The Evolution of Sex Determination in Plants

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Abstract

Separate sexes, i.e., the presence of male and female individuals in a species (= dioecy), do exist in flowering plants, despite being much less common than in animals. How becoming a male or a female (= sex determination) is achieved in dioecious plants is much less understood than it is in animals. On one hand, phylogenetic, ecological, and theoretical population genetics studies have provided a lot of information on what could be the evolutionary routes from hermaphroditism, the assumed ancestral sexual system in angiosperms, to dioecy, and what could be the genetics and the selective forces driving the evolution of males and females. On the other hand, genetic, molecular, and developmental data are scarce. Sex chromosomes have been described in a few dioecious species, and very recently two master sex-determining genes have been identified. We review here the theoretical findings on the evolution of dioecy and sex

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determination in plants and also discuss recent work on the genetics of the evolution of dioecy and on the molecular characterization of the first master sex-determining genes found in plants.

Keywords

Angiosperms · Sex determination · Sex chromosomes · Sexual systems · Dioecy · Monoecy · Gynodioecy

Introduction

Albeit rare, dioecious plant species, that is, species with male and female individuals (as typically found in most animal species) exist in flowering plants and represent 15,600 species (i.e., 5–6% of all species, Renner 2014). Sexual systems are very diverse in plants, but the most common sexual system is hermaphroditism with bisexual flowers, which suggests that this is the ancestral sexual system (Barret 2002). It is thus assumed that dioecy has evolved from hermaphroditism and it has done so repeatedly, as dioecy is widespread in plants and found in 43% of all families (Renner 2014). A total of 871 to 5000 independent origins of dioecy in plants have been estimated (Renner 2014). In most cases, these events are very recent and sometimes how dioecy has evolved can be tracked. Why dioecy has remained at a low frequency in angiosperms, whereas its frequency is much higher in animals (>95%, see Table 1) and in other land plant lineages such as gymnosperms (36%), mosses (50%), and liverworts (75%) are not yet clear (Käfer et al. 2017).

In animals, a range of sex determination mechanisms have been described (Bachtrog et al. 2014): genetic (sex chromosomes, polygenic systems, single gene) or environmental (e.g., temperature-dependent), and some information on sex determination mechanism is known for >90% of animal species (Table 1). In plants, we know much less on sex determination. Sex chromosomes have been reported in ~40 species out of the 871 to 5000 independent dioecious systems (Ming et al. 2011; Renner 2014; Muyle et al. 2017). Dozens of master sex-determining genes such as *Sry*, the male-determining gene in mammals, have been identified in animals (Bachtrog et al. 2014), and the gene network for sex determination is well

Table 1 Frequency of species with separate male and female individuals and the current state of knowledge on sex determination in animals and flowering plants

	Animals	Flowering plants
% of species with separate sexes	95%	5–6%
% of species with rough information on sex determination mechanisms[a]	>90%	<1%

From (Weeks 2012; Barrett 2002; Ming et al. 2011; Renner 2014; Bachtrog et al. 2014; Muyle et al. 2017)

[a] Genetic or environmental, presence of sex chromosomes

characterized in several groups such as vertebrates (Matson and Zarkower 2012). By contrast in plants, only two master sex-determining genes have been recently identified (Agaki et al. 2014; Murase et al. 2017). We do not yet know how widespread sex chromosomes are in plants, or whether other genetic mechanisms and environmental sex determination have evolved in this taxon, and we have not yet identified the vast majority of the molecular players involved in plant sex determination.

This current lack of knowledge strongly limits our understanding of dioecy both at functional and evolutionary levels, which we will see is based on a few well-studied species and some elegant theories, but with little empirical evidence to support them. This problem has major implications for the understanding of dioecy in crops, as ~20% of all crop species are dioecious, or derive from a dioecious progenitor. Examples of dioecious crops are kiwi, asparagus, hop, cannabis, date-palm, and persimmons, while crops such as papaya, grapevine, and strawberries are dioecious-derived. In species where only one sex (female in general) has an agricultural utility, the lack of genetic markers for sexing to select out the useless sex can generate huge costs, especially in trees where sexual maturity (and the opportunity of sexing individuals by looking at flowers) is reached only after several years. Moreover, our lack of knowledge on sex-determining genes prevents us from controlling the sexual system of crops.

