

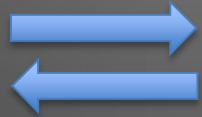
Transcriptional control of cell migration in the ascidian cardiogenic mesoderm

Lionel Christiaen
NYU Biology

KITP, UC Santa Barbara
August 19th 2011

The central dogma of systems biology

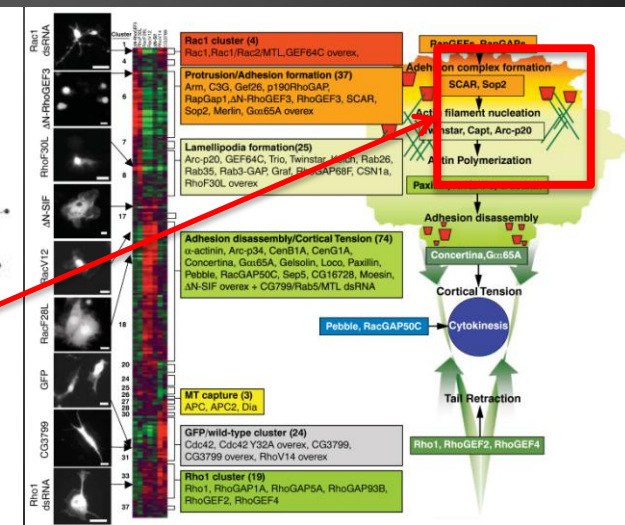
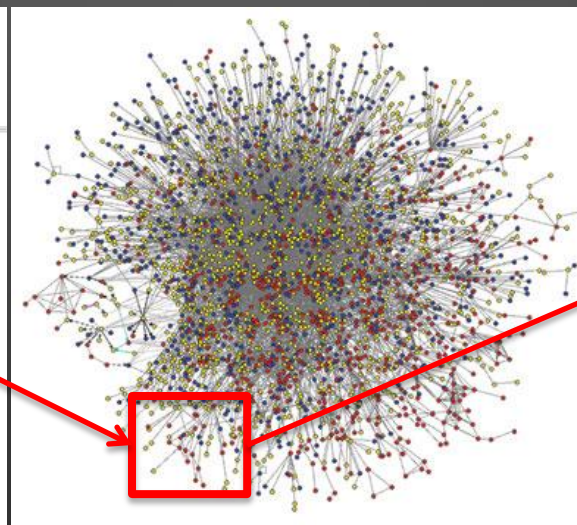
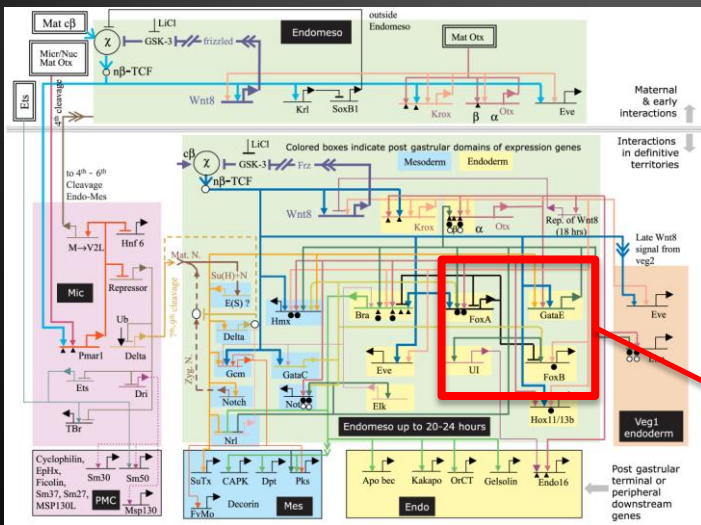
Gene
Regulatory
Network



Protein
Interaction
Network



Cellular
Activities
(Phenotypes)



Davidson *et al.* (2002) *Science*

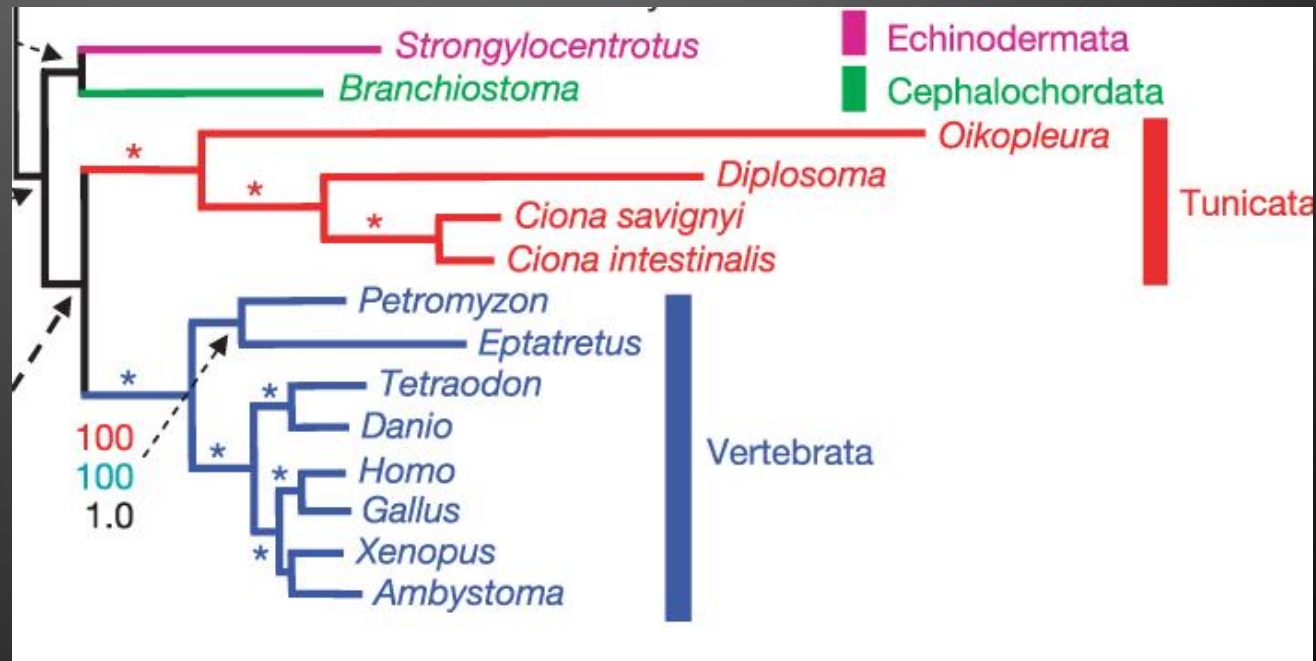
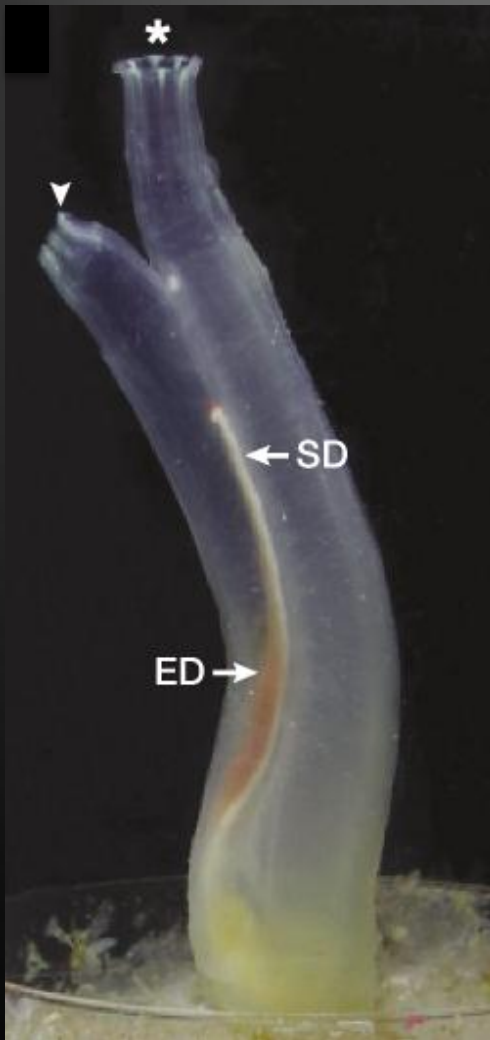
Li *et al.* (2004) *Science*

Bakal *et al.* (2007) *Science*

Talk outline

- Introduction to early development of the cardiogenic lineage in *Ciona*
- Transcriptional control and regulated effectors of TVC migration
- Atrial Siphon Muscle precursors re-activate a muscle program and turn on their own migration effectors
- Insights into the modular organization of the network from comparison with the Oral Siphon Muscles

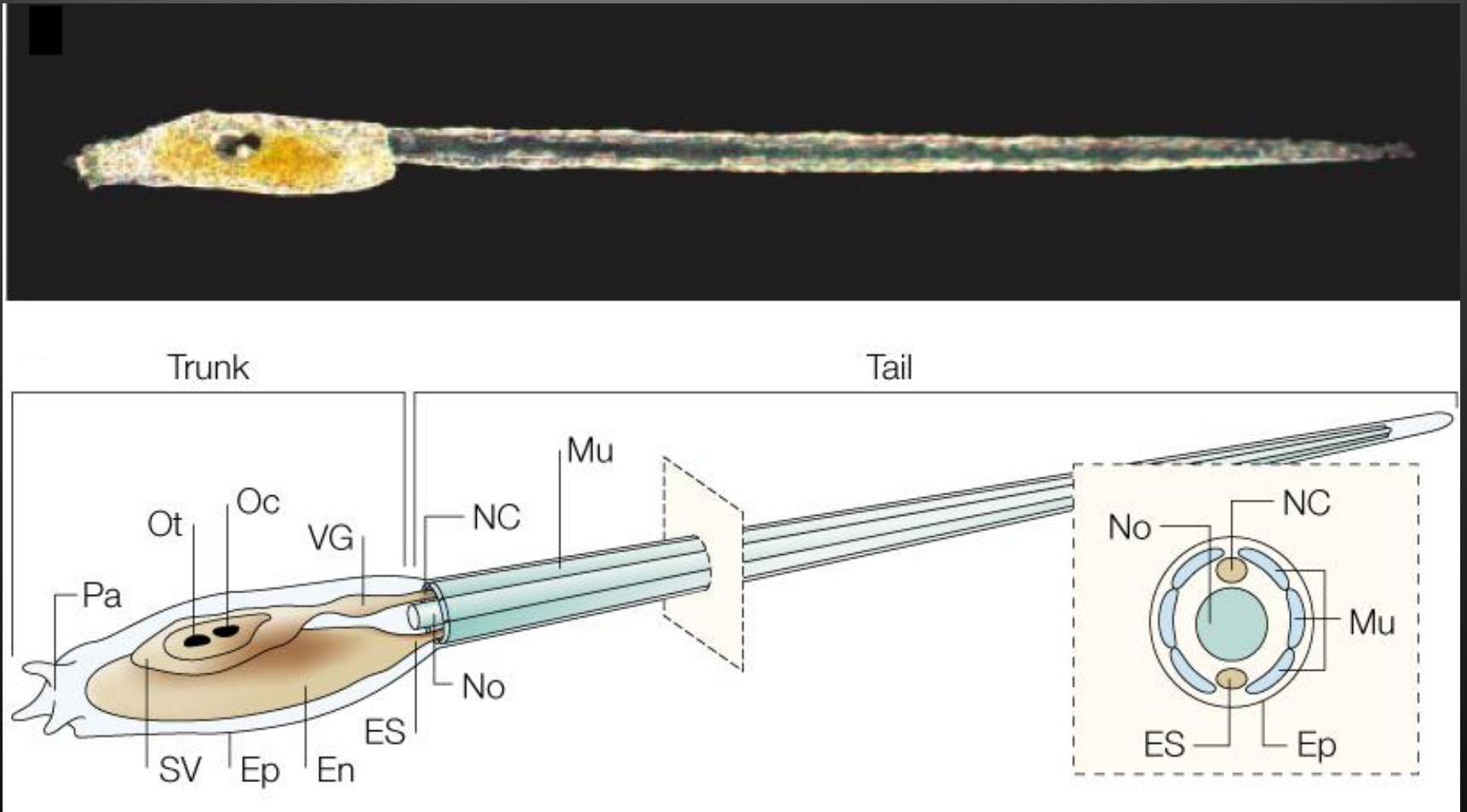
Tunicates are the closest living relatives to vertebrates



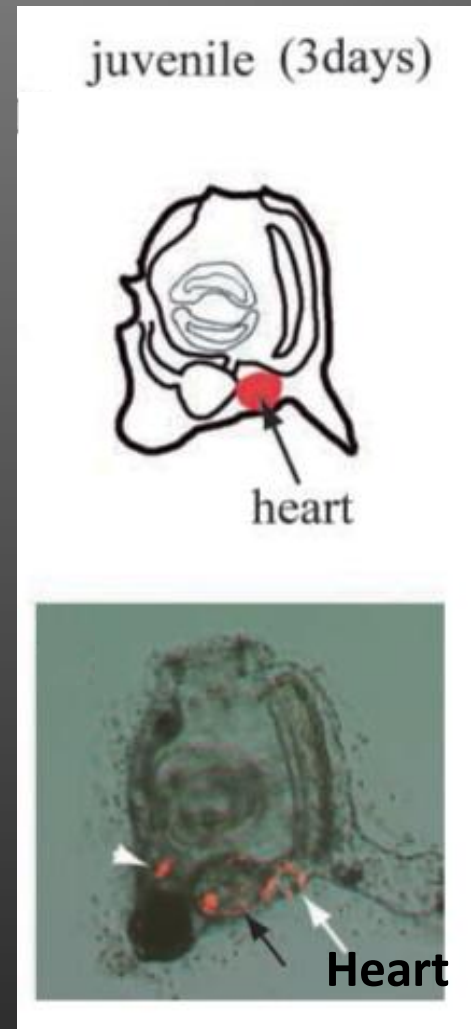
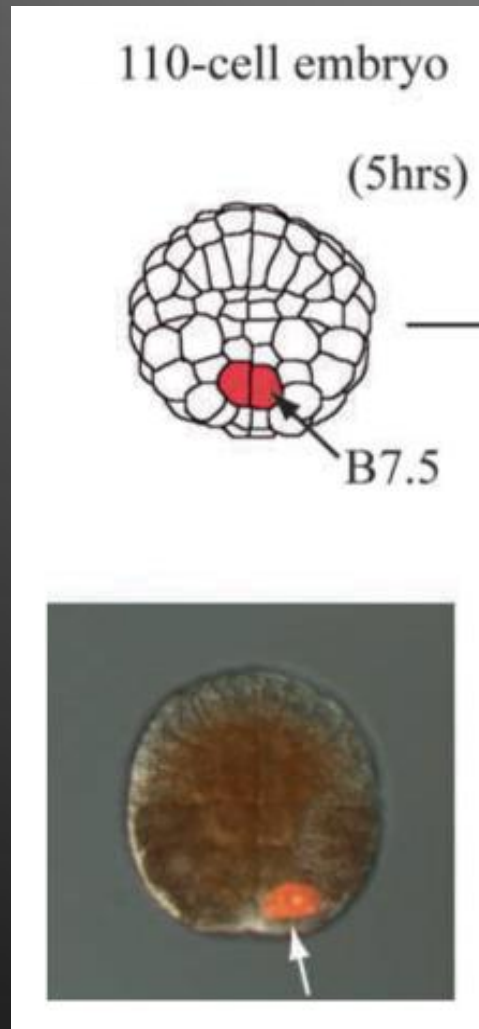
Delsuc et al. (2006) *Nature*

Satoh (2003) *Nat Rev Genet*

A typical chordate body plan



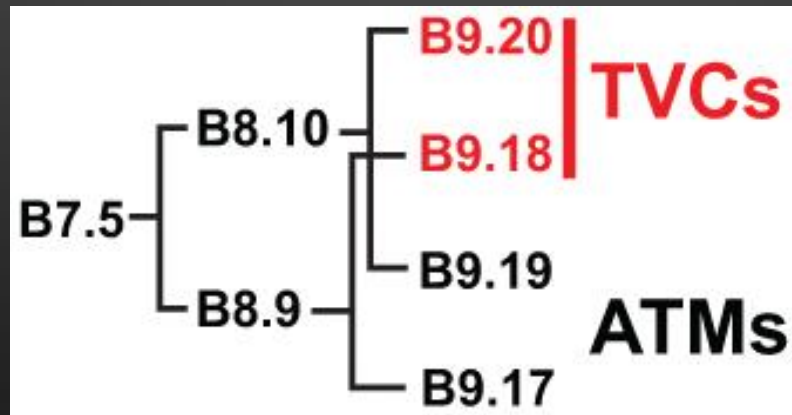
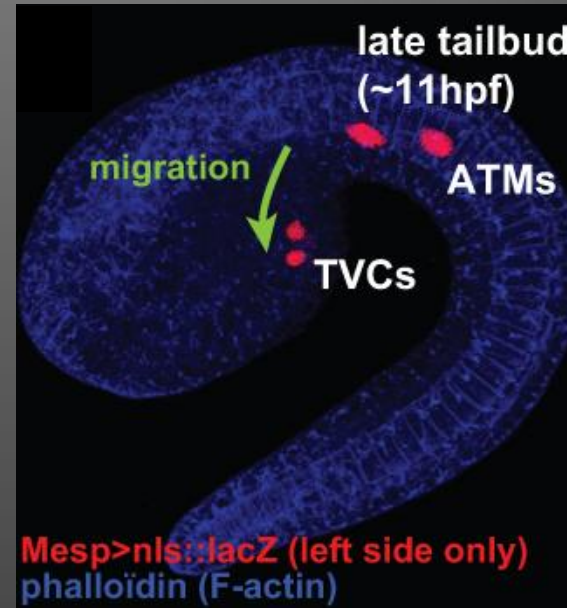
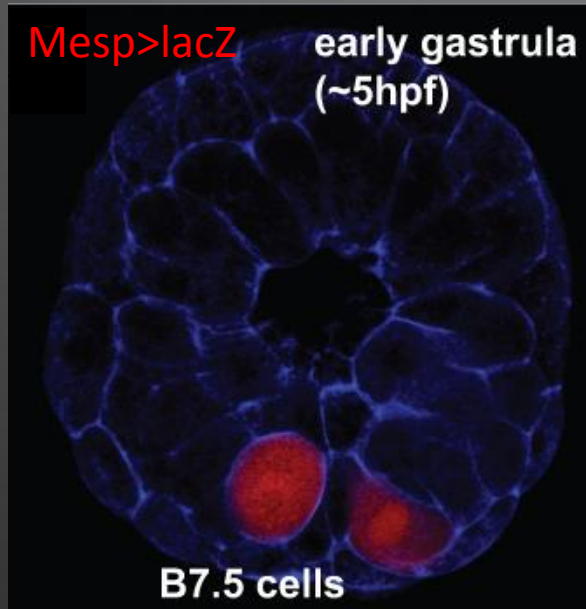
The Ascidian Heart originates from B7.5 cells



