Xenopus

Organizer Formation and Mesoderm specification

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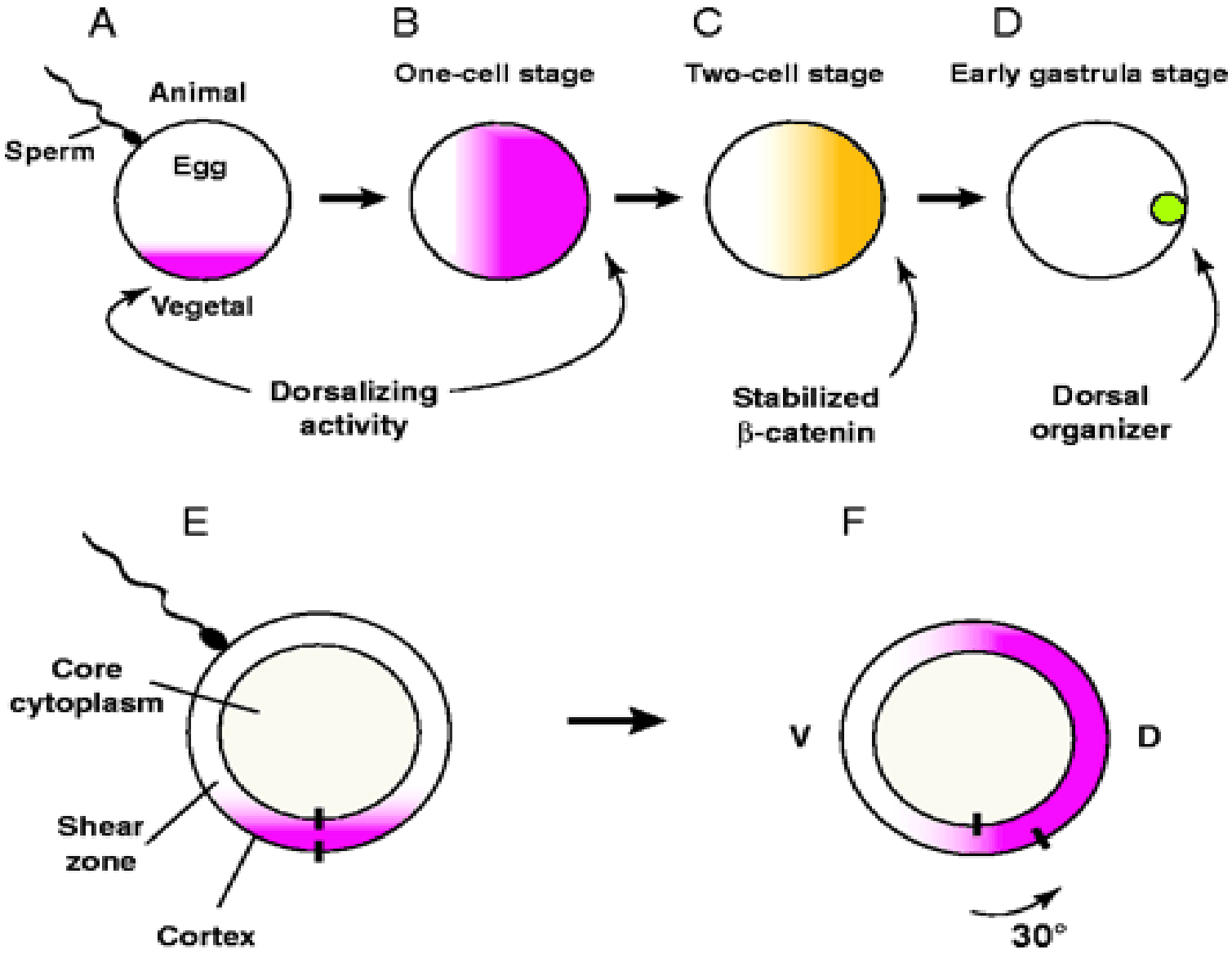
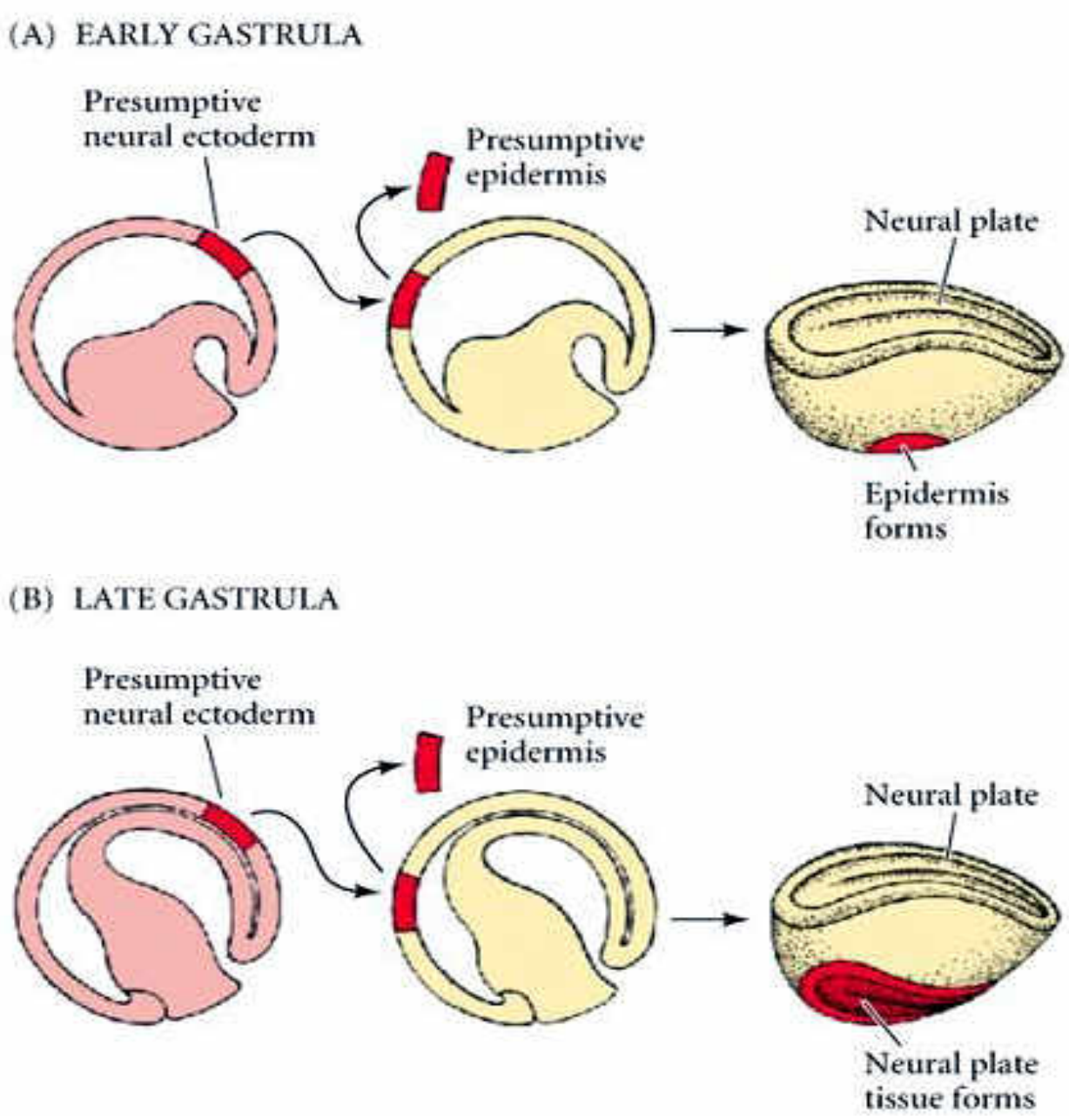
The future dorsal lip in amphibians usually forms opposite to the sperm entry point (Roux's sperm-imbibed silk thread experiment). The sperm brings in a large centriole. Microtubules form arrays under the egg cortex and drive a 30° rotation of the cortex with respect to the internal yolk. The rotation can be blocked by UV-irradiation of the vegetal (bottom) pole of the egg, or by nocodazole or other treatments that interfere with microtubule polymerization.

