# Effects of Microgravity on Cell Cytoskeleton and Embryogenesis

Presented in the Embryo Physics Course http://www.embryophysics.org
April 9, 2009

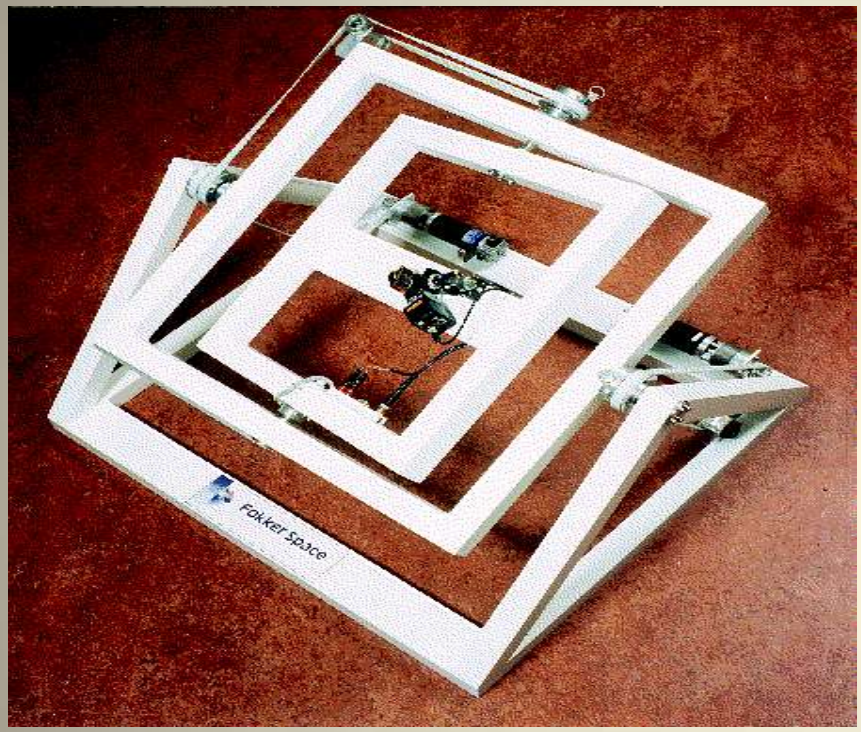
By
Susan Crawford-Young
Red River College
mudworks9@hotmail.com

# Development in Microgravity

By Susan Crawford-Young

Reference: Crawford-Young, “Effects of microgravity on cell cytoskeleton and embryogenesis”, International Journal of Developmental Biology, 50: 183-191 (2006)

# Microgravity



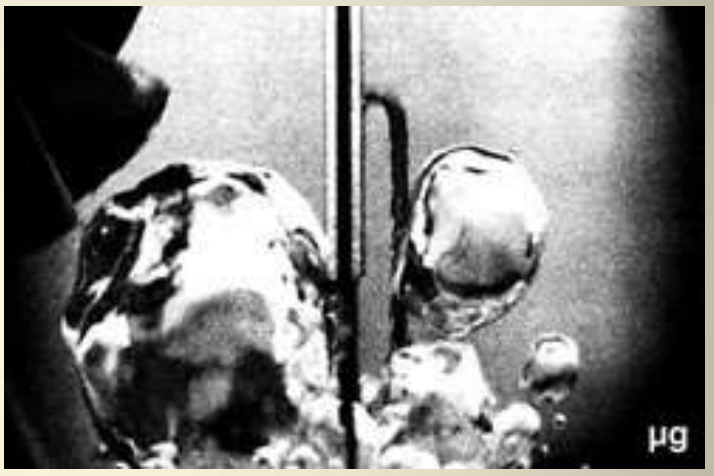

3D Clinostat partially imitates microgravity

Space around the earth [eol.jsc.nasa.gov]

