

**CELL DEATH DURING DEVELOPMENT**

**LLOYD A. GREENE  
DEPT OF PATHOLOGY**

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**KEY DEVELOPMENTAL PROCESSES**

**CELL PROLIFERATION**

**CELL MIGRATION**

**CELL DIFFERENTIATION**

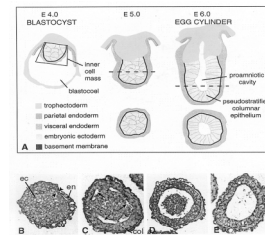
**CELL DEATH**

**FUNCTIONS OF DEVELOPMENTAL CELL DEATH**

**A. MORPHOGENESIS: SCULPTING/SHAPING STRUCTURES**

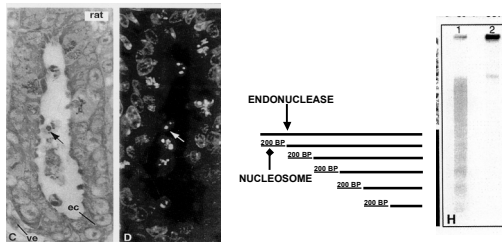
**CREATION OF CAVITIES AND TUBES**

**CELL DEATH AND FORMATION OF THE PROAMNIOTIC CAVITY FROM THE BLASTOCYST 1**



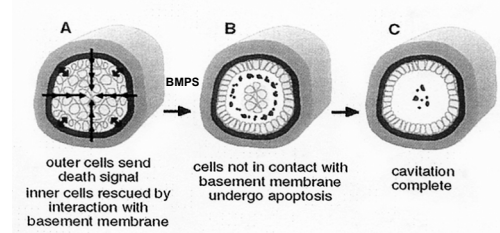
FROM: Coucouvanis and Martin. Cell 83: 279-287 (1995)

**CELL DEATH AND FORMATION OF THE PROAMNIOTIC CAVITY FROM THE BLASTOCYST 2**



FROM: Coucouvanis and Martin. Cell 83: 279-287 (1995)

**CELL DEATH AND FORMATION OF THE PROAMNIOTIC CAVITY FROM THE BLASTOCYST 3**



FROM: Coucouvanis and Martin. Cell 83: 279-287 (1995)

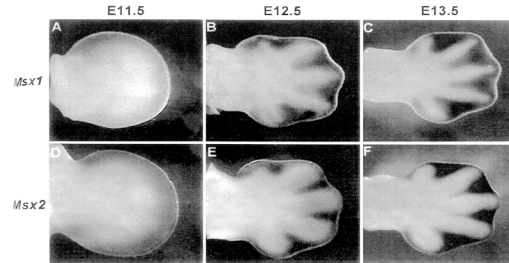
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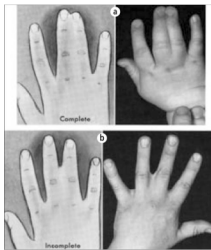
CREATION OF FORM (DIGITS)

**CELL DEATH AND FORMATION OF DIGITS 3**



FROM: Chen and Zhao, J. Exp. Zool. 282:691 (1998).

**HUMAN SYNDACTYLY**



SIMPLE

COMPLEX

FOR EXAMPLE: ONE  
FEATURE APERT  
SYNDROME - CAUSED BY  
ACTIVATING MUTATIONS IN  
FGF2 RECEPTORS

From: Flatt AE. Proc (Bayl Univ Med Cent). 2005 Jan;18(1):26-37.

**FUNCTIONS OF DEVELOPMENTAL CELL DEATH**

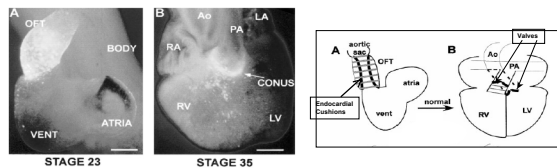
**A. MORPHOGENESIS: SCULPTING/SHAPING STRUCTURES**

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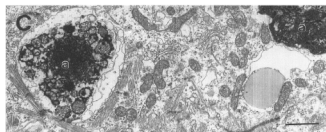
CREATION OF FORM (DIGITS)

TISSUE REMODELING (CARDIAC OUTFLOW TRACT)

