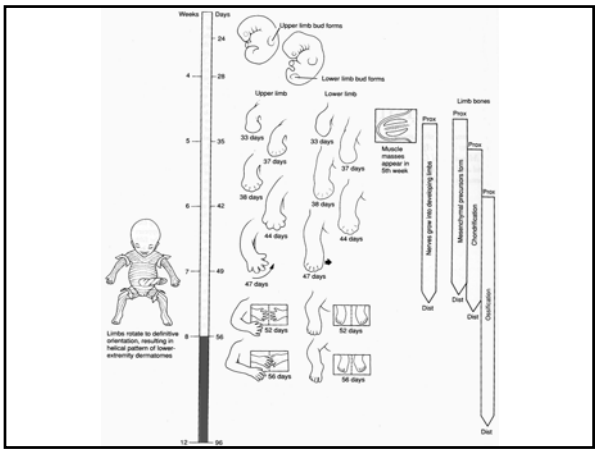
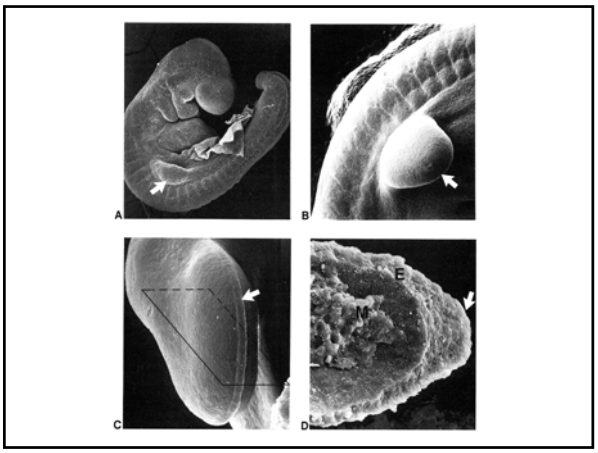
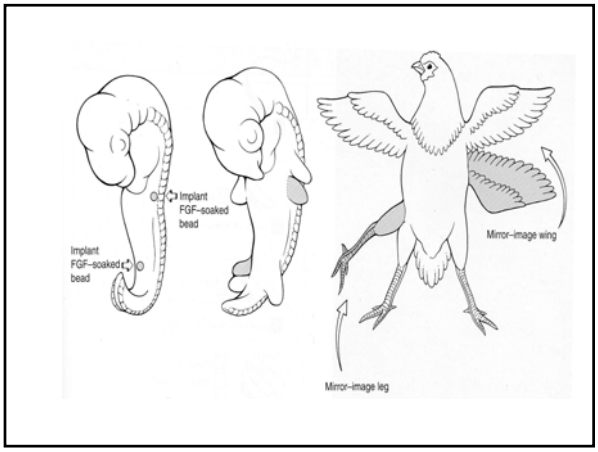






The same signal – FGF- induced the formation of either a wing or a leg. This and other evidence indicates that identity of the limb (how it responds to signals) resides in the mesenchyme (lateral plate mesoderm) which gives rise to the skeleton.

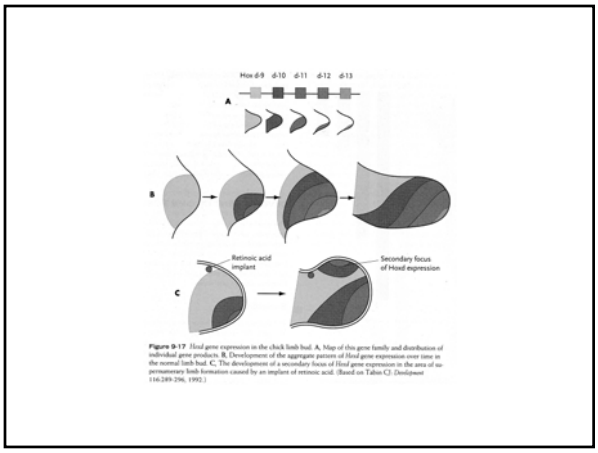
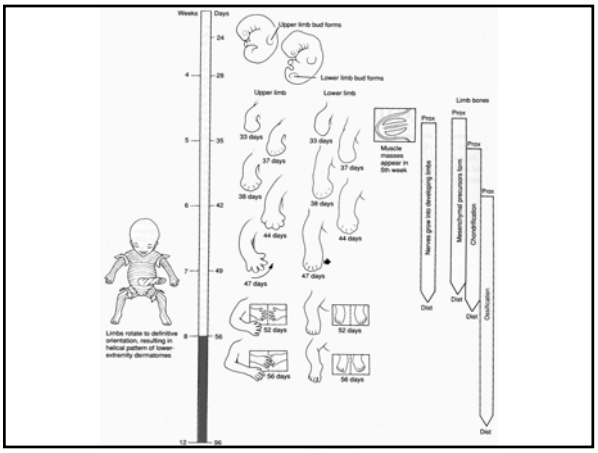
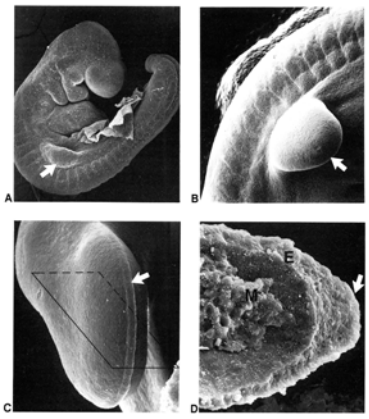
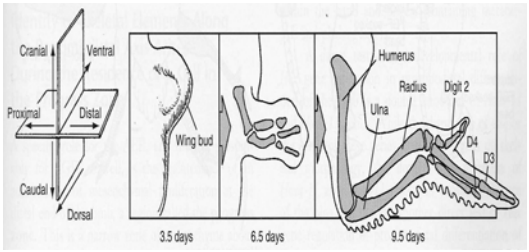
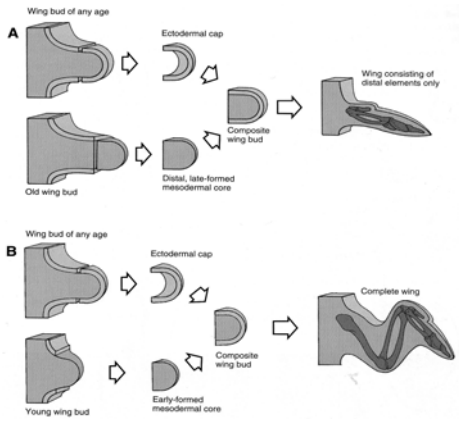


