Original article

Running head: **Chromosome number evolution in the Cyperaceae**

Title: **Inferring hypothesis-based transitions in clade-specific models of chromosome number evolution in the Cyperaceae**

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Abstract

Large-scale changes in chromosome number have been associated with diversification shifts in many lineages of plants. For instance, several ancient rounds of polyploidization events have been inferred to promote genomic differentiation and/or isolation and, consequently, angiosperm diversification. Dysploidy, although less studied, has been suggested to play also an important role in angiosperm diversification. In this article, we aim to elucidate the role of chromosomal rearrangements on lineage diversification by analyzing a

new comprehensive sedge (Cyperaceae) phylogenetic tree. Mode and tempo of chromosome evolution were inferred to be homogeneous in rate and process across the complete phylogeny as the null hypothesis. In order to discern patterns of diversification shifts and chromosome number changes within the family tree, we tested clade-specific chromosome evolution models for several subtrees according to previously reported increments of diversification rates. Results show that alternative hypotheses of different clade-specific models of chromosome evolution are significantly supported against the null hypothesis of a model with no transition events along the phylogeny. This could suggest a link between diversification and changes in chromosome number evolution. Our methodological approach may allow identifying different patterns of chromosome evolution, as found for Cyperaceae, for other lineages at different evolutionary levels.

Key words

ChromEvol, chromosome evolution, Cyperaceae, diversification rates, holocentric chromosomes, phylogeny

1. Introduction

Chromosomal rearrangements are frequent in eukaryotes and are in many cases correlated with differentiation and speciation (Coghlan et al., 2005). These rearrangements can be produced by a sole mechanism or a combination of translocations, aneuploidy, dysploidy and polyploidy (whole genome duplication; WGD) (Coghlan et al., 2005). Whereas some of these events could produce changes in the genome structure and linkage of genes (Butlin, 2005), others could affect directly the gene content through either deletions or duplications of DNA (Coghlan et al., 2005). These events may promote speciation by

provoking changes in species fitness, adaptability to new habitats, reproductive isolation and/or shifts in recombination rates (Butlin, 2005; Coghlan *et al*., 2005; Coyne and Orr, 2004; Navarro and Barton, 2003a, 2003b; Otto and Whitton, 2000; Rieseberg, 2001; Soltis *et al*., 2009).

In angiosperms, the role of polyploidy and its consequences on speciation have been intensely studied, with a particular interest in ancient polyploid events in some of the most species-rich lineages (Debodt et al., 2005; Smith et al., 2018; Soltis et al., 2009; Soltis and Soltis, 2016). This has led to an understanding of polyploidization as a possible driver for lineage radiation (Comai, 2005; Hegarty and Hiscock, 2007, 2008; Levin, 1983; Otto, 2007; Otto and Whitton, 2000; Soltis and Soltis, 2016, 2000; Van de Peer, 2011). On the other hand, although dysploidy (translocations, fusions and fissions that lead to changes in chromosome number) is more frequent than polyploidy and especially aneuploidy (duplication or deletion of an entire chromosome) in angiosperms (Grant, 1981), its consequences in diversification have been largely unexamined (though, see Gitaí *et al*., 2014; Lee and Namai, 1993, 1992; Orellana *et al*., 2007; Vallès *et al*., 2012; Vickery, 1995; Weiss-Schneeweiss *et al*., 2009). Dysploidy has recently been suggested to not represent a dead end through evolutionary time (Escudero et al., 2014).

Probabilistic models have been recently formulated for chromosome. These models vary in their complexity, with the simplest ones calculating the rate of gains and losses of chromosomes and changes in ploidy level along a phylogeny. More complex models allow identifying linear dependency between the current number of chromosomes and the rate of increasing and decreasing chromosome numbers. More recently, Freyman and Höhna (2018) expanded ChromEvol functions (Glick and Mayrose, 2014; Mayrose et al., 2010) with the ChromoSSE package in revBayes (Höhna et al., 2014). This software allows not only

detecting shifts in the mode of chromosome evolution during anagenetic processes but also during cladogenesis, that can be associated with diversification rate shifts. Moreover, BiChroM type models (correlated rates of phenotype and chromosome evolution; Zenil-Ferguson *et al*. 2017, 2018) can be integrated with the classic ChromEvol models. However, none of these new approaches considers the possibility of more complex models of chromosome evolution, with different parameters throughout the phylogeny. Here, we expand these studies by applying different models of karyotypic evolution to different clades. This approach is crucial to identify changes in the mode of chromosomal evolution as innovations that may be related to shifts in diversification rates.