**CELL DEATH AND CARDIAC MORPHOGENESIS**

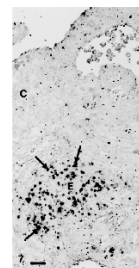


OFT = Outflow Tract  
RA = Right Auricle  
RV = Right Ventricle  
LA = Left Auricle  
LV = Left Ventricle  
PA = Pulmonary Artery  
Ao = Aorta  
a = Apoptotic Cardiomyocyte



From: Watanabe et al. Dev. Bio. 240: 274-288 (2001)

**CELL DEATH AND CARDIAC MORPHOGENESIS**



CELL DEATH IN CARDIAC  
OUTFLOW TRACT OF E13 MOUSE  
HEART AS REVEALED BY  
TUNEL STAINING (ARROWS)

From: Abdelwahid et al., Microscopy Res Tech. 58: 2002

**BLOCKADE OF DEATH IN DEVELOPING HEART OFT LEADS TO DOUBLE OUTLET RIGHT VENTRICLE (DORV)**

**CONTROLS**      **zVAD-fmk**

**A**      **B**      **C**

**E**

PV = Pulmonic Valve  
 RVOT = Right Ventricular Outflow Tract  
 Ao = Aorta  
 PA = Pulmonary Artery  
 OFT = Outflow Tract

From: Watanabe et al. Dev. Bio. 240: 274-288 (2001)

**FUNCTIONS OF DEVELOPMENTAL CELL DEATH**

**B. DELETION OF UNNEEDED STRUCTURES**

**KIDNEY: PRONEPHROS AND MESONEPHROS**

**BRAIN: CORTICAL SUBPLATE NEURONS**

**UROGENITAL SYSTEM: WOLFFIAN AND MÜLLERIAN DUCTS**

**REGULATION OF REPRODUCTIVE TRACT DEVELOPMENT 1**

EMBRYONIC RAT GENITAL RIDGES SHOWING:  
 MÜLLERIAN DUCT (MD)  
 WOLFFIAN DUCT (WD)  
 OVARY (Ov)  
 TESTES (Te)

**C**      Female      Male

FROM: Roberts et al., Devel. Bio. 208: 110 (1999)

**REGULATION OF REPRODUCTIVE TRACT DEVELOPMENT 2**

**MALE XY** → TESTES → MIS (MÜLLERIAN INHIBITING SUBSTANCE) → MÜLLERIAN DUCT → REGRESSION

TESTES → ANDROGEN → WOLFFIAN DUCT → MATURATION

**FEMALE XX** → DEFAULT PATHWAY → MÜLLERIAN DUCT → MATURATION

DEFAULT PATHWAY → WOLFFIAN DUCT → REGRESSION

**REGULATION OF REPRODUCTIVE TRACT DEVELOPMENT 3: CELL DEATH DURING MÜLLERIAN DUCT REGRESSION**

**A**      Müllerian ducts      Male      Female

apoptotic cells (%)

Stage	Male (%)	Female (%)
04.00	~10	~5
09.00	~25	~10
15.00	~45	~15
19.00	~35	~10
10.00	~30	~10

FROM: Roberts et al., Devel. Bio. 208: 110 (1999)

FROM: Xavier and Allard Mol Cell Endocrinol (2003)

**FAILURE OF MÜLLERIAN DUCT REGRESSION: PERSISTENT MÜLLERIAN DUCT SYNDROME**

Intraoperative appearance of a 15-month-old male with persistent mullerian duct syndrome.  
 T: Testis, SC: Spermatic cord, FT: Fallopian tube, F: Fimbria, U: Uterus

From: Boleken et al., Int Urol Nephrol. March 1, 2007

**FUNCTIONS OF DEVELOPMENTAL CELL DEATH**

**C. REGULATION OF CELL NUMBERS**

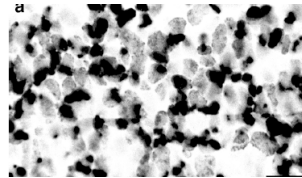
**NERVOUS SYSTEM:**

**GENERATION OF CORRECT NEURON NUMBER**

**MATCHING NEURONS AND TARGETS**

**MATCHING SCHWANN CELL AND OLIGODENDROCYTES WITH AXONS**

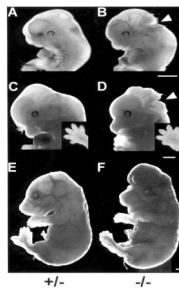
**DEATH OF PROLIFERATING VZ CELLS IN E14 MOUSE CEREBRUM**



