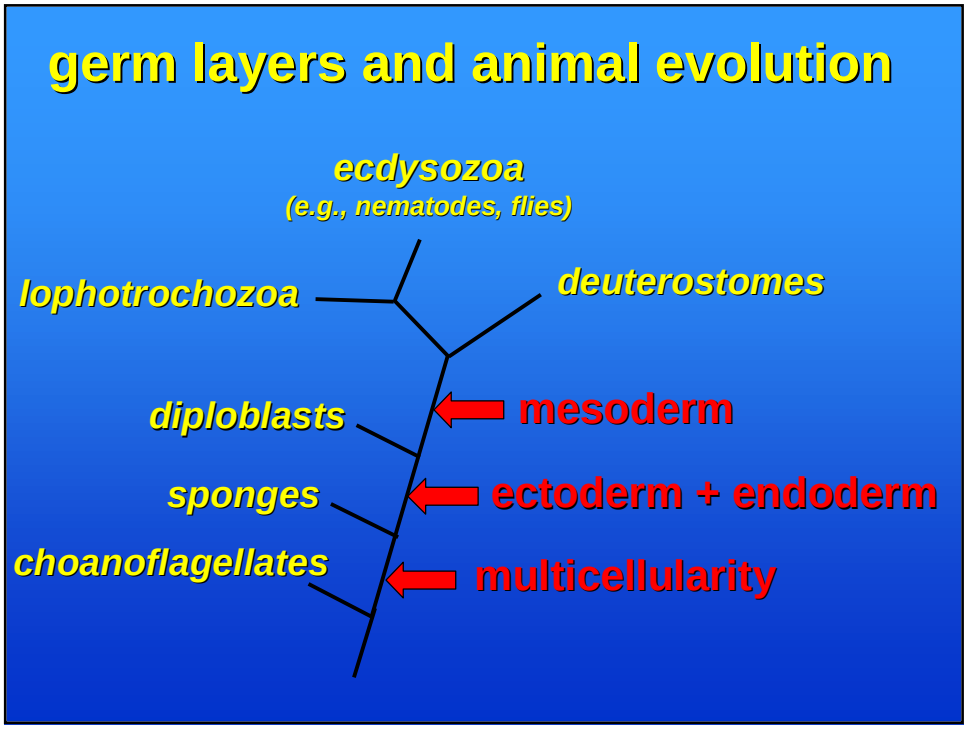
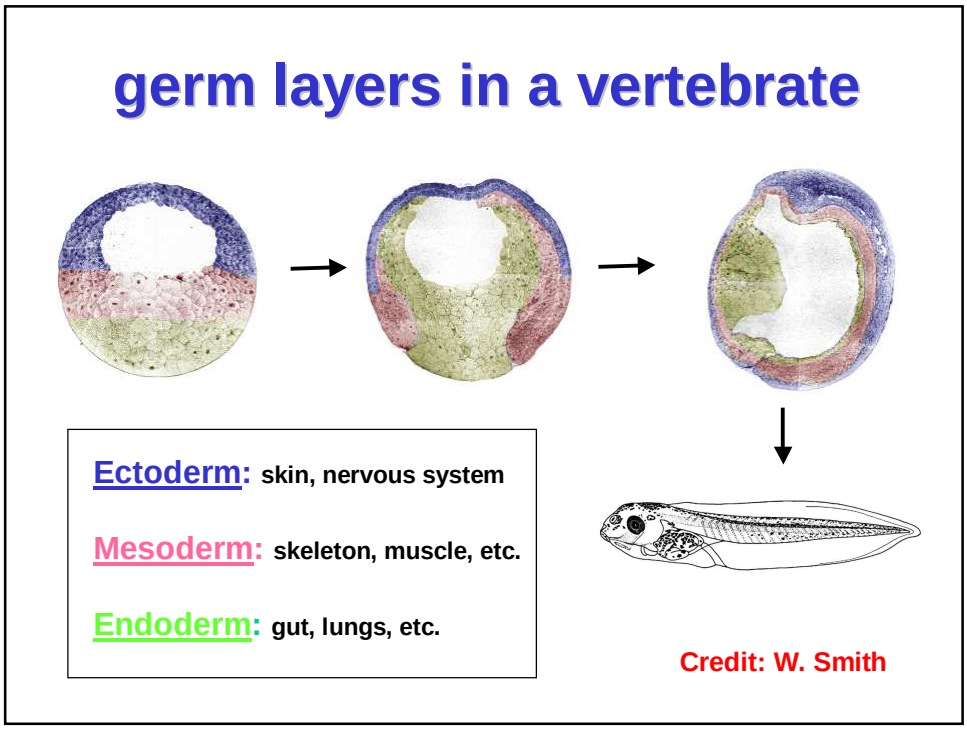

germ layers and animal evolution
ecdysozoa
(e.g., nematodes, flies)
lophotrochozoa
deuterostomes
diploblasts
mesoderm
sponges
ectoderm + endoderm
choanoflagellates
multicellularity


germ layers in a vertebrate
Ectoderm: skin, nervous system
Mesoderm: skeleton, muscle, etc.
Endoderm: gut, lungs, etc.
Credit: W. Smith