B7.5 blastomeres express *Mesp*



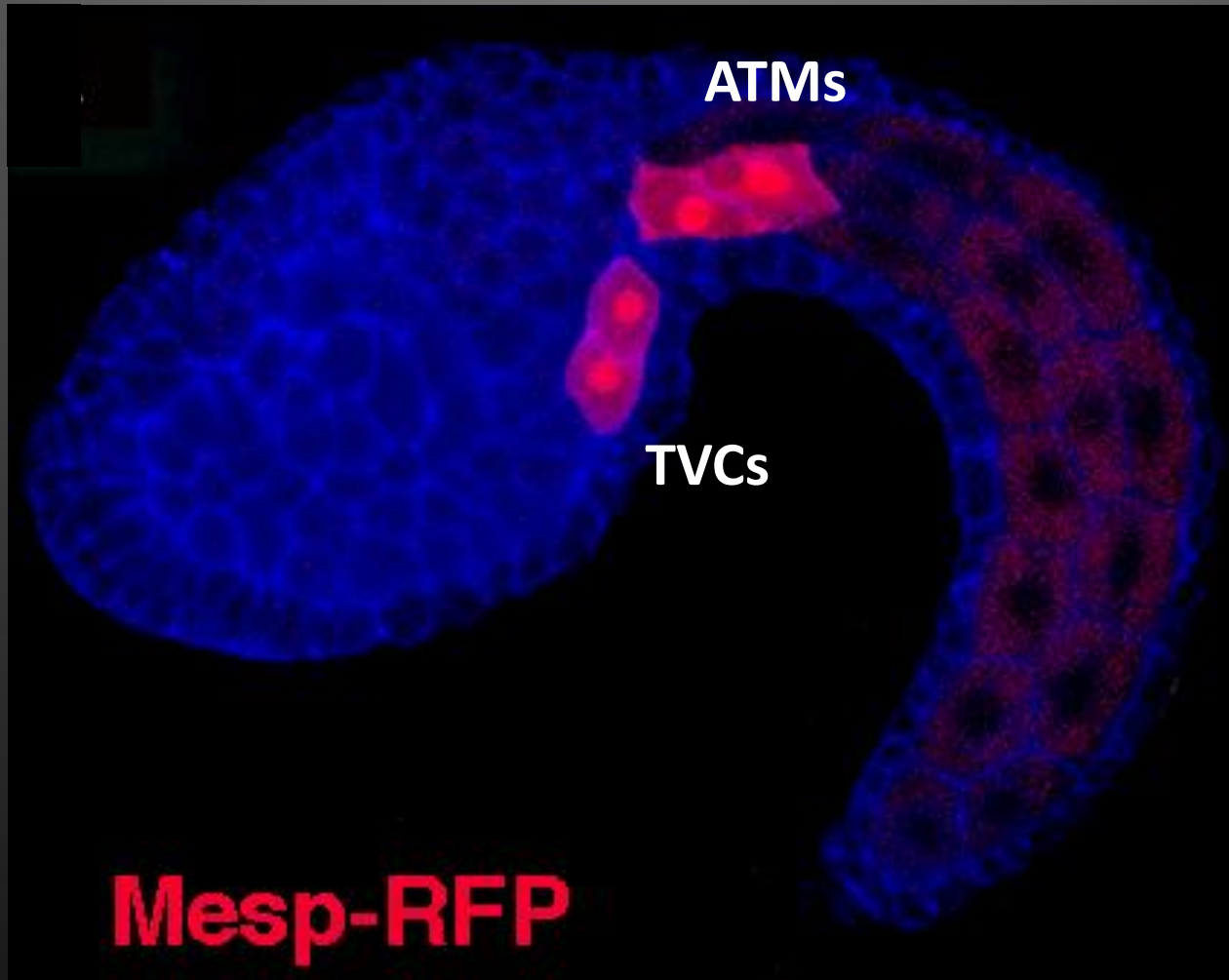
B7.5 blastomeres form TVCs and ATMs





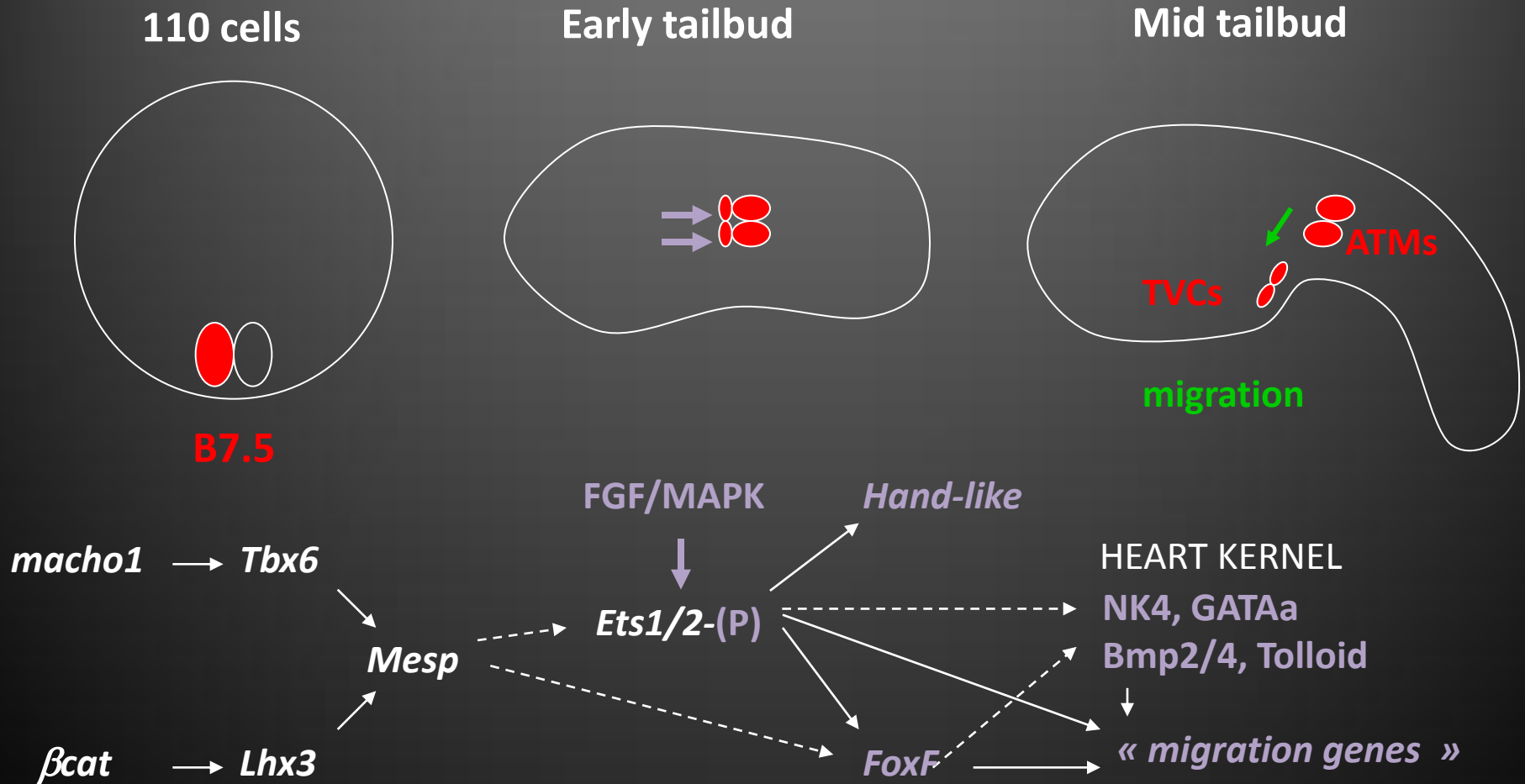
(Courtesy of Alberto Stolfi)

The B7.5-lineage gives birth to distinct cell types



(Davidson et al., 2006, Genes&Dev)

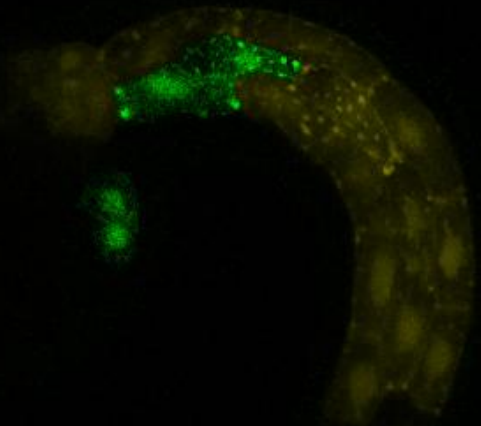
Transcriptional control of TVC fate specification and migration



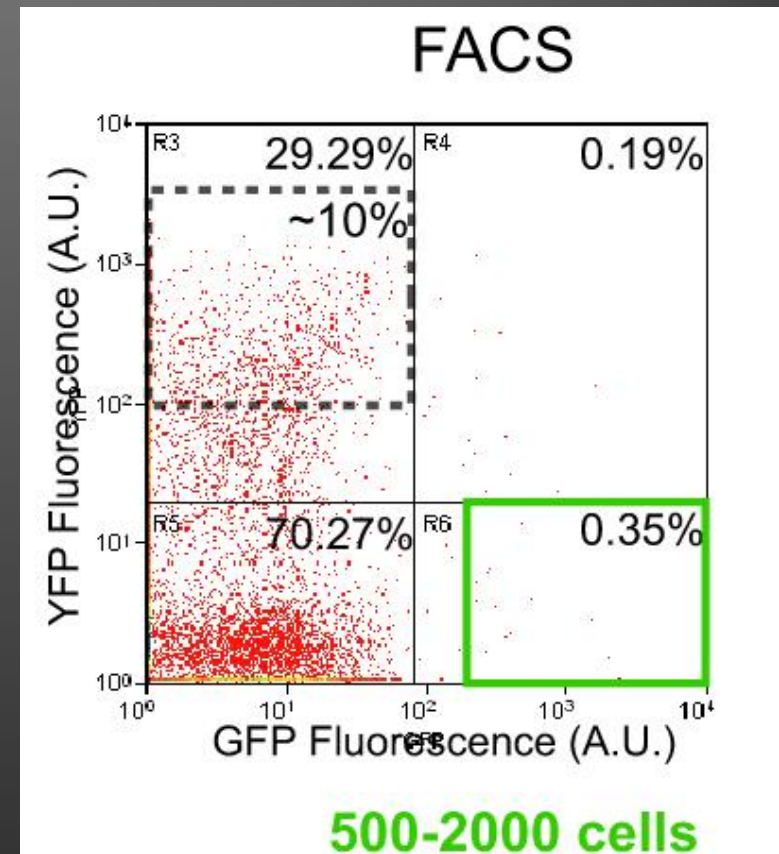
FACCS

Fluorescence Activated Ciona Cell Sorting

development & phenotype >75%

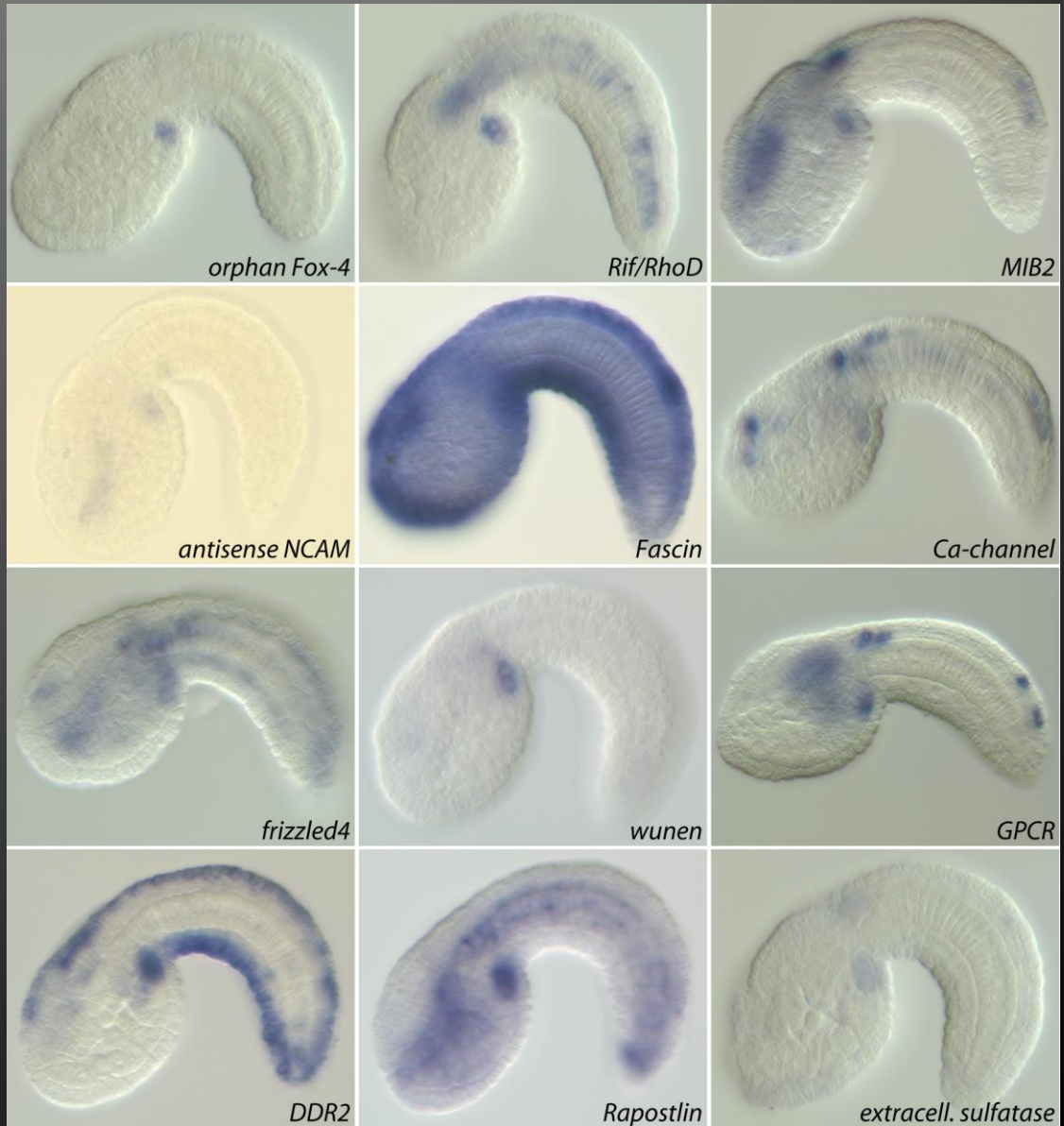
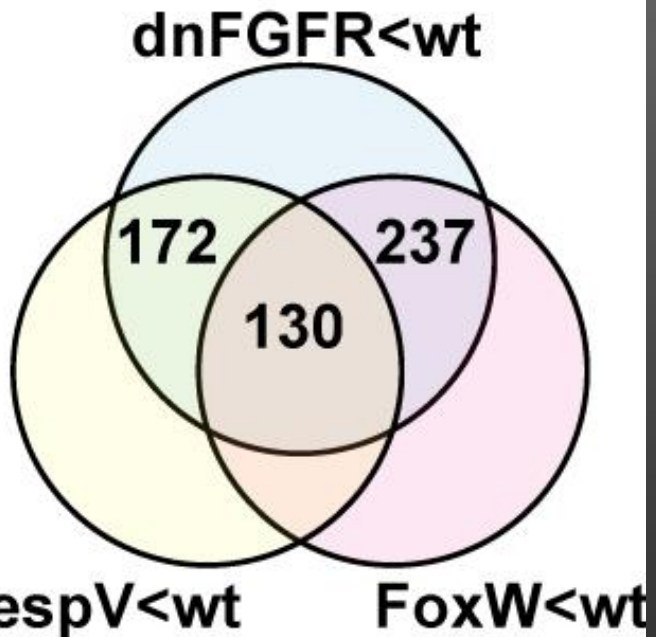


