The Gene Content of Mammalian and Avian Sex Chromosomes

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Sex chromosomes evolve specialised collections of genes, making them distinct from autosomes. Recent, high-quality, genomic sequencing of sex chromosomes allows us to trace the evolutionary history of the mammalian X and Y chromosomes and avian Z and W chromosomes, and examine how evolution has remodelled their gene content. The XY and ZW sex chromosomes arose from ordinary pairs of autosomes. Differentiation into specialised sex chromosomes initiated with the degeneration of gene content on the Y and W chromosomes and the preservation of gene content on the X and Z chromosomes. As these degeneration and preservation processes unfolded, the sex chromosomes also acquired new genes, many of which are located in large, nearly identical, segmental duplications called amplicons. Ampliconic sequences, independently evolving across multiple lineages, are a major driving force in sex chromosome evolution.

Introduction

Sexual reproduction evolved ~1 billion years ago. Sex chromosomes have evolved as one of the mechanisms to determine sex in a wide range of eukaryotes, including fungi, plants and animals. Across animals, two primary sex chromosome systems have evolved, XY and ZW systems. In mammalian XY species, females are XX and males are XY. The sex carrying a matching pair of sex chromosomes is reversed in avian ZW species, where males are ZZ and females are ZW. In both systems, the Y and W chromosomes are both morphologically smaller than the X or Z and are typically comprised of fewer genes, many of which are involved in sex-specific processes. The X and Z chromosomes are morphologically similar in many respects to autosomes, but also contain large collections of genes involved in sex-specific processes. Beyond the initial determination of sex, the gene content of sex chromosomes is specialised to govern additional sex-specific functions.

Sex Chromosomes as Former Autosomes

The mammalian X and Y chromosomes and avian Z and W chromosomes originated from autosomes (Bellott et al., 2010; Nanda et al., 1999). DNA sequence comparisons between mammals and birds show that in their most recent common ancestor, the current mammalian X and Y chromosomes were one pair of autosomes and the chicken Z and W chromosomes were another pair of autosomes. Thus, both XY and ZW systems evolved independently from different pairs of ancestral autosomes into highly specialised sex chromosomes.

The differentiation of the mammalian X and Y and avian Z and W chromosomes from autosomes represents reciprocal experiments of nature, where males are XY in mammals and females are ZW in birds. Similar evolutionary processes are suggested to govern the differentiation of both sex chromosome systems. Since the evolution of the mammalian X and Y chromosomes has been studied more extensively, this system will serve as the basis of comparison. Early differentiation of the X and Y chromosomes is thought to have initiated via the acquisition of a male
sex-determining gene, Sry, on one of the ancestral autosomes (Figure 1). Suppression of recombination around the Sry gene then occurred via a large inversion of the surrounding genomic region (Figure 1). It is also possible that an inversion existed before the formation of the male sex-determining gene, Sry. Large inversions naturally prevent X–Y recombination from occurring across the inverted genomic region, as the requisite base pair ing cannot be achieved. In this case, the resulting repression of recombination not only fixed the male sex-determining gene on one of the two ancestral autosomes, but also created the opportunity for sequences throughout this inversion region to diverge unchecked from those on the other ancestral autosome. Thus, the X and Y became distinct sex chromosomes, with the male sex-determining gene, Sry, specifying the Y chromosome.

Genes within an inverted region on the Y chromosome accumulate mutations because of the lack of recombination with the X chromosome. Over time, the lack of recombination within inverted Y chromosomal regions leads to the accumulation of deleterious mutations, sequence degeneration and loss of many Y-linked genes (Figure 1). Through recombination between the X chromosomes in females, the X chromosomes preserve both the ancestral autosomal gene order along the chromosome and the ancestral autosomal gene sequences. Therefore, within inversions, sequences of the homologous pairs of genes on the X and Y chromosomes diverge from one another over time. The extent of sequence divergence between homologous pairs of genes has been measured and used to cluster genes along the X chromosome according to the amount of time they have been nonrecombinant with their homologues on the Y chromosome. Five evolutionary ‘strata’ or physical groupings of genes of similar levels of sequence divergence between X and Y homologues are seen clearly along the human X chromosome, suggesting that five separate Y chromosome inversion events have successively expanded the nonrecombinant region over evolutionary time (Lahn and Page, 1999a; Ross et al., 2005). After becoming mostly nonrecombinant with the X, the Y chromosome continued to experience inversions and other genomic rearrangements, thus scrambling the locations of genes within each stratum on that chromosome. The oldest stratum (stratum 1) is the result of the first Y chromosome inversion event and is equivalent to the long arm of the human X chromosome. Sequence divergence decreases in the proximal to distal direction on the short arm of the human X chromosome (stratum 2–5) (Lahn and Page, 1999a; Ross et al., 2005). Over evolutionary time, the Y chromosome lost massive numbers of ancestral autosomal genes, leading to its diminished physical constitution as compared to the stately X chromosome.

