Computational Embryology:

Agent-Based Modeling and Simulation of External Genital Development

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NIH-NIDDK/AUA Workshop
"Congenital Anomalies of External Genitalia "
Linthicum MD, May 31-June1, 2018

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Scoping the Problem

Chemical regulation under the *Frank R. Lautenberg Chemical Safety for the 21st Century Act of 2016* (‘amended TSCA’) requires rapid affirmation of ‘low’ and ‘high’ priority substances based on unreasonable risk to vulnerable subpopulations and lifestages.

New approach methods (NAMs) based on HTS assays, complex *in vitro* systems, and *in silico* models can lead us in “*decoding the toxicological blueprint of active substances that interact with living systems*” [Sturla et al. 2014].

Anatomical homeostasis in a self-regulating Virtual Embryo

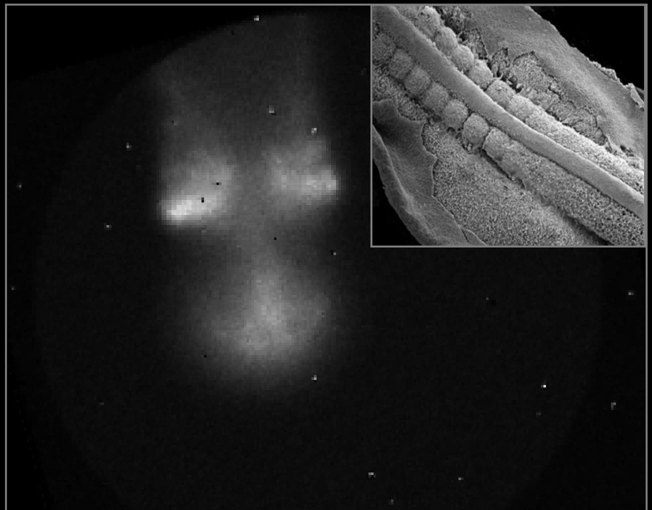
SOURCE: Andersen, Newman and Otter (2006) Am. Assoc. Artif. Intel.

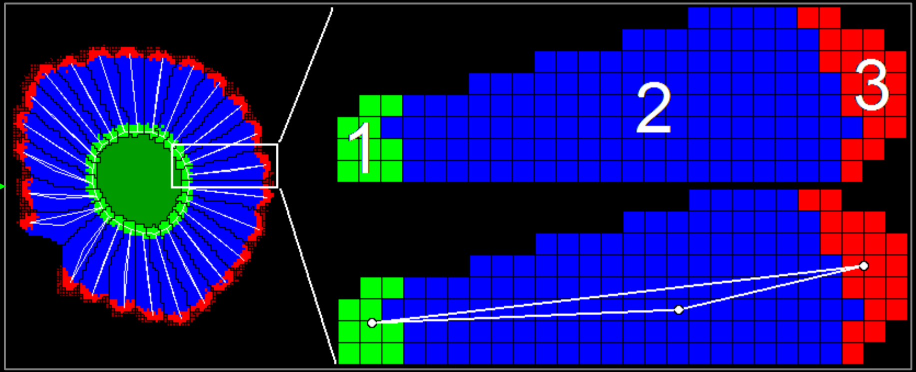
Agent-Based Models (ABMs):

in silico toxicology
is 3R's compliant!

- virtually reconstruct a morphogenetic field cell-by-cell and interaction-by-interaction (*compucell3d.org*)
- execute simulations that self-organize cells into higher-levels of biological structure and function (*emergent phenotypes*)
- simulate perturbations in the system - dose or stage response, critical pathways, non-chemical stressors, etc (*dynamics*)
- probabilistic rendering of where, when and how a developmental defect might occur (*mechanistic interpretation*)

