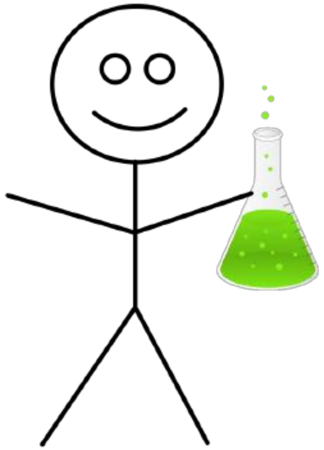
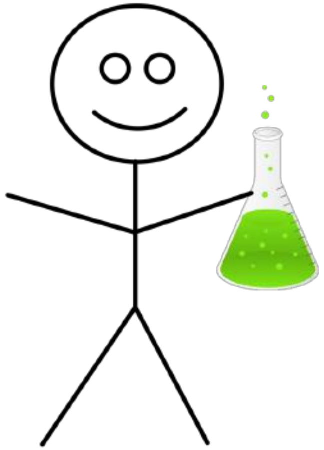
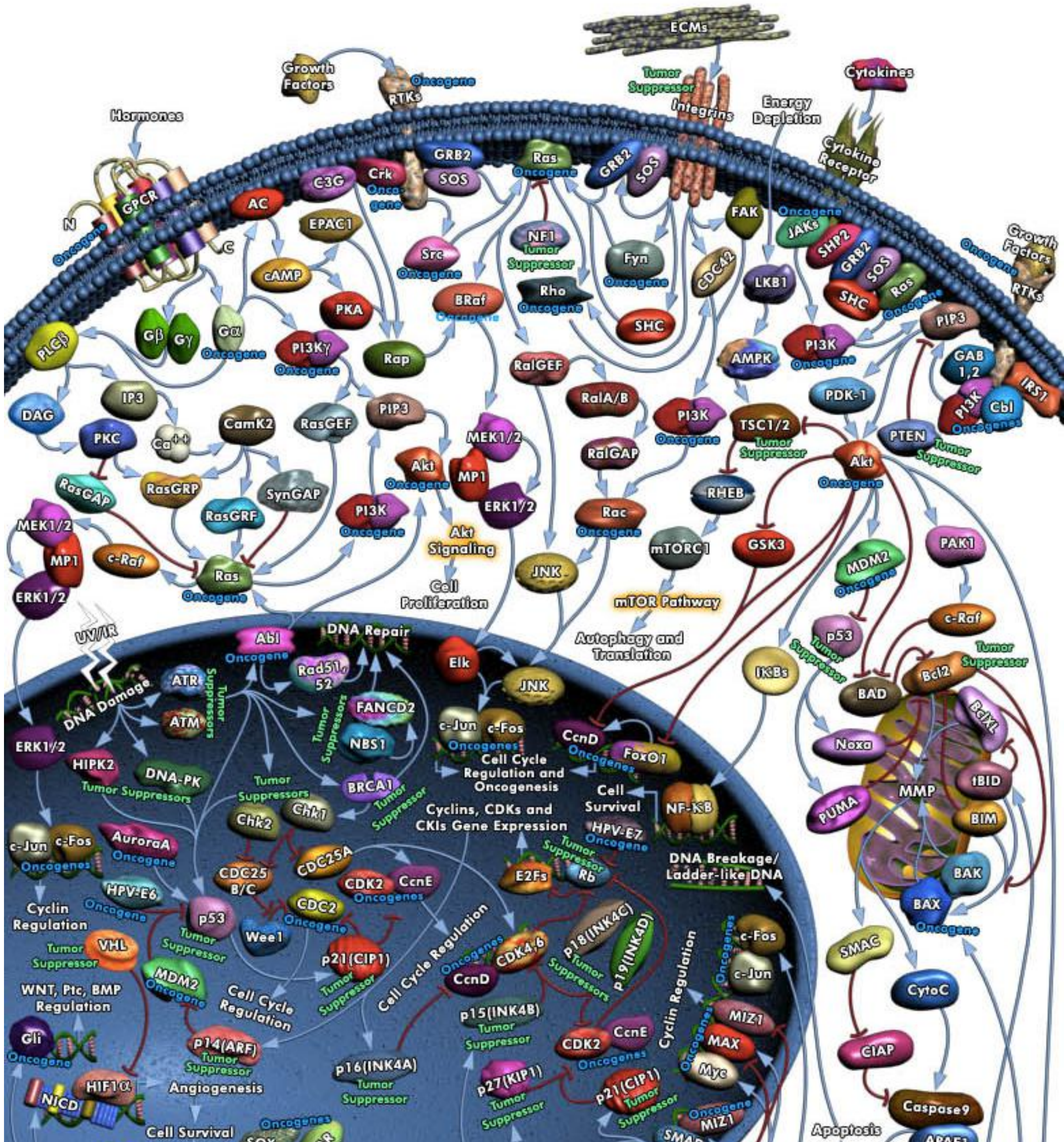
A Tool for Automated Inference of Executable Rule-Based Biological Models

Chelsea Voss, Jean Yang, Walter Fontana

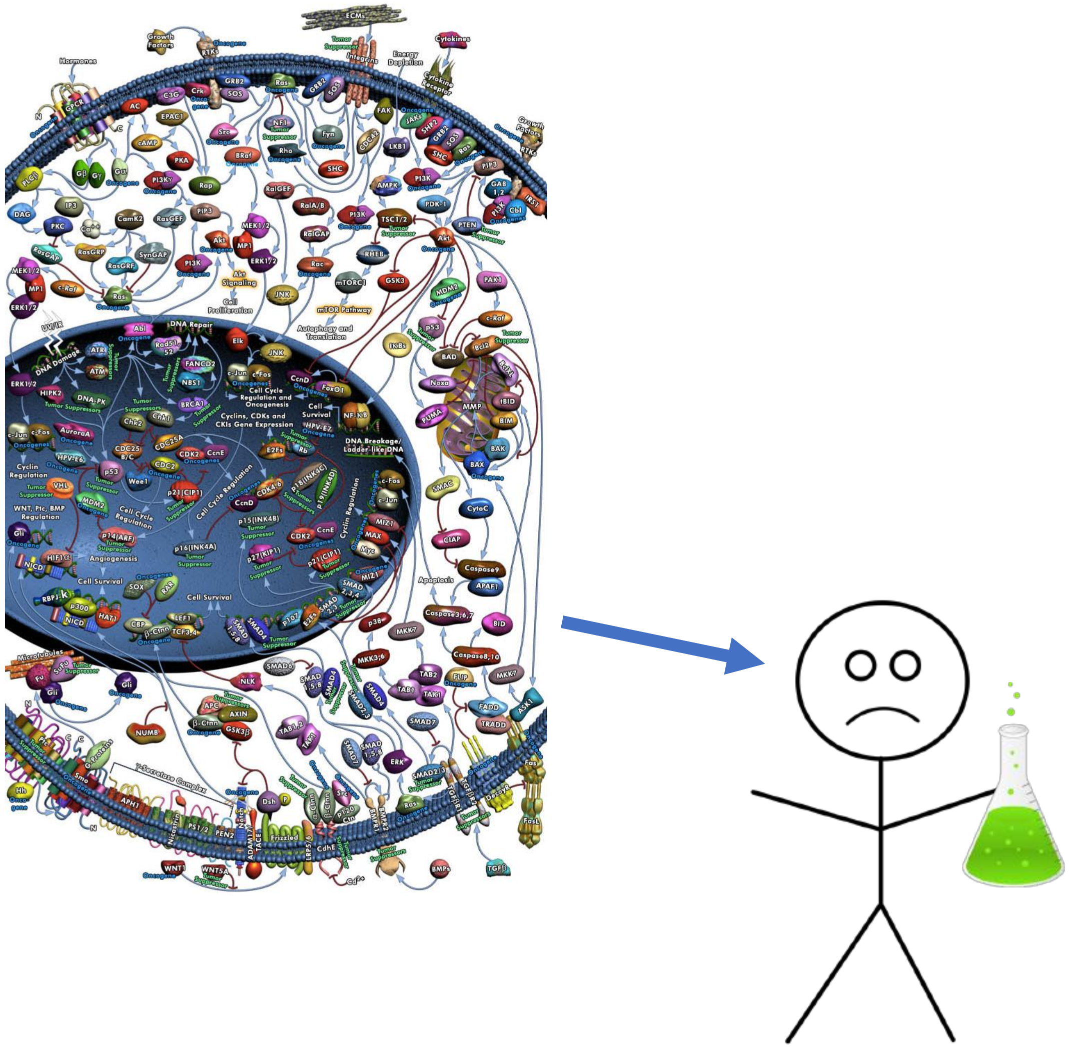
Static Analysis in Systems Biology, 2017

