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Chromosomes of parasitic wasps of the genus *Metaphycus* (Hymenoptera: Chalcidoidea: Encyrtidae)

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Abstract. Karyotypes of four species of the genus *Metaphycus* Mercet, 1917, namely, *M. flavus* (Howard, 1881) and *M. luteolus* (Timberlake, 1916) (both have n = 10 and 2n = 20), *M. angustifrons* Compere, 1957 (n = 9 and 2n = 18) and *M. stanleyi* Compere, 1940 (n = 5 and 2n = 10) were studied. The latter chromosome number, n = 5, is the lowest known one for the family Encyrtidae. A karyotype with n = 10 is considered ancestral for the genus *Metaphycus*. Karyotype evolution in this genus is likely to have occurred through chromosomal fusions.

Key words: chromosomes, karyotypes, Chalcidoidea, Encyrtidae, *Metaphycus*.

Introduction

The world fauna of the family Encyrtidae, a large group of the superfamily Chalcidoidea, contains more than 3700 species (http://www.nhm.ac.uk/research-curation/research/projects/chalcidoids/encyrtidae.html, on March 26, 2010). Chromosomes of only about 15 species of the family (i.e. less than 0.5% of described species) have been studied up to now (Gokhman, 2009). Moreover, karyotypes of the large genus *Metaphycus* Mercet, 1917 that includes many economically important parasitoids of soft scales (Homoptera: Coccidae) and related insects (Trjapitzin, 1989) remained fully unknown until recently. I have managed to study chromosomes of four species of this genus that are cultivated at the University of California at Riverside (USA) (UCR). Descriptions of these karyotypes are given below.

Material and methods

Air-dried preparations of mitotic chromosomes were obtained in August 2009 from cerebral ganglia of prepupae extracted from parasitized individuals of *Coccus hesperidum* Linnaeus, 1758 (Coccidae). Both parasitoids and their hosts were kept as laboratory stocks at UCR. The preparations were made according to the technique used by Imai et al. (1988) with certain modifications. Images of metaphase plates were obtained using light microscope Zeiss Axioskop 40 FL fitted with the digital camera AxioCam MRc (Table 1). Chromosomes were measured on digital micrographs of haploid metaphase plates using Zeiss AxioVision, their relative lengths (RL) and centromere indices (CI) were calculated (Table 2). However, since centromere position could not be precisely identified on chromosomes of certain species, CI values for those species were omitted in the table (see below). All chromosomes were arranged on karyo-

Table 1. Number of studied individuals and metaphase plates of *Metaphycus* species.

Species	No. individuals studied		No. metaphase plates studied	
	male	female	haploid	diploid
M. flavus	4	4	8	10
M. luteolus	1	3	2	10
M. angustifrons	6	2	10	3
M. stanleyi	4	1	17	2

Table 2. RL and CI values of chromosomes of *Metaphycus* species (mean ± SD). Numbers of metaphase plates used for morphometric analysis are given in brackets.

Species/ chromosome no.	***M. flavus*** **(4)**	***M. luteolus*** **(2)**	***M. angustifrons*** **(5)**		***M. stanleyi*** **(10)**	
	RL	RL	RL	CI	RL	CI
1	14.82 ± 1.25	12.00 ± 0.07	15.91 ± 0.78	48.51 ± 0.70	28.74 ± 1.99	47.96 ± 1.26
2	12.40 ± 1.13	11.91 ± 0.20	12.15 ± 0.46	7.79 ± 1.06	24.68 ± 1.55	37.10 ± 1.39
3	10.55 ± 0.46	11.28 ± 0.46	11.60 ± 0.34	21.59 ± 1.83	17.72 ± 1.36	38.43 ± 1.87
4	9.76 ± 0.33	11.02 ± 0.57	11.10 ± 0.43	22.72 ± 1.79	16.28 ± 1.17	38.08 ± 1.85
5	9.65 ± 0.33	10.72 ± 0.27	10.69 ± 0.31	23.07 ± 0.64	12.58 ± 1.54	43.78 ± 1.29
6	9.06 ± 0.34	9.78 ± 0.29	10.28 ± 0.08	22.22 ± 0.88		
7	8.96 ± 0.39	9.31 ± 0.30	9.84 ± 0.20	22.60 ± 0.87		
8	8.57 ± 0.29	8.46 ± 0.76	9.48 ± 0.40	21.32 ± 1.14		
9	8.31 ± 0.70	8.02 ± 0.51	8.95 ± 0.82	21.80 ± 0.65		
10	7.92 ± 0.53	7.50 ± 0.30				

grams according to the classification provided by Levan et al. (1964).

Results

Metaphycus flavus (Howard, 1881) (Fig. 1, a-c). n = 10, 2n = 20. Most chromosomes gradually decrease in size, except for the first pair (Fig. 1, b; or sometimes just one of the two homologues in the diploid set; Fig. 1, c) which is visibly longer than the others (see Table 2). All chromosomes appear to be either subtelocentric or acrocentric.