When a gene copy is lost from the Y chromosome, the dosage of gene expression from the remaining copy on the X chromosome may need to be adjusted for cellular survival. In mammals, balancing the gene expression between XX and XY cells is achieved via a combination of X-inactivation (where in XX females one X is randomly transcriptionally silenced per cell) and the upregulation of some gene expression from a single active X chromosome in both XX females and XY males (Deng et al., 2014). Once dosage compensation is achieved, geneticist Susumu Ohno predicted ancestral autosomal genes on the X chromosome would be retained over evolutionary time (Ohno, 1967). He predicted that as genes were lost on the Y chromosome, the X chromosome would compensate for this loss by upregulating the expression of X-linked genes two-fold in males and females (because one X chromosome is randomly inactivated). Gene movement from the X chromosome to an autosome would likely produce detrimental cellular effects as a result of the overexpression of the gene involved, because compensatory upregulation of expression was established on the X chromosome. Gene sequence comparisons between the X chromosomes of many mammalian species support Ohno’s hypothesis: the X-linked genes experiencing dosage compensation are highly conserved across many mammalian species. In addition, by applying Ohno’s prediction to the evolutionary strata of divergent genes on the X and Y, one would predict that the most divergent genes in stratum 1 are more likely to be regulated by dosage compensation than the less divergent genes in the younger strata (stratum 5). Indeed, stratum 1 contains more genes that experience dosage compensation than does stratum 5. The loss of genes from the Y chromosome and the establishment of dosage compensation for the remaining gene copies on the X chromosome has preserved almost the full complement (94%, 543/580) of the ancestral autosomal genes on the human X chromosome (Bellott et al., 2010; Ross et al., 2005).

See also: Mammalian Sex Chromosome Evolution

X–Y genes that are not enveloped in the inversions leading to strata formation lie within the pseudautosomal regions of the sex chromosomes (PAR, see Figure 2). PAR genes on the X and Y chromosomes are derived from autosomes (either ancestral autosomes or a more recent translocation of an autosome to a sex chromosome) and are nearly identical in sequence. PAR genes maintain the nearly identical sequence, because within the PAR the X and Y chromosomes can freely recombine, just like any pair of autosomes. Because males and females carry the same number of functional PAR gene copies, there is no need for dosage compensation of these genes. PARs are required for the X–Y sequence pairing and recombination as part of proper chromosome segregation during male meiosis. All mammals studied to date have PAR genes, but they represent only a few of the ~1000 protein-coding X-linked genes. Most ancestral autosomal genes have diverged along independent evolutionary trajectories on either the X or Y chromosome.

Mammalian X and Y Chromosomes

X chromosome

Males (XY) have only a single X chromosome and thus are more susceptible to the phenotypic effects of recessive X-linked gene mutations. For example, in humans, recessive X-linked gene mutations are responsible for many diseases that primarily occur in males: muscular dystrophy, hemophilia, red-green colour vision defects, and severe combined immunodeficiency to name a few. Females (XX) can be carriers of X-linked recessive gene mutations without any phenotypic effects, if they also carry normal copies of the genes. However, females with two copies of an X-linked recessive gene mutation will display the associated phenotypic effect. Consequently, a hallmark of an X-linked recessive gene mutation is a familial inheritance pattern...
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Figure 1 A simplified example of stepwise formation of strata and degeneration of Y-linked genes. This represents a generic mammalian X–Y chromosome pair. Sry evolves as the male sex-determining gene on the Y chromosome. An inversion on the Y chromosome leads to suppression of recombination with the X chromosomes and thus the formation of the first stratum (genes B, C, D, E). The lack of recombination on the Y chromosomes leads to the degeneration of genes. Subsequent inversions encompassing X–Y recombining portions of the sex chromosomes lead to the formation of additional strata (gene F).