Quantum cosmology

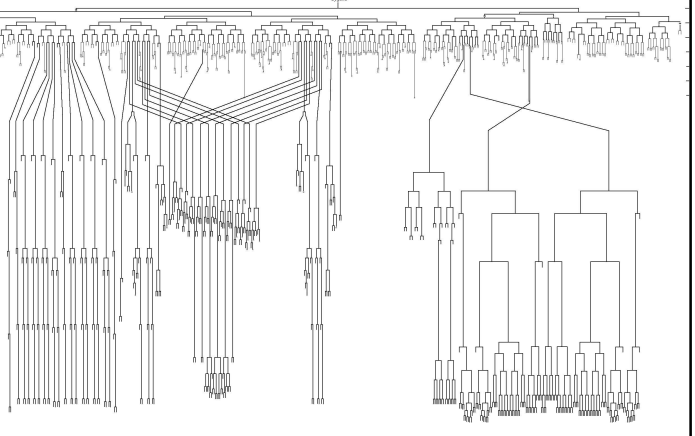
indeterminate

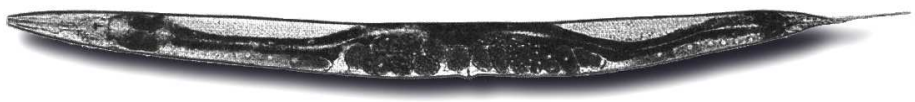
Nematode development

determinate

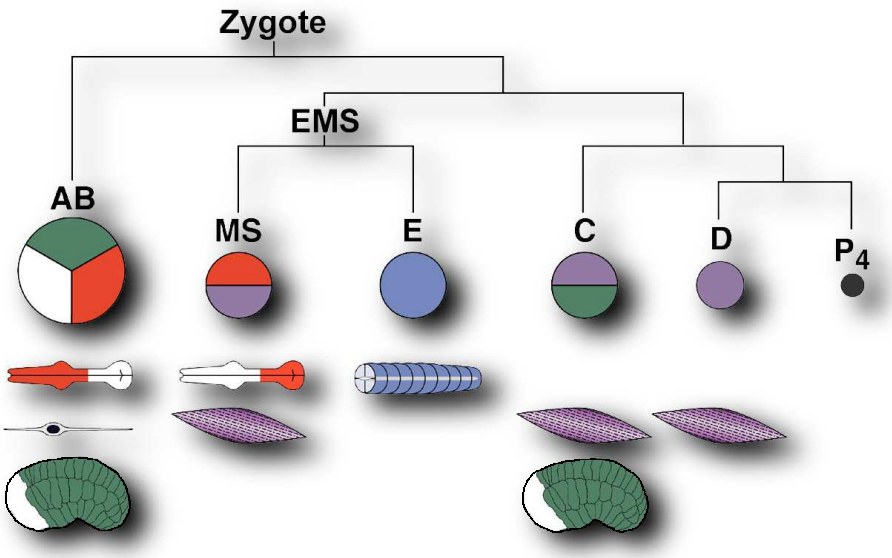
determinacy of *C. elegans* development

Sir John Sulston

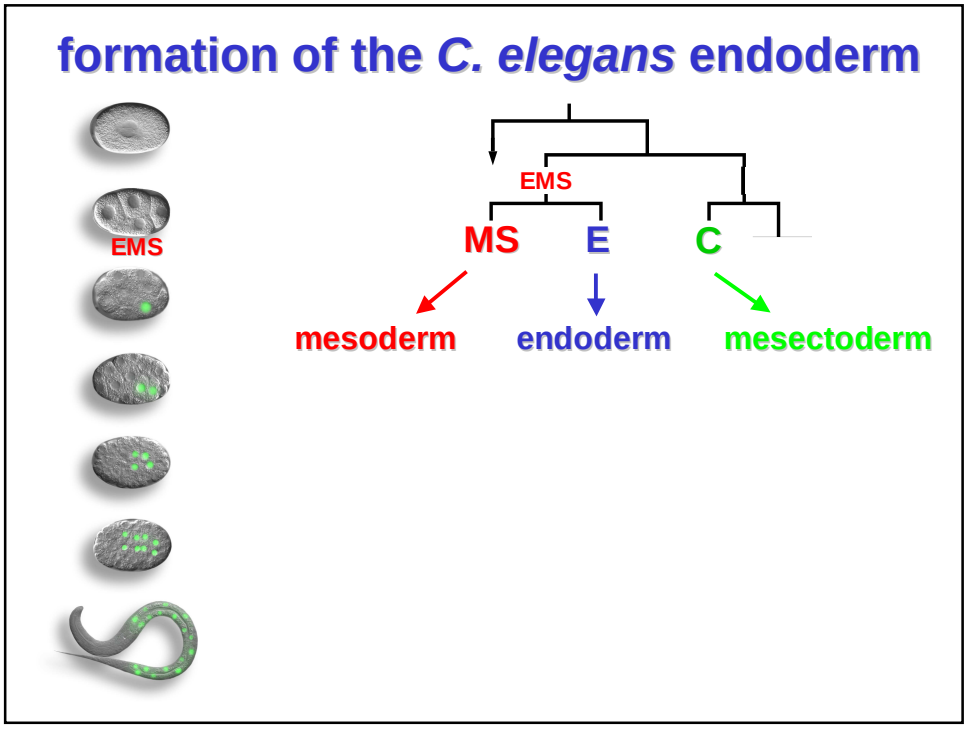


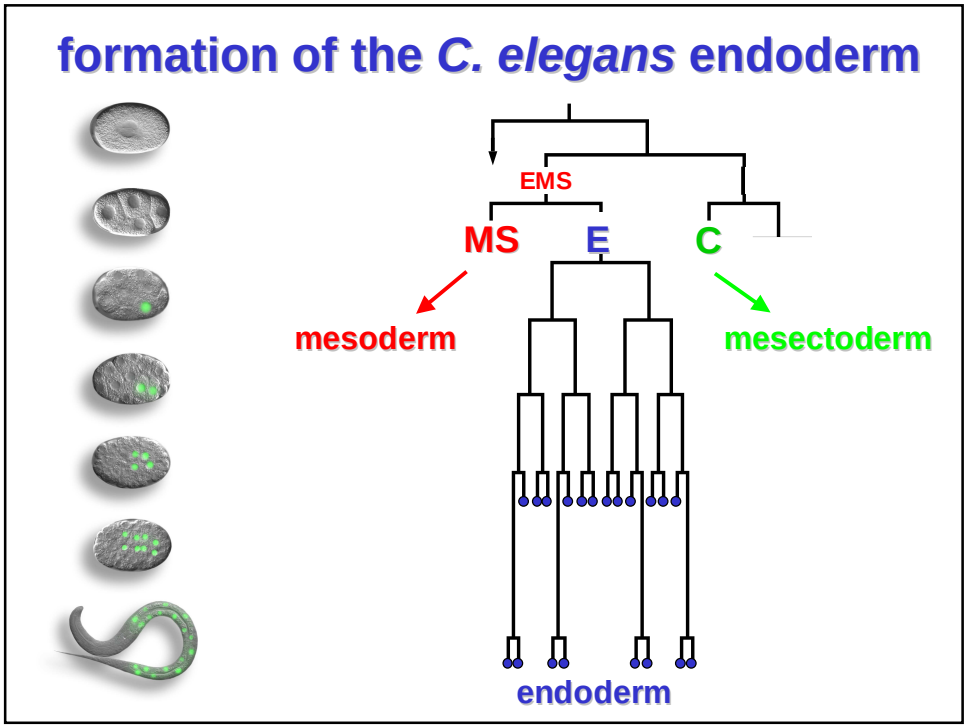


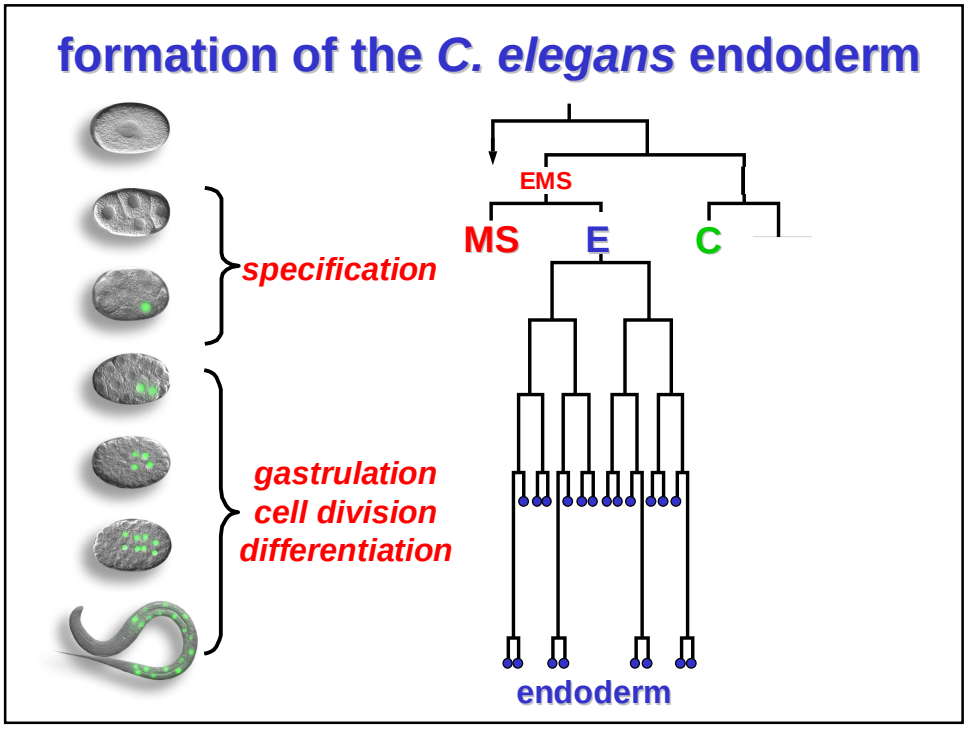
Major cell types made by founder cells

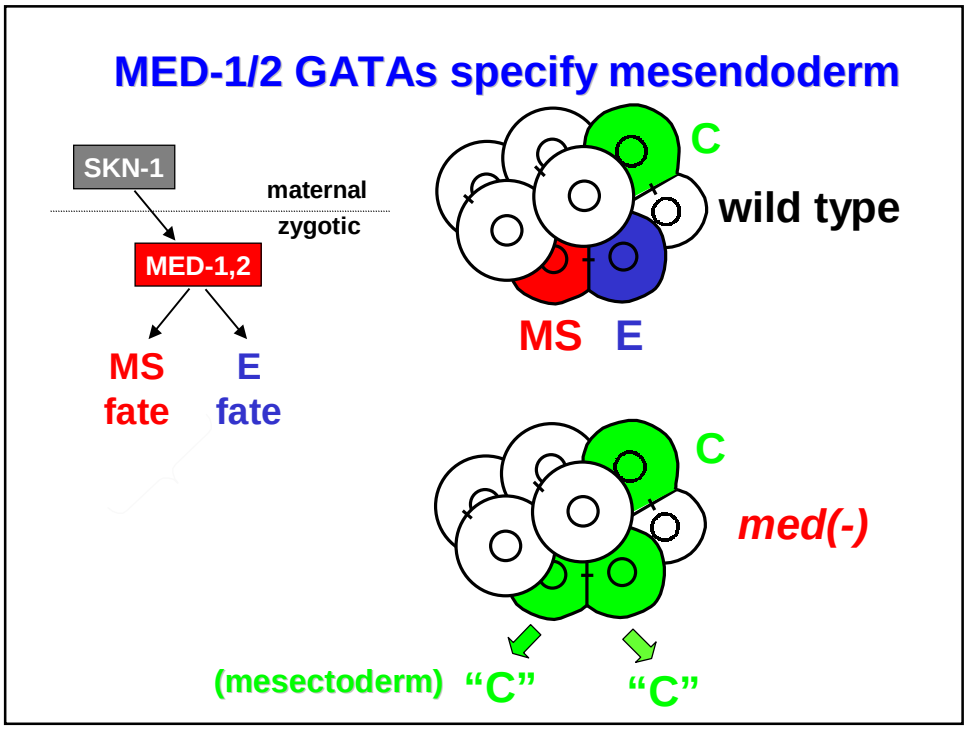


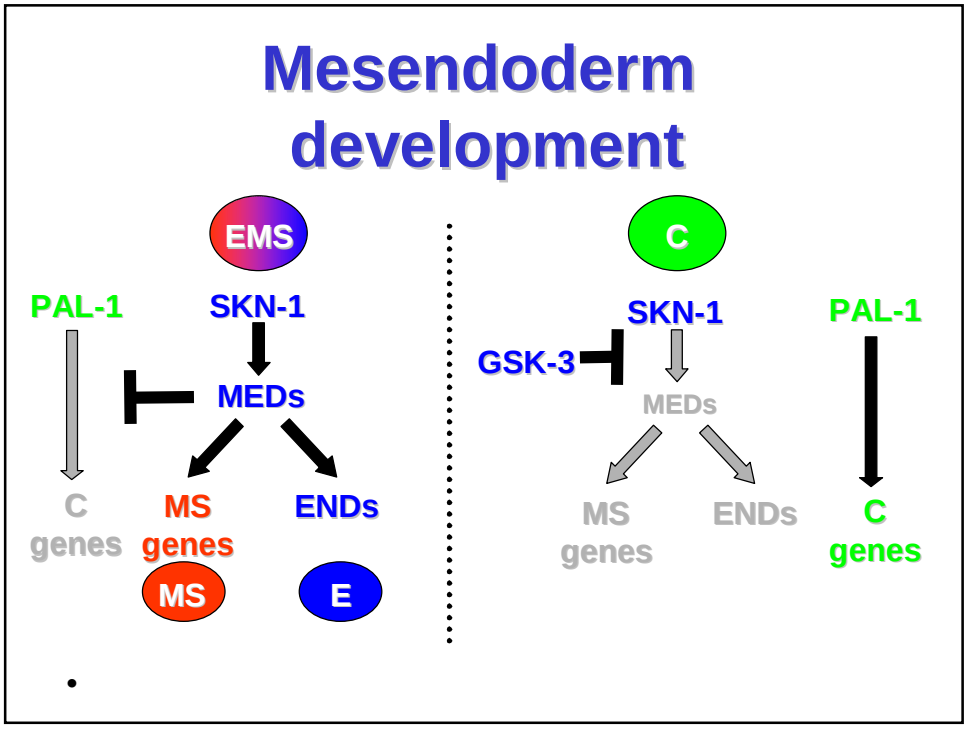
Gene regulatory cascade for mesendoderm

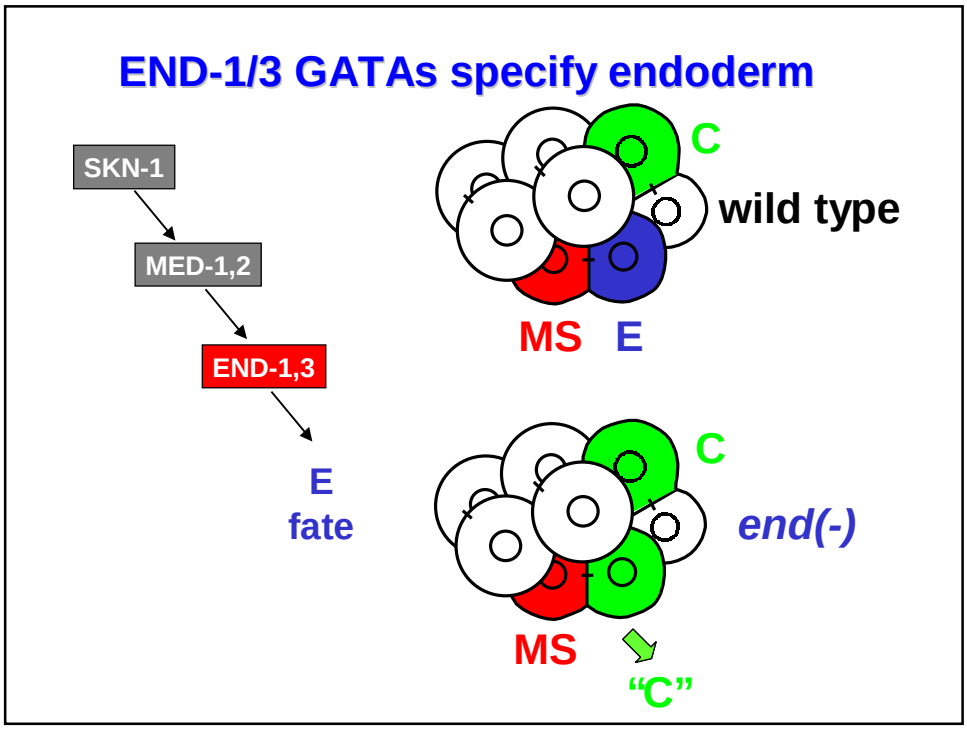

formation of the C. elegans endoderm
EMS
EMS
MS
E
C
mesoderm
endoderm
mesectoderm


formation of the C. elegans endoderm
EMS
MS
E
C
mesoderm
mesectoderm
endoderm


formation of the C. elegans endoderm
specification
gastrulation
cell division
differentiation
EMS
MS
E
C
endoderm

