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Anuran Haemoprotozoa Found in the Vicinity of Nagasaki City

2. *Dactylosoma ranarum* (Kruse, 1890)

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ABSTRACT : During a period of July to August, 1974, blood examinations of frogs and tadpoles collected from Mogi, near Nagasaki City, were carried out. *Trypanosoma rotatorium* (Mayer, 1843) was detected from the blood smears as reported in a previous paper (Miyata, 1976). From the smears at the same time, *Dactylosoma ranarum* (Kruse, 1890) was also found, and the following observations were obtained : 1) *D. ranarum* was found only from adult of *Rana rugosa* (larger than 4 cm in body size), and 8 out of 21 frogs had this parasite in their blood. 2) From all smears in which *D. ranarum* was detected, *T. rotatorium* was found except one case. 3) Two types of schizogony were recognized. In type A, 3∼10 nuclei were seen in the margin of round schizonts. In type B, fan-shaped schizonts were also seen in erythrocytes of frogs and in this case 3∼6 nuclei appeared at margin of broader end of the schizont. Gametocytes are slender and elongate forms. 4) No vector or mechanism of transmission was known.

The genus *Dactylosoma* Labbé, 1894, (type species: *D. ranarum* (Kruse, 1890) = *D. splendens* Labbé, 1894) was reported from the blood of amphibia, reptile, and fish, and at least 6 species of this geuns have been described. Among them, *D. ranarum* is a widely distributed and well-known parasite in the erythrocyte of frogs. *D. amonia* (Awerinzew, 1914) is described from the blood of *Chameleon fischeri* in West Africa. *D. salvelini* Fantham, Porter, and Richardson, 1942, is a parasite of *Salvelinus fontinalis* in Canada. *D. sylvatica* Fantham, Porter, and Richardson, 1942, is described from the blood of *Rana sylvatica* in Canada, however this species might be a synonym of *D. ranarum*. *D. taiwanensis* Manwell, 1964, is reported from *Rana limnocharis* in Taiwan. This species differs from *D. ranarum* in "being larger, and having a generally coarser appearence". The materials used by the present author is apparently resemble to this species. However, the author identified the present materials as *D. ranarum* tentatively, because he believes that those features emphasized hy Manwell are overlapping and not enough to raise a new species.

Fantham, Porter and Richardson (1942) described two species of malarial parasites

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(genus *Plasmodium*) from the blood of frogs and toads captured in Canada. According to their description, they observed pigment in their parasites. Actually *Dactylosoma* is similar to *Plasmodium* in its schizogony in the erythrocytes, but *Dactylosoma* has no pigment. Their malarial parasites are not yet confirmed by other researchers, but it is possible that they misidentified their parasites as *Plasmodium*, although they reported *Dactylosoma* spp. from other hosts in the same paper.

At present, any imformation concering the life cycle of the genus *Dactylosoma* is not known, and before going to describe many new species, the life cycle of the genus should be intensely studied.

In the first paper concerning anuran haemoparasites (Miyata, 1976), the author had reported the morphology of *Trypanosoma rotatorium* (Mayer, 1843), and in this paper several observations on *D. ranarum* will be described below.

MATERIALS AND METHODS

The materials and methods used were just the same as those described in the first paper of this series (Miyata, 1976).

RESULTS

The results of the blood examination were summarized in Tables 1 and 2 of the first paper (Miyata, 1976). *Dactylosoma ranarum* (Kruse, 1890) was found out in the erythrocytes of 8 out of 21 frogs of *Rana rugosa* Schlegel, which were captured in Mogi district, near Nagasaki City, during a period of July to August, 1974. The parasite was detected from larger frogs than 4 cm in size, but *D. ranarum* was never detected from smaller frogs than 4 cm or the tadpoles. From all the blood smears in which *D. ranarum* was detected, *Trypanosoma rotatorium* was also found except in one case (frog no. 1974−19, see Miyata, 1976).

Trophozoites: The youngest stages are rearly seen in the host erythrocytes as shown in Fig. 1, a and b. In more growing trophozoites, their sizes become larger and several vacuoles are usually observed in their cytoplasm (Fig. 1, c～h).

Schizonts and types of schizogony: Two types of schizogony could be distinguished morphologically.

Type A: In the youngest schizont, two chromatin dots situate at each end of round or elongate schizont (Fig. 1, i～k). In more advanced schizonts, 3 to 10 nuclei are seen in peripheral parts of round schizonts. The diameter of the largest schizont is 8μ, and 10 chromatin dots are observed. In this case (Fig. 1, q), the schizont appears to be in progress of further development, because it does not show any feature of splitting of the body.

