Russian Entomol. J. 32(4): 375–382

Chromosomes of Symphyta (Hymenoptera): current state and perspectives of research

Хромосомы сидячебрюхих перепончатокрылых (Symphyta): степень изученности и перспективы исследований

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KEY WORDS: Chromosome number, karyotype evolution, lower Hymenoptera, molecular cytogenetics, phylogenetic reconstructions.
КЛЮЧЕВЫЕ СЛОВА: Молекулярная цитогенетика, низшие перепончатокрылые, филогенетические реконструкции, число хромосом, эволюция кариотипа.

ABSTRACT. The current state and perspectives of karyotype research on the suborder Symphyta (Hymenoptera), or sawflies and horntails, are discussed. The haploid chromosome number (n) in the studied members of the group can vary from 5 to 35, although the ancestral state of this character is apparently closer to n = 25. Metacentrics and submetacentrics prevail in the karyotypes of the majority of Symphyta, but subtelocentrics and/or acrocentrics are often present in the chromosome sets of species with higher chromosome numbers. In the process of karyotype evolution, most horntails and sawflies retained higher n values, but this variable decreased in the superfamily Tenthredinoidea down to n = 10 and less. Prospective directions of chromosome research of the Symphyta imply a detailed investigation of the karyotype structure of these insects, including preparation of karyograms and morphometric analysis of chromosomes, as well as the chromosome study of horntails and sawflies by using techniques of molecular cytogenetic analysis.

РЕЗЮМЕ. Обсуждается нынешнее состояние и перспективы исследования кариотипов перепончатокрылых, относящихся к подотряду сидячебрюхих (Symphyta), или рогохвостов и пилильщиков. Гаплоидное число хромосом (n) у исследованных представителей данной группы может изменяться от 5 до 35, но анцестральное значение этого признака, очевидно, близко к n = 25. В составе кариотипов большинства Symphyta преобладают метацентрики и субметацентрики, однако в хромосомных наборах видов с высоким числом хромосом часто присутствуют субтелоцентрики и/или акроцентрики. В ходе эволюции кариотипа большинство пилильщиков и рогохвостов сохраняли высокие значения n, однако в надсемействе Tenthredinoidea произошло снижение этого показателя до n = 10 и ниже. Перспективные направления хромосомного исследования Symphyta, очевидно, связаны с детальным изучением структуры кариотипа этих насекомых, включая составление кариограмм и морфометрический анализ хромосом, а также с исследованием хромосомных наборов рогохвостов и пилильщиков с помощью методов молекулярной цитогенетики.

Introduction

The suborder Symphyta (= lower Hymenoptera), or sawflies and horntails, represents a relatively minor and the least advanced hymenopteran group. It includes more than 8,000 described species [Huber, 2017], but karyotypes of just about 400 members of this suborder, i.e., less than five per cent of its known species number, are studied up to now [Westendorff, 2006; Gokhman, 2023]. Nevertheless, chromosomes of this group were intensively studied during previous decades. Initial steps in this direction were summarized in the 1930s [Sanderson, 1932], when just a few sawfly species, mainly those of the family Tenthredinidae, were examined for the first time. In the middle of the 20th century, Maxwell [1958] and Sanderson [1971] significantly contributed to the karyotype research of the Symphyta, mostly of the families Diprionidae, Pergidae, Siricidae and a few other higher taxa. However, foundations of the modern karyotypic study of the whole suborder were laid by Naito [1982 onwards], who developed a simple and effective technique for obtaining chromosome preparations

How to cite this article: Gokhman V.E. 2023. Chromosomes of Symphyta (Hymenoptera): current state and perspectives of research // Russian Entomol. J. Vol.32. No.4. P.375–382. doi: 10.15298/rusentj.32.4.02

of sawflies. This technique is based on the karyotyping of haploid embryos, which, in turn, develop inside unfertilized mature eggs obtained via dissection of adult females and then incubated under artificial conditions. Using this method, as well as other techniques aimed for obtaining chromosome preparations, in the second half of the 20th century Naito, together with his co-authors, managed to study karyotypes of approximately two hundred species of the Symphyta, which mainly belonged to the family Tenthredinidae and Argidae [Naito, 1982]. Westendorff and co-authors [Westendorff *et al*., 1999 onwards] subsequently studied chromosome sets of European Tenthredinidae and some other subordinate taxa of the Symphyta. As a result of this research, an updated review of the known chromosome numbers and other karyotypic features of approximately 370 species of this suborder was published about two decades ago [Westendorff, 2006]. During the last years, I also studied chromosomes of several dozen sawfly species, which belong to the families Xyelidae, Tenthredinidae, Argidae and Cimbicidae [Gokhman, Kuznetsova, 2018a; present paper; Gokhman, *in prep.*]. This work therefore represents an update and a brief overview of the current knowledge of the karyotypes of lower Hymenoptera, as well as of the perspectives of the chromosome research on these insects.

