Developmental processes of tetrapod limbs

The patterning of Hox *genes*

The pattern of the *Hox* genes in the limbs involves only two *Hox* clusters, A and D, rather than all four as in the trunk. Front and hind limbs are patterned in an equivalent manner. The similarity in other aspects of development is sufficiently great that developmental biologists have coined special terms to refer simultaneously to the major elements of both limbs. **Stylopod** applies to the most proximate element of both the forelimbs and hind limbs (the humerus and the femur), **zeugopod** to the units formed by either the ulna and the radius or the tibia and fibula, and **autopod** to the more distal part of the limbs, including the carpals or tarsals (collectively termed the **mesopodials**), the metacarpals or metatarsals (together termed **metapodials**), and the digits and their included phalanges.

Morgan and Tabin (1994) showed that individual genes have different areas of overlap and may influence tissue development in different ways at different times during ontogeny. Both *Hoxa* and *Hoxd* genes are expressed in an anterior to posterior sequence within the limb, but more attention has been focused on *Hoxd* because of its anterior to posterior expression in the most distal portion of the limb, where the digits will develop. *Hoxd-9* is activated first and is expressed throughout the limb; *Hoxd-10* follows but is expressed only in the posterior half of the limb bud; and *Hoxd-11–13* are expressed in successively smaller areas of the posterodistal portion of the limb bud. These expression domains overlap one another, somewhat in the manner of Russian dolls, so that all five *Hoxd* genes are expressed in the posterodistal portion of the limb bud.

Targeted disruptions of *Hoxa* and *Hoxd* in the mouse indicate that development of the major limb elements in successively more distal portions of the limb bud is controlled by successive *Hox* groups (Davis et al. 1995): the shoulder girdle and pelvis by *Hox-9,* the humerus and femur by *Hox-10,* the ulna and radius, tibia and fibula, and the proximal carpals and tarsals by *Hox-11,* the distal tarsals and carpals by *Hox-12,* and the hands and feet by *Hox-13* (Fig. 10.14).

There are five overlapping areas of expression determined by the *Hox-D* series, extending from anterior to posterior: (1) d9 alone, (2) d9–10, (3) d9–11, (4) d9–12, (5) d9–13. The distribution of *Hox* genes in the distal portion of the limb has been used to suggest that they may be instrumental in establishing the number and identity of digits (Tabin 1992). Subsequently, Morgan and Tabin (1994) recognized that the changing domains of the various *Hox* genes during development made it impossible for them to code directly for specific digit number or morphology. They noted that in early development *Hoxd* genes are involved in regulating the growth of the undifferentiated limb mesenchyme, and suggested that the number of digits might be regulated by the amount of tissue produced at this stage. Later these genes regulate the maturation of the nascent skeletal elements, at which time the distinct morphology of the individual digits is determined. They concluded:

> *Hoxd* genes do not act in a simple combinatorial code for "digit identity." While they do contribute to the regulation of digit morphology, our current understanding of their action does not provide an indication of a constraint on potential morphologies. Either such a constraint remains to be discovered in the subtler aspects of *Hox* gene action or else one will have to look elsewhere for it. (pp. 185–6)