THE SECOND RECORD OF OCCURRENCE OF RHODAX? SP. IN TANKS OF TROPICAL FISHES IN JAPAN (TURBELLARIA, SERIATA, TRICLADIDA, CAVERNICOLA)

MASAHARU KAWAKATSU, SACHIKO TAMURA, IWASHIRO OKI, MASAYUKI TAKAI, and EUDOXIA MARIA FROEHLICH

INTRODUCTION: Historical Background to the Taxonomy

In 1946, MARCUS (pp. 133-142, 171-173, 236-241, pls. XXIV-XXVI, figs. 144-153, p. 250, pl. XXXI, fig. 5) reported a small and slender species of freshwater planarian under the name of *Rhodax evelinae*. This new species, collected from the vicinity of São Paulo, Brazil, was placed in a new paludicolen genus of the family Planariidae STIMPSON, 1857. MARCUS' original description cited above includes morphology, anatomy and histology of sexual specimens with sufficient figures. For many years, the species was known only from the type locality.

In July of 1982, a number of small, non-sexual specimens of planarian were collected from tanks for tropical fish culture (water temp., $22-26\,^{\circ}$ C) in the Biological Laboratory, Faculty of Science, Nagoya University, Nagoya, Honshû, Japan. This animal was reported as *Rhodax*? sp.; its chromosome numbers and karyotypes were studied by KAWAKATSU's team (cf. KAWAKATSU, OKI, TAMURA & YAMAYOSHI, 1985, pp. 2, 13–17, table 1, figs. 7 A–J, 8 C–D′, 9 C–E). Two different karyotypes were found in animals examined: 1) 3x = 24; 2) 3x = 24 & 3x + 1LB + 1SB = 25 + 1SB. These authors considered that Rhodax? sp. might have been imported with an Amazon sword plant, one of the most common aquatic plants occurring in tropical fish tanks in Japan.

SLUYS (1990), who studied the type material of *Rhodax evelinae*, gave a detailed taxonomic redescription of the species (pp. 14–16, figs. 1 and 2). And, he placed the genus *Rhodax* MARCUS, 1946, and the 3 other genera — *Opisthobursa* BENAZZI, 1972, *Balliania* GOURBAULT, 1978, and *Mitchellia* KAWAKATSU et CHAPMAN, 1983 — in the family Dimarcusidae MITCHELL et KAWAKATSU, 1972 (see also KAWAKATSU & MITCHELL, 1983, 1984 a, b). In his paper, SLUYS (*op. cit.*, p. 28) wrote as follows:

"...there is a fourth major lineage within the Tricladida which should be given the same categorical rank as the three lineages previously recognized. I propose to use the name Cavernicola, which has ecological roots but no such connotation, for the lineage from which at present we only know the genera *Rhodax*, *Opisthobursa*, *Balliania* and *Mitchellia* of the family Dimarcusidae".

In early September, 1993, KAWAKATSU was informed by FROEHLICH (in litt., São Paulo, Aug. 30) of the occurrence of Rhodax evelinae at a site other than the type locality. According to her information, several live specimens of this species, collected from an artificial lake located in Município Americana, Estado de São Paulo (approximately 135 km NW of São Paulo City), were brought to her

laboratory by Miss Regina Sawaya SÁFADI, a Master Course student of the Instituto de Biociências. These planarian specimens were obtained by cleaning roots of waterhyacinths (*Eichhornia crassipes*) collected from the lake. Unfortunately, all of the live specimens died within a few days.

Although FROEHLICH knows live specimens of *Rhodax evelinae* from the type locality (*i.e.*, São Paulo population), the discovery of the Americana population mentioned above provided her a second chance to see live specimens of this species. Tentative identification of the species was made, based upon external morphology alone. No photographs of live specimens were prepared. Their karyotypes were not studied. Additionally, the type locality of this species was lost in 1960–1970 due to the great expansion and urbanization of the city of São Paulo.