The cosmopolitan family of sedges (Cyperaceae, ca. 5500 species; Govaerts *et al*., 2017) is the tenth most species-rich angiosperm family. It has mainly diversified in the tropics, although genus *Carex* L., the most diversified genus of the family (ca. 2200 spp., 40% of species richness; Govaerts *et al*. 2017), and several other lineages are distributed mostly in temperate regions (Reznicek, 1990). Cyperaceae has the highest known chromosome number variation among all angiosperm families (2n=4–224; Roalson, 2008). Because of its high species richness and wide range of chromosome numbers, Cyperaceae constitutes a model taxon for incorporating studies of biodiversity with evolution and systematics (e.g. Hipp, 2007). This is especially true of the genus *Carex*, which alone displays a wide variation of chromosome number (2n=12–124; Hipp, 2007; Roalson, 2008) . Variation in the number of chromosomes and changes in the mode of evolution have been suggested as a possible driver of diversification in *Carex* (Escudero *et al*., 2012b, 2014). The huge continuous variation in chromosome number of this family is explained by the presence of holocentric chromosomes, which means that the kinetochoric activity is present along the chromosomes. By contrast, monocentric chromosomes have a clear primary constriction in which kinetochoric activity is

concentrated (Hipp et al., 2013; Melters et al., 2012; Mola and Papeschi, 2006). In lineages with holocentric chromosomes (see review in Márquez-Corro *et al*. 2017), fusions and fissions (termed symploidy and agmatoploidy, respectively; Escudero *et al*. 2014) are more common (Grant, 1981). This occurs even within species level, due to the characteristics of the kinetochoric plate (Hipp et al., 2013; Melters et al., 2012; Mola and Papeschi, 2006) that allows more or less constant C-values despite chromosome number variation (Escudero et al., 2014).

Four main shifts in diversification rate have been detected in Cyperaceae. Escudero *et al*. (2012b) found an increase in diversification rates in the non-*Siderostictae* clade (that comprises Core *Carex*, Caricoid *Carex* and *Carex* subgenus *Vignea*), which has been confirmed in a recent study by Spalink *et al*. (2016b). Escudero and Hipp (2013) used Hinchliff and Roalson's (2013) phylogeny to infer an additional shift in diversification rates in the clade including the tribes Scirpeae, Dulichieae, and Cariceae plus *Khaosokia caricoides* (SDC clade) and the tribes Fuireneae, Abildgaardieae, Eleocharideae, and Cypereae (FAEC clade). Spalink *et al*. (2016b) showed instead shifts in three different lineages inside the SDC+FAEC clade reported by Escudero and Hipp (2013). Thus, in addition to the shift in the non-*Siderostictae* clade (as in Escudero *et al*. 2012b), Spalink *et al*. (2016b) also found a shift in the FAEC clade and in the represented taxa of the C_4 photosynthetic pathway *Cyperus* within Cypereae 2 clade (within FAEC).

Different modes of chromosomal evolution are present in Cyperaceae. For example, *Carex* karyotype evolves mainly via agmatoploidy and symploidy (Heilborn 1924; Davies 1956), whereas polyploidy is more common in the rest of sedges (Escudero et al., 2012b). Thus, this hyperdiverse family and its wide range of karyotypic variation constitute an ideal lineage to study shifts in chromosome evolution and how they could be related with changes

in diversification rates. We hypothesize that some shifts in lineage diversification could be related, at least in part, with changes in the mode of chromosome evolution. This could be explained by the fact that chromosome evolution may lead to different mechanisms of adaptation (e.g. adaptive mutation perpetuated by fission events) and/or reproductive isolation that could drive differentiation and speciation (Butlin, 2005; Coghlan *et al*., 2005; Coyne and Orr, 2004; Navarro and Barton, 2003a, 2003b; Otto and Whitton, 2000; Rieseberg, 2001; Soltis *et al*., 2009). However, there is still possible that diversification rates shifts within the family are related with others characteristic rather than mode of chromosome number evolution, so further studies must be carried out.

The aims of this study are (i) to elucidate the role of chromosome evolution in the diversification of the sedge family using probabilistic models, and (ii) to evaluate the utility of nested models for studying chromosome evolution in diverse lineages. We hypothesize that transitions in the mode of chromosome evolution are closely preceded or followed by a shift in diversification rates in Cyperaceae. Our null hypothesis, by contrast, is that chromosome numbers change in the family at a constant rate, regardless of the diversification rate of independent clades.