Drop towers and airplanes are used as well to create free fall

# There is No Buoyancy in Microgravity

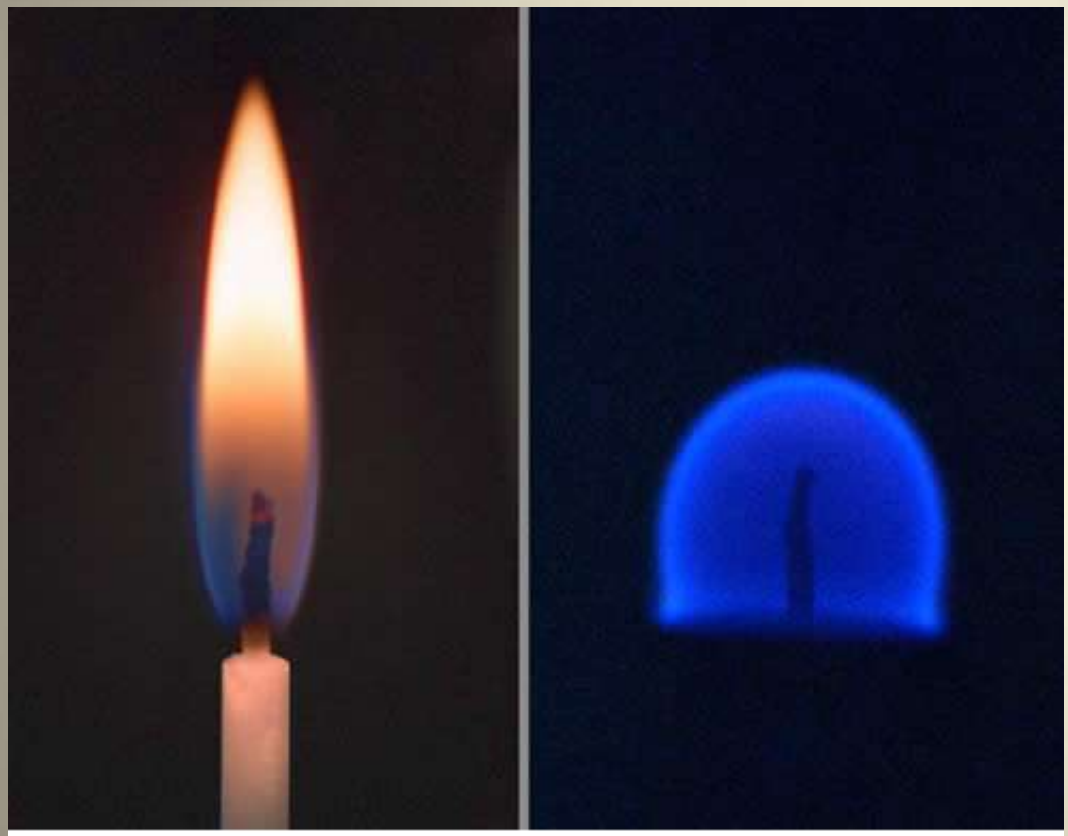

Boiling water in earth normal gravity 1G




Water boiling in Microgravity

In 1 g buoyancy dominates the flow in layers with dimensions larger than one centimeter. [spaceresearch.hamptonu.edu ]

# Convection does not work in microgravity

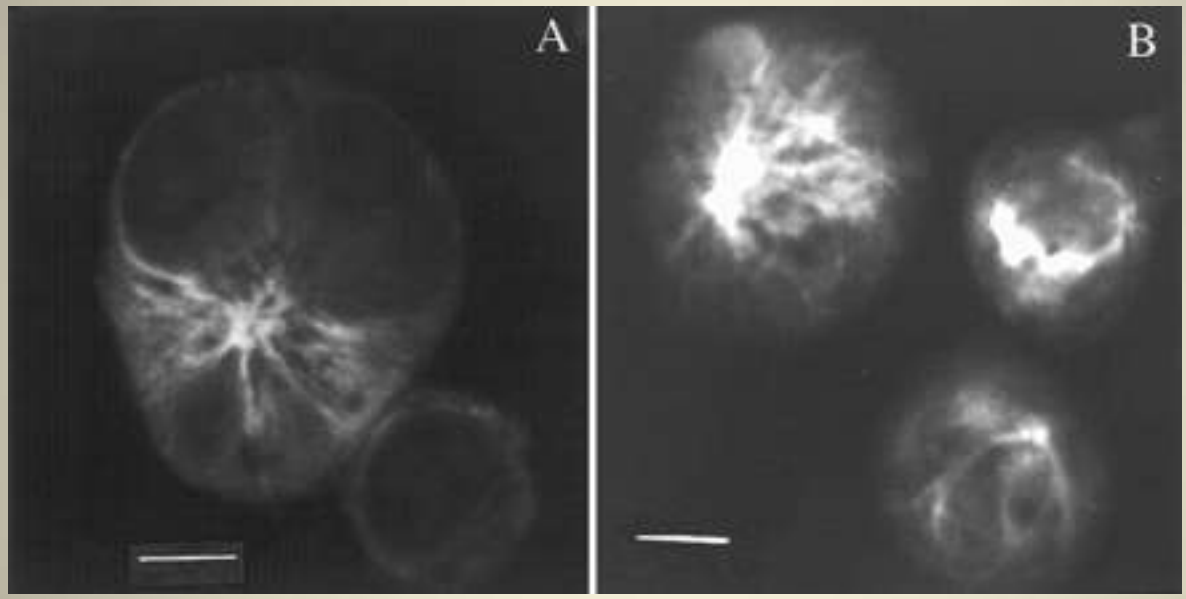


A candle in normal gravity (left) and in microgravity.

"In microgravity, the shape of the flame is spherical due to the inhibition of BDTC directed airflow and has a much lower temperature. On average, such a flame will burn 30-40 seconds before going out due to the fact that oxygen transport is limited to diffusionary levels and this is not sufficient to maintain combustion." [Nasa 2002]

# Microtubules in human lymphocyte cells after 4 hours of growth

ground control | microgravity




**Microtubules in human lymphocyte cells** after 4 hours of growth. In flown cells (microgravity), the microtubule filaments extended from poorly defined organizing centers (MTOCs) and were coalesced and shortened. In comparable ground controls, microtubules radiated in discrete filaments from organized MTOCs and branched toward the cell membrane. Bars represent 5µm. [Lewis et al. 1998]