Here we will present the data and theories that are currently available for dioecious plants. The first part of our chapter concerns theoretical aspects of dioecy and sex determination in plants, which have been developed despite the paucity of concrete data in this field of research. The second part concerns the data that have been accumulating at a slow pace for a long time. However, this situation is changing and we will discuss recent important empirical findings in plant sex determination.

Theories on the Evolution of Dioecy

The Routes to Dioecy

By both looking at the sexual system of close relatives of dioecious species and reasoning on how dioecy could evolve using population genetics, it was suggested several decades ago that dioecy has probably not evolved directly from hermaphroditism, but indirectly along various routes via different intermediates with distinct sexual systems (all sexual systems found in angiosperms and their frequencies are shown in Table 2). Two main routes have been proposed (Barrett 2002, see Fig. 1A and B): the first of these involves a monoecious intermediate (monoecy = presence of separate male and female flowers on the same individual in a population, Renner and Ricklefs 1995), while the second involves a gynodioecious intermediate (gynodioecy = presence in a population of individuals with bisexual flowers and others with female flowers, Charlesworth and Charlesworth 1978).

The gynodioecy-dioecy pathway has been well studied theoretically (Charlesworth and Charlesworth 1978; Charlesworth 1999), and there is some

Table 2 Frequency of sexual systems in angiosperm species

	Dimorphic individuals (two or more distinct forms)	Monomorphic individuals (only one phenotype)
Bisexual flowers	Heterostyly <1%	Monocliny ~ 80%
Female and bisexual flowers	Gynodioecy <1%	Gynomonoecy 2–3%
Male and bisexual flowers	Androdioecy ≪1%	Andromonoecy 1–2%
Female and male unisexual flowers	Dioecy 5–6%	Monoecy 6–7%

Adapted from Käfer et al. (2017)

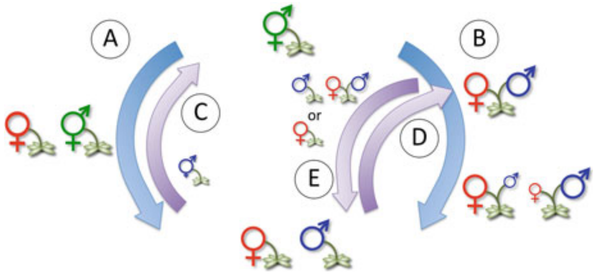

Fig. 1 The main evolutionary routes to dioecy. Two main routes from hermaphroditism (the assumed ancestral sexual system in angiosperms) to dioecy have been proposed: the gynodioecy-dioecy route (*A*) and the monoecy-dioecy route (*B*). It has been recently highlighted that dioecy may frequently revert to hermaphroditism (*C*, *D*), hence its rareness (Käfer et al. 2017). In the gynodioecy-dioecy route, inconstant males (males producing few viable seeds) may help reversions to hermaphroditism as drawn in C (Ehlers and Bataillon 2007). In the monoecy-dioecy route, there may be cycles between monoecy and dioecy (*D*, *E*). Some work suggests that in this case, androdioecy (males and monoecious individuals) and gynodioecy (females and monoecious individuals) may also evolve and may represent a situation of the re-evolution of dioecy after it has reverted to monoecy (e.g., Pannell et al. 2014). See text for details

phylogenetic evidence suggesting that many dioecious species may have evolved through a gynodioecious intermediate (Dufay et al. 2014). This pathway starts with a hermaphroditic population in which a male-sterility mutation producing females appears (Fig. 1A). Those females will reallocate all their reproductive energy to producing seeds and may produce more seeds than hermaphrodites. If the hermaphroditic population suffers from inbreeding depression, due for example to some self-fertilization, the females may not only produce more seeds but also seeds of better quality through obligate outcrossing. At some point, a female-sterility mutation producing males might appear. Those males will reallocate all their reproductive energy to producing pollen and will outcompete the hermaphrodites in fertilizing the females. The population has become dioecious.