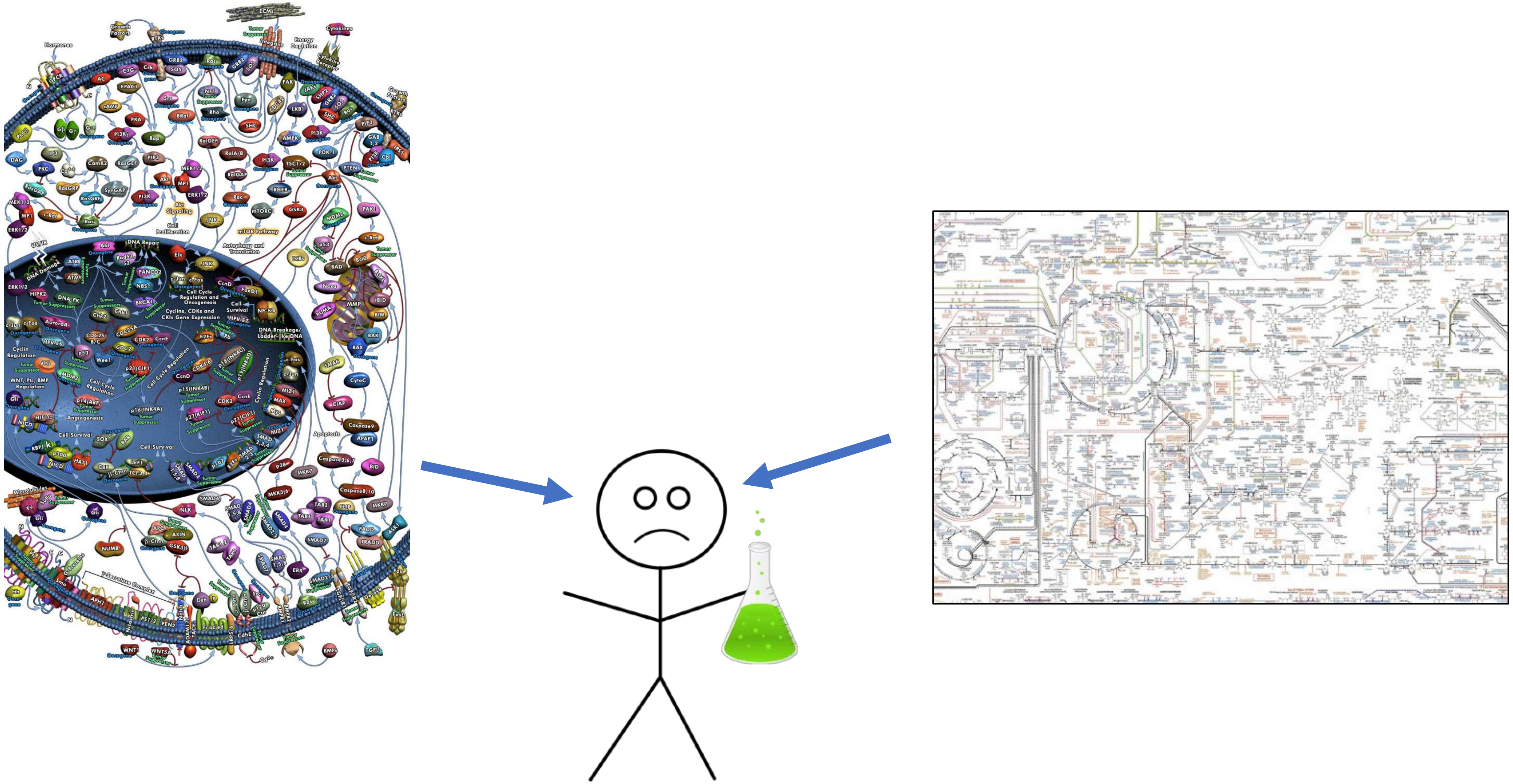


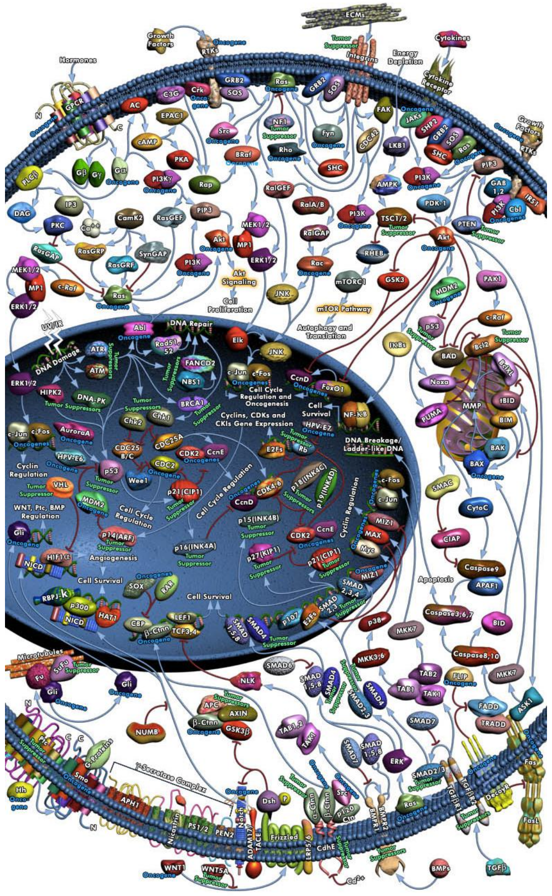




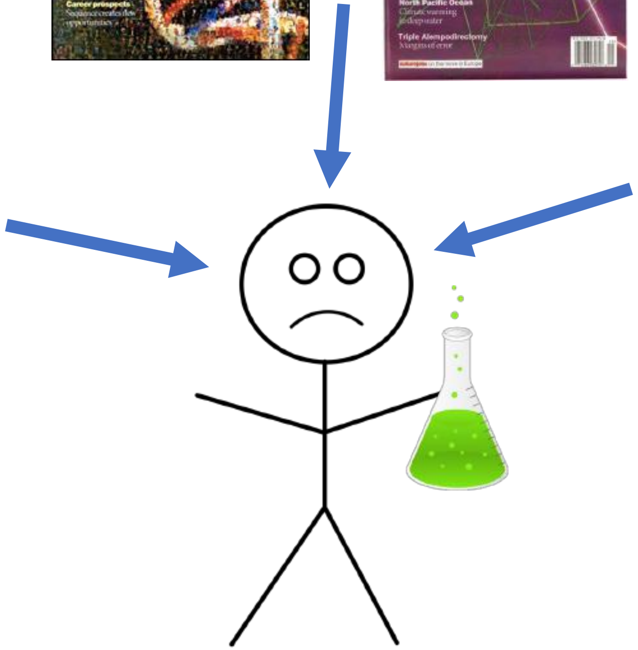
ECMs
Growth Factors
RTKs
Oncogene
Tumor Suppressor
Integrins
Cytokines
Energy Depletion
Cytokine Receptor
Hormones
GPCR
N
C
GRB2
SOS
Crk
C3G
Ras
AC
EPAC1
cAMP
PKA
Src
NF1
Fyn
Rho
SHC
FAK
CDC42
LKB1
JAKs
SHP2
PIP3
PLCβ
Gβ
Gγ
Gα
PI3Kγ
Rap
BRaf
RalGEF
AMPK
PI3K
GAB 1,2
Cbl
IRS1
PDK-1
PTEN
DAG
IP3
PKC
Ca++
CamK2
RasGEF
RalA/B
TSC1/2
Akt
RasGAP
RasGRP
SynGAP
RasGRF
MEK1/2
MP1
ERK1/2
RalGAP
RHEB
Rac
mTORC1
GSK3
MDM2
PAK1
c-Raf
Akt Signaling
Cell Proliferation
JNK
mTOR Pathway
Autophagy and Translation
UV/IR
DNA Damage
Abl
DNA Repair
Rad51, 52
ATR
ATM
Tumor Suppressors
FANCD2
NBS1
Elk
c-Jun
c-Fos
Oncogenes
CcnD
FoxO1
IKBs
p53
BAD
Bcl2
BclXL
HIPK2
DNA-PK
BRCA1
Chk1
Chk2
Cell Cycle Regulation and Oncogenesis
Cell Survival
NF-KB
Noxa
MMP
tBID
PUMA
BIM
Cyclins, CDKs and CKIs Gene Expression
HPV-E7
DNA Breakage/ Ladder-like DNA
AuroraA
CDC25A
CDC25 B/C
CDK2
CcnE
E2Fs
Rb
BAK
BAX
HPV-E6
Cyclin Regulation
VHL
Wee1
CDC2
p21(CIP1)
CDK4/6
p18(INK4C)
p19(INK4D)
SMAC
CytoC
MDM2
Cell Cycle Regulation
WNT, Ptc, BMP Regulation
Gli
p14(ARF)
p15(INK4B)
MIZ1
MAX
Myc
CIAP
HIF1α
NICD
Angiogenesis
p16(INK4A)
p27(KIP1)
Cyclin Regulation
Caspase9
Cell Survival
Apoptosis

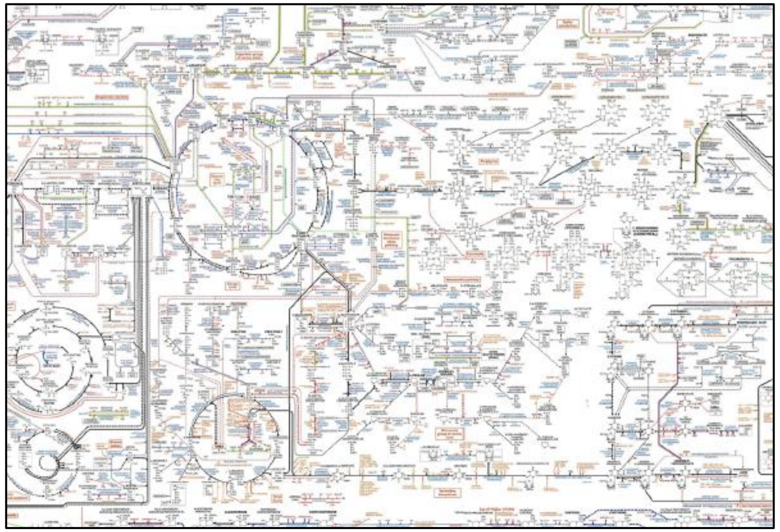



ECMs
Hormones
Growth Factors
Cytokines
DNA Repair
DNA Damage
Cell Survival
Apoptosis
Angiogenesis
mTOR Pathway
Autophagy and Translation
Cell Proliferation
DNA Breakage/ Ladder-like DNA
Cell Cycle Regulation and Oncogenesis
Cyclins, CDKs and CKIs Gene Expression