Tan = BrDU; Purple = ISEL/dying

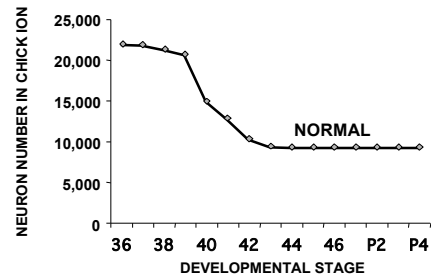
Biaschke et al J Comp Neurol 1998

**EMBRYOGENIC DEFECTS IN A MOUSE LACKING CASPASE-9**



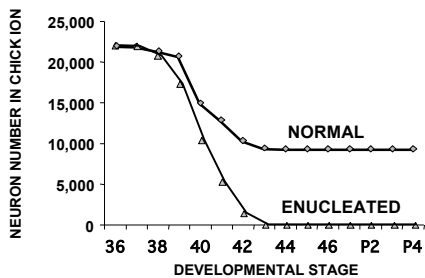
From: Kuida et al Cell:94: 325-337, 1998

**NORMAL DEVELOPMENTAL NEURONAL DEATH OCCURS AND IS REGULATED BY TARGET DERIVED TROPHIC FACTORS**



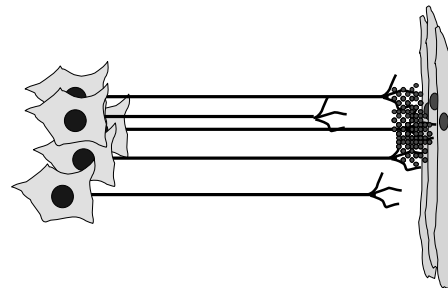
Clarke, Rogers & Cowan J. Comp. Neurol. 167: 125 (1976)

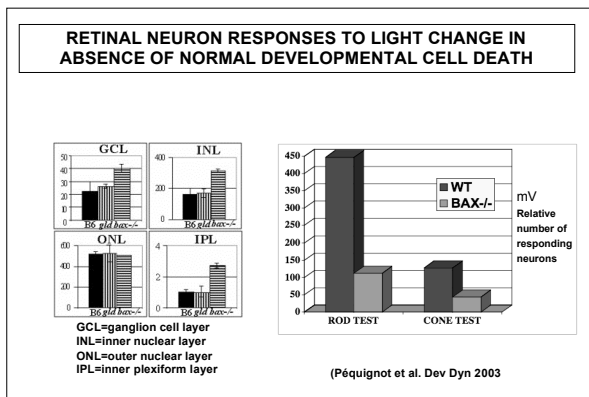
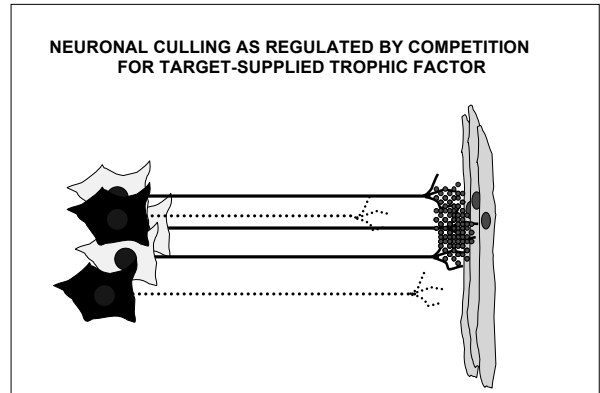
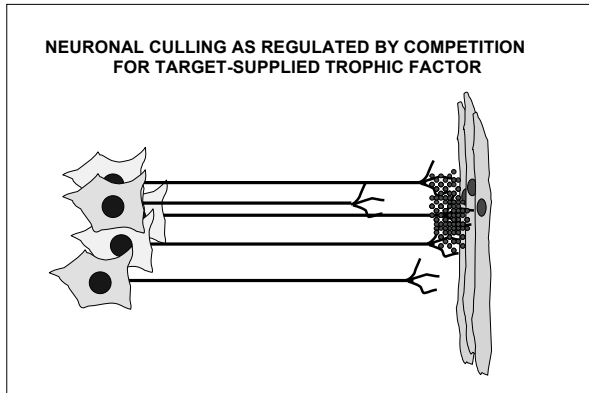
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Clarke, Rogers & Cowan J. Comp. Neurol. 167: 125 (1976)