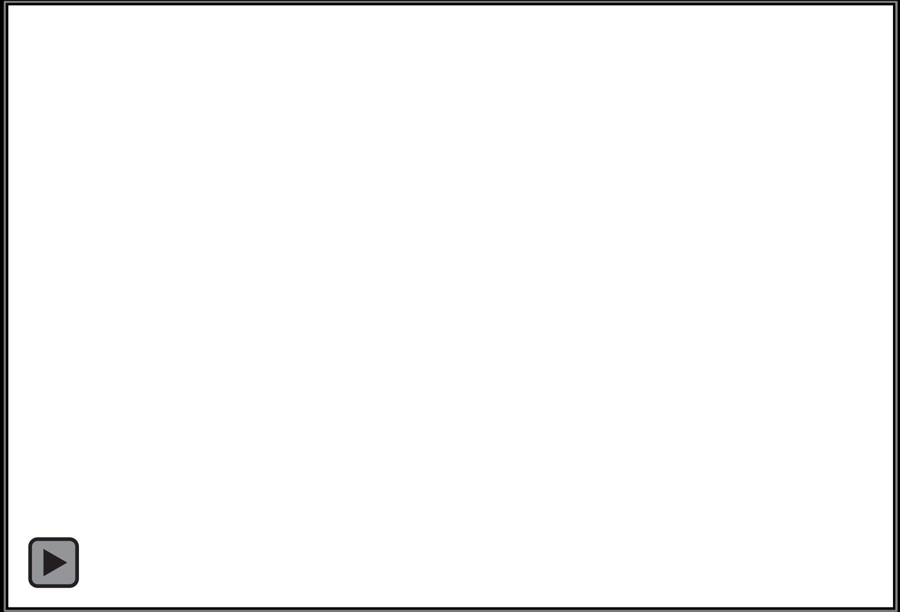
Modeling somite development

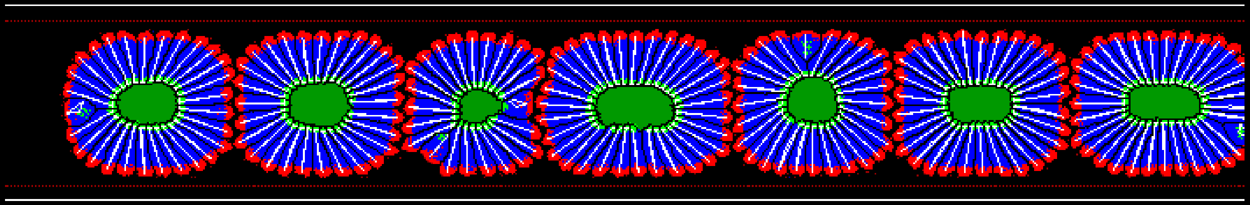

Hes1-EGFP time-lapse (3h)
Masamizu et al. 2006



Differential cell adhesion

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

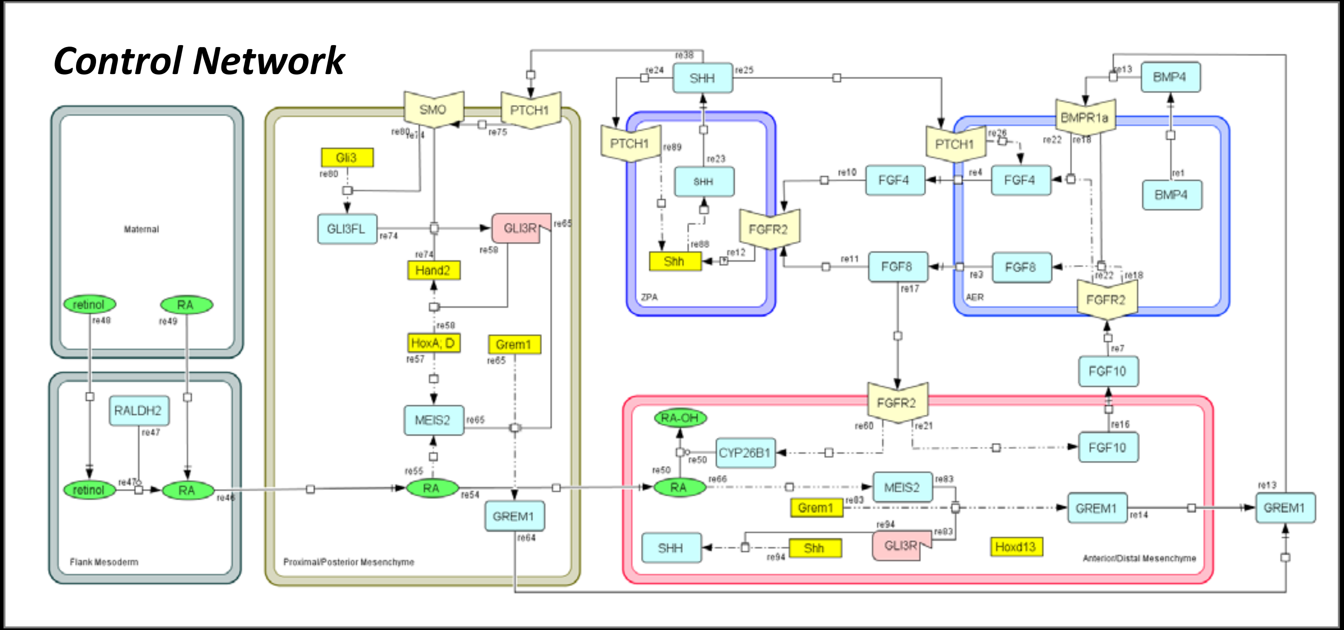


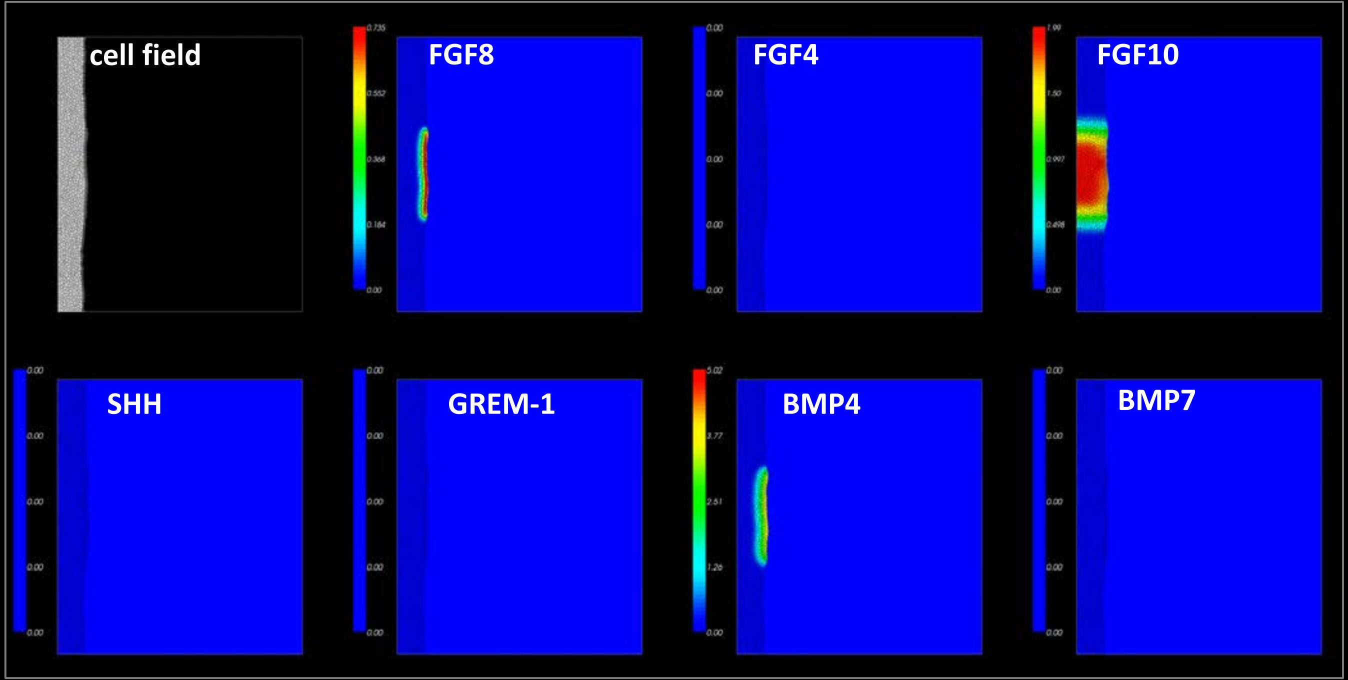


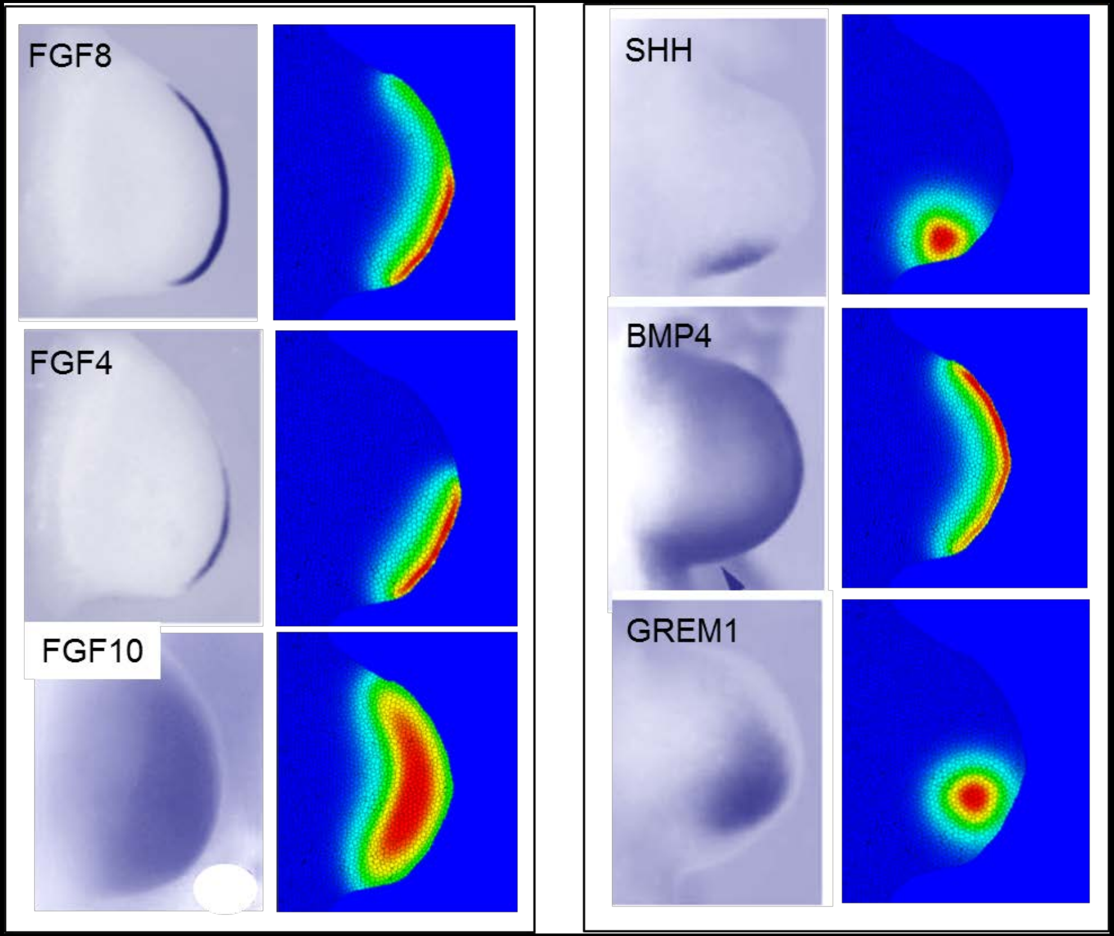
- *FGF8 wavefront restores sequentiality*
- *oscillatory clock improves regularity*