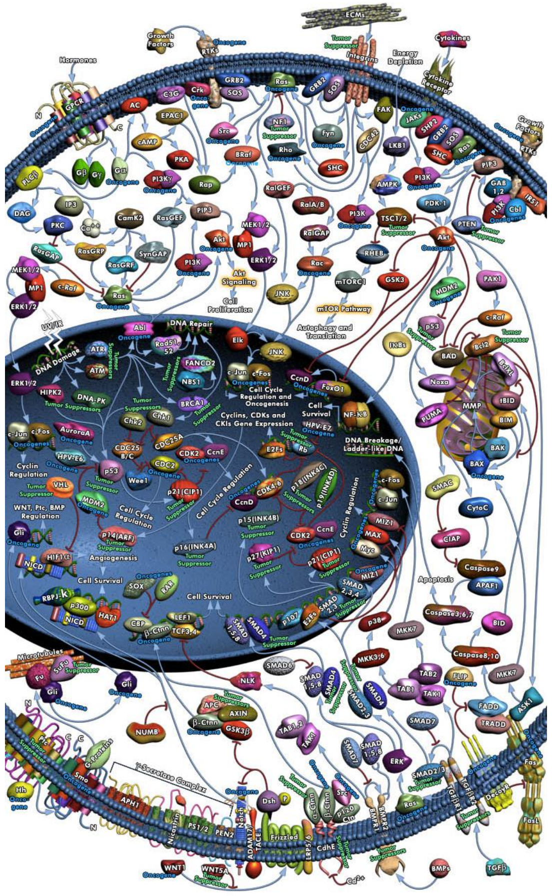


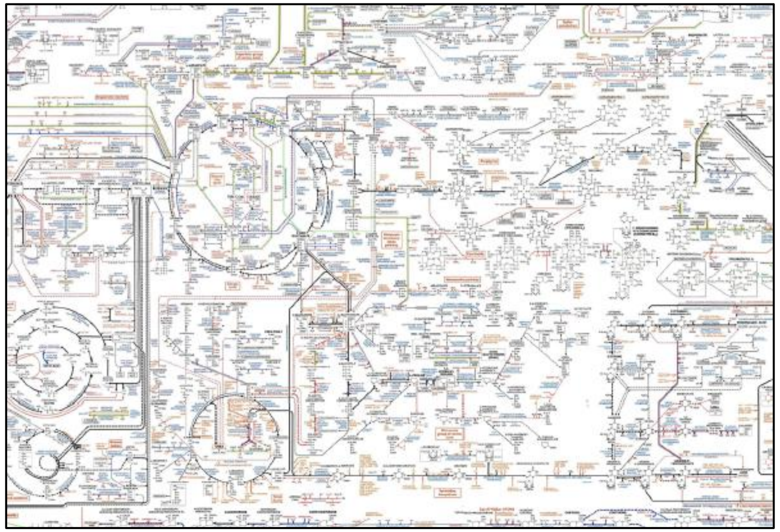

nature
Structure of the 30S ribosomal subunit
nature
the human genome
nature
Biochemical networks
Mapping the metabolic superhighways

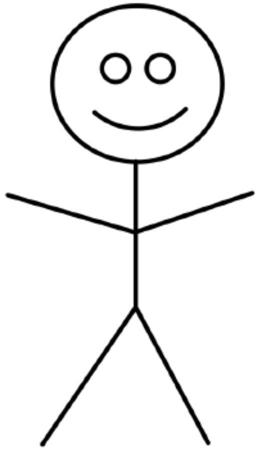




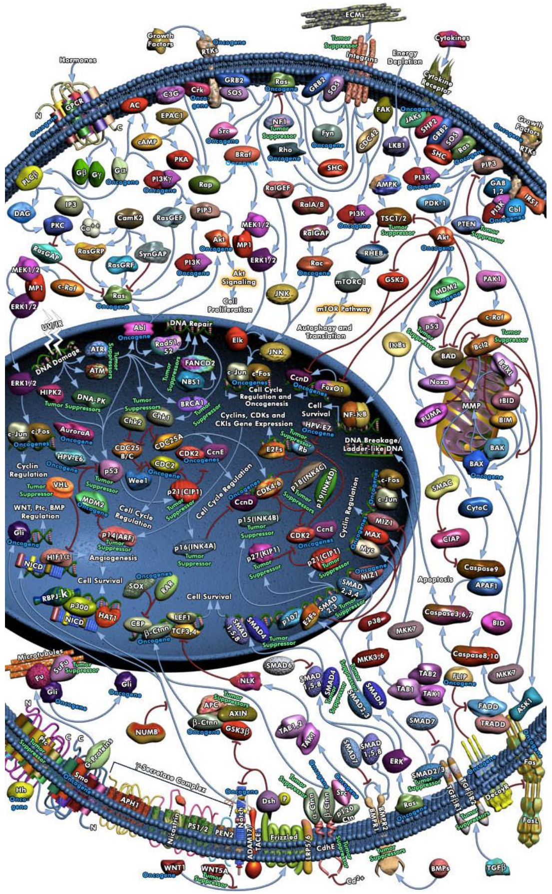
The need for biological models

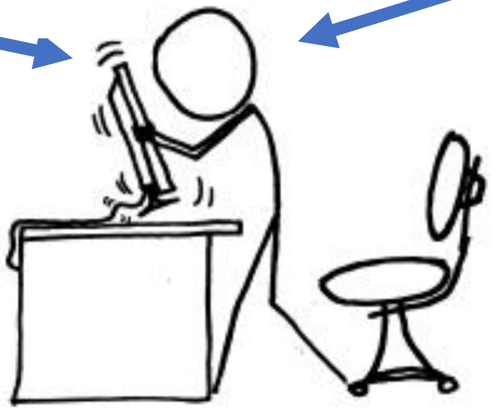




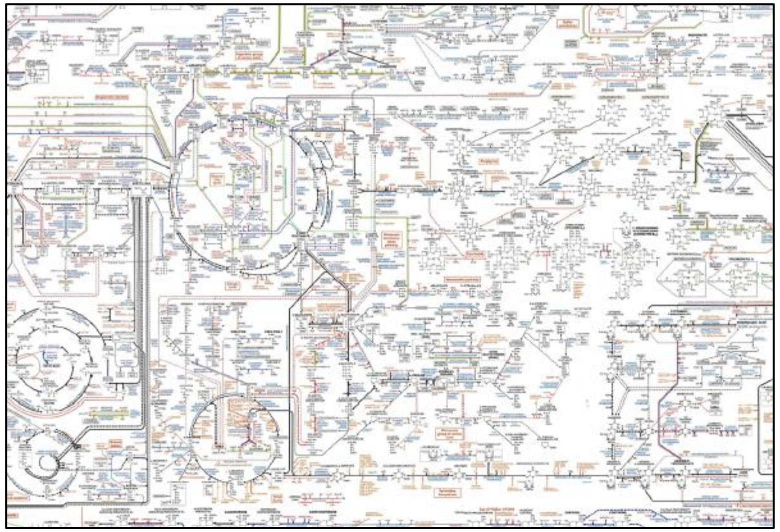


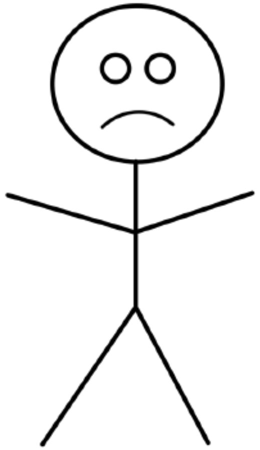
executable models for
"in silico" experimentation



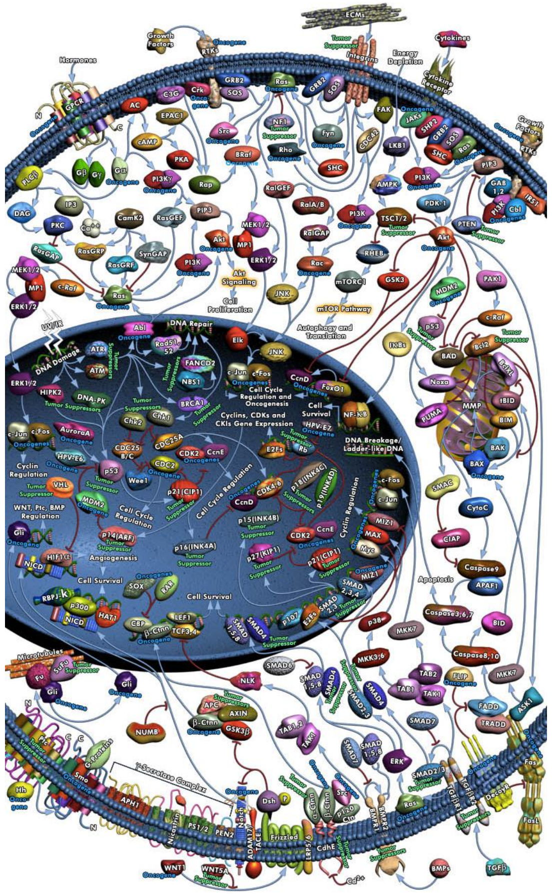


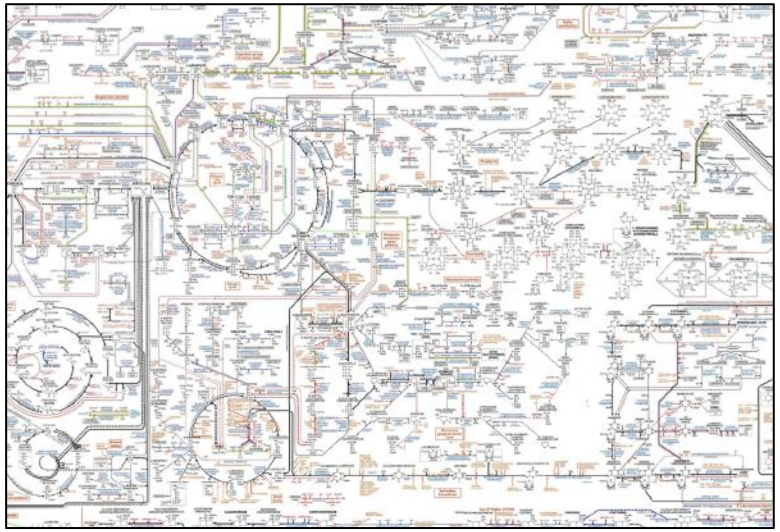
programming is hard



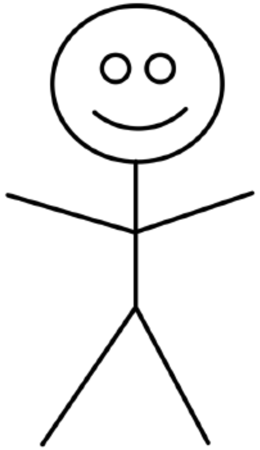


The need for *computer-generated* models





NLP



Some NLP output requires logical inference

Some NLP output requires logical inference

Executable model needs:

Mechanistic rules

Some NLP output requires logical inference

NLP produces:

Mechanistic rules

-

Non-mechanistic rules

-
-

Domain knowledge

-
-
-

Executable model needs:

Mechanistic rules

Some NLP output requires logical inference

NLP produces:

Mechanistic rules

-

Non-mechanistic rules

-
-

Domain knowledge

-
-
-

???

Executable model needs:

Mechanistic rules

Some NLP output requires logical inference

NLP produces:

Mechanistic rules

- MEK phosphorylates ERK1

Non-mechanistic rules

-
-

Domain knowledge

-
-
-

???