Material and methods

Unless otherwise stated, adult sawflies collected by the author in 2022–2023 near Ozhigovo, Russia (about 60 km SW Moscow: 55°28'N; 36°52'E) were mainly used in this study, except for *Abia fasciata*, which was collected near Bicaz, Romania (about 280 km N Bucharest: 46°54'N; 26°05'E). Chromosome preparations made in 2008 from early pupae of *Xyela julii*, which were also collected by the author near Ozhigovo, were re-examined. All specimens were preliminarily identified by the author; most identifications were then checked by S.A. Basov (Zoological Institute, Russian Academy of Sciences, St. Petersburg, Russia); *A. fasciata* was also identified by M. Prous (University of Oulu, Oulu, Finland). In terms of taxonomy and phylogeny of the Symphyta, I have chosen a somewhat conservative approach by generally following Malm and Nyman [2015].

Chromosome preparations were obtained according to the guidelines provided by Naito [1982] and Imai *et al*. [1988] with a few modifications. Adult females were dissected in small Petri dishes in distilled water, unfertilized mature eggs were extracted from their bodies and placed into the dishes on a filter paper soaked with distilled water, and then incubated for 3–4 days at room temperature. Haploid embryos were extracted from the eggs and dissected in 0.5% hypotonic sodium citrate solution containing 0.005% colchicine. The embryos were then transferred to a fresh portion of hypotonic solution and incubated for about 30 min at room temperature. The material was transferred onto a pre-cleaned microscope slide using a Pasteur pipette and then gently flushed with Fixative I (glacial acetic acid: absolute ethanol: distilled water 3:3:4). The tissues were disrupted using dissecting needles in an additional drop of Fixative I. Another drop of Fixative II (glacial acetic acid: absolute ethanol 1:1) was applied to the center of the area, and the more aqueous phase was blotted off the edges of the slide. The slides were then dried and stained with 3 per cent Giemsa solution for a few hours. In the case of *X. julii*, last-instar larvae collected from male cones of the Scotch pine, *Pinus sylvestris* Linnaeus, were allowed to pupate in the lab, and gonads of early pupae were then used for making chromosome preparations according to the protocol developed by Imai *et al*. [1988] (see above).

Mitotic divisions were studied and photographed using an optic microscope Zeiss Axioskop 40 FL fitted with a digital camera Axiocam 208 color (Carl Zeiss, Germany). To produce illustrations, the resulting images were handled with the image processing programs ZEN version 3.0 (blue edition) (Carl Zeiss) and GIMP version 2.10. Mitotic chromosomes were classified according to the guidelines provided by Levan *et al*. [1964].

Results

Family Xyelidae. *Xyela julii* (Brébisson, 1818), 2n = 50 (Fig. 1). Chromosome preparations obtained from 5 early pupae of *X. julii* have been studied. The first four pairs of chromosomes are slightly longer than the remaining ones, which gradually decrease in length. Although it is difficult to identify centromere positions in some shorter elements, chromosomes of all types are present in the karyotype of this species, including a few apparent metacentrics.

Family Cimbicidae. *Abia fasciata* (Linnaeus, 1758), n = 16 (Fig. 2). Chromosome set of a single female has been examined. All elements are apparently represented by subtelocentrics and/or acrocentrics forming a slow gradation in length, with the first ten chromosomes being slightly longer than the remaining ones.

Family Tenthredinidae. *Dolerus* (*Oncodolerus*) *eversmanni* Kirby, 1882, n = 8 (Fig. 3). As in the previous species, karyotype of a single specimen has been studied. All chromosomes are obviously bi-armed, i.e., either metacentric or submetacentric, and gradually decrease in size; the first and the last chromosomes are respectively more or less longer/shorter than the remaining ones.

