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Chromosomal polymorphisms involving telomere fusion, centromeric inactivation and centromere shift in the ant *Myrmecia (pilosula) n = 1*

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Abstract. Detailed karyological surveys of the ant *Myrmecia pilosula* species group, which is characterized by the lowest chromosome number in higher organisms ($2n=2$), were attempted. We revealed that this species has developed highly complicated chromosomal polymorphisms. Their chromosome numbers are in the range $2n=2$, 3, and 4, and six polymorphic chromosomes are involved, i.e., two for chromosome 1 (denoted as SM_1 and ST_1), three for chromosome 2 (A_2, A_2', and M_2), and $M_{(1+2)}$ for the $2n=2$ karyotype. We suggested that these chromosomes were induced from a pseudo-acrocentric (A_1^M) and A_2 as follows: (1) $A_1^M \rightarrow SM_1$ or ST_1 by two independent pericentric inversions; (2) $A_2 \rightarrow A_2' \rightarrow M_2$ by chromosomal gap insertion and centromere shift; and (3) $ST_1 + A_2 \rightarrow M_{(1+2)}$ by telomere fusion, where (3) means that the $2n=2$ karyotype was derived secondarily from a $2n=4$ karyotype. It is a noteworthy finding that active nucleolus organizer (NOR) sites, in terms of silver staining, are tightly linked with the centromere in this species, and that both the centromere and NOR of A_2 were inactivated after the telomere fusion.

Introduction

The ant *Myrmecia pilosula* (Fr. Smith) is one of the most primitive formicids in Australia, and is commonly called the 'Jack-jumper', due to its hopping behaviour. Although this species has long been considered as a single biological species (Clark 1954; Brown 1953), we found several years ago that it is a cytologically heterogeneous species, involving $2n=9$, 10, 31 and 32 chromosome forms (Imai et al. 1977). More recently, a unique '*pilosula*' colony having the lowest chromosome number in higher organisms ($n=1$ in males and $2n=2$ in workers) has been discovered by Crosland and Crozier (1986).

Encouraged by these findings, we have started an international cooperative programme aimed at a karyological and taxonomic survey of *M. pilosula*. Large-scale field studies were carried out in 1985 and 1987. During these surveys more than 150 '*pilosula*' colonies were collected from New South Wales, the Australian Capital Territory (A C T), Victoria, South Australia, and Tasmania.

It was revealed by these surveys that the chromosome numbers of *M. pilosula* range more divergently than previously thought, i.e., $2n=2$, 9, 10, 15, 17–32 (Crosland et al. 1988; Imai et al. 1988a). As there are some significant but subtle morphological variations as well as karyological differentiation, it is now clear that *M. pilosula* is a complex species including at least five separate but similar biological species. Details will be published elsewhere. We deal here with chromosomal polymorphisms in the species currently denoted *M. (pilosula) n = 1*, and discuss the origin of the karyotype with the lowest chromosome number in higher organisms ($n=1$).

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Materials and methods

The materials used here were named temporarily *Myrmecia (pilosula) n = 1* by R.W.T. (Imai et al. 1988a). *M. (pilosula) n = 1* is distinguished from other members of the *pilosula* group by its stocky form and details of cuticular sculpture and pilosity. It builds a small flat nest mound, with no pebbles on the mound. A total of 11 colonies have been collected from Canberra (A C T) (HI87-165, 235), the Mongarlowe/Charleyong district (N S W) (HI87-136, 148, 150, 151, 153, 154, 157), South Warrandyte (Vic.) (HI87-213), and Tidbinbilla (A C T), where HI87 is the code number (H. Imai, 1987). The Tidbinbilla colony was collected by M. Crosland in 1985. It was later sent to Japan and has been subsequently cultured by H.T.I. The colonies HI87-151, 165, and 235 are also being bred by H.T.I. at the National Institute of Genetics for C-banding and nucleolus organizer region (NOR) analysis.

Chromosomal preparations were made using cerebral ganglia of prepupa by the improved air-drying technique (for details see Imai et al. 1988a). The preparations were stained with Giemsa (3% in M/15 Sørensen's pH 6.8 phosphate buffer) for 10 min at room temperature. For the NOR silver staining, we followed the method of Howell and Black (1980). A brief outline of the technique is as follows: Two chemicals are used: (A), a 50% solution of $AgNO_3$, and (B), a gelatin solution with formic acid (50 ml distilled water: 1 g gelatin: 0.5 ml formic acid). Both solutions should be prepared freshly and filtered through a 0.22 μm Millipore filter. After observing the chromosomes first by Giemsa staining, the preparation is put into absolute glacial acetic acid for 10 min to remove the Giemsa completely. Six drops of solution (B) are added to the preparation with a 1 ml disposable syringe and then 3 drops of solution (A). Both solutions are mixed completely by flipping the slides, and covered with a fine nylon mesh