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Cytotaxonomical study of Dugesia indonesiana Kawakatsu (Tricladida Paludicola)

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SEZIONE III

(Botanica, zoologia, fisiologia e patologia)

Zoologia. — *Cytotaxonomical study of* Dugesia indonesiana *Kawakatsu* (*Tricladida Paludicola*). Nota di MARIO BENAZZI (*) e NICOLE GOURBAULT (**), presentata (***) dal Socio M. BENAZZI.

RIASSUNTO. — *Dugesia indonesiana* Kawakatsu, 1973 è una planaria d'acqua dolce diffusa nelle isole di Giava e Sumatra, che si riproduce prevalentemente per scissione. In esemplari raccolti nel Giardino Botanico di Bogor (Giava) gli Autori hanno riscontrato un corredo di 24 cromosomi, riferibile ad una condizione triploide, come confermano i dati cariometrici. Gli 8 elementi del corredo aploide presentano le caratteristiche generali proprie del cariotipo comunemente riscontrato nelle specie del gruppo *Dugesia gonocephala* s.l.

INTRODUCTION

In october 1973 one of us (1) collected some asexual specimens of a *Dugesia* of the *gonocephala* group from the Botanical Garden of Bogor (Java, Indonesia). In continuing culture this planarian has multiplied only by fission. Nonetheless, two or three animals showed external signs of sexuality after a few months and one of them reached full maturity. We have morphologically examined that specimen and determined the karyotype of the strain.

We present our results as a new contribution to knowledge of this *Dugesia* group so widely distributed in the Old World.

The first report on planarians from Java is that of de Beauchamp (1929). From the same Botanical Garden he recorded three species that he assigned to *Dugesia* (olim *Planaria*) *gonocephala* (Dugès), *Cura* (olim *Planaria*) *pinguis* (Weiss) and *Planaria* sp., the last two being represented by immature animals only. Recently Kawakatsu (1973) described *Dugesia indonesiana* from various localities in Sumatra and Java, including records of asexual material from the Botanical Garden of Bogor. He tried also to interpret de Beauchamp's data, stressing (p. 100–101) that the occurrence of *D. gonocephala* s. str. in the Far East is very doubtful and that de Beauchamp's *gonocephala* and *Planaria* sp. might be identical with *D. indonesiana*. In Kawakatsu's opinion even the planarian identified as *Cura pinguis* could be referred to his new species, although in a previous article he agreed with the identification of this planarian as a species of *Cura* (Kawakatsu, 1969).

From the previous data we inferred that our specimens could belong to a fissiparous strain of *Dugesia indonesiana*.

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(***) Nella seduta dell'8 febbraio 1975.

(1) N. Gourbault wishes to thank the Director of the Kebun Raya Tjibodas who allowed her to collect some aquatic fauna in different biotopes of the Botanical Garden.

Morphology of the species

1. *External features.*

The external characters leave no doubt that our population belongs to the genus *Dugesia*. The head-shape in particular corresponds closely to that of the *gonocephala* group and, moreover, to the description given for *D. indonesiana*. We have compared our living material with live *gonocephala* s.l. from several localities (Italy, Israel, Algeria) and found that the eyes are closest together in the Bogor population and slightly anterior to the auricles. The general colour of the body is dark brown.

Most of the *indonesiana* strains described by Kawakatsu attain a moderate to large size (15 to 30 mm); some are small (8 to 10 mm). All our specimens are small too; the sexual one measured 8.5×0.9 mm.

2. *Internal features.*

In sagittal section the sexual specimen we examined showed well developed gonads and copulatory organs. The testes, latero-dorsally located, are numerous and extend from the ovaries almost to the end of the body. Complete spermatogenesis can be seen in the largest ones.

The ovaries are situated posteriorly to the brain, after the fifth or sixth intestinal diverticula. They are very large and are represented by two lobes, the anterior one of which occupies nearly all the dorso-ventral space of the body. The ovaries are composed of a great number of oocytes, most of them of small size, with only a few in a more advanced stage of development. These ovarial characters are typical of the hyperplasic ovaries peculiar to ex-fissiparous specimens of *D. gonocephala* s.l. (Benazzi, 1974) and other species (Benazzi and Ball, 1973).

It is noteworthy that Kawakatsu (1973, p. 93) also observed very large ovaries in specimens from Kaliurang (Java), and whereas he did not interpret the meaning of these structures his obsevation demonstrates the general occurrence of the phenomenon in *D. indonesiana*. In fact all the populations from Indonesia can reproduce by fission because numerous regenerated worms were present in Kawakatsu's collections.

The general features of the copulatory complex of our specimen are similar to those described for *D. indonesiana* and therefore it is unnecessary to re-describe these organs.

Karyological data

We have studied the chromosome set of the cells in regenerative blastemas with the method commonly employed in our recent research (colchicine 0.3 % for 3 to 4 hours, then 2 % acetic acid for 5 minutes, lacto-aceto-orcein for 30 minutes, and squash). In all metaphase plates we have found 24 chromosomes.