Mesp>GFP
MyoD>YFP

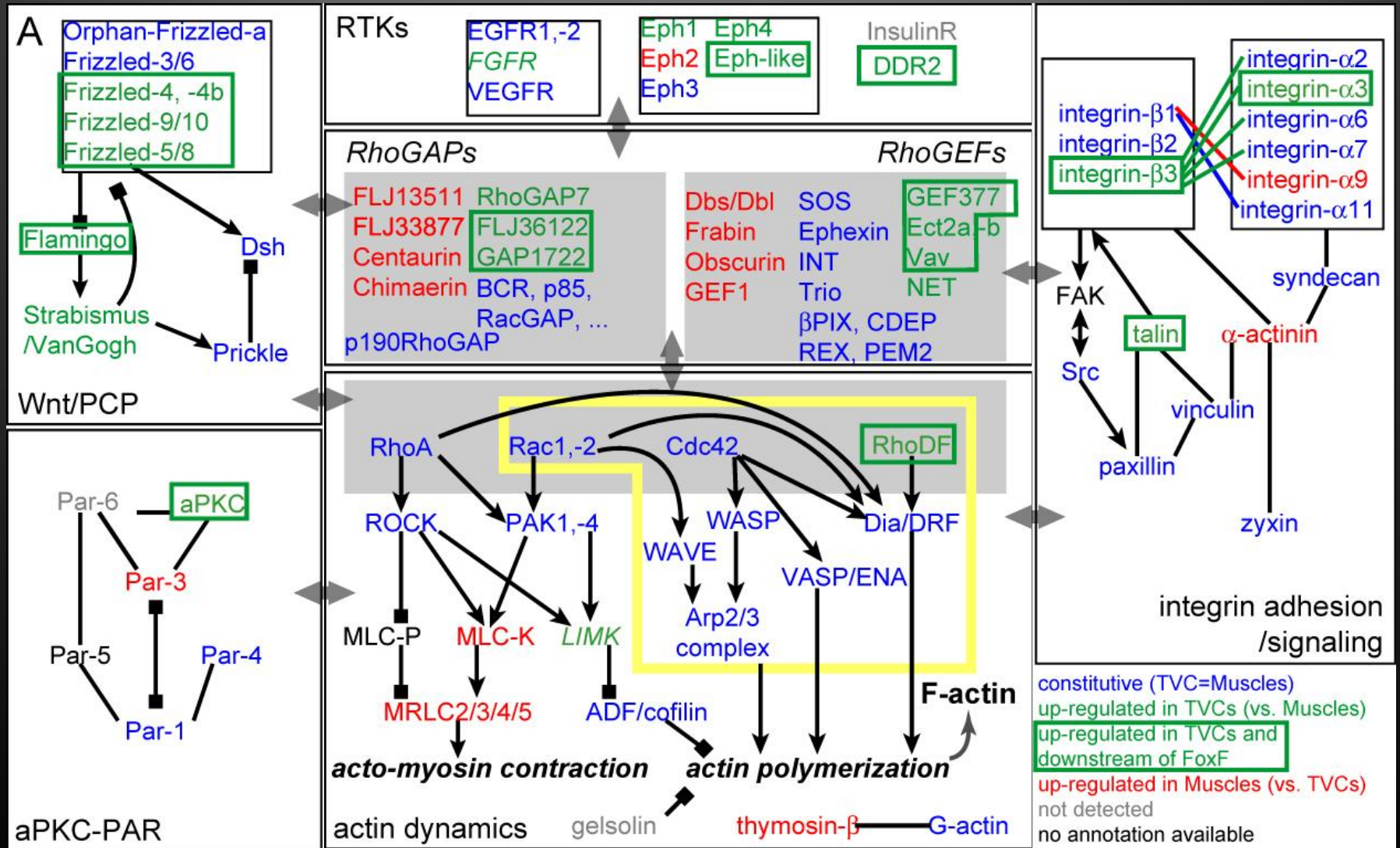


Whole Genome Microarrays

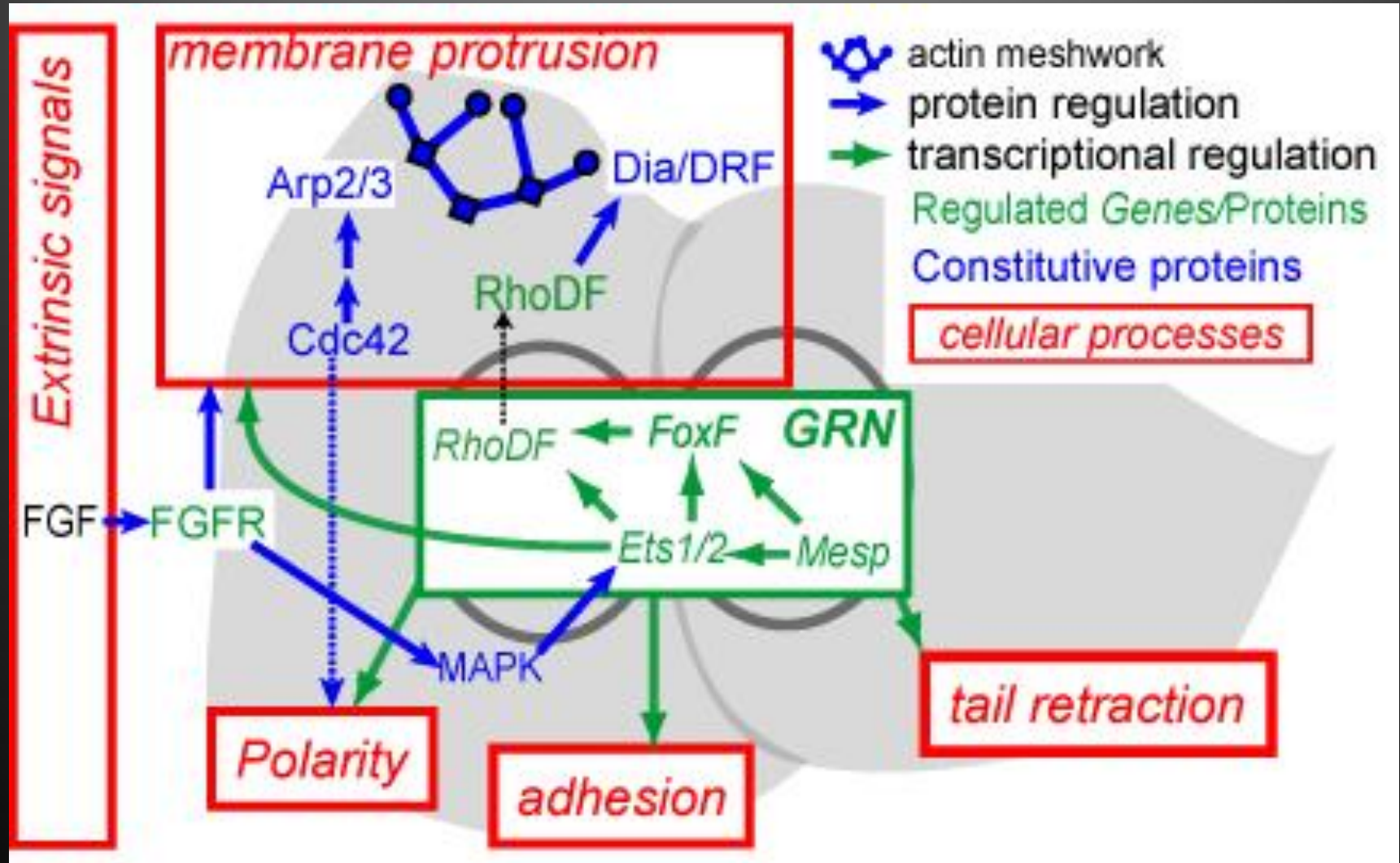
candidate migration genes



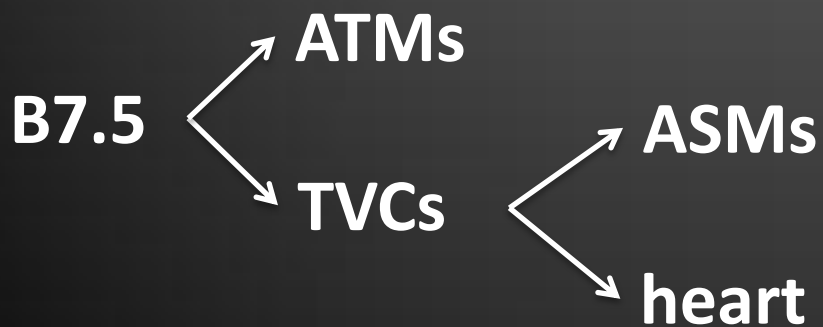
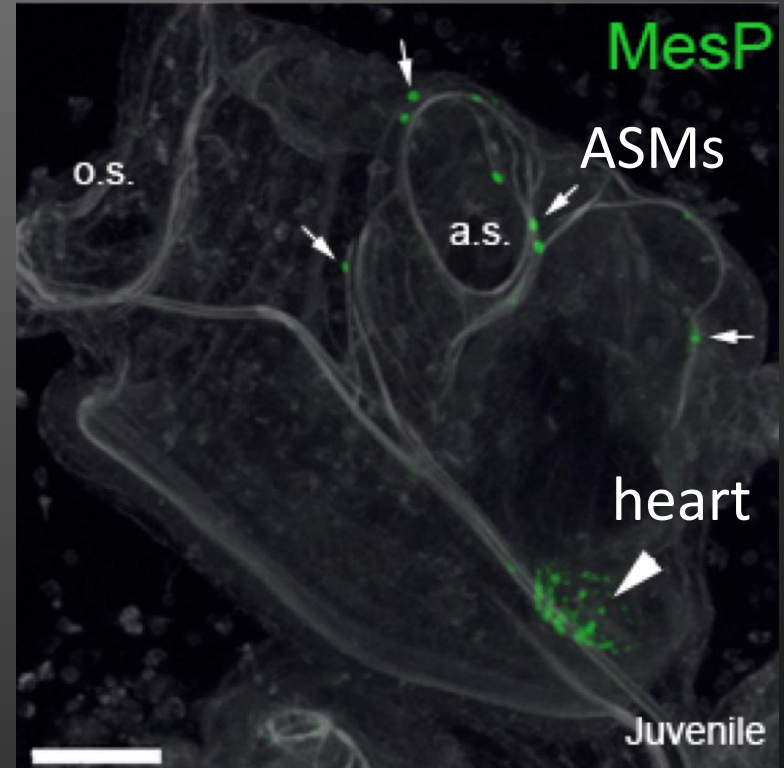
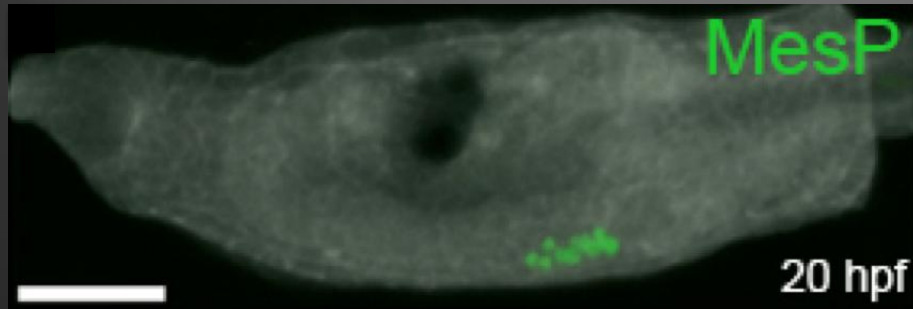
process-specific effectors genes



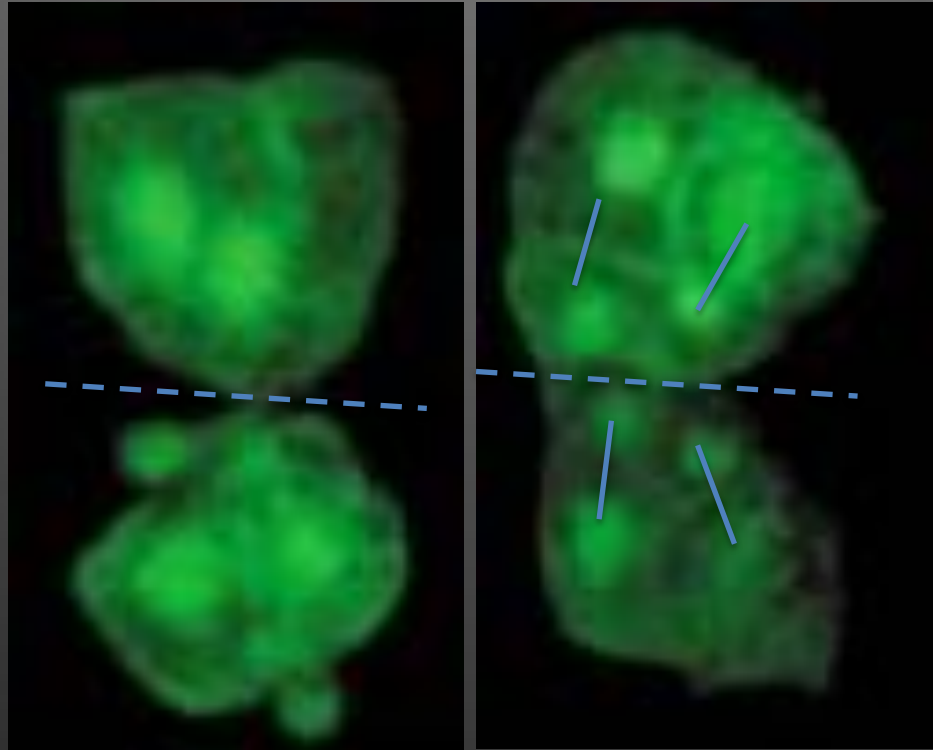
Summary model



TVCs from heart and atrial siphon muscles (ASM)

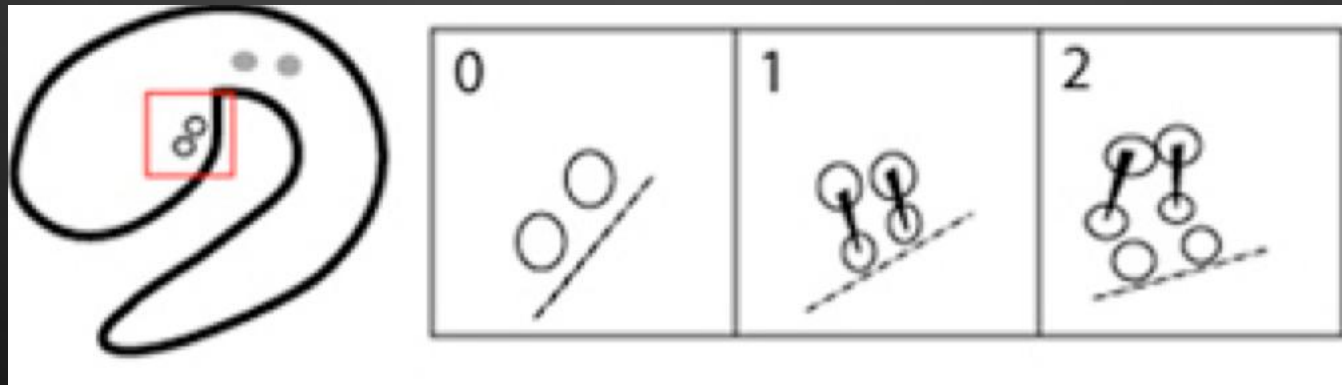
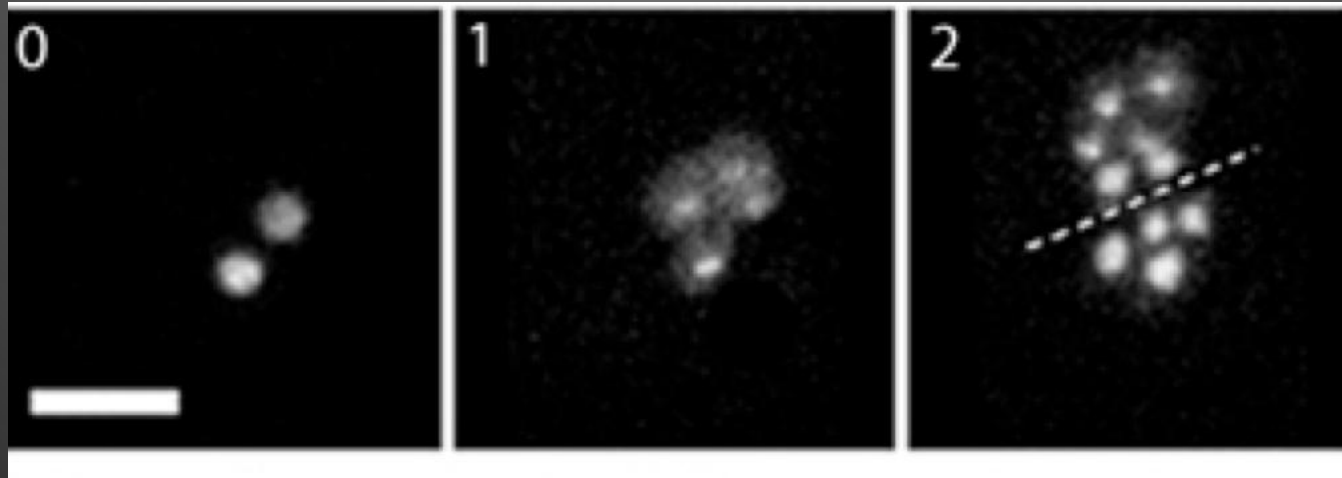


TVCs first divide asymmetrically

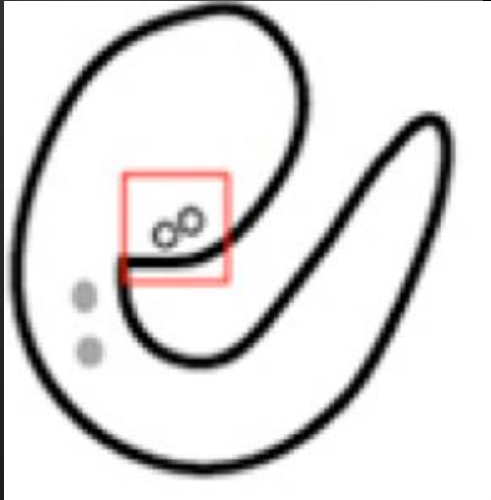


Davidson *et al* (2005) *Development*

TVCs first divide asymmetrically

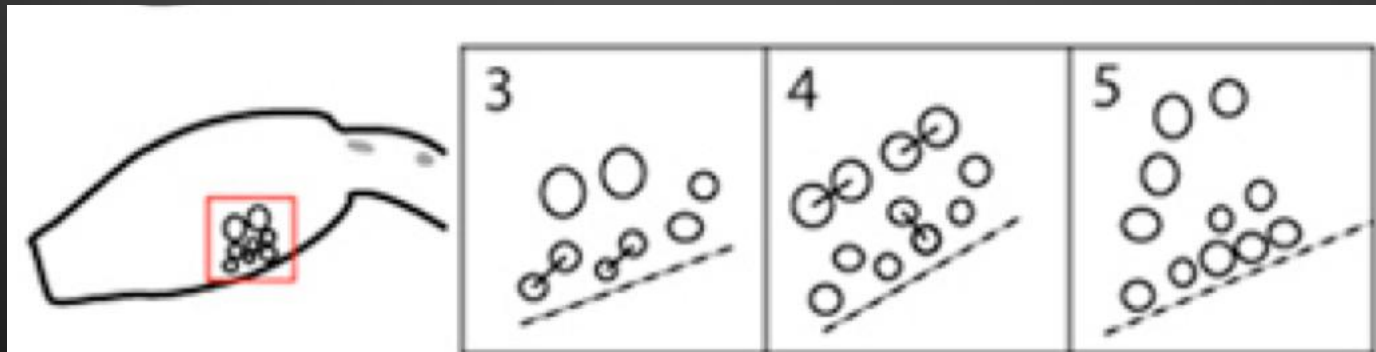
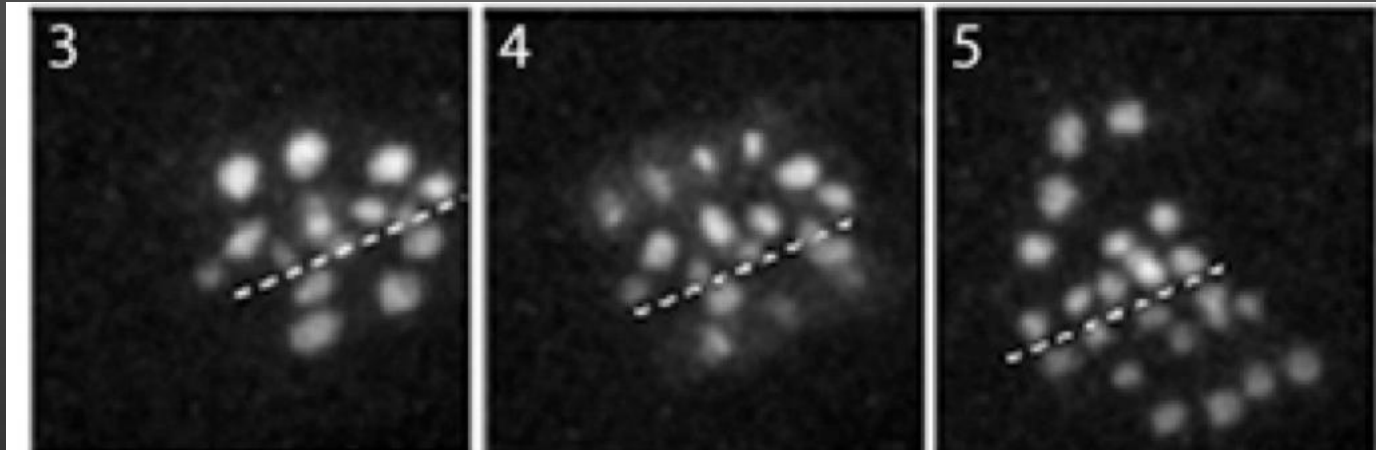