A strong co-occurrence of monoecy and dioecy in the angiosperms phylogeny suggests that the monoecy-dioecy pathway is widespread among angiosperms perhaps more widespread than the gynodioecy-dioecy pathway (Renner and Ricklefs 1995; Renner 2014). However, much less has been done on modeling the transition from hermaphroditism to monoecy and then to dioecy, and in particular, no population genetics model is available. In this pathway, the hermaphroditic population first evolves to a monoecious population in which both unisexual flower types can be found on the same individual (Fig. 1B). Then, it is assumed that disruptive selection (in which extreme values for a trait are favored over intermediate values) on the quantitative genetic variation in floral sex ratios within a monoecious population will gradually increase sex specialization, culminating in female and male individuals (Charnov 1982).

The androdioecy-dioecy pathway in which males would have evolved first in a hermaphroditic population is considered very unlikely (androdioecy = presence in a population of individuals with bisexual flowers and others with male flowers). First, contrary to monoecy and gynodioecy, androdioecy is extremely rare in angiosperms (Table 2). Second, in a hermaphroditic population where the individuals can self-fertilize, this pathway would require the production of a huge amount of pollen for the males to spread. The spread of females is much easier. A single example of androdioecy that may have evolved from hermaphroditism is found in *Phillyrea angustifolia* (Saumitou-Laprade et al. 2010), though for most androdioecious species, the scenario is believed to be different (Pannell et al. 2014). In *Mercurialis annua*, one of the best-studied androdioecious systems, it seems that androdioecy arose during cycles of evolution between dioecy and monoecy, and is an intermediate in populations evolving back to dioecy (Fig. 1D, E).

Reversions from dioecy, as shown in Fig. 1C and D, are probably much more frequent than we used to think and these could explain why dioecy is so rare in angiosperms (Käfer et al. 2017). Dioecy would be very easy to evolve (hence the numerous independent evolution of dioecy in angiosperms) but also easy to lose, in particular when population density drops and mates are difficult to find (Käfer et al. 2017).

The Genetics of the Transition to Dioecy

Only the genetics of the gynodioecy-dioecy pathway has been modeled. In the most popular model, both male-sterility and female-sterility mutations are nuclear (Charlesworth and Charlesworth 1978). If the male-fertility and female-fertility loci where these mutations appear are on the same chromosome, then selection will suppress recombination between these loci, which will create proto-sex chromosomes (Fig. 2a). A recessive male-sterility mutation and a dominant female-sterility mutation would typically create an XY system (Charlesworth and Charlesworth 1978), and once such a system is established, the sex chromosomes may then differentiate over time (Muyle et al. 2017).

Another possibility for the generation of sex chromosomes is Cytoplasmic Male Sterility (= CMS), which is well known in plants. Females carry a male-sterility mutation in the mitochondrial genome, the CMS mutation, and hermaphrodites