SOURCE: Dias et al. (2014) Science

Limb-bud outgrowth



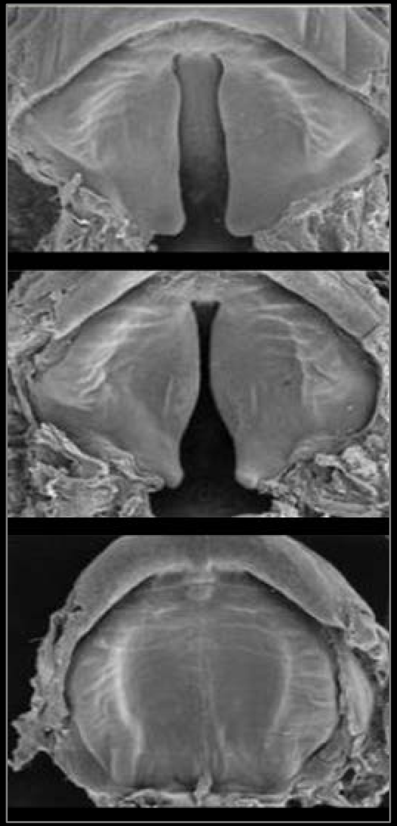




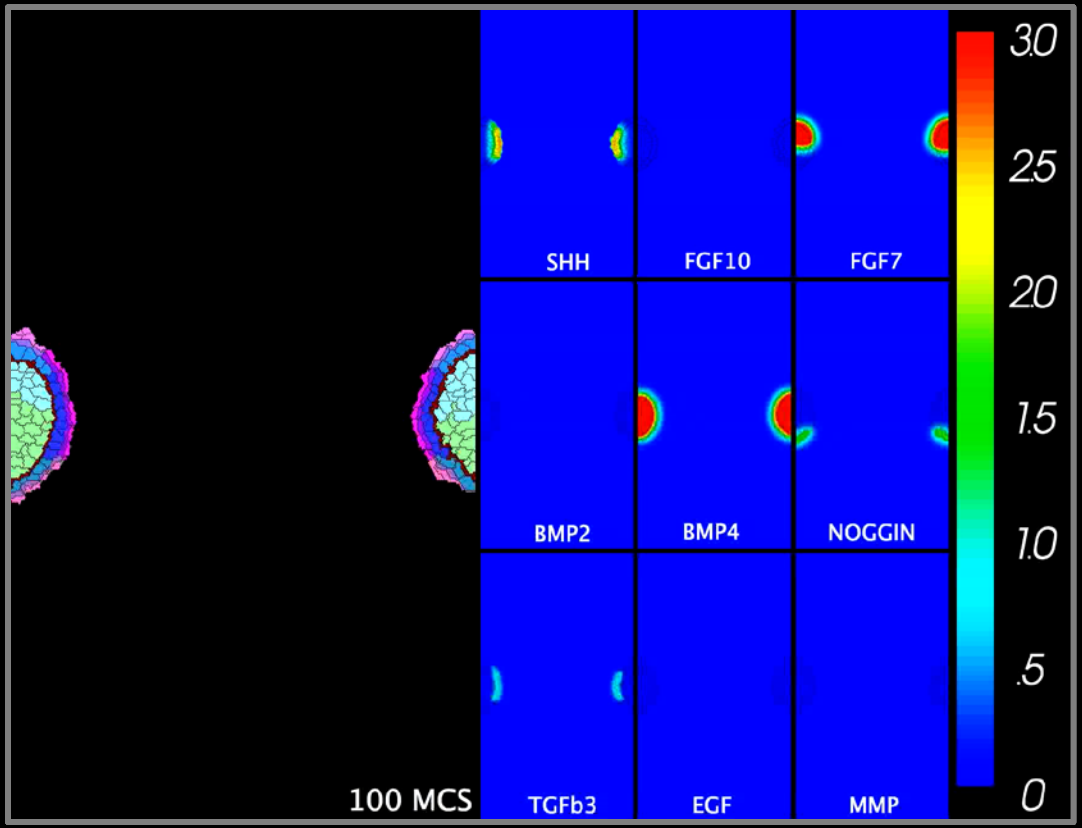
Palatal closure: *driven by medial edge epithelium (MEE) seam breakdown*

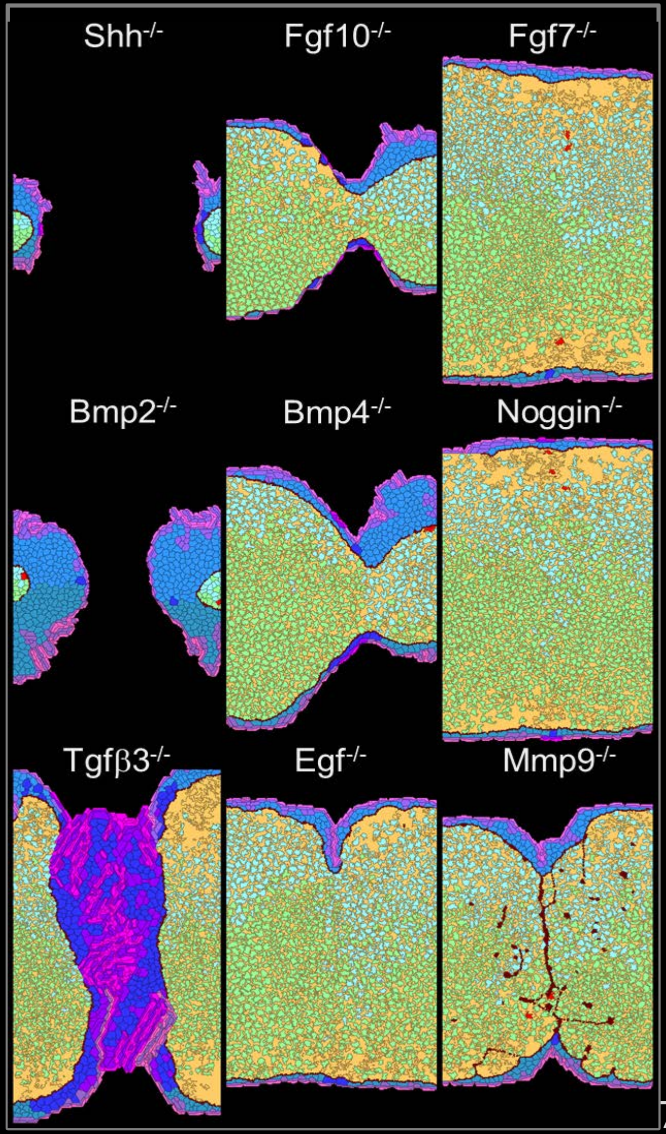
SEMs of human palate by K Sulik, UNC

Palatal fusion *in vivo*



Palatal fusion *in silico*

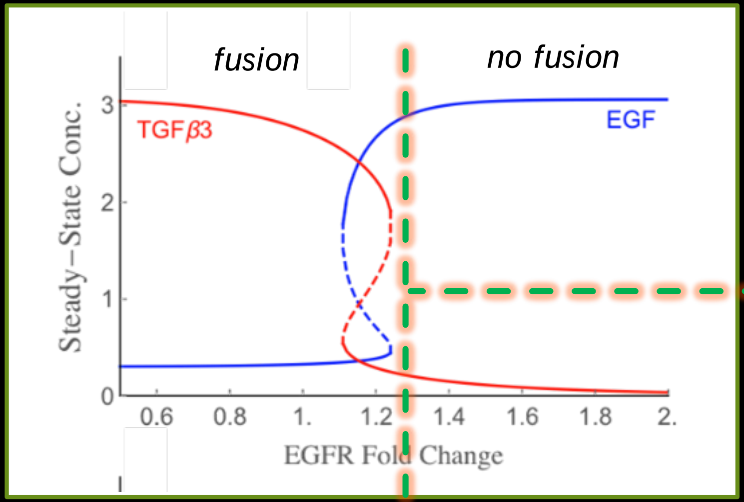




SOURCE: Hutson et al. (2017) Chem Res Toxicol

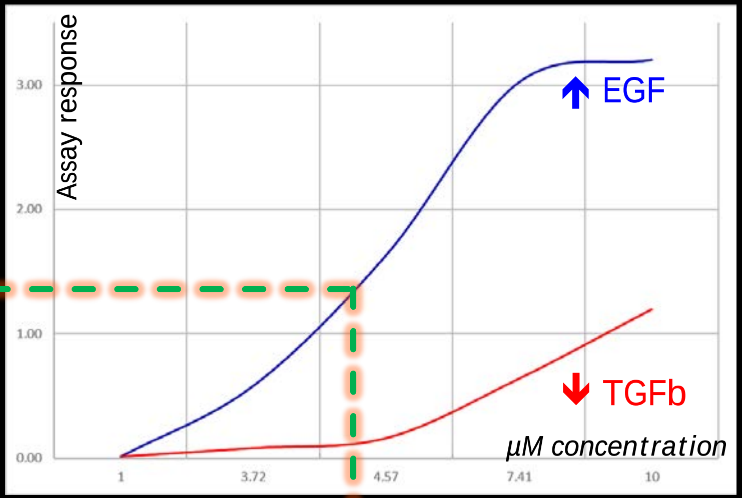
TGF-beta/EGF latch switch: *controls MEE breakdown*