Normally, the first cleavage bisects the "gray crescent". Isolated blastomeres give rise to perfectly normal duplicated embryos. However, if you experimentally alter the first cleavage so that one blastomere gets all the gray crescent cytoplasm and the other gets none, then the isolated blastomeres behave very differently. The one receiving the gray crescent material develops into a normal embryo, but the other forms a disorganized mass of tissue called a "belly piece" because it contains mainly ventral tissues. This suggests that some important cytoplasmic determinants are localized in the early vertebrate embryo. We are interested in the same questions that we addressed in Fly development. Where do the first asymmetries arise in vertebrate development? How are the anterior-posterior and dorsal-ventral axis established? How does embryonic patterning arise? When you look at a frog egg you can clearly see one maternally derived asymmetry. The animal-vegetal axis is obvious due to the pigmentation and yolk differences that are determined during oogenesis.

The Spemann-Mangold organizer, also known as the Spemann organizer, is a cluster of cells in the developing embryo of an amphibian that induces development of the central nervous system. Hilde Mangold was a PhD candidate who conducted the organizer experiment in 1921 under the direction of her graduate advisor, Hans Spemann at the University of Freiburg in Freiburg, Germany. The discovery of the Spemann-Mangold organizer introduced the concept of induction in embryonic development. Now integral to the field of developmental biology, induction is the process by which the identity of certain cells influences the developmental fate of surrounding cells. Spemann received the Nobel Prize in Medicine in 1935 for his work in describing the process of induction in amphibians. The Spemann-Mangold organizer drew the attention of embryologists, and it spurred numerous experiments on the nature of induction in many types of developing embryos.