Figure 9-17 *Hoxd* gene expression in the chick limb bud. **A**, Map of this gene family and distribution of individual gene products. **B**, Development of the aggregate pattern of *Hoxd* gene expression over time in the normal limb bud. **C**, The development of a secondary focus of *Hoxd* gene expression in the area of supernumerary limb formation caused by an implant of retinoic acid. (Based on Tabin CJ: *Development* 116:289–296, 1992.)

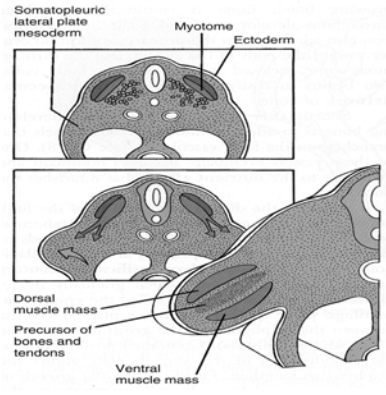


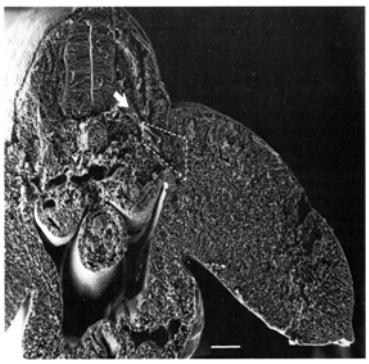


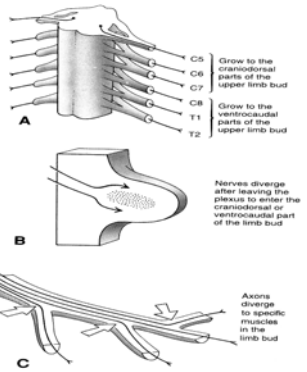


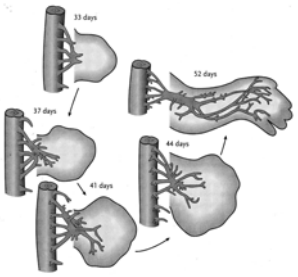


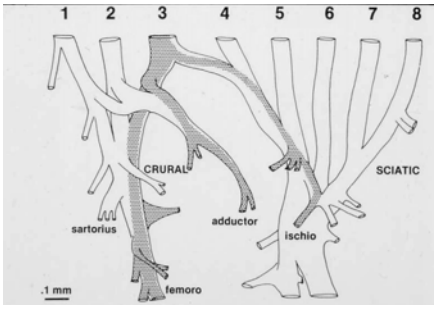
What would you predict would happen if you placed leg mesenchyme under the AER of the wing field?