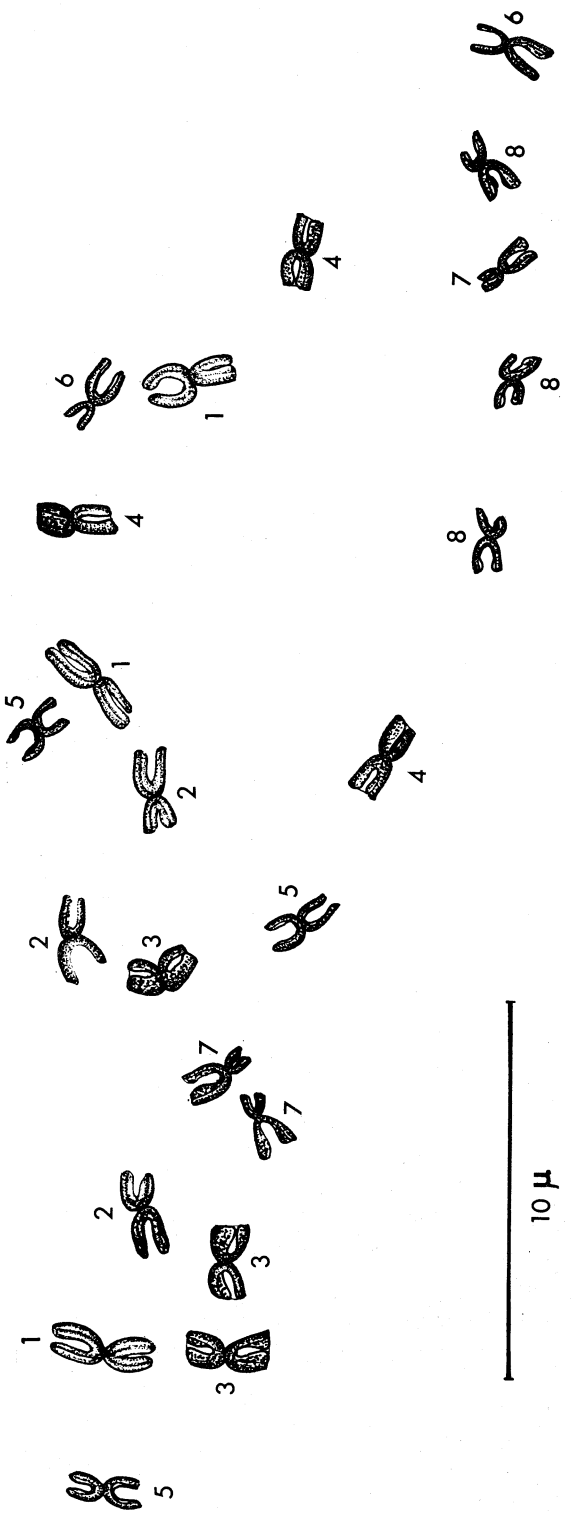

Fig. 1. – Metaphase plate of *Dugesia indonesiana*.

For the *D. gonocephala* species complex it is now well established that the haploid chromosome number is 8 (for a summary see Benazzi and Benazzi Lentati, in press). Therefore, the complement of 24 that we found might be referable to a triploid condition. This viewpoint is already supported by our karyometrical studies for these enable us to recognize 8 groups of three homologous chromosomes (fig. 1). Fig. 2 gives the idiogram of the species and is derived from the means calculated for 10 metaphase plates as shown in Table I. It can be seen that the relative lengths vary gradually from the longest to the shortest chromosome. Furthermore, the values of the centromeric indices are close; all the chromosomes are metacentric according to the nomenclature of Levan *et. al.* (1954). Thus it is impossible to further classify these 8 elements.

TABLE I

Relative length (r.l.) and centromeric index (c.i.) (means and standard deviation calculated for 10 metaphase plates) of the basikaryotype of the triploid Dugesia indonesiana.

r.l.	1	16,22 ± 1,01	5	11,76 ± 0,46
c.i.		45,80 ± 1,45		43,58 ± 2,37
r.l.	2	14,38 ± 0,50	6	11,35 ± 0,38
c.i.		44,92 ± 2,24		42,95 ± 2,57
r.l.	3	13,22 ± 0,25	7	10,66 ± 0,54
c.i.		45,64 ± 1,55		41,90 ± 3,83
r.l.	4	12,66 ± 0,37	8	9,47 ± 0,84
c.i.		43,11 ± 1,86		44,95 ± 3,82

DISCUSSION

From the data obtained from our laboratory culture, and that given in Kawakatsu's article it is apparent that *D. indonesiana* frequently, or in some populations exlusively, multiplies by fission. The few specimens of the fissiparous strains that reach sexual maturity probably are unable to lay fertile cocoons, as has often been observed in the *D. gonocephala* group.

