幹細胞を用いた
化学物質リスク情報共有化コンソーシアム
scChemRISC Stem Cell-based Chemical Risk Information Sharing Consortium (scChemRISC)

"Challenges and issues of new safety evaluation by next-generation technologies" – Tokyo, Oct 16-17, 2019

Profiling the ToxCast library with a pluripotent human (H9) embryonic stem cell assay

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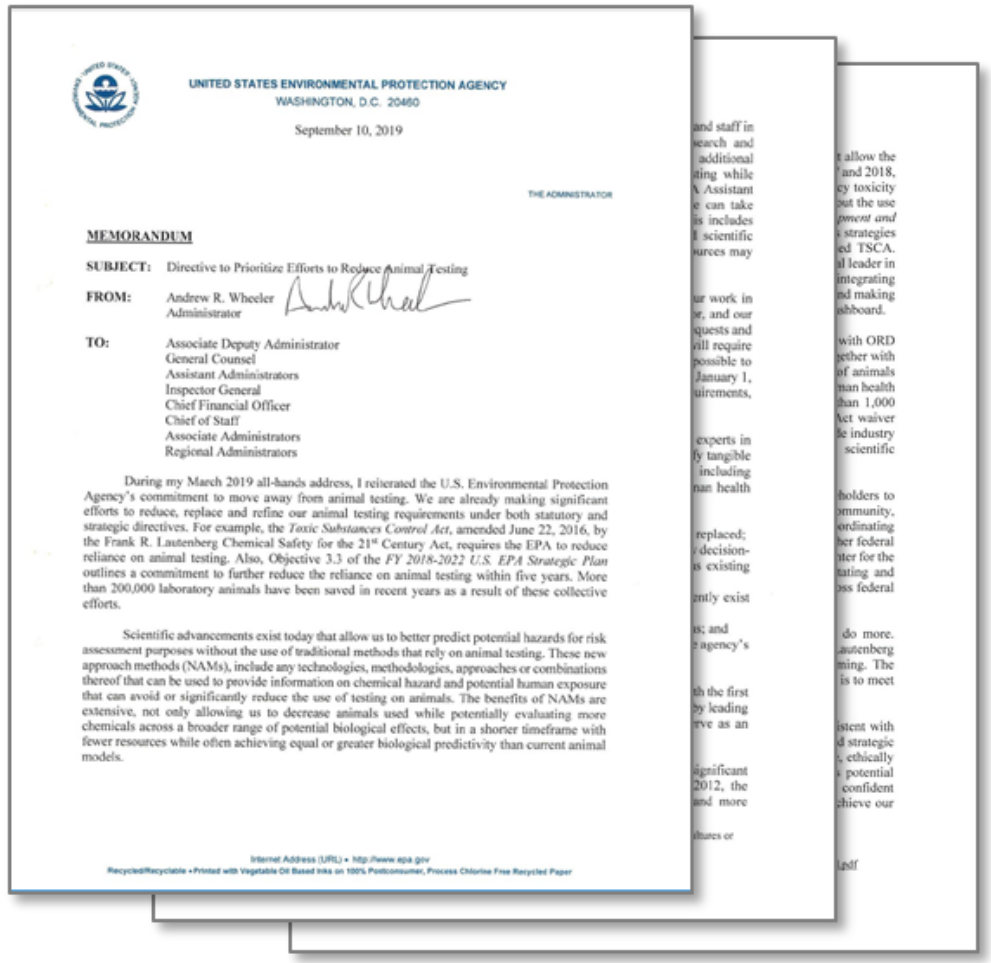
DISCLAIMER: The views expressed are those of the presenters and do not reflect Agency policy.

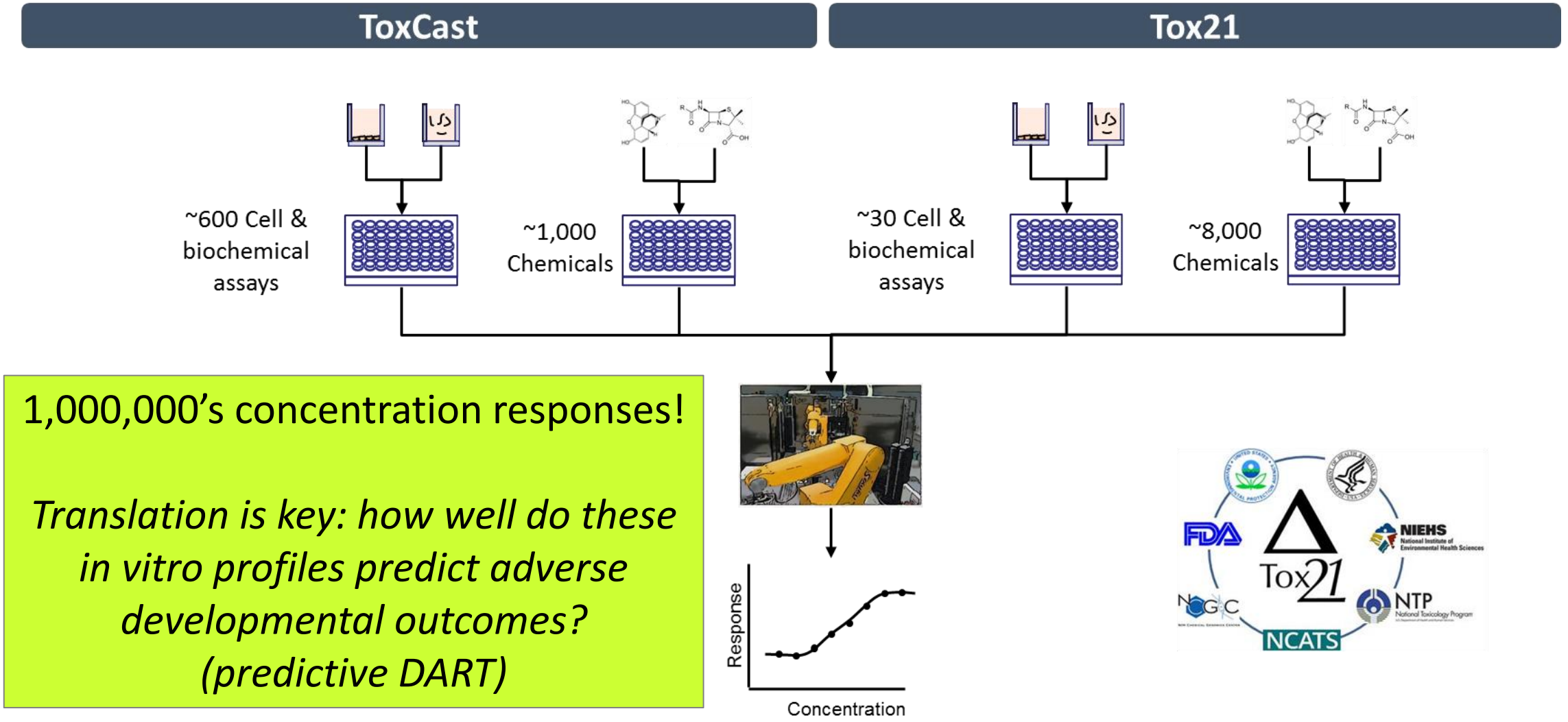
https://www.epa.gov/research/administrator-memo-prioritizing-efforts-reduce-animal-testing-september-10-2019



- EPA is evaluating <u>new approach methodologies</u> (NAMs) that can be used to quickly evaluate the human toxicity potential of chemicals with less reliance on animal testing.

- Sept 10 directive by Administrator Wheeler calls for reducing mammalian study requests 30% by 2025 and eliminating them by 2035.

Shifting toxicology to NAM-based approaches



1,000,000's concentration responses!

Translation is key: how well do these in vitro profiles predict adverse developmental outcomes? (predictive DART)

https://www.epa.gov/chemical-research/comptox-chemicals-dashboard

October is National Children's Health Month

Problem statement: *predictive DART*

- **Objective:** increase the diversity and relevance of assays in ToxCast that can be used to profile chemicals for potential adverse effects on human embryonic development.

- Chemical exposure to a pregnant woman has the potential to affect her unborn child, leading to adverse birth outcomes and/or risks to early child development.

- Traditional animal-based methods for assessing prenatal developmental toxicity (OECD TG 414) expose pregnant rats and/or rabbits during organogenesis and necropsy at term.

- Under reauthorized TSCA (2016) EPA must accelerate development of scientifically valid test methods to prioritize large numbers of chemicals with less reliance on animal testing.

devTOX^{qP} assay: *Stemina Biomarker Discovery, EPA contract EP-D-13-055*

Birth Defects Research Part B | Developmental and Reproductive Toxicology

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Original Article

Establishment and Assessment of a New Human Embryonic Stem Cell-Based Biomarker Assay for Developmental Toxicity Screening

Jessica A. Palmer ✉, Alan M. Smith, Laura A. Egnash, Kevin R. Conard, Paul R. West, Robert E. Burrier, Elizabeth L.R. Donley, Fred R. Kirchner

First published: August 2013 Full publication history

DOI: 10.1002/bdrb.21078 View/save citation

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Pages 343–363

Pluripotent H9 human embryonic stem cell metabolomics assay that "… identified the potential developmental toxicants in the test set with 77% accuracy (57% sensitivity, 100% specificity)."

Palmer et al. 2013

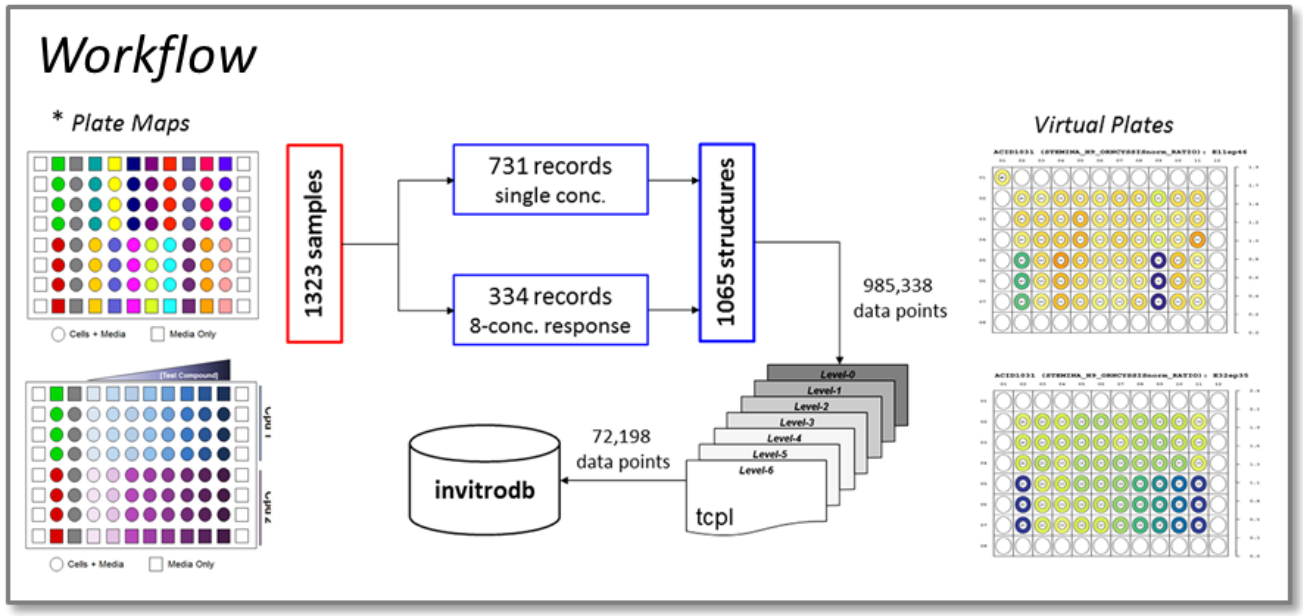
Ornithine release
urea cycle, polyamine & pyrimidine synthesis.

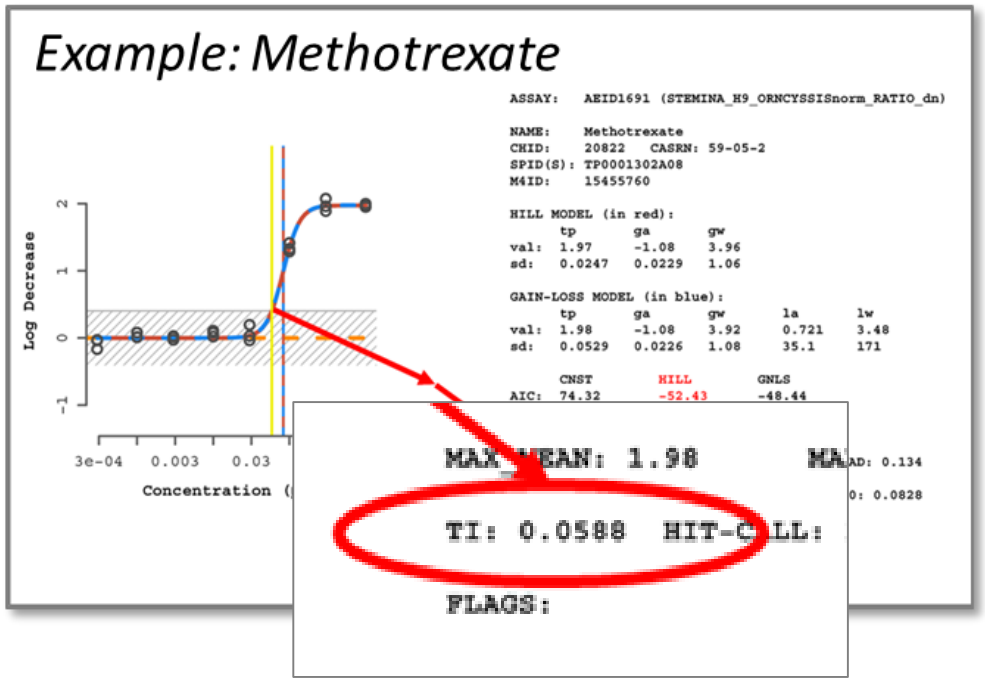
TI = ORN/CYSS

Cystine utilization
glutathione synthesis, redox cycling.

ToxCast_STM workflow: *ToxCast chemical library (1065 chemicals in Phase I/II)*

- pluripotent human embryonic stem cells (H9 line) exposed for 3-days
- concentration response on 334 chemicals; single concentration screen on 731 inactives
- (additional testing underway on 307 chemicals)
- data processed through the ToxCast pipeline (tcpl, level 6)
- readout is concentration that induces a critical drop in the biomarker (ORN/CYSS $\leq$ 0.76)

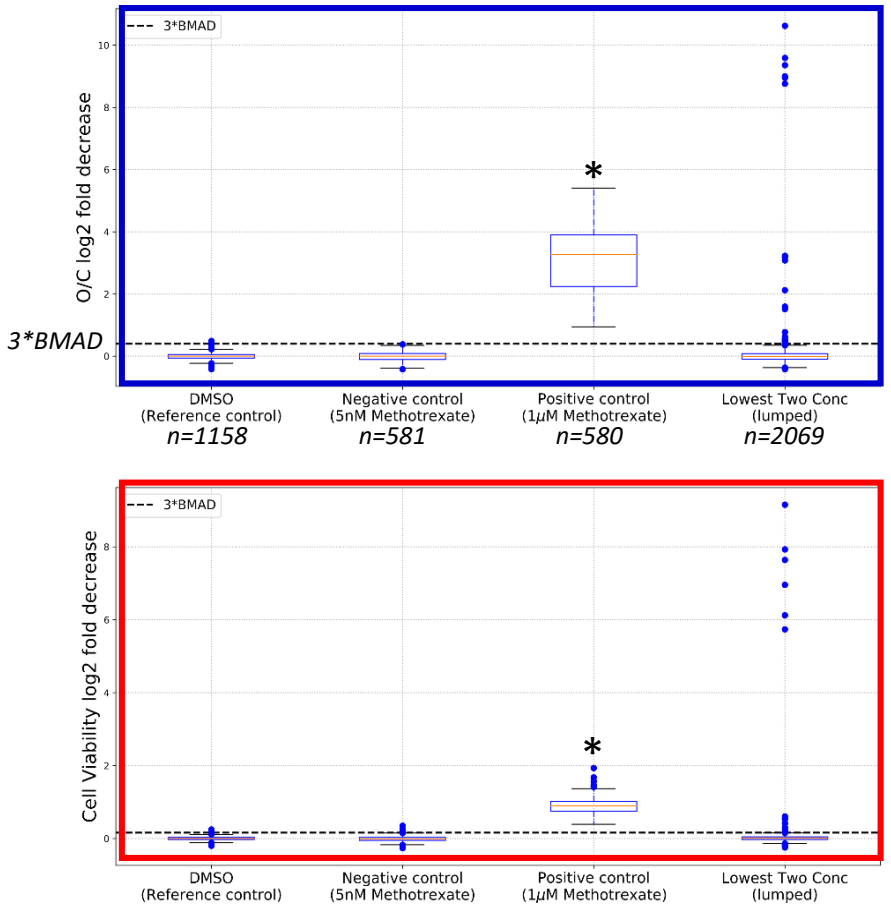




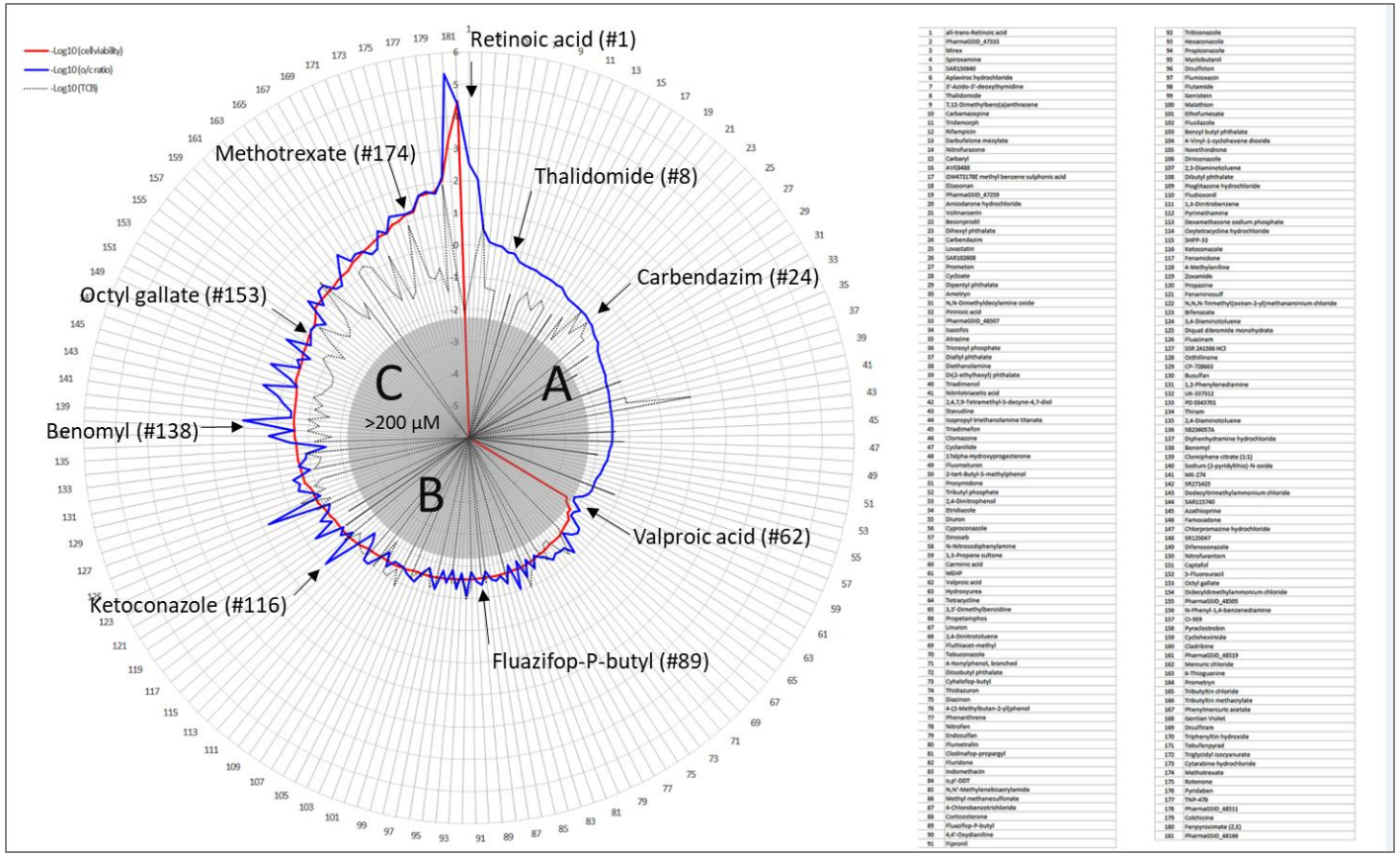
SOURCE: Zurlinden et al. (submitted)

