Review Lab 2
Animal Pole
Gray Crescent
Vegetal Pole

Micromere
Cleavage furrow
Macromere
Fertilization envelope
Dorsal lip blastopore
blastocoel
Area opaca

Area pelucida

Head process

Hensen’s node

Primitive groove

Primitive folds
epiblast

mesoderm

endoderm

Primitive groove

Primitive fold
Genes expressed in early frog development

• In the late blastula and early gastrula stages differences in mRNAs within cells can appear.

• In the following figures try to determine the germ layer of the labeled cells from your knowledge of the anatomy and fate map of the frog after you have studied them in the slides.
Genes expressed in early frog development

- Mixer encodes a transcription factor with a role in promoting endoderm formation.
- Xbra encodes a transcription factor expressed in mesoderm cells.
- Vg1 encodes a secreted growth factor of the TGF-beta family present in vegetal cells.
Early (B) and midgastrula (C-F) stage cross sections of the frog embryo showing ISH of mRNA for Mixer (B and C) and Xbra (D) (mRNA shown in dark blue)

Mesoderm (m) and endoderm (e) locations are shown in I. What is the germ layer expressing Mixer? Do all cells of this layer express Mixer? What cells express Xbra?
ISH of the Vg1 mRNA in frog oocyte (mRNA shown in white)

How can you determine the polarity of the oocyte? Where is the Vg1 mRNA expressed?
Lack of twinning in chicken eggs

- Although nearly always one streak (and therefore one embryo) develops per egg more than one can be experimentally induced.
  - Simply cutting the prestreak blastoderm into four sections allows each to form a miniature separate streak arising randomly anywhere from an original margin with the area opaca. (See next slide)

Considering the life cycle of egg-laying amniotes why might there be strong inhibitors of rival streak formation?

How might such an inhibitor act?
Figure 10.31 Chick twins produced by cutting unincubated blastoderms. [Courtesy of N. T. Spratt, Jr.]

Figure 10.32 Potentiality and polarity in chick blastoderm. Four individuals resulting from division of blastoderm into four segments. [After N. T. Spratt and H. Haas, J. Exp. Zool., 145:113 (1960).]
Because nutrients are in limited supply in the self-contained egg it is important to have only one offspring developing in the egg. Are there any examples of naturally occurring twins from eggs?

- Recent work suggests that a chick protein related to the frog Vg1 is released from one dominant early forming streak and spreads out by diffusion to act in surrounding cells as an inhibitor of rival streak formation.

- The ISH results on the next slide show that an isolated anterior half will, but the anterior half of an intact blastoderm will not, produce RNAs (e.g. brachury and chordin) that are characteristically expressed in the streak cells.
The isolated anterior half in B, C and D expresses brachury, chordin and crescent not normally expressed in the anterior of the blastoderm.
Application of Vg1 to one lateral side (A and B) gives one streak but two opposing applications give two streaks (C and D).

What conclusions can you make regarding the role of Vg1 in primitive streak formation?