# Microtubules in Altered Gravity


Normal Earth Gravity and Magnetic Field vertical and horizontal growth

A B

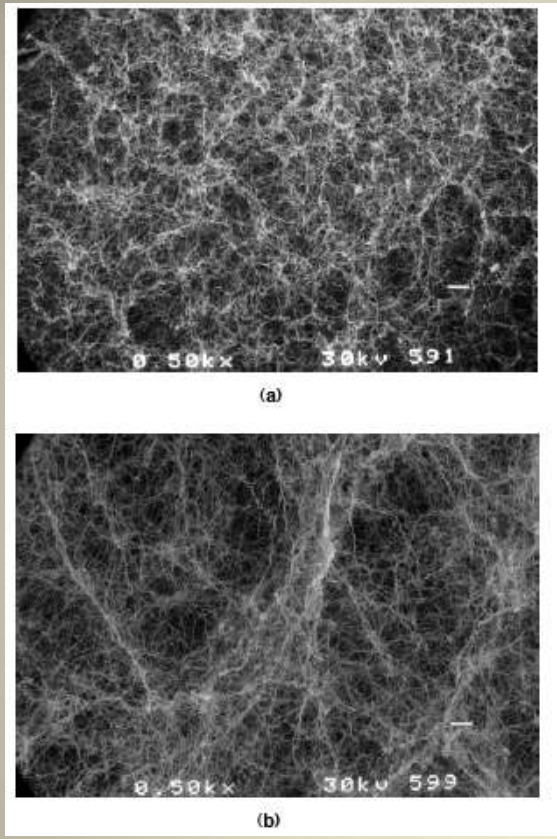
MT in simulated microgravity on earth, A without vibration, B with vibration