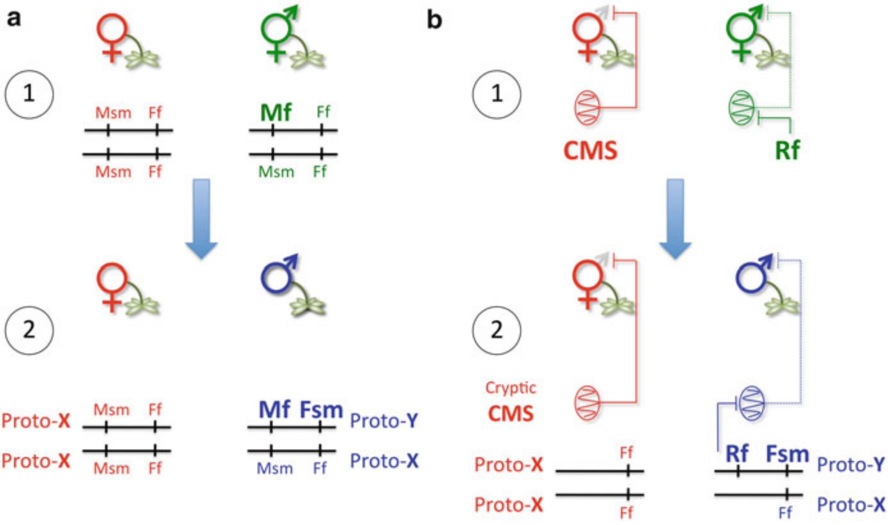

Fig. 2 The genetics of the evolution to dioecy from gynodioecy. The dominant model involves two nuclear mutations, which if on the same autosome will evolve into sex chromosomes (**a**). *Mf* male fertility gene (dominant), *Ff* female fertility gene (recessive), *Msm* male sterility mutation (recessive), *Fsm* female sterility mutation (dominant). Proto-X and proto-Y = nascent sex chromosomes. (**b**) An alternative model involves nucleo-cytoplasmic mutations, with gynodioecy being determined by a cytoplasmic-male-sterility factor (*CMS*) and a nuclear restorer of male fertility gene (*Rf*) system, in which case dioecy is determined by cryptic CMS, fixed in the population. If the Rf gene and a female fertility gene are on the same autosome, a proto-Y chromosome comprising the Rf gene (dominant) and a female sterility mutation (Fsm, dominant) may evolve

either have a male-fertile cytotype or carry a nuclear restorer-of-fertility (= Rf) gene that counteracts the CMS factor. It has been shown through modeling and computer simulations that the evolution of dioecy is less restrictive in nucleo-cytoplasmic than in nuclear gynodioecy (Schultz 1994; Maurice et al. 1994). CMS-Rf systems are engaged in evolutionary arms races. CMS-Rf systems arise from conflicts over what is the optimal reproductive system for mitochondrial genes versus nuclear genes. Mitochondrial genes are transmitted only through females, and a CMS mutation will clearly increase their transmission; females will spread in the population. Nuclear genes are transmitted through both males and females, and once CMS has evolved, there will be a strong selective pressure for Rf genes to evolve, increasing the frequency of the hermaphrodites in the population. Gynodioecy will be maintained through episodic invasions of a new CMS mutation followed by evolution of a new Rf gene and so on or through balanced polymorphism due to Rf genes having some costs that prevent their fixation (Delph et al. 2007). These dynamics can occasionally generate peaks of female frequency in the population, an ideal situation for males to spread, and simulations have shown that the evolution of dioecy from nucleo-cytoplasmic gynodioecy is, in fact, easier than from purely nuclear gynodioecy (Schultz 1994; Maurice et al. 1994).

This scenario too could create sex chromosomes (Fig. 2b). The genetics of the dioecy is expected to be different in the fully nuclear model (Fig. 2a) and in the nucleo-cytoplasmic model (Fig. 2b). If a dioecious species with an XY system evolved from a nucleo-cytoplasmic gynodioecious precursor, a CMS cytotype should be present in the dioecious species, in both sexes. CMS would generate the females; the males would be determined by a Y chromosome including an Rf gene (counteracting CMS) and a female-sterility gene (Fig. 2b). The CMS would be cryptic as the sex ratio would be balanced (not female-biased as in gynodioecy). However, some simulations suggest that dioecy might be unstable when arising from a nucleo-cytoplasmic context; males may disappear when a new unrestored CMS cytotype appears, if some inconstant males/hermaphrodites (still producing seeds) are present when this happens (Schultz 1994). This is why the evolution of dioecy through nucleo-cytoplasmic gynodioecy is usually considered less likely on theoretical ground (Charlesworth 1999), although empirical tests of these models are needed to tell how restricted/widespread they are.