TVCs first divide asymmetrically

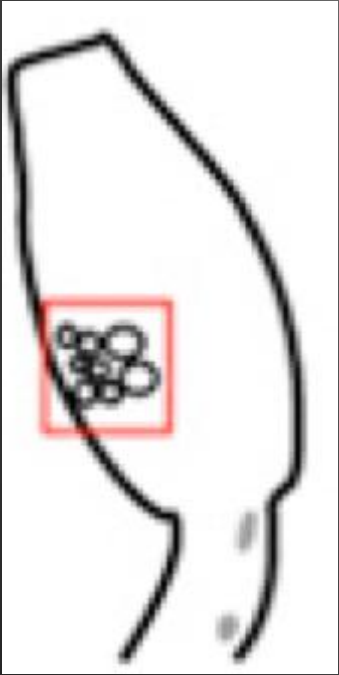


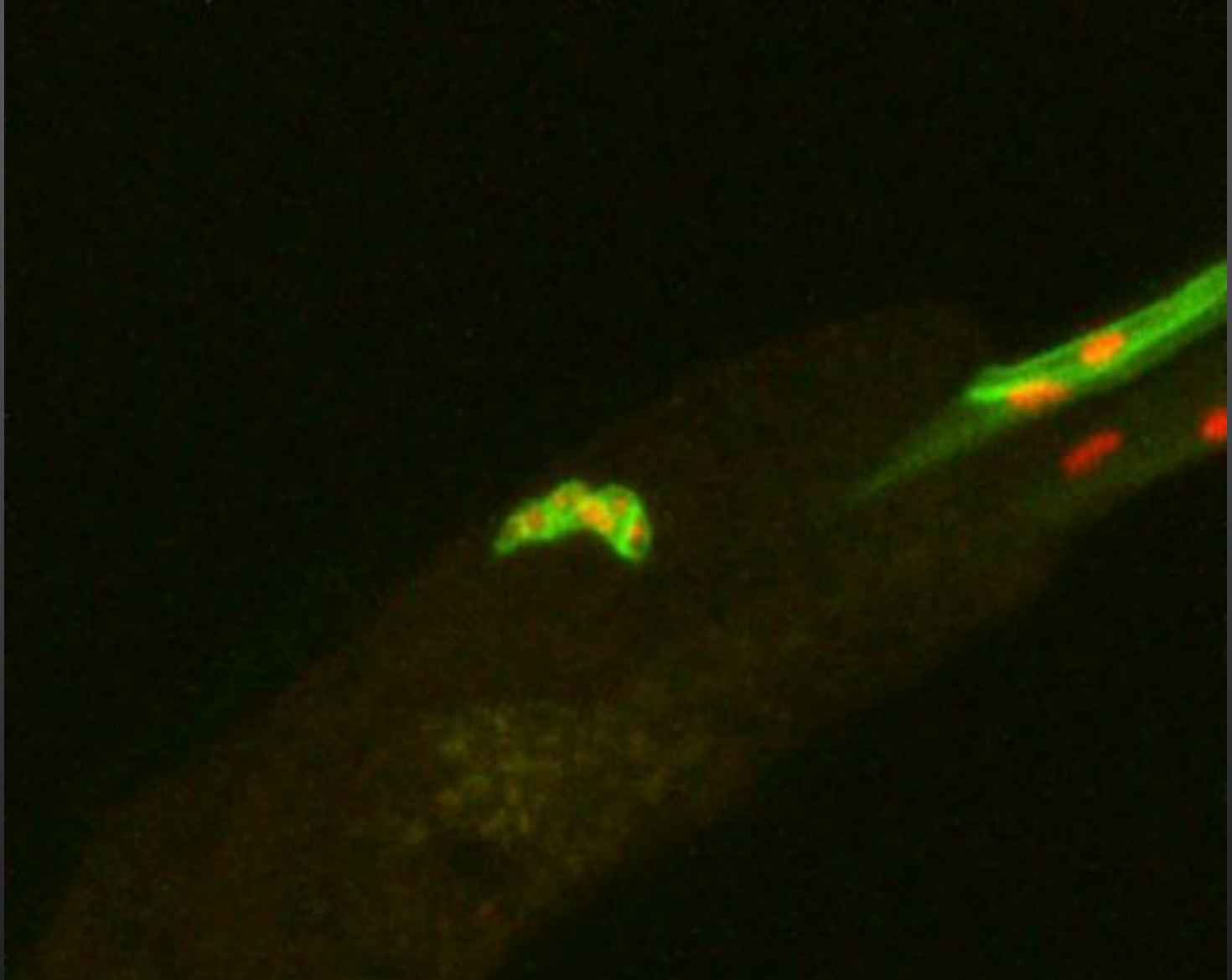
Mesp>H2B-GFP

lateral TVCs migrate again



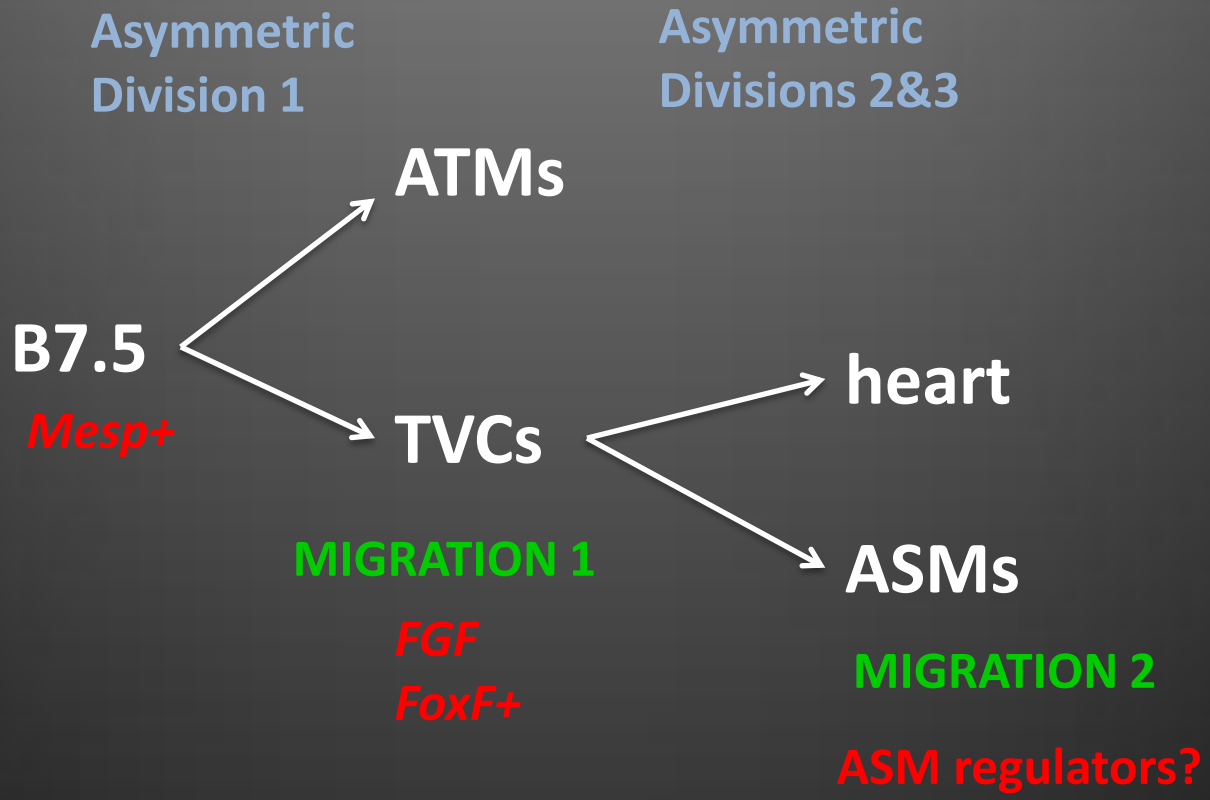
lateral TVCs migrate again





Mesp>H2B:mCherry

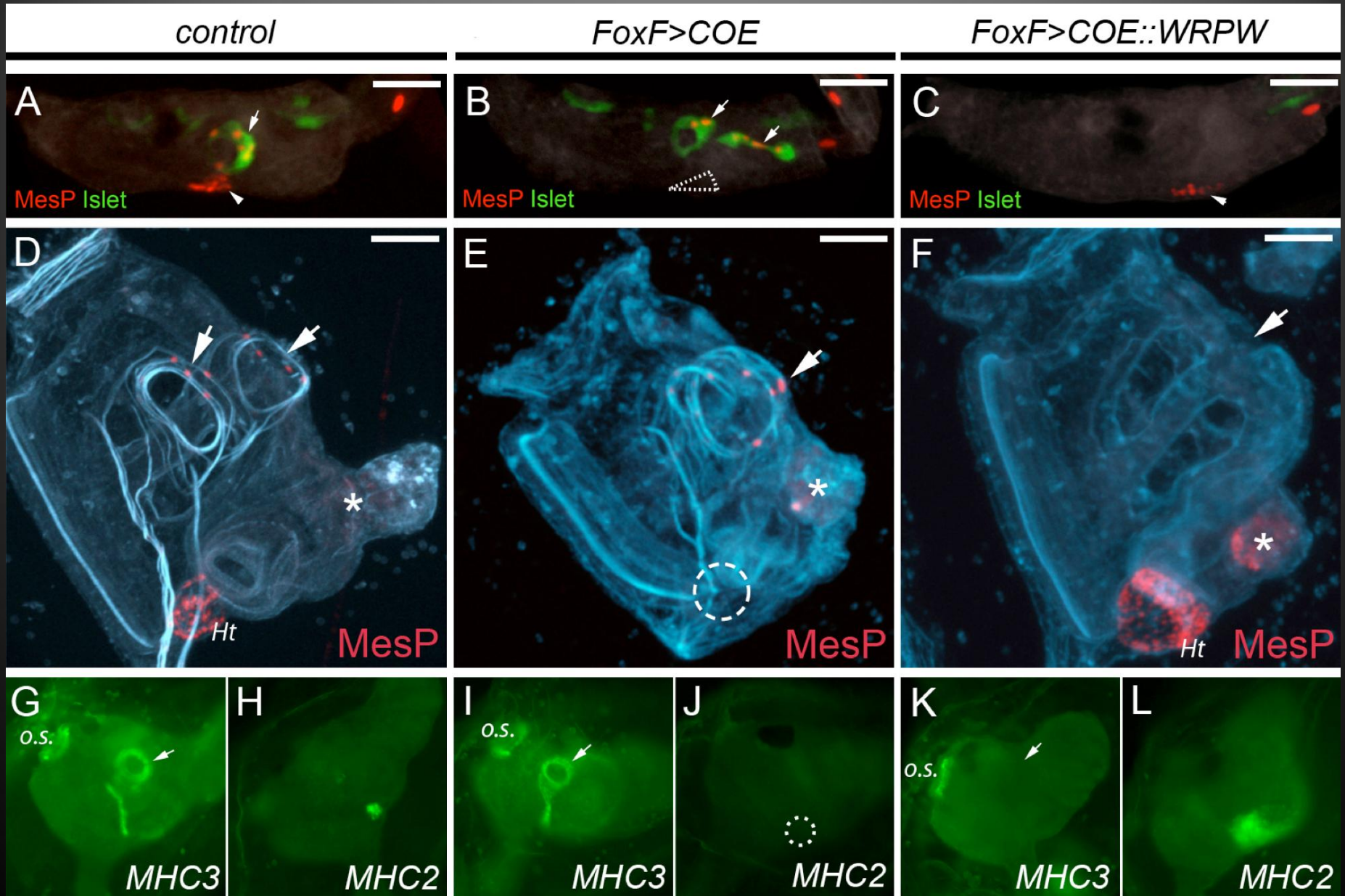
Mesp>PH:GFP



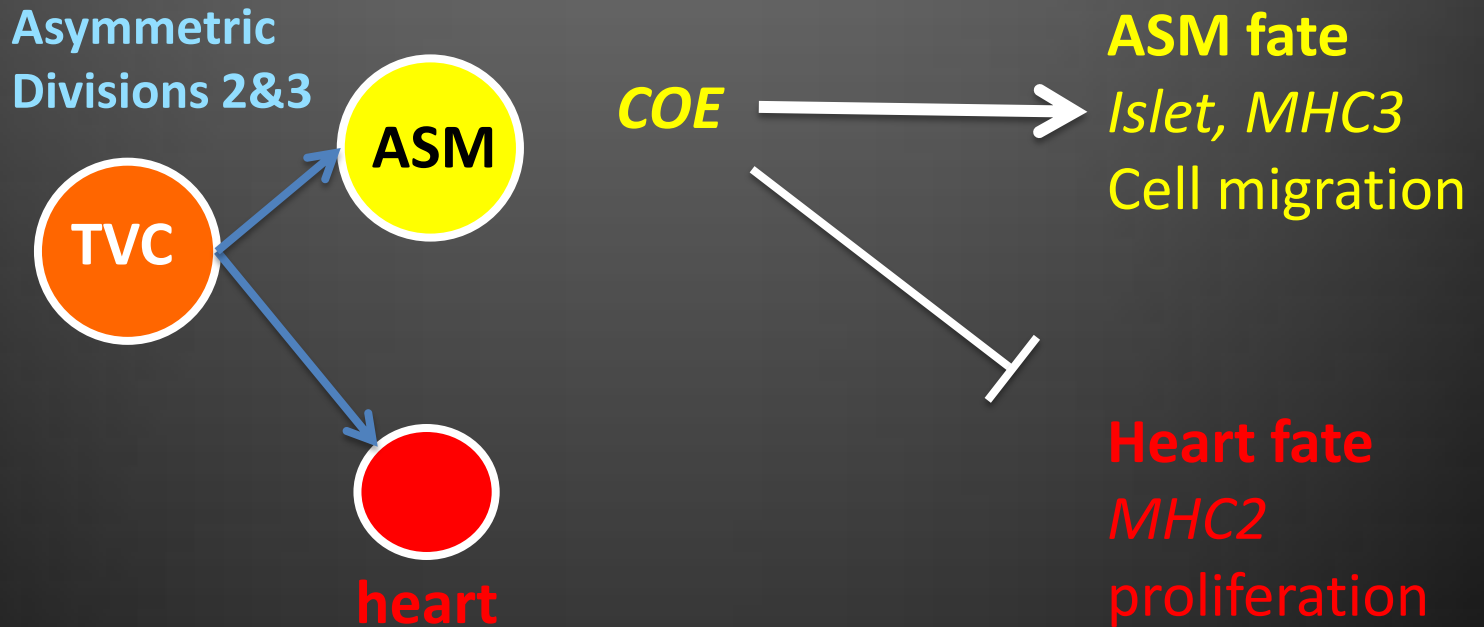
Early ASM-specific expression of *COE*



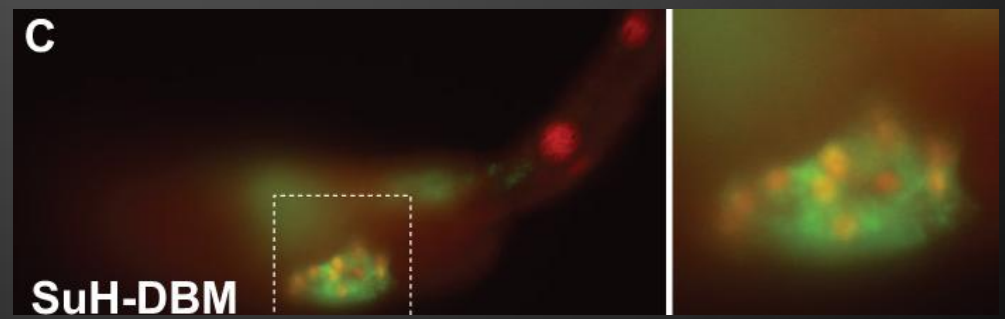
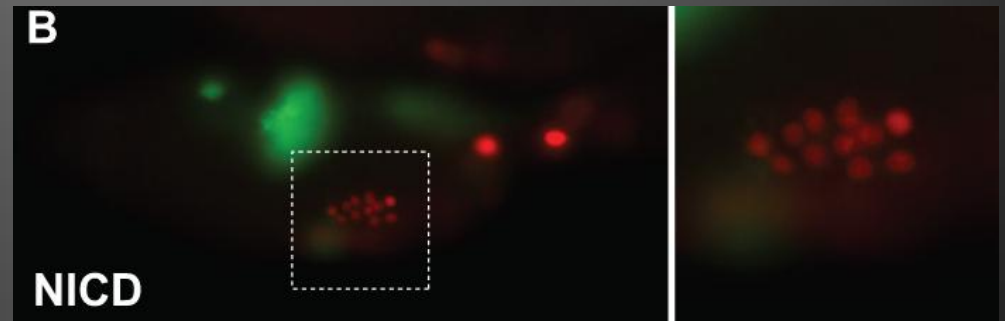
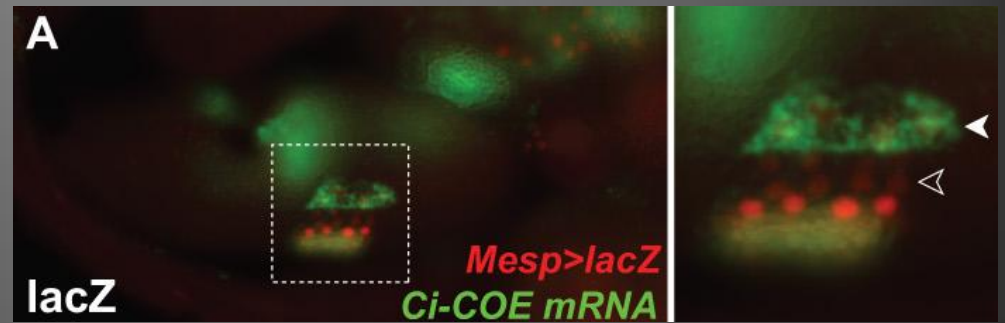
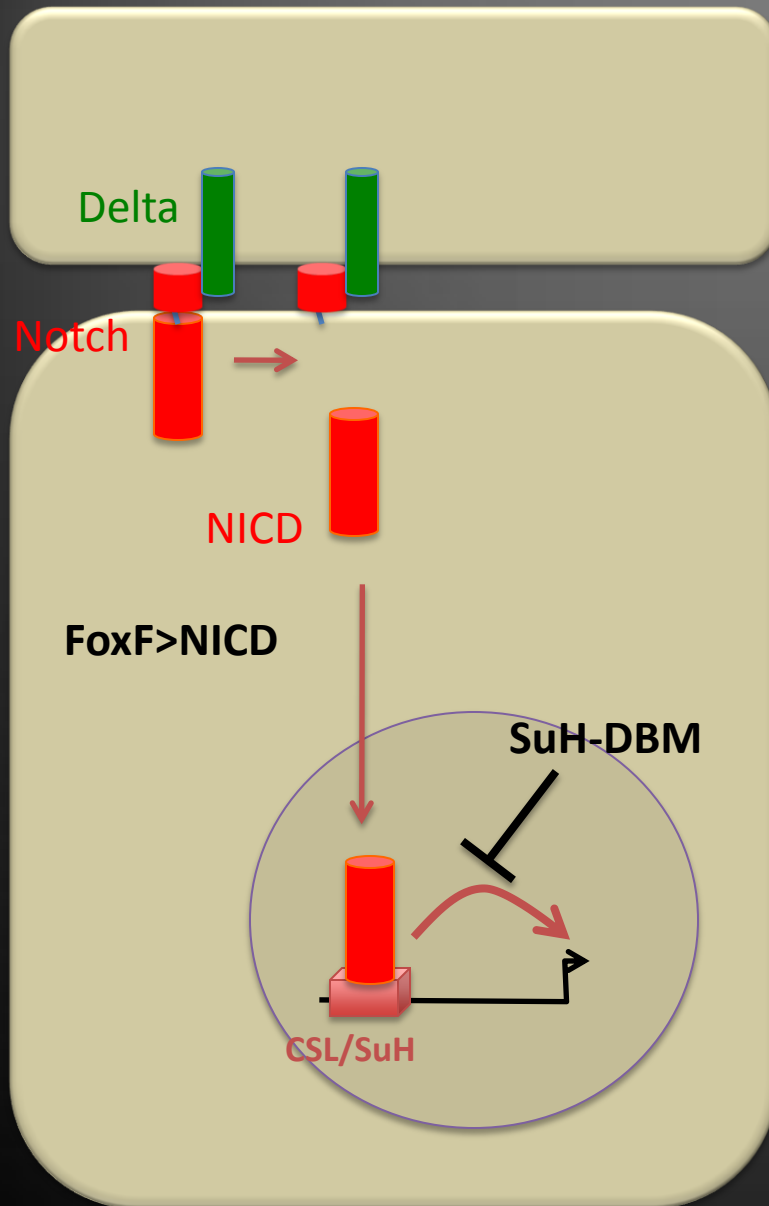
COE is necessary and sufficient for ASM specification



Simplest model