D. (*Poodolerus*) *gonager* (Fabricius, 1781), n = 8 (Fig. 4). Chromosomes of two females have been examined. As in the previous species, all chromosomes are visibly bi-armed. However, elements of the karyotype are more differentiated in terms of size than in *D. eversmanni*. For example, the last metacentric is about 3.5 times shorter than the first one, and the second chromosome is obviously longer than the third element.

Empria sexpunctata (Serville, 1823), n = 13 (Fig. 5). Chromosome set of a single individual has been studied. Although most chromosomes are bi-armed, at least three subtelocentrics/acrocentrics obviously present in the karyotype. The first submetacentric chromosome is substantially longer than the second one, which is, in turn, almost equal in length to the third submetacentric, as are 4th to 6th, 7th to 10th, and 11th to 13th chromosomes respectively.

E. pallimacula (Serville, 1823), n = 10 (Fig. 6). Karyotypes of three females have been examined. All elements of the chromosome set are obviously bi-armed. Possible groupings of the chromosomes by size include the first, second to 4th, 5th to 7th, and 8th to 10th elements.

Karyotype features of particular families of Symphyta

Superfamily Xyeloidea. Family Xyelidae. Chromosomes of the only member of this family, *Xyela julii*, have been studied up to now. Its haploid chromosome number, n = 25, was reported about 15 years ago [Gokhman, 2009], but a detailed description of the karyotype is given in the present paper (see above).

Superfamily Pamphilioidea. Family Megalodontesidae. Again, the chromosome set of a single species, *Megalodontes thor* Taeger, 2002 with n = 20 is currently known [Westendorff, 2006]. Subtelocentrics and/or acrocentrics apparently prevail in this karyotype.

Family Pamphiliidae. About 10 species of this relatively large family have been studied [Boato, Battisti, 2001]. In this group, the chromosome number ranges from n = 11 to n = 35. Due to an extensive variation, no obvious modal n value can be identified. In addition, *Cephalcia arvensis* Panzer, 1803 apparently represents a complex of morphologically similar cryptic species with different chromosome numbers, n = 23 to 29.

Superfamily Tenthredinoidea. By far, this is the best studied superfamily of sawflies, with about 370 examined species. This group is very diverse, in terms of both the chromosome number and other parameters of karyotype structure. Specifically, the haploid number in the Tenthredinoidea can vary from n = 5 to n = 22, and while metacentrics and submetacentrics generally predominate in the chromosome sets of this group, certain members have karyotypes with relatively high shares of subtelocentrics and/or acrocentrics (see below).

Family Blasticotomidae. No karyotype data are currently available for this family.

Family Pergidae. Six members of this relatively large family that belong to the genera *Perga* Leach, 1817, *Philomastix* Froggatt, 1890 and *Pterygophorus*