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

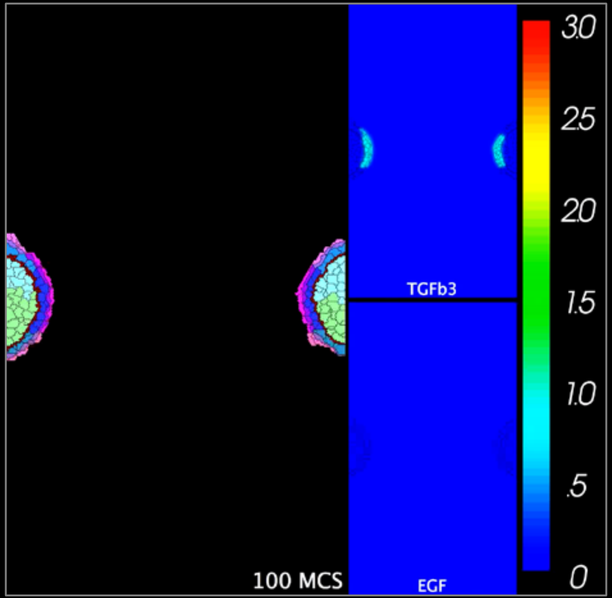
NOAEL = 10 mg/kg/day

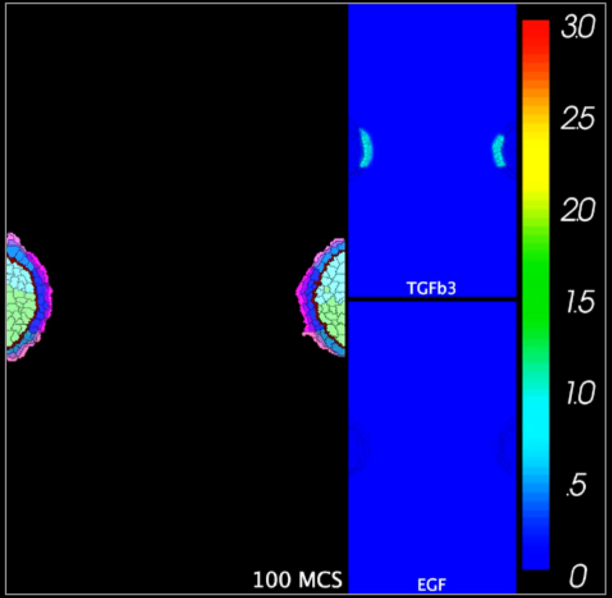
LOAEL = 30 mg/kg/day

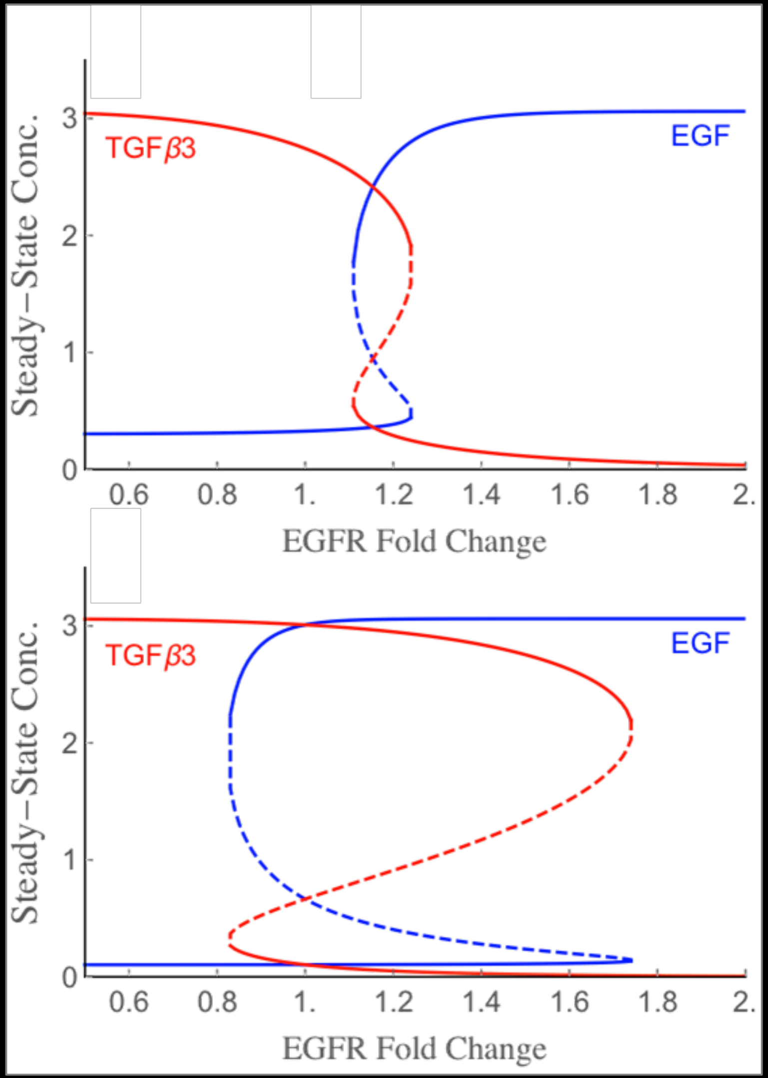
human HTTK model

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

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



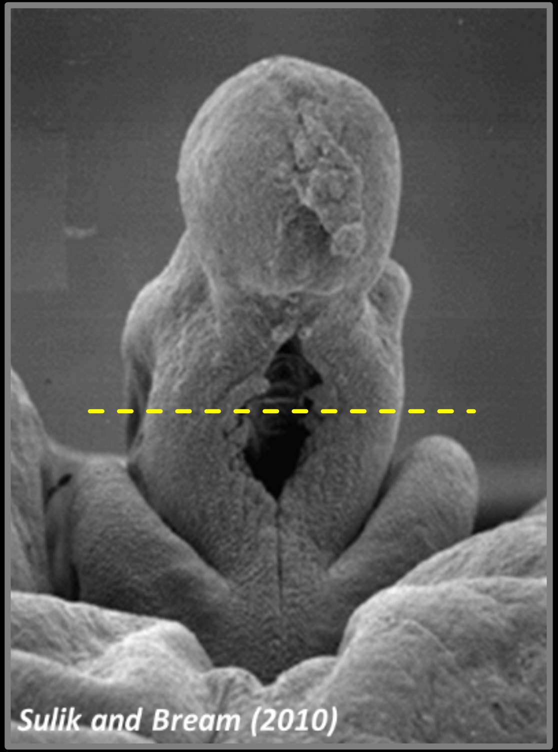


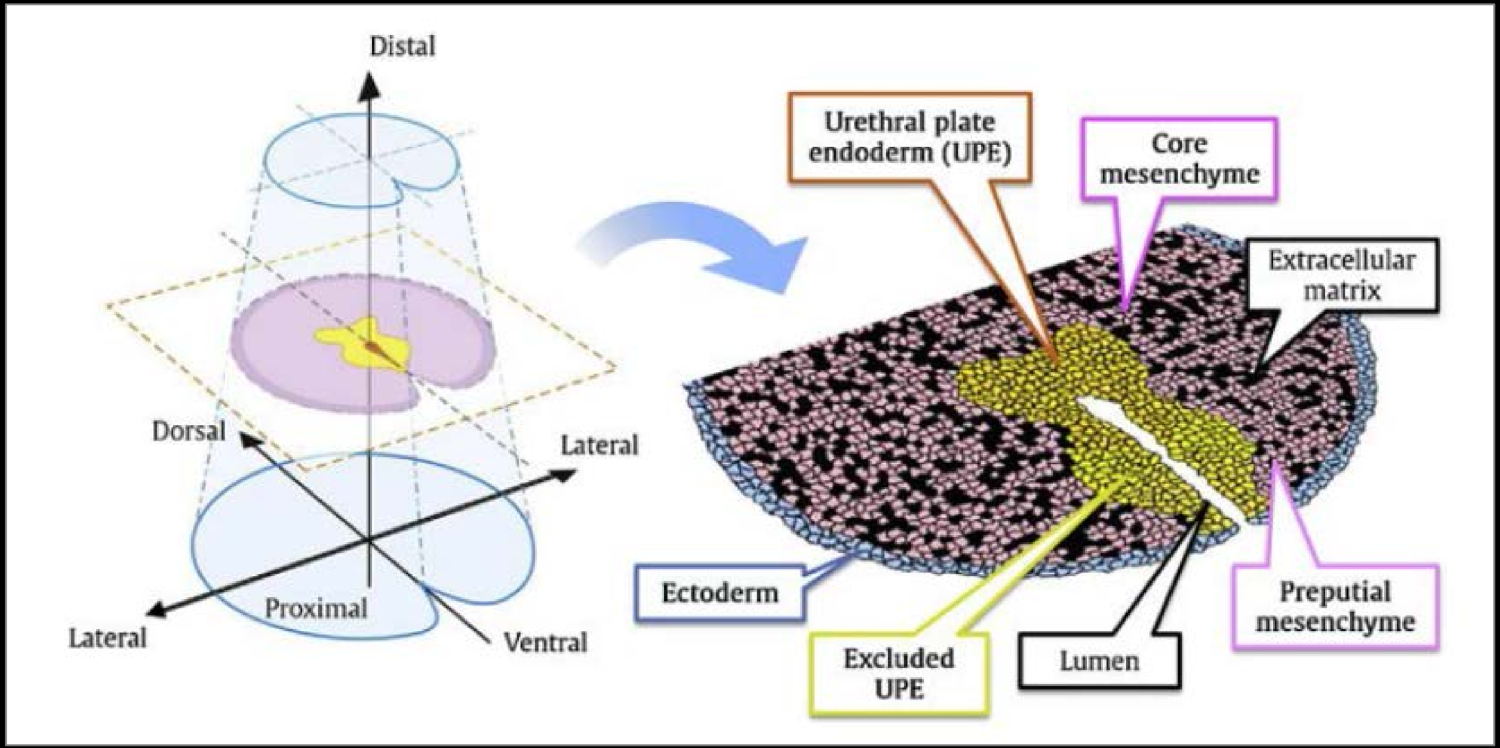


Narrow hysteresis:
less resilient but reversible

Broad hysteresis:
more resilient but irreversible