Figure 2 Mammalian and avian sex chromosomes generated with high-quality sequencing approaches. Each chromosome is drawn to scale and colour-coded, where the underlying sequence is ancestrally autosomal (yellow), ampliconic (blue), pseudoautosomal (green), heterochromatic (yellow with red lines) or other (grey). Centromeres are labelled as cen.
Y chromosome

In terms of genetic disease studies of sex chromosomes, the Y chromosome does not have the same rich history as the X chromosome. The Y chromosome has been viewed as genetically inert; large heterochromatic regions of sequence (~40 megabases, Figure 2) suggest that the Y chromosome is devoid of significant biological function. However, recent acquisition of high-quality genomic sequences of mammalian Y chromosomes from humans, chimpanzees, rhesus monkeys and mice reveal heterochromatic regions are only present on the human Y chromosome (Skaletsky et al. 2003). Thus, at least in mammals, three of the four Y chromosomes sequenced to date are largely comprised of euchromatic sequences, instead of heterochromatic sequences.

The euchromatic regions of mammalian Y chromosomes harbour genes critical to male sexual phenotypes, sexual identity and the production of sperm. Genes on mammalian Y chromosomes fall into two general classes: ancestral autosomal genes and newly acquired genes. Ancestral autosomal genes have homologous genes on the X chromosome and represent genes that date back to the most recent common ancestor of mammals and birds (Bellott et al., 2014). The number of Y-linked ancestral autosomal genes in mammals ranges from a low of 7 in mice to high of 18 in rhesus monkeys (Hughes et al., 2012; Soh et al., 2014). The Y chromosome acquired new genes by either retrotransposition or transposition from autosomes or arose de novo (Figure 3b). Many of the newly acquired genes are harboured within large, nearly perfect, segmental duplications, or amplicons, which have arisen independently in mammalian Y chromosomal lineages (Figure 3b). See also: Y Chromosome

Sex-Linked Gene Classes

Ancestral autosomal genes

The large blocks of sequence similarity between chicken chromosomes 1 and 4 and the human X chromosome highlight its former life as an ancestral autosomal. Because the gene density on the human X chromosome is reduced from the autosomal average of 12 genes per megabase to 7 genes per megabase, the human X chromosome is considered to be ’gene poor’ (Bellott et al., 2010; Ross et al., 2005). This lower gene density, however, is not due to the loss of ancestral autosomal genes, but is in part attributable to the accumulation of long interspersed elements-1 (LINE-1) and other retrotransposable elements. In fact, just as Susumu Ohno predicted, the human X chromosome has conserved 94% (543 of 580) of the ancestral autosomal genes (Bellott et al., 2010; Ohno, 1967). The conservation of ancestral autosomal genes on the mammalian X chromosome has been confirmed by numerous studies (Mueller et al., 2013). As discussed, the conservation of mammalian X-linked genes is considered to be a consequence of dosage compensation for the massive attrition of homologous genes on the Y chromosome.

In striking contrast to the conservation of gene content on mammalian X chromosomes, mammalian Y chromosomes retain only 2–3% (17 genes on the human Y chromosome) of the ancestral autosomal genes (Lahn and Page, 1999a; Skaletsky et al., 2003). Ancestral autosomal genes on the Y chromosome are typically single-copy and share a number of unique properties, suggesting that these common features may have led to their retention. Ancestral autosomal genes are broadly expressed across most cell types, are dosage sensitive and function in cellular housekeeping processes (Bellott et al., 2014; Cortez et al., 2014). Phenotypes associated with Turner syndrome (XO individuals) patients may be attributable to the loss of ancestral autosomal genes as well as to the loss of the genes within the PAR. The sequences of the ancestral autosomal genes on the Y chromosome have diverged significantly from their X-linked counterparts. This sequence divergence results in unique proteins and, in turn, unique protein functions may contribute to differences in cellular processes in XX and XY cells. See also: Evolution of the Mammalian X Chromosome