**NEURONAL CULLING AS REGULATED BY COMPETITION FOR TARGET-SUPPLIED TROPHIC FACTOR**



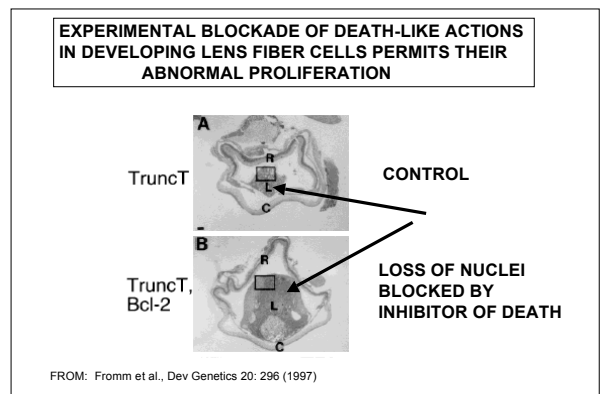
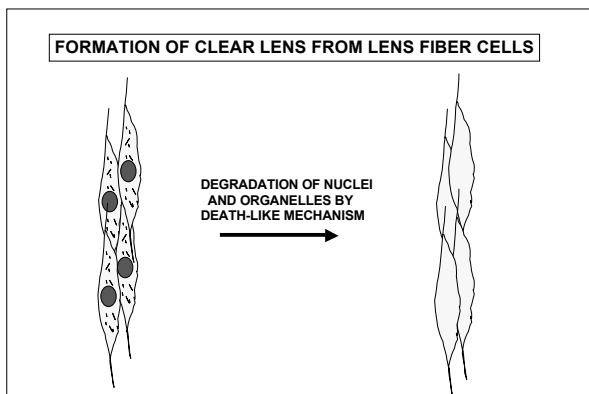


**FUNCTIONS OF DEVELOPMENTAL CELL DEATH**

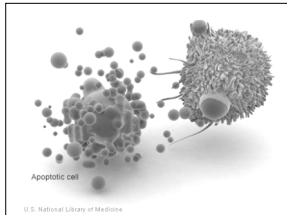
**E. PRODUCTION OF STRUCTURES WITHOUT ORGANELLES**

SQUAMOUS EPITHELIUM FROM KERATINOCYTES

**FORMATION OF LENS FROM LENS FIBER CELLS**



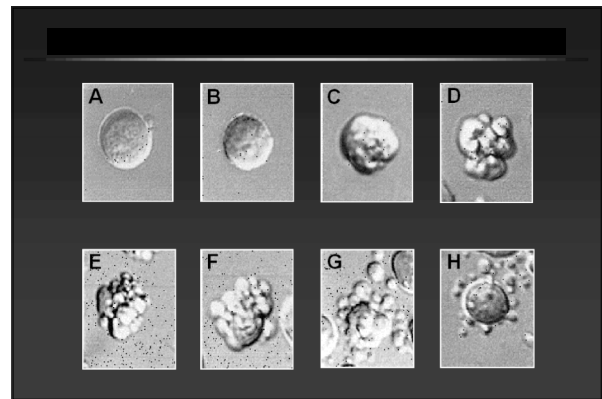
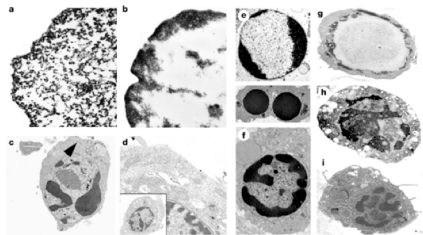
**HOW DOES DEVELOPMENTAL CELL DEATH OCCUR?**