2. Materials and Methods

2.1. Family Tree and Chromosome Counts

A new comprehensive phylogeny of Cyperaceae was created from NCBI GenBank database sequences of previous studies (e.g. Hinchliff and Roalson, 2013; Spalink *et al*., 2016b; Jiménez-Mejías *et al*. 2016a). This analysis included 1058 species out of the ca. 5500 circumscribed to Cyperaceae (Govaerts *et al*. 2017), and was based on a supermatrix alignment of the nuclear ribosomal genes ETS and ITS, the plastid genes *mat*K, *ndh*F, *rbc*L,

*ycf*6, and the chloroplast spacer region *trn*C-*ycf*6. Though we used the GTRCAT model in RAxML (Stamatakis, 2006) for computational purposes, the model parameters were individually calculated for five different partitions identified using PartitionFinder v2 (Lanfear *et al*., 2016). We converted the resulting maximum likelihood phylogeny to ultrametric using treePL (Smith and O'Meara 2012; see Fig.1, Appendix A). A total of eleven calibrations were placed on key nodes throughout the phylogeny based on fossil evidence (Jiménez-Mejías *et al*. 2016b; Spalink *et al*. 2016a, 2016b; Appendix B).

Species haploid numbers were collected from online databases IPCN (Index to Plant Chromosome Numbers, Goldblatt and Johnson 2017), CCDB (Chromosome Counts Database, Rice *et al*. 2015), and some chromosome number reports (see Appendix B). Chromosomes counts were downloaded for a total of 825 taxa that were included in the phylogeny (Appendix B).

Due to the holocentric characteristic of sedge chromosomes, counts can vary within single species (Roalson, 2008). Because we aimed to detect shifts in chromosome number evolution along the family tree, we assigned to the tips the most frequent number in the species dominated by symploidy/agmatoploidy series, and the record with the lowest chromosome number for species presenting polyploidy (see Appendix B).

2.2. Selecting the Best Scenario of Chromosome Evolution

We used ChromEvol v.2.0 (Glick and Mayrose, 2014; Mayrose et al., 2010) to model the mode of chromosome evolution. This software determines the likelihood of a model to explain the given data along the phylogeny, based on the combination of two or more of the following parameters: (i) gain or (ii) loss of a single chromosome, (iii) polyploidization, (iv) demi-polyploidization (half increment of the chromosome number) and (v) incremental

changes to the base number with regard to a rate of multiplication that is different from a regular duplication. Two additional parameters detect linear dependency between the current haploid number and the rate of (vi) gain and (vii) loss of chromosomes.

Shifts in diversification have been previously detected in four main nodes (1-4; Fig. 2) of Cyperaceae (SDC+FAEC, FAEC, non-*Siderostictae Carex* and C_4 *Cyperus*; Escudero *et al*., 2012b; Escudero and Hipp, 2013; Spalink *et al*., 2016b), so analyses were conducted independently not only for the complete phylogeny but also for the same phylogeny split in several combinations of subtrees (see below). These included clades that exhibit diversification rates shifts, the background phylogeny of these clades (i.e. pruned tree without the corresponding clade), and further combinations of clades and backgrounds. A similar methodology, but not with models of chromosome number evolution, has been previously used to infer transitions in continuous character evolution using Brownian and Ornstein-Uhlenbeck models (see Escudero *et al*., 2012a, 2010; Hipp, 2007; O'Meara *et al*., 2006). Specifically, we used the censored approach described by O'Meara *et al*. (2006). This approach breaks up the original tree in several subtress and the branches that connect the subtrees are excluded from the analyses. The main advantage of this approach is that assumptions are not made about when and how the trait shift occurs in the missing branch. We developed models ranging from the simplest (one model) to most complex (five models) scenarios, identifying the models that best fit the data by calculating the Akaike information criterion score with ChromEvol (AIC, Mayrose *et al*. 2010). In order to compare the simplest (one model) with the more complex scenarios (two to five models), the branches connecting the subtrees were removed in both the single model and two to five model cases. AIC weights (Wagenmakers and Farrell, 2004) were calculated and summed to infer the importance weights of a transition occurring on each specific clade.

In our specific study case, we defined four main clades (where shift in diversification rates were previously detected): (i) clade 1 is FAEC clade; (ii) clade 2 corresponds to non-*Siderostictae Carex* clade; (iii) clade 3 is C_4 *Cyperus*; and clade 4 conforms SDC+FAEC clade. Our chromosome modeling analyses were performed in up to five different subtrees: (i) subtree 1 is clade 1 after excluding clade 3; (ii) subtree 2 corresponds to clade 2; (iii) subtree 3 conforms clade 3; (iv) subtree 4 corresponds to clade 4 after excluding subtrees 1, 2, and 3; and (v) subtree 5 corresponds to the remaining phylogeny after excluding clade 4 (see Fig. 2).

3. Results

The best-fitting null model for the complete tree was Linear_Rate_Demi_Est, with an AIC score of 5501.84 (see Table 1). The Linear_Rate_Demi_Est model implies a constant rate of incremental/decremental change in chromosome number, polyploidy, and demi-polyploidy, and a linear relationship between the rate of incremental/decremental change and chromosome number (Mayrose et al., 2010).