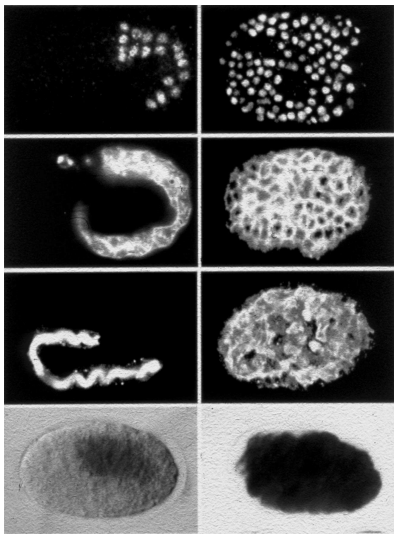

MED-1/2 GATAs specify mesendoderm
SKN-1
maternal
zygotic
MED-1,2
MS
fate
E
fate
C
wild type
MS
E
C
med(-)
(mesectoderm)
"C"
"C"


Mesendoderm development
EMS
PAL-1
SKN-1
MEDs
C genes
MS genes
ENDs
MS
E
C
SKN-1
GSK-3
MEDs
MS genes
ENDs
PAL-1
C genes


END-1/3 GATAs specify endoderm
SKN-1
MED-1,2
END-1,3
E fate
C
wild type
MS E
C
end(-)
MS
"C"

END-1/3 specify endoderm

SKN-1 → MED-1,2 → END-1,3 → E fate



wild type ubiquitous

END-1/3 specify endoderm

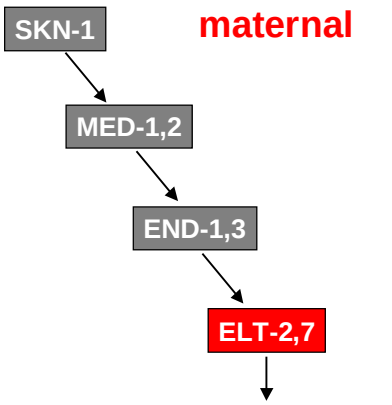
SKN-1 → MED-1,2 → END-1,3 → ELT-2,7 → E fate

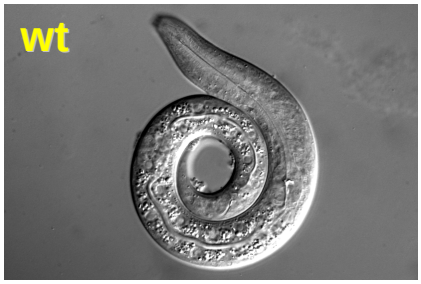
WT

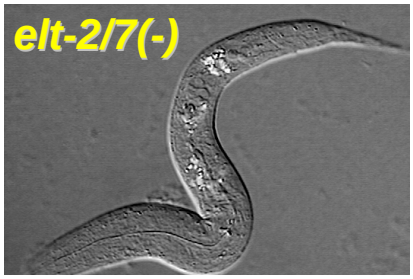
END-1::EnR

Shoichet, S.A., Malik, T.H., Rothman, J.H., and Shivdasani, R.A. *PNAS* 97, 4076 (2000)

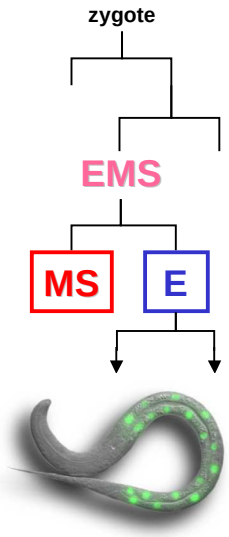
ELT-2/7 GATAs control gut differentiation