Metaphycus luteolus (Timberlake, 1916) (Fig. 1, d, e). n = 10, 2n = 20. The karyotype of this species is similar to that of the previous one. Nevertheless, chromosomes of the first pair are only slightly longer than the others (Table 2).

Metaphycus angustifrons Compere, 1957 (Fig. 1, f-h). n = 9, 2n = 18. Most chromosomes gradually decrease in size, except for the first pair, which is apparently metacentric (Table 2). Chromosomes of the second pair are obviously acrocentric, all other chromosomes are subtelocentric. In this species, a few metaphase plates with the so-

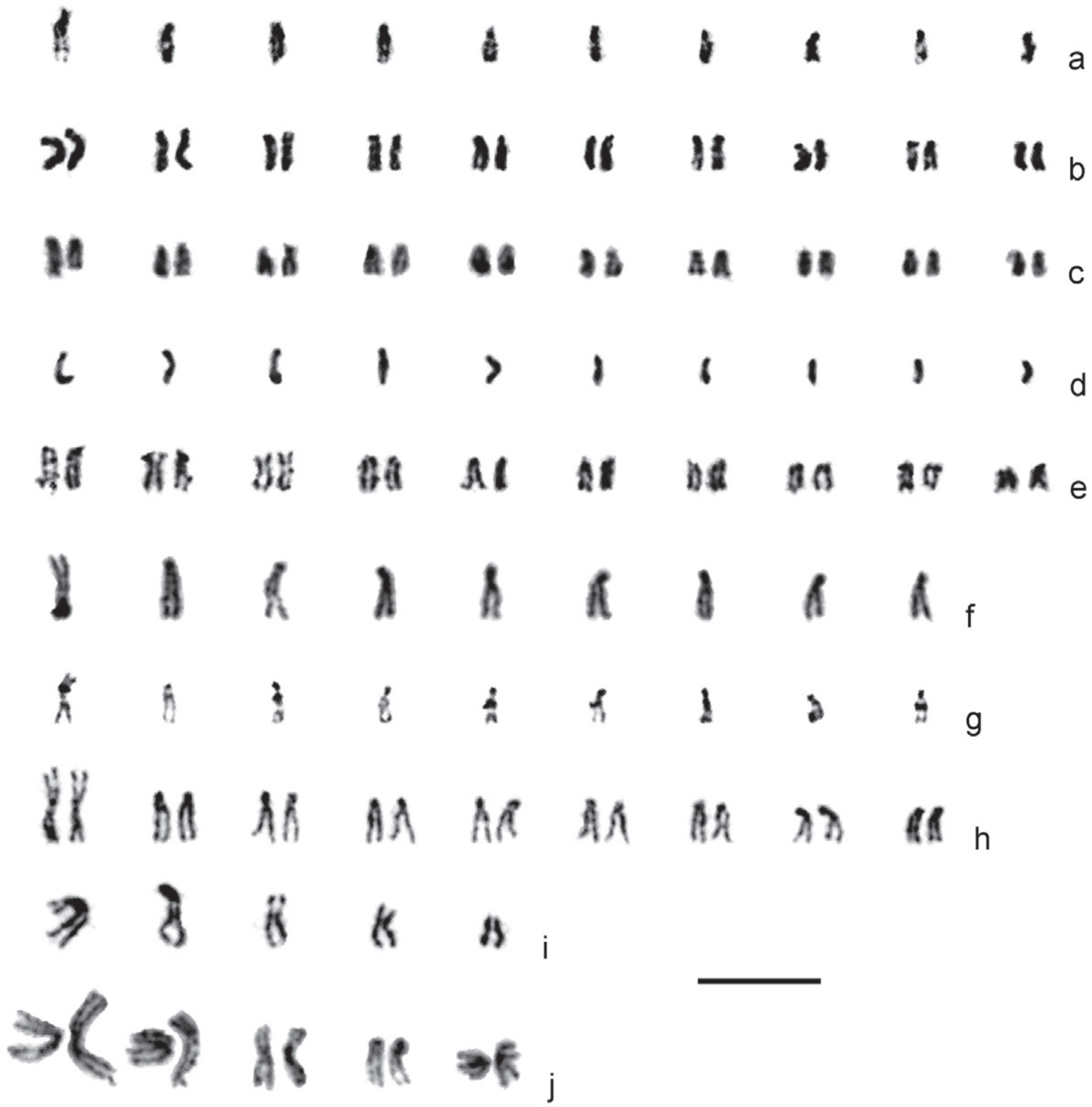

Fig. 1, a-j. Chromosomes of *Metaphycus* species. **a** - *M. flavus*, haploid karyogram. **b** - *M. flavus*, diploid karyogram, first pair of chromosomes homomorphic. **c** - ditto, first pair of chromosomes heteromorphic. **d** - *M. luteolus*, haploid karyogram. **e** - *M. luteolus*, diploid karyogram. **f** - *M. angustifrons*, haploid karyogram, routine chromosome staining. **g** - ditto, C-banding. **h** - *M. angustifrons*, diploid karyogram. **i** - *M. stanleyi*, haploid karyogram. **j** - *M. stanleyi*, diploid karyogram. Bar = 10 μm.

called spontaneous C-banding were found (Fig. 1, g). Shorter arms of all subtelocentrics are heterochromatic. Most chromosomes also carry visible intercalary (or sometimes pericentromeric) and weaker telomeric heterochromatic segments.

