

Cytogenetic analysis of three species of *Trypoxylon* (*Trypoxylon*) (Hymenoptera, Sphecidae, Larrinae)

Luiz Fernando Gomes, Silvia das Graças Pompolo and Lucio Antonio de Oliveira Campos

ABSTRACT

We describe the karyotypes of three species of wasps of the genus *Trypoxylon* (*Trypoxylon*), *T. sp1* ($2n = 32$, $2K = 24A + 8SM$), *T. asuncicola* ($2n = 32$, $2K = 22A + 9SM + 1M$), and *T. fabricator* ($2n = 32$, $2K = 12A + 18SM + 2M$). Tandem growth of constitutive heterochromatin and pericentric inversions after the occurrence of centric fission are discussed as mechanisms for karyotype differentiation among the three species.

INTRODUCTION

Among the Sphecidae, *Trypoxylon* is the second largest genus in number of species (Bohart and Menke, 1976). Having noted differences both at morphological and biological levels, Richards (1934) divided the genus into two subgenera, *Trypoxylon* and *Trypargilum*. The subgenus *Trypargilum* is restricted to the Western part of the Southern hemisphere, whereas the subgenus *Trypoxylon* is cosmopolitan (Bohart and Menke, 1976).

The cytogenetic study of insects has greatly advanced due to the use of simple and efficient techniques for numerical and structural characterization and for the visualization of band distribution, thus providing an unambiguous identification of chromosome complements.

Only 18 species of the superfamily Sphecoidea (Sphecidae) have been studied thus far in cytogenetic terms (Costa *et al.*, 1993; Hoshiba and Imai, 1993). Four of these species belong to the genus *Trypoxylon*

(Hoshiba and Imai, 1993). The objective of the present report was to provide more information for the understanding of karyotype evolution in the genus *Trypoxylon*.

MATERIAL AND METHODS

Three species of *Trypoxylon* (subgenus *Trypoxylon*) were studied cytogenetically: *T. asuncicola*, *T. fabricator* and *T. sp1*. collections sites, numbers of nests and numbers of individuals for each species are listed in Table I.

The cytogenetic preparations were obtained from cerebral ganglia and/or gonads of larvae in the prepupal phase by the technique of Imai *et al.* (1988). C-bands were obtained by the technique of Sumner (1972), modified by Pompolo and Takahashi (1990).

On average, 25 metaphases per individual were analyzed and those of better quality were selected and photographed with a Zeiss photomicroscope, using Agfa Copex Pan A.H.U. film.

Table I - Data about collection sites, number of nests, number of individuals, chromosome number and karyotype formula of the *Trypoxylon* species studied.

Species	No. of nests	No. of individuals		Chromosome number		Karyotype formula
		Male	Female	(n)	(2n)	
<i>T. sp1</i> ³	1	-	3	-	32	2K = 24A + 8SM
<i>T. asuncicola</i> ^{1,2}	8	12	20	16	32	2K = 22A + 9SM + 1M
<i>T. fabricator</i> ¹	2	2	6	16	32	2K = 12A + 18SM + 2M

Sites:

Viçosa-MG (1)

Mariana-MG (2)

Manaus-AM (3)

The karyotype was mounted by arranging the chromosomes in decreasing order of euchromatic arm size.

RESULTS

All species studied had the same chromosome number. A haploid complement (n) equal to 16 and a diploid complement (2n) equal to 32 was determined for *T. asuncicola* and *T. fabricator*. Only the diploid complement (2n=32) was determined for *T. sp1*, since only female organisms were analyzed (Table I). The distribution of constitutive heterochromatin in *T. asuncicola* and *T. fabricator*, demonstrated by C-banding, was similar for chromosomes of the same morphology, and occurred throughout the length of one arm in meta and submetacentric chromosomes, except for pair 6 of *T. asuncicola*, which showed pericentromeric distribution, and pair 16 of *T. fabricator*, which was fully C-band positive (Figures 2a and 3b). The C-banding technique was not used for *T. sp1*. However, even in a standard preparation, it was

possible to infer the pattern of heterochromatin distribution due to the differential pattern of chromosome staining. More intensely staining regions were observed, possibly representing heterochromatin, corresponding to the long arm in the submetacentric chromosomes and to the pericentromeric region in acrocentrics. This banding pattern is similar to that of the other two species (Figure 1).

DISCUSSION

In view of the hypothesis that more primitive Hymenoptera had a haploid chromosome number of seven to 10 (Imai, 1969), we believe that the data for the species analyzed here support the minimum-interaction hypothesis of Imai *et al.* (1986, 1988) and that the present karyotypes constitutions are the result of centric fissions and of later heterochromatin growth and pericentric inversions.