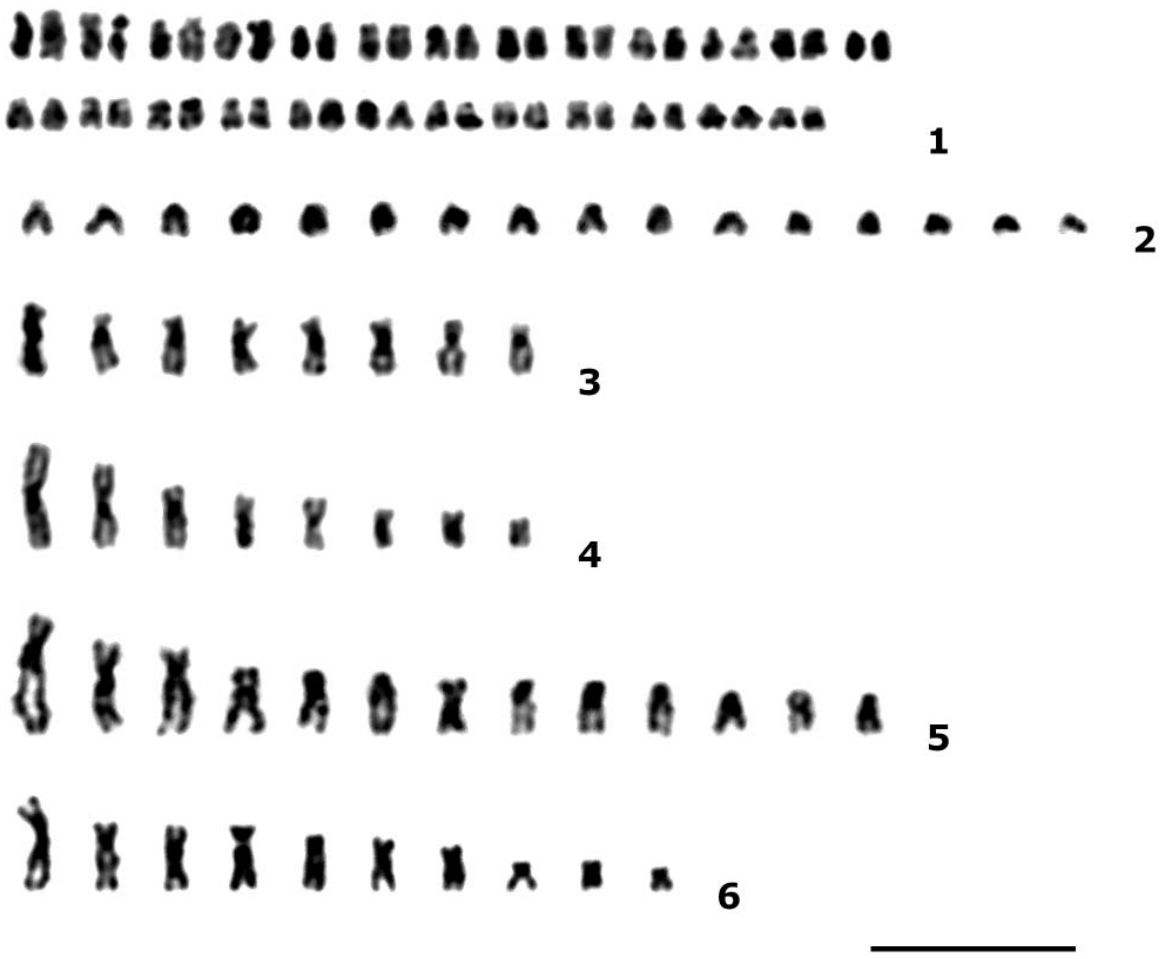

Fig. 1–6. Representative karyograms of Symphyta, diploid (1) and haploid (2–6) chromosome sets. Bar = 10 μm. 1 — X*yela julii*, 2 — *Abia fasciata*, 3 — *Dolerus eversmanni*, 4 — *D. gonager*, 5 — *Empria sexpunctata*, 6 — *E. pallimacula*.

Рис. 1–6. Репрезентативные кариограммы Symphyta, диплоидные (1) и гаплоидные (2–6) хромосомные наборы. Масштаб 10 мкм. 1 — *Xyela julii*, 2 — *Abia fasciata*, 3 — *Dolerus eversmanni*, 4 — *D. gonager*, 5 — *Empria sexpunctata*, 6 — *E. pallimacula*.

Klug, 1814 are studied [Maxwell, 1958]. Despite all these genera belong to separate subfamilies, all pergids turned to have n = 8.

Family Argidae. Karyotypes of 12 members of the genus *Arge* Schrank, 1802 are examined up to now [Maxwell, 1958; Naito, 1982; Westendorff, Taeger, 2002; Gokhman, *in prep*. etc.]. The chromosome number in this group ranges from n = 8 to n = 13, with 6 and 5 species having n = 8 and 10 respectively. Most chromosomes are metacentric; however, the proportion of subtelocentrics and/or acrocentrics apparently increases within higher-numbered chromosome sets. Interestingly, both n = 8 and 10 are recorded for *A. ustulata* (Linnaeus, 1758) [Greenshields, 1937; Gokhman, *in prep*.], which may indicate presence of cryptic taxa within this widely distributed morphospecies.

Family Heptamelidae. Chromosome sets of the two species that belong to this smaller family, *Pseudoheptamelus runari* Conde, 1932 and *Heptamelus ochroleucus* (Stephens, 1835), with n = 7 and 10 respectively, are currently known [Naito, 1982]. All chromosomes are obviously bi-armed, although a few subtelocentrics are apparently present in the karyotype of the former species.

Family Diprionidae. Karyotypes of about 40 members of this group have been studied up to now, with n varying from 6 to 15, and a clear maximum of 7 [Maxwell, 1958; Westendorff, 2006]. Most chromosome sets with lower n values contain only metacentrics and/or submetacentrics, but acrocentrics prevail in the high-numbered karyotypes. Two apparently cryptic members of the genus *Gilpinia* Benson, 1939, *G. polytoma* (Hartig, 1834) and *G. hercyniae* (Hartig, 1837), have different chromosome numbers, n = 6 and 7 respectively; moreover, the former species is arrhenotokous, whereas the latter one exhibits diploid thelytoky [Smith, 1941].