Executable model needs:

Mechanistic rules

Some NLP output requires logical inference

NLP produces:

Mechanistic rules

- MEK phosphorylates ERK1

Non-mechanistic rules

- MEK phosphorylates the ERK protein family
- Active ERK phosphorylates RSK

Domain knowledge

???

Executable model needs:

Mechanistic rules

Some NLP output requires logical inference

NLP produces:

Mechanistic rules

- MEK phosphorylates ERK1

Non-mechanistic rules

- MEK phosphorylates the ERK protein family
- Active ERK phosphorylates RSK

Domain knowledge

- When ERK1 is phosphorylated, it is active
- S151D-mutated ERK1 behaves as if always phosphorylated
- ERK1 and ERK2 are in the ERK protein family

???

Executable model needs:

Mechanistic rules

Some NLP output requires logical inference

NLP produces:

Mechanistic rules

- MEK phosphorylates ERK1

Non-mechanistic rules

- MEK phosphorylates the ERK protein family
- Active ERK phosphorylates RSK

Domain knowledge

- When ERK1 is phosphorylated, it is active
- S151D-mutated ERK1 behaves as if always phosphorylated
- ERK1 and ERK2 are in the ERK protein family

???

Executable model needs:

Mechanistic rules

- MEK phosphorylates ERK1
- MEK phosphorylates ERK2
- Phosphorylated ERK1 phosphorylates RSK
- Phosphorylated ERK2 phosphorylates RSK
- S151D-mutated ERK1 phosphorylates RSK

Mechanistic rules
Non-mechanistic rules
Domain knowledge

Mechanistic rules
Non-mechanistic rules
Domain knowledge

Models

Mechanistic rules
Non-mechanistic rules
Domain knowledge

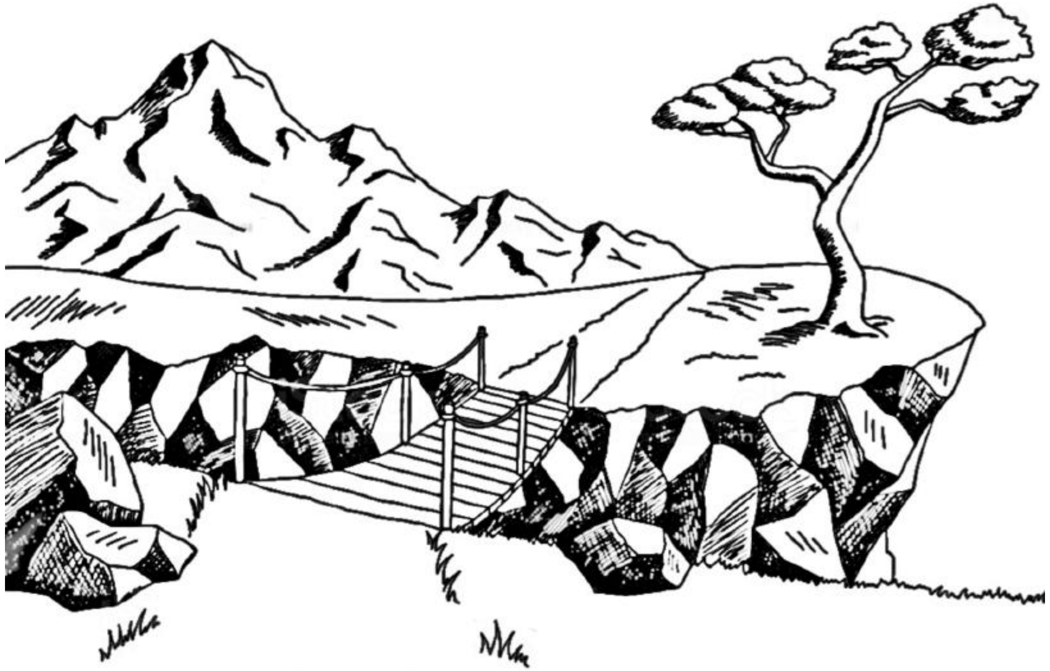
Space of possible *models*

Our contribution

Mechanistic rules
Non-mechanistic rules
Domain knowledge

→

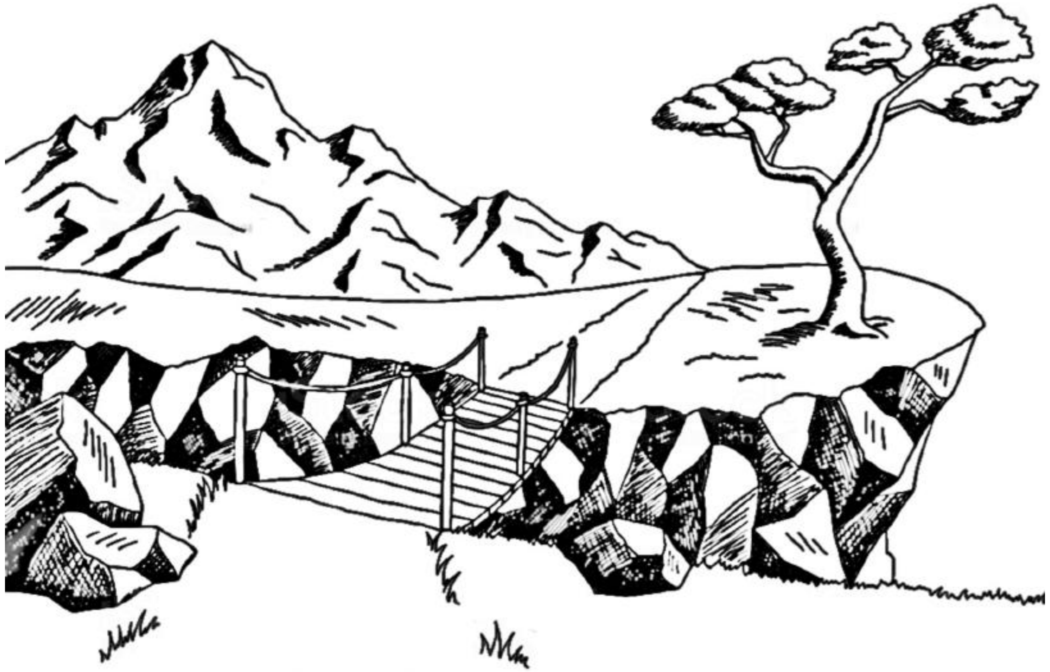
Space of possible models



Our contribution: how it works

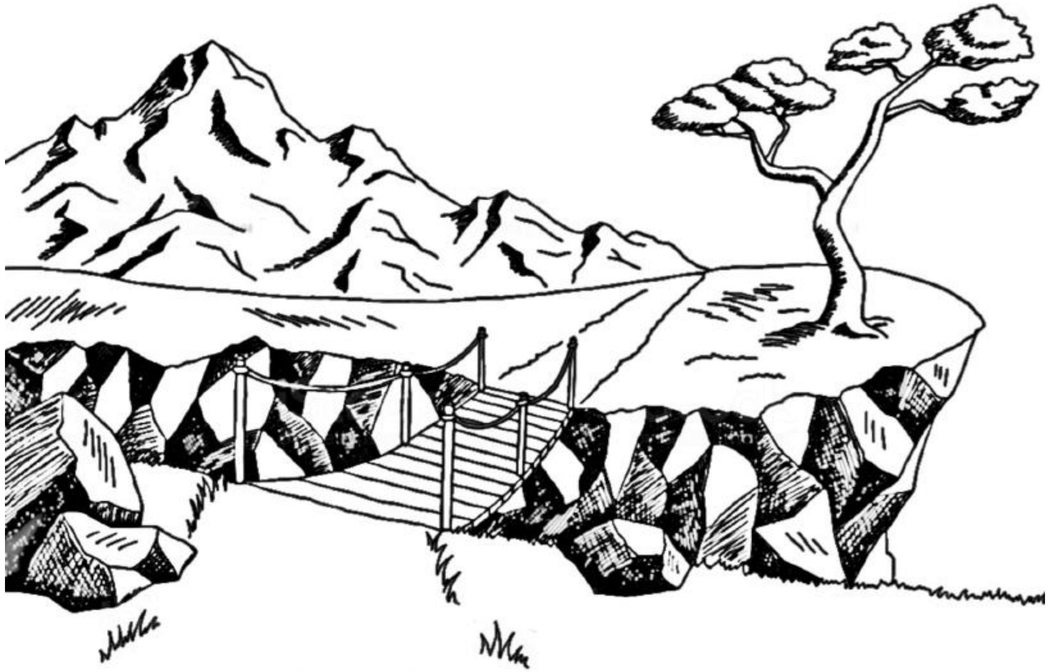
Mechanistic rules
Non-mechanistic rules
Domain knowledge

→ ***1: Predicates over models*** → *Space of possible models*



Our contribution: how it works

Mechanistic rules
Non-mechanistic rules
Domain knowledge
→ *1: Predicates over models* → ***2: Implement interpretation*** → *Space of possible models*



Our contribution: how it works

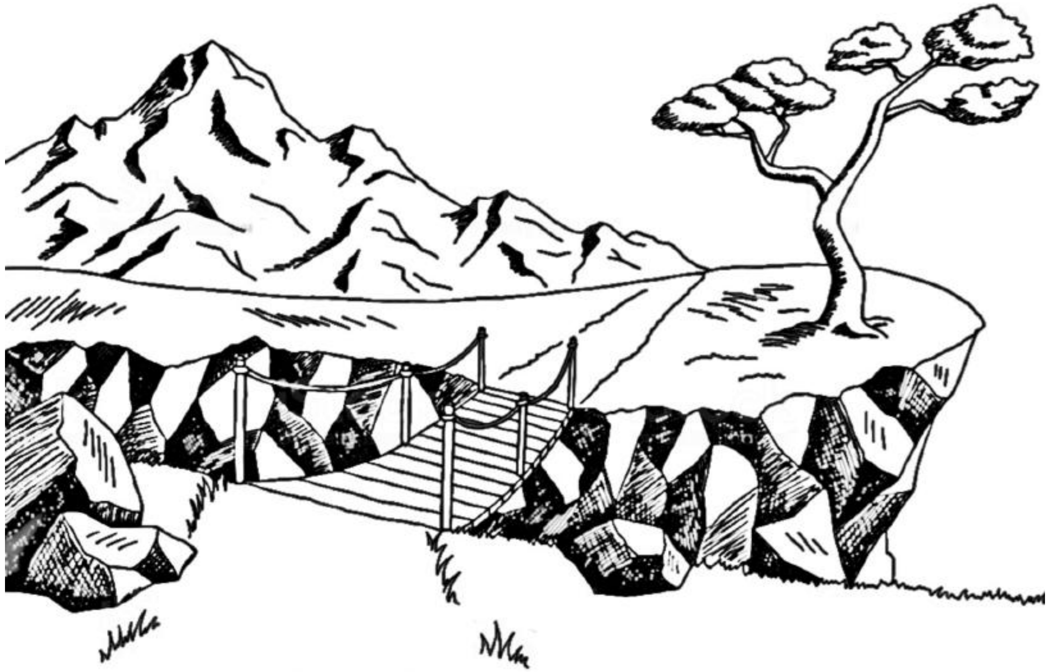
Mechanistic rules
Non-mechanistic rules
Domain knowledge