When the karyotypes of *T. asuncicola* and *T. sp1* (Figures 1, 2a and 2b) are compared, it can be seen that



Figure 1 - Karyotype of *Trypoxylon (Trypoxylon) sp1*. Standard staining of a female (2n=32). (Bar = 5 µm).

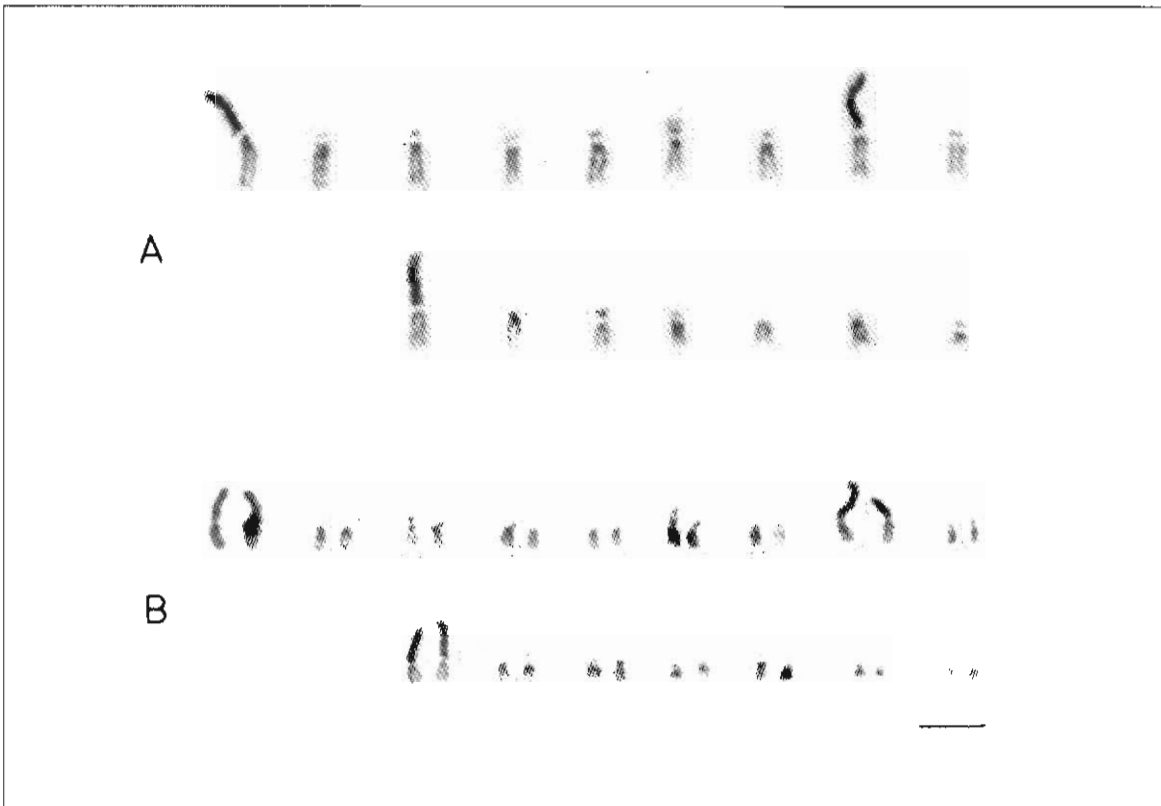


Figure 2 - Karyotype of *Trypoxylon (Trypoxylon) asuncicola*. a) C-band of a male (n=16). b) Standard staining of a female. (Bar = 5 μ m).