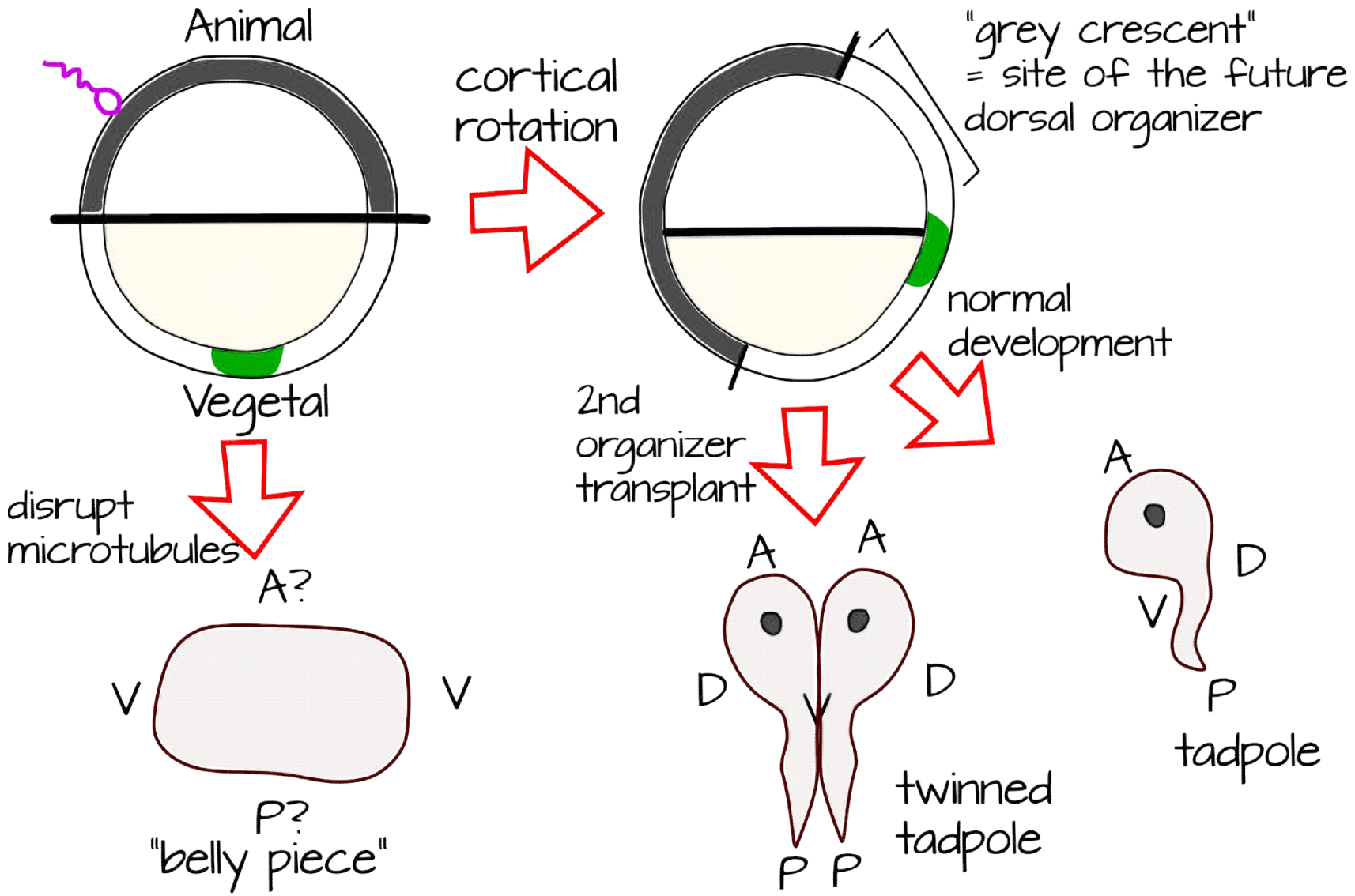

A
B
C
D
One-cell stage
Two-cell stage
Early gastrula stage
Animal
Sperm
Egg
Vegetal
Dorsalizing activity
Stabilized β-catenin
Dorsal organizer
E
F
Core cytoplasm
Shear zone
Cortex
V
D
30°


(A) EARLY GASTRULA
Presumptive neural ectoderm
Presumptive epidermis
Neural plate
Epidermis forms
(B) LATE GASTRULA
Presumptive neural ectoderm
Presumptive epidermis
Neural plate
Neural plate tissue forms

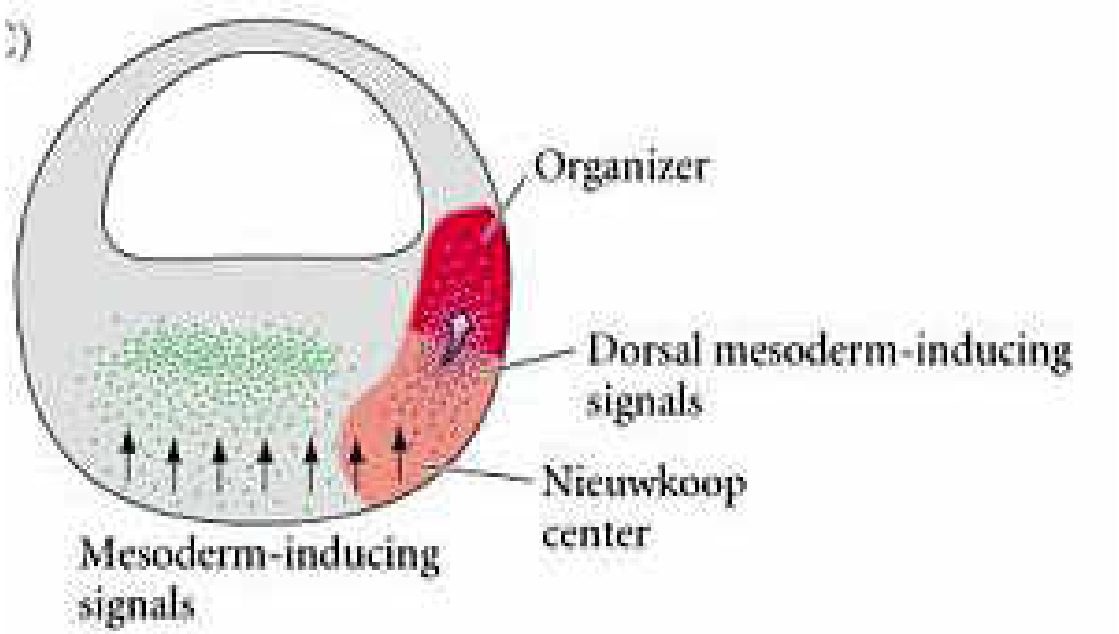
Translocation of the maternal dorsalizing activity.