Genital tubercle: abstraction for ABM-simulated urethral closure



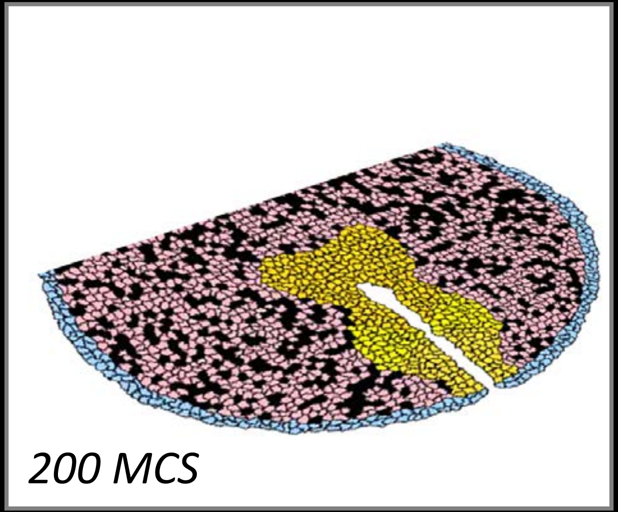


GT development modeled @ 1000 MCS/day of gestation (mouse):

- *E13.5 - urethral plate endoderm (UPE)*
- *E15.5 - ventral elevation of urethral folds*
- *E17.5 - urethral fusion and septation*

SOURCE: Leung et al. (2016) Reproductive Toxicology

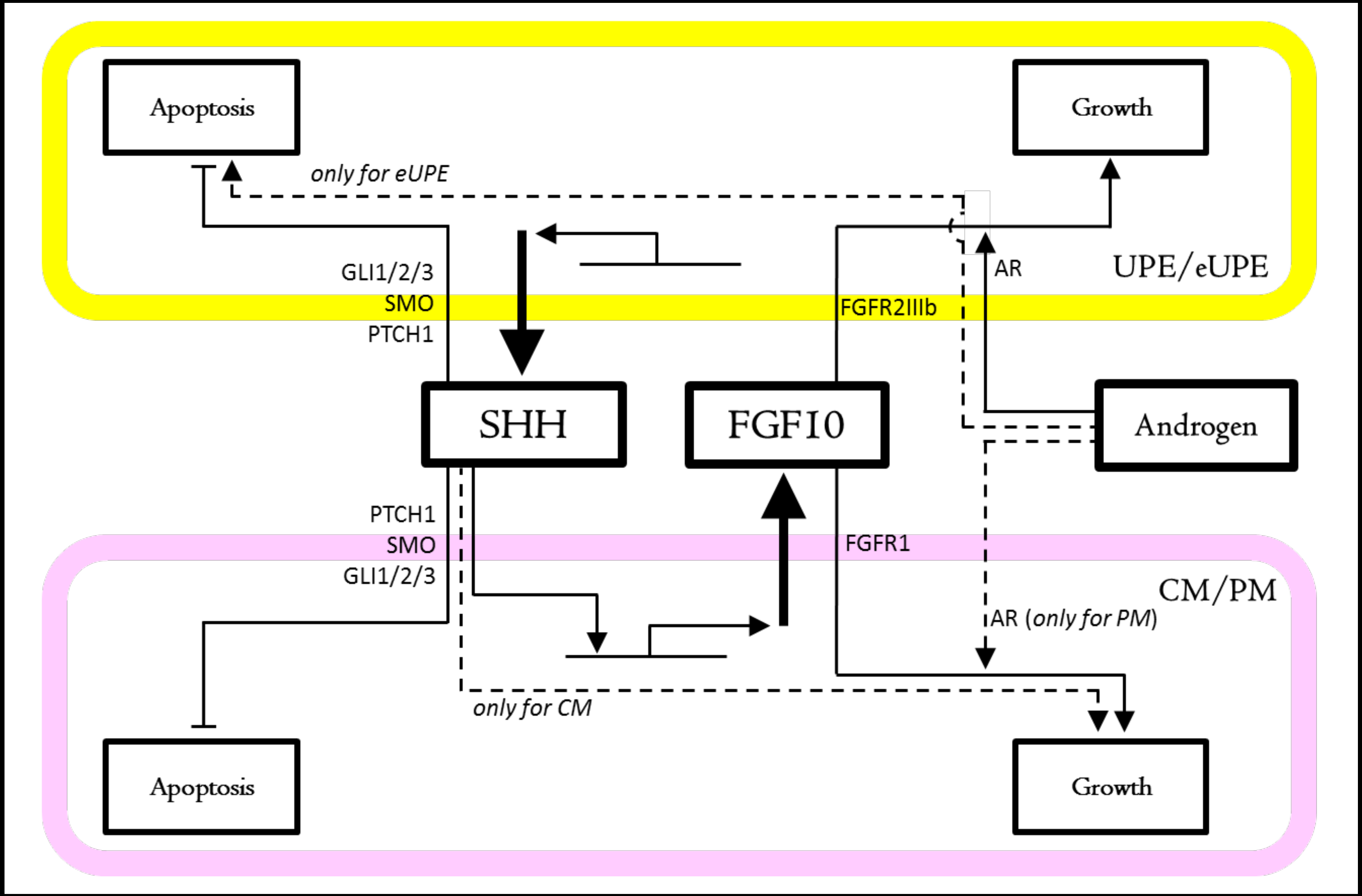
Control network: minimal requirements for dual-reciprocal signaling



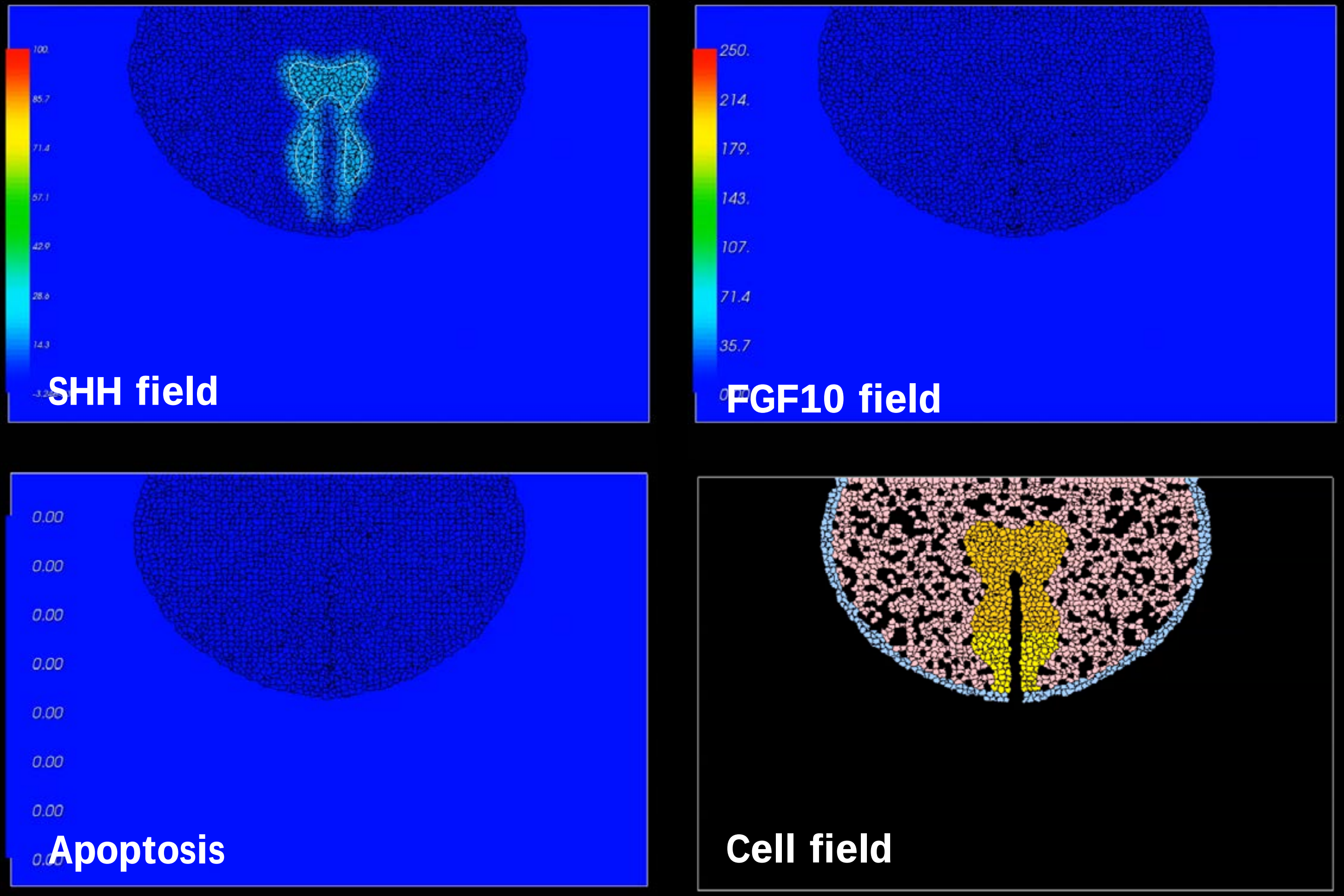
Biokinetic gradients:

FGF10 – simple diffusion
SHH – transcytosis (1/8 rate)
An – latched @2000 MCS (E15.5)

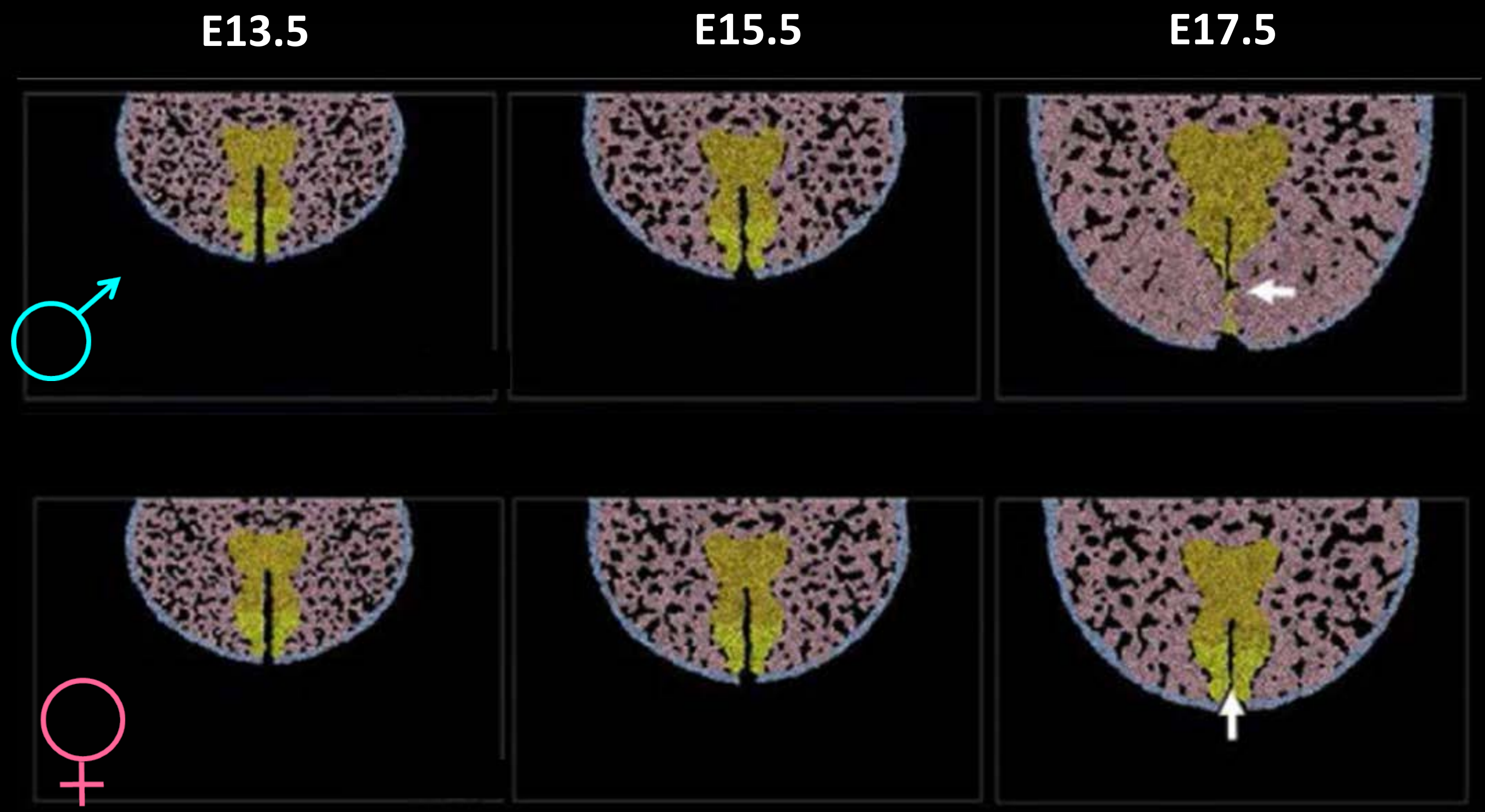
Note – model assumes androgen delivery by vascular perfusion