3: Create predicates →

1: Predicates over models

2: Implement interpretation →

Space of possible models



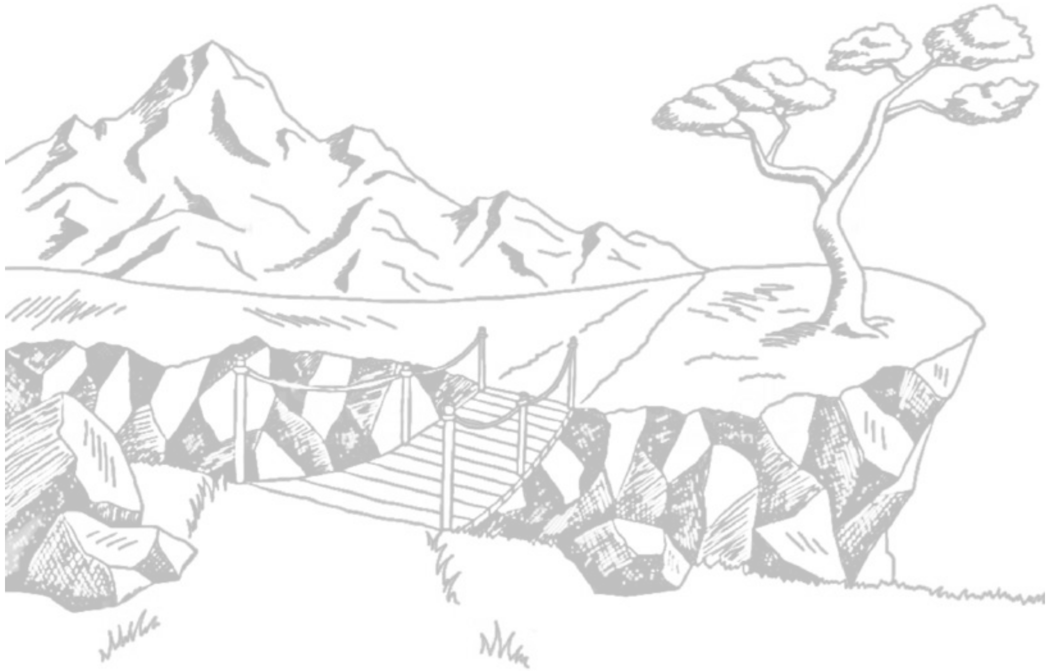
Mechanistic rules
Non-mechanistic rules
Domain knowledge

3: Create predicates

1: Predicates over models

2: Implement interpretation

Space of possible models



1: Predicates over models, in a logic

First, **choose a modeling language.**

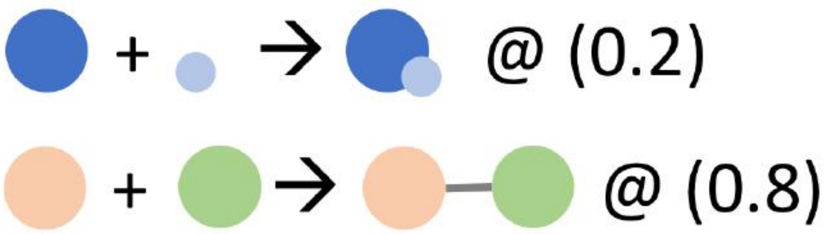
1: Predicates over models, in a logic

First, choose a modeling language: **Kappa.**

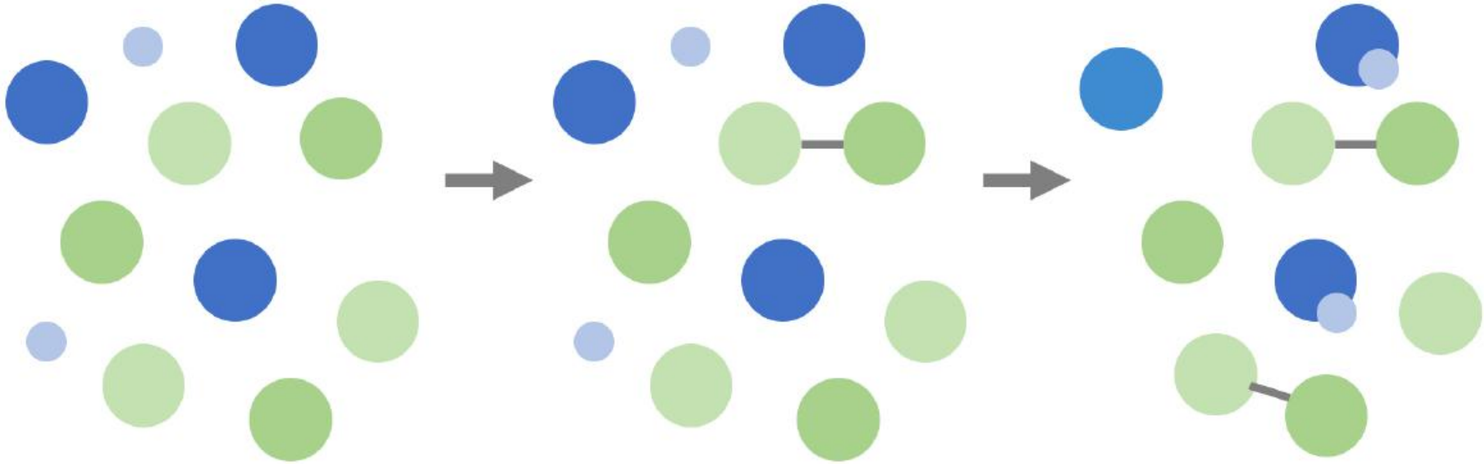
1: Predicates over models, in a logic

First, choose a modeling language: Kappa.

Kappa rules



Simulation of resulting system

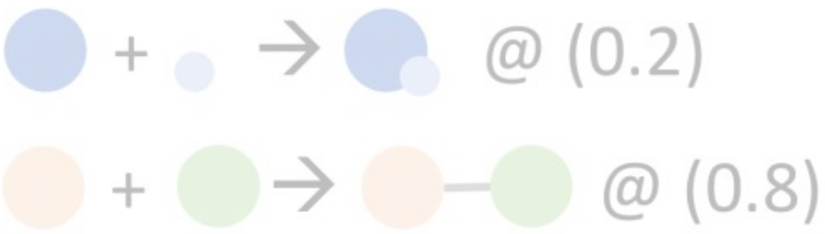


1: Predicates over models, in a logic

First, choose a modeling language: Kappa.

Why Kappa?

Kappa rules



Simulation of resulting system

1: Predicates over models, in a logic

First, choose a modeling language: Kappa.

Why Kappa?

Well-defined operational semantics
allow us to ***reason precisely.***

[Figure due to Danos et al. 2009: *Abstracting the ODE Semantics of Rule-Based Models: Exact and Automatic Model Reduction.*]

$$
\begin{array}{rcll}
E & ::= & \varepsilon \mid a\ ,E & \text{(expression)} \\
a & ::= & \emptyset \mid N(\sigma) & \text{(agent)} \\
N & ::= & A \in \mathcal{A} & \text{(agent name)} \\
\sigma & ::= & \varepsilon \mid s,\sigma & \text{(interface)} \\
s & ::= & n^{\lambda} & \text{(site)} \\
n & ::= & x \in \mathcal{S} & \text{(site name)} \\
\lambda & ::= & \epsilon \mid i \in \mathbb{N} \mid N@n \mid - & \text{(binding state)}
\end{array}
$$

(a) Syntax.

$$
\begin{gathered}
E\ ,A(\sigma,s,s',\sigma')\ ,E' \equiv E\ ,A(\sigma,s',s,\sigma')\ ,E' \\
E\ ,a\ ,a'\ ,E' \equiv E\ ,a'\ ,a\ ,E' \\
E \equiv E\ ,\emptyset \\
\frac{i,j \in \mathbb{N} \text{ and } i \text{ does not occur in } E}{E[i/j] \equiv E} \\
\frac{i \in \mathbb{N} \text{ and } i \text{ occurs only once in } E}{E[\epsilon/i] \equiv E}
\end{gathered}
$$

(b) Congruence.

$$
\begin{array}{rcl}
 & & \emptyset \models_{E_0} \emptyset \\
\lambda = \lambda_l & \Longrightarrow & n^{\lambda} \models^{N}_{E_0} n^{\lambda_l} \\
 & & n^{i} \models^{N}_{E_0} n^{-} \\
 & & n^{i} \models^{N}_{E_0} n^{\alpha_{E_0}(i,N@n)} \\
 & & \sigma \models^{N}_{E_0} \varepsilon \\
s \models^{N}_{E_0} s_l \wedge \sigma \models^{N}_{E_0} \sigma_l & \Longrightarrow & s,\sigma \models^{N}_{E_0} s_l,\sigma_l \\
\sigma \models^{N}_{E_0} \sigma_l & \Longrightarrow & N(\sigma) \models_{E_0} N(\sigma) \\
 & & E \models_{E_0} \varepsilon \\
a \models_{E_0} a_l \wedge E \models_{E_0} E_l & \Longrightarrow & a\ ,E \models_{E_0} a_l\ ,E_l
\end{array}
$$

(c) Matching.

$$
\begin{array}{rcl}
\emptyset[a_r] = a_r & & a_r[\emptyset] = \emptyset \\
\lambda_r \in \mathbb{N} \cup \{\epsilon\} \Longrightarrow n^{\lambda}[n^{\lambda_r}] & = & n^{\lambda_r} \\
n^{\lambda}[n^{-}] & = & n^{\lambda} \\
n^{\lambda}[n^{N@n}] & = & n^{\lambda} \\
\sigma[\varepsilon] & = & \sigma \\
(s,\sigma)[s_r,\sigma_r] & = & s[s_r],\sigma[\sigma_r] \\
N(\sigma)[N(\sigma_r)] & = & N(\sigma[\sigma_r]) \\
E[\varepsilon] & = & E \\
(a\ ,E)[a_r,E_r] & = & a[a_r]\ ,E[E_r]
\end{array}
$$

(d) Replacement.