On October 3, 1993, Miss Miyuki and Mrs. Kazuko KAWAKATSU (KAWAKATSU's daughter and wife) found many planarian specimens creeping on the inside surface of the glass wall of a tropical fish tank at a local tropical fish store, "The Fish Gallery — Sakana-no-Yakata", in the northeastern part of Sapporo. They brought several live specimens of this planarian to KAWAKATSU. Within a week, KAWAKATSU succeeded in collecting over 30 live specimens at "Sakana-no-Yakata" for taxonomic study.

Externally, the Sapporo specimens agree very well with previous descriptions and photographs of *Rhodax*? sp. from the Nagoya population (cf. KAWAKATSU, OKI, TAMURA & YAMAYOSHI, 1985). There were no sexual specimens in this Sapporo population. Photographs of live specimens were prepared in KAWAKATSU's laboratory. About 10 specimens were sent to TAMURA (Ôsaka in Honshû) for chromosomal study. An additional 10 specimens were sent for culturing to TAKAI (Saga in Kyûshû) following his method of breeding of planarians (cf. TAKAI, 1989). The remaining specimens were placed in a tropical fish aquarium in Miss KAWAKATSU's room at home (Sapporo in Hokkaidô). Unfortunately, animals of both TAKAI's and KAWAKATSU's cultures disappeared within a month.

The karyological data obtained by TAMURA showed that the karyotypes of animals from the Sapporo population agree with those of animals from the Nagoya population of *Rhodax*? sp. (cf. KAWAKATSU, OKI, TAMURA & YAMAYOSHI, 1985). TAMURA's draft of idiograms was examined carefully and confirmed by OKI.

FROEHLICH, who received a complete set of the data on *Rhodax*? sp. of the Nagoya and the Sapporo populations from KAWAKATSU, has studied the comparative morphology of animals found in Japan (based upon KAWAKATSU's photographs, sketches and observation notes in *litt*.) and MARCUS' (1946) original description and figures of *Rhodax evelinae* (including her memory of external morphology of animals). She concluded that specimens of *Rhodax*? sp. from Japan are very similar to those of *Rhodax evelinae*.

The second record of occurrence of *Rhodax*? sp. in Japan has been listed in the 1993 Åbo Symposium preprint, abstract and the 1995 publication: The Biology of Turbellaria and Some Related Flatworms (cf. KAWAKATSU, OKI, TAMURA, TAKAI, TIMOSHKIN & PORFIRJEVA, 1993, p. 2, fig. 1; OKI, TAMURA, TAKAI & KAWAKATSU, 1993, p. 91, 1995, pp. 71–74).

The purpose of the present paper is to provide photographs of live specimens of Rhodax? sp. from the Sapporo population and their idiograms, together with an additional discussion on the possible accidental distribution route of Brazilian freshwater planarian species into Japan.

MATERIALS AND METHODS

KAWAKATSU's Specimen Lot No. 2013 was given to the Sapporo specimens of Rhodax?

sp. used in the present study (see the above section). Chromosomes were observed according to the technique described in the previous papers (cf. OKI, TAMURA & KAWAKATSU, 1976; OKI, TAMURA,

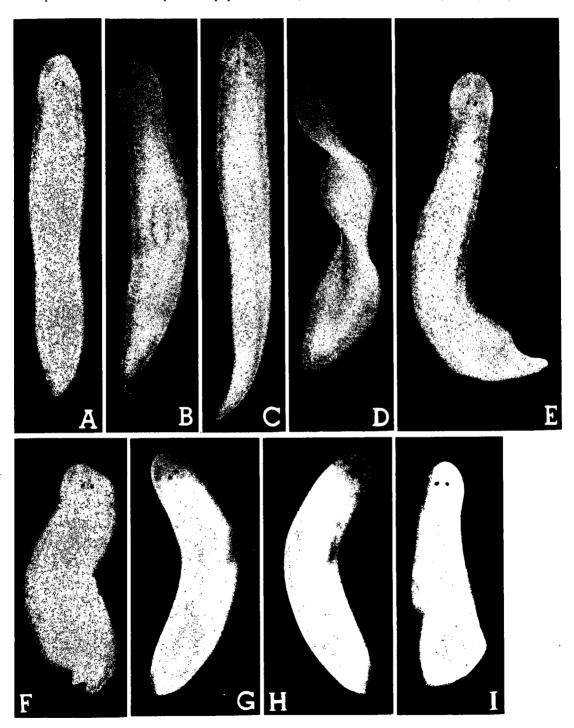


Fig. 1. Photographs of Rhodax? sp. from the Sapporo population (Specimen Lot No. 2013). A-F: Two live specimens. G-I: Two preserved specimens. G, dorsal view; H, ventral view.

