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Ann Burke, at Wesleyan University [16] in Middletown, Connecticut, studied the evolution of body structure in vertebrates. Craig Nelson, at the University of Connecticut in Storrs, Connecticut, also studied how vertebrate animals evolved. These two scientists collaborated with Bruce Morgan and Cliff Tabin, at Harvard Medical School [18] in Boston, Massachusetts. The focus of Tabin's research was on how genes [5] help produce structures in developing vertebrate animals, while Morgan focused on how genes [5] formed appendages in vertebrates. The group came together in 1995 to investigate to study how different developmental processes evolved. The group studied chicks (gallus gallus [19]) and mouse [7] (mus musculus [20]) embryos. They studied twenty-three Hox genes [5] in chicks and sixteen Hox genes [5] in mice, all of which they grouped into thirteen paralogue groups, or groups that are
genetically related after duplication occurs within the genome \[21\]. The researchers analyzed the expression of those groups in mice and in chicks to analyze the similarities and differences of Hox gene expression across species.

The group analyzed the boundaries of Hox gene expression of chick \[6\] and mouse \[7\] embryos by means of in situ hybridization. In situ hybridization is a technique that uses complementary DNA (cDNA) templates, or double stranded DNA synthesized from messenger RNA templates, to locate a specific nucleic acid sequence, and it helps the sequence of interest to become visible. Bruce, Burke, Nelson, and Tabin used in situ hybridization to find the relationship between Hox gene expression, or the amount of RNA produced by each Hox gene, and morphological boundaries along the anterior-posterior body axis in chick \[6\] and mouse \[7\] embryos. Specifically, the four scientists focused on sixteen Hox genes \[5\] that they thought functioned in axial development, or development of the central part of the body in chicks and mice. There were four kinds of Hox gene clusters, called Hoxa, Hoxb, Hoxc, and Hoxd.

Paralogue group four showed expression, or produced RNAs, within the cervical (neck) region of both the chick \[6\] and mouse \[7\]. Specifically, Hoxa-4, Hoxb-4 Hoxc-4 genes \[5\] expressed within the cervical region in the organisms. Hoxa-4 expressed in the anterior cervical vertebrae, and Hoxb-4 and Hoxc-4 express toward the middle cervical vertebrae. The researchers observed Hoxb-8, Hoxc-8, Hoxd-8 genes \[5\], all three members of the eighth paralogue group, in both the chick \[6\] and mouse \[7\] embryos. In both mouse \[7\] and chick \[6\] embryos, the Hoxc-8 genes \[5\] functioned in the cells bordering vertebra, the fifth thoracic vertebrae in the chick \[6\], and the sixth thoracic vertebrae in mice. Hoxd-8 and Hoxb-8 resulted in an unclear anterior-posterior area of gene expression in both species. The entire ninth paralogue expressed close to the end of the thoracic vertebrae in both animals, showing gene expression for four segments behind the forelimb in the chick \[6\] and nine segments behind the forelimb in the mouse \[7\]. All Hox10 paralogues expressed close to the lower spine (lumbosacral) region in both organisms, with Hoxd-10 expressed at the first sacral vertebra in both the chick \[6\] and mouse \[7\] embryos. Paralogue groups eleven through thirteen did not show gene expression as close together as the other groups, although groups eleven through thirteen all showed gene expression in the sacral and tail region of the embryos.

In their research report, the researchers hypothesized from their results that Hox genes \[5\] played a role in vertebrate evolution \[17\]. The researchers noted that the Hox gene expression of genes \[5\] in paralogue groups four through thirteen showed remarkably similar morphological expression among the chick \[6\] and mouse \[7\] embryos. They also noted that despite the significantly different overall body structure between the two organisms, specific anatomical areas, such as the vertebrae, showed a correlation in the types of individual genes \[5\] that were expressed. The researchers suggested that the correlation demonstrates that Hox genes \[5\] function in the segmentation \[11\] of the anterior-posterior vertebrate axis in both organisms. The researchers proposed that the minor shifts in developmental spacing of the genes \[5\], as noted with paralogue group nine, are caused by expansion or shortening of a specific body region, and not from differential expression of the genes \[5\] themselves. From those observations, the researchers argued that Hox genes \[5\] play a crucial role in the evolution \[17\] of axial variation and thus the evolution \[17\] of tetrapods.

The researchers hypothesized that the full range of Hox genes \[5\] were present in the common ancestor of tetrapods and fishes. Although lacking the axial regions seen in tetrapods, they claimed that it is almost certain that the Hox genes \[5\] played a role of anterior-posterior
In 1995, researchers Ann Burke, Craig Nelson, Bruce Morgan, and Cliff Tabin in the US studied the genes that regulate the construction of vertebra in developing chick and mouse embryos, they showed similar patterns of gene regulation across both species, and they concluded that those patterns were inherited from an ancestor common to all vertebrate animals. The group analyzed the head-to-tail (anterior-posterior) axial development of vertebrates, as the anterior-posterior axis showed variation between species over the course of evolutionary time. Along those axes, they showed where Hox genes produced RNAs. Hox genes have the homeobox, a portion of DNA contributes to the generation of the body plans of animals, plants, and fungi. In the 1995 study, the researchers compared the expression patterns of Hox genes across the chick and mouse embryos, showing where the patterns were similar and where they differed. Based on those comparisons, they argued that Hox genes were present in the ancestors of tetrapods and fishes, and that Hox genes function in the segmentation of the anterior-posterior vertebrate axis in both chick and mouse embryos.

**Sources**