The analysis of separate subtrees showed a significant decrease in AIC scores (see Table 1). In the best-fitting model ($\Delta AIC = -207.56$), a transition in the model of karyotype evolution was observed in each of the analyzed subtrees except for the subtree 4 (clade 4, SDC+FAEC; Fig. 2, Appendices C-D). In this case, subtree 4 and 5 displayed the same model, a Base_Num model, with 0.07 fission events/Myr, 0.70 fusion events/Myr and a rate of base-number multiplication of $0.2e^{-3}$ events/Myr with a base haploid number x = 13. Further transitions are inferred for subtrees 1 (FAEC clade excluding subtree 3), 2 (non-*Siderostictae Carex*) and 3 (C_4 *Cyperus* lineage). Because these transitions include linear rates parameters, we specify the events per chromosome number and million years (hereafter iMyr) and the range of fission and fusion rates using the minimum and maximum chromosome

number in each subtree (see Appendix E).

TABLE 1. Akaike information criterion (AIC) values, difference (ΔAIC) from the null scenario (no transitions) and AIC weights for each scenario. Importance weights for no transition scenario and for each clade appear together with brief comments on the right side of the table.

Transition scenarios†	AIC	ΔAIC	AIC weight	Conclusions
Null	5501.84	0.00	$6.41e^{-46}$	No transition events
1	5382.08	−119.76	$6.51e^{-20}$	A single transition event, either in FAEC clade (1), non-*Siderostictae Carex* (2), C_4 *Cyperus* (3) or SDC+FAEC clade (4)
2	5369.57	−132.27	$3.38e^{-17}$	
3	5420.74	−81.11	$2.62e^{-28}$	
4	5467.23	−34.61	$2.10e^{-38}$	
1,2	5330.73	−171.11	$9.20e^{-09}$	Scenarios of two transition events
1,3	5345.63	−156.21	$5.34e^{-12}$	
1,4	5369.09	−132.75	$4.31e^{-17}$	
2,3	5311.06	−190.78	$1.72e^{-04}$	
2,4	5377.40	−124.44	$6.75e^{-19}$	
3,4	5387.07	−114.77	$5.36e^{-21}$	
1,2,3	***5294.28***	***−207.56***	***$7.55e^{-01}$***	Scenarios of three transition events. The best scenario suggest a sole mode of chromosome number evolution through sedges, with exception of clades 1, 2 and 3
1,2,4	5333.07	−168.77	$2.84e^{-09}$	
1,3,4	5332.64	−169.20	$3.53e^{-09}$	
2,3,4	5302.58	−199.26	$1.19e^{-02}$	
1,2,3,4	5296.63	−205.21	$2.33e^{-01}$	Most complex scenario, Four transition events. This case is not much worse than the scenario 1,2,3 (ΔAIC=2.35), and would support transition events in lineages 1, 2, 3 and 4

The best scoring scenario is indicated with bold italics.

†Each number corresponds to a transition in the mode of chromosome evolution for the respective clade.

On the subtree 1 (FAEC clade excluding subtree 3), the mode of evolution changed to the Linear_Rate_Demi model, with negligible constant rates of fusion or fission (0

events/Myr), 0.03 duplication events/Myr (either demi-polyploidization or WGD), and a linear rate of $8.2e^{-3}$ fission events/iMyr and $5.2e^{-3}$ losses events/iMyr (linear and net rates of 0.02–0.45 fission events/Myr and 0.02–0.29 fusion events/Myr). The C_4 *Cyperus* lineage retained the Linear_Rate_Demi_Est model, with 13.68 fission events/Myr, 9.98 fusion events/Myr, 0.22 duplication events/Myr, 1.59 demi-polyploid events/Myr, and a rate of -0.15 fission events/iMyr and 0.75 fusion events/iMyr (linear rate of -0.90–-12.30 fission events/Myr and 4.50–61.50 fusion events/Myr, and net rate of 12.78–1.38 fission events/Myr and 14.48–71.48 fusion events/Myr). Finally, the non-*Siderostictae Carex* best model was Linear_Rate_Demi_Est, with a constant rate of 2.50 fission events/Myr, 2.13 fusion events/Myr, $2.7e^{-3}$ duplications events/Myr, 0.01 demi-polyploidy events/Myr, and a linear rate of 0.02 fission events/iMyr and 0.07 fusion events/iMyr (linear rate of 0.14–1.30 fission events/iMyr and 0.49–4.55 fusion events/iMyr, and net rate of 2.64–3.80 fission events/Myr and 2.62–6.68 fusion events/Myr).