Newly acquired genes

In addition to ancestral autosomal genes, the gene content of mammalian sex chromosomes has been bolstered by the acquisition of new genes by both transposition and retrotransposition from autosomes (Figure 3b). The study of retrotransposition to and from the X chromosome in humans and mice shows that in both species the X chromosome is a common origin and recipient of retrotransposition events (Emerson et al., 2004). Whether acquired by transposition or retrotransposition, newly acquired genes are subject to the unique natural selective pressures of the sex chromosomes. For example, the transcriptional silencing of the sex chromosomes during male meiosis, known as meiotic sex chromosome inactivation (MSCI), is considered to be a selective force for genes critical to spermatogenesis to move off the X chromosome and onto autosomes (Potrzebowski et al., 2008; Wang, 2004). Alternatively, evolutionary theory posits that the single X chromosome in males naturally makes the X chromosome a selectively advantageous location for male-benefit genes (Rice 1984). In females, beneficial genes on the X chromosome need to either have dominant effects or be present in two copies to confer a selective advantage. See also: Meiotic Sex Chromosome Inactivation

Retrotransposition and transposition of autosomal genes has also governed gene movement onto the Y chromosome. For example, on the human Y chromosome, retrotransposition explains the acquisition of CDY from the autosomal gene CDYL.
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Duplication of pre-existing ancestral sex-linked genes

Ancestral autosome

Evolved sex chromosome

Gene type

Ampliconic

Retrotransposition, transposition and subsequent duplication of autosomal genes

Ancestral autosome

Evolved sex chromosome

Gene type

Single-copy

Duplication

Ampliconic

Figure 3  Mechanisms of new gene acquisition and the development of amplicons on the sex chromosomes. Newly acquired sex-linked genes have evolved via duplication of a preexisting sex-linked gene (red vertical line indicates gene) or gene movement from an autosome to the sex chromosomes. (a) Large genomic regions surrounding preexisting sex-linked genes are duplicated (amplicons, shown as blue arrows) and maintained with a high level of sequence identity between the duplicated ampliconic sequences. (b) Autosomal genes are retrotransposed or transposed onto the sex chromosomes and subsequently duplicated.

(Lahn and Page, 1999b) and transposition explains the acquisition of the DAZ gene from its autosomal copy DAZL (Saxena et al., 1996). In each of these cases, and others, the newly acquired genes exhibit testis-specific expression. In addition to retrotransposition and transposition of genes onto the Y chromosome, some Y-linked genes have apparently arisen de novo. The origins of de novo genes are unknown and they typically do not contain protein domains suggestive of function. For all three types of newly acquired genes on the sex chromosomes, a common theme exists: newly acquired genes are present in multiple copies. In many cases, the multiple copies are adjacent to each other, suggesting that a single gene is newly acquired and subsequently duplicated.

Ampliconic genes

Amplicons are large, nearly identical, inverted or tandem segmental duplications that are greater than 10 kilobases in size. Ampliconic sequences and their associated genes are enriched on both the X and Y chromosomes (Figure 2), as compared to autosomes. Amplicon harbour genes, which preexisted on the sex chromosomes (Figure 3a) or were newly acquired (Figure 3b). Genes in these ampliconic repeats are expressed predominantly in testicular germ cells. Recently, a concerted effort was made to determine the correct genomic sequence of X-ampliconic regions in humans and mice (Mueller et al., 2013). In total, 232 and 109 X-linked genes are ampliconic in mice and humans, respectively (Mueller et al., 2008; Mueller et al., 2013). Comparison of mouse and human sequences shows amplicons evolved independently in each species after they diverged from a common ancestor. Convergent evolution of amplicons in mice and humans suggests that they serve an important function in reproductive strategies and in the development of testicular germ cells.

While ampliconic genes vary substantially in copy number, most are found in palindromic-oriented arrangements. The total number of gene copies in mammalian Y-amplicons ranges from 696 gene copies in mice to 12 gene copies in rhesus monkeys (Hughes et al., 2012; Soh et al., 2014). The massive number of gene copies on the mouse Y chromosome is the result of repeated duplication of just three genes (Soh et al., 2014). The palindromic nature of amplicons promotes intrachromosomal gene conversion, preventing Y-chromosome genes from accumulating inactivating mutations, replacing nonfunctional alleles with functional copies (Rozen et al., 2003). This mechanism slows or possibly stops the attrition of Y chromosomal genes. This suggests that once a gene is newly acquired on the Y chromosome the gene is duplicated and the gene duplication is inverted to form a self-preserving palindrome. In addition, amplicons may provide the benefits of forming unique chromatin configurations enabling gene expression despite male MSCI and performing essential functions during spermatogenesis (Mueller et al., 2008). Thus, in addition to transposition off the sex chromosomes, incorporation into large palindromic repeat structures represents another gene mechanism for tolerating the unique natural selective pressures applied to the sex chromosomes.