MT in simulated 1g in space vertical and horizontal growth

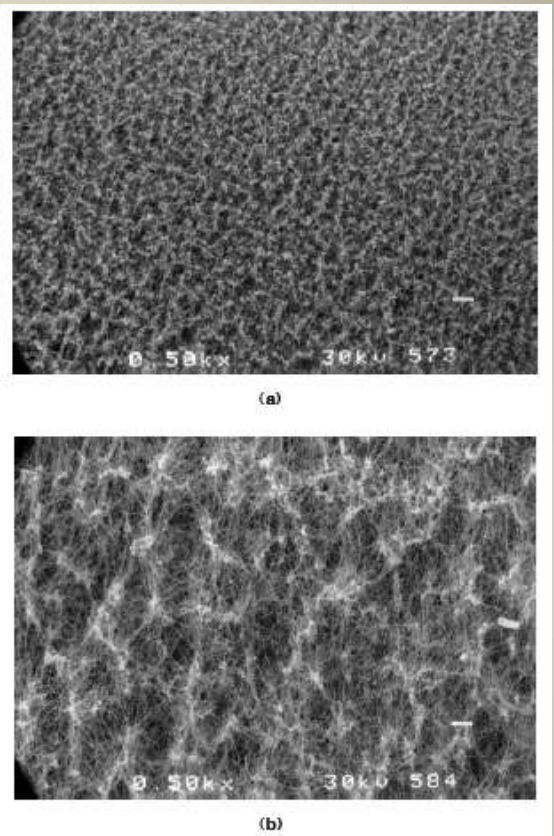
MT in Space Microgravity

Papaseit et al. 2000, Glade et al. 2006

# Collagen Gels

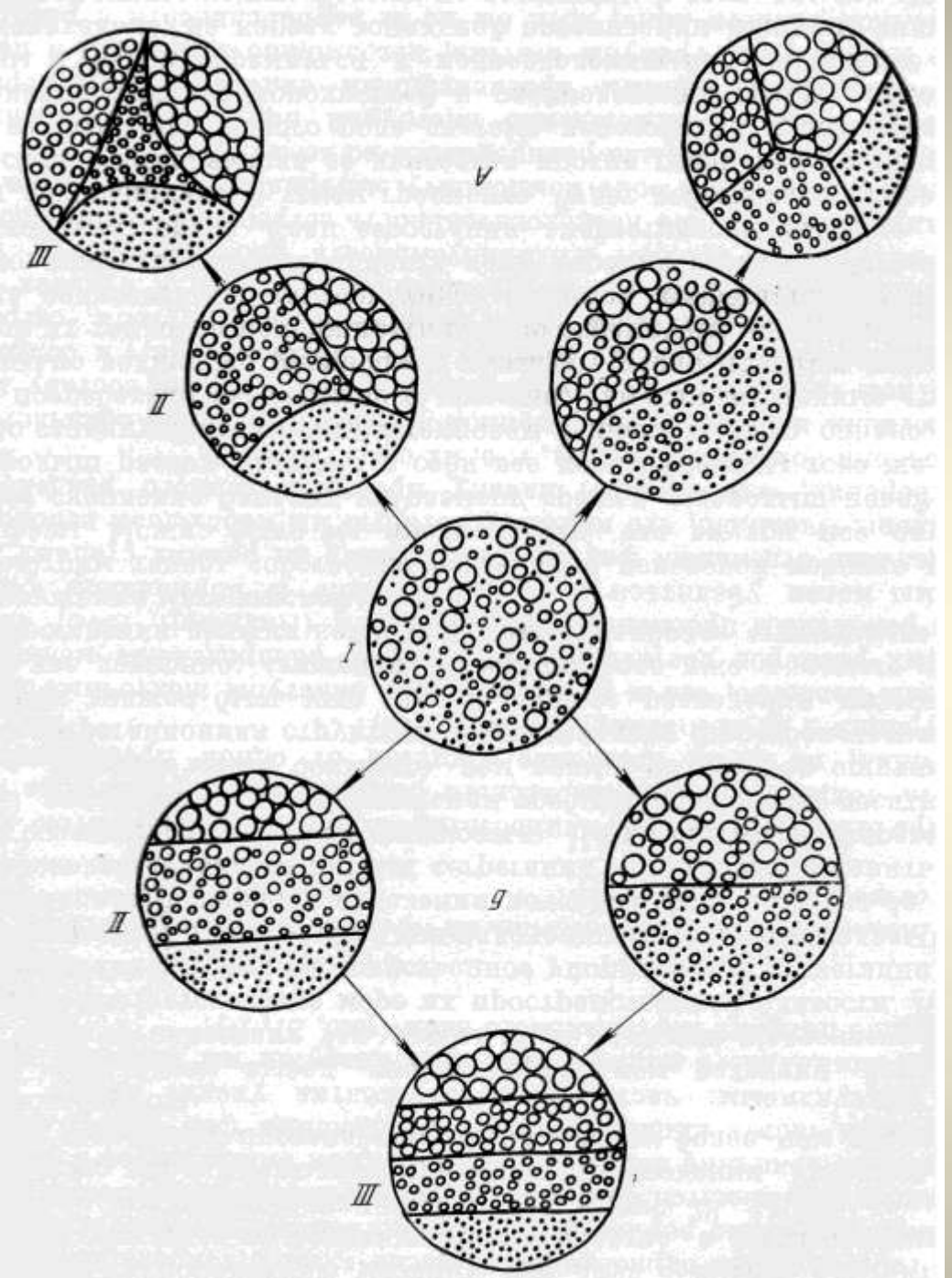



Ground Gel earth gravity




Flight gel microgravity

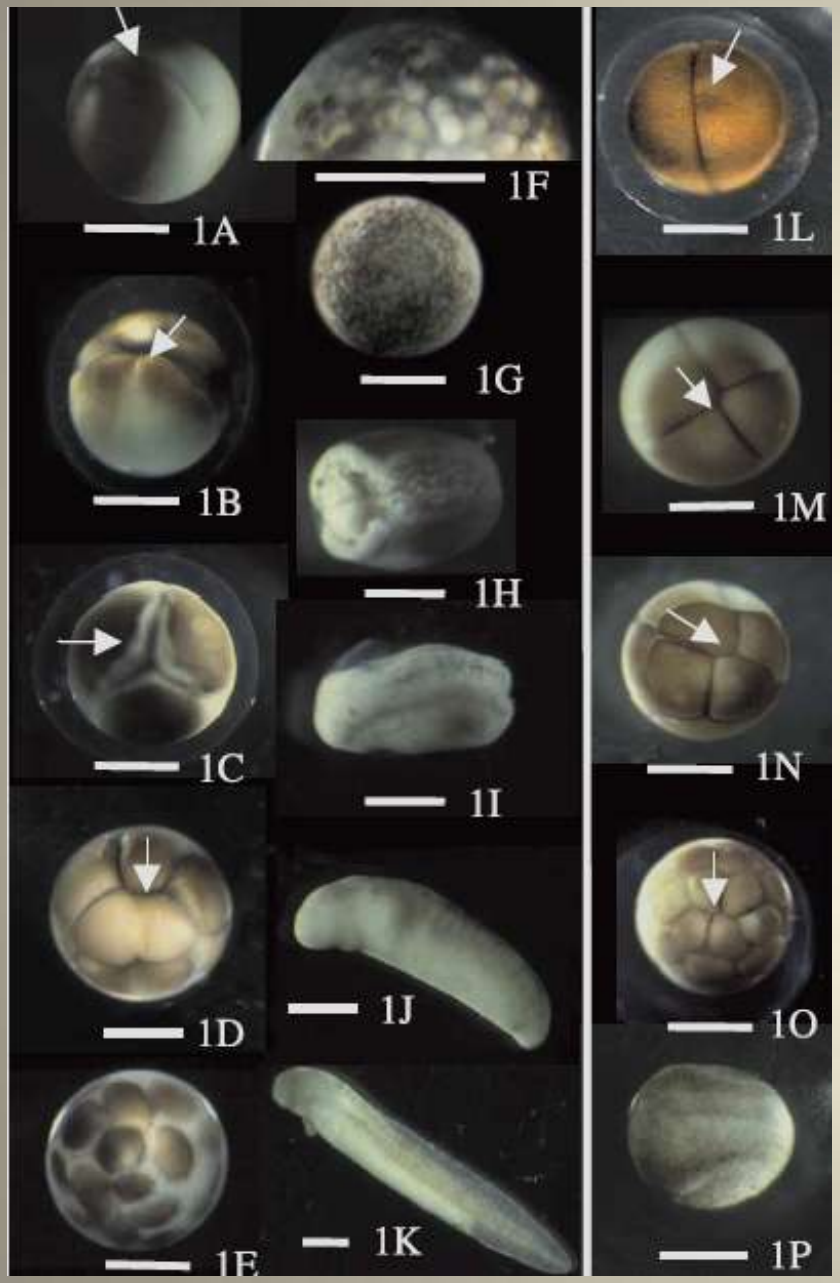
The more granular appearance of the ground gels relative to the highly uniform flight gels (a) close to the interface (b) near the center of the gel. Microgravity makes a difference in patterning at a microscopic level [Roedersheimer et al. 1997].



