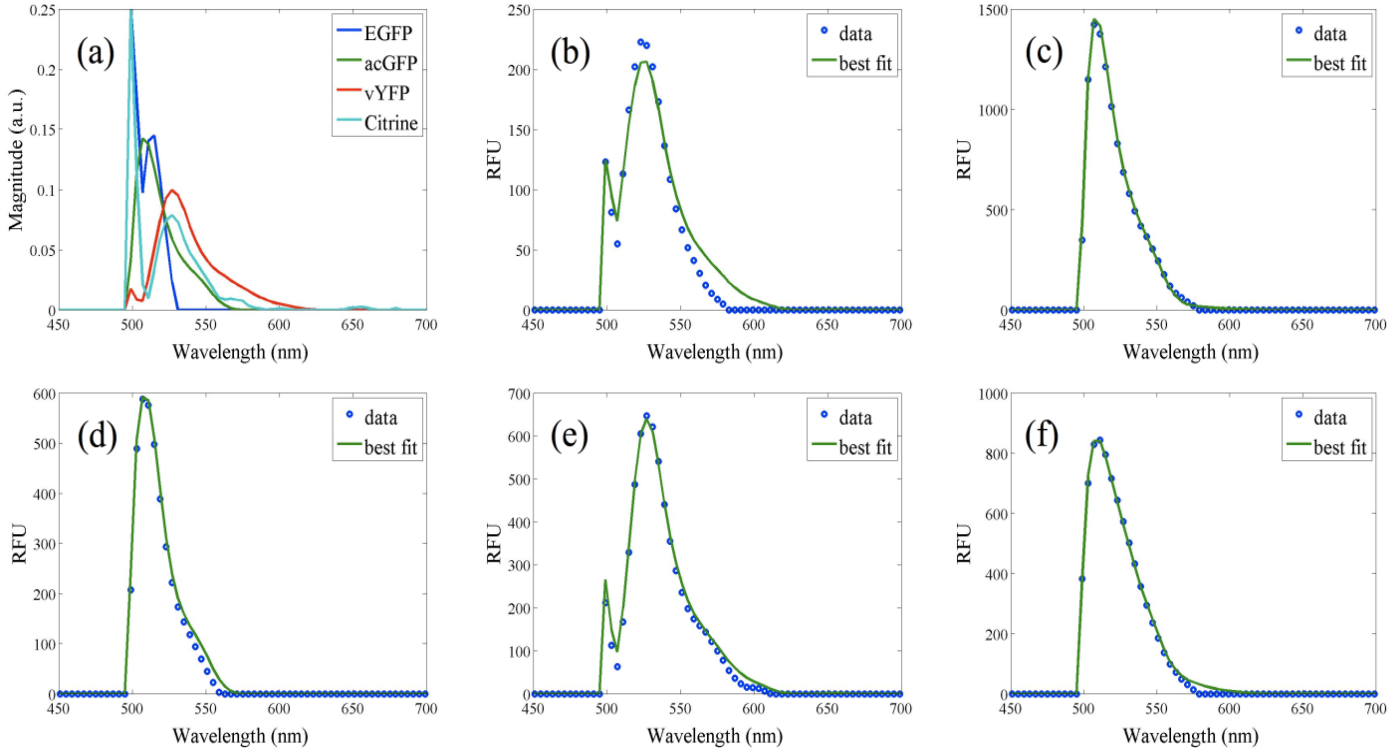
- Algorithm:

$$\begin{pmatrix} F_1 \\ F_2 \\ \vdots \\ F_m \end{pmatrix} = \begin{pmatrix} X_{11} & X_{12} & \cdots & X_{1n} \\ X_{21} & X_{22} & \cdots & X_{2n} \\ \vdots & \vdots & \ddots & \vdots \\ X_{m1} & X_{m2} & \cdots & X_{mn} \end{pmatrix} \begin{pmatrix} A_1 \\ A_2 \\ \vdots \\ A_n \end{pmatrix}$$