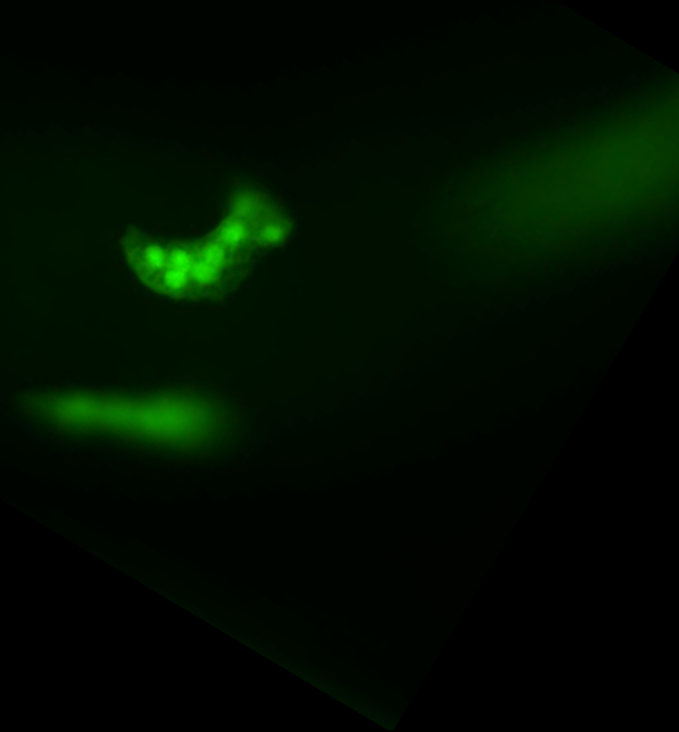


Notch signaling inhibits *COE* expression



Over-expression of NICD blocks the second migration

50 μ g FoxF>mCherry
(CONTROL)

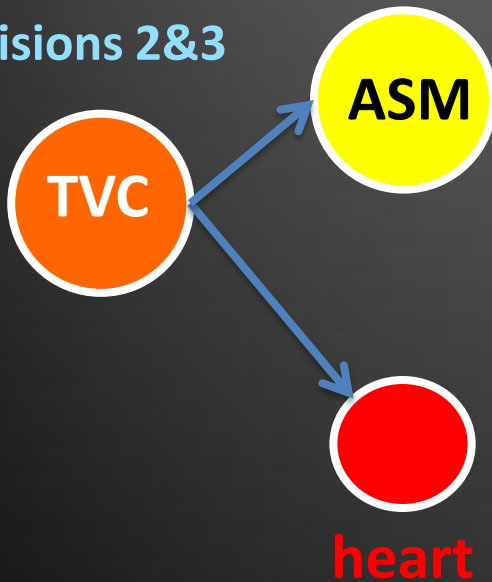


50 μ g FoxF>NICD
(CONTROL)

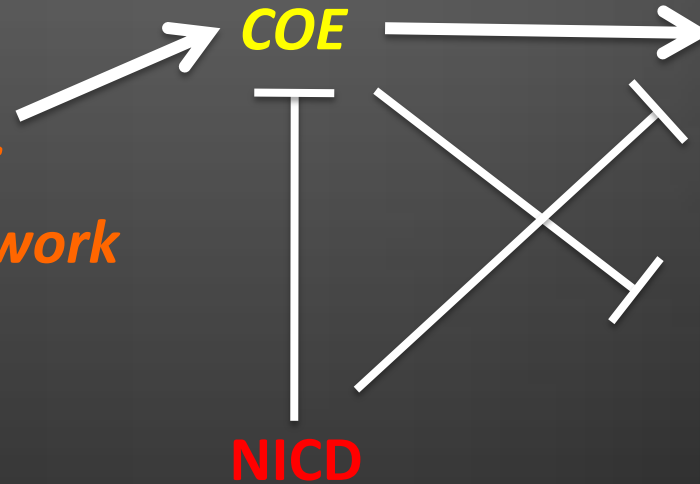


Simplest model

Asymmetric
Divisions 2&3



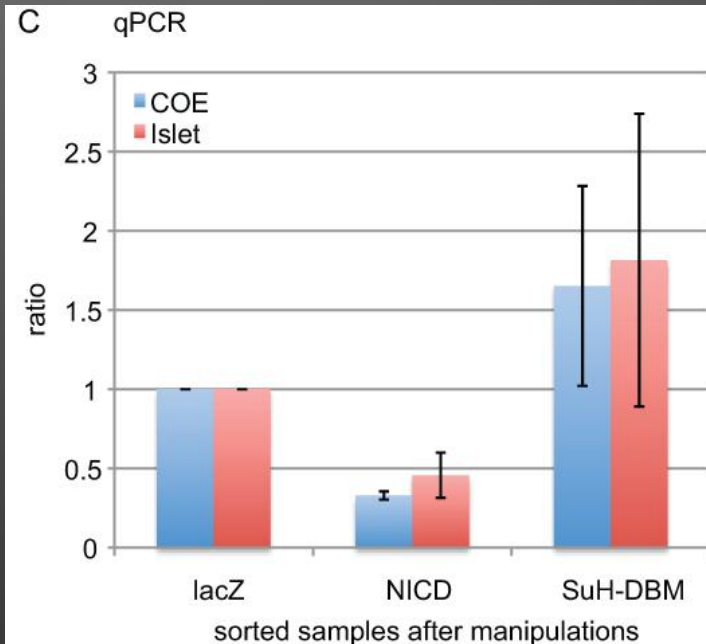
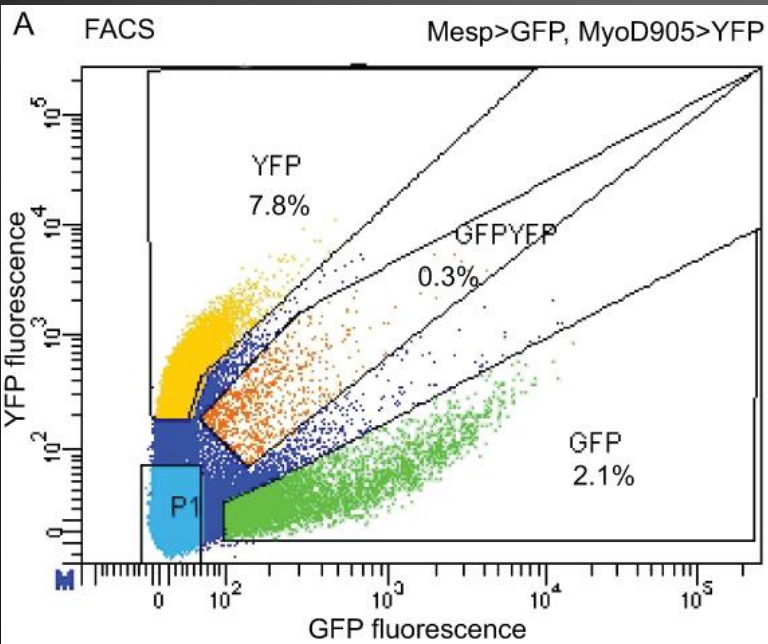
*TVC
network*



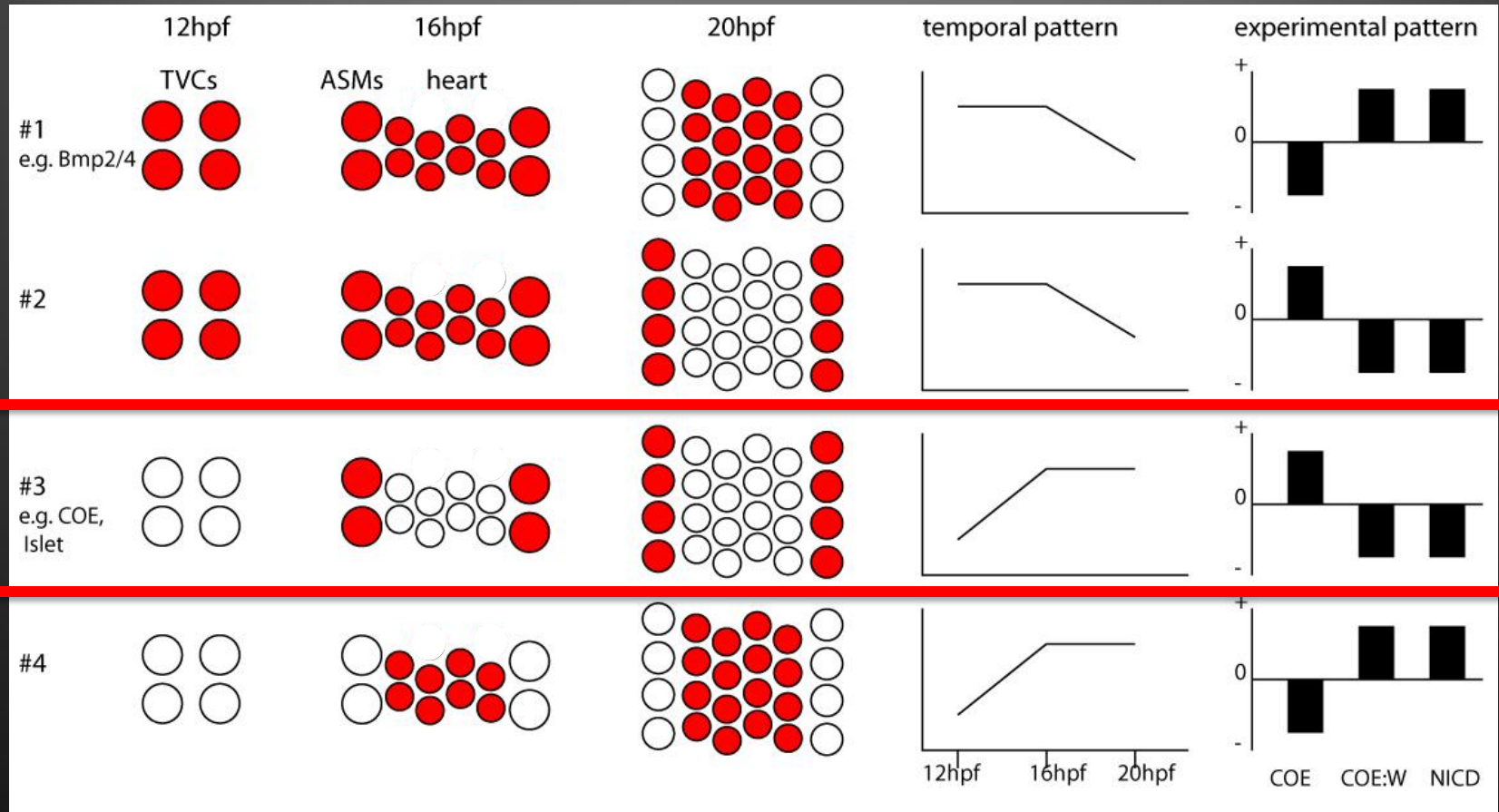
ASM fate
Islet
Cell migration

Heart fate
proliferation

FACS and qPCR confirm that Notch signaling inhibits *COE* and *Islet* expression in the TVC lineage