Type B: Fan-shaped schizonts are also seen in erythrocytes, and in this case 3 to 6 chromatin dots appear at the end of broader part of the schizont. In this type, splitting of

the body is apparently observed as shown in Fig. 1, s~w. In this type and type A, neither residual body nor malarial pigment was observed.

Gametocytes : So-called gametocytes are slender, elongated forms, containing a chromatin dot and sometimes a few dark bodies and vacuoles (Fig. 2, a~i). The dark bodies are different from malarial pigment in its colour. Gametocytes are usually seen in the host erythrocytes, but sometimes slender forms which are stained darkly with Giemsa are detected outside of the erythrocytes (Fig. 2, j~l).

Tissues or organs of the frogs examined extensively, but any other stages of the parasite were not found.

DISCUSSION

Tanabe (1931) described *Dactylosoma ranarum* (Kruse, 1890) from *Rana nigromaculata* collected in Korea, and he observed three types of schizogony in the blood of frogs as shown in Fig. 3. His types Ⅰ and Ⅱ accord to types A and B in the present paper, respectively. However, Tanabe's type Ⅲ was not seen by the present author. According to Tanabe, his type Ⅲ is very much similar to that of *Dactylosoma mariae* Hoare, 1930, described from a fish (*Haplochromis* sp.) in the Lake Victoria, Africa. *D. mariae* is now transfered to the geuns *Babesiosoma* Jakowska and Nigrelli, 1956. The initial schizont is similar to the type Ⅰ, but with development, the body increases in length as well as width, and nuclear division occures by binary fission. The two nuclei arrange respectively at either end of an enlarged body (Fig. 3, k). The nucleus at each end divides as shown in Fig. 3, m ~ o. The nuclear division does not occure at the same time at each end, then different number of nuclei are seen. In this type, seven-nucleate schizonts are the most advanced ones among schizonts observed by Tanabe. In the type Ⅰ (type A in the present paper), 16 merozoites might be produced at their maximum. In the type Ⅱ (type B in the present paper), Tanabe also believed that 16 merozoites as a maximum may be produced at the end of the schizogony. According to the present author's observation, only 3 to 6 chromatin dots were counted, but 3 to 4 times of nuclear division (8 to 16 merozoites) might occure in this type.

At present, the fate of schizonts of type Ⅰ is not known, but if only one type of schizogony is really present, the following explanation might be thought. Before splitting of the body, nuclei might move to one end of schizont, then merozoites might be produced at the point. This tentative explanation will be supported by the presence of the same maximum number of nuclei in both types of schizogony. Tanabe's type Ⅲ seems to be apparently an intermediate form between type Ⅰ and type Ⅱ.

In other explanation, mature schizonts of type Ⅰ disappear from peripheral blood or heart blood before splitting the body, and merozoites might be produced in other organ of tissues. The present author examined several organs such as liver, spleen, and lung, however, any imformation concerning this matter could not be obtained.

Vector or mechanism of transmission of *Dactylosoma* is not known. Nöller (1913) examined the possibility of transmission by the leech, *Hemiclepsis marginata*, but he could