- Translocation of the dorsalizing activity leads to β-catenin stabilization and the formation of the dorsal organizer. Between the time of fertilization and the first embryonic cell division, a maternally deposited dorsalizing activity (red) moves from (A) the vegetal pole to (B) the prospective dorsal region. (C) By the two-cell stage, maternal β-catenin (yellow) has become asymmetrically stabilized in the region that has received the dorsalizing activity.

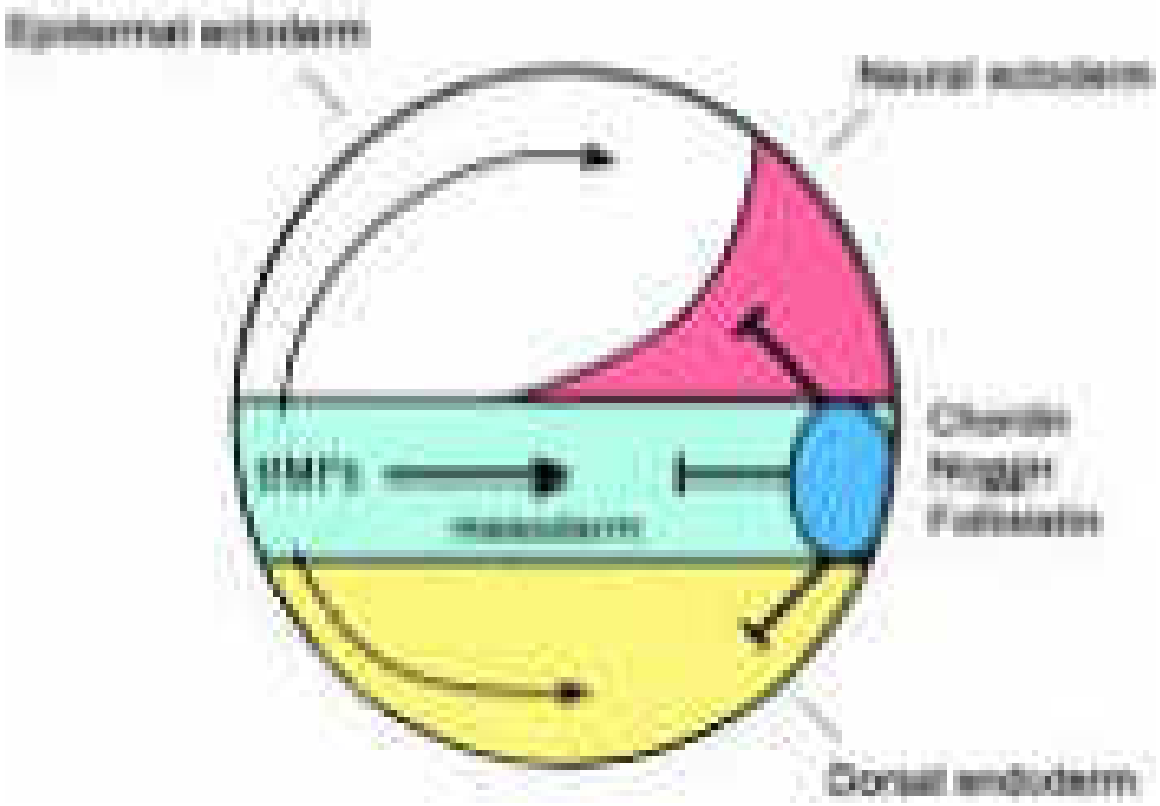
(D) Stabilized β-catenin activates genes of the dorsal organizer (green circle; also called the Nieuwkoop and Spemann organizers) in the dorsal equatorial region, as shown in an early gastrula embryo. (E,F) The dorsalizing activity translocates in the same direction as cortical rotation. (E) The dorsalizing activity (red) resides in the shear zone, an area of looser cytoplasm that forms between the outer cortex of the egg and the dense core cytoplasm following fertilization. The black bars at the vegetal pole mark the starting positions of the core and the cortex early in the first cell cycle. (F) During the first cell cycle, the cortex rotates relative to the core, moving about 30° towards the dorsal side in the same direction as the dorsalizing activity. This process is called cortical rotation, as represented by the displacement of the outer black bar