# Frog Egg in Gravity and Microgravity

Illustration of how microgravity can control segregation of the yolk granules inside the egg: arrows show a sequence of segregation steps in microgravity (upwards arrows) and normal gravity (downwards arrows): differently sized yolk granules are shown by differently sized circles [Dorfman & Cherdantsev 1994]

# Salamander Egg Growth in Space




A to K: mG-embryos fertilized, developed and fixed on board MIR. L to P: 1G- ground control embryos. Arrows indicate the animal pole. Abnormal pigmented area, Enlarged intercellular space, abnormal three-cell stage, D: Five blastomeres instead of 4 at the animal hemisphere, Yolk platelets between blastomeres. F: detached cells. G H: neurula at stage 15. Loss of neurectodermal cells. I: neurula at stage 20. Neural tube not closed in the dorsal region. J,K Normal aspect. Bar¼1 mm.

[Gualandris-Parisot et al. Biol. Sci. Space. 19, 3–11 (2002)]

# Salamanders that developed in Microgravity

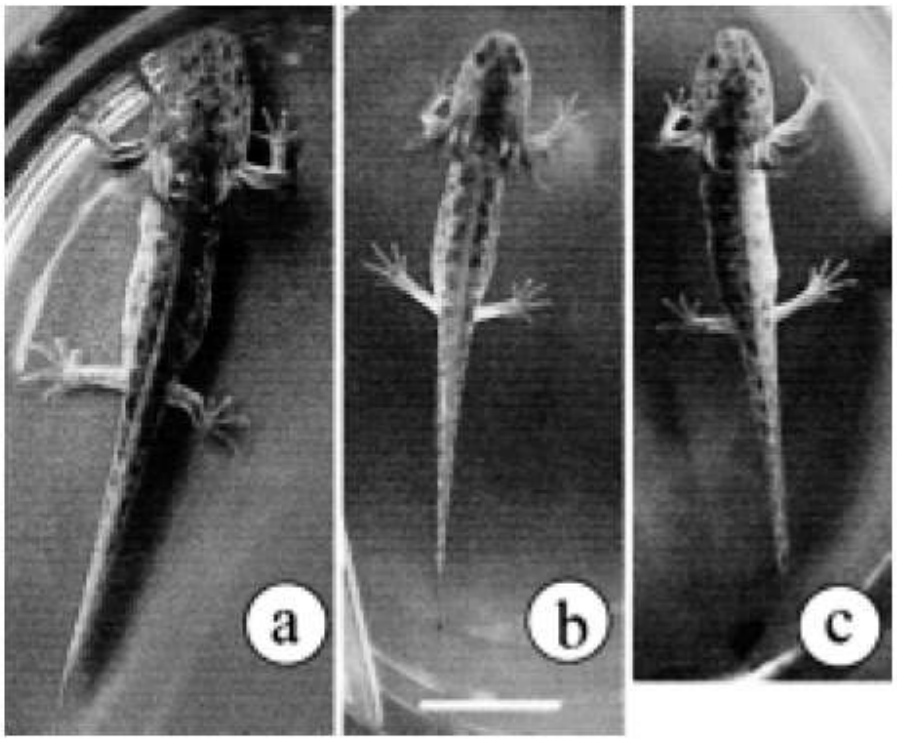
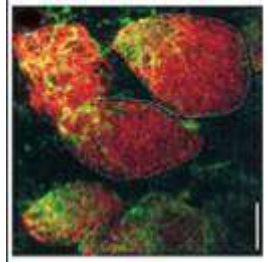



**Fig. 4.** Larvae at the beginning of metamorphosis (stage 55). Larvae born and reared up to the hatching stage onboard the Mir space station during the (a) Cassiopèe and (b) Pègase missions, then reared in laboratory conditions. (c) Ground control larva at the same developmental stage. Bar, 20 mm.

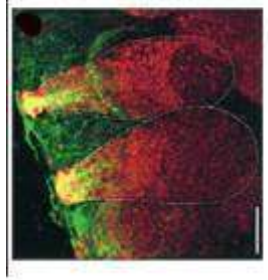
Odd cell division and problems with neural tube closure

Gualandris-Parisot et al. 2001



Cells in space micro-gravity

MT in Space Microgravity

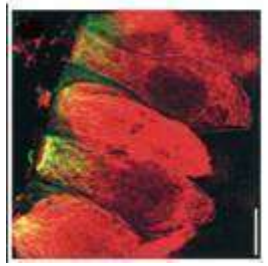


Cells in simulated 1g in space

MT in simulated 1g in space

A B

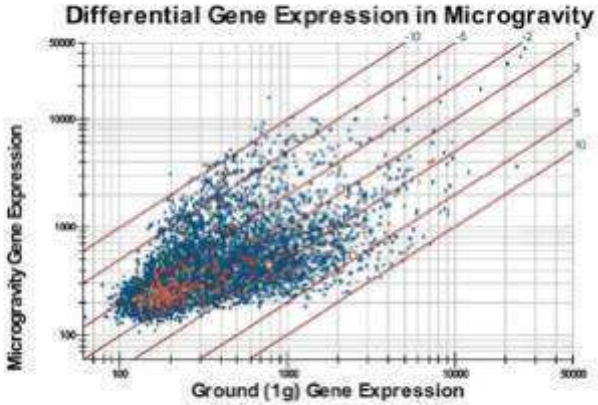
MT in simulated microgravity on earth, A without vibration, B with vibration