ToxCast_STM results

Plate-level controls and tcpl samples



Positivity on 181 of 1065 (17%) ToxCast chemicals (Phase I, II)



Targeted biomarker (o/c) gives the teratogenic potential

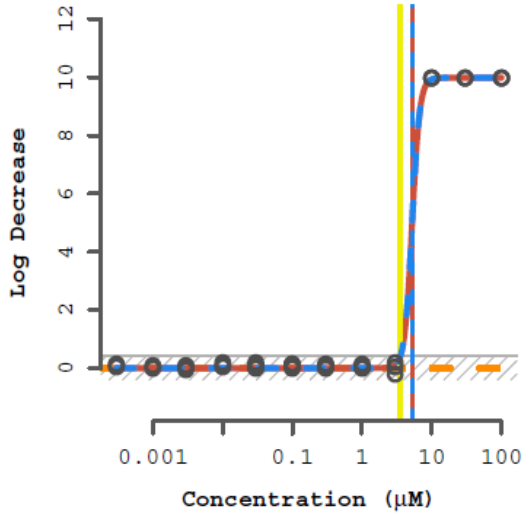
11% loss of cell number (cv) gives general cell consequence

Median AC50 of the ToxCast cytotoxicity burst

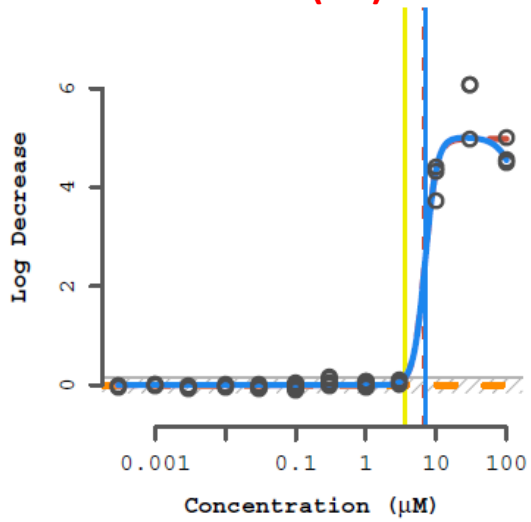
SOURCE: Zurlinden et al. (manuscript in preparation)

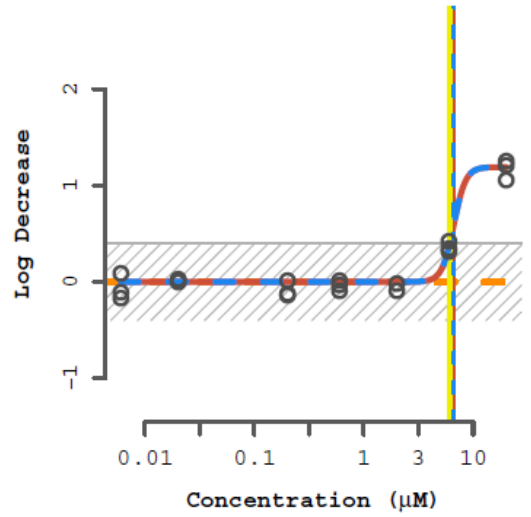
Metabolic pair: *Benomyl and its conversion product (Carbendazim)*

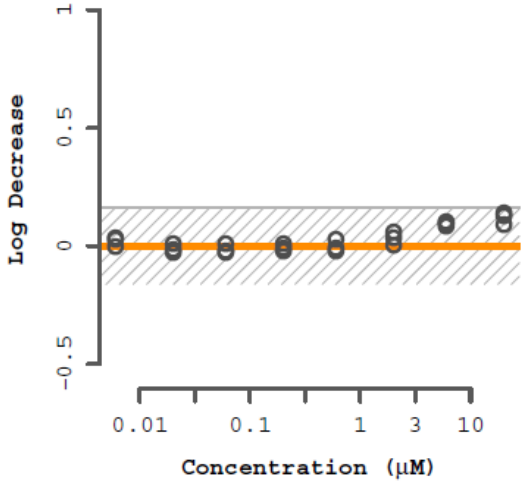
Targeted biomarker (TI)

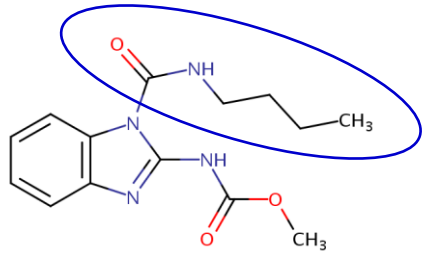


Viable cell number (CV)







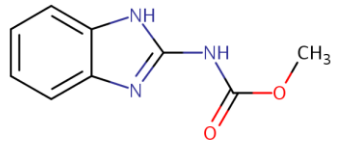


Benomyl

TI = 3.53 µM, CV = 3.63 µM

dLEL rat = 62.5 mg/kg/day (< mLEL)

dLEL rabbit = 180 mg/kg/day (mLEL)



Carbendazim

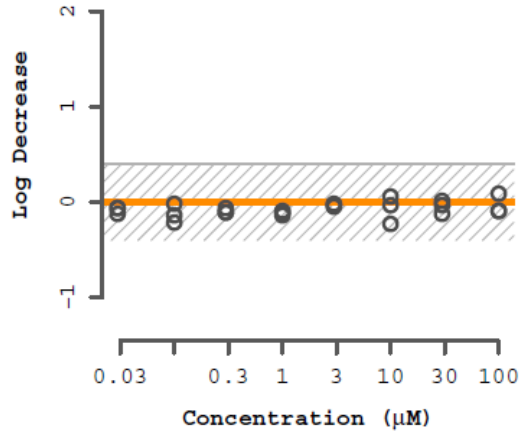
TI = 6.12 µM, CV = no effect

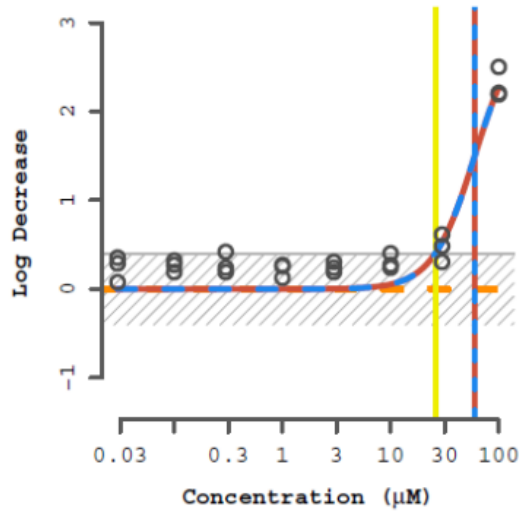
dLEL rat = 20 mg/kg/day (< mLEL)

dLEL rabbit (no ToxRefDB entry)

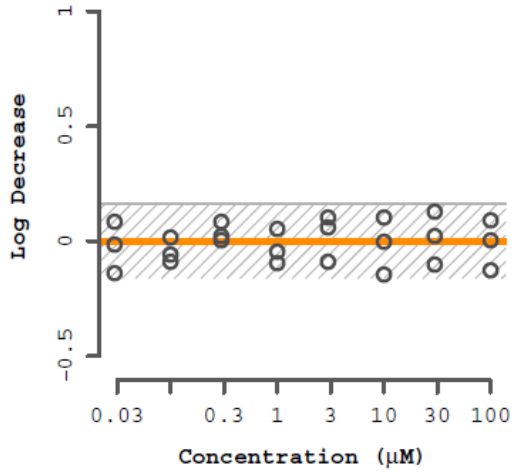
Stereoisomers: *R-enantiomer (Fluazifop-P-butyl) is the active herbicide*

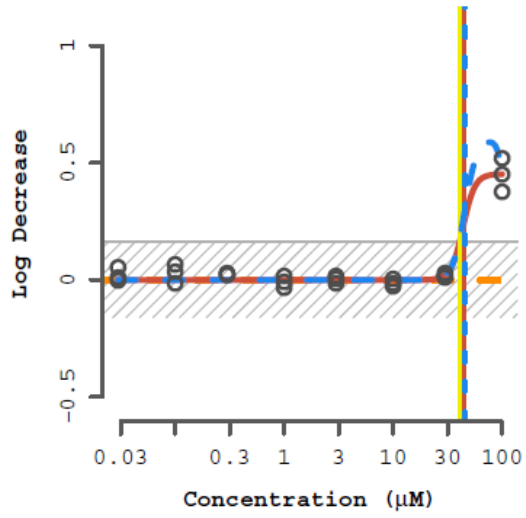
Targeted biomarker (TI)

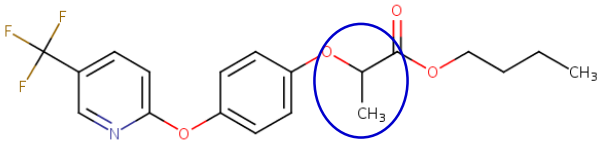




Viable cell number (CV)





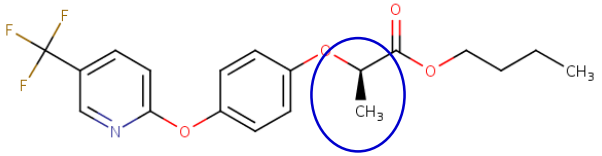


Fluazifop butyl

TI = not active, CV = no effect

dLEL rat = 10 mg/kg/day (< mLEL)

dLEL rabbit = 90 mg/kg/day (mLEL)



Fluazifop-P-butyl

TI = 26 µM, CV = 40.8 µM

dLEL rat = 5 mg/kg/day (< mLEL)

dLEL rabbit = 50 mg/kg/day (mLEL)

DevTox Performance Check

- ToxCast has 42 benchmark compounds often used to validate alternative DevTox platforms[1].
- Accuracy = 78.6% (0.65 sens, 1.00 spec) consistent with pharma-trained model.

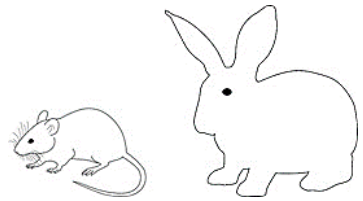
How does the STM prediction do with ToxRefDB (v1) prenatal developmental toxicity studies?

[1] Genschow et al. 2002; West et al. 2010; Daston et al. 2014; Augustine-Rauch et al. 2016; Wise et al. 2016

CASRN	Chemical	HTC[1] (µM)	CV[2] (µM)	TI[3] (µM)	Preg.class[4]	STM class[5]
302-79-4	all-trans-Retinoic acid	10	NA	0.003	X	TP
69-74-9	Cytarabine hydrochloride	1	0.083	0.054	D	TP
59-05-2	Methotrexate	1	0.062	0.059	X	TP
147-24-0	Diphenhydramine hydrochloride	100	3.76	0.588	B	TP
50-35-1	Thalidomide	100	NA	1.27	X	TP
51-21-8	5-Fluorouracil	1	1.45	2.02	D	TP
298-46-4	Carbamazepine	100	NA	2.29	C	TP
55-98-1	Busulfan	100	4.91	2.31	D	TP
13292-46-1	Rifampicin	10	NA	2.46	C	TP
19774-82-4	Amiodarone hydrochloride	10	NA	5.1	D	TP
75330-75-5	Lovastatin	20	NA	5.1	X	TP
3056-17-5	Stavudine	100	NA	32.5	C	TP
2392-39-4	Dexamethasone sodium phosphate	100	21.8	37.7	C	TP
53-86-1	Indomethacin	100	44.1	72.7	D	TP
127-07-1	Hydroxyurea	1000	237	74.9	D	TP
127-01-1	Valproic acid	1000	271	155	D	TP
4376-20-9	MEHP	500	NA	167	D	TP
57-41-0	5,5-Diphenylhydantoin	100	NA	NA	D	FN
51-52-5	6-Propyl-2-thiouracil	100	NA	NA	D	FN
10043-35-3	Boric acid	40.7	NA	NA	NTP	FN
4449-51-8	Cyclopamine	10	NA	NA	D	FN
6055-19-2	Cyclophosphamide monohydrate	20	NA*	NA	D	FN
56-53-1	Diethylstilbestrol	10	NA	NA	X	FN
107-21-1	Ethylene glycol	100000	NA	NA	NTP	FN
57-30-7	Phenobarbitol sodium	100	NA*	NA	D	FN
81-81-2	Warfarin	100	NA	NA	X	FN
69-72-7	Salicylic acid	1000	1795	513	C	TN
103-90-2	Acetaminophen	100	NA*	NA	B	TN
79-06-1	Acrylamide	36	NA	NA	NTP	TN
50-78-2	Aspirin	100	NA*	NA	C	TN
80-05-7	Bisphenol A	100	39.4	NA	NTP	TN
94-26-8	Butylparaben	100	NA	NA	GRAS	TN
58-08-2	Caffeine	500	NA	NA	B	TN
464-49-3	D-Camphor	20	NA	NA	C	TN
131-11-3	Dimethyl phthalate	100	NA	NA	NTP	TN
59-30-3	Folic acid	100	NA	NA	A	TN
54-85-3	Isoniazid	8.8	NA*	NA	C	TN
57-55-6	1,2-Propylene glycol	1000000	246664	327552	NTP	TN
68-26-8	Retinol	10	NA	NA	A	TN
81-07-2	Saccharin	100	NA	NA	A	TN
134-03-2	Sodium L-ascorbate	20	NA*	NA	A	TN
599-79-1	Sulfasalazine	100	NA*	NA	B	TN
	True Positive Rate (sensitivity)		*0.29*	*0.65*		
	True Negative Rate (specificity)		*0.94*	*1*		
	Overall Accuracy		***55.00%***	***78.60%***	***(MCC = 0.647)***	

SOURCE: NCCT, manuscript in preparation

Binary classification model: *fetal endpoints (dLEL) from ToxRefDB*



- **Key point**: BAC 78% (0.63 sensitivity, 0.91 specificity, n=127) where evidence for DevTox is strong, but drops as evidence weakens due to ↓sensitivity.

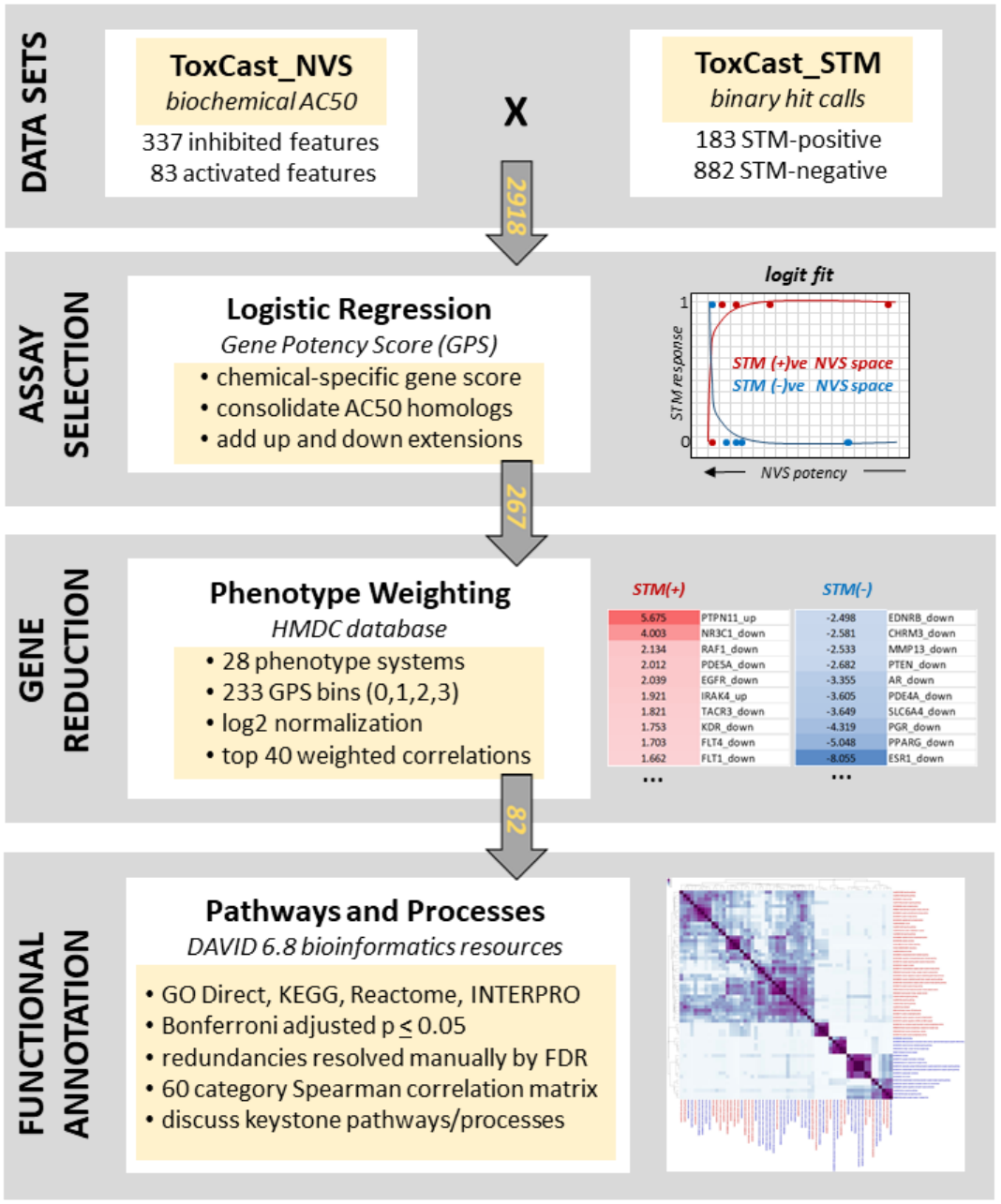
		Stringency Filter Applied to DevTox Anchor			
Condition[2]	***Base***[3,4]	***Low***[3,5]	***Medium***[3,6]	***High***[3,7]	***BM-42***[3]
TP	85	60	35	19	17
FP	14	37	23	9	0
FN	217	127	51	11	9
TN	116	208	176	88	16
n	432	432	285	127	42
sensitivity	0.281	0.321	0.407	0.633	0.654
specificity	0.892	0.849	0.884	0.907	1.000
Rand ACC	46.5%	62.0%	74.0%	84.3%	78.6%
PPV	0.859	0.619	0.603	0.679	1.000
NPV	0.348	0.621	0.775	0.889	0.640
BAC	60.3%	62.0%	68.9%	78.4%	82.0%
MCC	0.190	0.202	0.332	0.554	0.647

in vivo

in vitro

TP	FP
FN	TN

SOURCE: Zurlinden et al. (submitted)