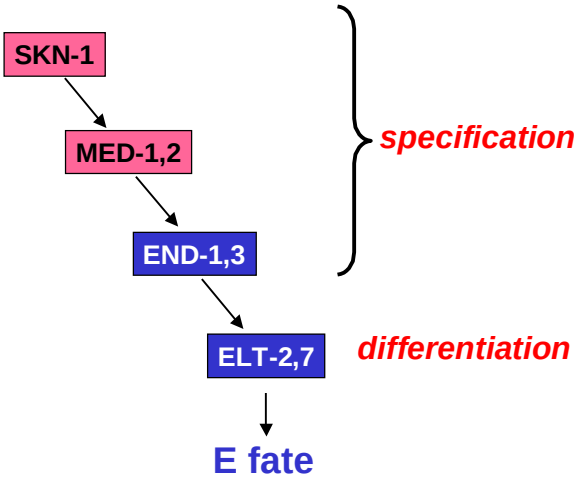


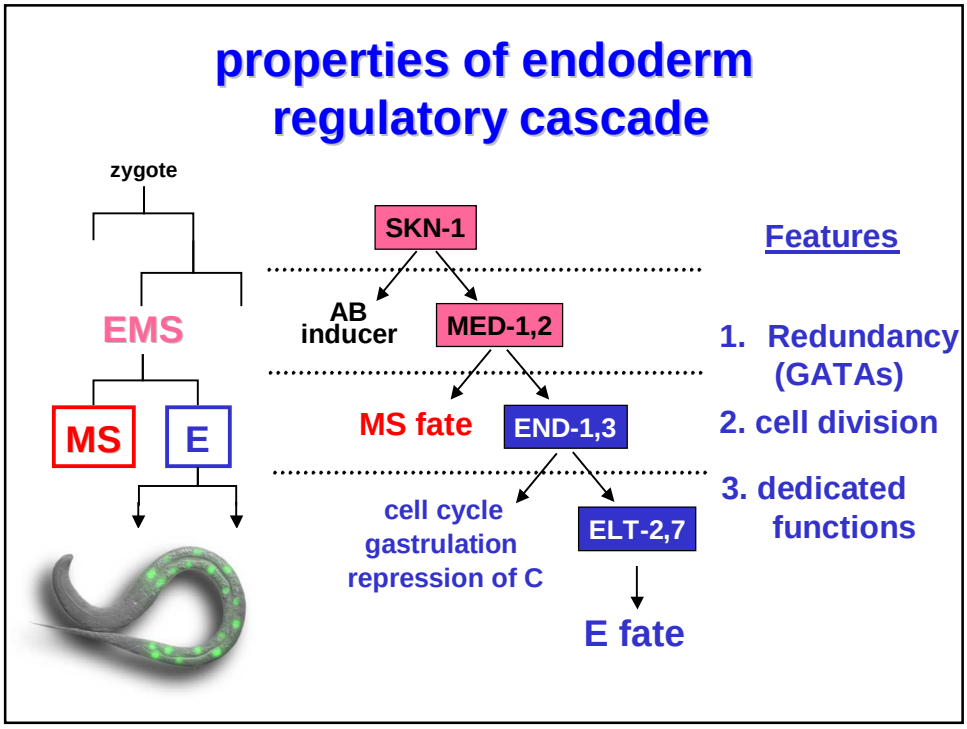


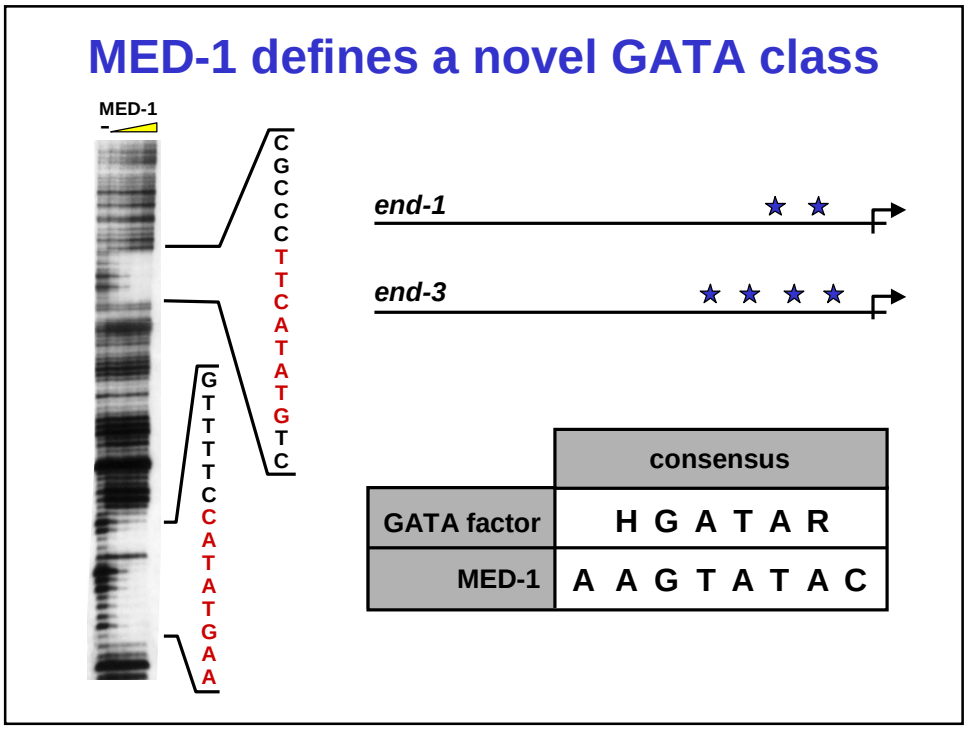


properties of endoderm regulatory cascade






properties of endoderm regulatory cascade
zygote
EMS
MS
E
SKN-1
AB inducer
MED-1,2
MS fate
END-1,3
cell cycle
gastrulation
repression of C
ELT-2,7
E fate
Features
1. Redundancy (GATAs)
2. cell division
3. dedicated functions

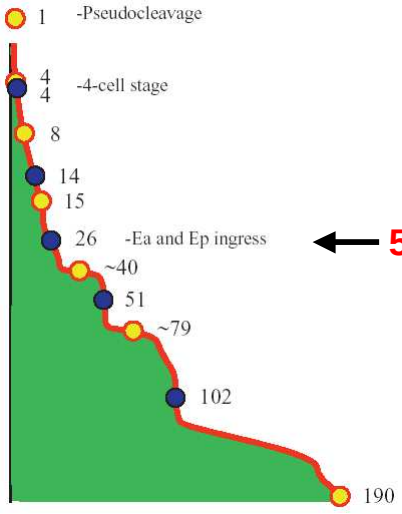

MED-1 defines a novel GATA class
MED-1
CGCCCTTCATATGTC
GTTTTCCATATGAA
end-1
end-3
consensus
GATA factor
H G A T A R
MED-1
A A G T A T A C

fourteen putative MED targets

A A G T A T A C $N_{25\text{-}100}$ A A G T A T A C

gene	product	MED sites
F58E10.2	*end-1*	★ ★
F58E10.5	*end-3*	★ ★ ★ ★
F35H8.7	*wee-1.1*	★ ★ ★
ceh-20/F31E3.2	homeobox	★ ★
F58G4.4	LAG-2-like	★ ★
C32E12.5	Sox family (HMG)	★ ★ ★
ZK849.2	RCC1	★ ★
T07D1.2	unknown	★ ★
ZK177.10, ZK177.1	T-box/unknown	★ ★ ★ ★ ★ ★ ★ ★
T11A5.5	oxygen transport	★ ★ ★
C17C3.7, C17C3.10	bHLH (2)	★ ★
B0303.8, 9	unknown	★ ★ ★ ★

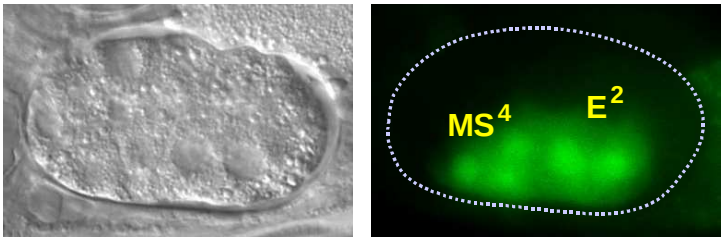
correlation with embryonic transcriptome