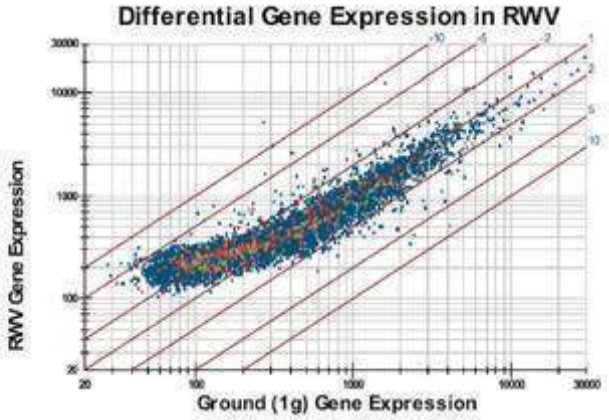
Cells in earth normal gravity

MT on earth

Differential Gene Expression $\mu g$




Differential Gene Expression simulated $\mu g$




Earth Normal Gravity (Differential Gene Expression is a straight line)

Changes in (A) rat utricular cells, (B) microtubules, and (C) gene expression between microgravity in space, simulated 1G in space, simulated microgravity on earth, and earth normal gravity (A) red- calretinin, green- a-tubulin (B) blue- 45°, yellow- 135° crystal orientation (C) green- shear stress and heat shock proteins, red- transcription factors (A) Gaboyard et al. 2002; (B) Papaseit et al. 2000, Glade et al. 2006; (C) Hammond et al. 2000

# Cells in Microgravity

- A variety of cell types have been sent into the microgravity environment of space for study.
- Many cell types recovered to a certain extent after 24 to 48 hours.
- The cytoskeleton of the cells including Microfilaments, Intermediate Filaments and Microtubule structure were abnormal.
- Some cells recovered better than others, with bone cells and lymphocytes as two cell types that have the most difficulty.

# Artificial Microgravity

- Produced in clinostats
- Rotating wall vessels
- Drop towers
- Diving airplanes

None of these methods produces the same effects as microgravity. The drop towers and airplane maneuvers do produce it but only for a short time.

# *Xenopus* in Simulated Microgravity for 48 hours starting at mid-gastrulation


Control

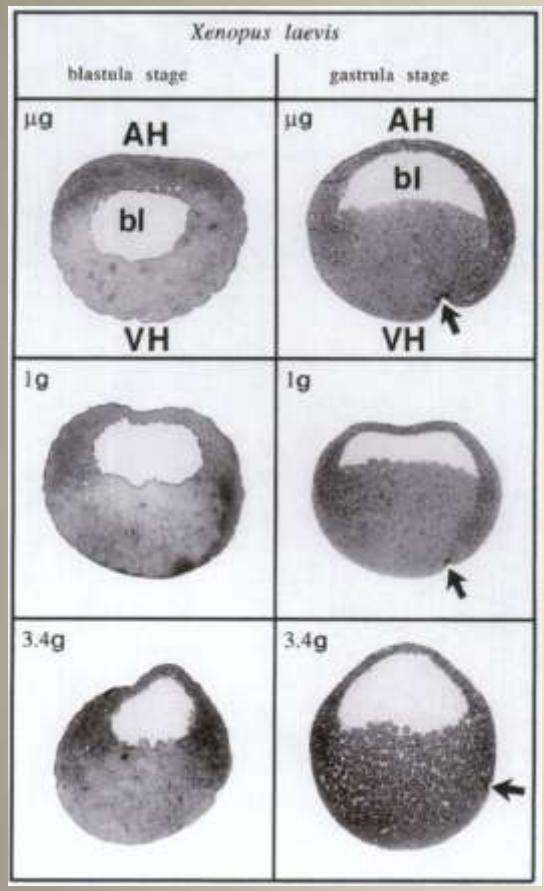
Slow Turning Lateral Vessel 1

Group placed on the rotor base of STLV 2

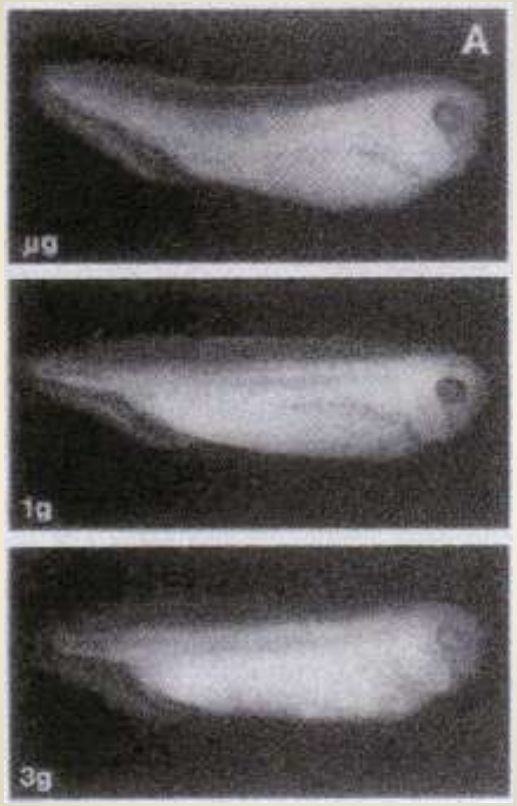
Exposure to rotation or to vibration strongly diminished the Hoxa2 mRNA staining in all areas. Exposure of *Xenopus embryos* to simulated microgravity increases the incidence of skeletal malformation. A study of neural crest derived tissue.