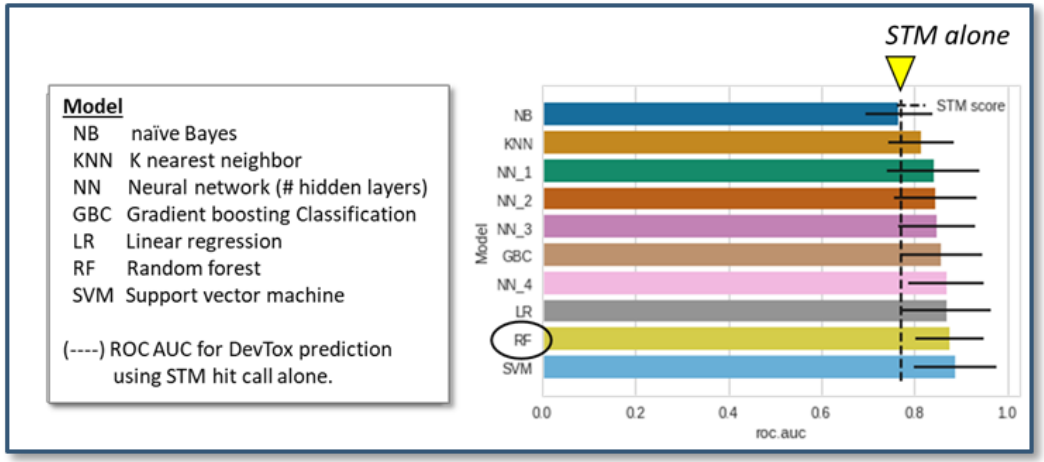
Keystone Pathways

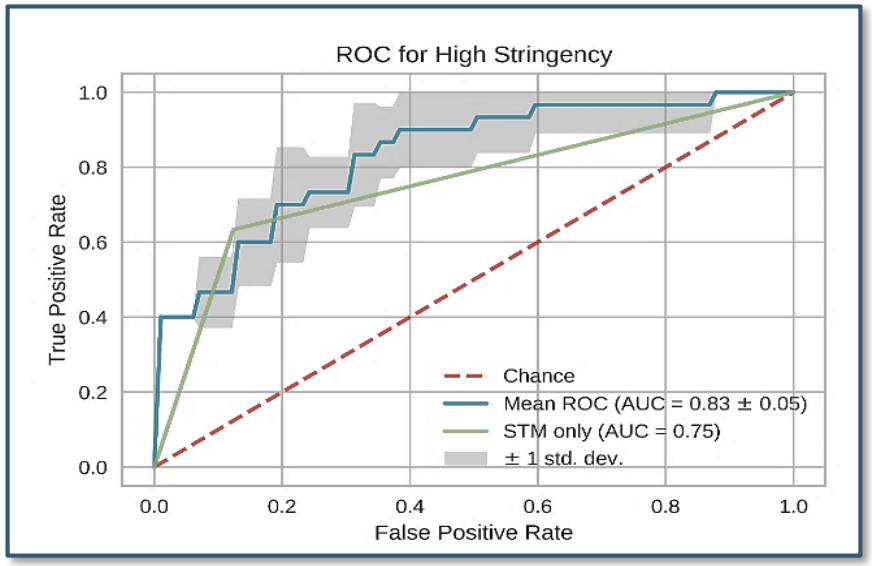
- Functional annotations inferred from mining STM response against biochemical (NVS) features.
- What we can and cannot say about the applicability domain with regards to biochemical targets:

Can machine learning to mine the ToxCast portfolio pick up some of the biology that may be missed by the hESC biomarker?

Key Point: potent MIEs may define what the STM response can and cannot predict.

DevTox mined to >800 ToxCast features





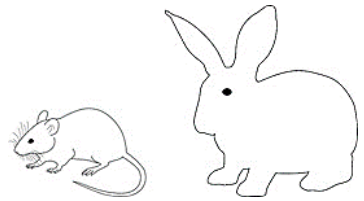
- ML with 5-fold cross-validation on train/test split;
- ~200 ToxCast features correlated with DevTox;
- STM was the top-weighted feature;
- 3 other features tied for next-most informative.

Feature	*Assay read-out (what the feature measures)*
STM_ORN/CYSS_dn	critical effect of the hESC biomarker
ATG_CRE_cis_up	cis-acting reporter activation via cAMP/CREB
ATG_NRF2_ARE_cis_up	NFE2L2 antioxidant response element
ATG_PXR_cis_up	cis-acting reporter activation via PXR/PXRE

- sets up a hierarchical rules-based decision workflow:
  - Rule 1: STM(+) & CREB3(-) predicts TP (86.4%)
  - Rule 2: CREB3/NRF2/PXR (+) overrides STM(+) as TN
  - Rule 3: STM(-) & PXR(+) OR NRF2(+) predicts TN (91.3%)
  - Rule 4: STM(-) & CREB3/NRF2/PXR(-) condition predicts TN (83.3%)

SOURCE: Todd Zurlinden (work in progress)

Refined binary classification model: *ToxCast augmentation (+)*



- **Key point**: Augmenting the hESC response with ToxCast data for 3 adaptive pathways (UPR, ARE, XME) improved positive predictive value to BAC up to 88%.

Condition	Low	Low+	Medium	Medium+	High	High+
TP	60	50	35	33	19	19
FP	37	24	23	13	9	3
FN	127	137	51	53	11	11
TN	207	220	175	185	88	94
n	431	431	284	284	127	127
sensitivity	0.321	0.267	0.407	0.618	0.633	0.633
specificity	0.848	0.902	0.884	0.976	0.907	0.969
Rand ACC	61.9%	62.6%	73.9%	89.8%	84.3%	89.0%
PPV	0.619	0.676	0.603	0.875	0.679	0.864
NPV	0.620	0.616	0.774	0.902	0.889	0.895
BAC	61.9%	64.6%	68.9%	88.9%	78.4%	87.9%
MCC	0.201	0.222	0.331	0.679	0.554	0.676

SOURCE: Todd Zurlinden (work in progress)

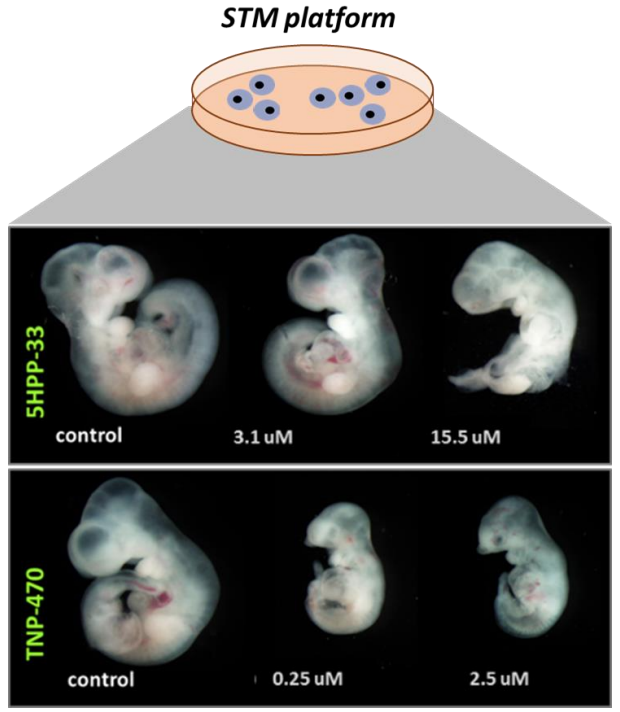
hESC (predicted) vs rat WEC (observed)

5HPP-33: *synthetic thalidomide analog*

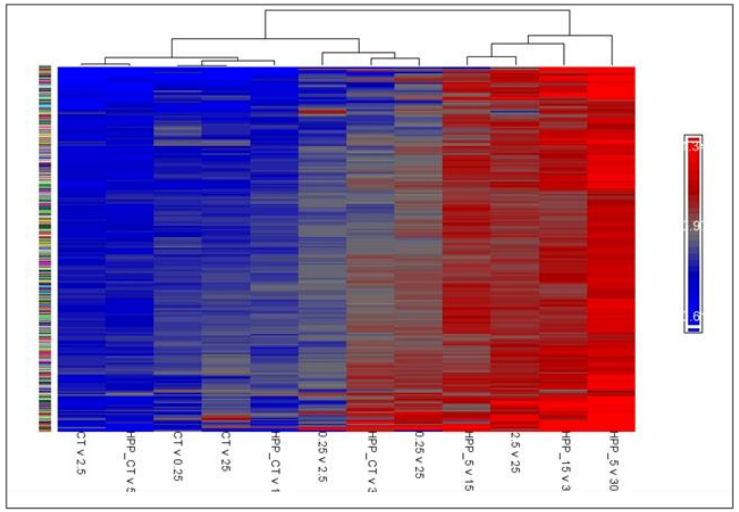
- T.I. predicted 9.5 µM
- AC50 observed 21.2 µM (embryo viability)

TNP-470: *synthetic fumagillin analog*

- T.I. predicted 0.01 µM
- AC50 observed 0.04 µM (dysmorphogenesis)



RNAseq: exposure-based potential for DevTox at 4h correlated with changes in common for pathways regulating splicesome-RNA metabolism and proteasome-ubiqutination.



SOURCE: Ellis-Hutchings et al. (2017) Reprod Toxicol; Siali et al. (2019) Curr Opin Toxicol

BIOCHEMICAL
- NVS (380)

REPORTER
- ATG (161)
- Tox21 (252)

CELLULAR
- ACEA (5)
- APR (103)
- hESC (2)

CELL-CELL
- BSK (96)
- mESC (2)

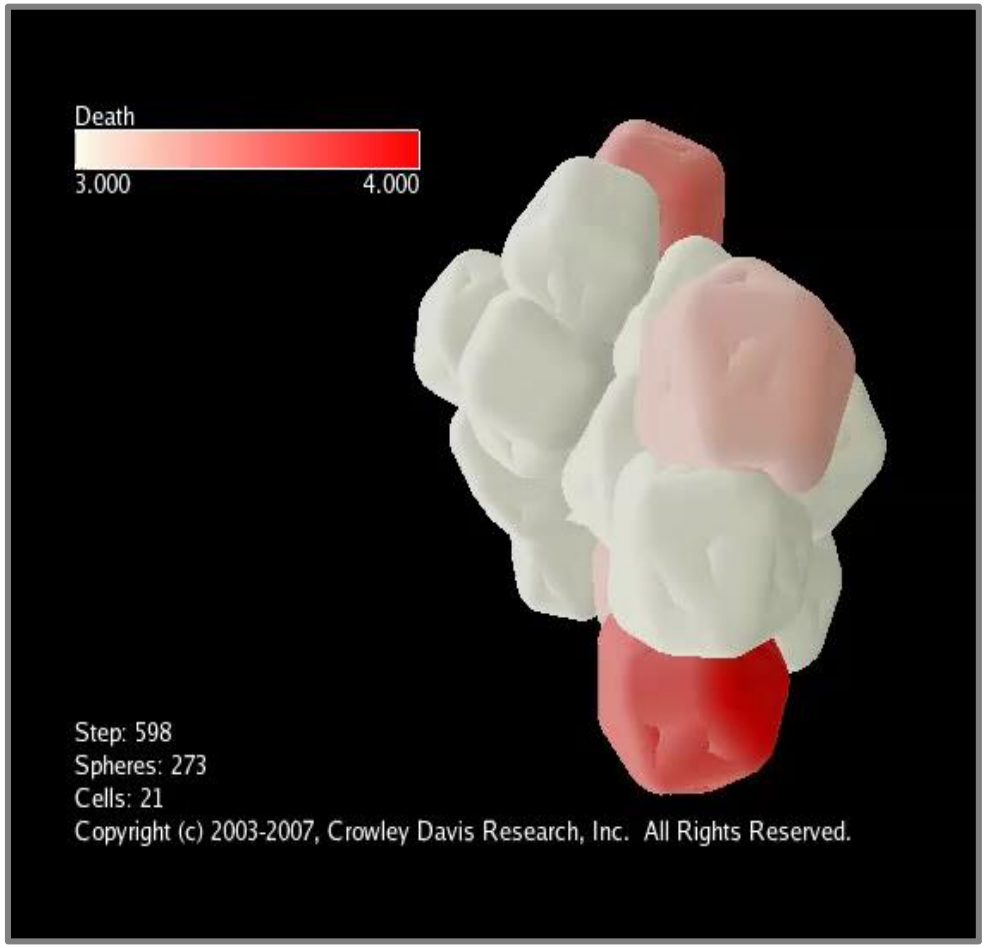
EMBRYOLOGY
- OCM
- ABM

Recursive partitioning (v2): mining *in vitro* bioactivity profiles of 1000 features in ToxCast/Tox21 HTS assays to assess the value of the STM dataset for predicting human developmental toxicity (*in progress*).

Bring in the embryology to better understand mechanisms and translate NAMs

Anatomical homeostasis in a self-regulating ‘Virtual Embryo’

- EA for self-regulation (fitness measure) - simulation executes randomly paired agents (parent cells) that generate daughter cells mutated in their rules.
- You only need to specify the goal of the computation; EA searches rule-space using ‘survival of the fittest’ (good solutions propagate, poor solutions discarded).



SOURCE: Andersen, Newman and Otter (2006) Am. Assoc. Artif. Intel.

Agent-Based Models (ABMs)

- nature-inspired *agents* (cells) and *rules* (behaviors) are set into motion as a self-organizing virtual system, using an open-source modeling environment (CompuCell3d.org).

- soft-computing uses ‘fuzzy logic’ to simulate forces or properties governing cell fate and behavior where rules are inexact or knowledge incomplete (computational intelligence).

- can change course in response to a particular situation or stimulus, such as genetic errors or biomolecular lesions introduced from real world data (dynamic translation).

- probabilistic rendering of where, when and how a particular condition might lead to an adverse developmental outcome (cybermorphs).

Translating cellular lesions into quantitative phenotypes

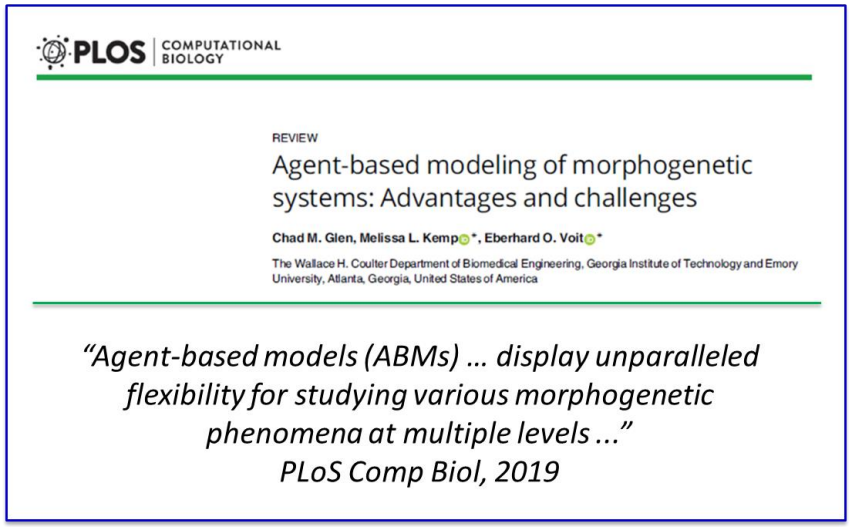
Core Developmental Processes
Patterning (Sets up Future Events)
Timing (Clocks and Oscillators)
Differentiation (Cell Diversification)
Morphogenesis (Tissue Organization)

Cellular Primitives
Growth (Proliferation)
Growth (Volume Increase)
Death (Apoptosis)
Differentiation (Function)
Adhesion (Differential Hypothesis)
Shape (Geometry)
Motility (Cell Migration)
Extra Cellular Matrix (Remodeling)

Morphogenetic Movement
Folding
Epiboly
Convergent Extension
Branching Morphogenesis
Cell Condensation
Cell Sorting
Trans-Differentiation
Cavitation
Involution/Invagination
Tractional Forces

Directed Cell Movement
Contact Guidance (Boundaries)
Haptotaxis (ECM Tracks)
Chemotaxis (Chemical Signals)

- Morphogenesis is fundamentally complex; the hallmark resides in the ability of cells to interact with one another.
- Genetic signals setup spatial information that cells then translate into a coordinated biological response.
- Just as 'the Cell' is the basic unit of biology, so too should it be the computational unit ('Agent') for modeling the embryo.


PLOS COMPUTATIONAL BIOLOGY

REVIEW

Agent-based modeling of morphogenetic systems: Advantages and challenges

Chad M. Glen, Melissa L. Kemp*, Eberhard O. Voit*

The Wallace H. Coulter Department of Biomedical Engineering, Georgia Institute of Technology and Emory University, Atlanta, Georgia, United States of America

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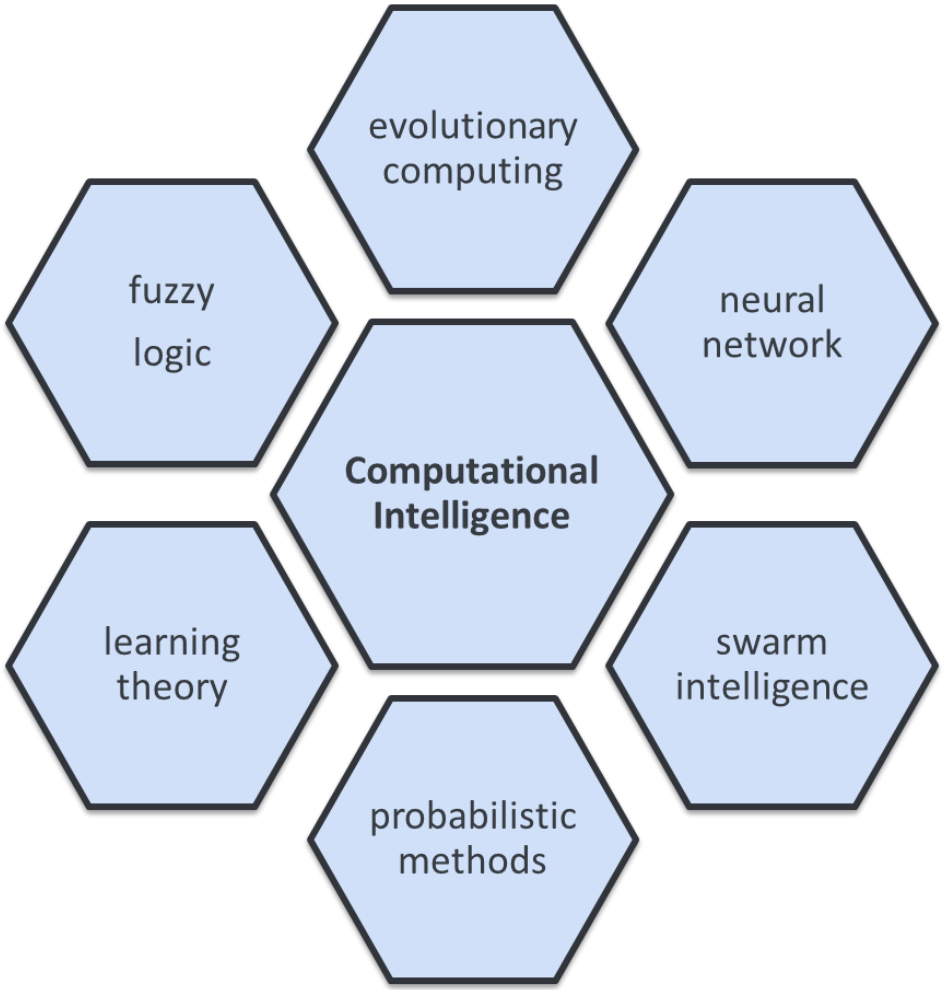
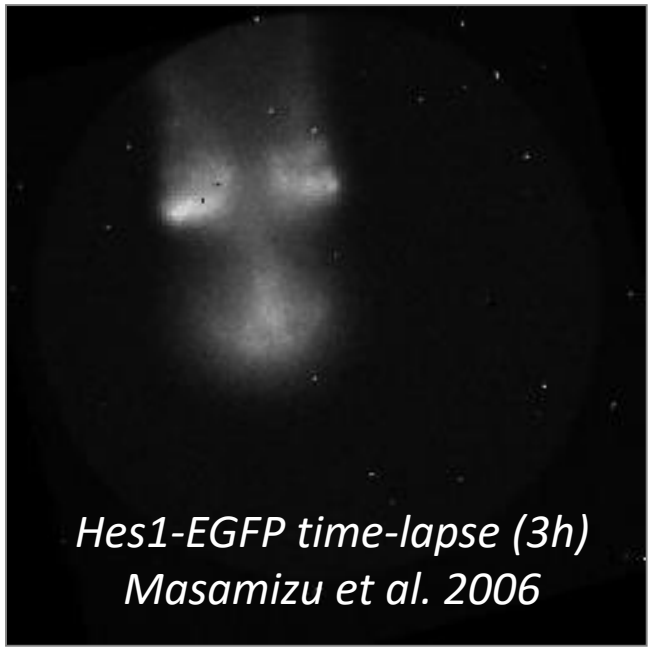
PLoS Comp Biol, 2019