YAMAYOSHI & KAWAKATSU, 1980, p. 4, fig. 4; KAWAKATSU, OKI, TAMURA, OGREN, YAMADA & MURAYAMA, 1990, p. 10, fig. 20; OKI, TAMURA, OGREN & KAWAKATSU, 1991, pp. 164-165, fig. 1).

DESCRITPTION

Order SERIATA BRESSLAU, 1933
Suborder TRICLADIDA LANG, 1884
Infraorder CAVERNICOLA SLUYS, 1990
Family D i m a r c u s i d a e MITCHELL et KAWAKATSU, 1972
Genus Rhodax MARCUS, 1946
Rhodax? sp.

External features. Photographs of live specimens of Rhodax? sp. from the Sapporo population are shown in Fig. 1 (A-E). Non-sexual specimens in life measure 3-5 mm in length and 0.4-0.5 mm in width. The head is of a rounded shape with a gentle swelling on either side (a pair of non-conspicuous auricular sense organ). It was observed in creeping animals that the anterior end formed a slight obtuse protrusion. Behind the head, the body reaches its greatest width at the level of the short pharynx, which is inserted anterior to the middle of the body. The posterior end of the body is bluntly pointed (Fig. 1 A-E).

A slightly thick adhesive region was observed at the antero-ventral end of the body of the Sapporo specimens. In live specimens, this organ is recognizable as a small swelling. Its histological description, based upon the animal from the Nagoya population, was given in a previous paper (cf. KAWAKATSU, OKI, TAMURA & YAMAYOSHI, 1985, p. 14; see also MARCUS, 1946, pp. 236-237, fig. 144, z, *i.e.*, "área adesiva").

The two eyes are situated on the dorsal side of the head at a somewhat posterior level. The distance between them is about one-sixth of the head width at the level of the eyes (Fig. 1 A-E).

The bodies of small specimens are translucent or milky white. Somewhat larger specimens show a pale brown coloration on the dorsal surface of the body; small brownish pigments occur at the pharyngeal region. The shape of both anterior and posterior intestinal trunks can be seen through the body surface (Fig. 1 A–E).

Karyological observation. Idiograms of the animal examined from the Sapporo population are shown in Fig. 2 (A and B). The specimen has two different types of cells intermingled in one body: triploid cells and triploidic aneuploidy cells (3x = 24 & 3x + 1LB + 1SB = 25 + 1SB) (Fig. 2 A and B). The triploidic karyotype consists of 4 sets of metacentric chromosomes (labeled Nos. 1, 2, 3, and 6 in the idiograms) and 4 sets of submetacentric chromosomes (labeled Nos. 4, 5, 7, and 8) (Fig. 2 A and B). The large B-chromosomes (LB) is a submetacentric (Fig. 2 B).

The karyotypes of the Sapporo specimen reported in the present paper agree with those of the Nagoya population (cf. KAWAKATSU, OKI, TAMURA & YAMAYOSHI, 1985, p. 15, fig. 8 C-D ').

Remarks on accidental distribution. This is a continuation of the "Taxonomic remarks" for Rhodax? sp. of the Nagoya population (cf. KAWAKATSU, OKI, TAMURA & YAMAYOSHI, 1985, pp. 15-17).

The discovery of the Nagoya population was some 13 years ago (see the "Introduction"). According to tropical fish catalogs issued in Japan, importations of various kinds of tropical fishes and water-plants from abroad — especially, Hong Kong, Singapore, Australia, Europe (Germany and The Netherlands), South and East Africa, North America, and South America (Peru, Brazil) — have greatly increased now in Japan. Fig. 3 (left) shows a copy of the Varig Air Cargo invoice for a direct



Fig. 2. Idiograms of *Rhodax*? sp. from the Sapporo population. A: 3x=24. B: 3x+1LB+1SB=25+1SB. A and B were found in the body of a single specimen.

import of tropical fishes from Peru (Lima) to Japan (Tôkyô). This cargo arrives in less than 15 hours. Fig. 3 (right) shows a copy of a domestic air cargo from the consignee in Tôkyô (Japan Pet Fish Trading Co., Ltd.) to the Fish Gallery in Sapporo (*Rhodax*? sp. was found in this shop).