Empirical Data on Dioecious Plants

Evolution of Dioecy Through Gynodioecy

Available data suggest that gynodioecy and dioecy are found associated in the same genus more than expected by chance in angiosperms (Dufay et al. 2014). Some case-studies have provided clear phylogenetic evidence that dioecy can evolve through gynodioecy, as in the *Silene* genus, for example (see below). The expected reallocation from female to male functions in hermaphrodites co-occurring with females has been documented in several subdioecious (anatomically cosexual, but functionally male or female) and gynodioecious species and suggests an ongoing transition to dioecy (Spigler and Ashman 2012).

There are two types of gynodioecy: nuclear and nucleo-cytoplasmic, on which the current models are based (see previous section and Fig. 2 and Table 3). Despite the fully nuclear model being the dominant model in the literature on dioecy, this type of gynodioecy is rare; nucleo-cytoplasmic gynodioecy is much more common (Delph et al. 2007). Species showing nucleo-cytoplasmic gynodioecy typically have a CMS-Rf system (Touzet and Meyer 2014).

Silene latifolia, a dioecious plant, is often presented as the typical example of the evolution of dioecy through gynodioecy (Fig. 3). It has indeed several gynodioecious relatives and dioecy has probably followed the gynodioecy pathway in the *Silene* genus (Desfeux et al. 1996). *Silene latifolia* has a X/Y chromosome pair, which is probably ~5 million years old (Rautenberg et al. 2010). These sex chromosomes are very large (X: 400 Mb, Y: 550 Mb) and still not fully sequenced (Papadopulos et al. 2015). Although sex-determining genes are still unknown in *S. latifolia*, three sex-determining regions of a few Mb have been identified on the Y chromosome using a mutant collection (Fig. 3c, Zluvova et al. 2007), among which

Table 3 Sex determination mechanisms in dioecy and in gynodioecious and monoecious intermediates

Sexual systems	Sex determination	Genes
Gynodioecy	Mostly nucleo-cytoplasmic	CMS: Chimeric mitochondrial genes Rf: Pentatricopeptide-repeat (PPR) family proteins
Monoecy	Phytohormonal	
	Cucurbits: Ethylene pathway	Cucurbits: Genes of the ethylene pathway (2 enzymes, 1 transcription factor)
	Maize: Gibberellic acid pathway	Maize: *Silkless* gene *sk1, Tasselseed* family genes *TS1* and *TS2, tasselseed4* microRNA
Dioecy	Sex chromosomes (XY, ZW)[a]	Persimmon: OGI small RNA/MeGI Asparagus: MYB transcription factor

See text for references

[a] In XY systems, the males are heterogametic (have a pair of nonidentical sex chromosomes), while in ZW systems, it the females are heterogametic