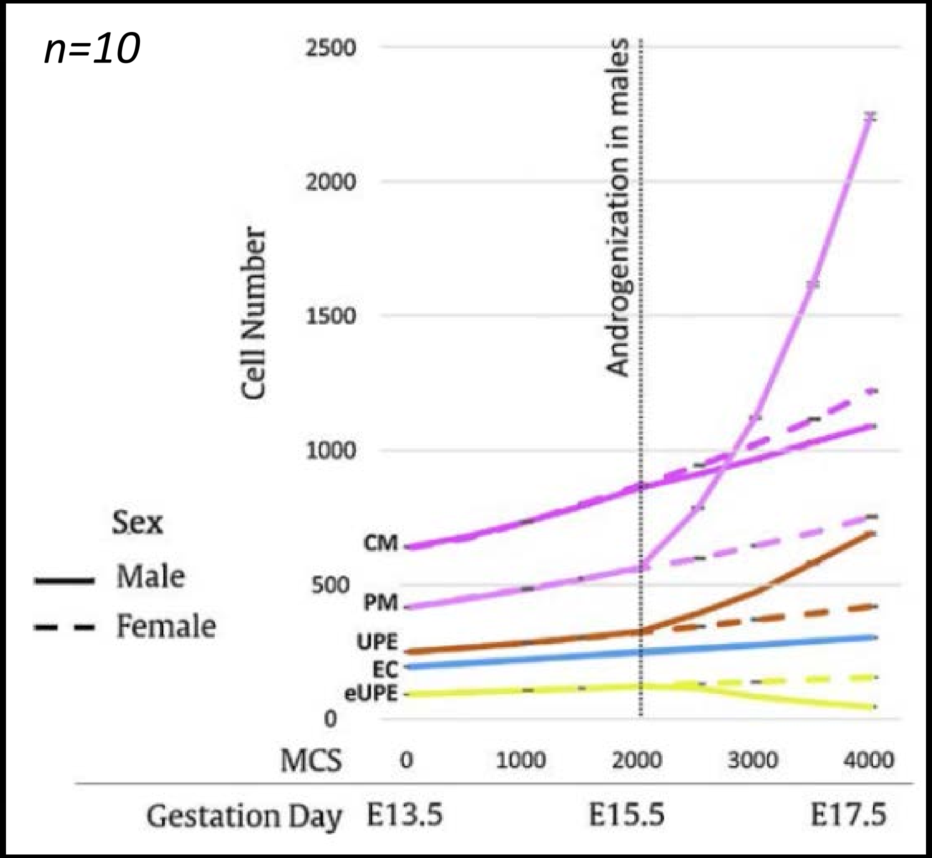


Androgenization: executed @ 2000 MCS as a continuous field → AR activation



Sexual dimorphism

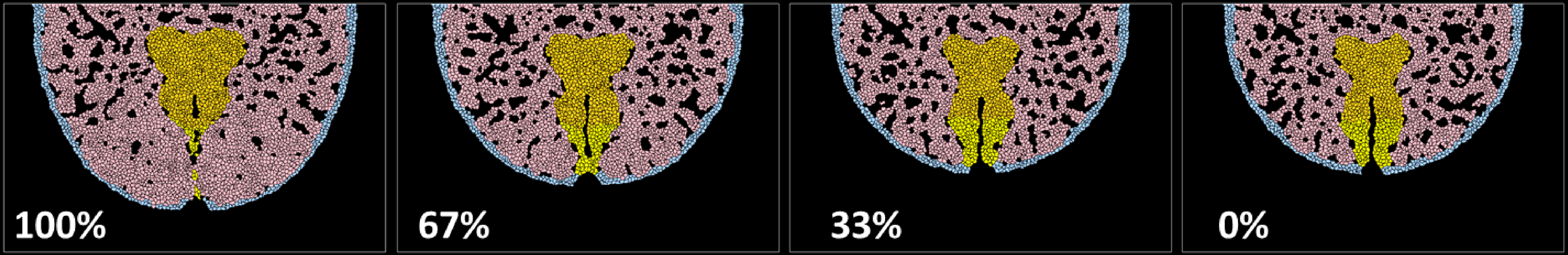


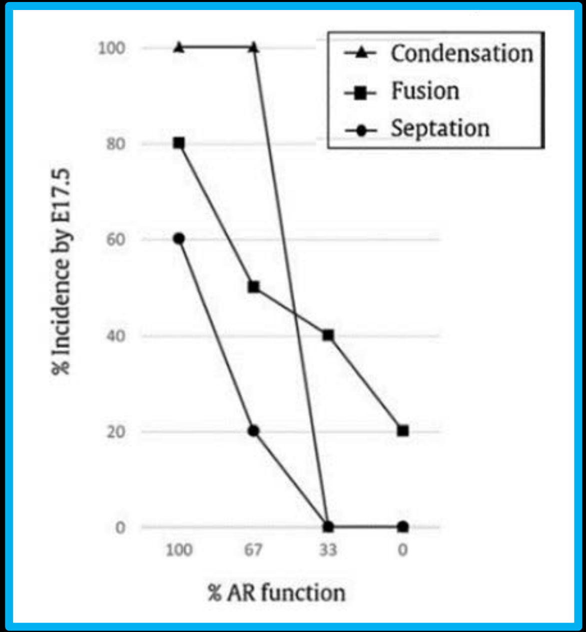


Urethral closure is an emergent property, driven by:

- *urethral plate endoderm (apposition, contact, fusion and centralization)*
- *preputial mesenchyme (proliferation, condensation, migration)*
- *androgen-dependent effects mediated by FGFR2-IIIb signaling*

Androgen disruption: *closure rates @4000 MCS* $\int androgen\ supply$

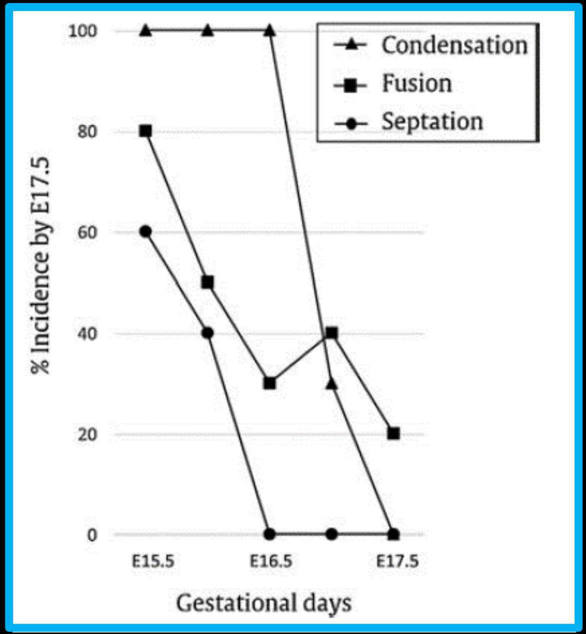




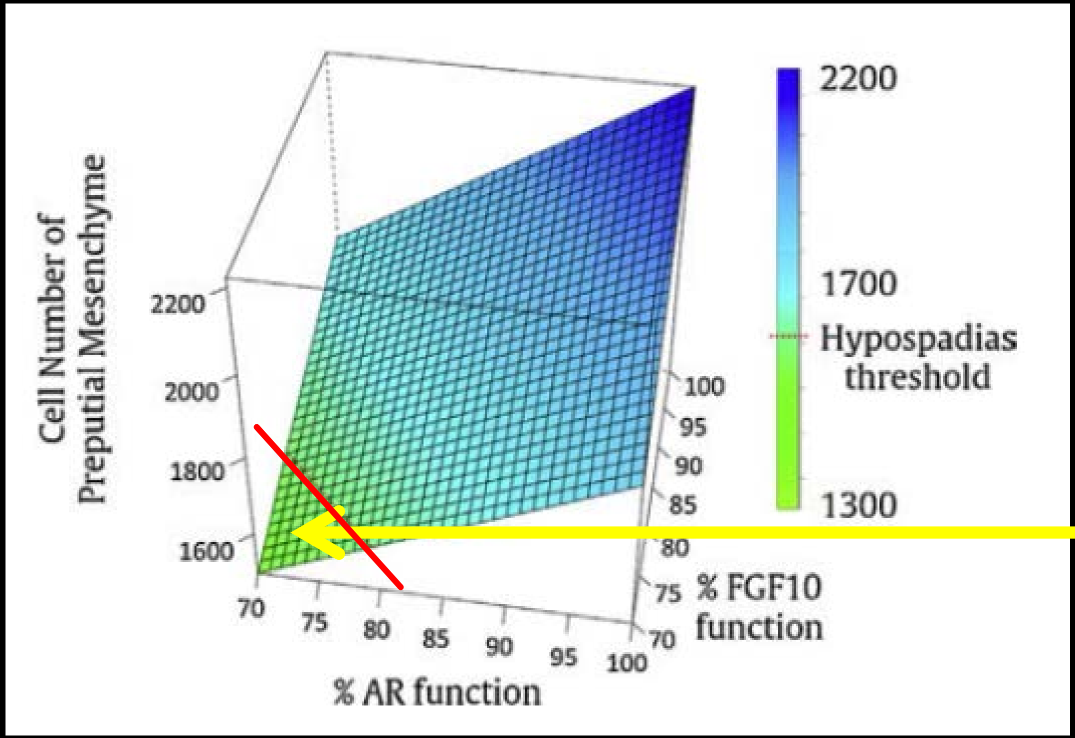
Closure indices (simulated, n=10)