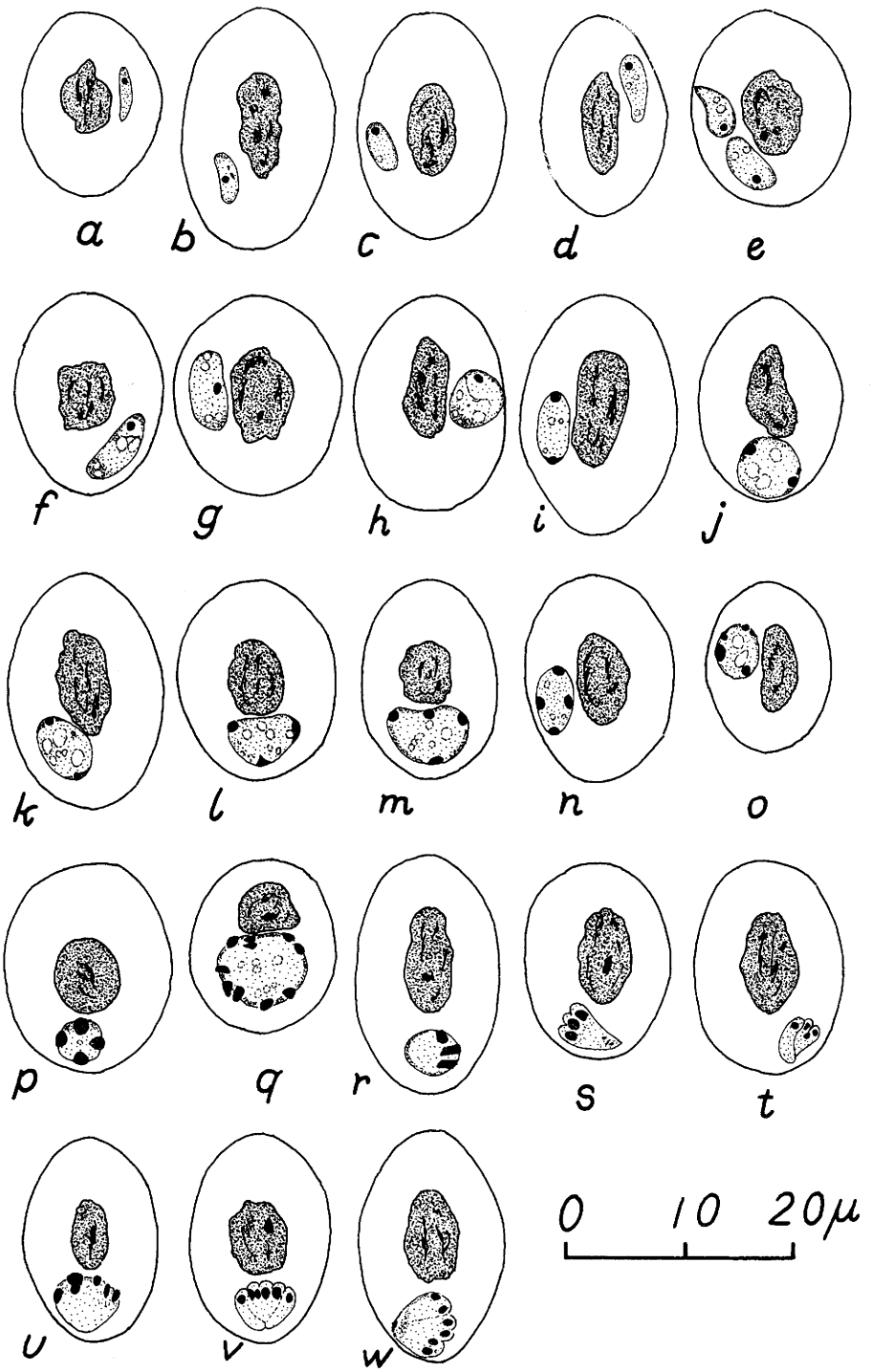

Fig. 1 Trophozoites (a~h) and schizonts (i~w) of *Dactylosoma ranarum*.