The results of the remaining AIC scores of model selection and combination are included in Appendix D, with the best-fitting models depicted in Figure 2. Analysis output files with all the inferred chromosome rate transitions of every model studied are available online at github.com/jimarcor/ChromEvolCyp.

4. Discussion

4.1. Chromosome Evolution Modes on Cyperaceae

The sedge phylogeny presented here is the most comprehensive family tree published to date, with more than twice as many taxa as previous analyses (Hinchliff and Roalson, 2013; Spalink et al., 2016b). This phylogeny allows studying evolutionary processes more thoroughly in Cyperaceae. We also use a new approach for inferring modes of chromosomal

evolution across this phylogeny. By separately analyzing the full tree and subtrees, we have clarified our understanding of chromosome evolution along the Cyperaceae phylogeny.

The null hypothesis of a single mode of chromosome evolution on the sedges' phylogeny is consistently rejected by the analyses (Table 1). This approach appears to be useful for studying transitions in chromosome evolution at higher taxonomic levels and could be used at finer evolutionary levels as well (e.g., analyzing groups of close species). Our results are particularly relevant in the study of clades containing species with holocentric chromosomes, whose labile karyotypes could exhibit heterogeneous modes of evolution.

The best-fitting model of karyological evolution in Cyperaceae suggests multiple model transitions throughout the family phylogeny. These include distinct modes of evolution in the C_4 *Cyperus* clade, in non-*Siderostictae Carex* clade, and the FAEC clade excluding C_4 *Cyperus*). We found no support for a distinct mode of chromosome evolution at the origin of the SDC+FAEC clade.

Chromosome numbers seem to have evolved primarily by fusion (Fig. 2, Appendices D-F) until diversification of the non-*Siderostictae Carex* and FAEC clades. The shift at the non-*Siderostictae Carex* (Table 1-2) is mainly related to a massive increase in the rate of chromosome fissions and fusions. This clade also includes the former genera *Kobresia*, *Schoenoxiphium*, *Uncinia* and *Cymophyllus* (Global Carex Group, 2015), in which no or few genome duplications have been inferred (Davies, 1956; Hipp et al., 2009; Hoshino, 1981; Wahl, 1940). Accordingly, non-*Siderostictae Carex* shows here the lowest polyploidy rates of all subtrees with the exception of the remaining SDC clade and early divergent lineages (from Rhynchosporeae to Mapania clades, see Fig. 2) that show the lowest (in the transition non-*Siderostictae Carex* a soft increase of polyploidy rates was detected). Models regarding this clade imply the evolution of chromosomes by events of agmatoploidy (fission) and symploidy

(fusion). This phenomenon has been suggested to occur in *Carex* (Davies, 1956; Hipp et al., 2009; Hoshino, 1981; Wahl, 1940), but it has never been statistically tested at the genus level. *Carex* along constitutes ca. 40% of the Cyperaceae species of the sedges family (Govaerts et al., 2017). Therefore, understanding whether diversification rate shifts are related to karyotypic change is key to comprehending chromosome evolution as the result, trigger, or part of the speciation process and whether this change is mediated by intrinsic factors (e.g. linkage disequilibrium), extrinsic factors (e.g. reinforcing ecological speciation), or both.

TABLE 2. Importance weights for each clade and weight for the null scenario of no transitions. In bold are those sums with the highest probability of a transition to occur.

Transition scenarios by clades	**AIC weight sum**
Null	$6.41e^{-46}$
1	**0.988**
2	**1.000**
3	**1.000**
4	0.245

A second transition in mode of karyological evolution corresponds to the FAEC clade excluding C_4 *Cyperus* (Table 1-2). This shift in the mode of chromosome evolution is dominated by a decrease of the rate of fusion events, and a slightly increase of fission events as chromosome number grows (Fig. 2, Appendices D-F). Chromosome duplication seems to have no large effect, and thus, karyotypes are likely to remain largely stable within this clade, particularly in lineages such as *Fimbristylis* and *Eleocharis* (though, some instances of duplication may be evident in *Schoenoplectus* and *Schoenoplectiella*). This pattern could suggest the possibility of constraints against chromosome number evolution in this clade, although the selection process that would cause such results remains obscure.