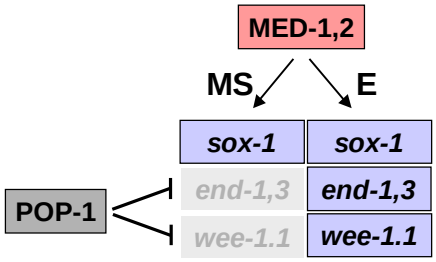


5/12 detected

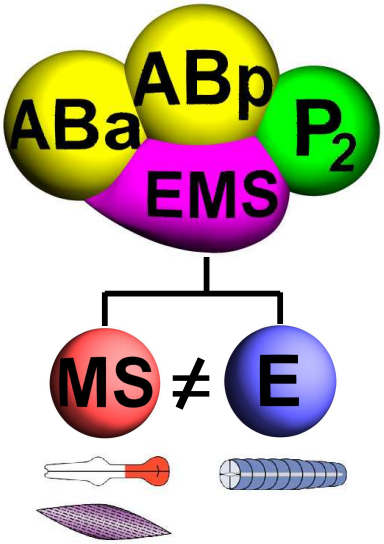
Baugh *et al.*, 2003

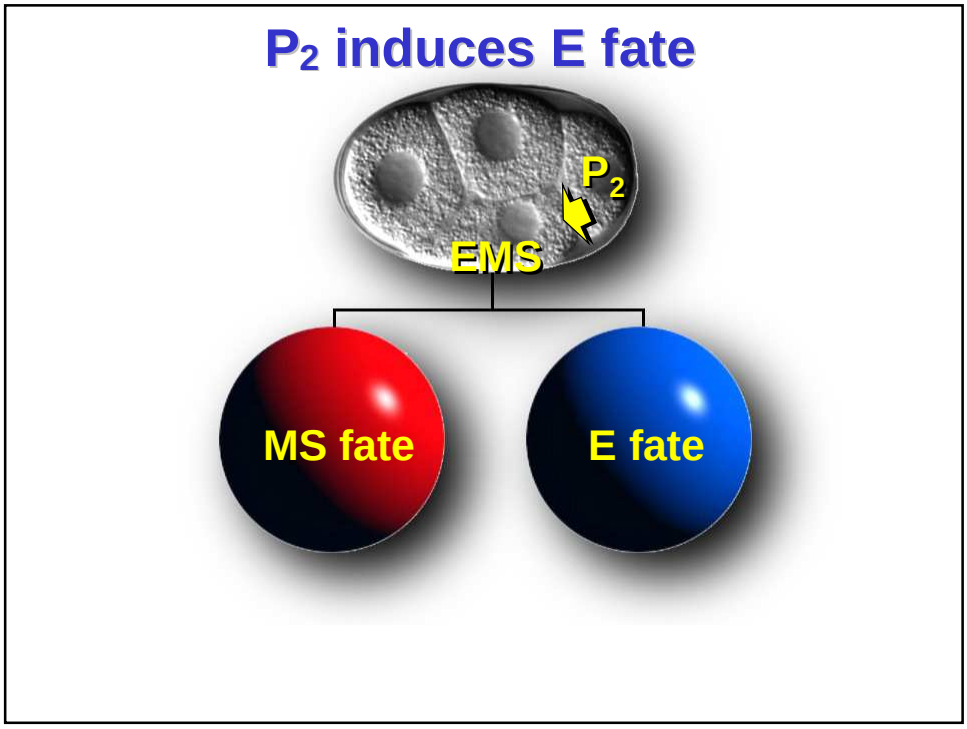
sox-1::GFP in E, MS descendants

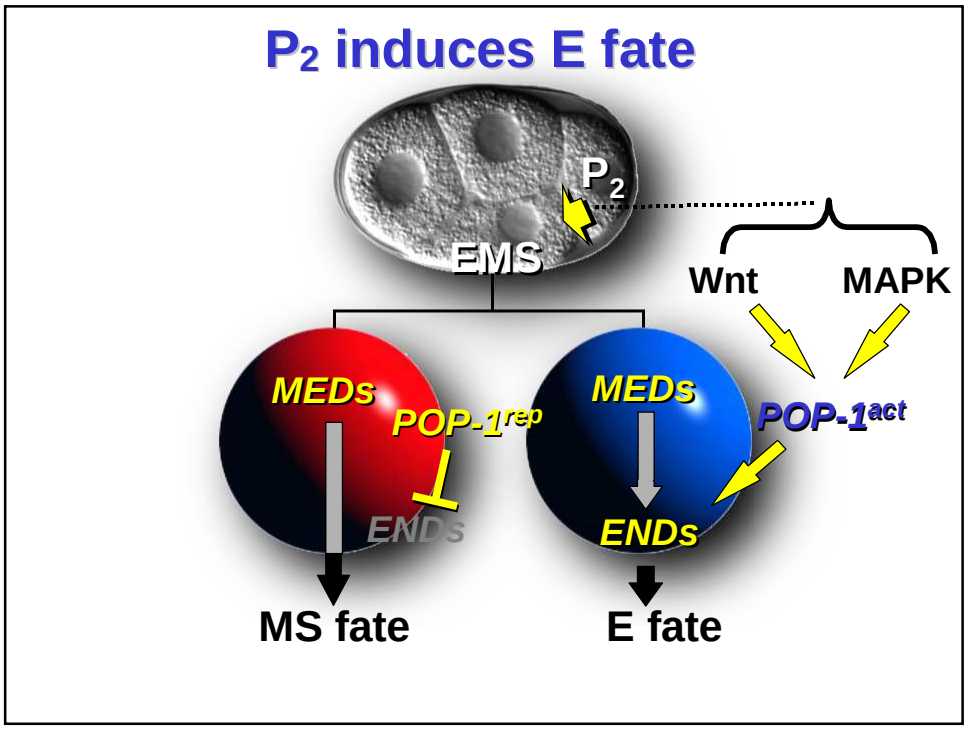




Asymmetric cell division



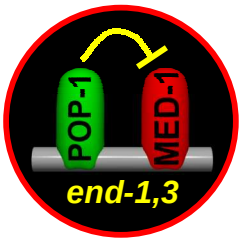

P2 induces E fate
P2
EMS
MS fate
E fate


P2 induces E fate
P2
EMS
Wnt
MAPK
MEDs
POP-1rep
ENDs
MEDs
POP-1act
ENDs
MS fate
E fate

Models for POP-1 repression

MS

MED displacement



MED inhibition

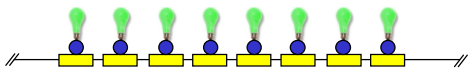
in vivo detection of protein-DNA interactions