According to the recent article printed in The Asahi Newspaper (Asahi-Shinbun) (Fig. 4), the reporter who stayed at Manaus in Brazil stated that the number of specimens of tropical fishes collected annually in the Estado de Amazonas for commercial dealings may reach over 42 million (approximately 60 species). The lost specimens in the transportation from their localities (Barcelos on the Rio Negro for Cardinal Tetra, etc.) to Manaus also reach huge numbers (over 80 per cent of the whole!). Suvivors will be exported to the United States, Japan, etc. Air cargo from Manaus to Tôkyô via São Paulo requires over 30 hours.

The artificial migration of small, aquatic, invertebrate animals is probably an everyday event.

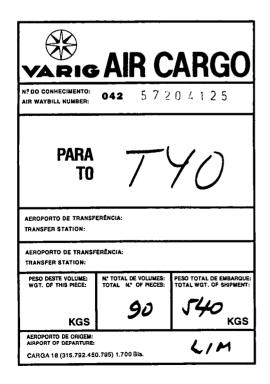




Fig. 3. Invoices of the Varig Air Cargo (left) and a domestic air cargo (right) for importation and transportation of tropical fishes. For other explanations, see text.

(伊刊)

强烈

恐れがある魚の全面禁漁に踏み切ること ったり……。原因は、日本など先進国で 魚は小ぶりのものばかり目立つようにな の数が激減したり、二片を超すはずの大 だ。ブラジル政府はとの秋から、絶滅の の熱帯魚ブームを受けた乱獲にあるよう で生態系に異変が起きている。特定の魚

(マナウスヘブラジル)=磯村健太郎 熱帯魚の宝庫であるアマゾン奥地の川

カージナルといろ小さな鶫 レラン課長は話した。 以内に絶滅するだろう」と

師が約二千人いる。 捕まえ

IBAMAの報告では、

合め、ピラルクー派を禁じ

期間は取りあえず五年 「そうしなければ十年

その何倍にも上る。

が、実際に捕獲されるのは

グロ川沿いの町バルセロス

カージナル流が盛んなネ

水が汚れると死ぬ確率が高 て、酸素を詰めたビニール

段が付いたこともある。ア メスのつがいで百万円の値 のような形の魚は、オスと

マゾナス州ではディスカス

袋に小分けされる。フンで

には、マナウスの輸出業者

らエサは与えない。主要な くなるので、二日ほど前か

のは減ってしまった。 たが、最近これほどの大も ・ニーニ・五がとされてき は、淡水魚としては世界最 が生息する。成魚は体長二 大級のピラルクーといら魚 アマゾン川やその源流に ブラジル政府の環境・資 ブラジル めったにいない」。 実態調査をしている。アマ 五年前の十分の一以下に減 ゾナス州を聞べてきたジョ 各州に支部を置き、河川の 国外へ延び出す大掛かりな 音できる軽飛行機を使い、 漁が横行している。 川で発 ピラルクーの稚魚を狙う密 ンピアとの国境近くでは、 あり、輸出もされる。コロ の愛好者や水族館に人気が た。しかし、この魚は世界 ら取るのは認められてき べるので、一・五紅以上な 「確認できるのはせいぜい ゼ・レラン管理課長(四)は 顧研究所(IBAMA)は 一・二片級。一・八片級も 関査の結果、漁獲量は十 地元民はピラルクーを食