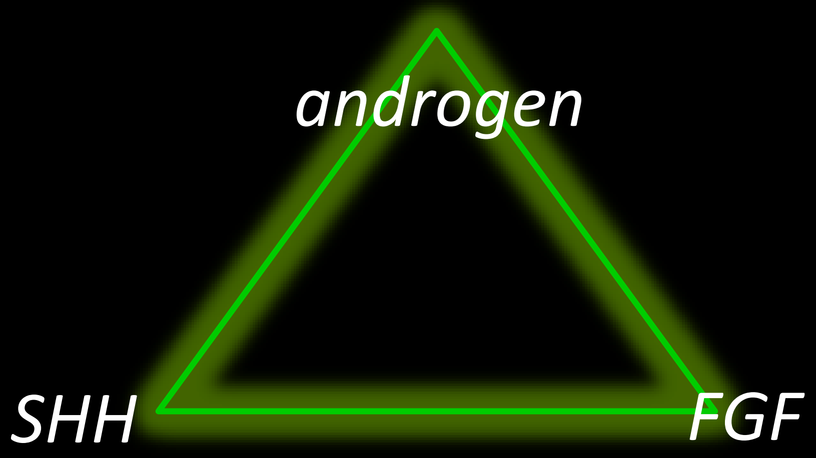
LEFT: androgen insufficiency

RIGHT: delayed virulization



Gene-environment interaction *(sensitivity analysis)*





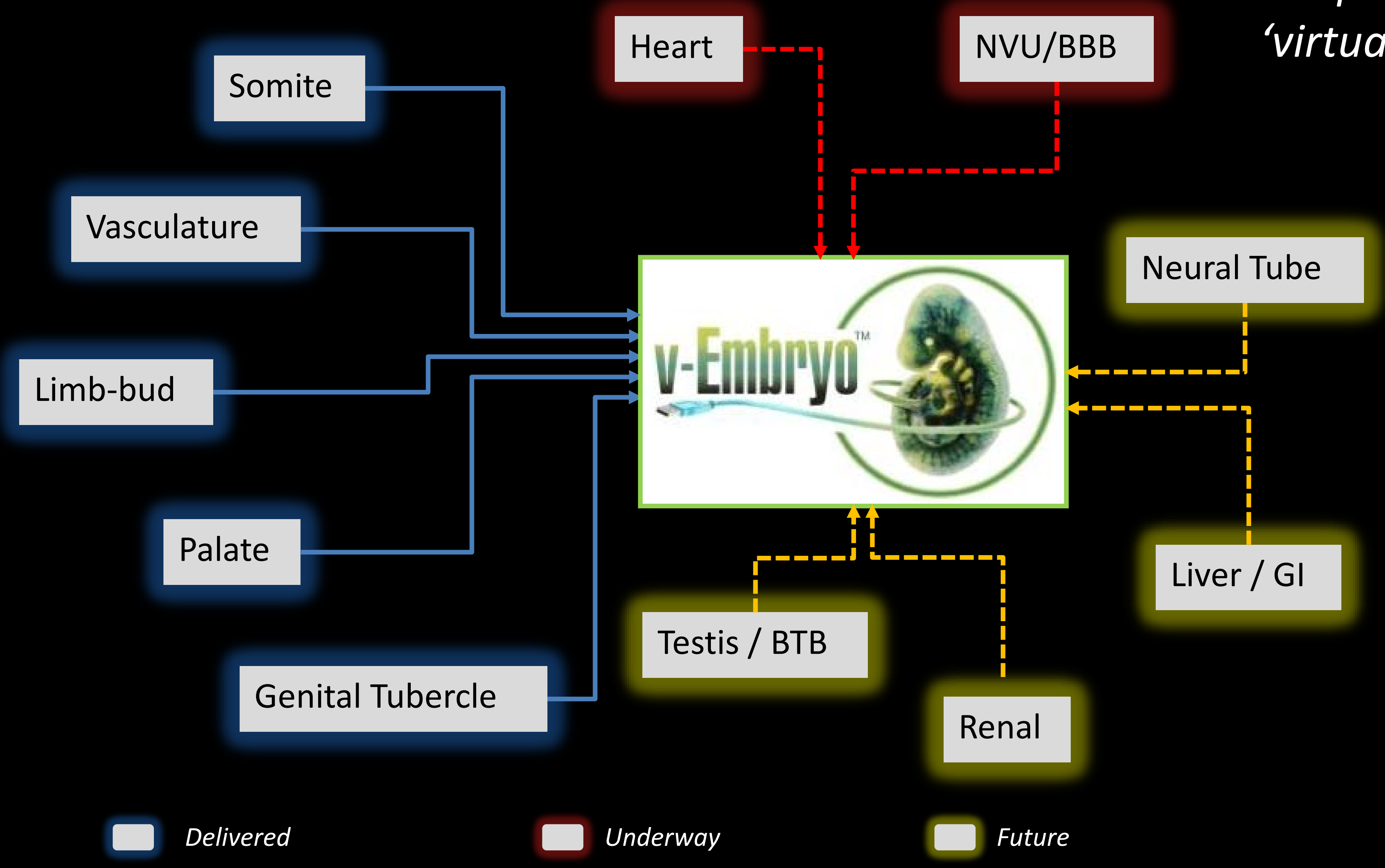
High risk for hypospadias

Multi-disturbance surface plot for individual risk factors:

- genetics (e.g., FGFR polymorphism)
- metabolism (e.g., SHH cholesterol-ification)
- environmental exposure (e.g., androgen disrupters)

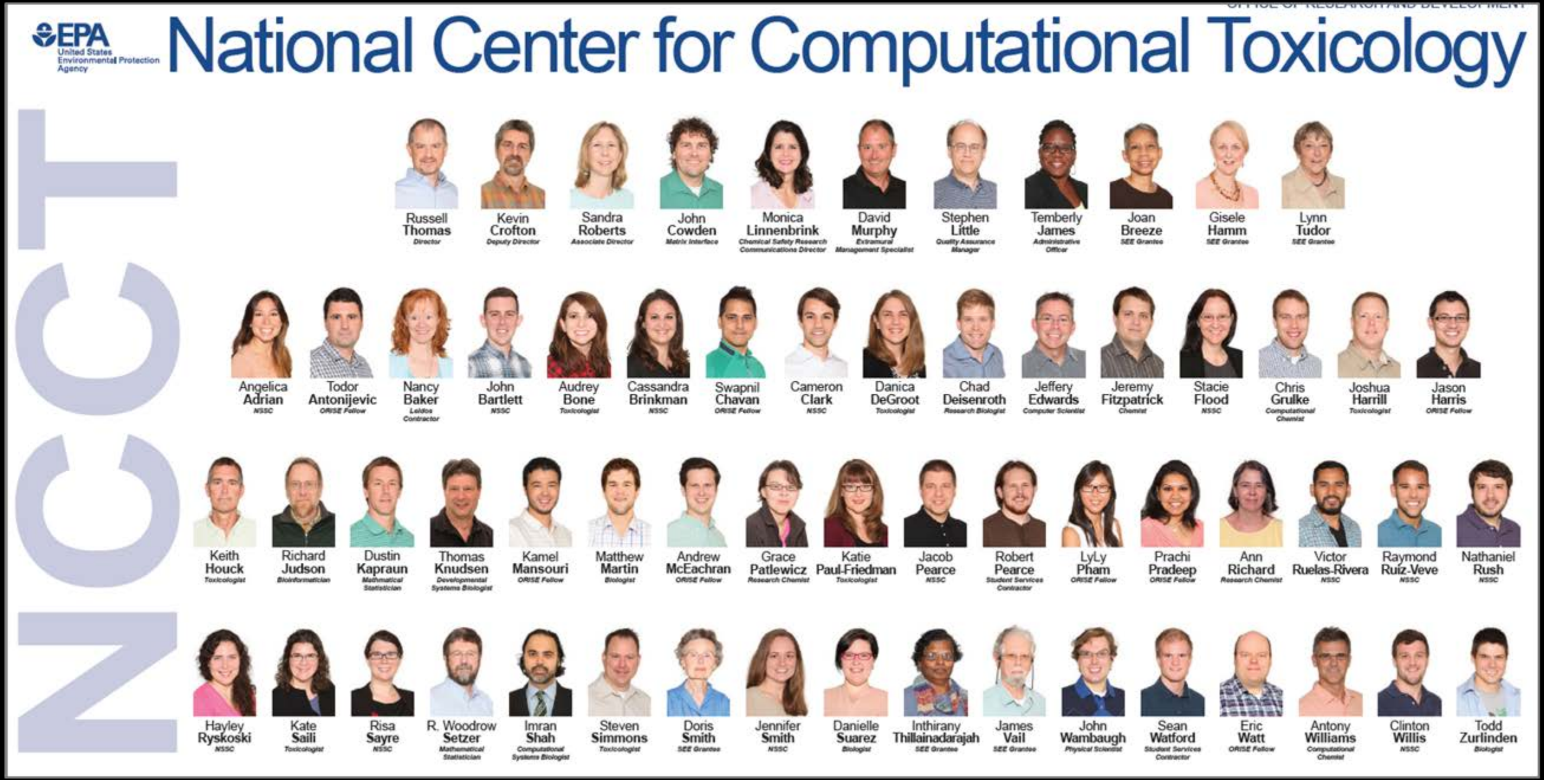
Grand Challenge:

a predictive
'virtual embryo'



Special Thanks

- Max Leung – NCCT (now CalEPA)
- Kate Saili – NCCT
- Todd Zurlinden – NCCT
- Nancy Baker – Leidos / NCCT
- Richard Spencer – ARA / EMVL
- James Glazier – Indiana U
- Sid Hunter – NHEERL / ISTD
- Kyle Grode – NHEERL (now Nikon)
- Andrew Schwab – NHEERL/ISTD
- Barbara Abbott – NHEERL/TAD
- Imran Shahe - NCCT
- Shane Hutson – Vanderbilt U (VPROMPT)
- Brian Johnson – U Wisconsin
- Aldert Piersma – RIVM, The Netherlands
- Nicole Kleinstreuer – NCCT (now NTP)
- George Daston – Procter & Gamble Co.
- Ashley Seifert – U Kentucky
- Martin Cohn - (U Florida)



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