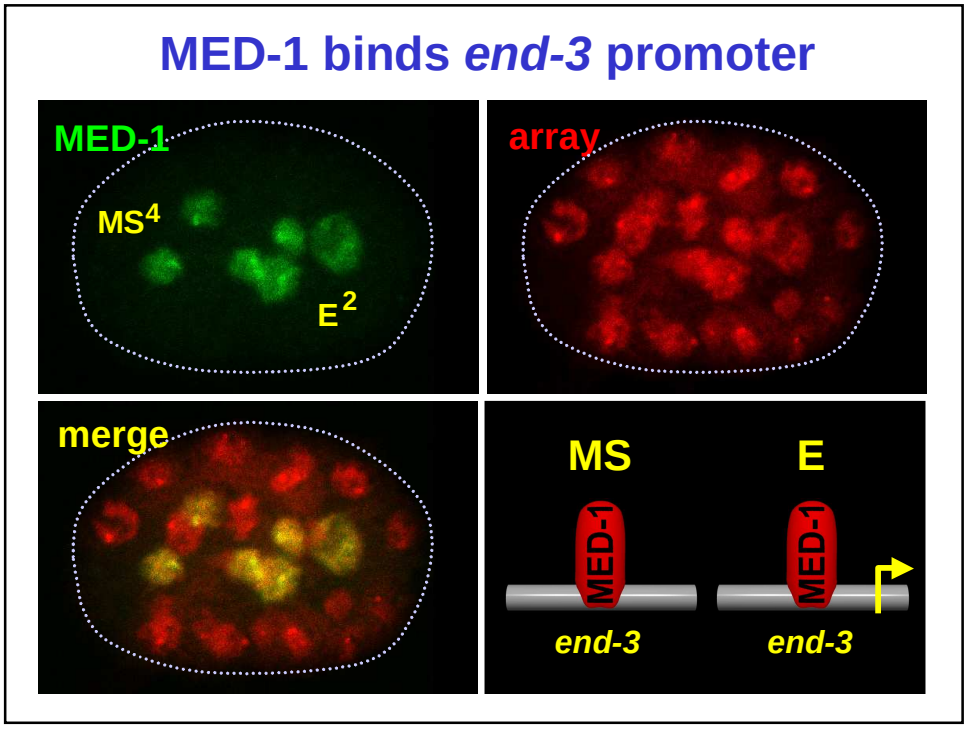
fusion protein

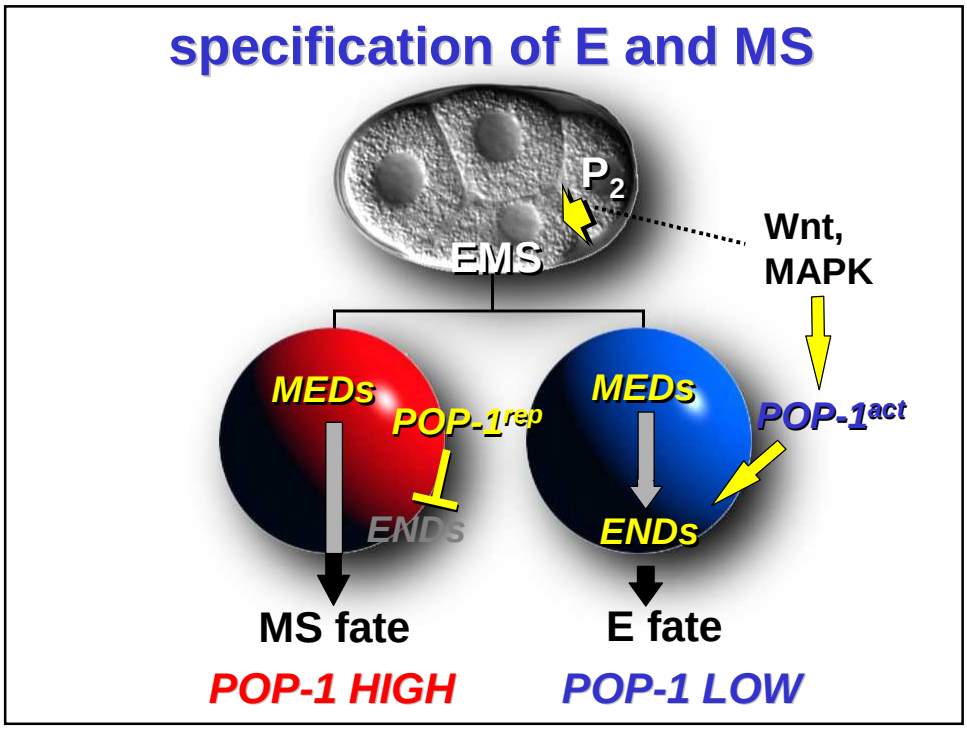
GFP

transcription factor

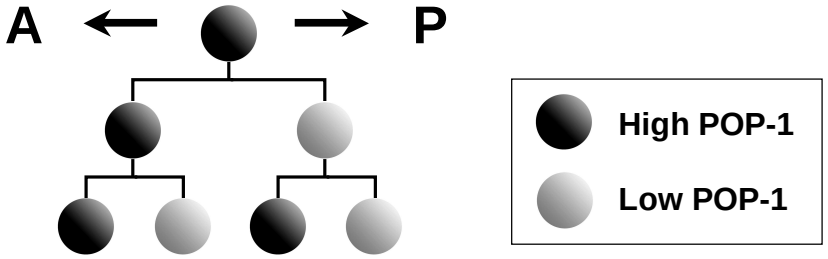
target array



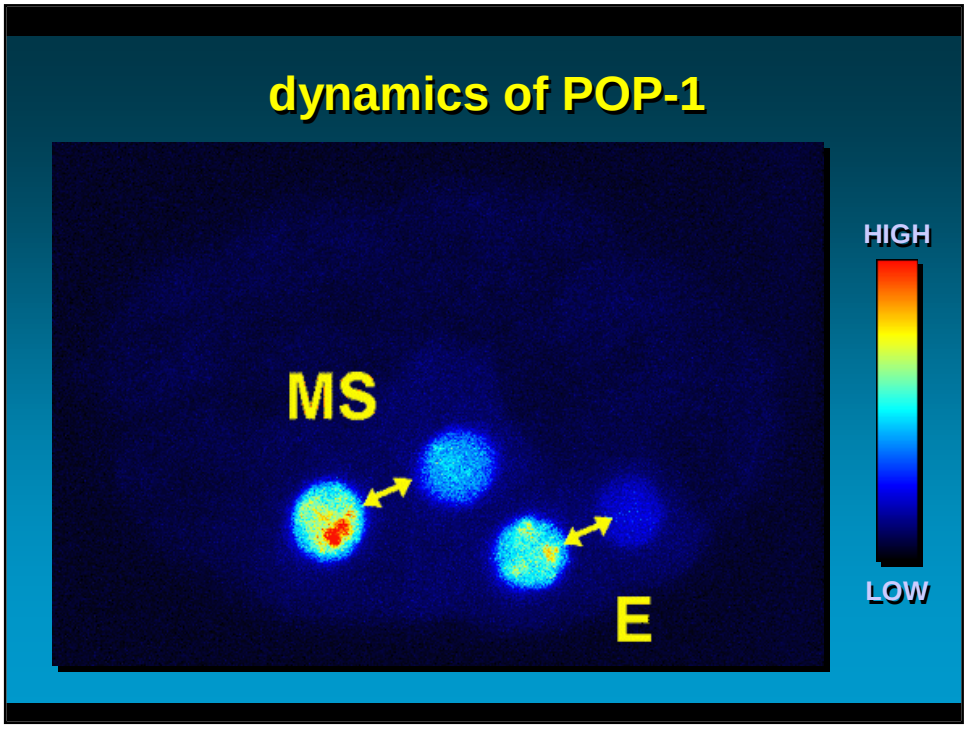

MED-1 binds end-3 promoter
MED-1
MS4
E2
array
merge
MS
E
MED-1
MED-1
end-3
end-3

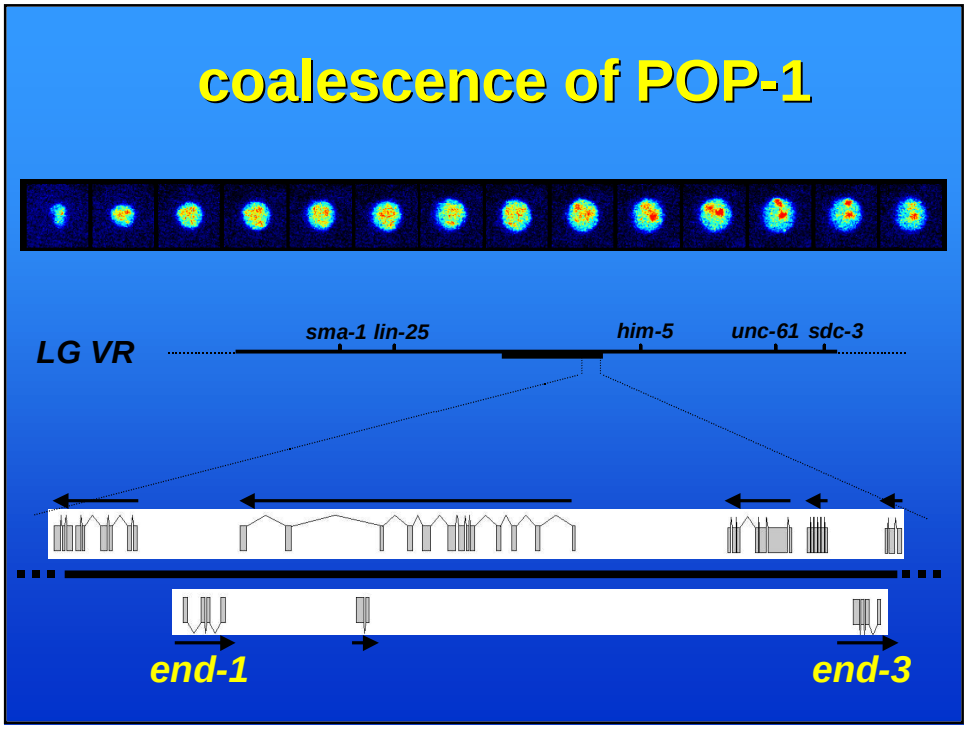

specification of E and MS
P2
EMS
Wnt,
MAPK
MEDs
POP-1rep
ENDs
MEDs
POP-1act
ENDs
MS fate
E fate
POP-1 HIGH
POP-1 LOW