Computational Intelligence

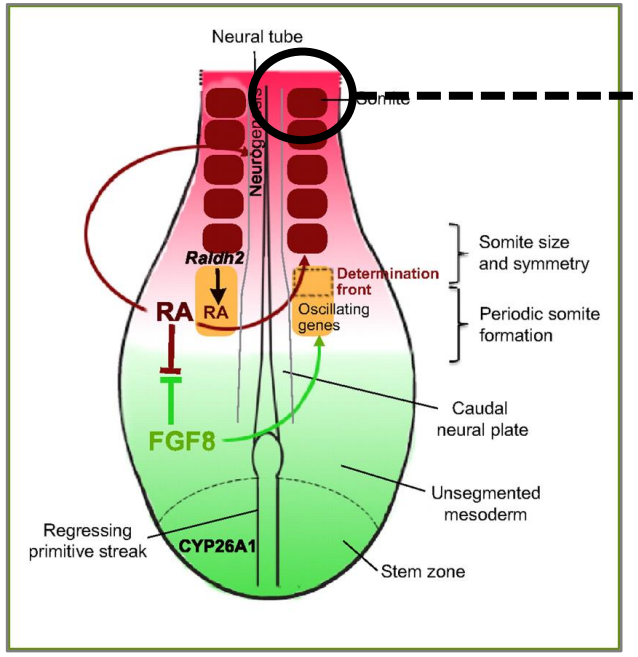
https://www.thescientist.com **May, 2019**

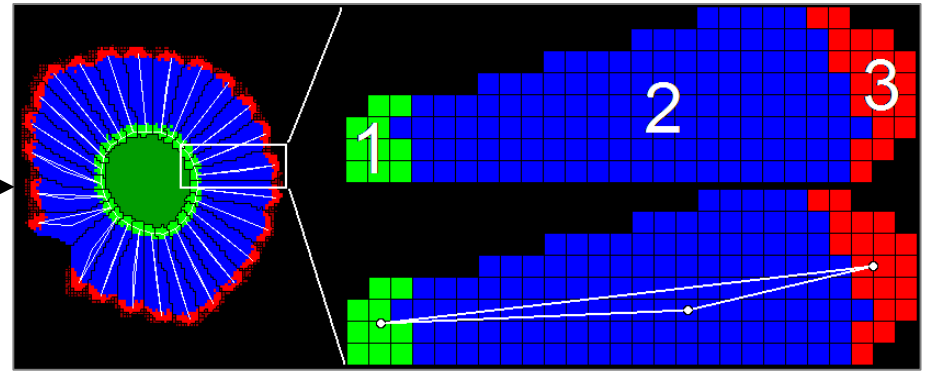


Somite development



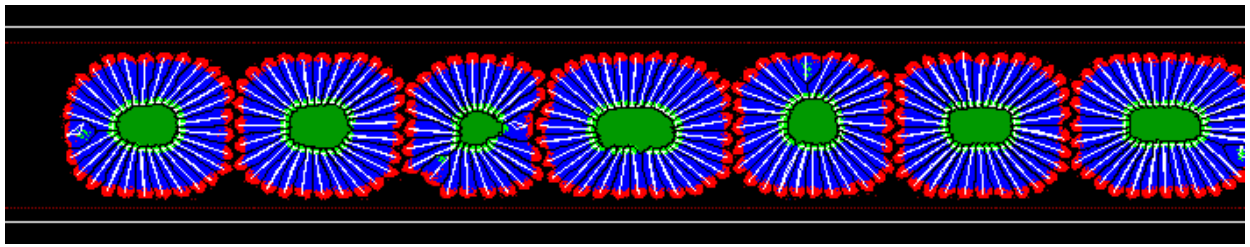
Hes1-EGFP time-lapse (3h)
Masamizu et al. 2006



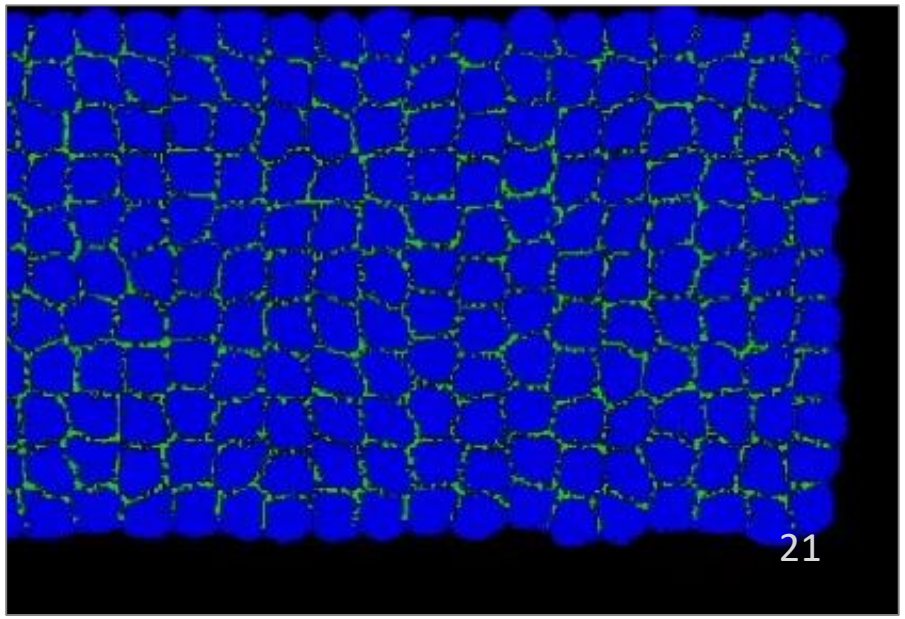


Differential cell adhesion

- *clock genes do not oscillate*
- *somites form simultaneously*

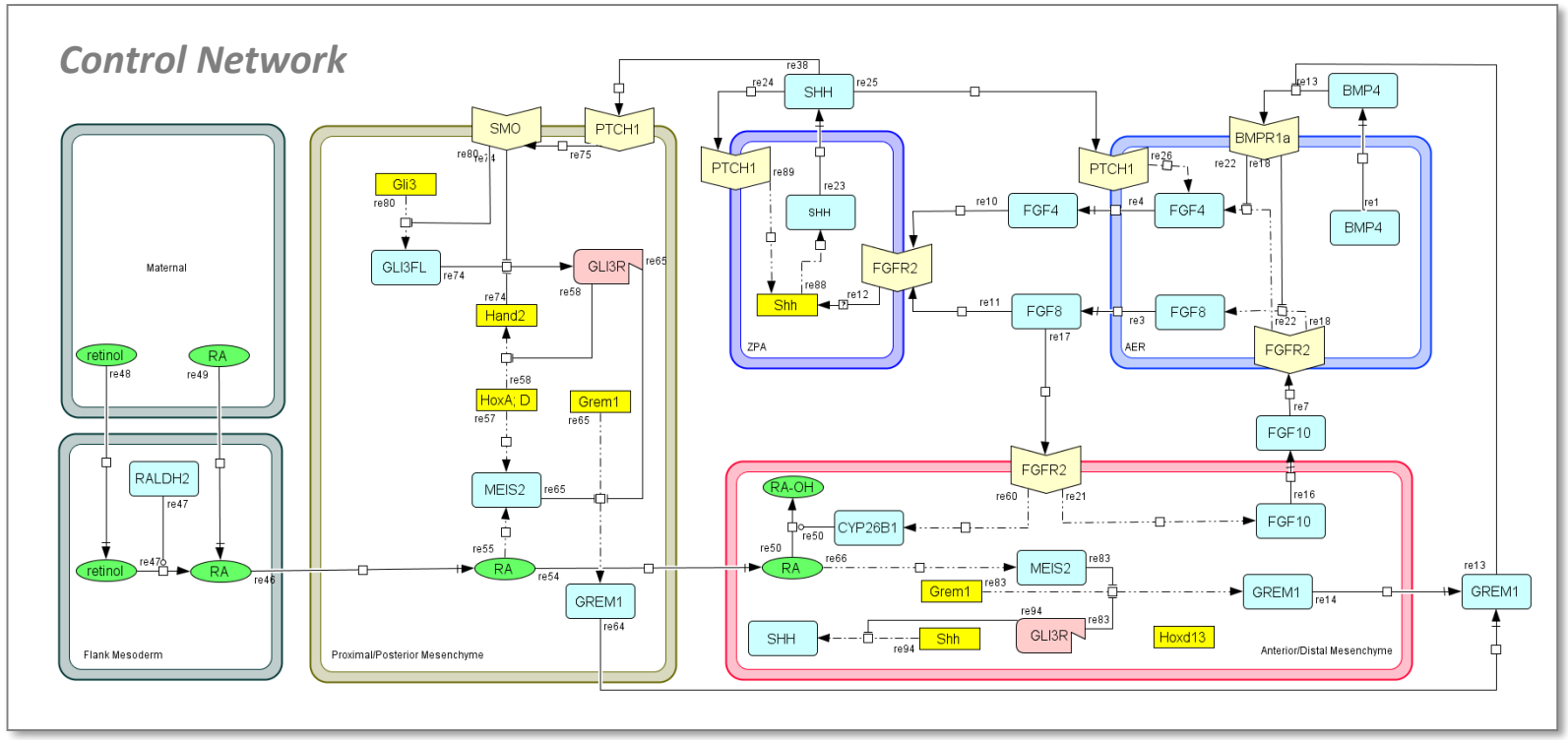


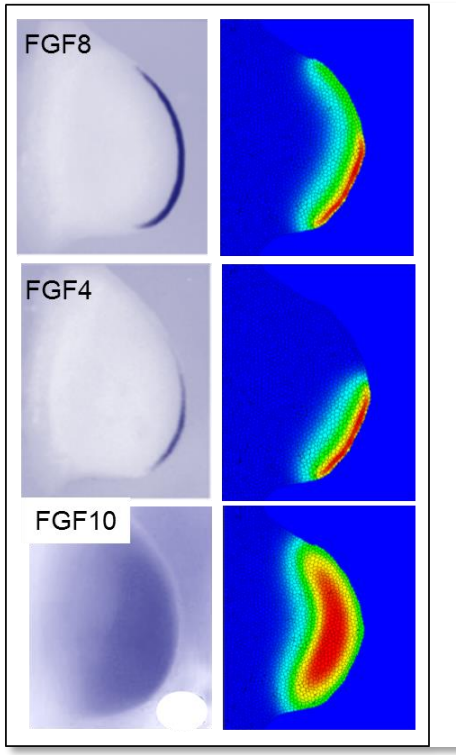
- *FGF8 wavefront restores sequentiality*
- *oscillatory clock improves regularity*

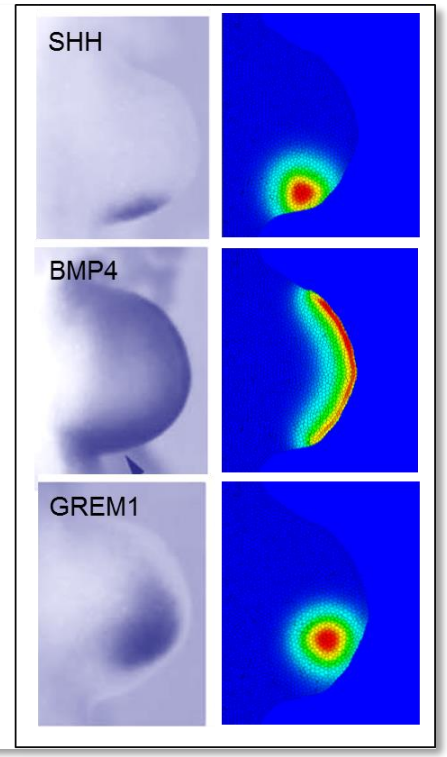


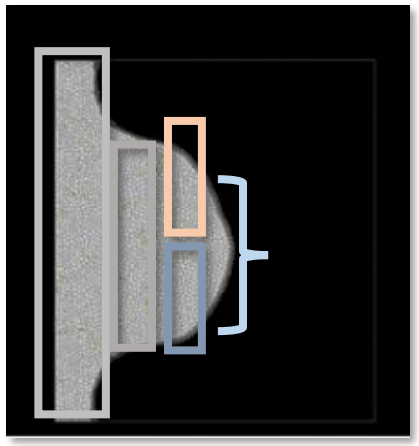
SOURCE: Dias et al. (2014) Science

Translating genetic control circuits into phenotypes with C.I.

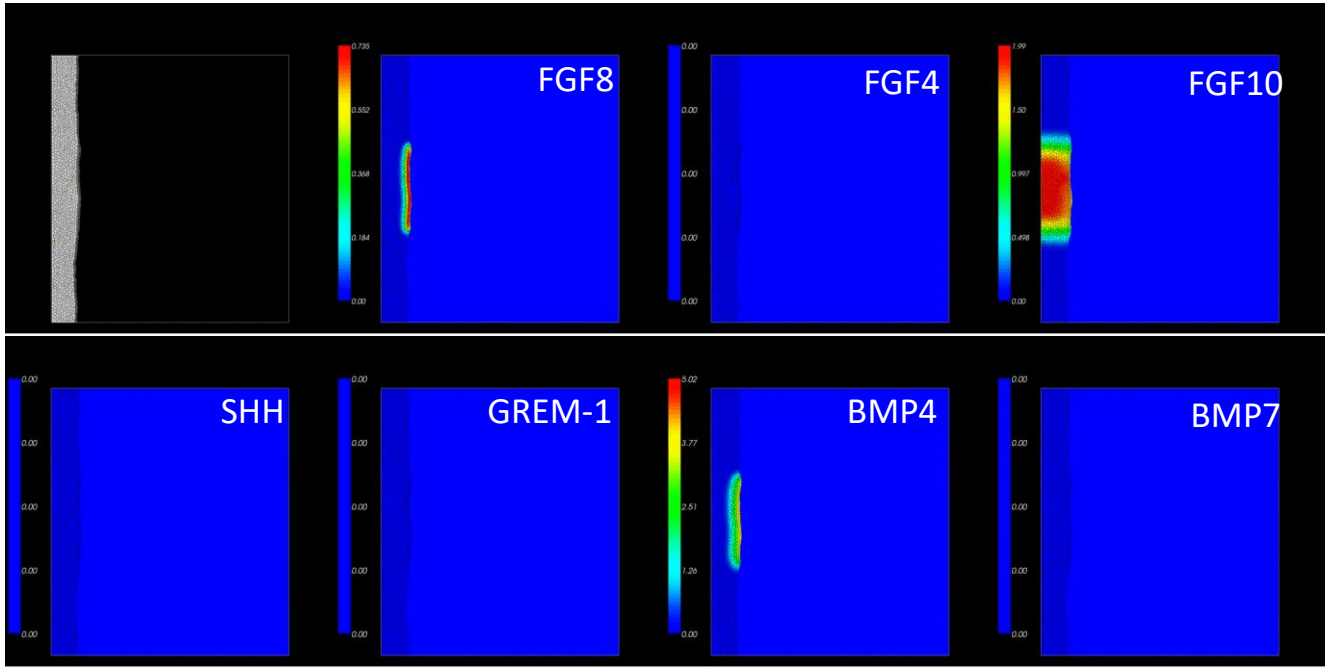


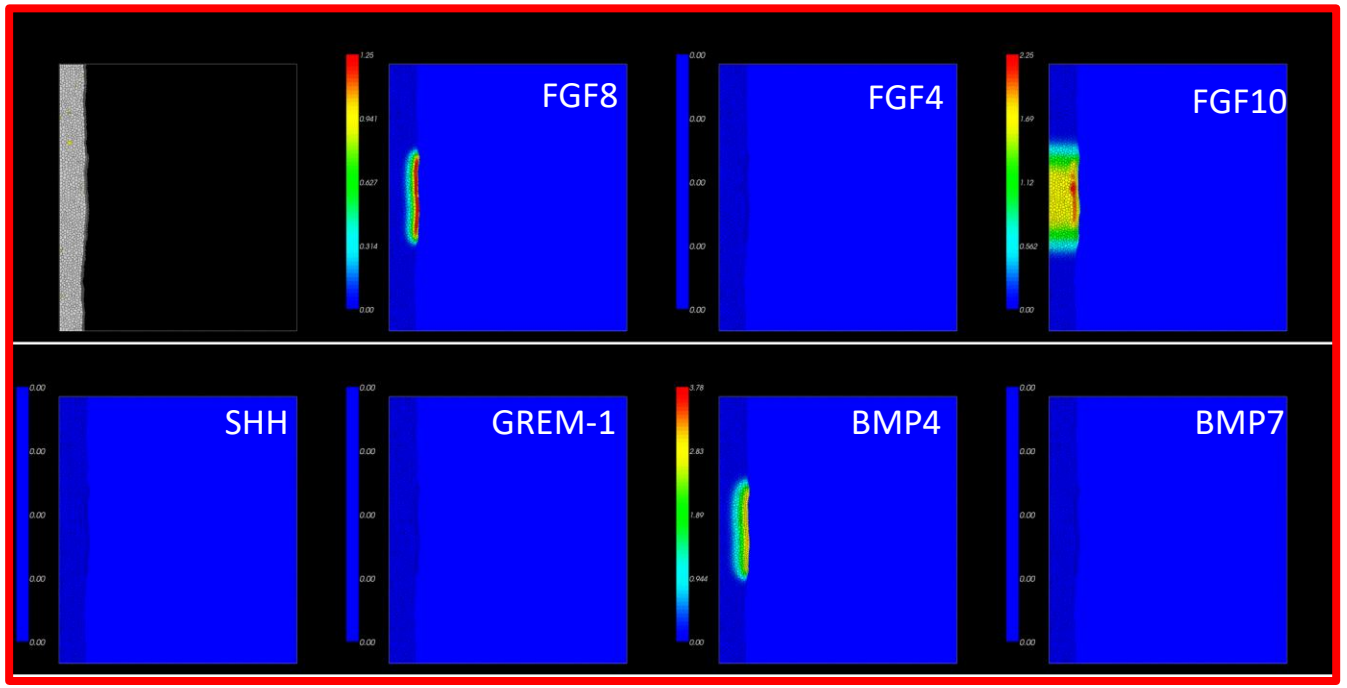






- biological wiring diagram maps cell-cell signaling
- we code the signal-response for individual cell types
- and enable ‘steppables’ of individual cell behavior in CompuCell3d.org
- executing the simulation triggers signal-response behaviors
- can quantitatively monitor emergent properties