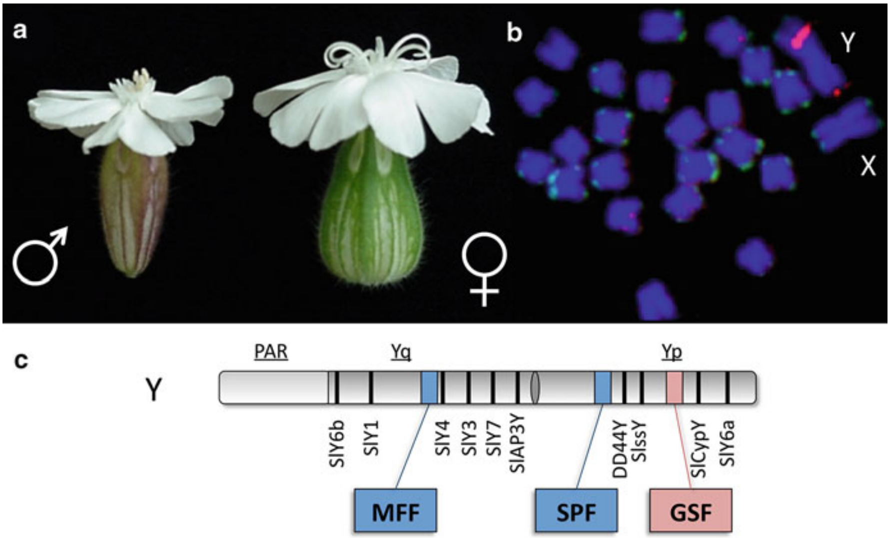

Fig. 3 The dioecious plant *Silene latifolia*. (**a**) Flowers from male and female individuals. (**b**) A karyotype of a *S. latifolia* male individual (from Hobza et al. 2007). (**c**) The *S. latifolia* Y chromosome and the three sex-determining regions identified using mutants. These regions comprise two male-fertility regions: *MFF* male fertility factor, *SPF* stamen promoting factor, and one female-sterility region: *GSF* gynoecium sterility factor. Well-known Y-linked genes are indicated. *PAR* pseudoautosomal region (X-Y recombining region)

are a female-sterility region and a male-fertility region, as would be expected from theory (Fig. 2).

The gynodioecious *Silene* species are of the nucleo-cytoplasmic type (e.g., *Silene vulgaris*, *Silene nutans, Silene acaulis*), but their CMS and Rf genes have not yet been identified (Bernasconi et al. 2009). There are several indirect lines of evidence indicating cryptic CMS in *S. latifolia,* including biased sex ratios in crosses with close dioecious relatives (Taylor 1994). One of the sex-determining regions (MFF, see Fig. 3c), when deleted, gives a phenotype that closely resembles that of females in gynodioecious close relatives; they undergo anomalous anther development, with problems of cell proliferation in the tapetum (a cell layer feeding the developing pollen grains), which results in pollen-less anthers (Zluvova et al. 2007). Also, a cross of a *S. latifolia* female and a *Silene viscosa* hermaphrodite resulted in a 100%-female F1 (Zluvova et al. 2005), which can be easily explained by CMS transmitted by the *S. latifolia* mother for which the *S. viscosa* father did not have the corresponding Rf.

Fragaria (strawberry) is another genus in which dioecy probably evolved through the gynodioecy-dioecy pathway. The gynodioecious *Fragaria vesca* subsp. bracteata has a CMS-Rf system involving several Rf and Rf inhibiting loci (Ashman et al. 2015). Interestingly, one of the chromosomes harboring these loci has evolved into ZW chromosomes in the dioecious *F. virginiana* and *F. chiloensis*, which raises the possibility that these Rf loci are involved in sex determination in those dioecious relatives.

Evolution of Dioecy Through Monoecy

Phylogenetic evidence that monoecy and dioecy are associated is very strong as shown by angiosperm-wide, plant-family-focused and case studies (Renner and Ricklefs 1995; Barrett 2002; Renner 2014; Käfer et al. 2017). It is quite clear that the model developed for the gynodioecy-dioecy pathway does not apply to the monoecy-dioecy pathway (Golenberg and West 2013). In monoecious plants, male and female flowers do not develop randomly on the plant, instead the sex of flowers depends on their location. The sexual identity of flowers appears to result from gene networks that are responsive to information on the position of the flower in the plant, which are mediated by hormones. This is a starting point for evolving master sex-determining genes that is very different from that in the gynodioecy-dioecy pathway (Golenberg and West 2013). The genetics of monoecy is starting to be deciphered, in cucurbits and in grasses, and both groups also include dioecious species.

