FOGA I - IS THERE SUCH A THING AS A GENE? = FORMATTING THE GENOME FOR PROTEIN SYNTHESIS

Dilemma in choosing lecture style:

• Schematic presentation of all exceptions to classical view

• In-depth examination of basic model system & discussion of general principles

Lectures I & III - model system approach; lecture II - schematic approach
Diauxic Growth - a Cognitive Problem (Monod, 1942)

Glc 50% Lac

50% Glc, 50% Lac

67% Glc, 33% Lac
Historical Deconstruction of *lac*

1947

1961

1985
Cis-dominance of operator mutations

\[ \Rightarrow \text{operator does not encode protein} \]

- \( I^+ O^+ Z^- Y^+ / I^- O^+ Z^+ Y^- = \text{inducible for } Z \text{ and } Y \)

Constitutive \( I^- \) mutation is \textit{recessive} for both the active \( lacZ \) sequence in \textit{cis} and for the active \( lacY \) sequence in \textit{trans}

- \( I^+ O^+ Z^- Y^+ / I^+ O^c Z^+ Y^- = \text{constitutive for } Z \text{ but inducible for } Y \)

Constitutive \( o^c \) mutation is \textit{dominant} in \textit{cis} and conveys its constitutive phenotype to the adjacent active \( lacZ \) sequence, but the same mutation is \textit{recessive} in \textit{trans} and does not affect expression of the active \( lacY \) sequence on another DNA molecule
Cartoon of \textit{lac} operon induction

\url{http://www.blc.arizona.edu/marty/411/Modules/catrep.html}
Composite Nature of Operator, Promoter and Crp Sites

---lacI---| >------lacZ-------

O3 | CRP | P-35 | P-10 | O1 | O2

O1         CRP
AATTGTGAGCGGATAAACAAATT
AATTCTTACGCCACTACATT

AATTGTTATC
G
GATAACAATT

TGTTAGTTAGCTCACT
ACTGAACTAACACTACAAA
LacI Repressor Binding to \( lacO \) Sites and DNA Looping - Cooperativity Plus

Derepression by Allosteric Effect of Inducer Binding to Repressor
LacZ Converts Lactose to Allolactose Inducer
Cooperative Interactions in Crp
Stimulation of Transcription from $lacP$

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Role of Glucose in Regulating cAMP Synthesis

Boolean interactions in \textit{lac} operon regulation

Operations involving \textit{lac} operon products
\[\text{LacY} + \text{lactose(external)} \Rightarrow \text{lactose(internal)} \] \hspace{1cm} (1)
\[\text{LacZ} + \text{lactose} \Rightarrow \text{allolactose (minor product)} \] \hspace{1cm} (2)
\[\text{LacI} + \text{lacO} \Rightarrow \text{lacI-lacO} \text{ (repressor bound, \textit{lacP} inaccessible)} \] \hspace{1cm} (3)
\[\text{LacI} + \text{allolactose} \Rightarrow \text{LacI-allolactose (repressor unbound, \textit{lacP} accessible)} \] \hspace{1cm} (4)

Operations involving glucose transport components and adenylate cyclase
\[\text{IIA}^{\text{Glc-P}} + \text{glucose(external)} \Rightarrow \text{IIA}^{\text{Glc}} + \text{glucose-6-P(internal)} \] \hspace{1cm} (5)
\[\text{IIA}^{\text{Glc-P}} + \text{adenylate cyclase(inactive)} \Rightarrow \text{adenylate cyclase(active)} \] \hspace{1cm} (6)
\[\text{Adenylate cyclase(active)} + \text{ATP} \Rightarrow \text{cAMP} + \text{P~P} \] \hspace{1cm} (7)

Operations involving transcription factors
\[\text{Crp} + \text{cAMP} \Rightarrow \text{Crp-cAMP} \] \hspace{1cm} (8)
\[\text{Crp-cAMP} + \text{CAP} \Rightarrow \text{Crp-cAMP-CRP} \] \hspace{1cm} (9)
\[\text{RNA Pol} + \text{lacP} \Rightarrow \text{unstable complex} \] \hspace{1cm} (10)
\[\text{RNA Pol} + \text{lacP} + \text{Crp-cAMP-CRP} \Rightarrow \text{stable complex, initiate \textit{lacZYA} mRNA} \] \hspace{1cm} (11)

Partial computations
\[\text{No lactose} \Rightarrow \text{lacP inaccessible} \] \hspace{1cm} (3)
\[\text{Lactose} + \text{LacZ(basal)} + \text{LacY(basal)} \Rightarrow \text{lacP accessible} \] \hspace{1cm} (1, 2, 4)
\[\text{Glucose} \Rightarrow \text{low IIAGlc-P} \Rightarrow \text{low cAMP} \Rightarrow \text{unstable transcription complex} \] \hspace{1cm} (5, 6, 7, 10)
\[\text{No glucose} \Rightarrow \text{high IIAGlc-P} \Rightarrow \text{high cAMP} \Rightarrow \text{stable transcription complex} \] \hspace{1cm} (5, 6, 7, 8, 9, 11)

Overall computation = \textbf{IF} lactose present \textbf{AND} glucose not present
\AND \text{cell can synthesize active LacZ and LacY,}
\textbf{THEN} transcribe \textit{lacZYA} from \textit{lacP}
General Principles Deducible from *lac* Operon Case

* Functional requirement to interpret cognitive inputs
* Systemic (modular-interactive) nature of each protein molecule and DNA element
* Weak interactions, specific binding & cooperativity essential aspects of molecular computations in cells
* Layering of weak and “fuzzy” interactions provides overall precision to integrated cellular responses
* Allostery (the fact that binding of one ligand affects binding a distinct ligand) confers communication and processing capabilities on individual molecules
* Repetition in DNA and proteins ==> specific logical operations arise through combinations of basic circuit elements (complex regulatory regions in DNA, domain accretion & swapping in proteins)
* Cells use chemical symbols to represent physiological information (allolactose, phosphorylation of IIA\textsuperscript{Glc}, cAMP levels)
* No fundamental separation between control molecules and execution molecules ==> no “Cartesian” dualism in *E. coli* cell
* Participation of DNA directly in formation of repression and transcription nucleoprotein complexes ==> no Turing separation of “machine” and “tape” (also seen in computations that do not involve DNA)
Further Topics Pertinent to Continued Relevance of Classical Genetic Concepts

- Modular nature of proteins, protein machines and protein networks
- Complexity of transcriptional regulatory determinants
- Multiple proteins from a single DNA segment (overlapping coding regions, alternative splicing)
- Variation and *de novo* assembly of coding sequences
- Networked determination of phenotypic characters