Metaphycus stanleyi Compere, 1940 (Fig. 1, i, j). n = 5, 2n = 10. All chromosomes of this species are obviously biarmed (mostly metacentric) and generally larger than those of the previous ones. Apart from karyotypes of preceding species, most chromosomes of *M. stanleyi* can easily be distinguished from the others by their length and centromere position.

Discussion

Karyotypes of the studied *Metaphycus* species show a considerable variation in terms of both chromosome number and chromosomal morphology. Surprisingly, substantial differences regarding length of the first chromosome have been found at least between the studied populations of *M. flavus* and *M. luteolus* with n = 10, although these species are sometimes considered synonymous (Guerrieri, Noyes, 2000). The two other species also have different n values, i.e. 9 and 5. The latter chromosome number, n = 5, is the lowest known one not only for the genus *Metaphycus*, but for the whole family Encyrtidae as well (the second lowest n value, n = 8, has been recorded for the genus *Copidosoma* Ratzeburg, 1844; see Gokhman, 2009 for review). The data obtained therefore indicate further blurring the border between the so-called "high-numbered" and "low-numbered" chalcid families. This phenomenon has recently been observed also in the Eupelmidae, Eurytomidae and Torymidae (Gokhman, Gumovsky, 2009).

Since the haploid karyotype with eleven acrocentric or subtelocentric chromosomes is considered ancestral not only for the Encyrtidae, but also for the superfamily Chalcidoidea as a whole (Gokhman, Gumovsky, 2009), the closest n value found in *Metaphycus flavus* and *M. luteolus*, n = 10, is likely to be ancestral for the genus *Metaphycus*. If this is true, chromosome numbers of *M. angustifrons* and *M. stanleyi*, n = 9 and 5 respectively, must be derived, and they have therefore originated through apparently subsequent chromosomal fusions. The fact that all shorter arms of subtelocentrics are heterochromatic in the former species, also favours this hypothesis because those arms can easily be lost during the fusion process. On the other hand, strong intercalary heterochromatic segments are infrequent on chromosomes of parasitic wasps (Gokhman, 2009), and this aberrant feature may be correlated with massive insertions of certain DNA sequences recently found in the Encyrtidae (Gillespie et al., 2005).

Finally, the present study has potential practical implications. Immature stages of the above mentioned parasitoids of the genus *Metaphycus* can easily be sorted to sex and species by their karyotypes. Population sex ratios of the studied *Metaphycus* species can therefore be evaluated at the prepupal stage using this technique (see also Gokhman, 2009).

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References

Gillespie J.J., Munro J.B., Heraty J.M., Yoder M.J., Owen A.K., Carmichael A.E. 2005. A secondary structural model of the 28S rRNA expansion segments and D3 for chalcidoid wasps (Hymenoptera: Chalcidoidea) // *Mol. Biol. Evol.* 22: 1593-1608. (doi:10.1093/molbev/msi152).

Gokhman V.E. 2009. Karyotypes of parasitic Hymenoptera. Dordrecht. 183 p. (doi:10.1007/978-1-4020-9807-9).

Gokhman V.E., Gumovsky A.V. 2009. Main trends of karyotype evolution in the superfamily Chalcidoidea (Hymenoptera) // *Comp. Cytogenet.* 3: 63-69. (doi:10.3897/compcytogen.v3i1.1).

Guerrieri E., Noyes J.S. 2000. Revision of European species of genus *Metaphycus* Mercet (Hymenoptera: Chalcidoidea: Encyrtidae), parasitoids of scale insects (Homoptera: Coccoidea) // *Syst. Entomol.* 25: 147-222. (doi:10.1046/j.1365-3113.2000.00099.x).

Imai H.T., Taylor R.W., Crosland M.W.J., Crozier R.H. 1988. Modes of spontaneous chromosomal mutation and karyotype evolution in ants with reference to the minimum interaction hypothesis // *Japanese J. Genet.* 63: 159-185.

Levan A., Fredga K., Sandberg A.A. 1964. Nomenclature for centromeric position on chromosomes // *Hereditas.* 52: 201-220.

Trjapitzin V.A. 1989. Parasitic Hymenoptera of the Fam. Encyrtidae of the Palaearctic. Leningrad. 488 p. (In Russian).

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