Olson et al. 2009 in press

# Embryos in altered gravity (clinostat)




Final morphology



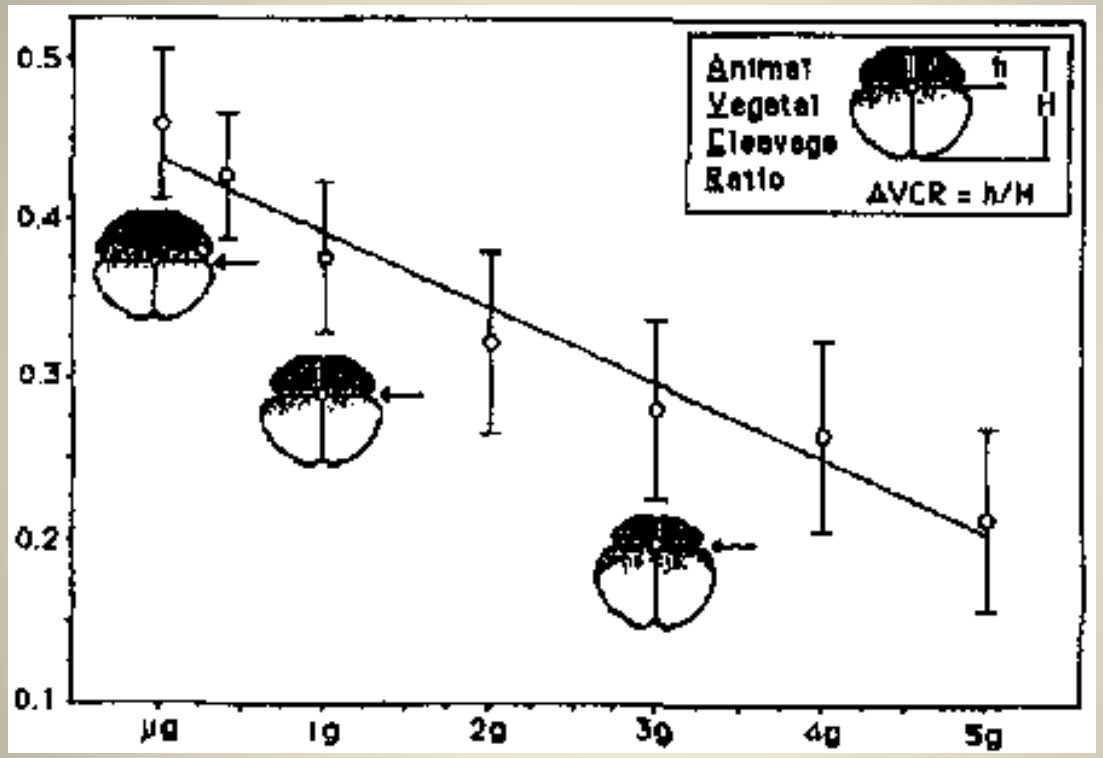

Micro gravity
Large head and arched back

Earth Normal gravity

3 g Thickened body

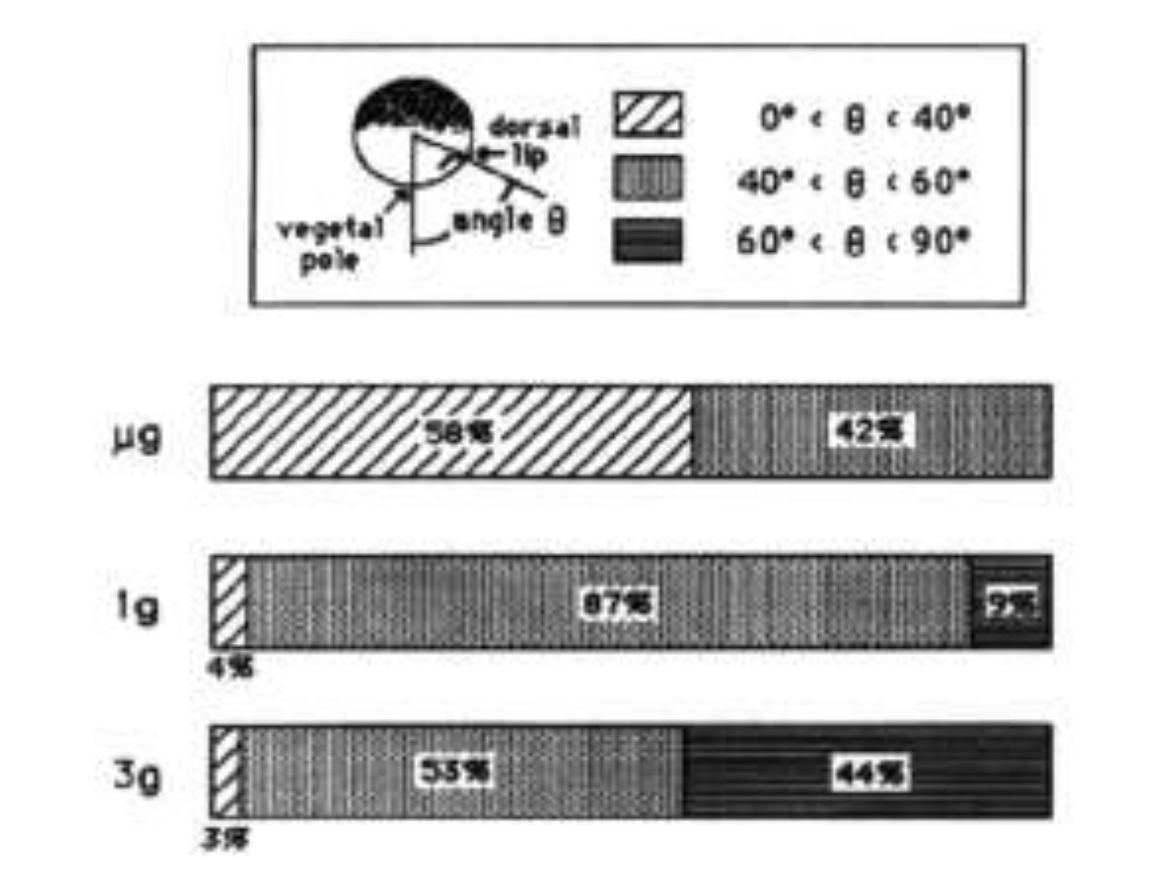
There may be a change in the density gradient of eggs exposed to altered gravity in a clinostat. If left in altered gravity the overall morphology of the hatchling is changed. [Neff 1993]