<b>APOPTOTIC DEATH</b>	vs	<b>NECROTIC DEATH</b>
PRESENT IN DEVELOPING TISSUES		RESPONSE TO CELL INJURY, TOXINS
CYTOPLASMIC BLEBBING		
CELLULAR & NUCLEAR PYKNOSIS		CELL & NUCLEAR SWELLING
CHROMATIN CONDENSATION		
DNA DEGRADATION BY ENDONUCLEASES (FORMATION OF DNA LADDER)		RANDOM DNA DEGRADATION
FORMATION OF MEMBRANE-LIMITED APOPTOTIC BODIES		LOSS OF MEMBRANE INTEGRITY & LOSS OF CYTOPLASMIC CONTENTS
PHAGOCYTOSIS OF APOPTOTIC BODIES		
ABSENCE OF INFLAMMATORY RESPONSE		INFLAMMATORY RESPONSE

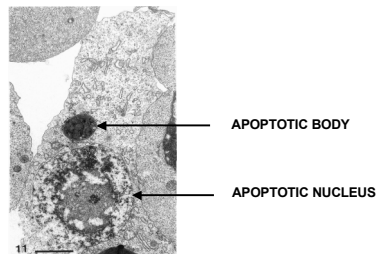
Kerr, Wyllie and Currie

**ELECTRON MICROSCOPIC VIEWS OF NORMAL (a) AND APOPTOTIC (b-i) NUCLEI**



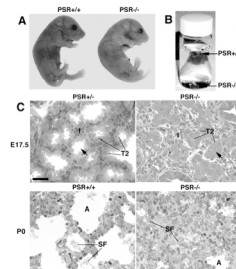
**APOPTOTIC BODIES AND NUCLEI ARE CLEARED BY PHAGOCYTOSIS**

EM OF A MACROPHAGE ENGULFING AN APOPTOTIC BODY AND APOPTOTIC NUCLEUS IN THE VENTRICULAR WALL OF THE DEVELOPING MOUSE HEART



From: Abdelwahid et al. Anat. Rec. 256:208 (1999)

**MICE MUTANT FOR PHAGOCYTOSIS SHOW DEFECTIVE LUNG DEVELOPMENT AND RESPIRATORY SYMPTOMS SIMILAR TO THOSE IN HUMAN RESPIRATORY DISTRESS SYNDROME (RDS))**



From: Li et al., Science: 302 (2003)

**WHAT ARE THE MECHANISMS BY WHICH CELLS DIE DURING DEVELOPMENT?**

THERE ARE EVOLUTIONARILY CONSERVED MECHANISMS THAT GOVERN DEVELOPMENTAL CELL DEATH

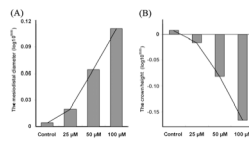
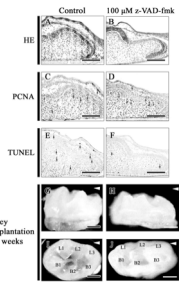


Robert Horvitz

**CASPASES**

- FAMILY OF EXECUTORS OF APOPTOTIC DEATH
- CYSTEINE PROTEASES THAT CLEAVE AFTER ASP
- CONSTITUTIVELY PRESENT AS INACTIVE FORMS
- ACTIVATED BY CLEAVAGE VIA INTERACTION WITH COFACTORS SUCH AS APAF1 AND CYTOCHROME C
- WHEN ACTIVATED, CLEAVE CELLULAR SUBSTRATES, LEADING TO APOPTOTIC DEATH

**BLOCKING CELL DEATH WITH CASPASE INHIBITORS AFFECTS TOOTH DEVELOPMENT**



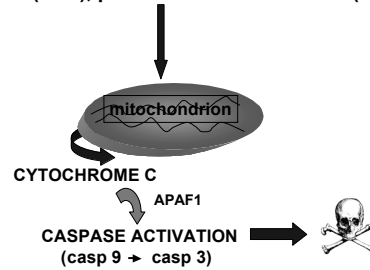
Control	25 μM	50 μM	100 μM	Control	25 μM	50 μM	100 μM
1.00	1.00	1.00	0.97	1.16	0.93	1.29	0.88
1.00	0.89	0.89	0.81	0.89	0.89	0.81	0.81