Family Cimbicidae. Chromosomes of five members of this family are known, with two species having n = 8, the third one n = 9, and another two — n = 16 [Westendorff, 2006; present paper]. Westendorff and Taeger [2002] report at least two metacentrics/submetacentrics in the karyotype of *Abia candens* Konow, 1887 with n = 16, but they admit on being unsure about the centromere positions due to the small size of chromosomes. In *A. fasciata* with the same n value, all chromosomes apparently look like subtelocentrics and/or acrocentrics (Fig. 2, see above).

Family Tenthredinidae. Chromosomes of approximately 300 species of this vast group are examined up to now [Naito, 1982; Westendorff, 2006; Gokhman, *in prep*]. In this family, the chromosome number ranges from n = 5 to n = 22, with an obvious maximum at n = 10 [Naito, 1982; Westendorff, 2006], mostly due to the prevalence of the members of the subfamily Tenthredininae among the studied species. As in many other sawflies, metacentrics and submetacentrics predominate in the chromosome sets of this group, but the proportion of subtelocentrics and/or acrocentrics tends to increase with the chromosome number. However, karyotypes of certain subordinate taxa of the Tenthredinidae can have somewhat different parameters. For instance, species of the genus *Athalia* Leach, 1817 *s.l.*, which is sometimes considered as a member of a separate family Athaliidae [Niu *et al*., 2022], can have either n = 6 or n = 8, with a single exception of *A. scutellariae* Cameron, 1880 with n = 7 [Westendorff, 2006]. Analogously, n = 6-9 were found in the relatively large subfamily Nematinae [Naito, 1982; Westendorff, 2006]. On the other hand, all four studied members of the genus *Empria* Lepeletier et Serville, 1828, i.e., *E. pallimacula*, *E. parvula* (Konow, 1892), *E. sexpunctata* and an unidentified species from Japan, appeared to have different chromosome numbers: n = 10, 11, 13 and 15 respectively [Westendorff, 2006; present paper]. Moreover, I have found n = 8 in *Dolerus* (*Oncodolerus*) *eversmanni* (Fig. 3), as in most other members of this genus, although Naito [1982] apparently reports 7 pairs of chromosomes in *D.* (*O.*) *obscurus* Marlatt, 1898, which is usually treated as a subspecies of *D.* (*O.*) *eversmanni*. In addition, certain groups of apparently cryptic species have been identified in the family Tenthredinidae. For example, *Tenthredo* (*Tenthredo*) *arcuata* Förster, 1771, *T.* (*T.*) *brevicornis* (Konow, 1886) and *T.* (*T.*) *notha* Klug, 1817 are close to each other in terms of external morphology, but they have different chromosome numbers. i.e., n = 10, 18 and 20 respectively [Westendorff, 2006].

Superfamily Cephoidea. Family Cephidae. Up to now, karyotypes of only three members of this group are known [Westendorff, Taeger, 2002]. All these sawflies belong to different genera and display a wide range of chromosome numbers, from n = 9 to n = 26, and therefore it is currently impossible to identify the modal n value for the Cephidae.

Superfamily Siricoidea. Family Anaxyelidae. No chromosomal data are available for this smaller family.

Family Siricidae. Chromosome sets of five species of this group have been studied [Sanderson, 1971]. Among these, three members of the genus *Sirex* Linnaeus, 1760 have n = 8 with obviously bi-armed chromosomes, whereas two species of *Urocerus* Geoffroy, 1762 have n = 13 and 18. Although n = 8 therefore represents a formal modal number for the family, it can hardly be considered as the putative ancestral character state for the Siricidae in general.

Superfamily Xiphydrioidea. Family Xiphydriidae. No karyotype data are available for this group.

Superfamily Orussoidea. Family Orussidae. As in the previous family, no chromosomal data are available for this group.