$\alpha_E(i, A@n) = A'@n'$ where the site n' in the agent A' is the unique site distinct from the site n in A in the pattern E that is tagged with i.

(e) Look-up function.

$$
\frac{\begin{cases} E_0,E_1 \text{ are mixtures, } r = E_\ell, E_r \in R \\ E_0 \equiv E_0',\ E_0' \models_{E_0'} E_\ell,\ E_0'[E_r] \equiv E_1 \end{cases}}{E_0 \longrightarrow_r E_1}
$$

(f) Transitions.

Fig. 3. Syntax and operational semantics.

1: Predicates over models, in a logic

First, choose a modeling language: Kappa.

Second, **devise a logic for quantifying over models.**

1: Predicates over models, in a logic

First, choose a modeling language: Kappa.

Second, **devise a logic for quantifying over models.**

Datatypes:

- **Graphs** represent the state of a Kappa system
- **Rules** are sets of <graph, action> pairs
    - action rewrites graph, creates new graph
- **Models** are sets of rules

[Conversations with Husson & Krivine, 2015-2016]

1: Predicates over models, in a logic

First, choose a modeling language: Kappa.

Second, **devise a logic for quantifying over models.**

Datatypes:

- **Graphs** represent the state of a Kappa system
- **Rules** are sets of <graph, action> pairs
  - action rewrites graph, creates new graph
- **Models** are sets of rules

Predicates:

- **Atomic predicates** specify a set of rules
- **Predicates** specify a set of models

[Conversations with Husson & Krivine, 2015-2016]

Atomic predicates

```
class AtomicPredicate:

   Top
   Bottom
   Equal
   PreLabeled, PostLabeled
   PreUnlabeled, PostUnlabeled
   PreParent, PostParent
   PreLink, PostLink
   PreHas, PostHas
   Add, Rem
   DoLink, DoUnlink
   DoParent, DoUnparent
   Named
```

Atomic predicates

```
class AtomicPredicate:

    Top
    Bottom
    Equal
    PreLabeled, PostLabeled
    PreUnlabeled, PostUnlabeled
    PreParent, PostParent
    PreLink, PostLink
    PreHas, PostHas
    Add, Rem
    DoLink, DoUnlink
    DoParent, DoUnparent
    Named
```

Predicates

```
class Predicate:

    And
    Not
    Or
    Implies
    ModelHasRule
    ForAllRules
    Top
    Bottom
```

Example predicate syntax tree

```
a = Agent('a')
b = Agent('b')
p = And(
        ModelHasRule(lambda r:
            PregraphHas(r, a.bound(b))),
        ModelHasRule(lambda r:
            PostgraphHas(r, a.unbound(b))))
```

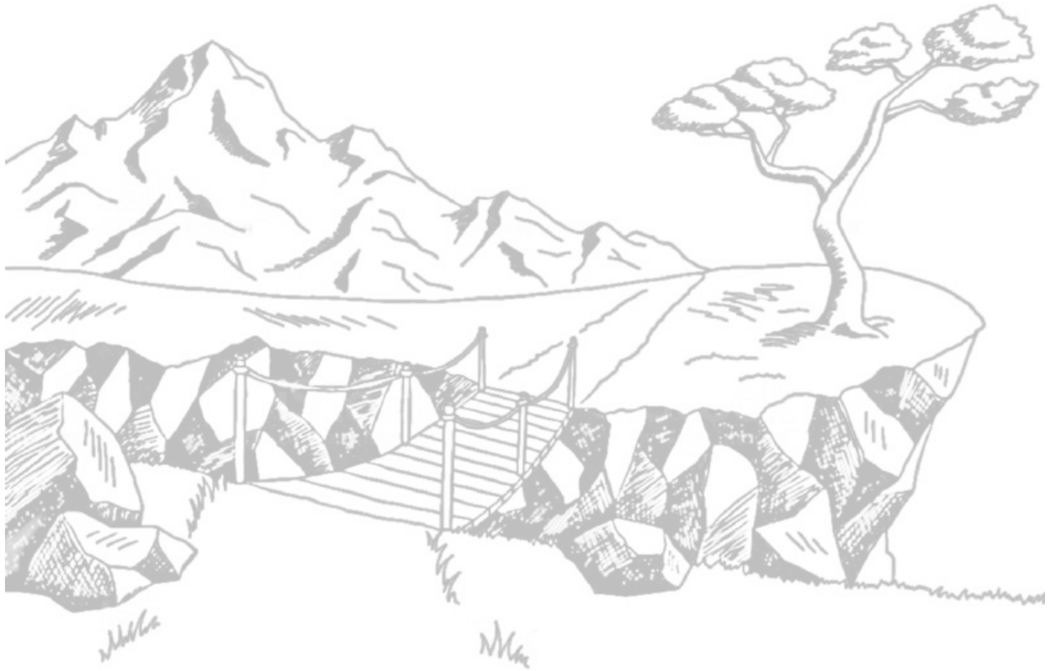
Mechanistic rules
Non-mechanistic rules
Domain knowledge

3: Create predicates

1: Predicates over models

2: Implement interpretation

Space of possible models



2: Implement interpretation of predicates

- Solving predicates in this logic is **reducible to first-order logic**

2: Implement interpretation of predicates

- Solving predicates in this logic is **reducible to first-order logic**
- Workhorse: **Z3 Theorem Prover**

2: Implement interpretation of predicates

- Solving predicates in this logic is **reducible to first-order logic**
- Workhorse: **Z3 Theorem Prover**

2: Implement interpretation of predicates

- Solving predicates in this logic is **reducible to first-order logic**
- Workhorse: **Z3 Theorem Prover**
  - Demo at http://rise4fun.com/z3

2: Implement interpretation of predicates

- Solving predicates in this logic is **reducible to first-order logic**
- Workhorse: **Z3 Theorem Prover**
  - Demo at http://rise4fun.com/z3
  - High-performance satisfiability solver

2: Implement interpretation of predicates

- Solving predicates in this logic is **reducible to first-order logic**
- Workhorse: **Z3 Theorem Prover**
  - Demo at http://rise4fun.com/z3
  - High-performance satisfiability solver
  - Wide variety of datatypes supported: arithmetic, fixed-size bit-vectors, extensional arrays, datatypes, uninterpreted functions, and quantifiers

2: Implement interpretation of predicates

- Solving predicates in this logic is **reducible to first-order logic**
- Workhorse: **Z3 Theorem Prover**
  - Demo at http://rise4fun.com/z3
  - High-performance satisfiability solver
  - Wide variety of datatypes supported: arithmetic, fixed-size bit-vectors, extensional arrays, datatypes, uninterpreted functions, and quantifiers

z3

Microsoft Research

Is this formula satisfiable?

```
(declare-fun x () Int)
(assert (>= 5 x))
(check-sat)
(get-model)
```

2: Implement interpretation of predicates

- Solving predicates in this logic is **reducible to first-order logic**
- Workhorse: **Z3 Theorem Prover**
  - Demo at http://rise4fun.com/z3
  - High-performance satisfiability solver
  - Wide variety of datatypes supported: arithmetic, fixed-size bit-vectors, extensional arrays, datatypes, uninterpreted functions, and quantifiers

Is this formula satisfiable?

```
(declare-fun x () Int)
(assert (>= 5 x))
(check-sat)
(get-model)
```

2: Implement interpretation of predicates

- Solving predicates in this logic is **reducible to first-order logic**
- Workhorse: **Z3 Theorem Prover**
  - Demo at http://rise4fun.com/z3
  - High-performance satisfiability solver
  - Wide variety of datatypes supported: arithmetic, fixed-size bit-vectors, extensional arrays, datatypes, uninterpreted functions, and quantifiers

Is this formula satisfiable?

```
(declare-fun x () Int)
(assert (>= 5 x))
(check-sat)
(get-model)
```

```
sat
```

2: Implement interpretation of predicates

- Solving predicates in this logic is **reducible to first-order logic**
- Workhorse: **Z3 Theorem Prover**
  - Demo at http://rise4fun.com/z3
  - High-performance satisfiability solver
  - Wide variety of datatypes supported: arithmetic, fixed-size bit-vectors, extensional arrays, datatypes, uninterpreted functions, and quantifiers

Z3

Microsoft Research

Is this formula satisfiable?

```
(declare-fun x () Int)
(assert (>= 5 x))
(check-sat)
(get-model)
```

```
sat
(model
  (define-fun x () Int
    5)
)
```

2: Implement interpretation of predicates

- Solving predicates in this logic is **reducible to first-order logic**
- Workhorse: **Z3 Theorem Prover**
- Using Z3 to interpret our predicates
  - Declare **Z3 datatypes** to represent
  - **Recursively build** Z3 predicates from our predicate classes
  - Use `(check-sat)` and `(get-model)`

2: Implement interpretation of predicates

- Solving predicates in this logic is **reducible to first-order logic**
- Workhorse: **Z3 Theorem Prover**
- Using Z3 to interpret our predicates
- Value added:
  - **Extract models**
  - Detect **inconsistencies** (if P is our facts so far and Q is a new predicate, and P/\Q is unsatisfiable, then Q is inconsistent with the existing facts)
  - Detect **redundancy** (if Q is a new fact, and P => Q, then Q is redundant)
  - Detect **ambiguity** (if model M satisfies predicate P, and P/\¬(model=M) is satisfiable, then P has multiple solutions)

Usage example: Inconsistency checking

```
>>> from syndra.engine import macros, predicate
```

Usage example: Inconsistency checking

```
>>> from syndra.engine import macros, predicate
>>> x = macros.directly_phosphorylates("MEK", "ERK")
```