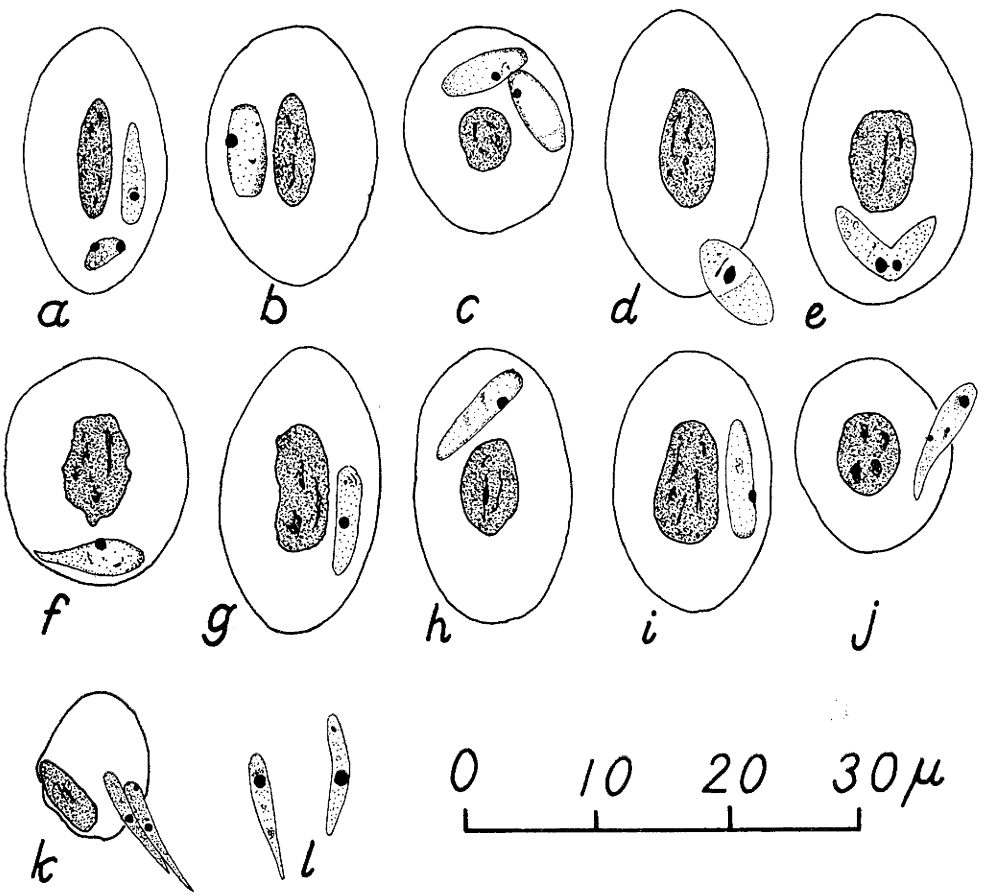

Fig. 2 Gametocytes of *Dactylosoma ranarum*.

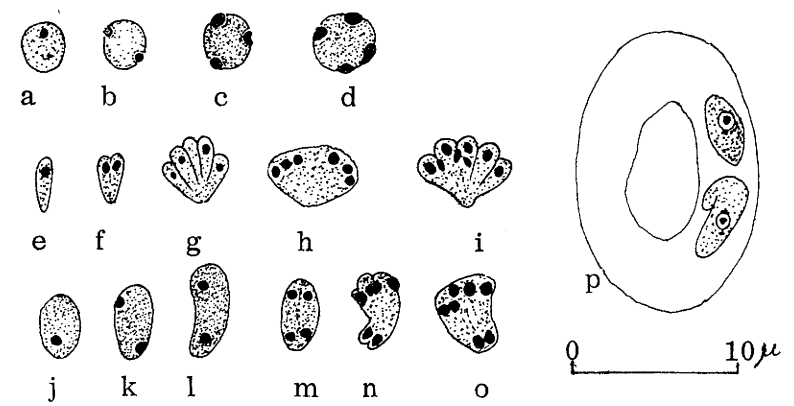

Fig. 3 Three types of schizogony in *Dactylosoma ranarum*.
(a~o. Red cells are not presented. modified after Tanabe, 1931)

not success to show any evidence. He expressed his opinion that "fish louse" *Argulus foliaceus* is a probable vector without showing any clear reason. In the present study, *D. ranarum* was never found from tadpoles or young frogs. Moreover, in most cases where *D. ranarum* was detected, *Trypanosoma rotatorium* was also observed. The leech is still suspicious as the possible vector, since it is known as a vector of *T. rotatorium* (see Miyta, 1976). But, if so, it is difficult to explain why young frogs and tadpoles did not infect with *D. ranarum*. Much more studies on the life cycle of *D. ranarum* will be needed.

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長崎市郊外で発見された無尾類の住血原虫　2．*Dactylosoma ranarum*（Kruse，1890）

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前報（Miyata，1976）に続き，この論文では，長崎市郊外茂木のツチガエル（*Rana rugosa*）の赤血球内から検出された *Dactylosoma ranarum*（Kruse，1890）について報告する．*Dactylosoma* 属はピロプロズマと近縁であると考える学者もあるが，反面マラリア原虫（*Plasmodium* 属）と赤血球内における無性生殖の仕方が類似しており，ただマラリア色素をもたない点が区別点として強調されている．従ってこの原虫の研究はマラリア原虫の起源を解明する上で大変重要な意味をもっている．得られた成績は次の通りである．1）*D. ranarum* は体長（鼻端より肛門まで）4 cm 以上のツチガエル成体からのみ検出された．21匹のツチガエルの成体のうち8匹から *D. ranarum* が検出された．しかし 4 cm 以下のツチガエルやそのオタマジャクシからは検出されず，また検査した5匹のトノサマガエル（*Rana nigromaculata*）からも *D. ranarum* は発見されなかった．3）次の2型のシゾゴニイをみとめることができた．A型：シゾントはまるく，その周縁に3～10コの核が並んでいる．B型：扇形のシゾントで，その広くなっている側の縁に3～6コの核が並んでいる．また生殖母体ではないかと考えられる虫体は細長く，時には赤血球外に出ているのが観察される．4）*D. ranarum* が検出されたツチガエル8匹のうち，1匹を除き，その血液内に *Trypanosoma rotatorium*（Mayer，1843）が寄生しているのが観察された．この両原虫の媒介者は，あるいは共通しているのではないかと疑われるが，*D. ranarum* の媒介者についても全く何も判明しなかった．

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