General features of chromosome sets of Symphyta

As in many other hymenopterans [Crozier, 1975; Gokhman, Kuznetsova, 2018b], arrhenotoky, i.e., development of males from unfertilized eggs, apart from females, is characteristic of the overwhelming majority of the Symphyta. Nevertheless, a moderate number of species with diploid thelytoky, when, on the contrary, unfertilized eggs give rise only to females [Gokhman,

Kuznetsova, 2018b], are apparently known in this suborder [Smith, 1941; Sanderson, 1971]. In addition, I believe that those members of the Tenthredinidae, in which unfertilized eggs gave rise to diploid embryos under Naito's experiments [Naito, 1982], also likely belong to this group. Moreover, *Pachyprotasis youngiae* Inomata et Naito from the same family is the only known thelytokous triploid member of the Symphyta [Naito, Inomata, 2006]. On the other hand, not only diploid, but also triploid males can be produced in the laboratory via strict inbreeding at least in *Athalia rosae ruficornis* Jakowlew, 1888 [Naito, Suzuki, 1991].

Among different superfamilies of the Symphyta, the haploid chromosome number (n) can vary from n = 5 to n = 35 [Westendorff, 2006]. On the other hand, the modal, i.e., the most frequent, numbers for the majority of superfamilies belong to the much narrower interval. For example, Tenthredinoidea include several families with modal n values of different families fall into the range of n = 7 to 10 [Naito, 1982; Westendorff, 2006]. Karyotypes mostly containing obviously bi-armed chromosomes, i.e., metacentrics and submetacentrics, prevail in many groups of the Symphyta. Nevertheless, chromosome sets, which mainly harbor elements with either hardly visible or virtually absent shorter arms, i.e., subtelocentrics and acrocentrics, are known for a few taxa, mostly with higher chromosome numbers (see, e.g., Fig. 2).

Almost all data on symphytan karyotypes were therefore obtained from studies of routinely stained chromosomes and, even in those cases, karyograms of chromosome sets were rarely presented (see, e.g., [Nishimoto *et al*., 2014]). Moreover, karyotypes of these insects were never analyzed using morphometric analysis of chromosomes, although related sawfly species with the same haploid number can strongly differ in other characteristics of the karyotype structure, e.g., chromosome size (Figs 3–4).

In addition to the routine staining, a few karyotypes of the family Tenthredinidae were studied using either "traditional" or "modern" techniques of chromosome banding (see [Gokhman, 2023]). Specifically, chromosomes of several members of the genera *Tenthredo* Linnaeus, 1758 and *Rhogogaster* Konow, 1884 were subjected to C- and AgNOR-bandings, which reveal distribution of the constitutive heterochromatin and nucleolus organizing regions (NORs) respectively [Kuznetsova *et al*., 2001]. In these sawflies, C-banding visualized pericentromeric and sometimes interstitial heterochromatin, whereas silver nitrate staining detected a single NOR per haploid set in the only studied species, *T.* (*Eurogaster*) *mesomela* Linnaeus, 1758. Similar heterochromatin distribution was also found in *Diprion pini* (Linnaeus, 1758) (Diprionidae) [Rousselet *et al*., 1998]. At least in *T.* (*E.*) *mesomela*, NOR location was confirmed by chromosome staining with chromomycin A_3 (CMA_3), which reveals CG-rich chromosome segments, often represented by NORs. However, among the three examined species, *T. arcuata* was the only one having two CMA_3-positive bands. On the contrary, staining with AT-specific fluorochrome, 4',6-diamidino-2-phenylindole (DAPI), did not detect any clusters on chromosomes of these sawflies, similarly to most other hymenopterans (see, e.g., Gokhman [2009]). Moreover, fluorescence *in situ* hybridization (FISH) with rDNA probe, another comprehensive tool for visualizing NORs, revealed four rDNA sites on the chromosomes of *Athalia rosae* (Linnaeus, 1758) [Matsumoto *et al*., 2002]. Nevertheless, analogous experiments with the rDNA probe detected by a non-fluorescent marker, visualized single rDNA clusters in the karyotypes of both diprionids, *D. pini* and *Neodiprion abietis* (Harris, 1841) [Rousselet *et al*., 1998, 2000]. Similarly, DNA probe with the vitellogenin gene detected a single subterminal signal on a particular chromosome of *A. rosae* [Matsumoto *et al*., 2002]. Sumitani *et al*. [2005] also used FISH to locate a single copy of the *white* gene orthologue on chromosomes of the same species.