Usage example: Inconsistency checking

```
>>> from syndra.engine import macros, predicate
>>> x = macros.directly_phosphorylates("MEK", "ERK")
>>> y = predicate.Not(x)
```

Usage example: Inconsistency checking

```
>>> from syndra.engine import macros, predicate
>>> x = macros.directly_phosphorylates("MEK", "ERK")
>>> y = predicate.Not(x)
>>> x_and_y = predicate.And(x, y)
```

Usage example: Inconsistency checking

```
>>> from syndra.engine import macros, predicate
>>> x = macros.directly_phosphorylates("MEK", "ERK")
>>> y = predicate.Not(x)
>>> x_and_y = predicate.And(x, y)
>>> print x_and_y.check_sat()
```

Usage example: Inconsistency checking

```
>>> from syndra.engine import macros, predicate
>>> x = macros.directly_phosphorylates("MEK", "ERK")
>>> y = predicate.Not(x)
>>> x_and_y = predicate.And(x, y)
>>> print x_and_y.check_sat()
False
```

Usage example: Redundancy checking

```
>>> from syndra.engine import macros, predicate
```

Usage example: Redundancy checking

```
>>> from syndra.engine import macros, predicate
>>> x = macros.directly_phosphorylates("MEK", "ERK")
```

Usage example: Redundancy checking

```
>>> from syndra.engine import macros, predicate
>>> x = macros.directly_phosphorylates("MEK", "ERK")
>>> y = macros.phosphorylated_is_active("ERK")
```

Usage example: Redundancy checking

```
>>> from syndra.engine import macros, predicate
>>> x = macros.directly_phosphorylates("MEK", "ERK")
>>> y = macros.phosphorylated_is_active("ERK")
>>> z = macros.directly_activates("MEK", "ERK")
```

Usage example: Redundancy checking

```
>>> from syndra.engine import macros, predicate
>>> x = macros.directly_phosphorylates("MEK", "ERK")
>>> y = macros.phosphorylated_is_active("ERK")
>>> z = macros.directly_activates("MEK", "ERK")
>>> x_and_y_imply_z =
        predicate.Implies(predicate.And(x, y), z)
```

Usage example: Redundancy checking

```
>>> from syndra.engine import macros, predicate
>>> x = macros.directly_phosphorylates("MEK", "ERK")
>>> y = macros.phosphorylated_is_active("ERK")
>>> z = macros.directly_activates("MEK", "ERK")
>>> x_and_y_imply_z =
        predicate.Implies(predicate.And(x, y), z)
>>> print x_and_y_imply_z.check_sat()
```

Usage example: Redundancy checking

```
>>> from syndra.engine import macros, predicate
>>> x = macros.directly_phosphorylates("MEK", "ERK")
>>> y = macros.phosphorylated_is_active("ERK")
>>> z = macros.directly_activates("MEK", "ERK")
>>> x_and_y_imply_z =
        predicate.Implies(predicate.And(x, y), z)
>>> print x_and_y_imply_z.check_sat()
True
```

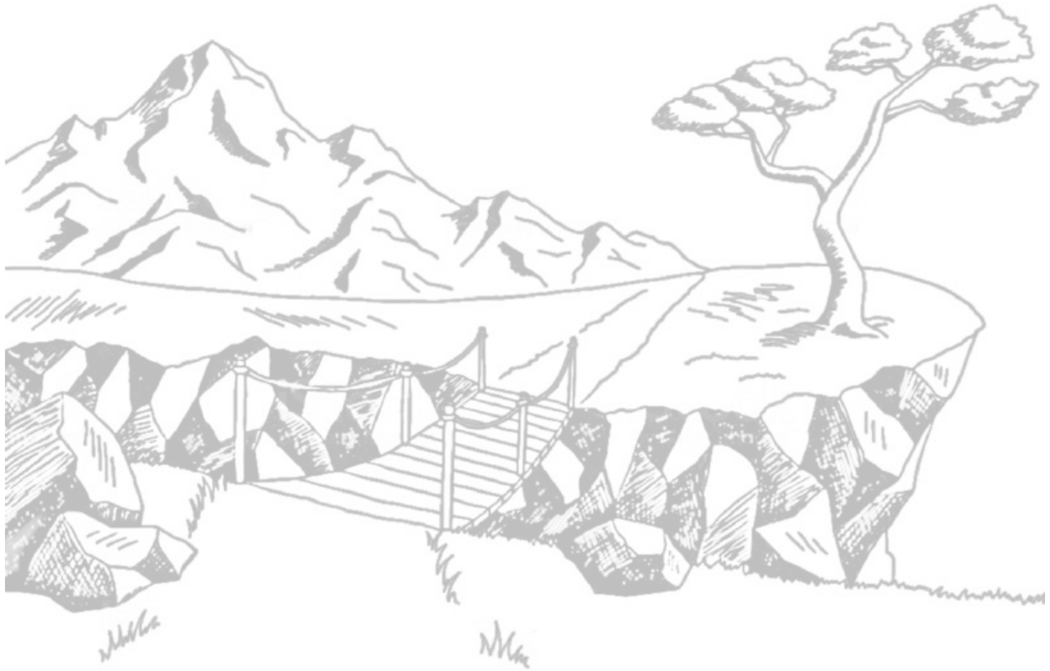
Mechanistic rules
Non-mechanistic rules
Domain knowledge

3: Create predicates →

1: Predicates over models

2: Implement interpretation →

Space of possible models



3: Tools for creating predicates

- **Macros**

3: Tools for creating predicates

- **Macros**

A phosphorylates B →

```
PreLabeled(A, phosphorylated) ∧
PreUnbound(A, B) ∧
PostLabeled(A, phosphorylated) ∧
PostBound(A, B)
```

3: Tools for creating predicates

- **Macros**

A phosphorylates B $\rightarrow$ `PreLabeled(A, phosphorylated)` $\wedge$ `PreUnbound(A, B)` $\wedge$ `PostLabeled(A, phosphorylated)` $\wedge$ `PostBound(A, B)`

- `directly_phosphorylates`
- `phosphorylated_is_active`
- `directly_activates`
- `negative_residue_behaves_as_if_phosphorylated`

3: Tools for creating predicates

- **Macros**
- **Interface with INDRA**

[INDRA: Gyori et al. *From word models to executable models of signaling networks using automated assembly*. 2017]

3: Tools for creating predicates

- **Macros**
- **Interface with INDRA**
  - `indra.statements.Phosphorylation`
  - `indra.statements.Activation`
  - `indra.statements.ActiveForm`

[INDRA: Gyori et al. *From word models to executable models of signaling networks using automated assembly*. 2017]

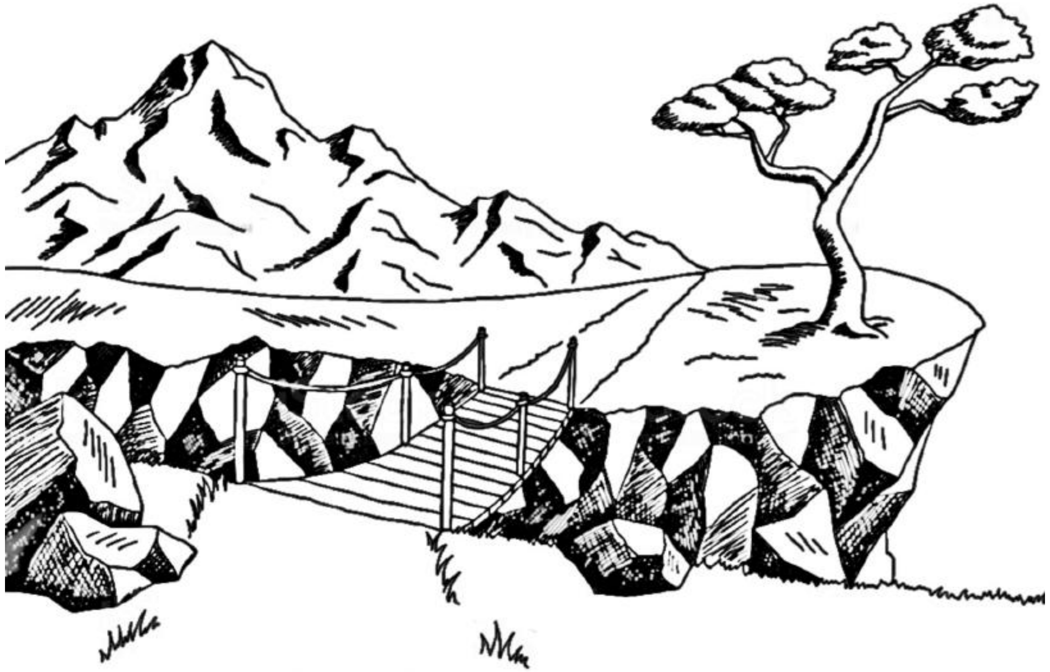
Mechanistic rules
Non-mechanistic rules
Domain knowledge