Using FACS and microarrays to identify heart and ASM-specific transcripts

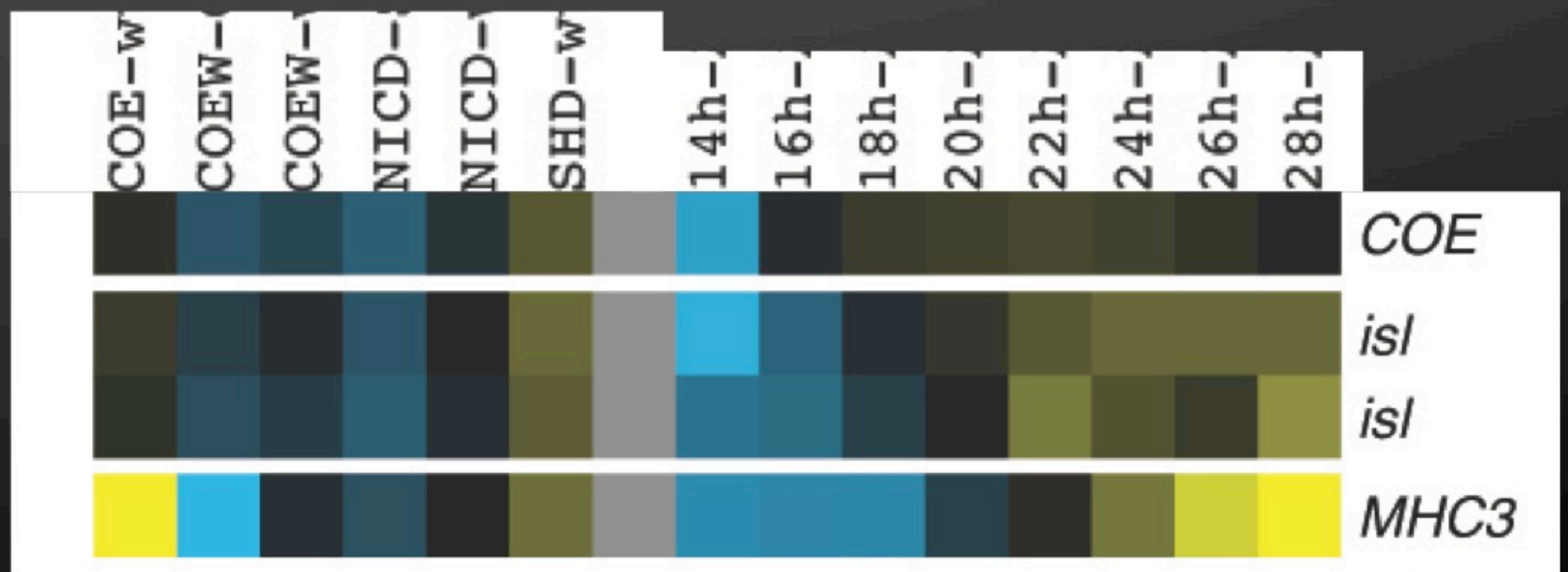
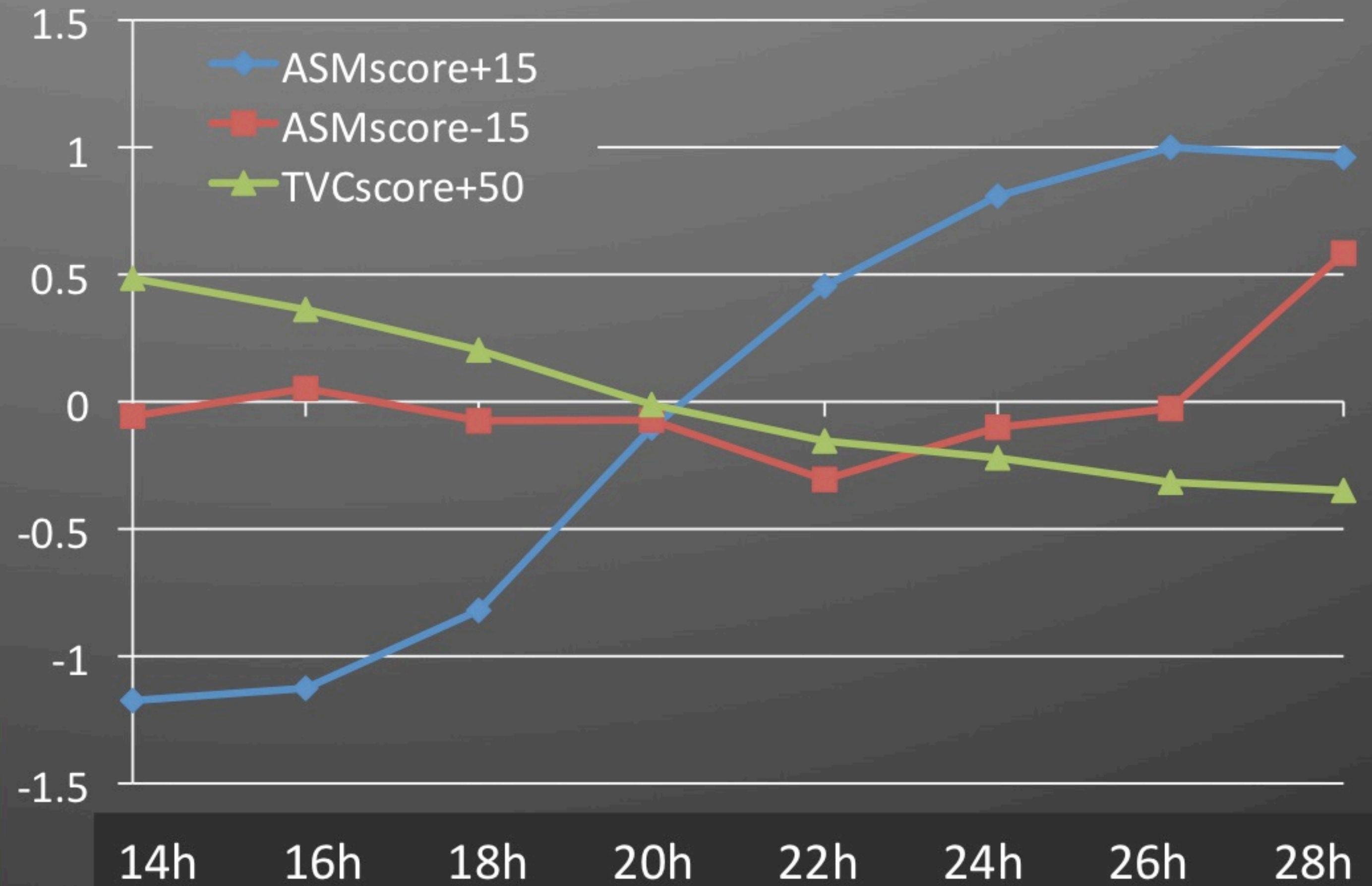
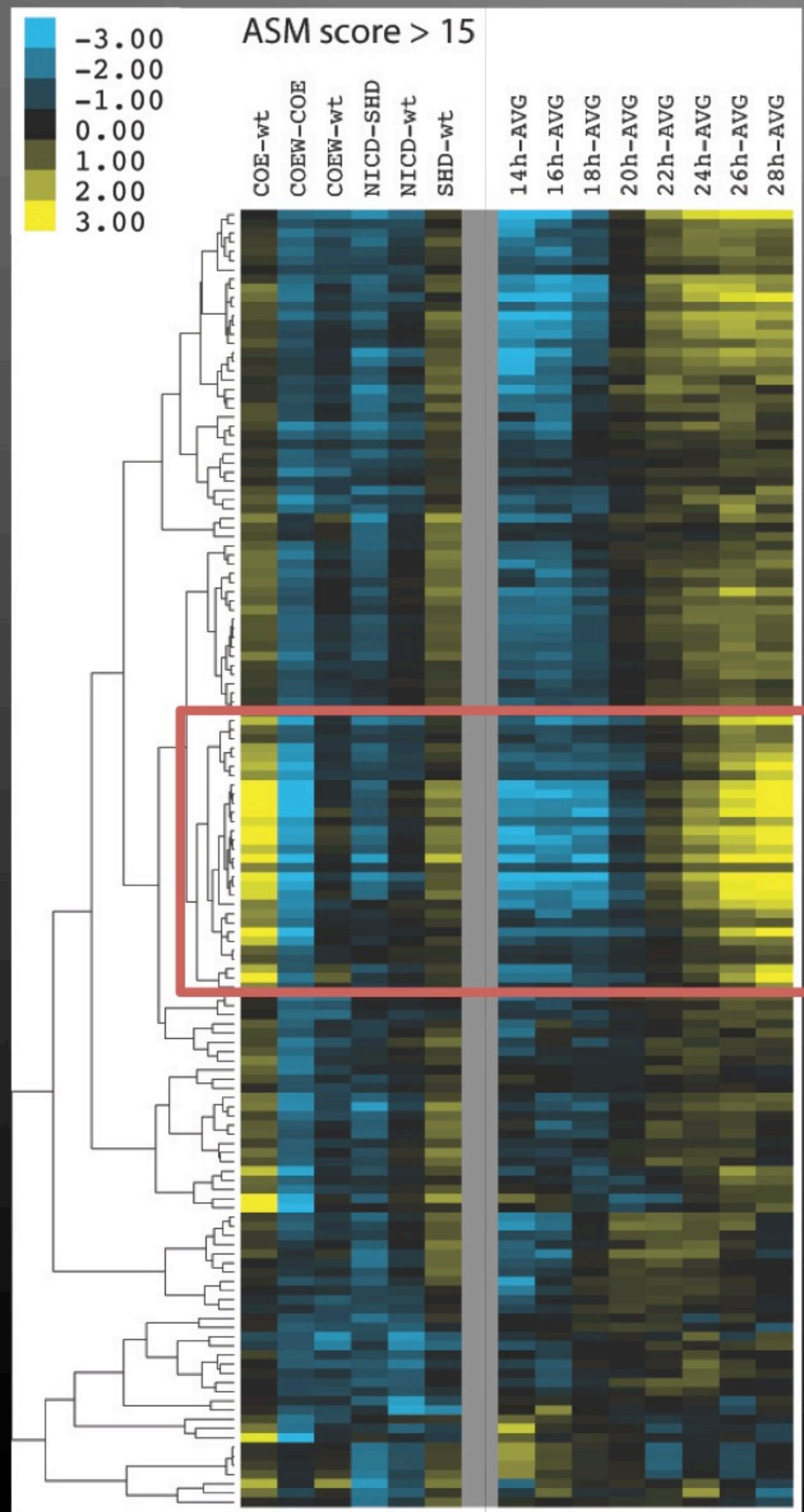


5 conditions: control (lacZ), NICD, SuH-DBM, COE, COEWRPW

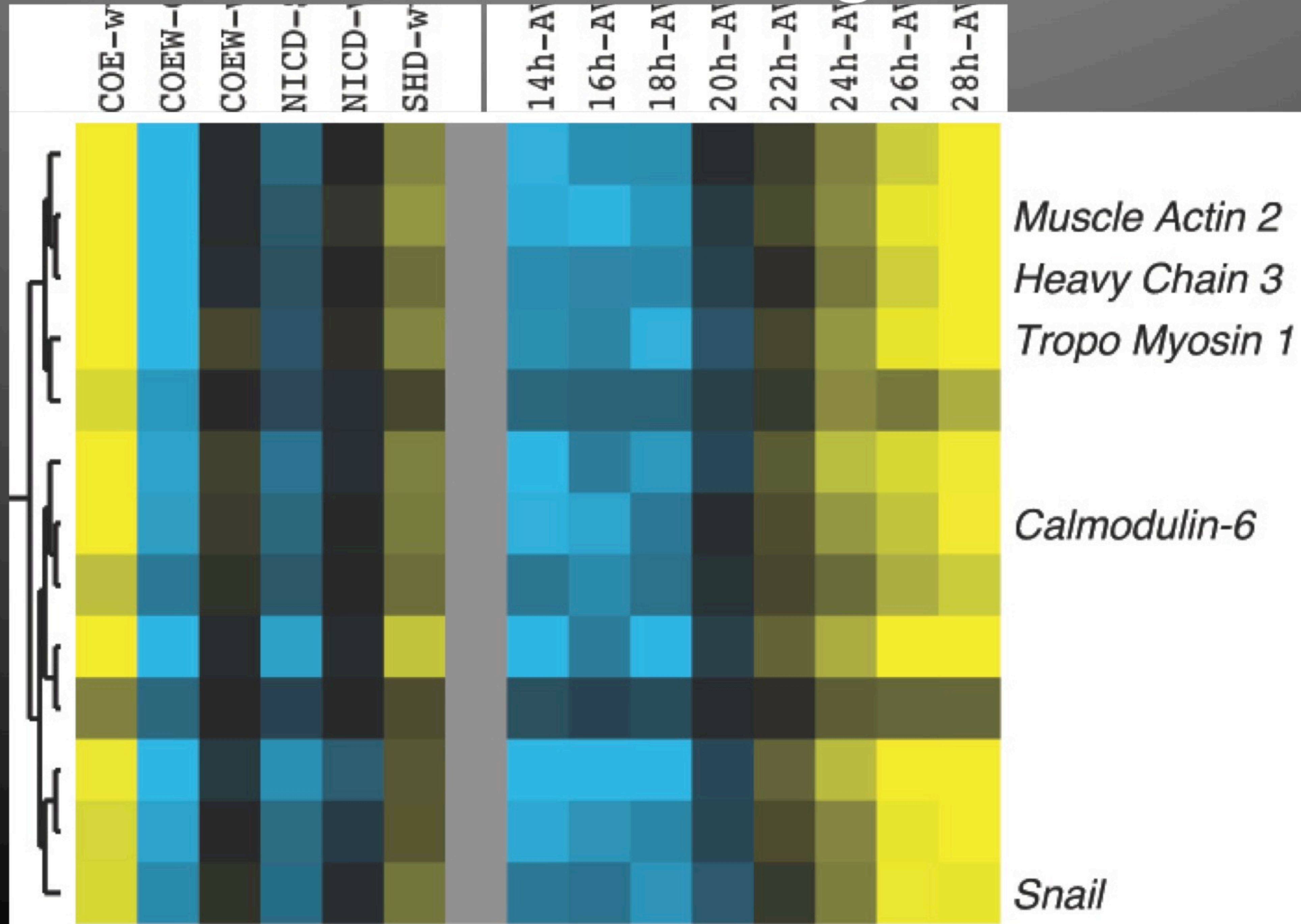
8 time points : 14h, 16h, 18h, 20h, 22h, 24h, 26h, 28h

ASM score = 20h(WT) x [(COE-COEWRPW)+(SuHDBM-NICD)]

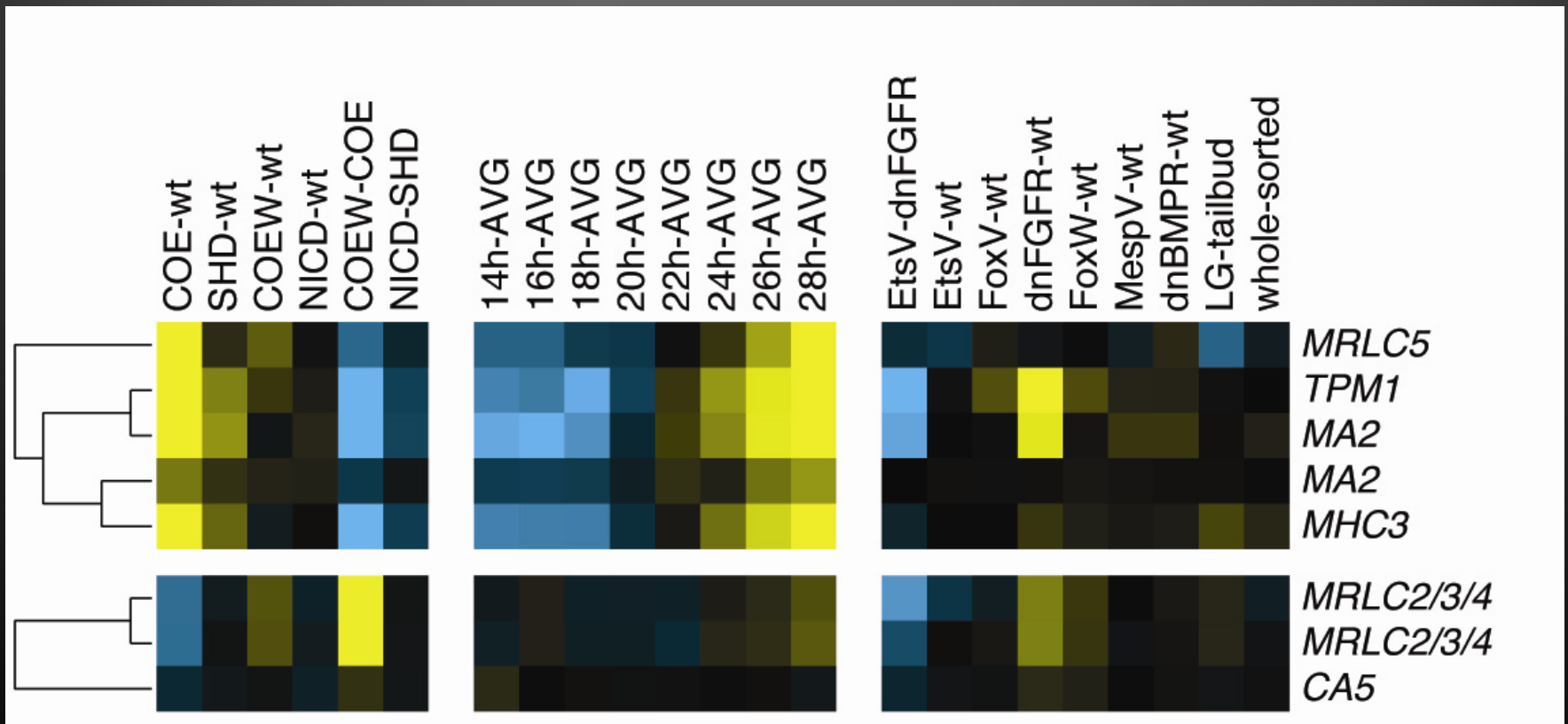
Most ASM candidates show *de novo* expression



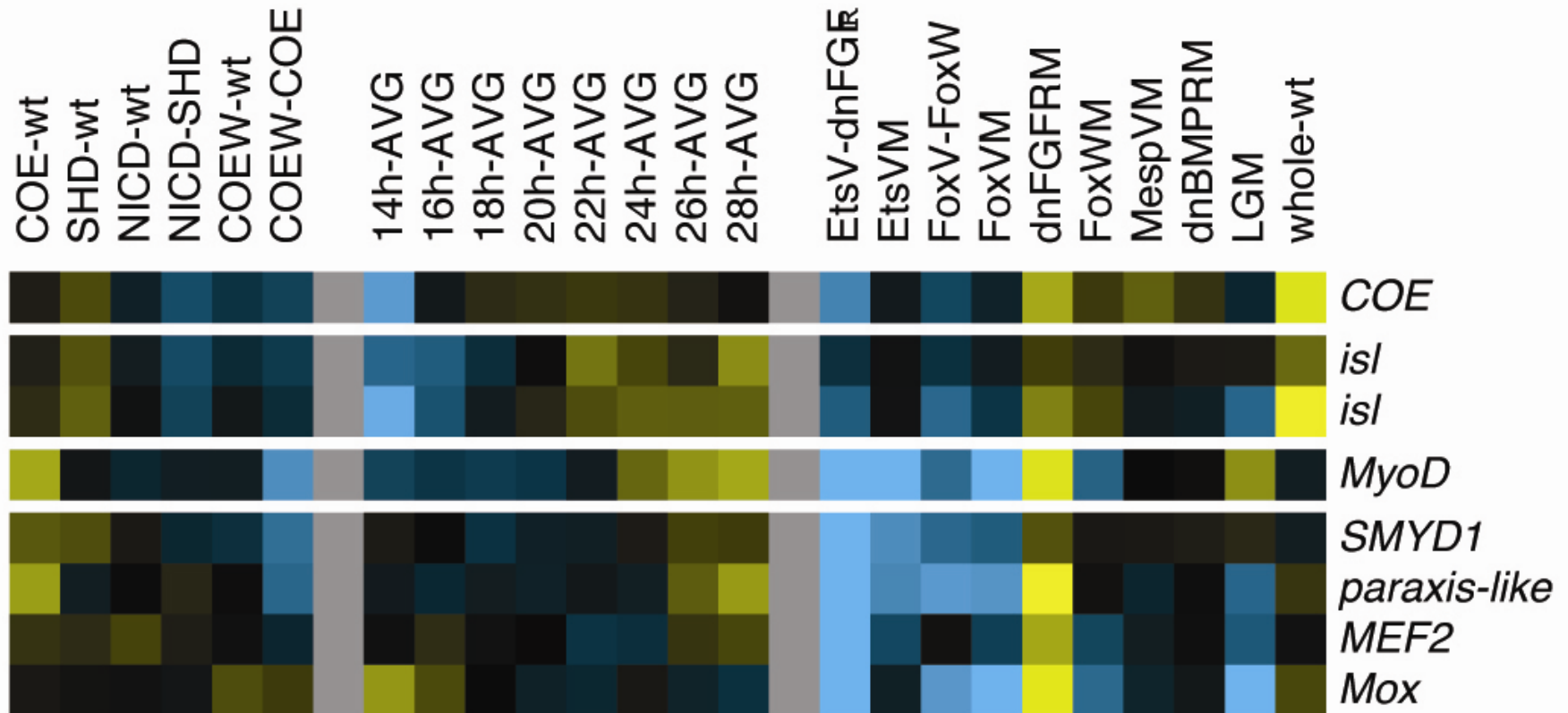
Several candidate ASM genes are skeletal muscle genes



Re-activation of muscle gene batteries?