In melon (*Cucumis melo*) and cucumber (*Cucumis sativus*), three key genes, the *monoecious* (*M*), *androecious* (*A*) and *gynoecious* (*G*) genes, interact to determine the sexual identity of a given flower with respect to its position on the plant. Combinations of mutations of these genes produce female individuals, male individuals, and hermaphroditic individuals with bisexual flowers, and it was possible to create an artificially dioecious population in melon using these mutations (Boualem et al. 2015). The *M* gene inhibits the development of stamens (male organs), which results in a female flower. The *M* gene is inhibited by the *G* gene, which also suppresses the development of carpel (female organs). When expressed, the *G* gene turns a flower into a male one. The *A* gene inhibits the *G* gene and thus produces female flowers. Its expression is probably under the influence of the information on the position of the flower in the plant. If the *M* gene is nonfunctional (and *A* and *G* are functional), hermaphroditic flowers are produced. Both *A* and *M* genes encode for enzymes (*ACS-11* and *ACS-7*, respectively) which are involved in ethylene biosynthesis, an important hormone that regulates many plant processes. The *G* gene encodes a C2H2 zinc finger transcription factor (*WIP1*). These three genes thus work coordinately to control the sexual phenotype of flowers in monoecious cucumis species (Boualem et al. 2015).

Maize (*Zea mays*) is monoecious with two types of unisexual inflorescences located, respectively, at the plant apex (tassels = male inflorescences) and leaf axils (ears = female inflorescences). Many mutants affecting the sexual phenotype of flowers have been identified, contributing to the choice of this species for studies of the molecular mechanisms of flower development in grasses (Li and Liu 2017). The *Silkless* gene *sk1* was identified as the master gene for pistil identity. This gene interacts with the *Tasselseed* family genes *TS1* and *TS2,* which are responsible for the arrest of pistil development. On the other hand, male flower development is controlled by the *tasselseed4* microRNA, a member of the miR172 family of microRNAs (miRNAs), which regulates the APETALA2 family of transcription factors and inhibits pistil development. A genetic hierarchy is beginning to emerge from the analysis of single and double mutants of these genes.

The Master Sex-Determining Genes in Dioecious Plants

The only sex determination mechanism known thus far in flowering plants is based on sex chromosomes, which have been reported in a minority of dioecious plants (<1%, Tables 1 and 3). For the vast majority of dioecious plants, the basis of sex determination, either genetic or environmental, is unknown. Both XY systems, in which males are heterogametic (have different sex chromosomes), and ZW systems, in which females are heterogametic, have been found, although the latter seem to be rarer than the former (Ming et al. 2011). Of the ~40 sex chromosome systems currently described, ~20 are heteromorphic (i.e., sex chromosomes are clearly distinguishable using cytogenetics). Examples of heterogametic systems are *S. latifolia* and *Coccinia grandis*, a dioecious cucurbit with an XY system, which happens to have the most heteromorphic sex chromosomes known in plants (Sousa et al. 2013). In these two species, the Y is larger than the X, a situation that has not been encountered in any animal species thus far. The remaining ~20 known plant sex chromosome systems are homomorphic (i.e., the sex chromosomes are indistinguishable using cytogenetics). One example is *Carica papaya* (papaya), the only plant species for which the sex chromosomes (XY as it happens) are fully sequenced and assembled (Wang et al. 2012). Interestingly, in this species a modified Yh determines XYh hermaphrodites in addition to XY males and XX females, and this Yh was probably selected during the domestication process of papaya (VanBuren et al. 2015). It is possible that many dioecious plants have yet unidentified homomorphic sex chromosomes, which are more difficult to detect, as in many cases dioecy has evolved recently and young sex chromosomes tend to be homomorphic (Ming et al. 2011; Muyle et al. 2017). Importantly, among the ~40 plant species with known sex chromosomes, in almost all cases the master sex determining genes have not yet been identified.