FGF8
FGF4
FGF10
SHH
GREM-1
BMP4
BMP7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FGF10
SHH
GREM-1
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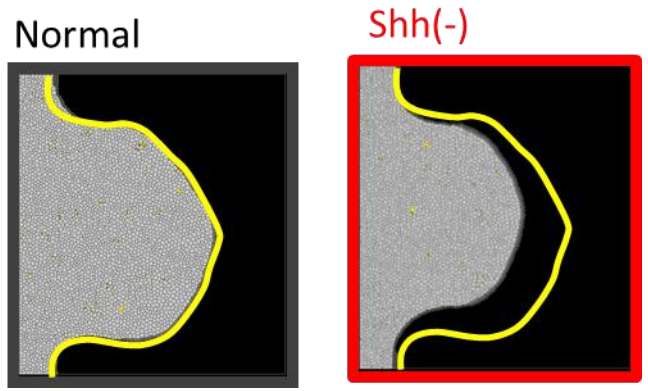
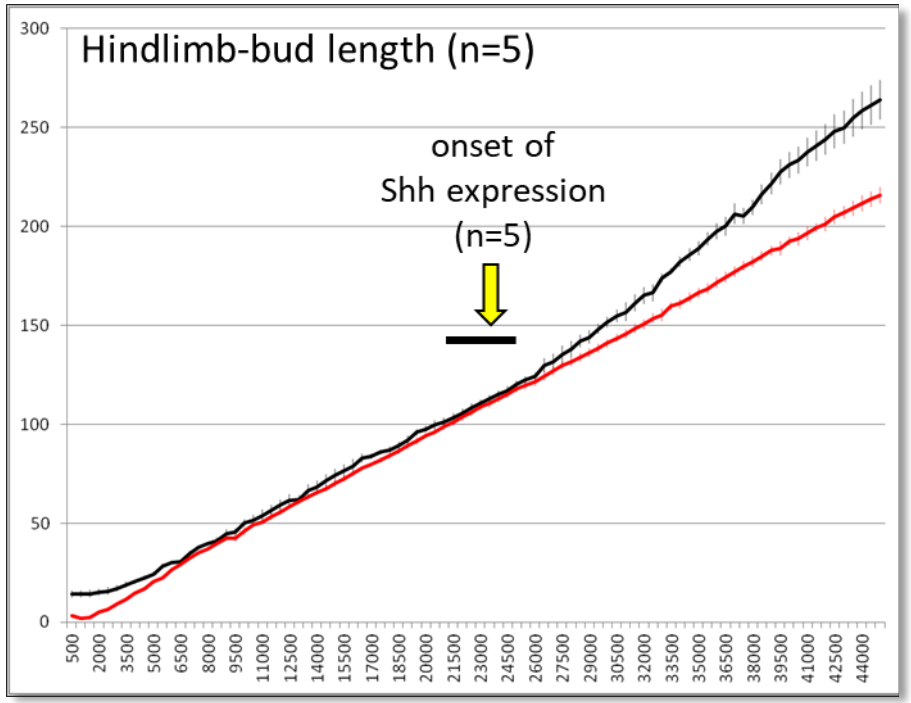

Normal
Shh(-)


Hindlimb-bud length (n=5)
onset of
Shh expression
(n=5)
300
250
200
150
100
50
0
500
2000
3500
5000
6500
8000
9500
11000
12500
14000
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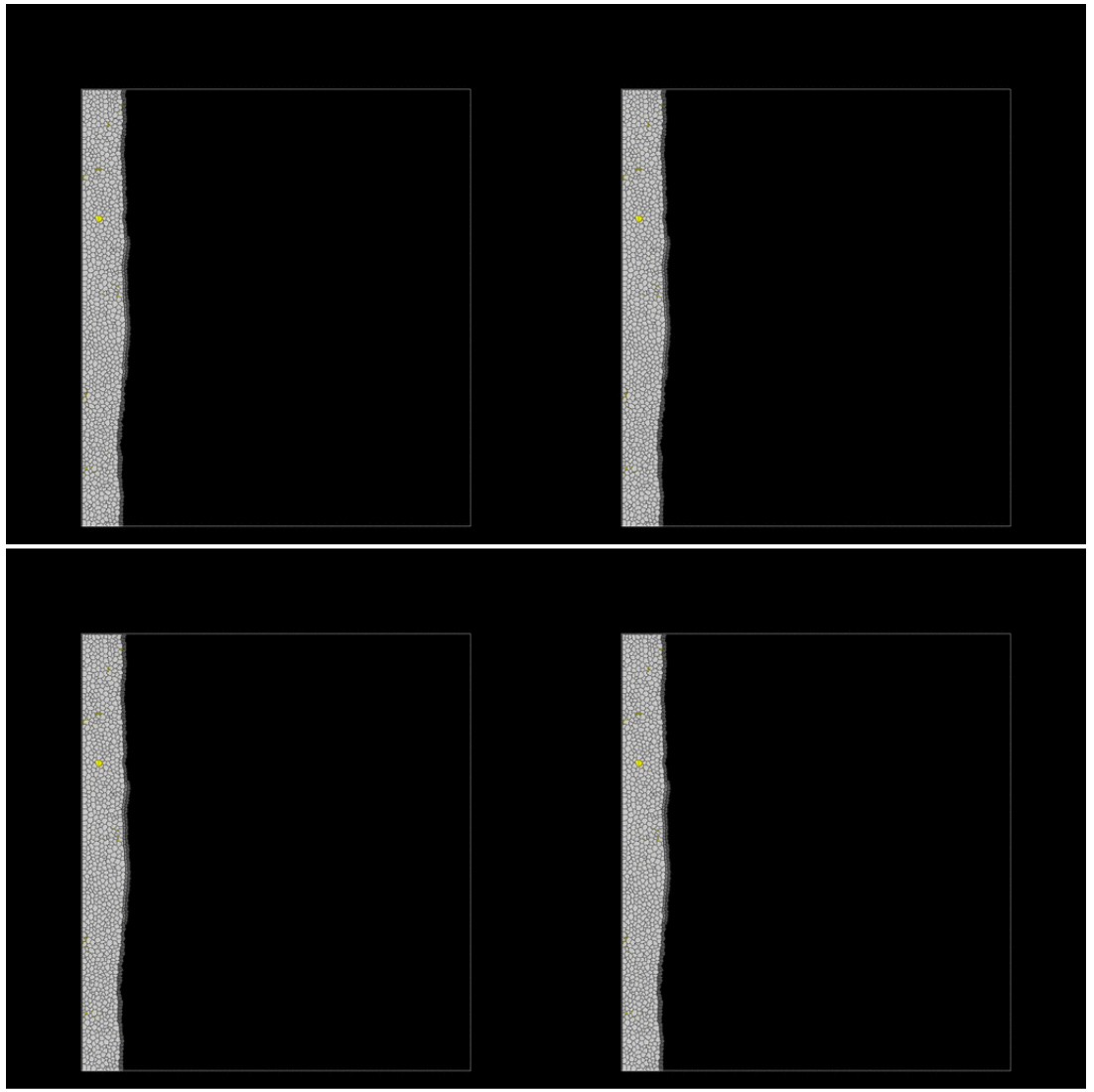
Introducing cellular lesions into the swarm …

- SI addresses collective behavior of a complex self-organizing system emerging from local interactions.
- Agents work together in closed-loop systems (e.g., flocks, schools, colonies, swarms) → phenotype.
- Subtle details in the simulation can greatly influence the outcome (checkpoints?).



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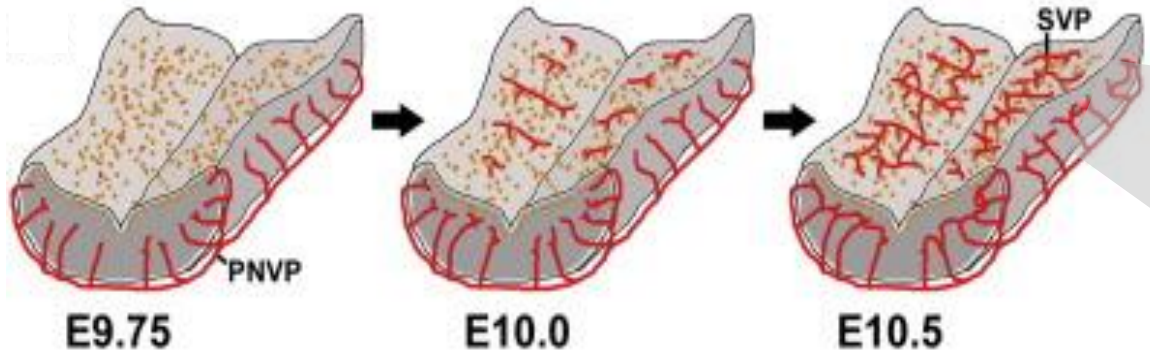
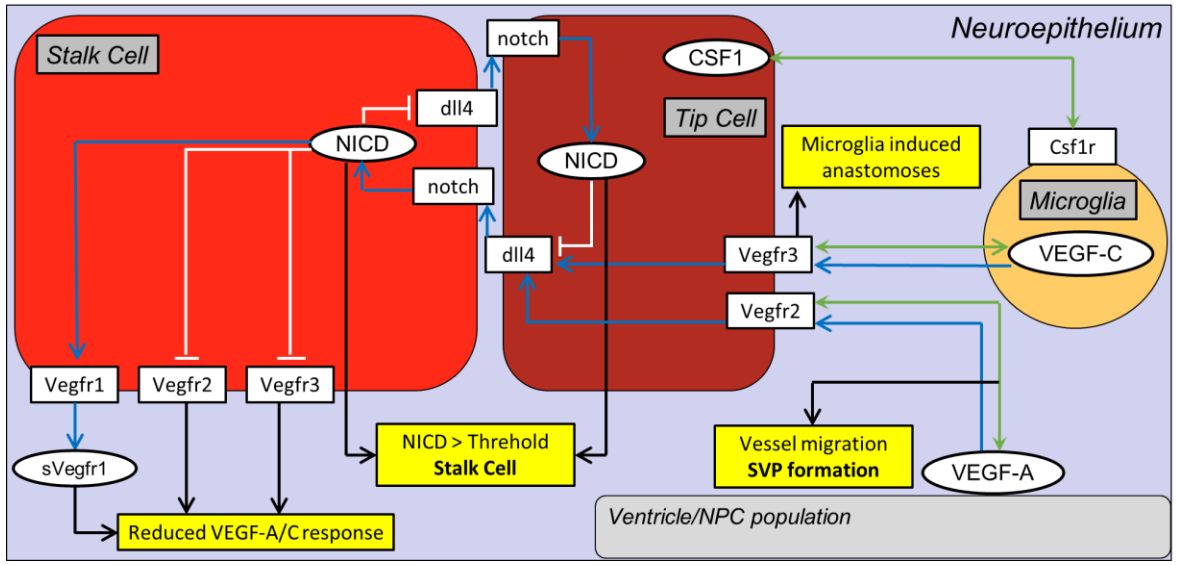
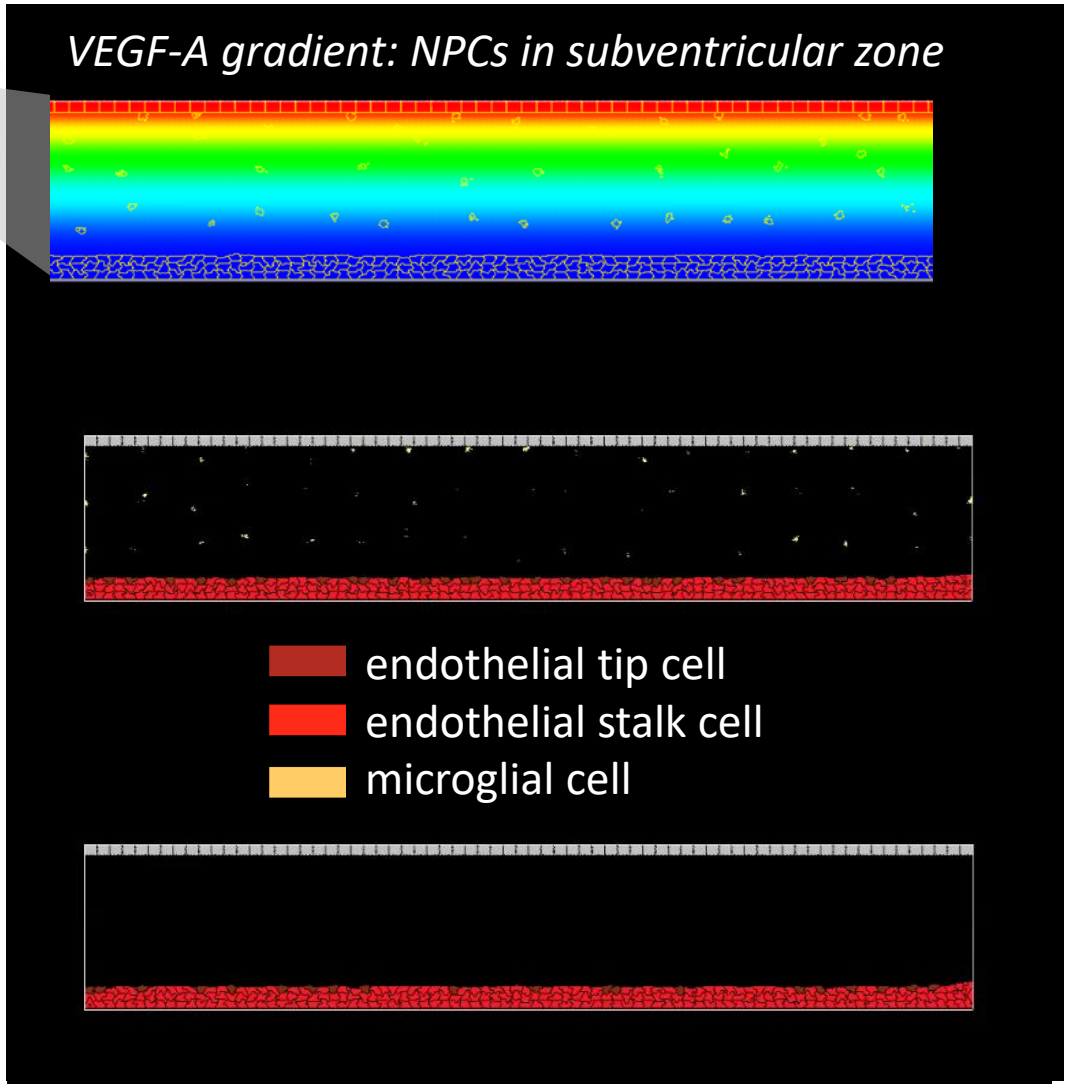
PLoS Comp Biol, 2019

Brain angiogenesis

Tata et al. (2015) *Mechanism Devel*

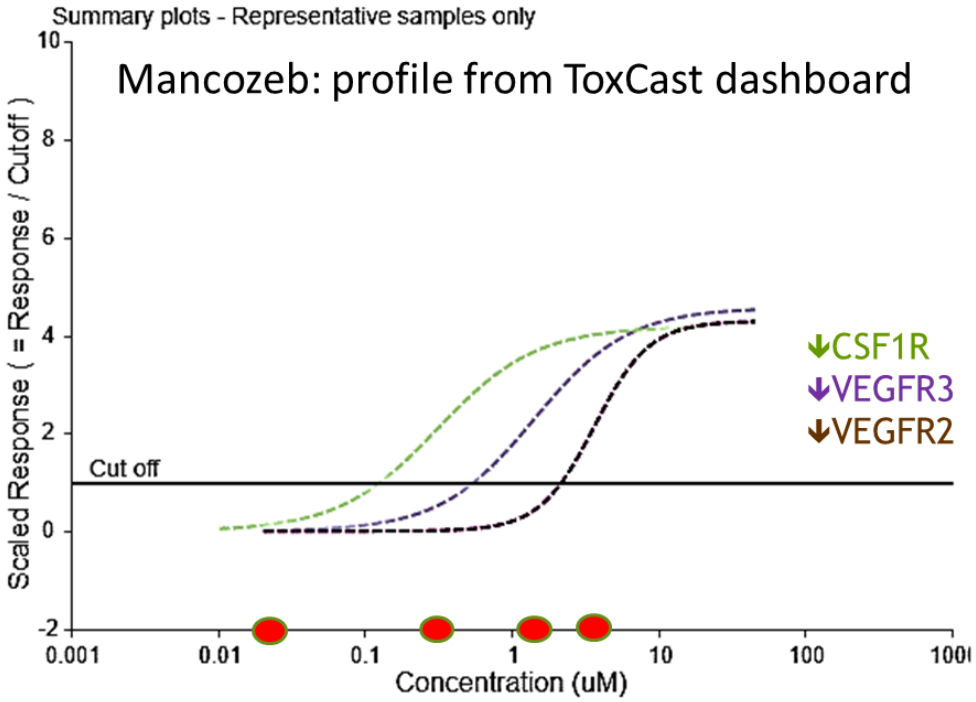


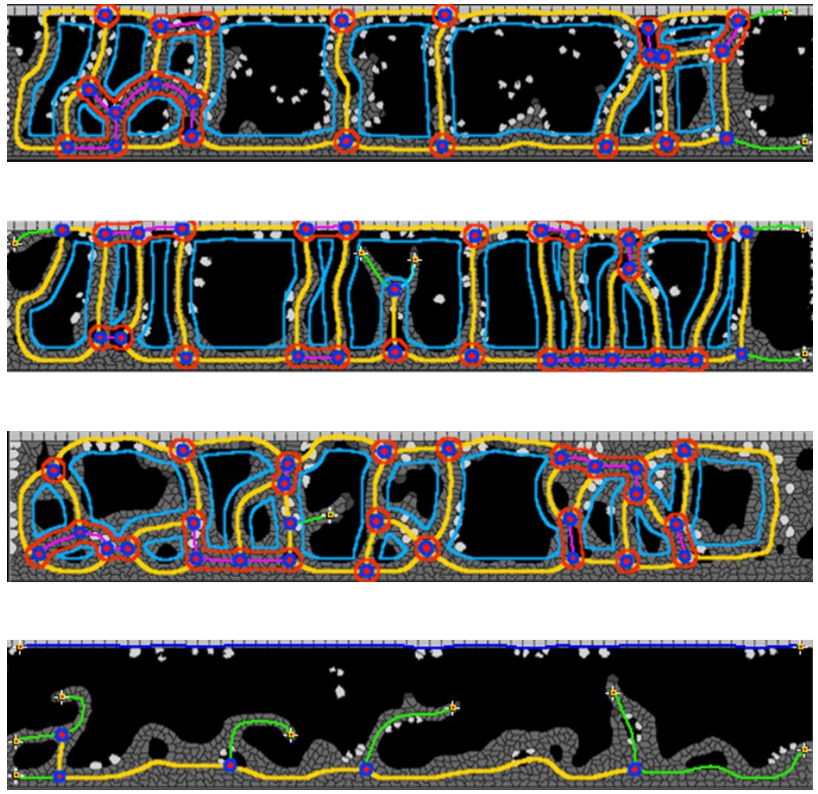




SOURCE: Zurlinden, Kate Saili (2018) – NCCT, unpublished

Executing a simulated dose-response





0.03 µM

0.3 µM

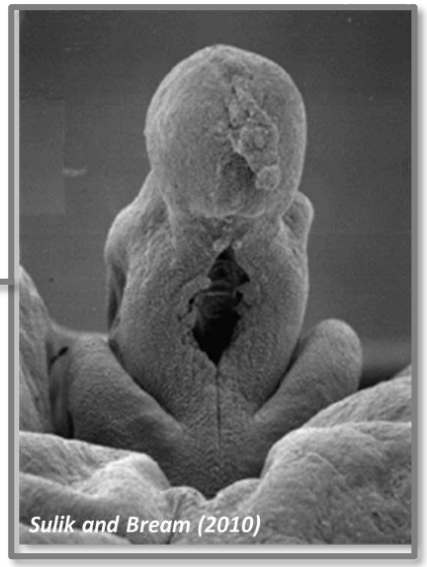
2.0 µM

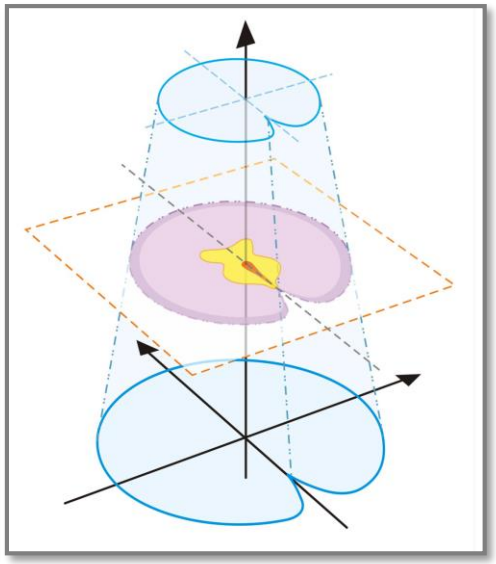
6.0 µM

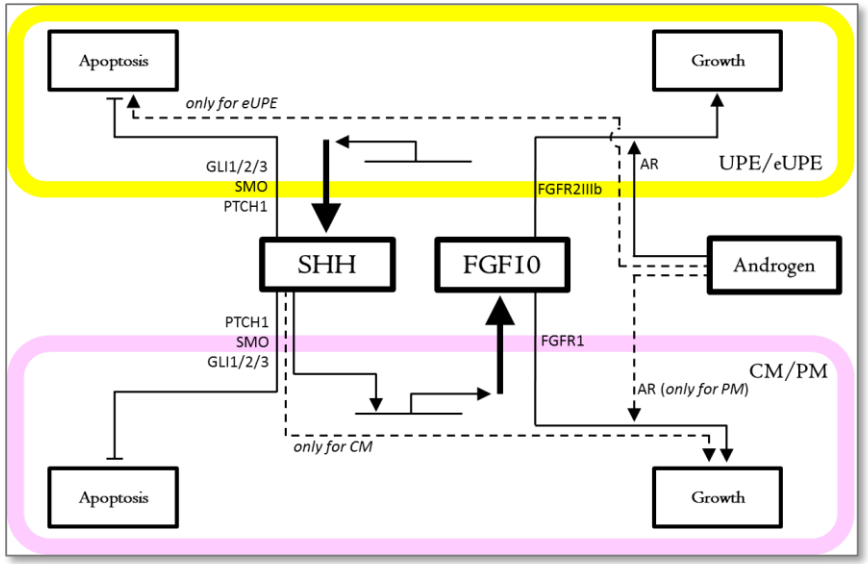
Critical concentration of Mancozeb on brain angiogenesiis:

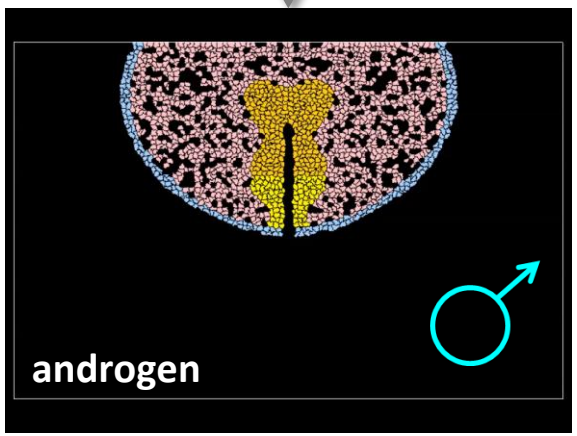
- predicted from *in silico* model ~0.5 µM (Zurlinden, NCCT)
- observed in 3D organotypic culture model of the hNVU ~0.3 µM (Daly, UWisc)

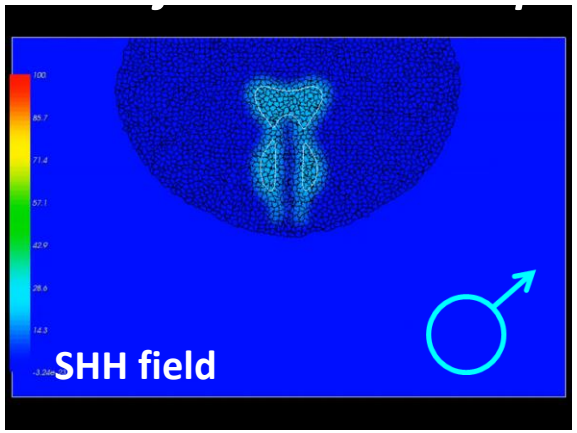
Sexual dimorphism: *genital tubercle development*

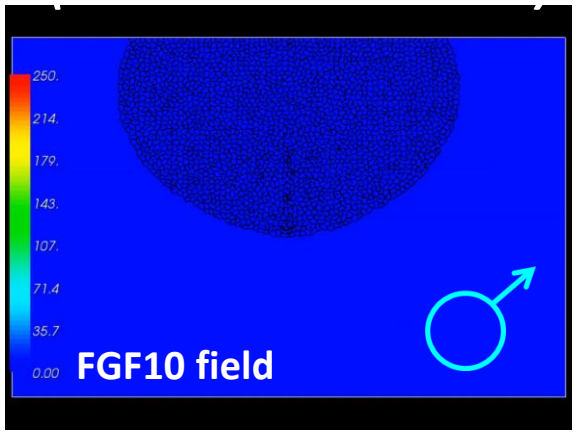


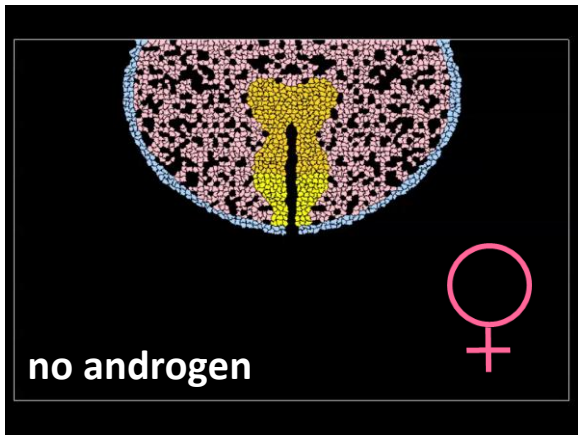




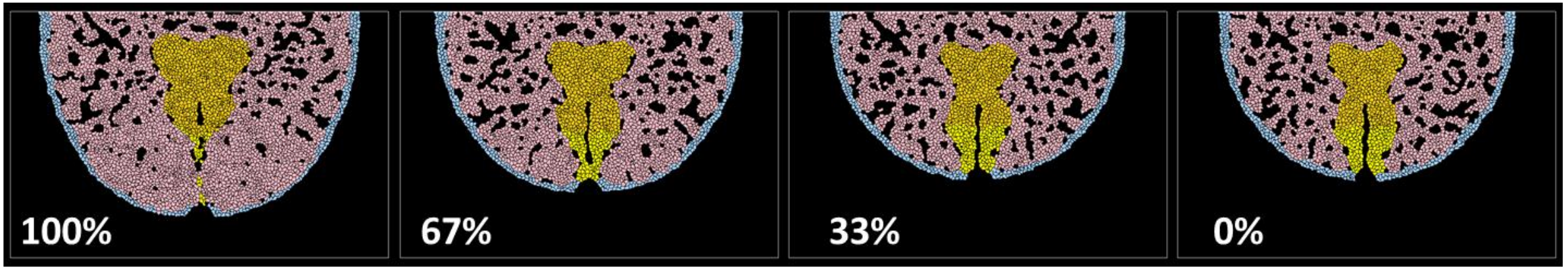


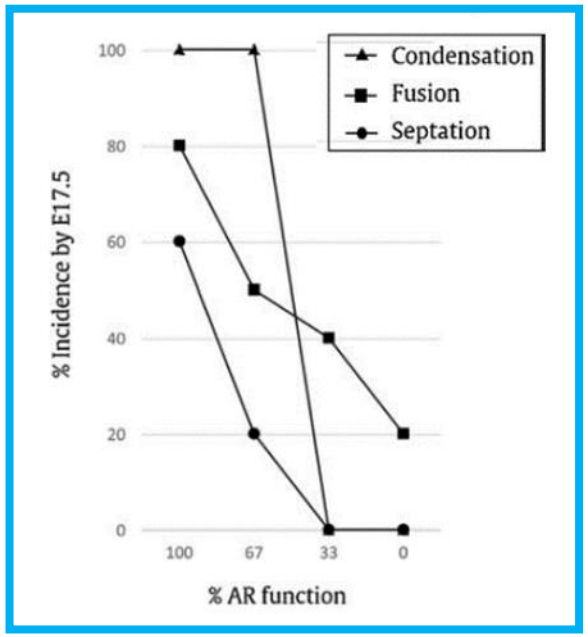






Androgen virulization: *closure rates @4000 MCS ∫ androgen supply*

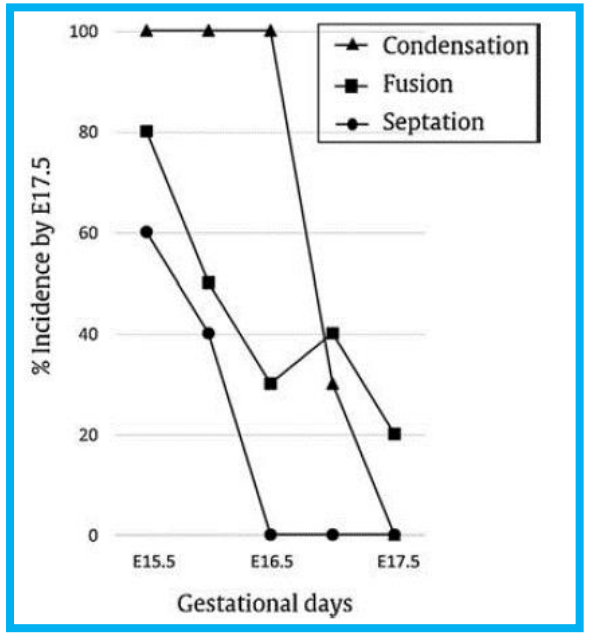




Closure indices (simulated, n=10)

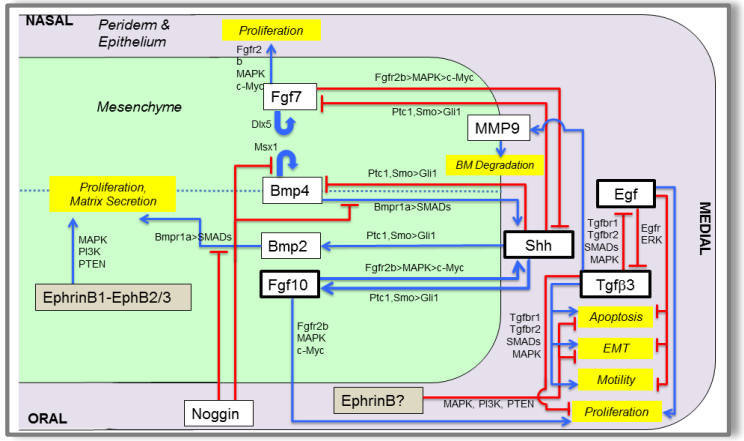
LEFT: androgen insufficiency

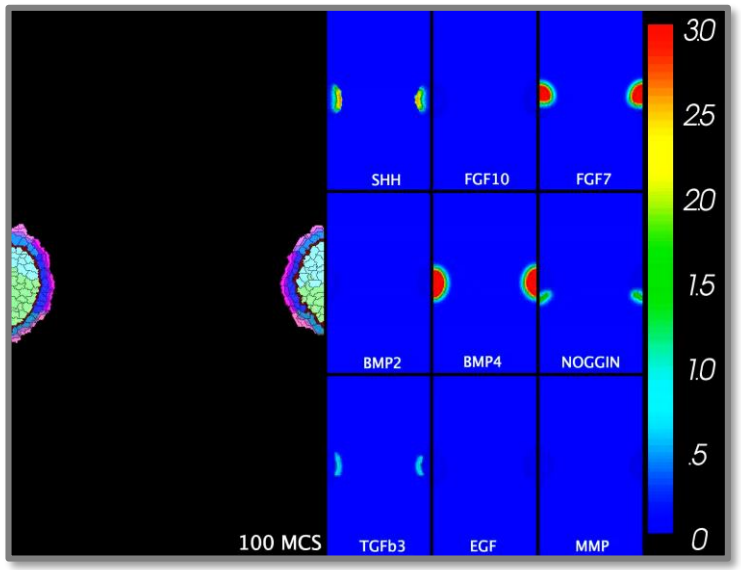
RIGHT: delayed virulization



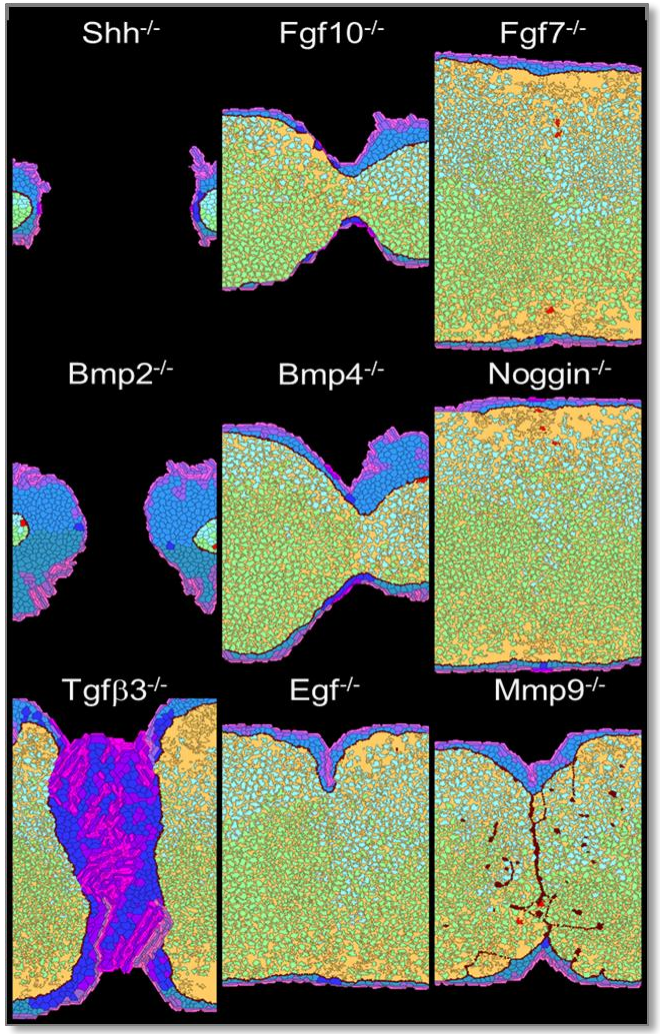
Palatal fusion

Control network

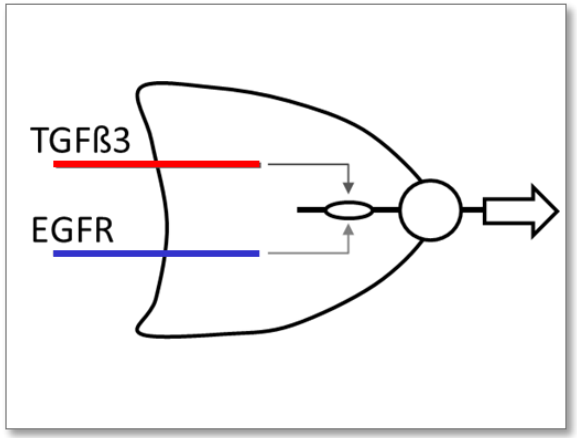


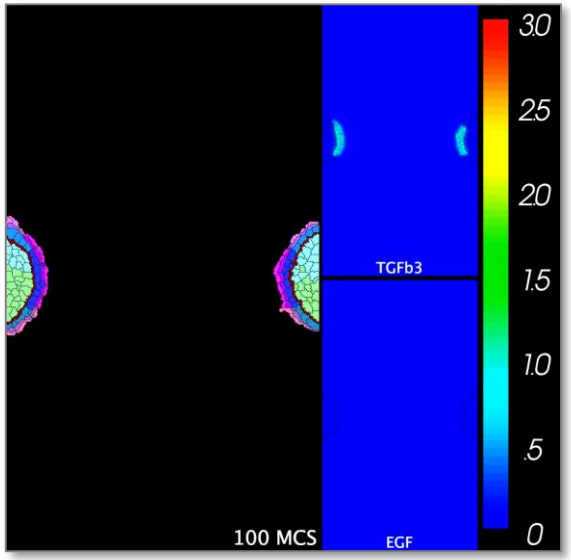


Cybermorphs



Bistable switch





SOURCE: Hutson et al. (2017) Chem Res Toxicol

Smart model …

Chemical Research in Toxicology

Article

pubs.acs.org/crt

Computational Model of Secondary Palate Fusion and Disruption

M. Shane Hutson,*[†,‡] Maxwell C. K. Leung,[‡] Nancy C. Baker,[§] Richard M. Spencer,[§] and Thomas B. Knudsen*[,∥]

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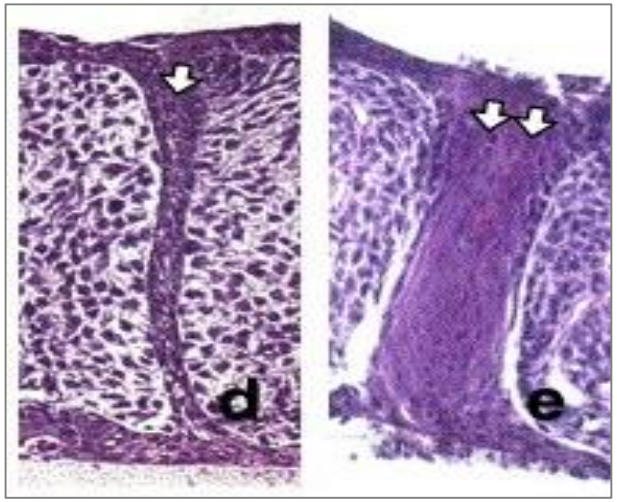
[‡]Oak Ridge Institute for Science & Education, Oak Ridge, Tennessee 37832, United States
[§]Leidos, Research Triangle Park, Durham, North Carolina 27711 United States
[∥]National Center for Computational Toxicology, Office of Research & Development, U.S. Environmental Protection Agency, Research Triangle Park, Durham, North Carolina 27711, United States

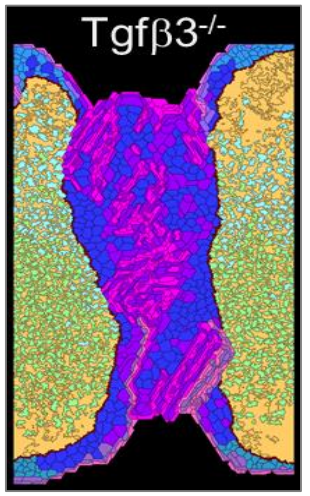
Supporting Information

ABSTRACT: Morphogenetic events are driven by cell-generated physical forces and complex cellular dynamics. To improve our capacity to predict developmental effects from chemical-induced cellular alterations, we built a multicellular agent-based model in CompuCell3D that recapitulates the cellular networks and collective cell behavior underlying growth and fusion of the mammalian secondary palate. The model incorporated multiple signaling pathways (TGFβ, BMP, FGF, EGF, and SHH) in a biological framework to recapitulate morphogenetic events from palatal outgrowth through midline fusion. It effectively simulated higher-level phenotypes (e.g., midline contact, medial edge seam (MES) breakdown, mesenchymal confluence, and fusion defects) in response to genetic or environmental perturbations. Perturbation analysis of various control features revealed model functionality with respect to cell signaling systems and feedback loops for growth and fusion, diverse individual cell behaviors and collective cellular behavior leading to physical contact and midline fusion, and quantitative analysis of the TGF/EGF switch that controls MES breakdown—a key event in morphogenetic fusion. The virtual palate model was then executed with theoretical chemical perturbation scenarios to simulate switch behavior leading to a disruption of fusion following chronic (e.g., dioxin) and acute (e.g., retinoic acid) chemical exposures. This computer model adds to similar systems models toward an integrative "virtual embryo" for simulation and quantitative prediction of adverse