An enormous variation in the so-called telomeric motifs, i.e., short DNA repeats located at the telomeres, i.e., terminal segments of chromosomes, has been recently revealed among different clades of the order Hymenoptera [Zhou *et al*., 2022; Lukhtanov, Pazhenkova, 2023]. Nevertheless, we used FISH to discover that chromosomes of Symphyta, i.e., two members of the genus *Tenthredo*, *T.* (*Tenthredo*) *omissa* (Förster, 1844), and *T.* (*Endotethryx*) *campestris* Linnaeus, 1758 (the latter sawfly was wrongly identified in our paper as a species of the genus *Taxonus* Hartig, 1837), carry the canonical insect telomeric motif, TTAGG, at their telomeres [Gokhman, Kuznetsova, 2018a], and therefore suggested the presence of this repeat in the lower Hymenoptera as the ancestral character state for the whole order. This hypothesis was later confirmed for other Tenthredinidae, as well as for the members of Cephidae and Orussidae, using bioinformatic approaches [Zhou *et al*., 2022; Lukhtanov, Pazhenkova, 2023].

Taxonomic implications of chromosome study of Symphyta

Related species of sawflies often have similar karyotypes, and this can be observed, for example, in the genus *Macrophya* Dahlbom, 1835 (Tenthredinidae), in which some species have chromosome sets n = 10 with a very large first metacentric, whereas some other members of *Macrophya* have 8 chromosomes that gradually decrease in size [Westendorff *et al*., 1999]. In the genus *Athalia*, different chromosome numbers, n = 6 and 8, are correlated with certain morphological and ecological traits of most species [Abe, 1988]. Moreover, there are several reports of different chromosome numbers and other karyotypic features for apparently the same sawfly species (see [Westendorff, 2006] for review). However, it is often difficult to choose between wrong taxonomic identifications and the presence of morphologically cryptic species, especially if those reports are presented by different researchers. Nevertheless, the presence of true cryptic taxa is already detected in the families Pamphiliidae, Tenthredinidae and Diprionidae, and it is also supposed for Argidae (see above).

Pathways of karyotype evolution of Symphyta

An analysis of chromosomal variation in the suborder Symphyta using the phylogenetic reconstruction provided by Malm and Nyman [2015] (Fig. 7) suggests that the putative ancestral karyotype of the lower Hymenoptera apparently resembled the chromosome set of the only studied member of the most basal superfamily Xyeloidea, *Xyela julii*, with an n value around 25, and a considerable proportion of subtelocentrics and acrocentrics. Consequently, most other families of sawflies and horntails apparently retained higher chromosome numbers (i.e., about n = 20), except for those of the Tenthredinoidea with the modal ones of n = 7 to 10. In addition, the view of ancestral higher n values for most superfamilies of the Symphyta is further supported by the estimate of n = 14 to 19 as the initial chromosome number for the Apocrita [Gokhman, 2007]. Since the modal n values in the Tenthredinoidea are substantially lower than those characteristic of any other large group of the Symphyta, an obvious decrease in the chromosome number has therefore occurred in the former superfamily [Gokhman, 2007], although this general pattern does not exclude an increase in this parameter within certain genera and/or species of the Tenthredinoidea [Rousselet *et al*., 2000] (also see above).

Due to the scarce use of modern techniques of chromosome study of the Symphyta, little can be said at