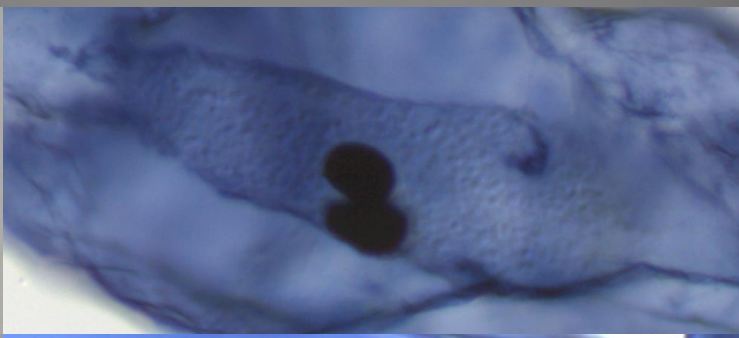
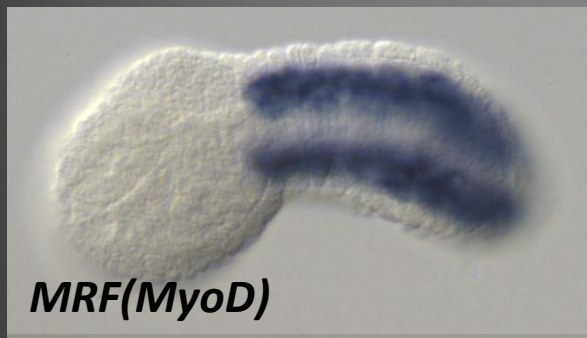


Essential Muscle Regulators are activated downstream of COE

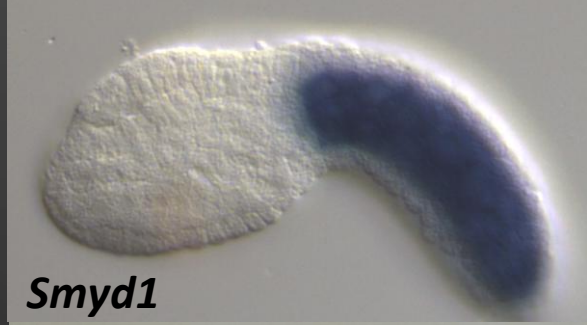


14/18 new
ASM markers
Also in OSM

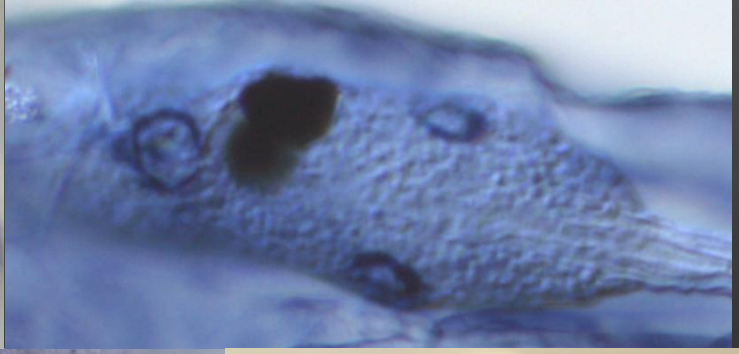
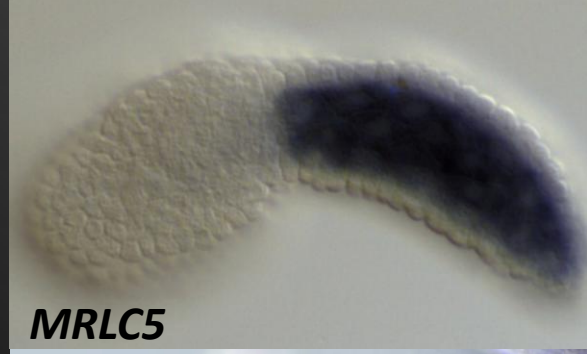
MRF(MyoD)



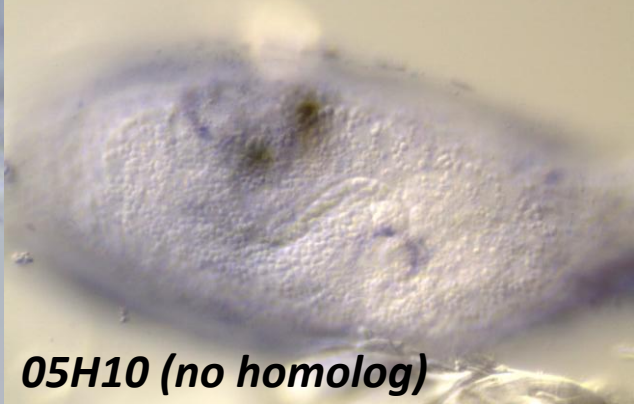
Smyd1



MRLC5



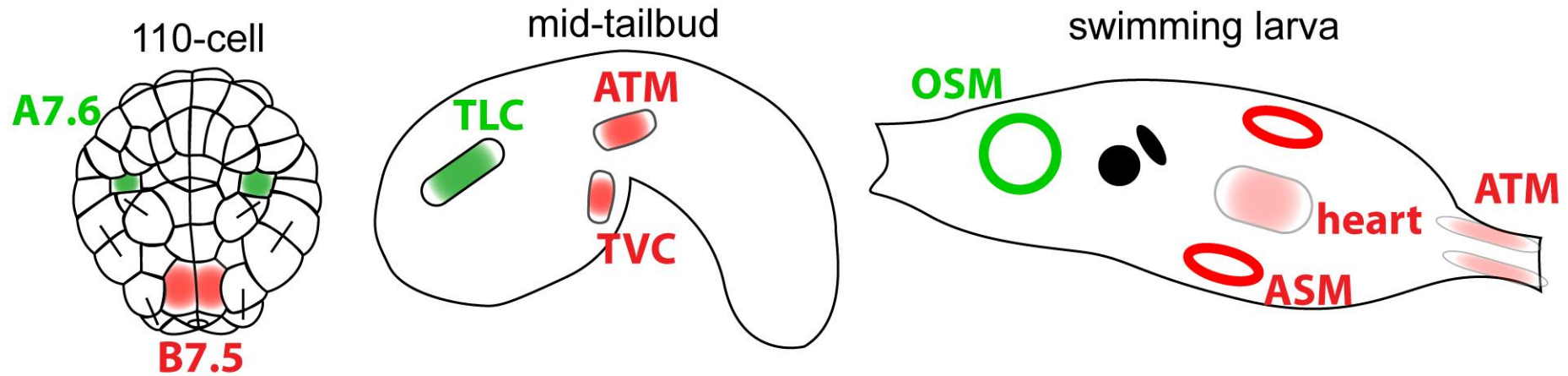
Col6a3



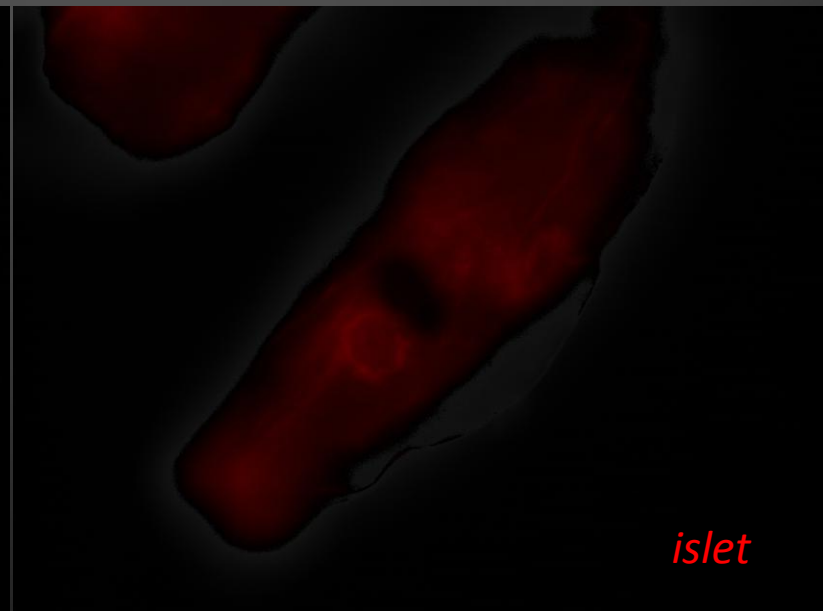
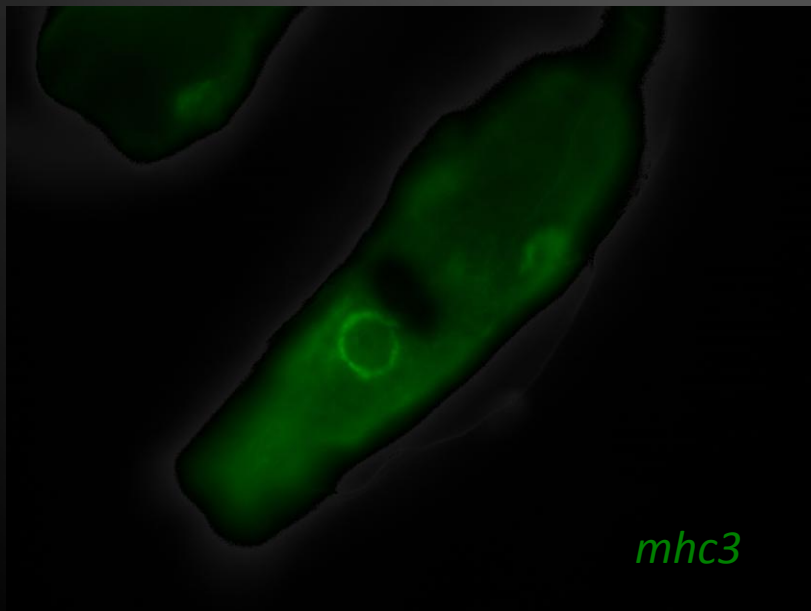
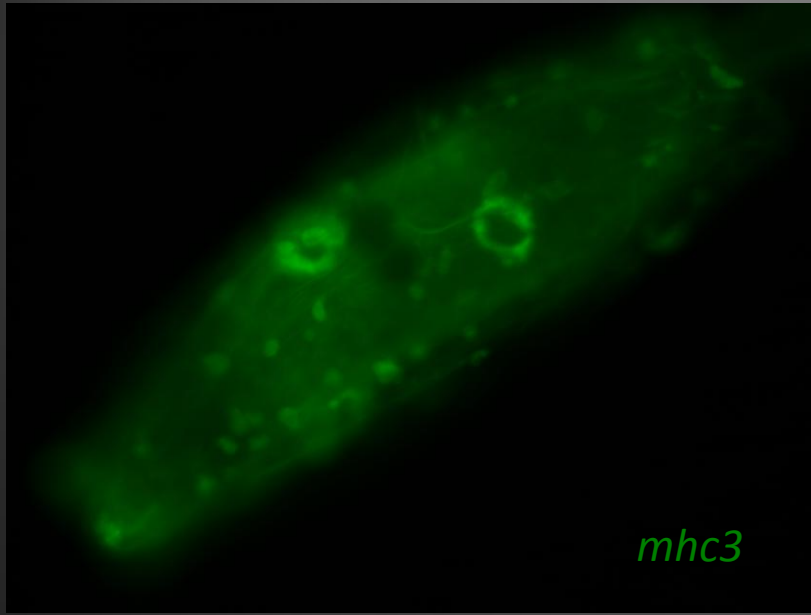
05H10 (no homolog)

Florian Razy

ASM and Oral Siphon Muscles (OSM) derive from distinct lineages



COE, islet and MHC3 are co-expressed in ASM and OSM



The COE enhancer is expressed

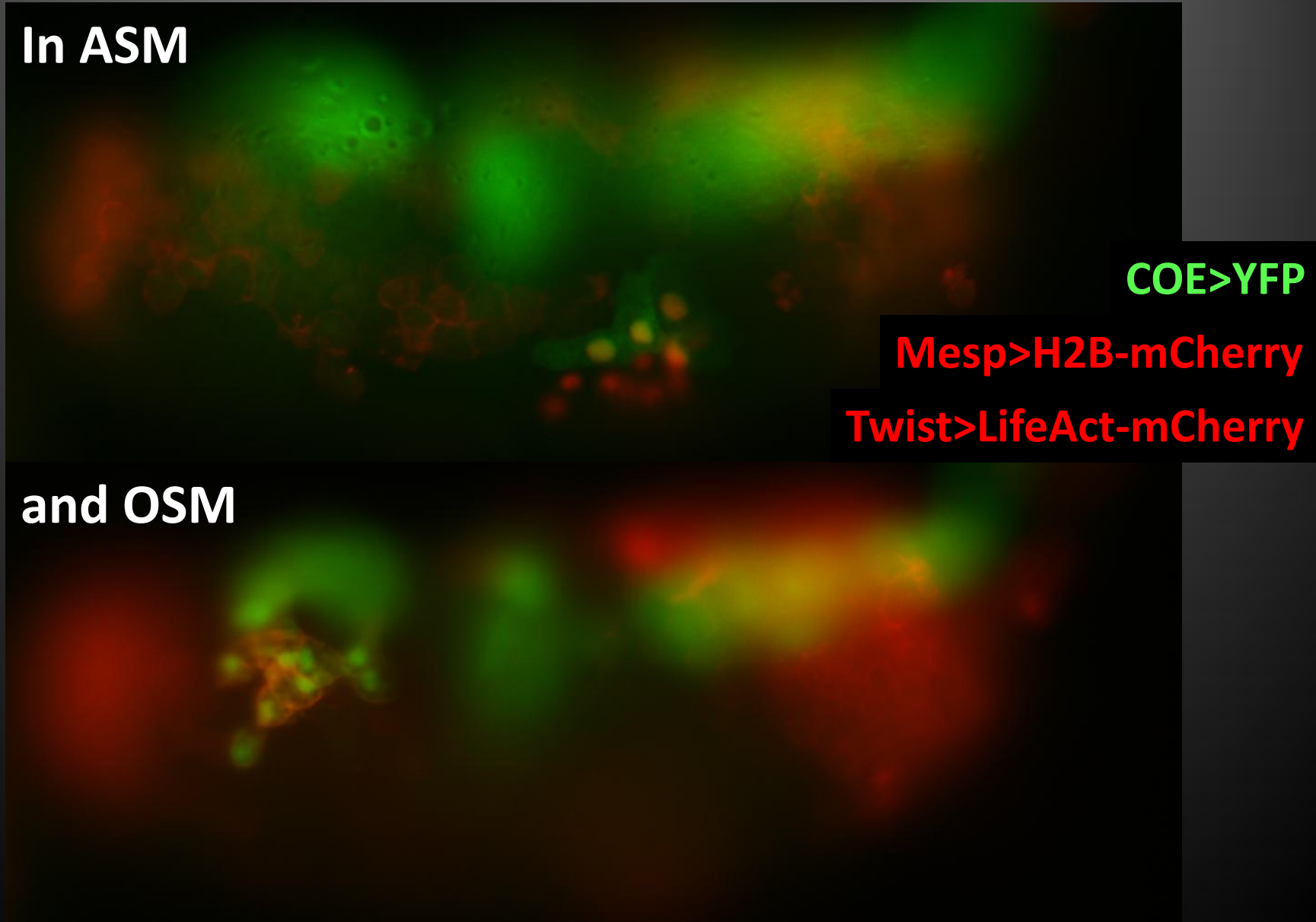
In ASM

COE>YFP

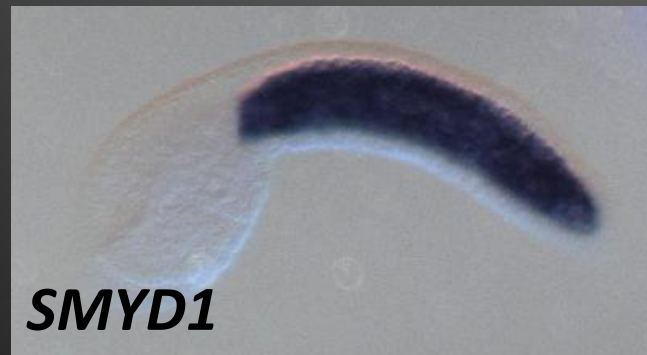
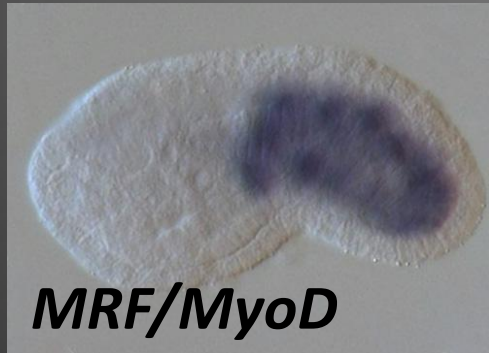
Mesp>H2B-mCherry

Twist>LifeAct-mCherry

and OSM



Skeletal Muscle regulators are expressed in muscle precursors but not *COE* and *Islet*



Independent activation of a “muscle regulatory module” in the ASM and

OSM(?)

I-ary and II-ary Muscles

OSM

ASM

ATM

A7.6/TLC inputs

B7.5/TVC inputs

Macho-1, ZicL
Tbx6 genes

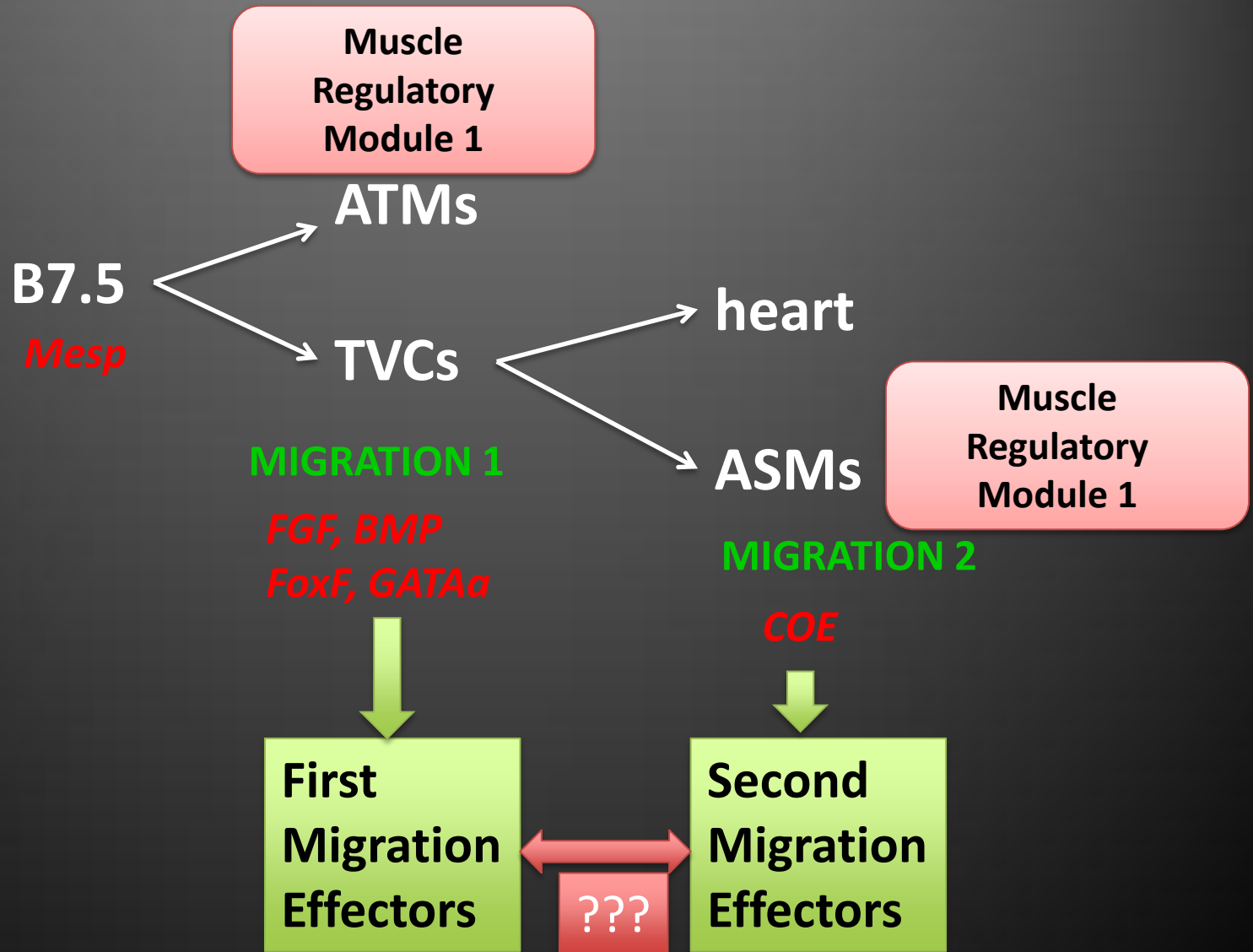
COE, Islet
Siphon Muscle plug-in

Generic Muscle Regulators
(MyoD, MEF2, SMYD1, Paraxis)