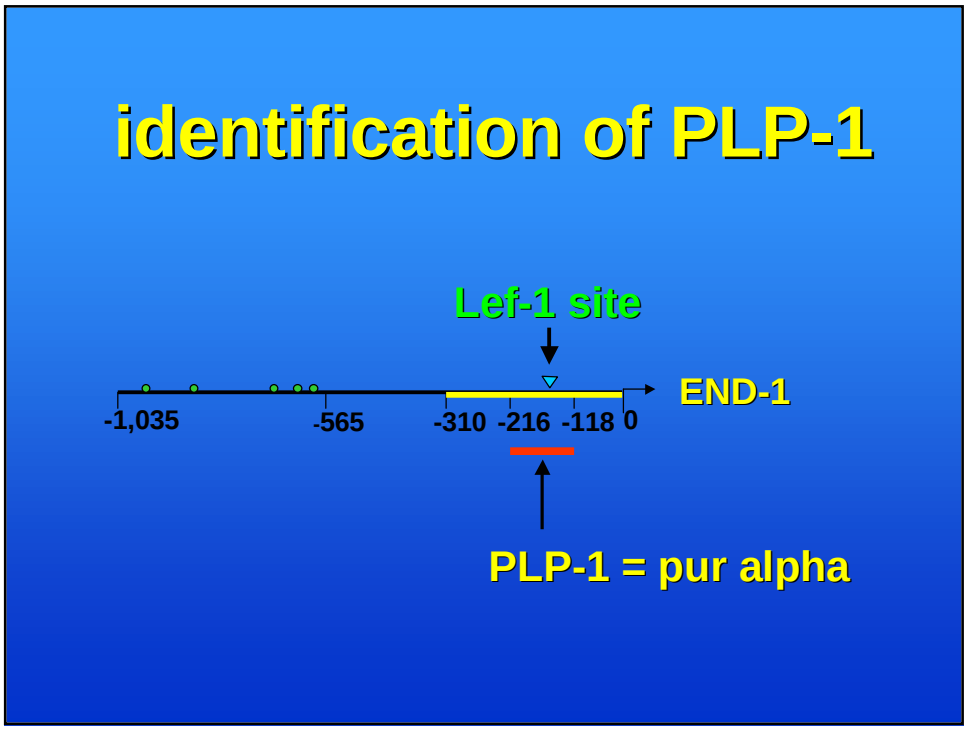
recursive POP-1 asymmetry



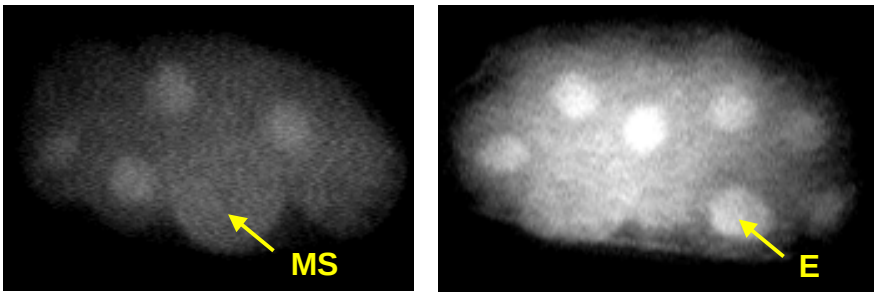
Lin *et al.* (1998) Cell *92*, 229–239



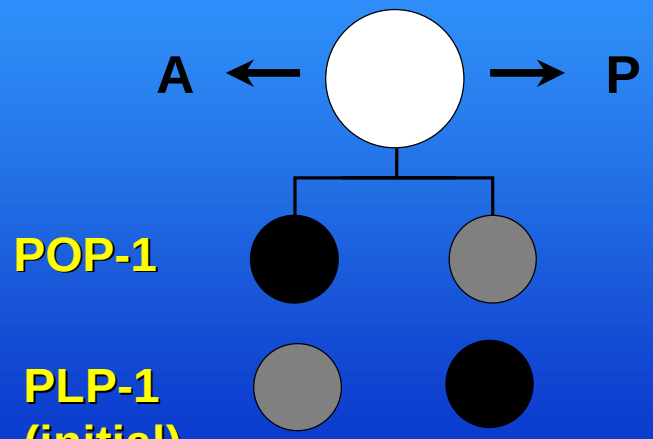

coalescence of POP-1
LG VR
sma-1 lin-25
him-5
unc-61 sdc-3
end-1
end-3

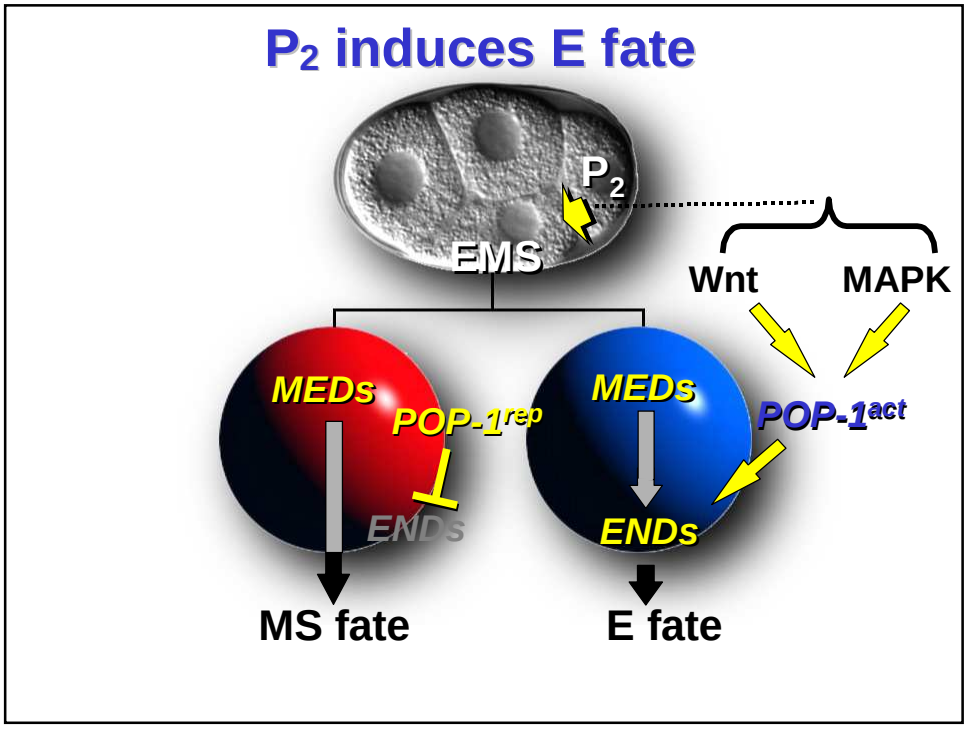

identification of PLP-1
Lef-1 site
END-1
-1,035
-565
-310
-216
-118
0
PLP-1 = pur alpha

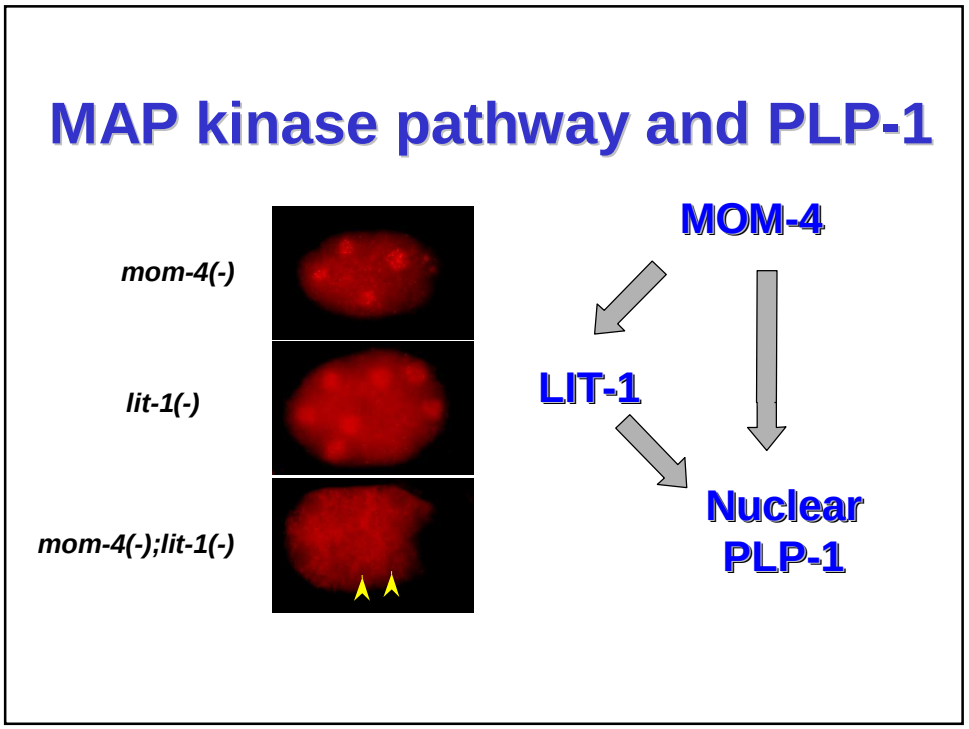
asymmetry of nuclear PLP-1



POP-1 and PLP-1 in asymmetry




P2 induces E fate
P2
EMS
Wnt
MAPK
MEDs
POP-1rep
MEDs
POP-1act
ENDs
ENDs
MS fate
E fate


MAP kinase pathway and PLP-1
mom-4(-)
lit-1(-)
mom-4(-);lit-1(-)
MOM-4
LIT-1
Nuclear
PLP-1

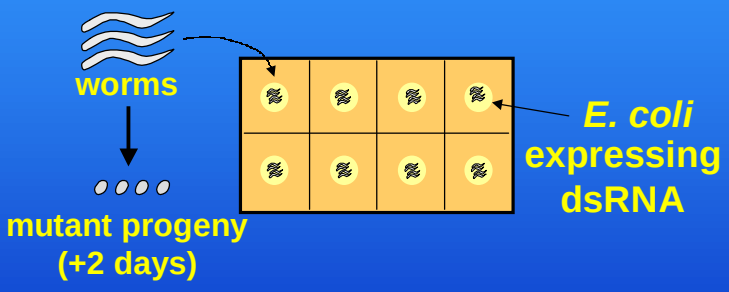
RNAi screen

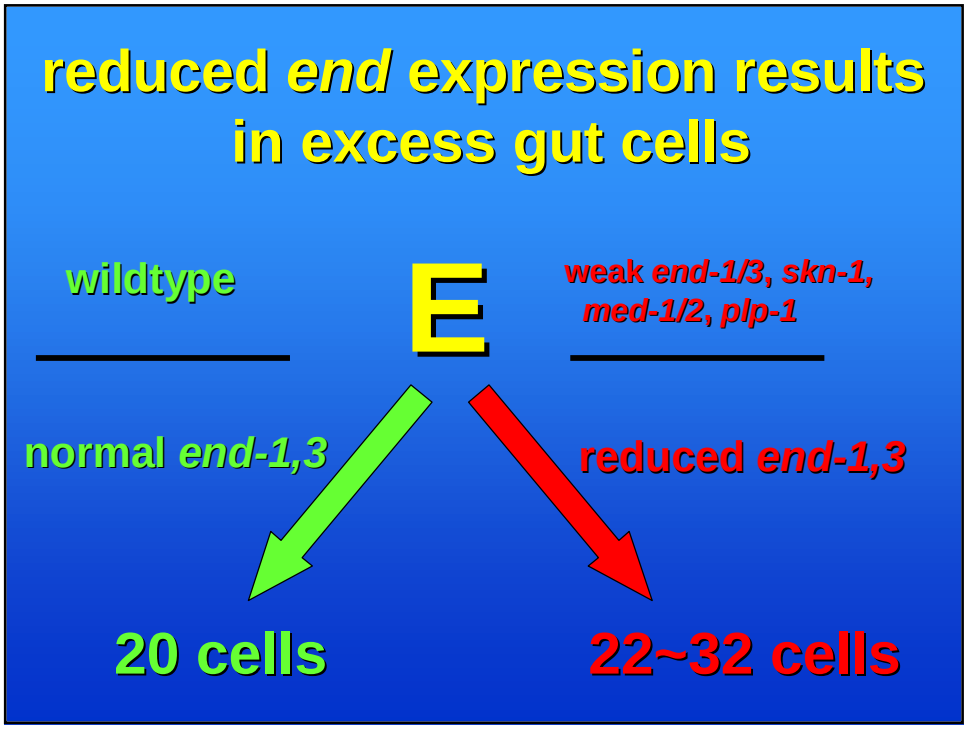
dsRNA → "instant gene knockout"