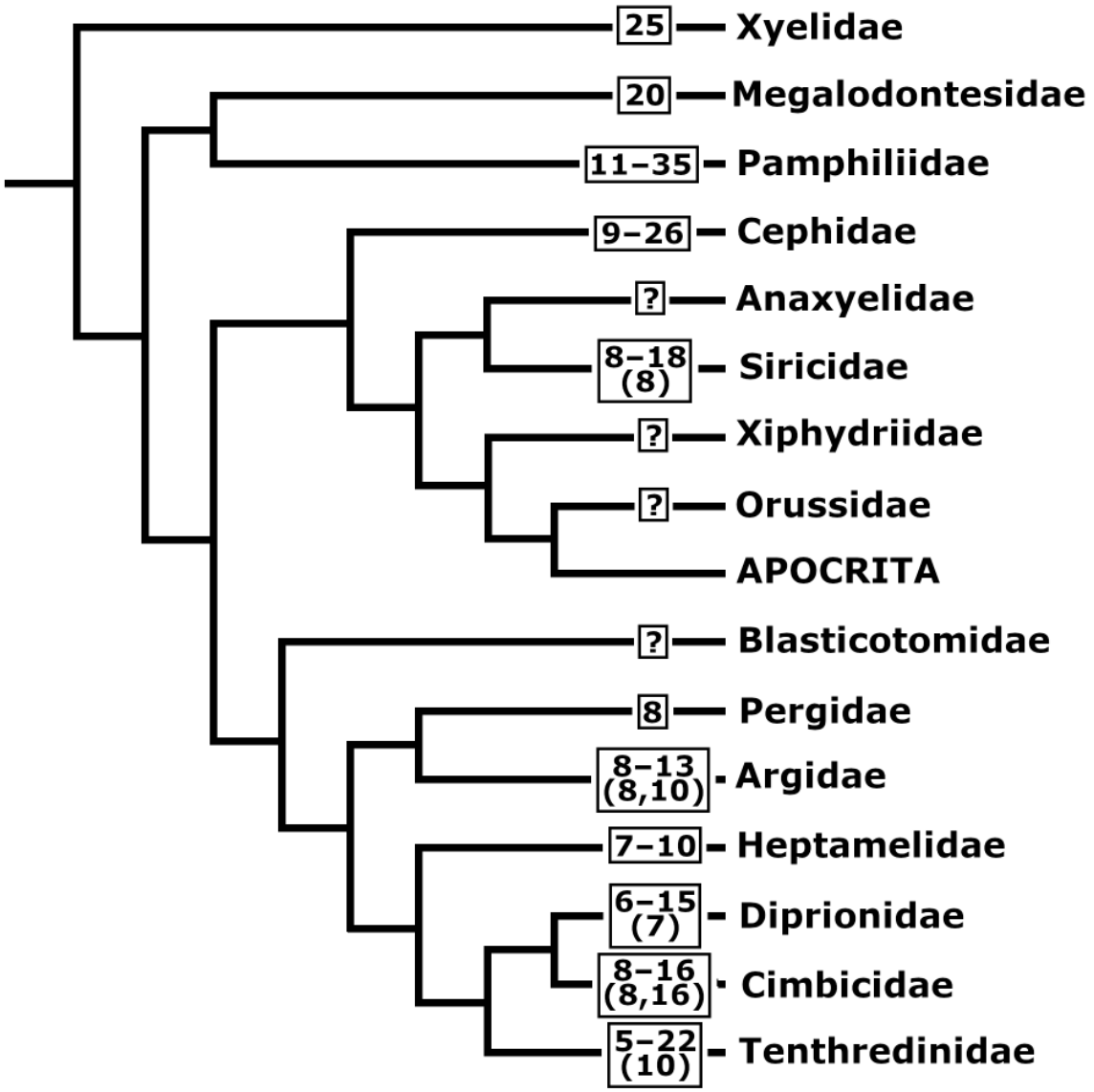

Fig. 7. Phylogeny of symphytan families (simplified after Malm and Nyman [2015]) with indications of variation ranges of haploid chromosome numbers and their modal values.
Рис. 7. Филогения семейств Symphyta (упрощено по: [Malm, Nyman, 2015]) с указанием пределов изменчивости гаплоидных хромосомных чисел и их модальных значений.

present about possible chromosomal rearrangements that underlie the evolutionary changes of their karyotypes. Nevertheless, as noted above, species with higher chromosome numbers often tend to have an increased proportion of subtelocentrics/acrocentrics in the chromosome set (Figs 5–6), and this feature suggests that centric fusions/fissions can be involved, depending on the direction of change in the chromosome number [Westendorff, 2006]. In addition, differences in the chromosome size between related species with the same n values can be explained by other rearrangements, such as translocations or deletions/duplications of the constitutive heterochromatin (see, e.g., [Gokhman, 2009]).

Conclusions and further perspectives of the chromosome study of Symphyta

Discussing the current state of karyotype research on the Symphyta in terms of the best/least studied groups, we can conclude that chromosomes of Tenthredinidae are fairly well known, but data on the karyotypes of Anaxyelidae, Blasticotomidae, Xiphydriidae and Orussidae are entirely missing, and in some other families, i.e., Xyelidae, Megalodontesidae, Cimbicidae, Pergidae, Siricidae and Cephidae, only several species are studied. Furthermore, quite a few papers on the subject were published during the last ten years [Nishimoto *et al*., 2014; Gokhman, Kuznetsova, 2018a], and karyotypes of many common species remain unknown. Nevertheless, I believe that further chromosome study of the Symphyta using both conventional and advanced techniques will have important implications for the genetic, taxonomic and phylogenetic research of the lower Hymenoptera.

Acknowledgements. The author is very grateful to S.A. Basov and M. Prous for identifying sawflies as well as for providing important information on their taxonomy. The present study was supported by the Russian Science Foundation (grant no. 23-24-00068).

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