Reviewer Comment: *"Crucial mechanisms occurring during palate fusion, especially opposing palatal shelf adhesion, are not considered in the model. In fact, the main reason why Tgf-b3 KO mice have cleft palate is a failure of opposing MEE adhesion, leading to separation of palatal shelves after their initial contact. Even in those strains in which palatal shelves adhere partially, I have never seen a MES as the one shown in Fig. 5."*

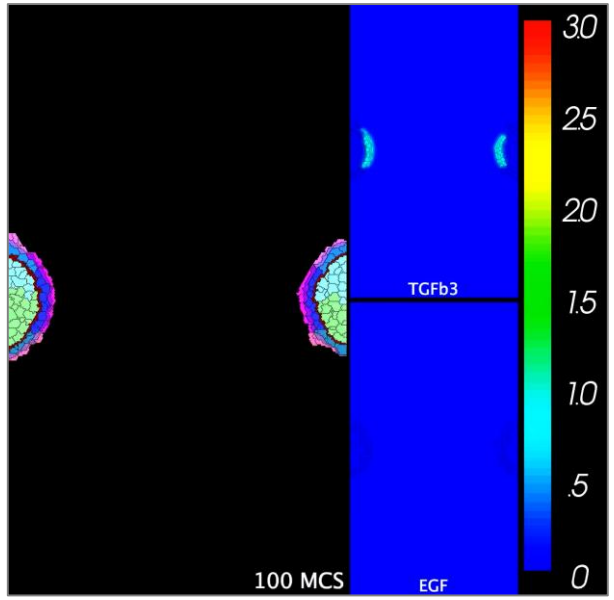
Our Response: TGF-b3 knockout mouse palates transduced with ALK vectors *in vitro. (from Dudas et al. 2004).*

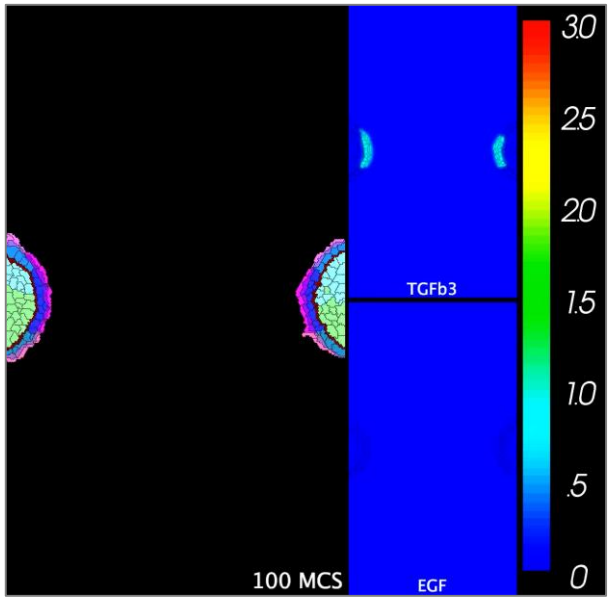


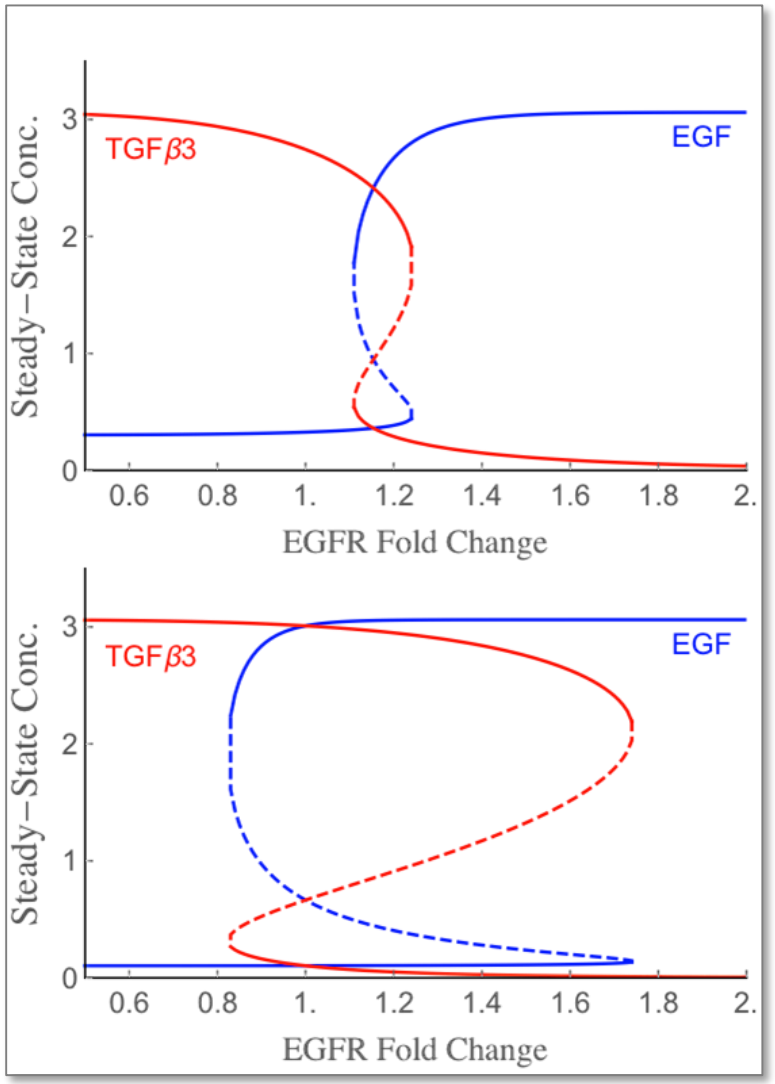


SOURCE: Hutson et al. (2017) Chem Res Toxicol

Messin' with the switch: *two scenarios for bistable dynamics*







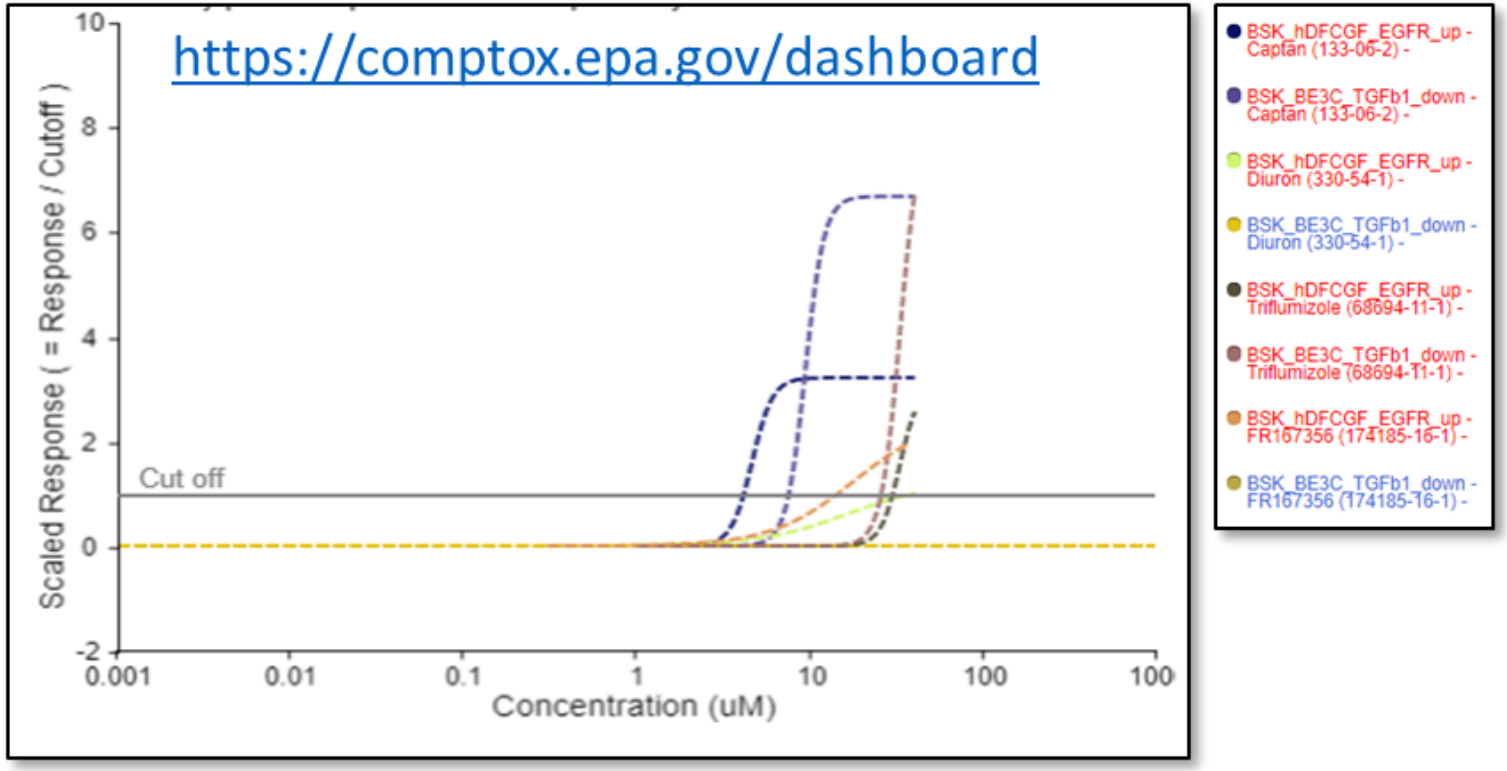
Narrow hysteresis:
less resilient but reversible

Broad hysteresis:
more resilient but irreversible

ToxCast dataset: *39 ↑EGF-signaling; some also ↓TGF-beta signaling*

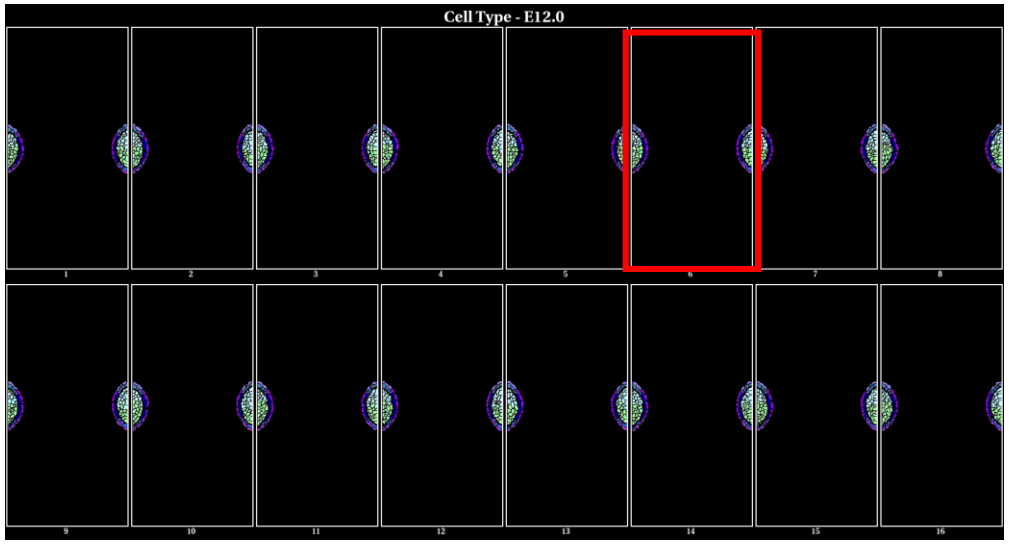
ChemicalName	*EGFR_up (uM AC50)*	*TGFb1_down (uM AC50)*	*STM (uM TI)*	*ToxRefDB (low)*
Carbaryl	0.07	1000.00	2.92	POS
Captafol	1.02	3.76	0.35	POS
Fipronil	1.18	1000.00	66.01	POS
Fluazinam	2.39	2.48	10.75	POS
Thiram	4.45	6.95	8.26	POS
Linuron	10.91	1000.00	30.94	POS
Maneb	0.01	1000.00	NEG	POS
Propoxur	1.67	1000.00	NEG	POS
Captan	4.59	7.15	NEG	POS
Bendiocarb	8.75	1000.00	NEG	POS
Raloxifene hydrochloride	12.40	15.94	NEG	POS
Tri-allate	19.19	x	NEG	POS
Triflumizole	32.71	19.88	NEG	POS
Butachlor	32.71	17.85	NEG	POS
Rotenone	0.82	1000.00	0.05	NEG
Zoxamide	14.22	17.37	16.13	NEG
Diuron	16.51	1000.00	68.06	NEG
Forchlorfenuron	0.02	1000.00	NEG	NEG
Azamethiphos	0.89	1000.00	NEG	NEG
Methylene bis(thiocyanate)	1.14	5.93	NEG	NEG
2-(Thiocyanomethylthio)benzothiazole	2.28	6.48	NEG	NEG
Methyl isothiocyanate	4.60	1000.00	NEG	NEG
Bromacil	20.50	1000.00	NEG	NEG
Diphenylamine	32.71	5.95	NEG	NEG
TNP-470	7.78	3.97	0.02	x
PharmaGSID_48511	12.19	11.22	0.02	x
4-Pentylaniline	0.00	x	NEG	x
Monobutyl phthalate	0.01	1000.00	NEG	x
Estrone	0.03	1000.00	NEG	x
SAR102779	0.05	12.95	NEG	x
Niclosamide	0.58	1000.00	NEG	x
CP-457920	3.50	1000.00	NEG	x
Perfluoroundecanoic acid	6.81	4.76	NEG	x
1,2-Benzisothiazolin-3-one	8.22	11.91	NEG	x
SB243213A	10.24	x	NEG	x
Phenolphthalein	16.26	x	NEG	x
FR167356	17.65	1000.00	NEG	x
SB281832	34.72	1000.00	NEG	x
p,p'-DDT	38.17	x	NEG	x

ChemicalName	*EGFR_up (uM AC50)*	*TGFb1_down (uM AC50)*	*STM (uM TI)*	*ToxRefDB (low)*
Fluazinam	2.39	2.48	10.75	POS
Captan	4.59	7.15	NEG	POS
Diuron	16.51	1000.00	68.06	NEG
FR167356	17.65	1000.00	NEG	x

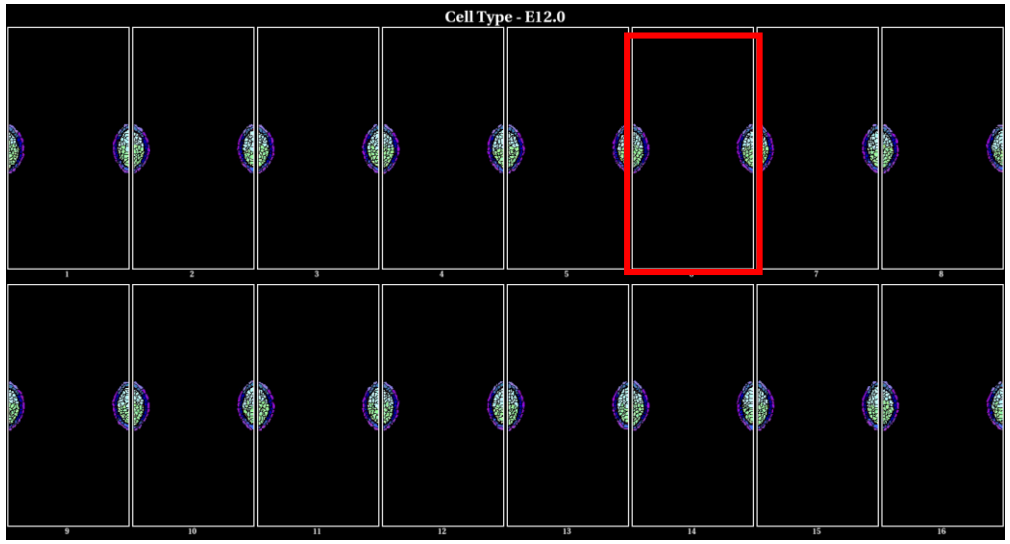


In silico **dose-response:** *translating ↑EGFR conc. profile into a critical dose*