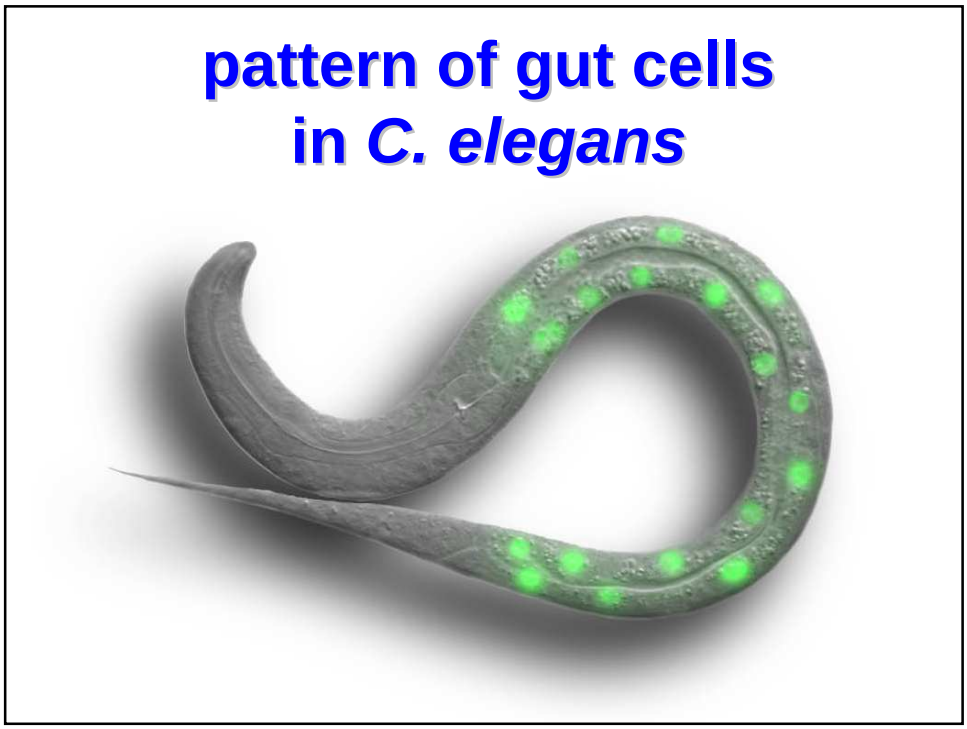
feed bacteria expressing dsRNA

genome-wide screen (~19,000 genes)

RNAi library screen



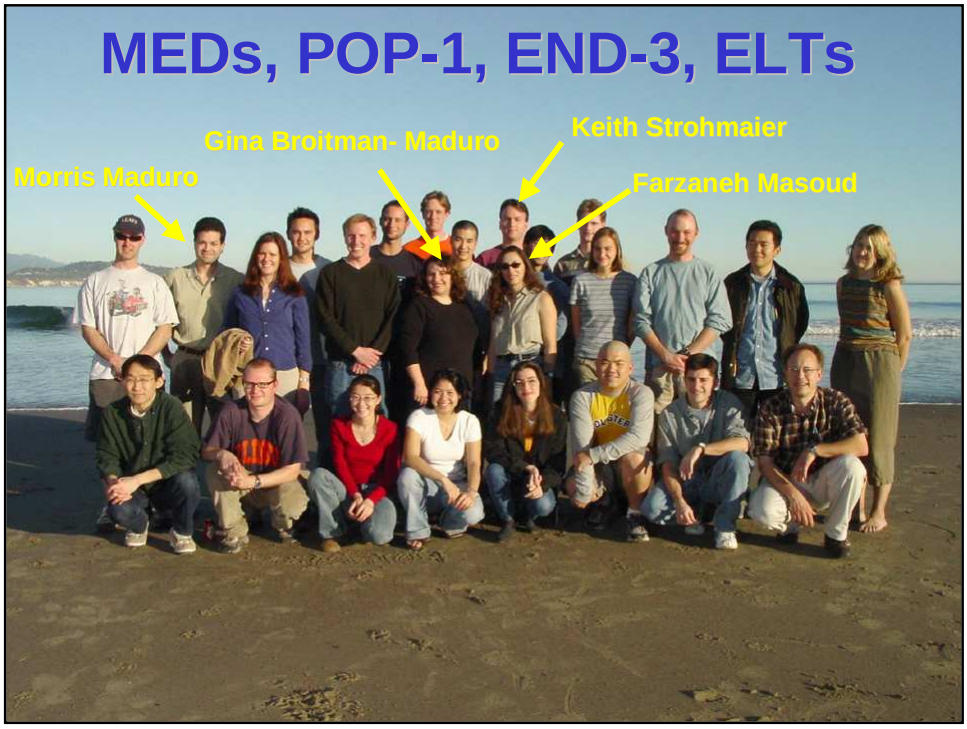

reduced end expression results in excess gut cells
wildtype
E
weak end-1/3, skn-1, med-1/2, plp-1
normal end-1,3
reduced end-1,3
20 cells
22~32 cells

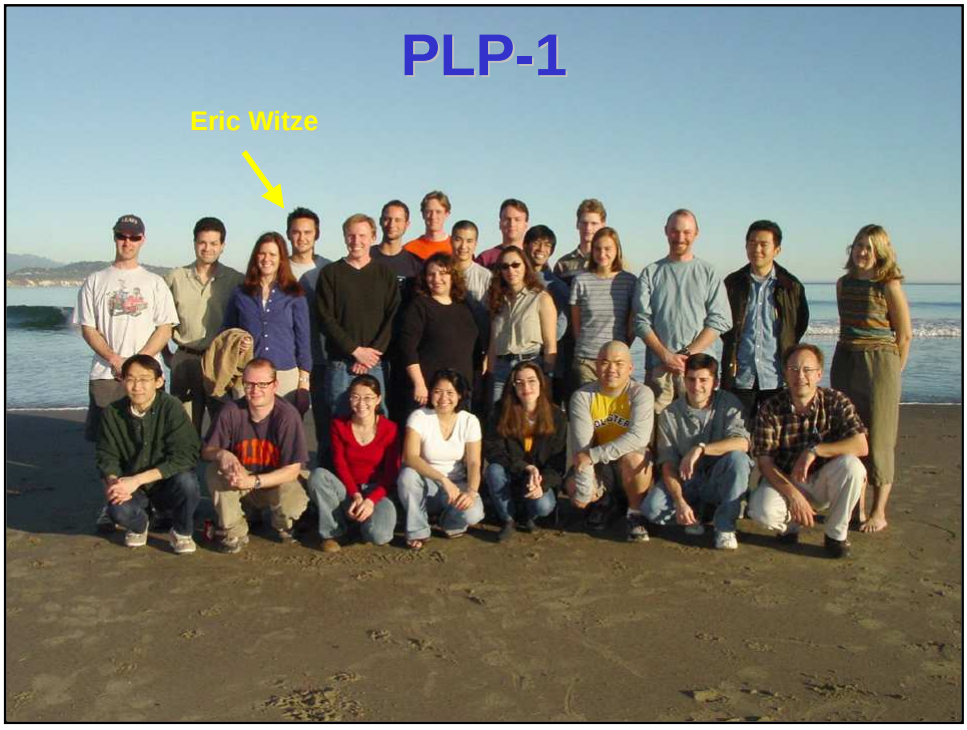

pattern of gut cells in C. elegans

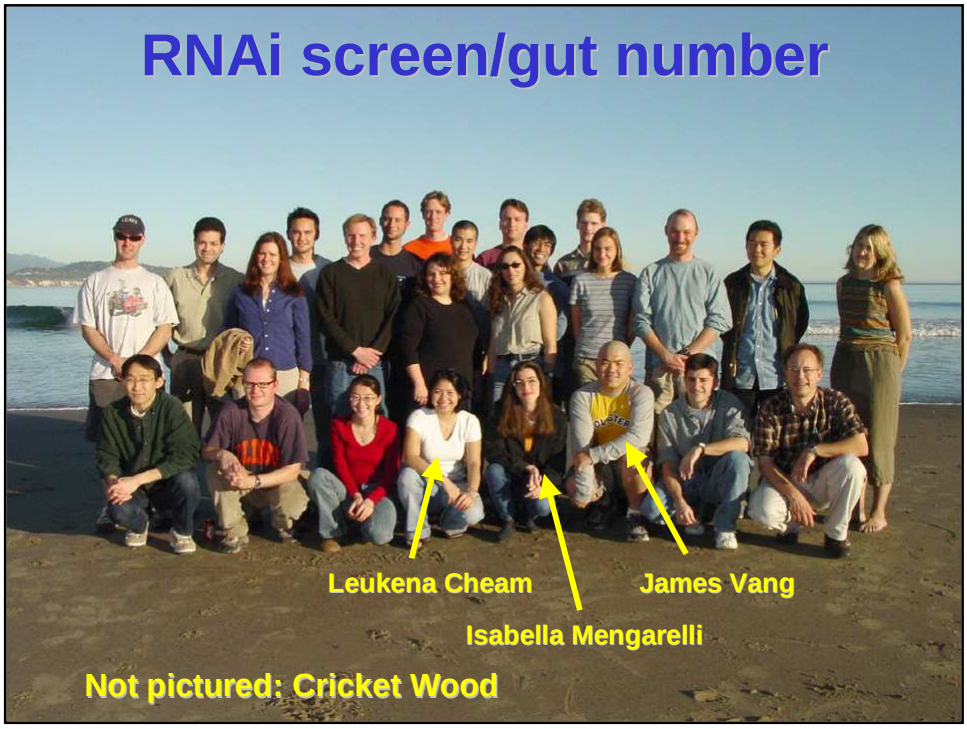
RNAi screen summary

	Lethal		Viable	
Phenotype	N (248)	%	N (2229)	%
I slight excess	13	5%	7	0.3%
II large excess	9	3%	0	0%
III subnormal	0	0%	4	0.2%
IV faint express.	1	0.4%	37	1.7%
V no expression	12	5%	0	0%
VI abn. pattern	37	15%	8	0.4%

Research Group


MEDs, POP-1, END-3, ELTs
Gina Broitman- Maduro
Keith Strohmaier
Morris Maduro
Farzaneh Masoud


PLP-1
Eric Witze


RNAi screen/gut number
Leukena Cheam
James Vang
Isabella Mengarelli
Not pictured: Cricket Wood