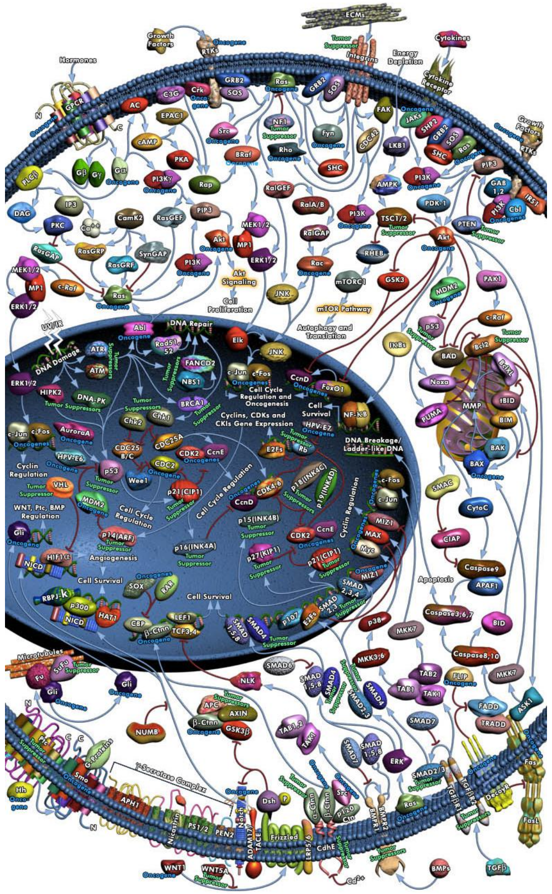
3: Create predicates →

1: Predicates over models

2: Implement interpretation →

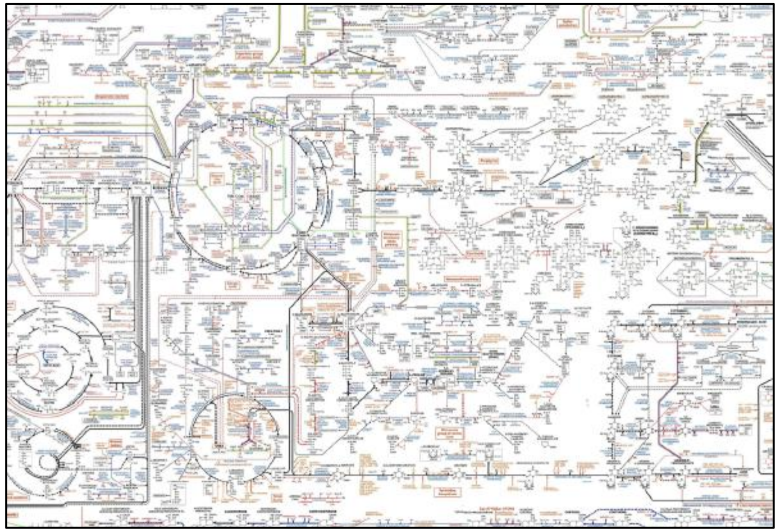
Space of possible models

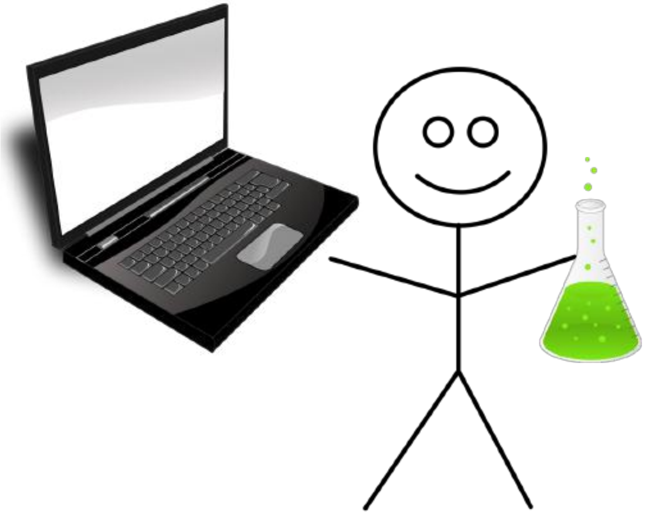


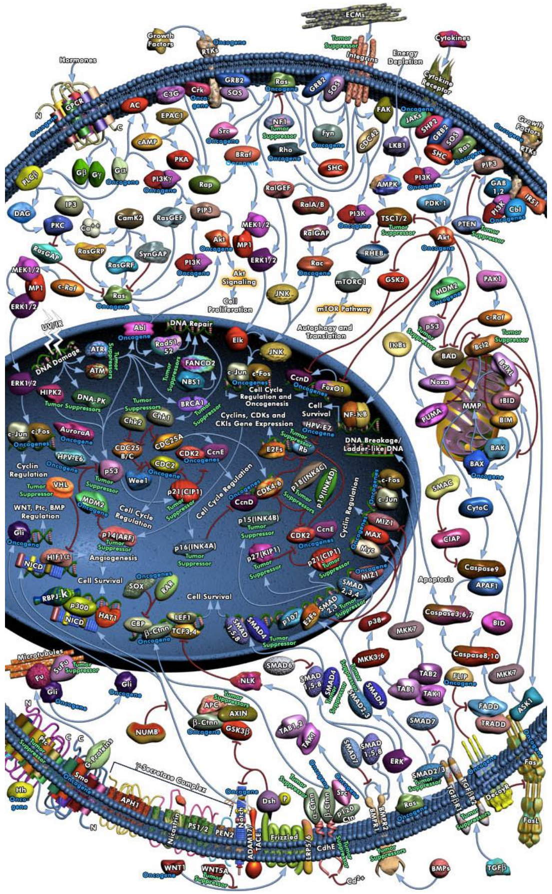



nature
Structure of the 30S ribosomal subunit
nature
the human genome
nature
Biochemical networks
Mapping the metabolic superhighways

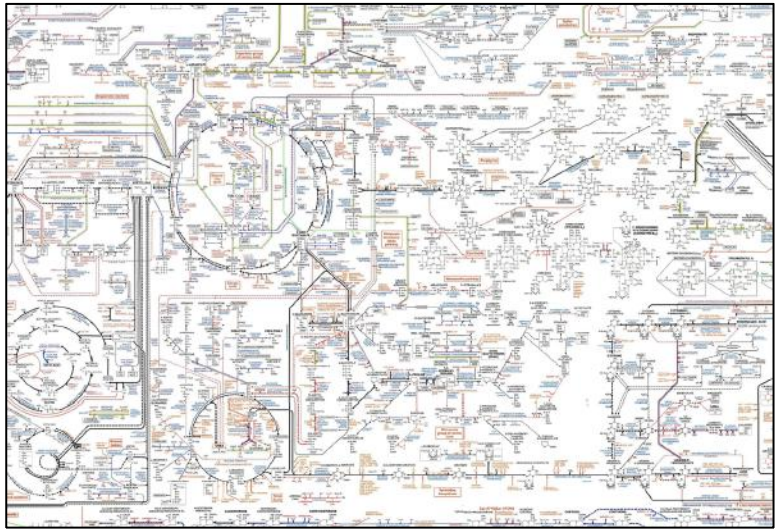
NLP





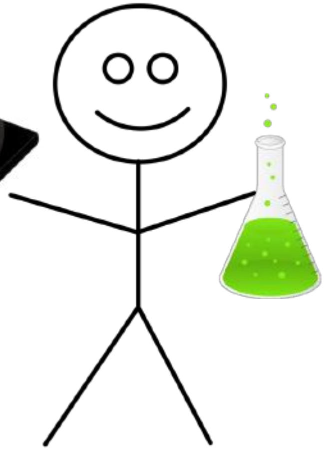


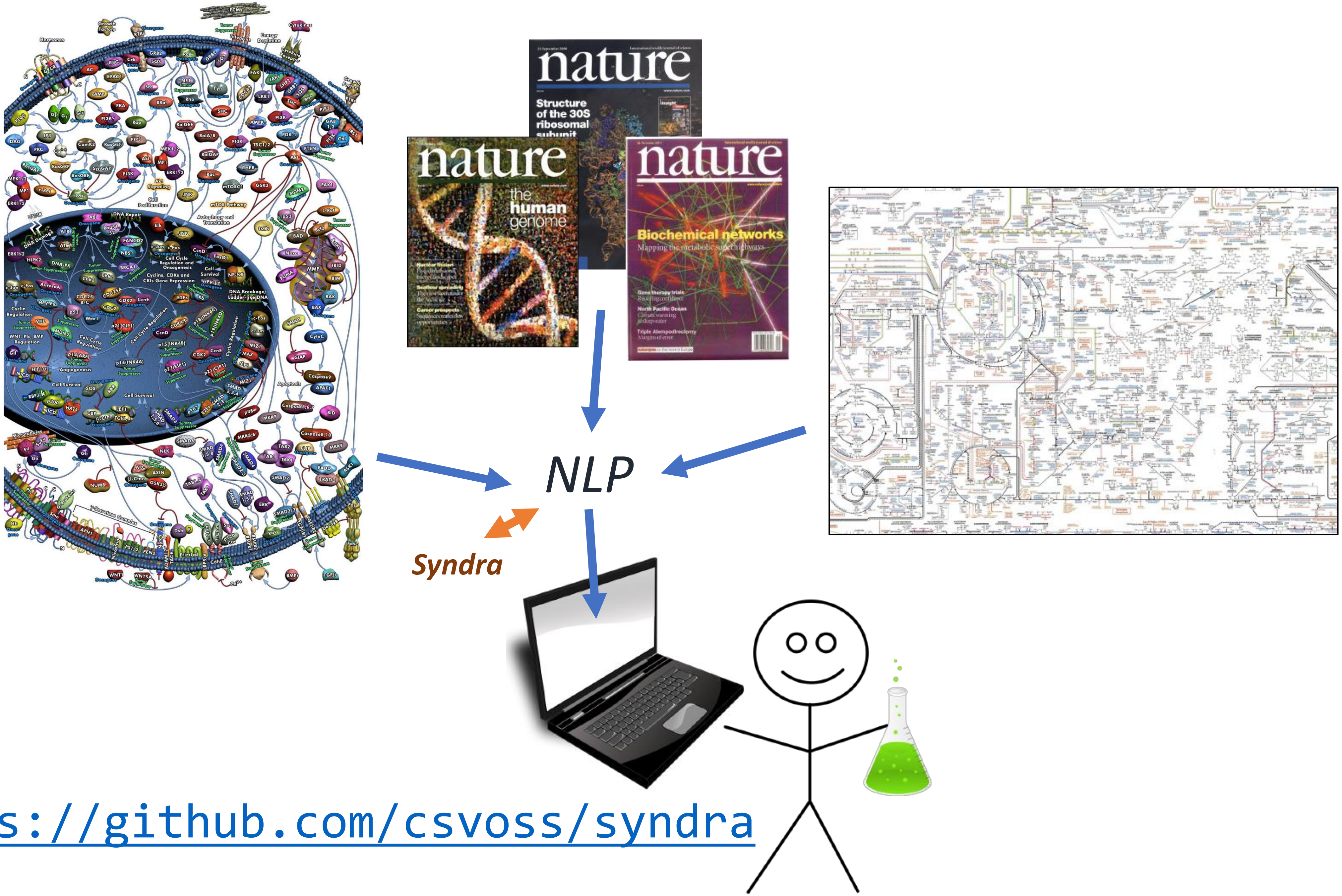

nature
Structure of the 30S ribosomal subunit
nature
the human genome
nature
Biochemical networks
Mapping the metabolic superhighways



NLP

Syndra





https://github.com/csvoss/syndra