# AVCR Graph
# Animal Vegetal Cleavage Ratio



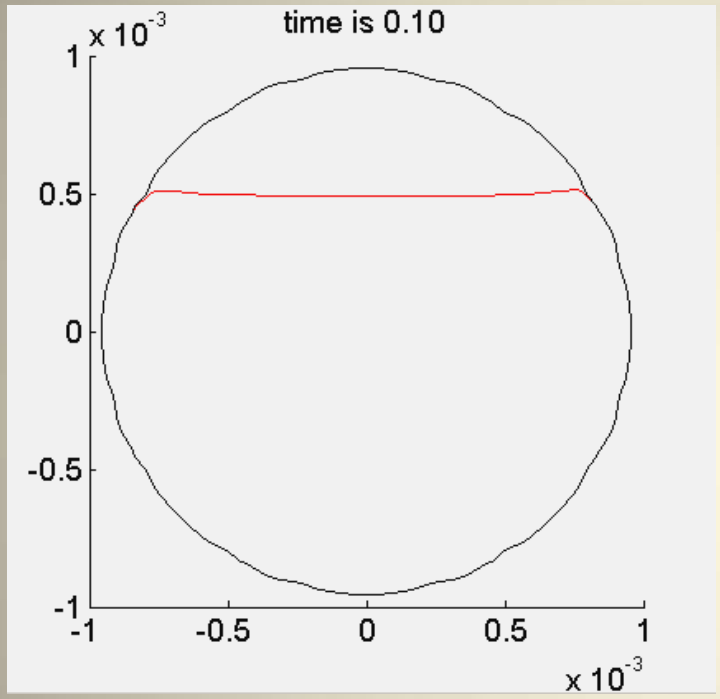

"The definition of AVCR (inset) and the relationship between mean AVCR and the strength of the gravitational field (*Xenopus*)." [Neff 1993]

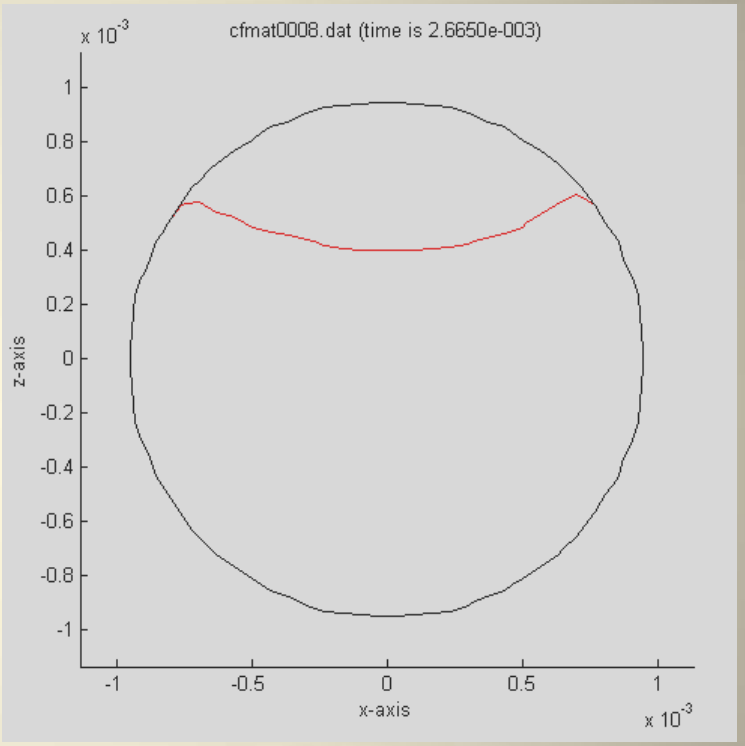
# Dorsal Lip of the Blastopore Angle




"Distribution of angular separations between the vegetable pole and the initial involution site of the dorsal lip of the blastopore" [Neff 1993]

# Fluid simulation of two immiscible fluids in a sphere




Start of the simulation

Final position of the fluid in microgravity

Using ComFlo the lighter fluid is on top. The parameters are $\rho_1$ = 1.3 x water (1300 kg/m$^3$), $\rho_2$ = 0.8 x water (800 kg/m$^3$), $\mu_1$ viscosity = 10 x water, $\mu_2$ = 10 x water, surface tension is about 0.1 of water so $\sigma$ is 10. This makes the surface tension x density seven times smaller than that of water. The contact angle $\theta = 90^o$. Thanks to Comron Nouri & Roel Luppes.