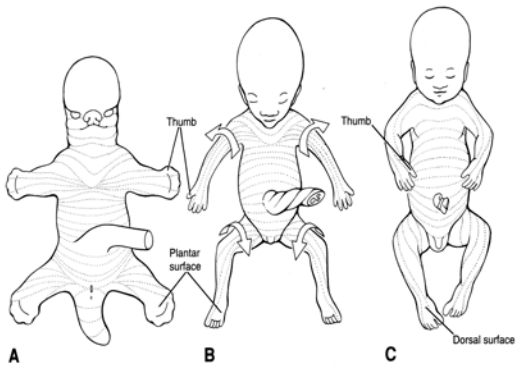












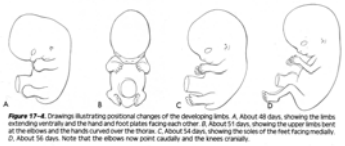
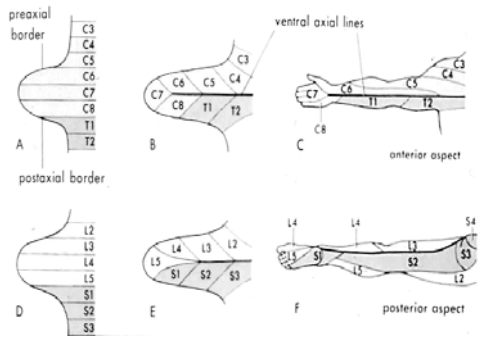
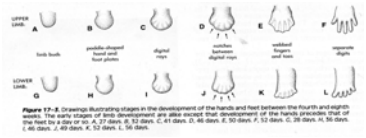
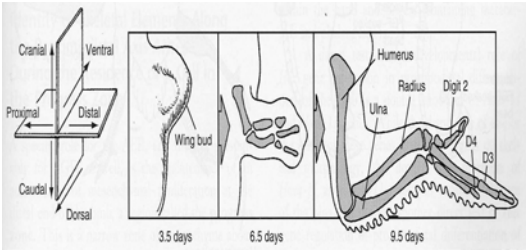


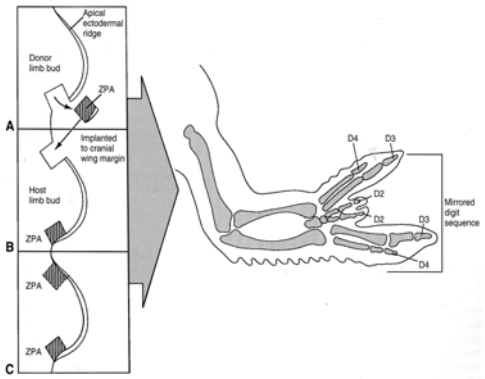
Figure 17–4. Drawings illustrating positional changes of the developing limbs. *A*, About 48 days, showing the limbs extending ventrally and the hand and foot plates facing each other. *B*, About 51 days, showing the upper limbs bent at the elbows and the hands curved over the thorax. *C*, About 54 days, showing the soles of the feet facing medially. *D*, About 56 days. Note that the elbows now point caudally and the knees cranially.





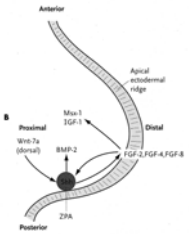
Limb is patterned as it develops.





What are the known characteristics of the ZPA?

1. If ZPA is transplanted (at the correct time) to an ectopic
site it will respecify tissue.
2. It acts to induce the correct A/P pattern via
a concentration gradient.
3. Acts over a very narrow time frame.
4. The activity of the ZPA is universal among
quadrapeds and is the same for the fore limb and hind limb.



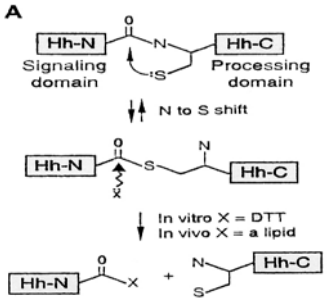
1. What is the polarizing signal?
   (a) Sonic hedgehog (Shh) is the polarizing signal.
   (b) Beads soaked in Shh can substitute for the ZPA.
   (c) The notochord (which makes Shh) can substitute for the ZPA.

2. Concentration gradient.

As suggested by the transplantation experiments, the polarizing activity stimulates differentiation of the mesenchyme in a concentration dependent manner.

This has been verified by using fibroblasts that have been transfected with Shh. Depending on the number of cells used to substitute for the ZPA different patterns of digit formation occur.

How is the concentration gradient of SHH established? Mechanism I.



What induces the ZPA?

In vitro ZPA loses polarizing activity if cultured alone.

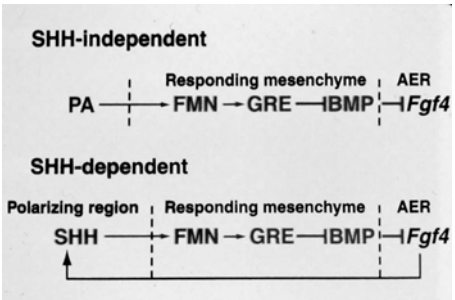
In vitro ZPA retains activity if cultured with AER.

If one ablates the AER completely, the ZPA is lost.

As in the leg/wing experiment FGF (4/8) can replace the AER.

FGF is made in the right place (over ZPA)

Its diffusion is limited by binding to ECM.



In summary:

Two sources of mesoderm limb: lateral plate forming cartilage and bone; somite derived cells forming muscle.

The trunk level of the lateral plate mesoderm determines whether it become forelimb or hindlimb.

The ectoderm is a signaling center regulating growth and it participates in patterning this outgrowth.

Because limb innervation arrives just as the muscle masses are forming, the segmental pattern of innervation will be altered as limb rotate.

The ZPA in the posterior aspect of the hand plate is induced by FGF from the posterior aspect of the AER. The ZPA in turn makes Shh. The concentration gradient of this signal results in patterned digit formation.