Avian Z and W Chromosomes

Avian species whose genomes have been studied to date share the same Z and W chromosomes with the last common avian ancestor. Because of the limited number of high-quality Z and W chromosome sequence assemblies, our understanding of avian
Z and W chromosome evolution and gene content is more limited than for the well-studied mammalian X and Y chromosomes. Sex chromosome aneuploidies played an important role in discovering the necessity of the Y chromosome in mammalian male sex determination. However, sex chromosome aneuploidies are rare in birds, making it more difficult to dissect the function of the W chromosome. Despite these setbacks, the avian ZW and mammalian XY systems appear to share a similar history, evolving from a pair of autosomes into a pair of highly diverged sex chromosomes. ZW chromosomes display features such as the formation of strata, the acquisition of new genes and the formation of ampliconic sequences.

Similar to the mammalian XY system, the avian ZW chromosomes evolved from a pair of autosomes, albeit from a different pair of autosomes than the mammalian X and Y chromosomes (Bellott et al., 2010; Nanda et al., 1999). The sequence of the chicken Z chromosome shares many of the same features found on the mammalian X chromosome: retention of the majority of its ancestral autosomal gene content, low gene density compared to avian autosomes, high density of LINE-1, and the existence of ampliconic repeats. A substantial fraction of Z chromosome gene content is comprised of ampliconic genes expressed in the testis (Bellott et al., 2010). Large inversions and subsequent degeneration of the W chromosome genes also appear to have left evolutionary strata (Nam and Ellegren, 2008), although the number of strata and the strata boundaries are not as clear as in mammals.

In contrast to the mammalian XY system, sex determination in the ZW system appears to be governed by Z-dosage and not a dominant female-determining gene carried on the W chromosome. Gene knockdown studies of a Z-linked gene, DMRT1 (Smith et al., 2009), results in feminisation of gonad, indicating DMRT1 is required for male sex determination. The hypothesis is that two copies of DMRT1 (in ZZ males) are necessary for male sex determination, while a single copy of DMRT1 (in ZW females) specifies female sex determination. It will be of great interest to identify other factors that fit into the male and female sex determination pathways in relationship to DMRT1.

### Unusual Mammalian Sex Chromosome Systems

Although most mammalian sex determination pathways rely on the XY sex chromosome system that has developed since the evolutionary divergence of mammals and birds, there are a few exceptions. For example, among New World monkeys, red howler males have acquired a neo-Y chromosome function due to a translocation of a Y chromosomal segment onto an autosome (Consigliere et al., 1996). In addition, rodents have a bizarre collection of sex chromosome systems. For example, mole vole (Eliobius lutescens) males and females are both XO, where the Y chromosome is lost and the Sry gene is translocated onto the X chromosome, suggesting a X0/X0 system. Wood lemming (Myopus schisticolor) males are XY and females are XX, X*X, and X*Y, where a mutation on X* overrides Y-governed sex determination (Fredga et al., 1976). The most bizarre of all mammalian sex chromosome systems belong to monotremes. For example, platypus males have five different X chromosomes that pair with five Y chromosomes to form a chain during meiosis (Grutzner et al. 2004). Most likely, additional sex chromosome systems will be discovered by the study of other species of New World Monkeys and rodents, because both lineages exhibit high rates of chromosomal rearrangements, including translocations. As costs of sequencing continue to fall, exciting new high-quality sequence assemblies of these unusual sex chromosome systems will provide insights into the mechanisms of sex determination and the evolutionary processes that have shaped their gene content. See also: Evolution and Organization of Monotreme Sex Chromosomes

### Conclusions

Millions of years of evolution have shaped the gene content of mammalian XY and avian ZW sex chromosomes to be distinct from that of autosomes. Sex chromosomes are constantly remodelling to preserve and protect both ancestral autosomal genes and newly acquired genes for specialised functions. Most of the ancestral autosomal genes are found on the X or Z chromosomes. Unique to the sex chromosomes, many of the newly acquired genes are arranged in multiple copies and in large ampliconic sequences. Studies of sex chromosome systems of other vertebrates will be important to determine if the evolutionary features of XY and ZW gene content are either unique to mammals and birds or common among many lineages.

### References


**Further Reading**