The only dioecious plant species for which master sex determining genes have been identified are persimmons (Akagi et al. 2014) and possibly asparagus (Murase et al. 2017) (Table 3). In persimmons (*Diospyros lotus*), sex determination is driven by a Y-specific sex determinant, *OGI* (Japanese for "male tree"), which encodes a small RNA, and its target, the autosomal *MeGI* gene (Japanese for "female tree"), a homeodomain transcription factor that regulates anther fertility. Transformed tobacco and *Arabidopsis thaliana* plants clearly suggest that *MeGI* is a feminizing gene and *OGI* acts by suppressing *MeGI* expression. In XY individuals, *OGI* is present and inhibits the expression of *MeGI*, which results in anther development and the arrest of carpel development, and the individuals are consequently male. In XX individuals, *MeGI* is expressed, carpel development proceeds, and anther development is repressed, resulting in female individuals. The *OGi-MeGI* system seems to be conserved in several *Diospyros* species and could be >50 million years old.

In asparagus (*Asparagus officinalis*), a plant with X and Y chromosomes, sex determination is controlled by a single locus, the *Mating* (M) locus. Transcriptome and RT-PCR analysis showed that a *myeloblastosis-like* (*MYB*) gene, *Male Specific Expression 1* (*MSE1*), is specifically expressed in males during early anther development and exhibits tight linkage with the Y chromosome, as well as loss-of-function on the X chromosome (Murase et al. 2017). Knockout of *MSE1* orthologue in *A. thaliana* produces female plants, as expected for a male-determining gene. *MSE1* has a male-specific expression pattern in several dioecious asparagus species, which suggests it plays a master male-determining role in these species too. It is also present in hermaphroditic species, which suggests that another master sex-determining gene, suppressing the female development program, is yet to be discovered in asparagus.

In asparagus, dioecy may have evolved through gynodioecy (Dufay et al. 2014) and current data are consistent with more than one master sex determining gene, which is expected from models for the genetics of the gynodioecy-dioecy pathway. In persimmons, in which dioecy may have evolved through the monoecy-dioecy pathway (Renner 2014), a Y-autosomal gene duet seems to be enough to determine male and female, and there seems to be a single master sex determining gene on the Y, as is the case for mammals, in which *Sry* is the master control gene. These considerations suggest that the genetics of the monoecy-dioecy and gynodioecy-dioecy pathways may be different (Renner 2016).

Conclusions and Perspectives

At this stage, it is of course difficult to draw general conclusions about sex determination in plants as genetic, molecular, and developmental data are too scarce. This scarcity is partly explained by the difficulty in studying sex chromosomes in plants. First, it is likely that many dioecious plants have young and homomorphic sex chromosomes, which cannot be identified simply looking at female/male karyotypes and requires more sophisticated methods. Second, sequencing sex chromosomes is notoriously difficult, even in animals (Muyle et al. 2017). However, new approaches to sequence sex chromosomes have been developed recently, and this could boost the field of plant sex determination (Muyle et al. 2017).

To understand better the gynodioecy-dioecy pathway, more master sex-determining genes of dioecious species that evolved from this pathway should be identified, in particular to understand whether fully nuclear or nucleo-cytoplasmic mutations have been more common. The monoecy-dioecy route is well established, although monoecy could also evolve from dioecy. More theoretical work is needed to understand the evolutionary forces underlying the monoecy-dioecy pathway. The genetics of this pathway is not clear either, but recent findings on genes controlling monoecy will surely give a boost to this field of research. Also, some dioecious plants might have evolved via minor pathways, i.e., androdioecy or heterodistyly. Study of these cases will certainly be needed to form a global picture of the evolution of dioecy and sex determination in plants.

Dioecy is rare in angiosperms, but this is not the case in other land plant lineages such as gymnosperms, mosses and liverworts. In haplo-diploid plants, males and females can be haploid and carry a third type of sex chromosomes called UV (Muyle et al. 2017). In order to gain an even wider picture of sex determination in plants, these groups must also be studied.

Cross-References

- ▶ Alternation of Generations in Plants and Algae

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