m=data points,
n=fluorphores

Solve for A with least squares fit to minimize F-**X**A

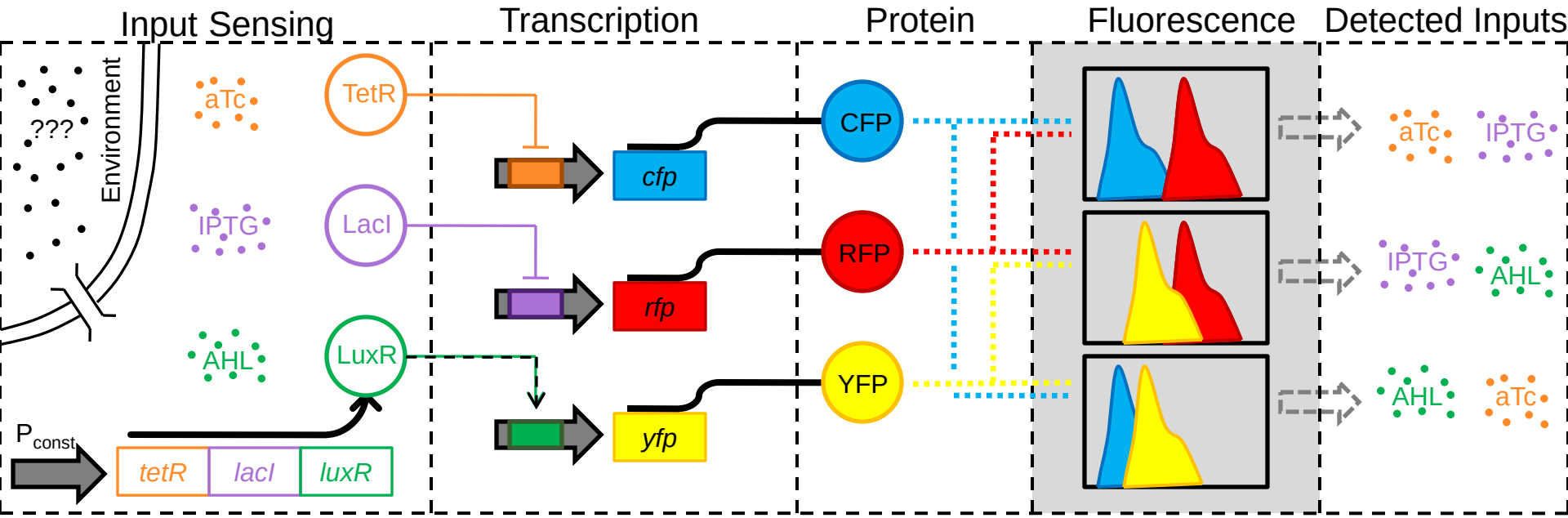
Approach 3: Preliminary Results



OD_{600}	Mix B		Mix C		Mix D		Mix E		Mix F	
	Meas.	Fit	Meas.	Fit	Meas.	Fit	Meas.	Fit	Meas.	Fit
acGFP	0	0.003	0.050	0.099	0.046	0.040	0	0.004	0.030	0.062
Citrine	0	0.006	0.070	0.039	0	0.002	0.081	0.014	0.027	0.017
EGFP	0.053	0.040	0	0	0.048	0.060	0	0	0.032	0.011
vYFP	0.048	0.029	0	0.006	0	0	0.082	0.100	0.027	0.037

Approach 3: Spectral Unmixing

- Cell simply transduces signal and logic is implemented in the analysis



Comparison Metrics

- Difficulty of implementation:
  - Design effort
  - Iterative tweaking
- Performance:
  - Accuracy
  - Strength/balance of output signals
  - Minimum detectable concentration
  - Response time
- Field use
  - Cost of manufacturing

Acknowledgements

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Questions?