Animal
Vegetal
cortical rotation
"grey crescent"
= site of the future
dorsal organizer
normal development
2nd organizer transplant
disrupt microtubules
A?
V
V
P?
"belly piece"
A
A
D
D
V
P
P
twinned tadpole
A
D
V
P
tadpole

Nieuwkoop center signaling

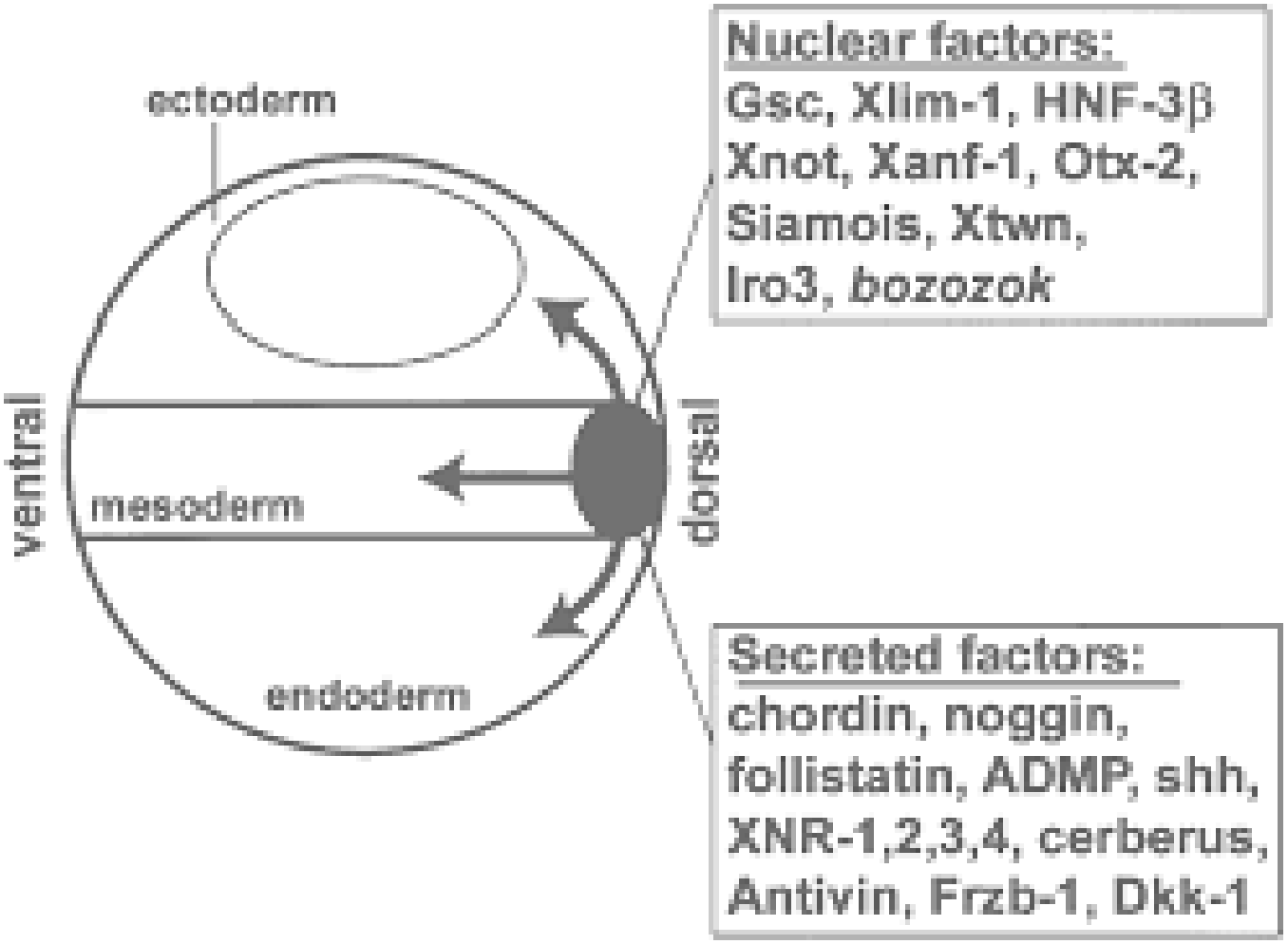


The work of Nieuwkoop, and Dale and Slack, and others is summarized in the model shown above. They hypothesized that only ectodermal and endoderm fate existed in the earliest embryo. Mesodermal