The high rates of fusions, fissions, demi-polyploidization and duplications in the C_4

Cyperus clade contrast remarkably with the karyotype stability of the FAEC clade (Fig. 2, Appendices D-F). Lowest haploid numbers in this clade correspond to a polyploid series; *Cyperus brevifolius* (=*Kyllinga brevifolia*), for instance, also presents high chromosome number ranges due to duplication (n = 9–86; Roalson, 2008). Polyploidy has also been suggested previously for *Cyperus esculentus* (Arias et al., 2011; De Castro et al., 2015), and has been reported as frequent throughout the clade (see Roalson, 2008). Though neo-polyploids generally do not feature higher diversification rates (Mayrose et al., 2011), this *Cyperus* lineage (ca. 760 species; Larridon *et al*., 2013) would constitute a counterexample of that trend. Nevertheless, although high rates of fission and fusion have been detected, these parameters could be the byproduct of a biased chromosome dataset. Since there are few species represented in this clade and chromosome data depends on the current published reports, high fusion and fission rates can be due to the inability to detect further duplications and demi-polyploidization. In this case, lineage diversification could suggest a link with the mode of chromosome evolution towards an evolutionary scenario dominated by incremental changes to ploidy. Alternatively, this increase in the diversification rate could be related with other innovative mechanisms of the lineage, such as the evolution of the C_4 photosynthetic pathway (Larridon et al., 2013). Therefore, genome duplications and shifts in the photosynthetic pathway could have acted in concert.

Although a clear correspondence between chromosome number transitions and diversification rates shifts cannot be assured in this study, strong evidence is found in shifts in chromosome evolution modes through the family tree that might suggest a link. Nevertheless, as exemplified by the *Cyperus* lineage, this relationship could also be related to other evolutionary process such as the development of C_4 photosynthetic pathway. Further research is required to accurately test the relationship between chromosome model evolution

transitions and shifts in diversification rates. The results of these studies could provide new insight into the macroevolutionary processes that explain these patterns.

4.2. Final Remarks

Summing up, this study proposes (i) the use of single model vs. complex models (i.e. two to five different models) of chromosome evolution as a feasible approach to the study of chromosome evolution; (ii) that, for Cyperaceae, the statistical support for a complex transition scenario was much higher than a simple model of chromosome number evolution; (iii) a clear pattern of high rate of duplications, and possibly fusions and fissions, as the main mean of chromosome evolution for, at least, part of the lineage of C_4 *Cyperus* species, (iv) very high rate of agmatoploidy and symploidy in genus *Carex* (except *Siderostictae* clade), (v) karyotype stability (low rates of chromosome evolution) through most FAEC clade lineages.

Figure captions

FIGURE 1. Summarize infographic of the methodology followed in the study.

FIGURE 2. Best-fitting scenarios of chromosome evolution for the Cyperaceae phylogeny. Numbered clades correspond to those in which a shift in diversification rate have been detected (1, FAEC clade; 2, *Carex* lineage; 3, C_4 *Cyperus* lineage; 4, SDC+FAEC clade). Akaike information criterion (AIC) of the best-fitting scenario (AIC_1) appear next to the phylogeny, compared (ΔAIC) to the null hypothesis AIC score (AIC_0).

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Supporting Information:

Appendix A: Phylogenetic tree in nexus format
Appendix B: Calibrations for Cyperaceae phylogeny and list of haploid chromosome number used in the analysis
Appendix C: AIC scores for model selection
Appendix D: AIC scores for scenario comparison
Appendix E: Best model parameters
Appendix F: Family tree with chromosome data

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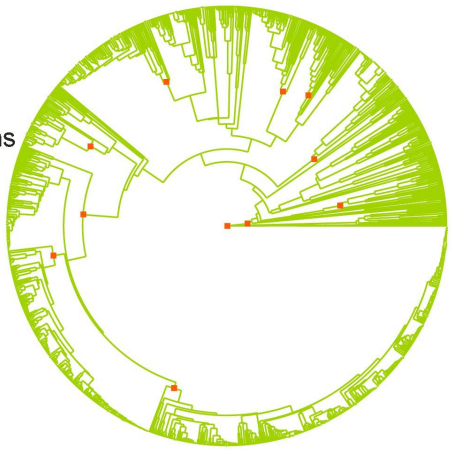
1. PHYLOGENY CONSTRUCTION

11calibrations

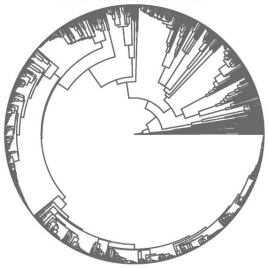
1058 taxa of sedges



2. MODELS OF EVOLUTION

Pruned down to 825 taxa with chromosome data

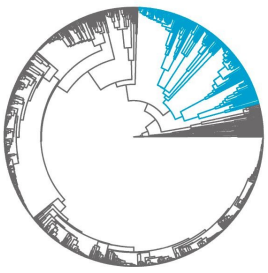
No transition



Compare the null hypothesis against complex scenarios of two to five transition events

vs.