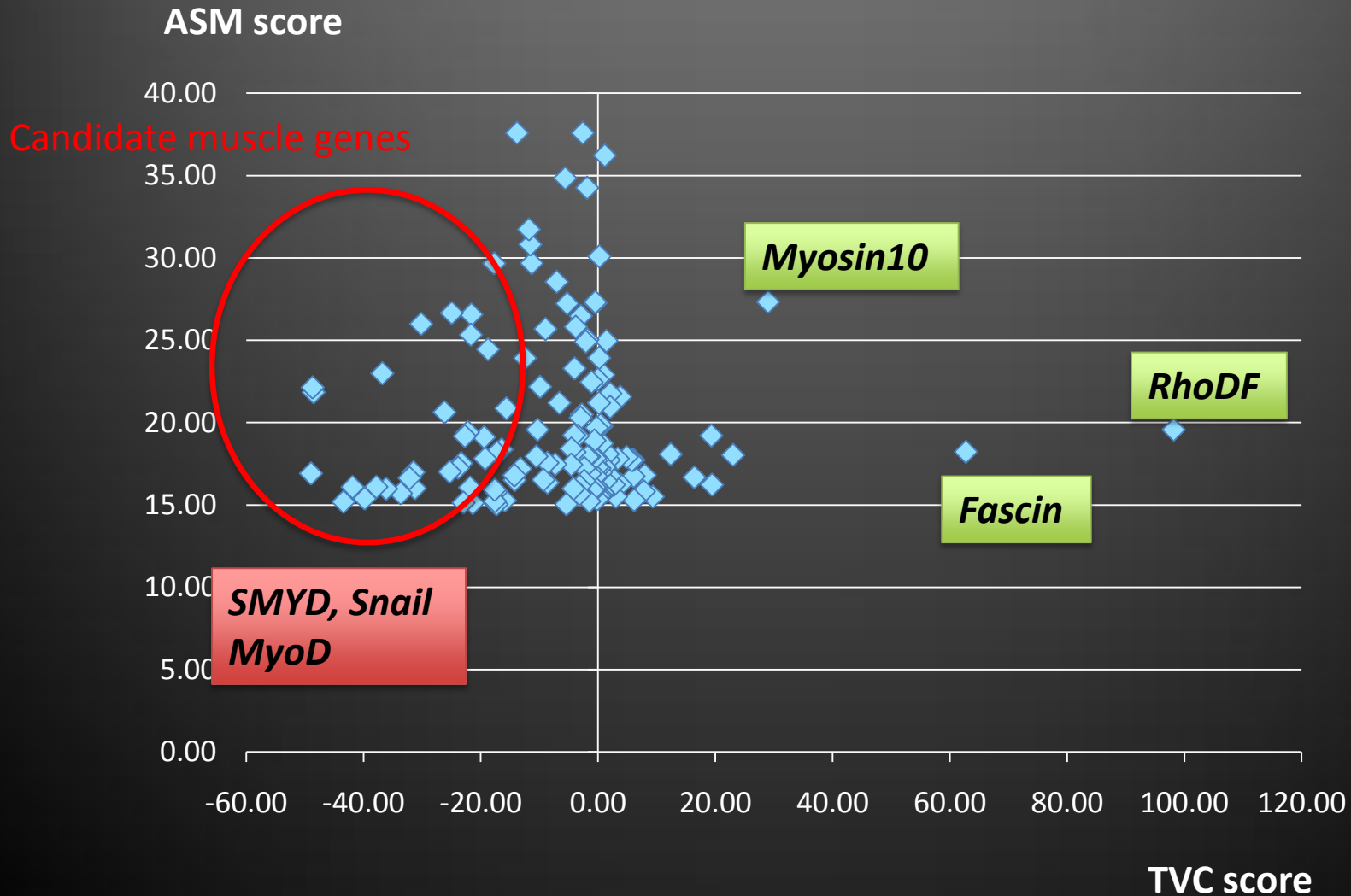
Muscle Differentiation Genes
(Actin, Myosin, Tropomyosin, etc...)

Muscle Regulatory Module 1

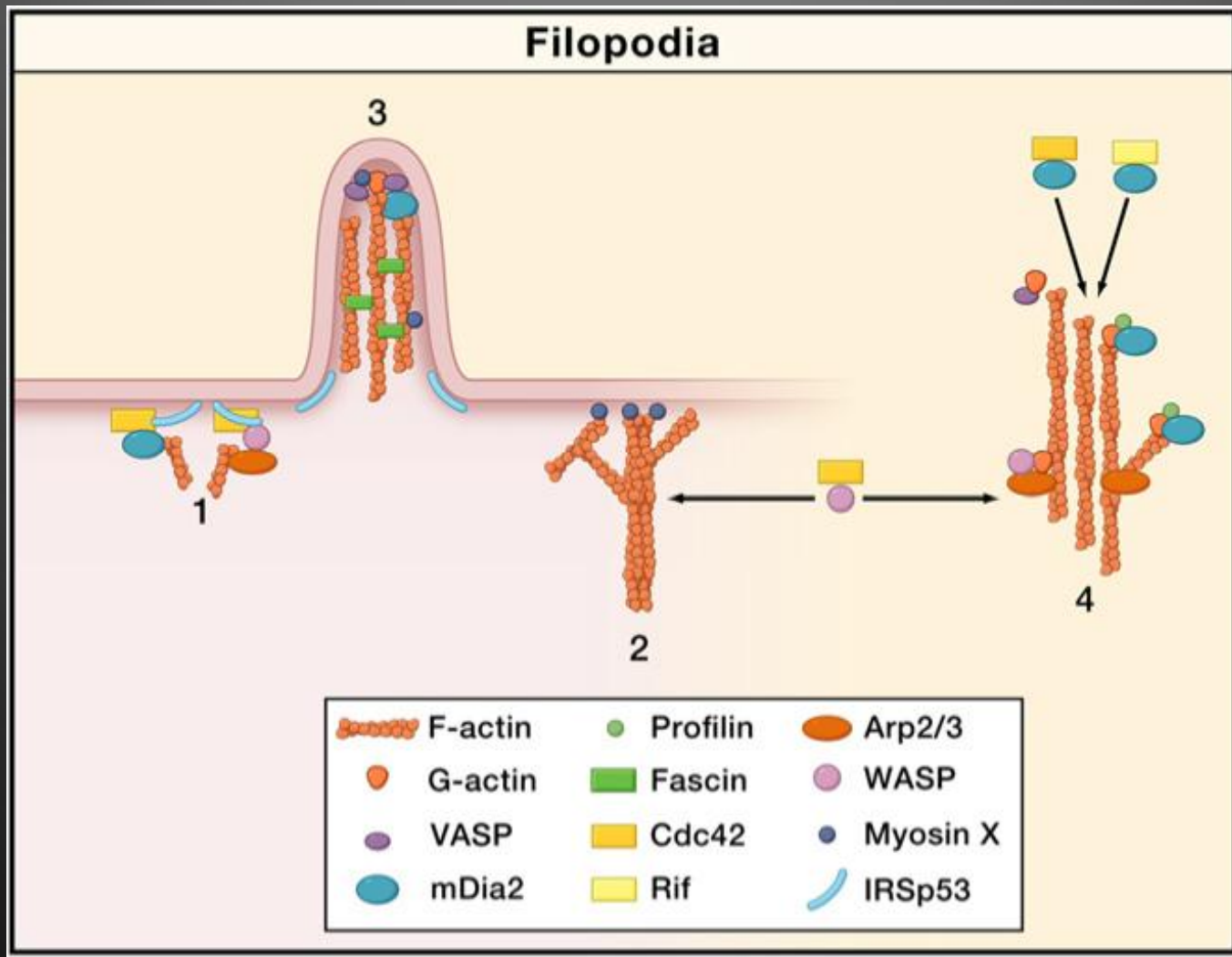
Searching for migration gene batteries

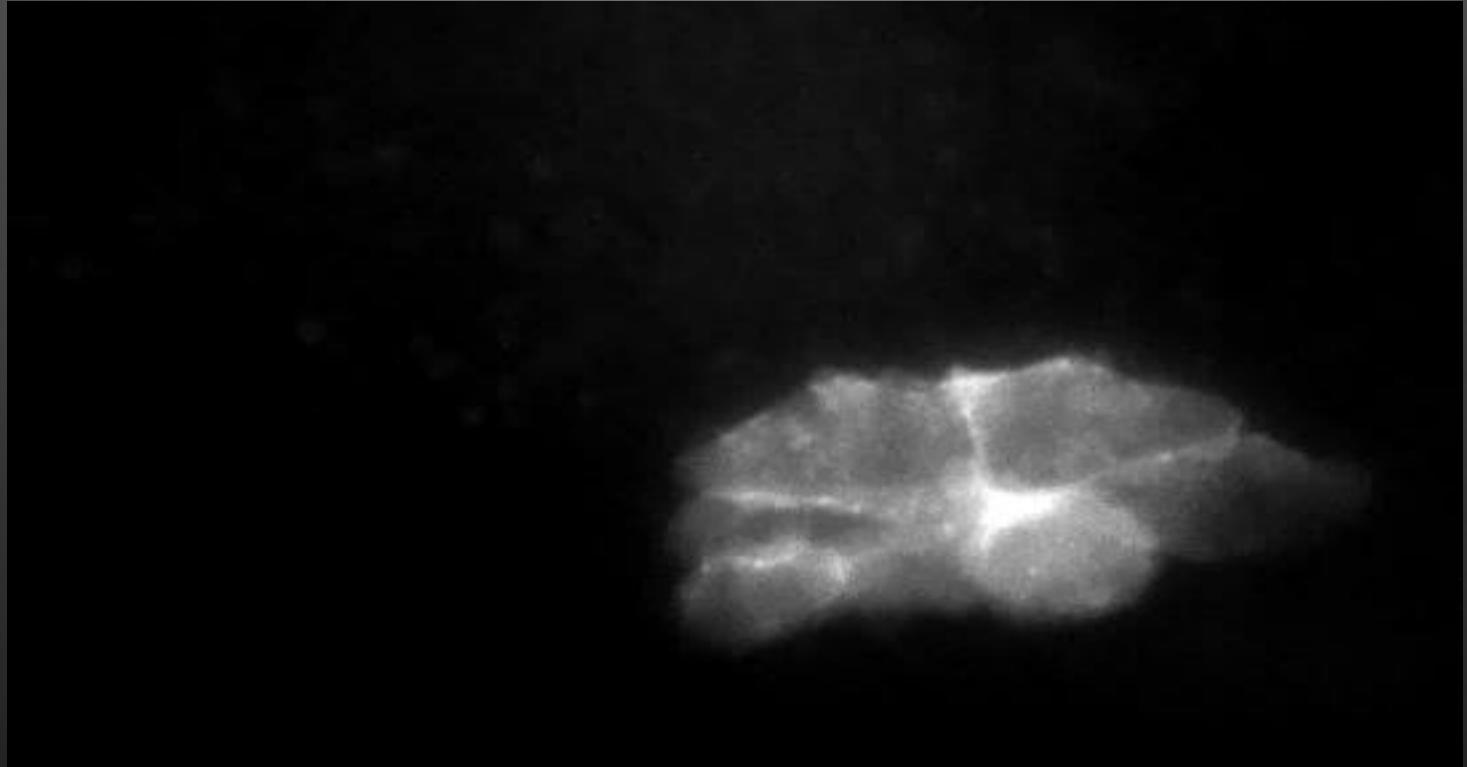


Most ASM candidates have low TVC scores

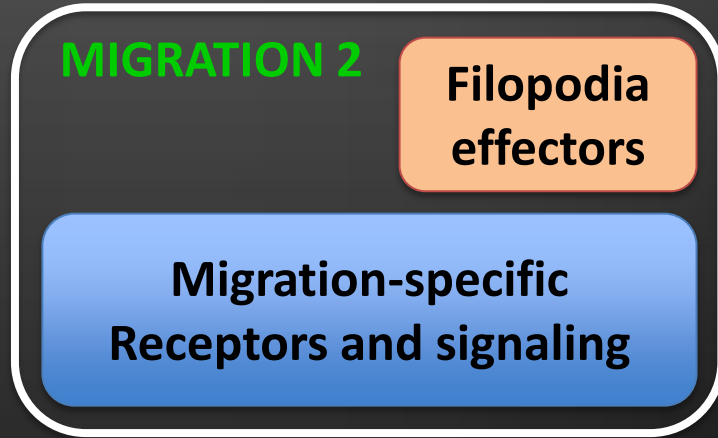
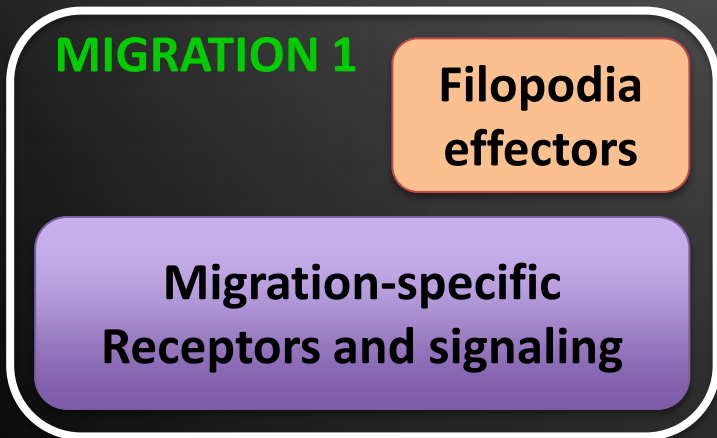
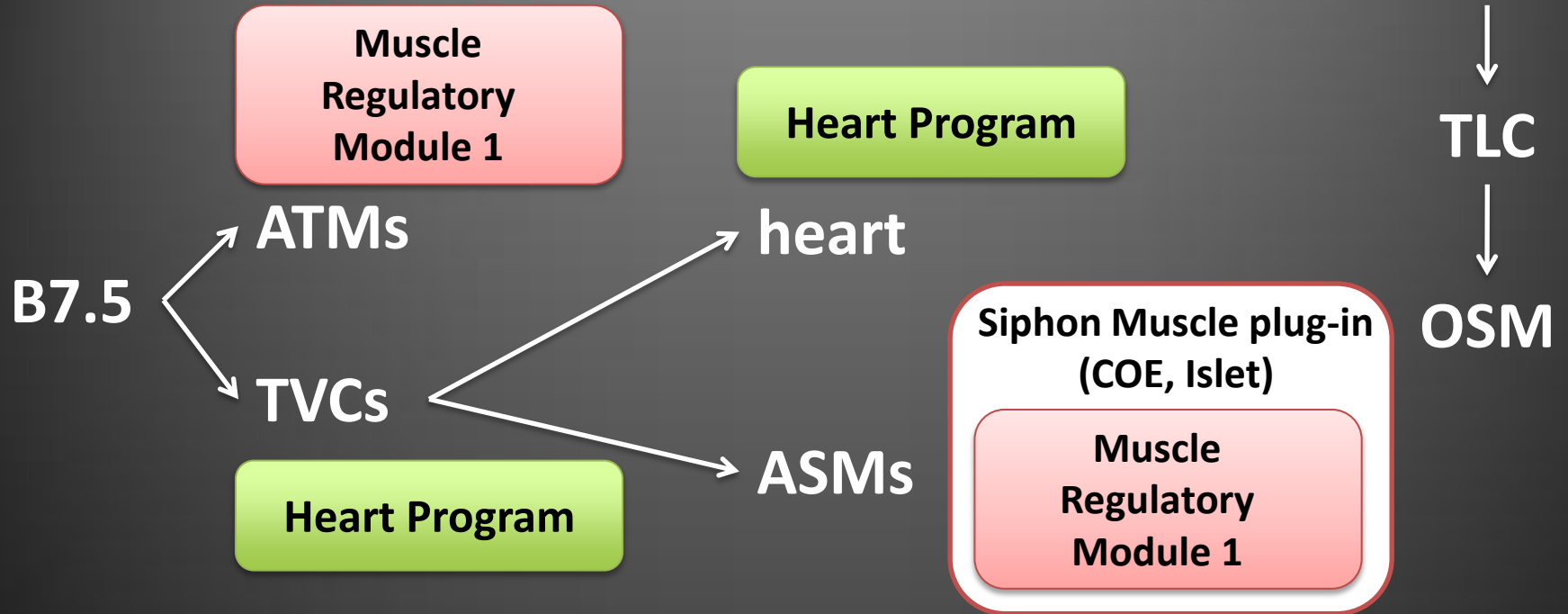


RhoDF, Fascin and Myosin10 homologs have been implicated in the formation of filopodia





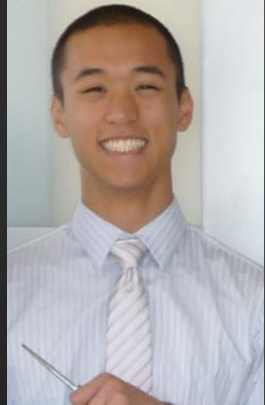
summary



Acknowledgements

Eric

(COE enhancer)



Ali
(Notch)



Florian
(FACS/array)



Renee
(HESb)



Theadora



Emily

Yusuff



Yasmin



Katherine



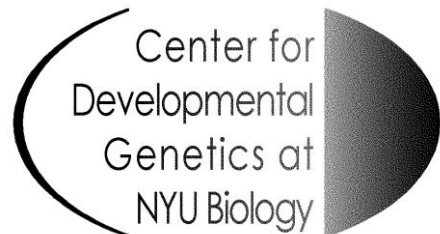
Wei



Dylan



Nikki



Levine lab
Alberto Stolfi
T. Blair Gainous
Mike Levine