fate is induced by signals from the vegetal cells to the marginal zone cells. Nieuwkoop's Center represented a special zone represented by the dorsal most vegetal cells that had the ability to induce dorsal mesoderm or organizer function. The next obvious question was to find and characterized these postulated mesoderm inducing signals.

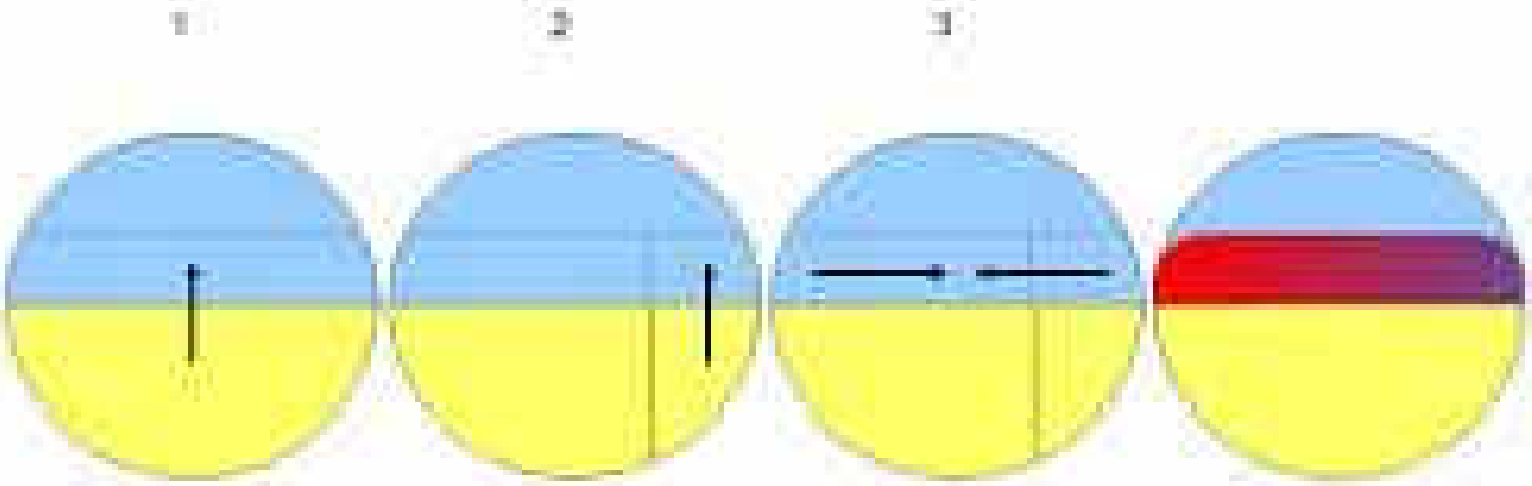


Spemann-Mangold organizer secreted factors antagonize ventral signals provided by BMPs