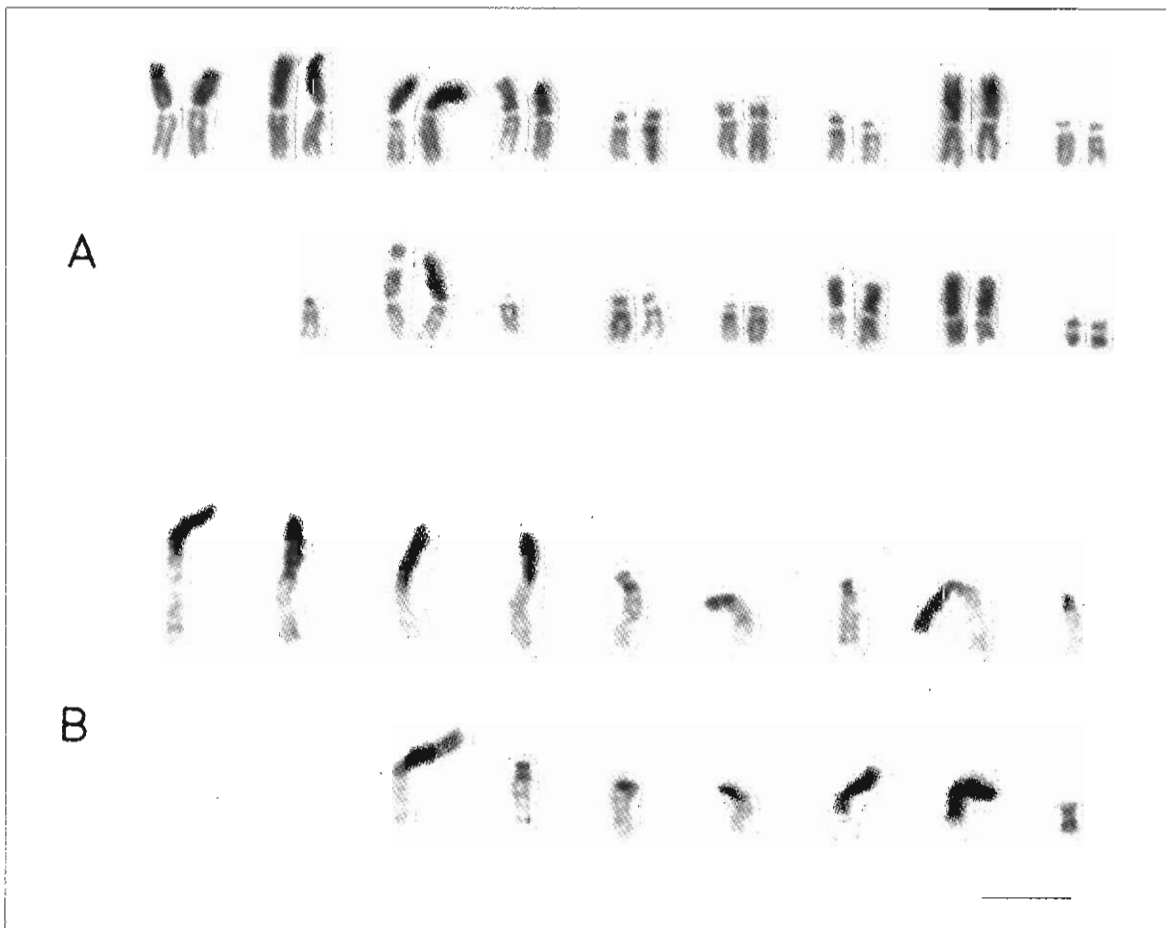


Figure 3 - Karyotype of *Trypoxylon (Trypoxylon) fabricator*. a) C-band of a male (n=16). b) Standard staining of a female. (Bar = 5 μ m).

these are quite close species and that the difference between them consists of a variation in centromere position in some smaller chromosomes, resulting in different arm numbers. The mechanism responsible for this karyotypic variation may be rearrangements of the pericentric inversion type. Another difference is the presence of a heteromorphic pair (pair 8) in all female *T. asuncicola* specimens, and the presence of a lighter interstitial region in the heterochromatic arm of one of the homologues in pair 10 in *T. sp1*. This structure was also detected in *T. fabricator* (Figure 3a and b) and may correspond to the nucleolar organizer region.

The heteromorphism of pair 8 in *T. asuncicola* (Figure 2b) may be due to translocation or duplication of the long arm in one homologue.

The karyotype of *T. fabricator* differs from that of the above species by presenting a larger number of two-armed chromosomes (M-SM), or pseudoacrocentric chromosomes (A^M) in the nomenclature proposed by Imai et al. (1988), due to tandem growth of constitutive heterochromatin in one arm.

The presence of this heterochromatin further supports the minimum-interaction hypothesis, according to which these karyotypes are indeed derived from an ancestral type with a small number of chromosomes, mainly by centric fission. This hypothesis assumes that the telocentric chromosomes that originated by centric fission present telomeric instability and that tandem growth of constitutive heterochromatin transforms these telocentrics into acrocentrics or pseudoacrocentrics, thus reestablishing telomeric stability (Imai et al., 1986, 1988).

Comparison of these data with those obtained for the four species studied by Hoshihara and Imai (1993) shows that the number of chromosomes with one heterochromatic arm increases with an increase in the number of chromosomes, a fact that, according to these investigators, strongly indicates a high telomeric instability of telocentric chromosomes in Hymenoptera.

RESUMO

Neste trabalho são descritos os cariótipos de três espécies de vespas do gênero *Trypoxylon* (*Trypoxylon*): *T. sp1* ($2n = 32$, $2K = 24A + 8SM$), *T. asuncicola* ($2n = 32$, $2K = 22A + 9SM + 1M$) and *T. fabricator* ($2n = 32$, $2K = 12A + 18SM + 2M$). Crescimento em tandem de heterocromatina constitutiva e inversões pericêntricas após a ocorrência de fissão cêntrica são discutidos como os principais mecanismos para a diferenciação entre os cariótipos das três espécies.

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