1



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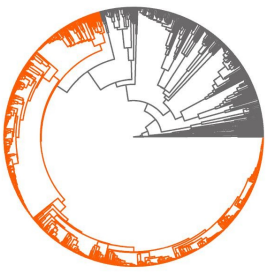

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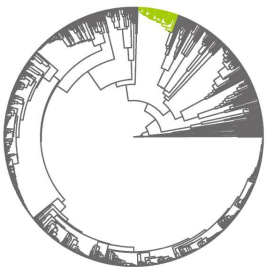

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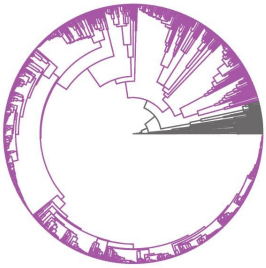

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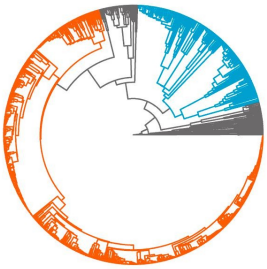

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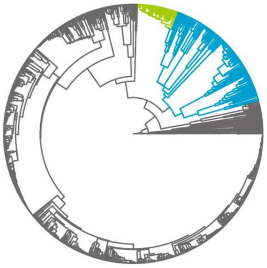

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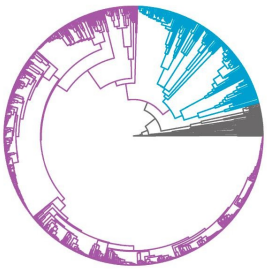
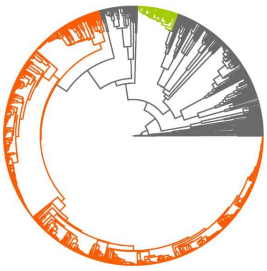


2, 3



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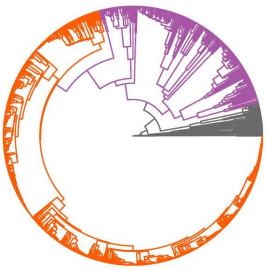

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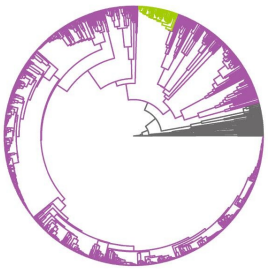
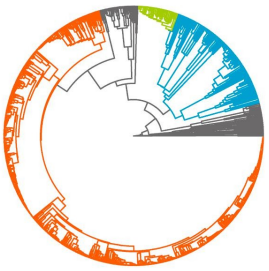


1, 2, 3



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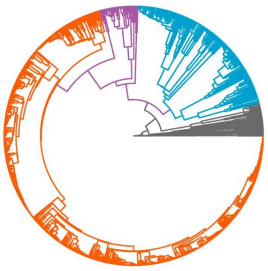

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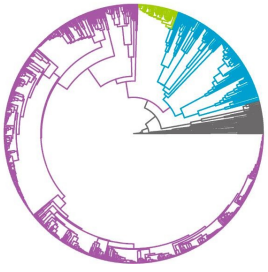

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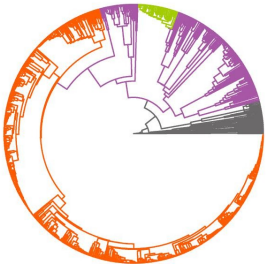
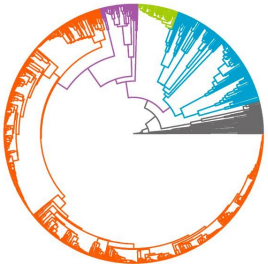
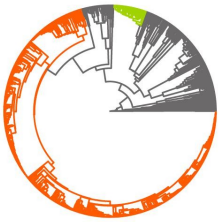