With regard to the karyology of the planarian we studied it is important to compare its complement with those of the allied species. Chromosome researches have been accomplished on species from Europe (Benazzi Lentati *et. al.*, 1951, 1956; Dahm, 1958, 1963; Dutrillaux and Lenicque, 1971), Africa (Dahm, 1967) and Japan (Dahm, 1963; Teshirogi and Itagaki, 1965). From

these studies it appear that the chromosomes of the haploid set show common patterns in all the species, although some differences in the centromere location may be noticed. Unfortunately most of the studies have not been completed with karyometrical data so a fruitful comparison is not possible. We may consider, however, the chromosome measurements made by Dutrillaux and Lenicque (l.c.) on a Hungarian fissiparous race with 16 to 24 chro-

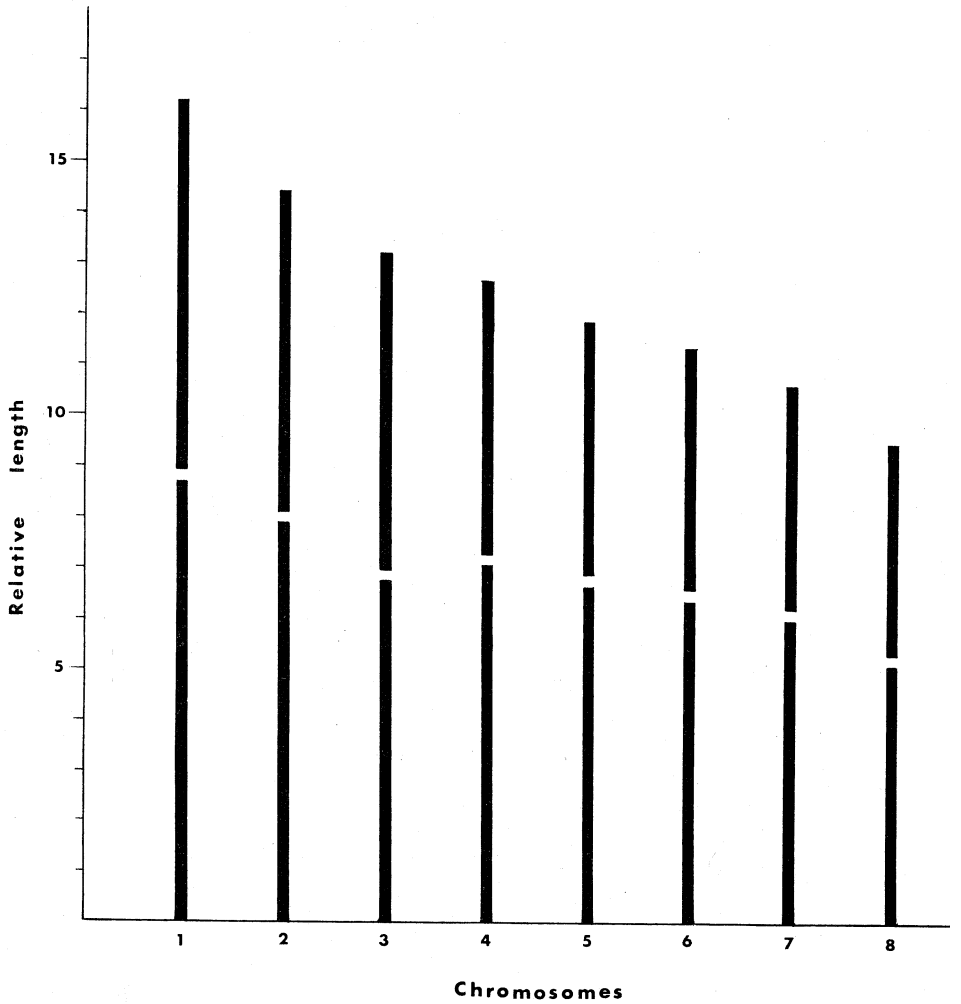

Fig. 2. – Idiogram of the basikaryotype of *Dugesia indonesiana.*

mosomes in somatic cells, and those made by Benazzi *et al.* (unpublished data summarized in Benazzi *et al.*, 1972), in three sexual, diploid populations from Germany, Corsica and Corfu. In these studies it was found that the relative length of the chromosomes decreased gradually and rather similarly in the different populations. The largest element was almost twice the shortest. The centromeric indices all belonged in the metacentric class, although some

variations occurred. In the Bogor population these variations are less pronounced and this character may also be found in the karyotype of *D. japonica* in which, according to Teshirogi and Itagaki (l.c.), all elements are metacentric with a centromere more or less medially located.

It is possible to conclude from the new data derived from the Indonesian planarian that in all the specie (or microspecies) of the *D. gonocephala* group hitherto studied the 8 chromosomes of the haploid complement have not displayed marked morphological variation.

A final point to discuss is the triploid condition of the Bogor population. The occurrence of polyploidy or aneuploidy in the *gonocephala* group is well known. In strains reproducing sexually triploidy is connected with special chromosome cycles (for a summary cf. Benazzi Lentati, 1970 and Benazzi and Benazzi Lentati, in press). In fissiparous populations aneuploidy or heteroploidy, with variable and often high chromosome numbers, are the most frequent conditions. The eutriploid condition of the fissiparous Bogor population therefore is an interesting occurrence.

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