Control	PCNA	TUNEL
100 μM z-VAD-fmk	163 ± 1.8	45 ± 1.8*
100 μM z-VAD-fmk	163 ± 1.8	83 ± 1.8*

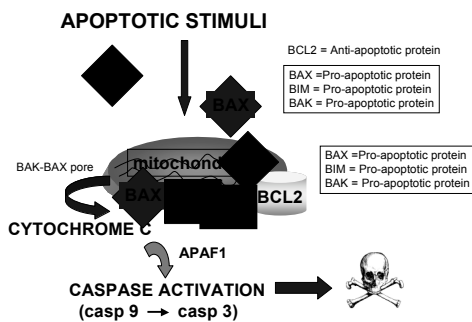
From: Kim et al., J Dent Res 85: 530 (2006)

**THE MITOCHONDRIAL PATHWAY OF APOPTOTIC DEATH - 1**

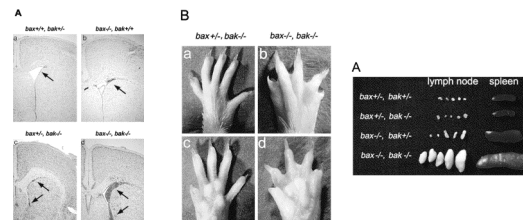
APOPTOTIC STIMULI (absence of survival factors (FGF), presence of death factors (BMPs))



**THE MITOCHONDRIAL PATHWAY OF APOPTOTIC DEATH - 2**



**ABNORMALITIES IN MICE LACKING BAX AND BAK**



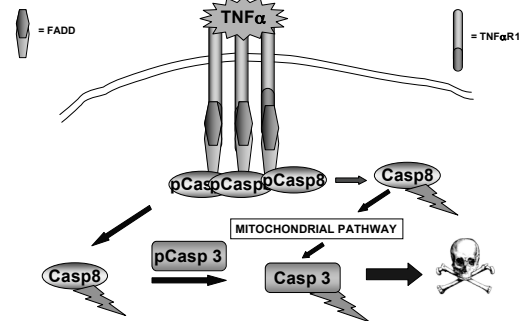
Arrow shows neuroprogenitors in ventricular zone of embryonic brain

From: Lindsten et al. Mol Cell 6:1389 (2000)

**DEATH PROMOTING RECEPTORS AND LIGANDS  
THE EXTRINSIC PATHWAY TO APOPTOTIC DEATH**

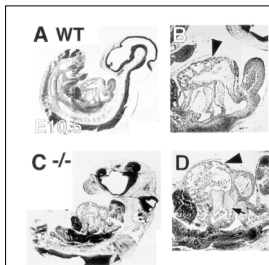
LIGAND	RECEPTOR
TNF $\alpha$	TNF $\alpha$ R1
FAS ligand	FAS
TRAIL	TRAIL-R

**THE RECEPTOR-MEDIATED PATHWAY OF APOPTOTIC DEATH**



**MICE LACKING FADD DIE DURING EMBRYOGENESIS  
AND HAVE MULTIPLE DEFECTS**

Low power view:  
A,C



B,D: Ventricular  
Myocardium

Arrowhead shows  
abnormal developing  
trabeculae; arrow  
normal endocardial  
cushion.

From: YEH et al. Science 279: 1954 (1998)

**CONCLUSIONS**

- CELL DEATH IS A MAJOR REGULATOR OF NORMAL EMBRYOGENESIS
- IT OCCURS AT ALL STAGES OF EMBRYONIC DEVELOPMENT AND IN MULTIPLE ORGAN SYSTEMS AND PLAYS A VARIETY OF ROLES
- FAILURE OF NORMAL CELL DEATH DURING EMBRYOGENESIS LEADS TO A VARIETY OF DEVELOPMENTAL DEFECTS
- DEVELOPMENTAL CELL DEATH IS GENERALLY APOPTOTIC IN NATURE
- THE GENERAL MECHANISMS OF APOPTOTIC CELL DEATH ARE BECOMING UNDERSTOOD
- MUTATION OF SPECIFIC APOPTOTIC GENES LEADS TO DEVELOPMENTAL ABNORMALITIES