1, 2, 3, 4

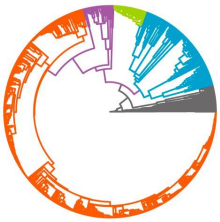


3. SCENARIO COMPARISON

AIC = **5501.84***



AIC = 3550.03 + 343.06 + 1417.97* = **5311.06**

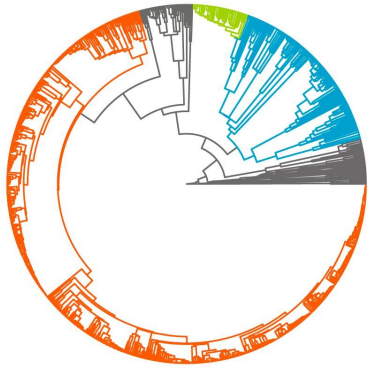


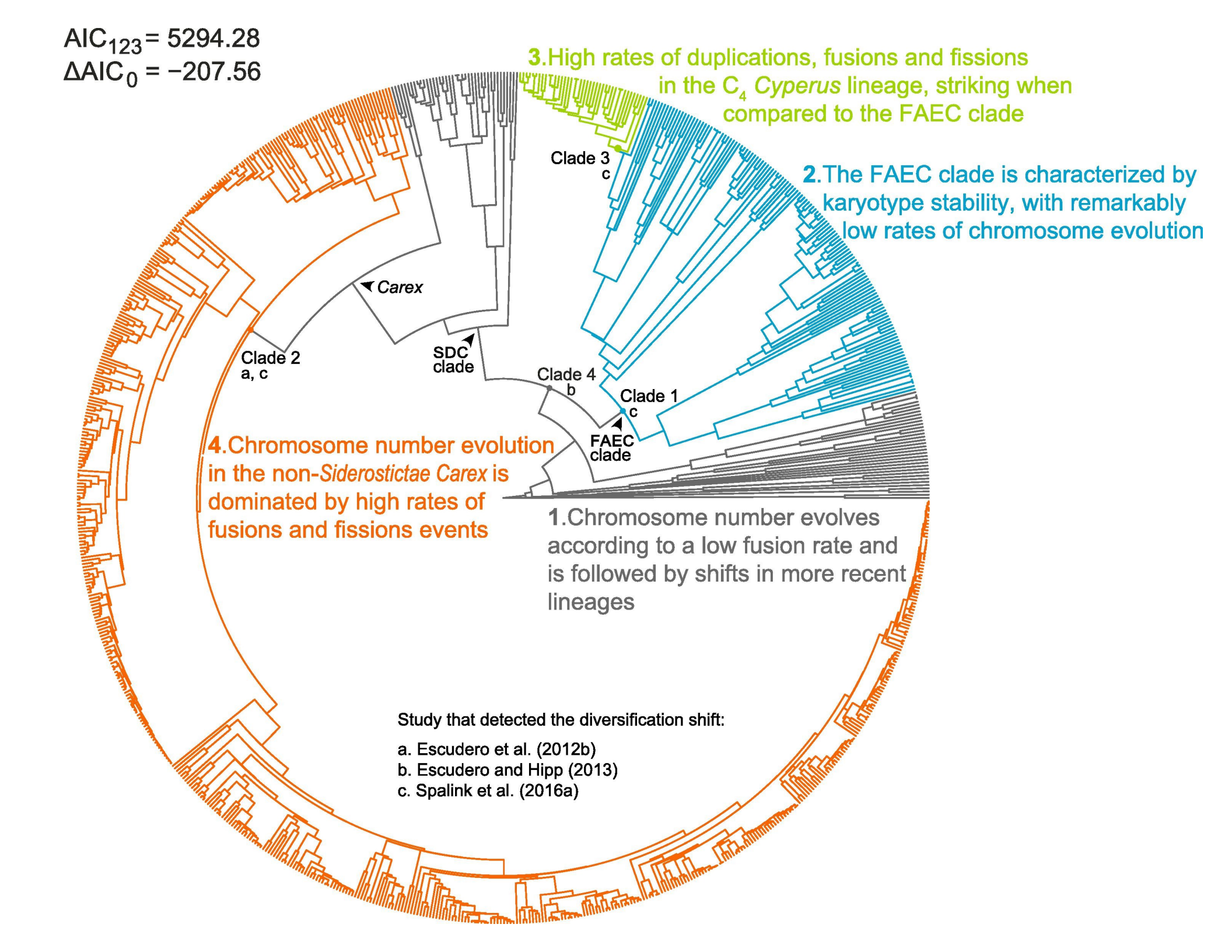
AIC = 3550.03 + 343.06 + 277.70 + 908.68 + 217.16 = **5296.63**

* When the same model was applied to two or more subtrees, loglikelihoods and AIC values were calculated excluding branches that connected the subtrees.

4. BEST TRANSITION SCENARIO

Choose the best scenario (see Fig. 2)




AIC_{123} = 5294.28
ΔAIC_0 = −207.56
3.High rates of duplications, fusions and fissions in the C_4 *Cyperus* lineage, striking when compared to the FAEC clade
2.The FAEC clade is characterized by karyotype stability, with remarkably low rates of chromosome evolution
Clade 3
c
Carex
Clade 2
a, c
SDC
clade
Clade 4
b
Clade 1
c
FAEC
clade
4.Chromosome number evolution in the non-*Siderostictae Carex* is dominated by high rates of fusions and fissions events
1.Chromosome number evolves according to a low fusion rate and is followed by shifts in more recent lineages
Study that detected the diversification shift:
a. Escudero et al. (2012b)
b. Escudero and Hipp (2013)
c. Spalink et al. (2016a)