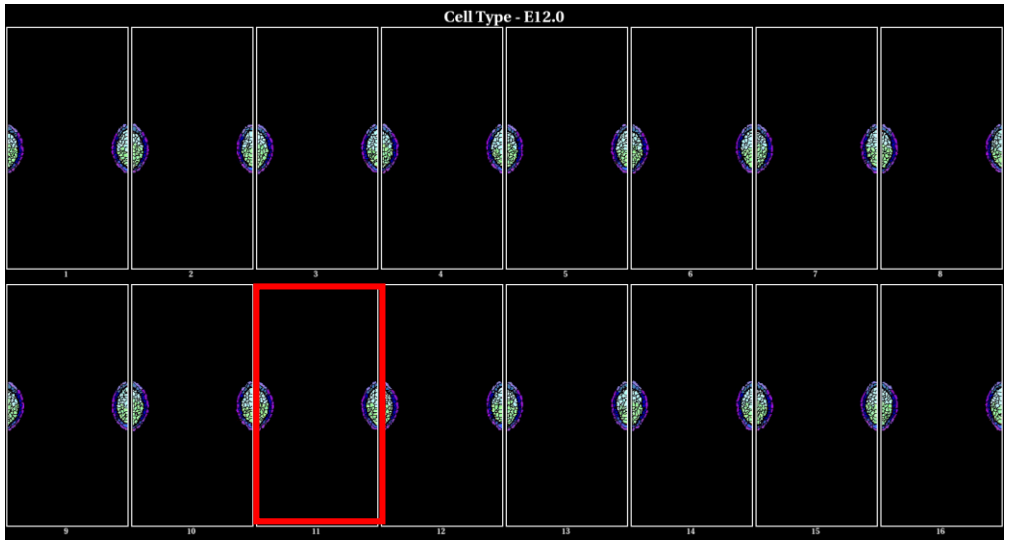
Fluazinam



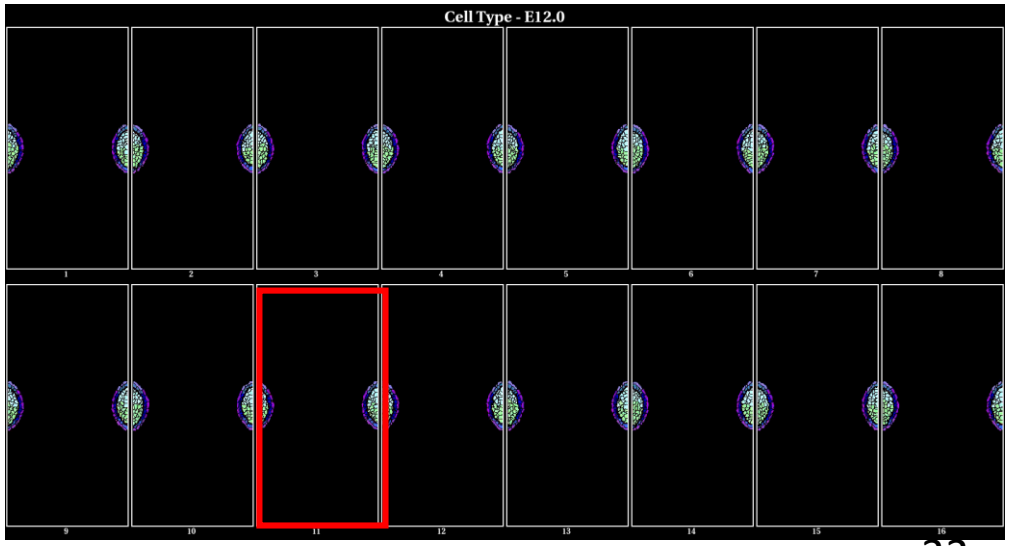
Captan



Diuron

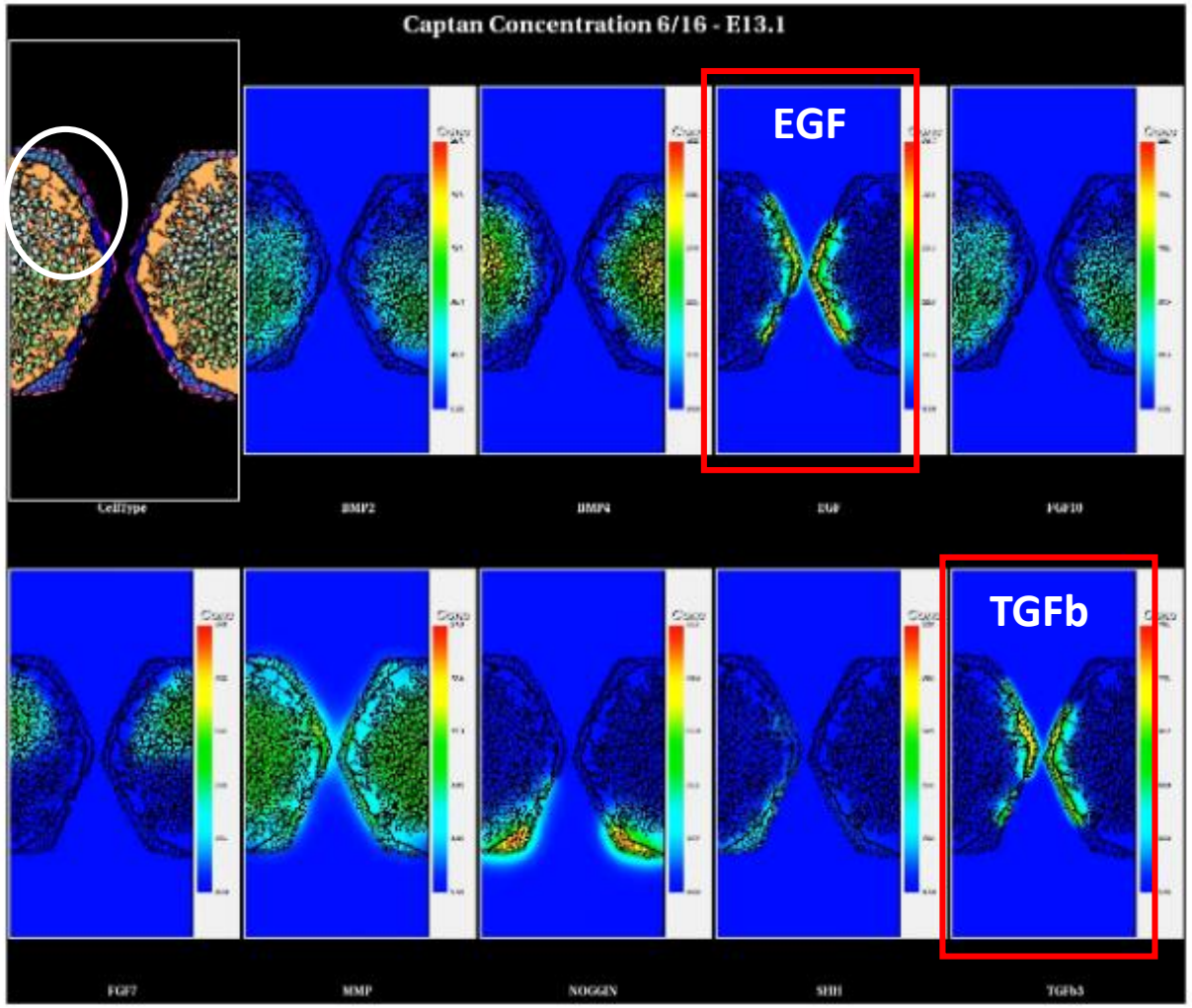


FR167356

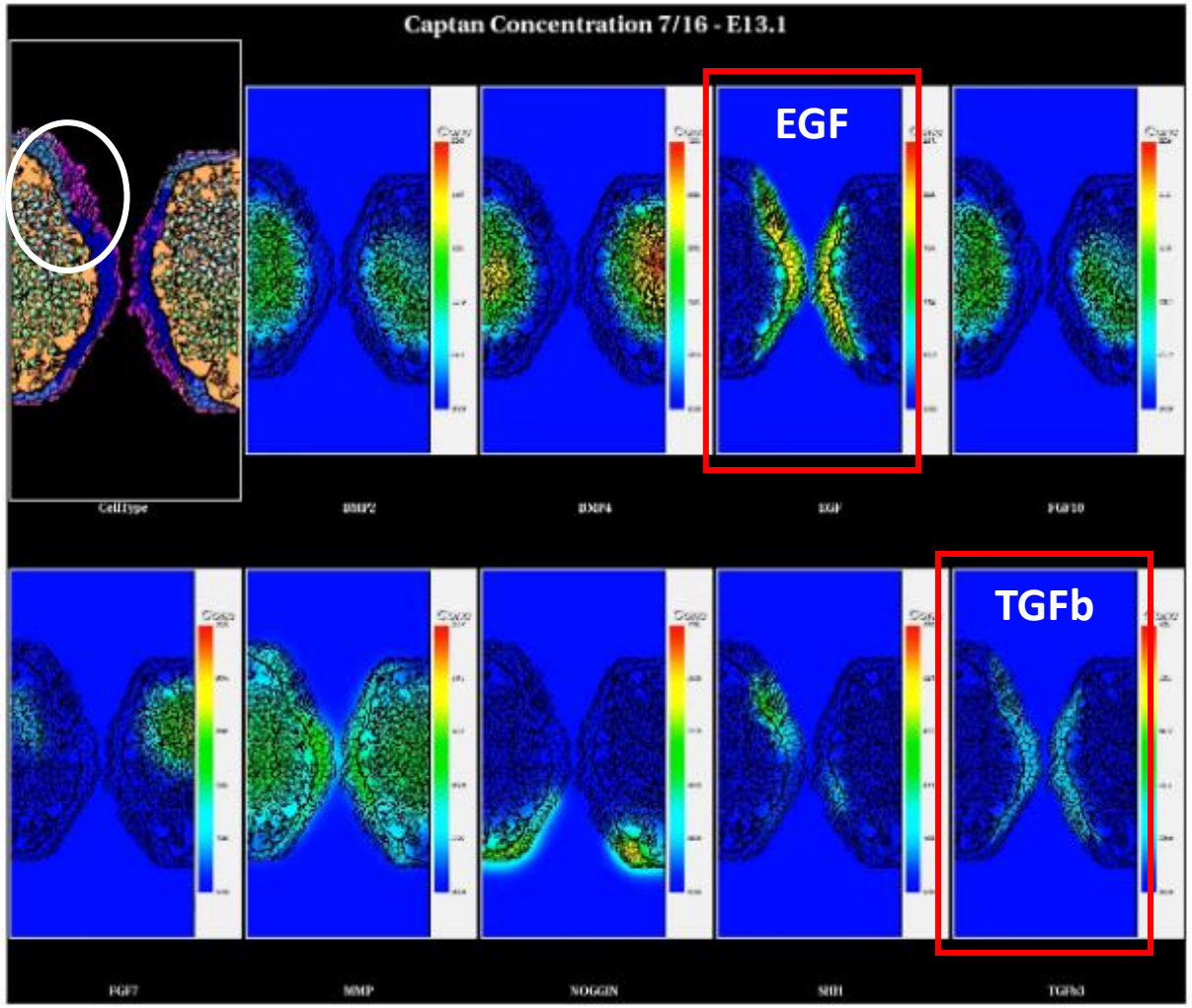


Pathogenesis: *simulating the prefusion alterations*

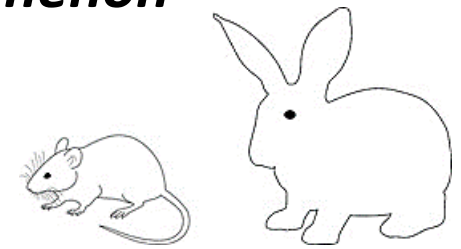
pre-critical dose



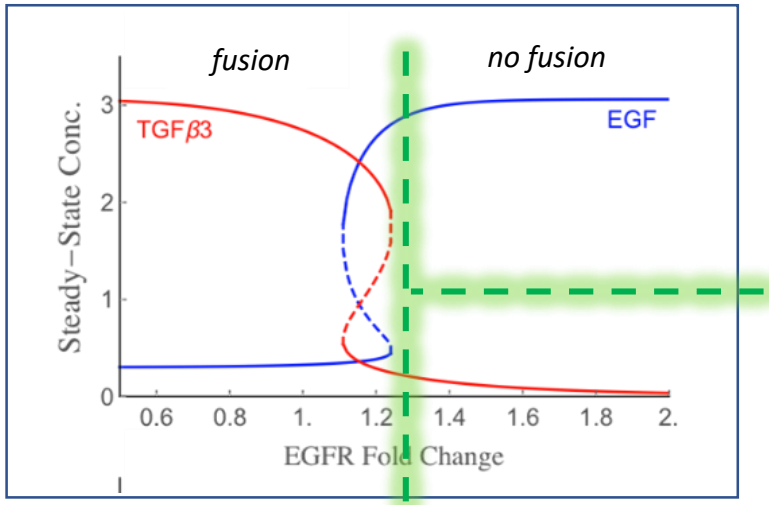
post-critical dose



Predictive model: *modeling the critical phenomenon*

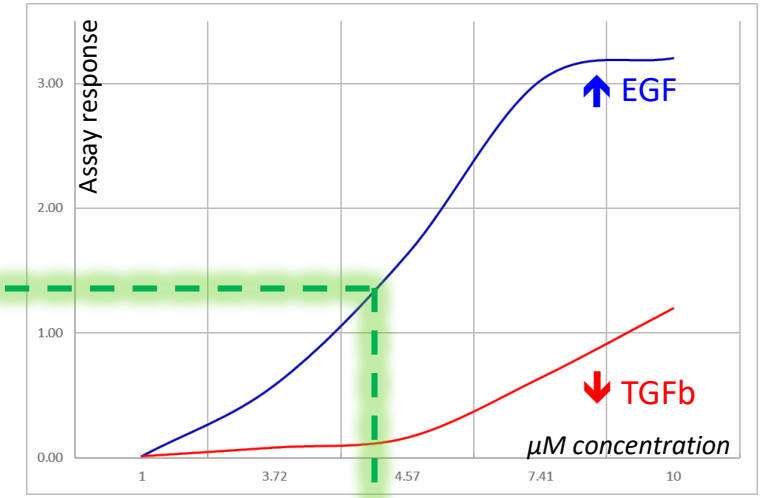


INPUT: switch dynamics



tipping point predicted by computational dynamics (hysteresis switch)

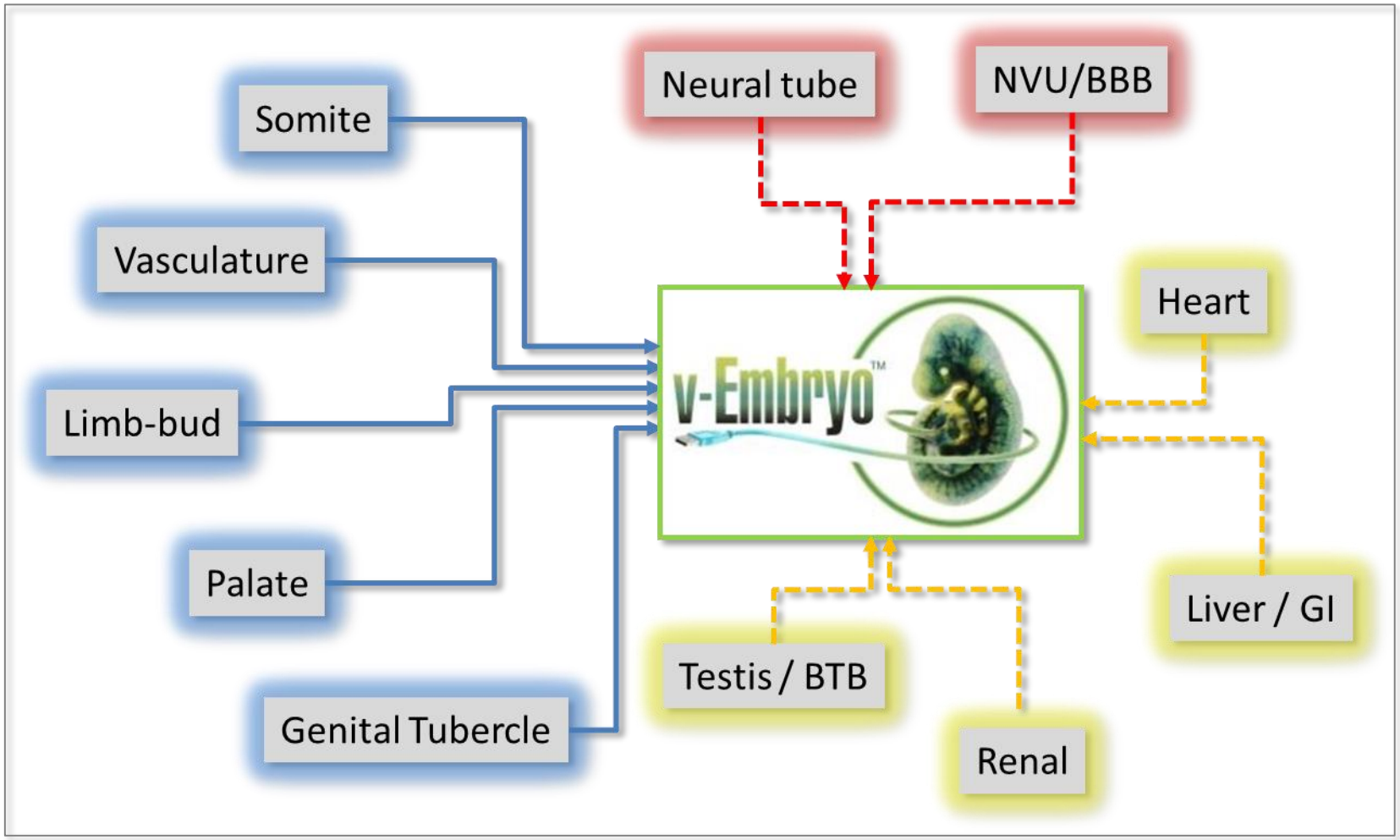
Captan in ToxCast



OUTPUT: tipping point mapped to concentration response (4 μM)

Captan in ToxRefDB
NOAEL = 10 mg/kg/day
LOAEL = 30 mg/kg/day

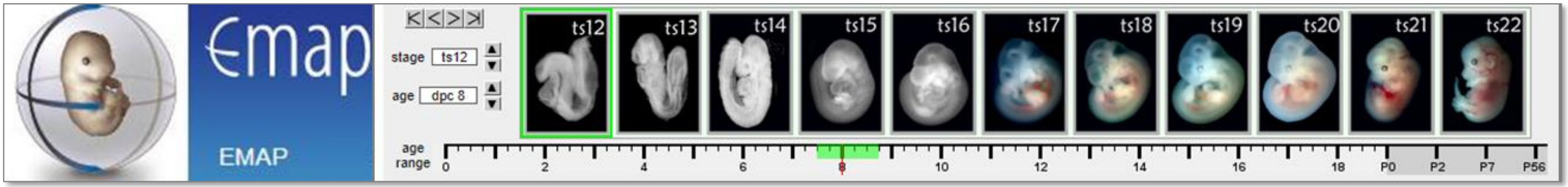
human HTTK model
2.39 mg/kg/day would achieve a steady state of 4 μM in fetal plasma



VTLS

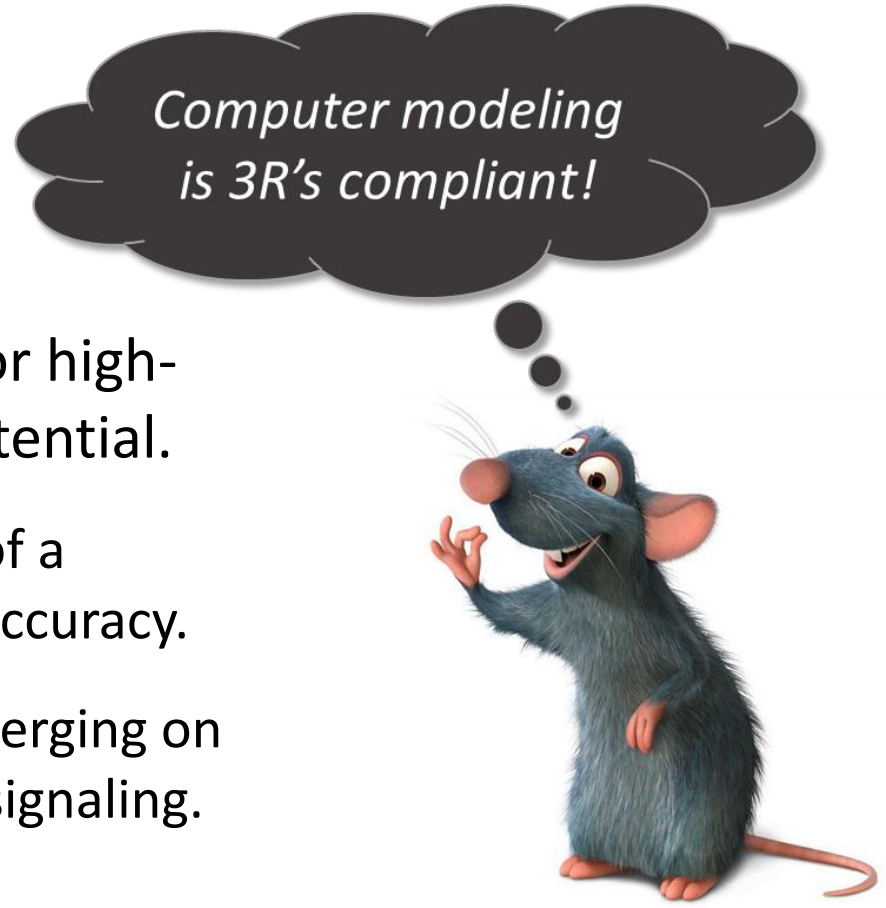
- *access to models & simulations*
- *VT-KB (knowledgebase)*
- *Literature mining*
- *tied to ToxCastDB*
- *high-performance computing*

vtls.epa.gov/



https://www.emouseatlas.org/emap/ema/home.php

Summary and Conclusions



1. Several new approach methods (NAMs) are available for high-throughput screening chemical inventories for DevTox potential.

- STM assay in ToxCast gives an exposure-based readout of a chemical's DevTox hazard potential with 84% balanced accuracy.
- Assay sensitivity predicted high for kinase signaling converging on FoxO signaling but weak for estrogenic (ESR1) and G(q) signaling.

2. Cell ABMs recapitulate morphogenesis cell-by-cell and interaction-by-interaction as an embryonic system advances in time.

- Computer models simulate key events in AOPs to render mechanistic predictions and critical phenomena for DevTox.

https://www.pinterest.com/courtney1882/disney-ratatouille/

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