Factors for signaling



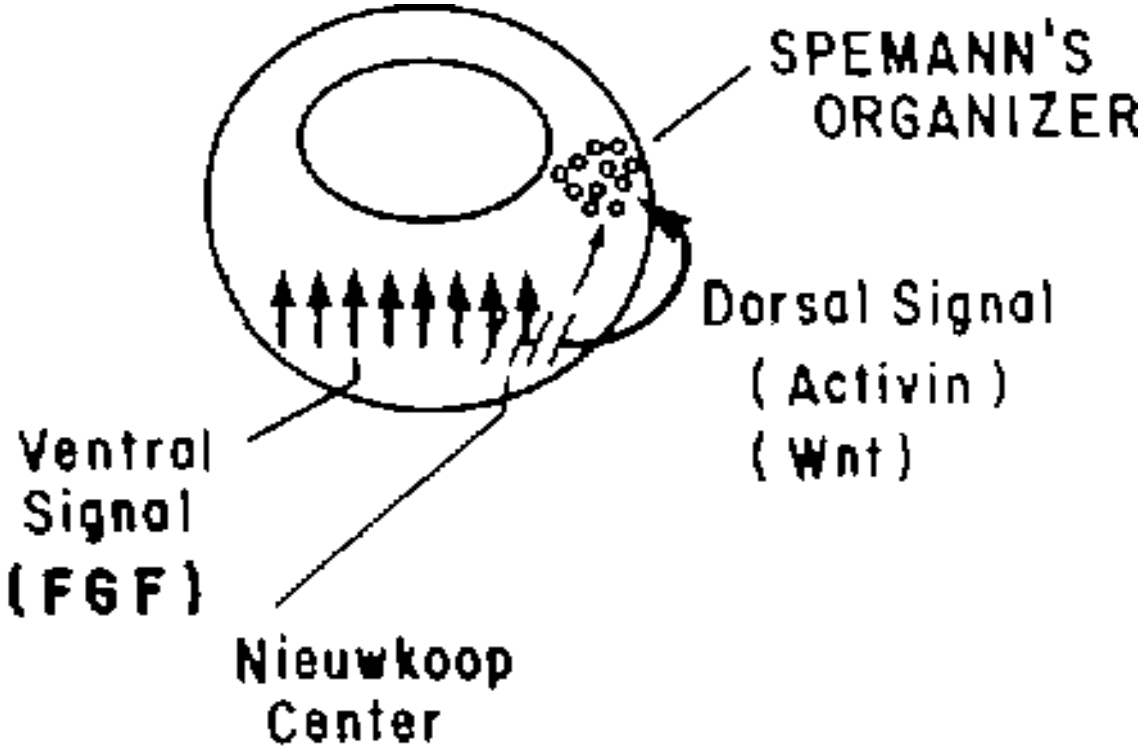
Mesoderm specification model



(1) Vegetal region, specifies mesoderm as ventral
(2) Nieuwkoop center specifies the Spemann organizer
(3) Organizer dorsalizes the adjacent mesoderm by inhibiting the ventralizing (default) signal.

The result is a (a) region that is fated to become mesoderm, (b) a division of that mesoderm into 2 or more sub-types: dorsal and ventral mesoderm.

Mesoderm Induction



A ventral signal is released radially by the vegetal cells (e.g., FGF), inducing the entire marginal zone to become mesoderm. On the dorsal side an additional signal (perhaps activin- or wnt-Yike) is released by vegetal cells of the Nieuwkoop center, inducing Spemann organizer tissue in the overlying marginal zone cells.