The single cell zygote contains all of the information required for the development of an adult organism. Understanding how this information is decoded, processed, and deciphered to create the amazing structures we animals are is a major incomplete scientific challenge. How are instructions for organizing body forms encoded as instructions at the molecular level? What information determines which part of the developing embryo will become the future head and what part the future limb? It is amazing how symmetry is maintained in the evolution of complex body designs. Understanding the action of a group of genes, called homeobox genes, has improved our current knowledge of these aspects of developmental genetics and the evolution of complex body forms.

Homeobox genes were first discovered in the fruit fly Drosophila melanogaster. Mutations in single genes were noted to produce flies that had normal body parts in abnormal places: legs where antennae were supposed to be, duplication of body segments producing an extra pair of wings, or the growth of an eye on a wing. It takes hundreds of genes to produce limbs, body segments with wings, and eyes; yet here were single genes that controlled all of those genes necessary to produce complex normal structures and place the parts in an abnormal location. Several of these control genes were sequenced and found to be similar in structure and function. They were named homeobox genes, from the Greek “homeosis,” which is the replacement of one body part by another. Each gene contained a 180 base-pair in length (the homeobox) sequence, which when expressed produces a 60 amino acid length protein. This protein then goes back into the nucleus, folds into three helices, and attaches at proper points on the DNA to act as transcription factors that control a host of genes.

Homeobox genes are found in all animals and many plants and fungi. They predate even the divisions of the living kingdoms. They are highly conserved (ie, the sequence that affects the thorax of the fly similarly affects the thorax of vertebrates).

Certain homeobox genes (HOX genes) are linked together in a cluster and are transcribed in an order that corresponds to the temporal and structural development of an embryo. Transcription begins at the 3’ end of the strand where the head structures are controlled and proceeds down to the 5’ end where the genes that control the hind structures are found. The timing, sequence, and dosage of expression of several developmental genes that play an important role in embryonic
patterning and cell differentiation are controlled by these genes. These genes, which regulate many other genes, have been aptly called the “Master Genes.”

The HOX genes determine cell polarity, patterning of the anteroposterior axis, and many dorsal and ventral details. HOX genes are not linked to specific morphologies or cell types but are instrumental in providing abstract spatial information. Each cell in the embryo receives positional information that leads to its destiny and fate. These cells receive instructions from the HOX genes and undergo one or more of the following: cell division, migration, differentiation, or cell death. Thus, these groups of cells, known as functional domains, become committed to form body structures such as limbs and organs. The boundaries of HOX domains often coincide with morphological transitions. The complexity of the HOX gene has been instrumental in the evolution of animals with increasingly complex body forms. Evolution takes place in part by increasing the number and complexity of the HOX genes and their clusters. Linear duplication allows for increasing segments (just ask the centipede or earthworm). When by chance a HOX gene is duplicated in a cluster, the original gene can control the formation of what it vitally did initially and the duplicated gene can then mutate and create specialized new structures such as the addition of mouth parts, wings, limbs, or new organs. Humans have 4 clusters of 10 HOX genes each. These 40 specialized homeobox genes define the placement of the “pieces” necessary to construct a human.

Homeobox genes are present on every human chromosome. Their total number in the human genome exceeds 300. Eye development in animals is linked to a set of homeobox genes known as the PAX family. PAX genes are found to control eye development in all animals including both simple and complex forms. Humans have 9 unlinked genes. (PAX1 through PAX9). The PAX6 gene is a paradigm for our current understanding of the molecular genetics of mammalian eye development. Unlike HOX genes, PAX genes are not clustered but are scattered throughout the genome.

Transcription of DNA to mRNA and subsequent translation of mRNA to protein is the central dogma of molecular genetics. Human PAX6 (Figure 1) is transcribed as a 2.7-kb mRNA and encodes a 422-amino acid transcriptional regulator protein that includes several DNA-binding domains that control gene expression: a paired box domain (PRD) at the N terminal, a paired-type homebox domain (HD), and the PST (proline, serine, and threonine-rich sequence) transregulatory domain at the C terminal. The homeodomain is the most highly conserved region of the PAX6 protein. The paired domain is composed of two distinct DNA-binding subdomains, the N-terminal subdomain (NTS) and the C-terminal subdomain (CTS), which bind respective DNA sequences. The N-terminal PRD includes an in-frame 14-amino acid alternatively spliced exon (5a), which is separated from the HD by a linker region. Hence, depending on how the mRNA is spliced, PAX6 can regulate expanded or restricted set of genes. Exon 5a appears to function as a molecular indicator that specifies target genes. Both upstream and downstream modulation of PAX6 occurs. PAX6 is required for formation of the lens placode, an ectodermal thickening that precedes lens development. Because development of most ocular structures is greatly dependent on normal induction of the lens, the critical role played by PAX6 is evident.

Human PAX6 was identified as a candidate gene for aniridia during the search for the genes responsible for WAGR syndrome (Wilms tumor, aniridia, genitourinary malformations, and mental retardation) caused by a 11p13 deletion. Aniridia results from PAX6 haploinsufficiency, the loss of function of one copy of the gene. Mutations in PAX6 have been shown to cause a spectrum of congenital anomalies of the eye. These include Peter’s sequence, congenital cataract with late onset corneal dystrophy, autosomal dominant keratitis, and foveal hypoplasia. Congenital optic nerve anomalies including optic nerve hypoplasia, coloboma of the optic nerve head, optic nerve aplasia, and morning glory disc anomaly can also occur. Extraocular sites of PAX6 expression include the developing olfactory system, brain, neural tube, and endocrine pancreas.

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