合、魚を入れる容器には酸

機も付いていない。 索を補う装置も、水温調整

ろ、愛好家の間で人気を呼 ある」と話す業者もいた。 分の二が死んでいたことも

日本がパブル景気のと

カスがすみ替くといった生 た。その結果、本来いるは カスはしばしば川に放され

燃系の乱れが起きている。 ずのない川に別種のディス

最近では「ピラニアの出

んだディスカスという円盤

問かかる。 ほとんどの場 スまでは四百世、約十六時 用する答船を使う。マナウ た魚の輸送には地元民が利 と契約した熱帯魚専門の激

経由し、三十時間以上。 本への空輸はサンパウロを けは約三〇%を占める。日 輸出先は米国で、アジア向

異なる。ところがマナウス

ディスカスは生息する川

への輸送中、弱ったディス によって色や模様が微妙に

「日本に行いた時には、三

\$6 の五月から七月は禁漁だ が、効果は上がっていな 帯魚の八割を占め、産卵期 マナウスから輸出される熱 年前の三分の一に減った。 事務所提供=の漁獲量も十 帮魚=写真上、世界自然保 酸基金 (WWF) ブラジル

ウスに替くまでに約八割の

輸出業者によると、マナ

ウスに集中している。同州 熱帯魚が輸出される。だ けで年間約四千二百万匹の からは約六十和、合法分だ 者が十四あり、半数はマナ アマゾナス州には輸出菜 魚が死ぬ。業者は漁師に、 量に取る。 生き残った分しか代金を払

Ħ

が死ぬのを計算に入れて大 わない。だから類師は、魚 マナウスでは輸出に向け

ナウスで、磯村写す

にも応じる。出荷準備 飼っておき、急な注文 ら届けられた熱帯魚を に小分けしている=マ 大手輸出業者は地方か

に豊富に魚をそろえなけれ も往文にとたえられるよう しくなるばかりだ。いつで 大手は「菜者間の競争は激 の業者に届いている。ある 催促が、日本からマナウス 荷鷺を増やしてくれ」との

るかも」と興味をひくアイ 数をする。「珍価が見つか 業者や選好者をあて込んだ 出し売り込む必要もある」。 熱帯魚の原産地に案内し、 団体旅行を企画している。 ばならない。珍種を見つけ 別の業者は、日本の輸入

だ。心をなごませてくれる 熱帯魚たちの受難は続く。 はまだまだ成長する勢い など、日本での熱帯魚産業 デアだ。取った魚を日本へ 水槽ごと熱帯魚を貸す商売 送る手続きも代行する。 銀行や病院、バーなどに

輸出拠点までに8割、昇天、 ▼ 空輸途中にも、昇天、▼ また乱獲

MAは今月中にも、食用を っていた。このためIBA

— 68 —

ACKNOWLEDGEMENT

The authors wish to thank to Dr. Robert W. MITCHELL (Professor Emeritus of Texas Tech University), Bandera, Texas, U. S. A., for his kind reading of a draft of the present paper.

REFERENCES

BENAZZI, M., 1972. Notizie preliminari sulle planarie raccolte nella Seconda Spedizione Lincea in Messico. Att. Acad. Naz. Lincei, Rend. Cl. Sci. Fis., Mat. Nat., (8), 52: 403-405.

BRESSLAU, E., 1933. Turbellaria. In: KÜKENTHAL, W. & T. KRUMBACH (eds.), Handbuch der Zoologie, Bd. 2 (1 Hälfte). Vermes Amera: 52-293. Walter de Gruyter & Co., Berlin und Leipzig.

GOURBAULT, N., 1978. Une nouvelle planaire primitive d'origine marine, Turbellarié Triclade de Polynésie. Cahiers Biol. Mar., 19: 23-36 + pl. 1.

KAWAKATSU, M. & CHAPMAN, P., 1983. *Mitchellia sarawakana* gen. et sp. nov. (Turbellaria, Tricladida, Maricola), a new freshwater planaria from Water Polo Cave, Sarawak, East Malaysia. Jour. Speleol. Soc. Japan, 8: 21–34.

KAWAKATSU, M. & MITCHELL, R. W., 1983. Record of a cave-adapted planarian (Turbellaria, Tricladida, Maricola) from Guatemala. Annot. Zool. Japon., 56: 291–298. 1984a. *Oahuhawaiiana kazukolinda* gen. et sp. nov. (Turbellaria, Tricladida, Maricola), a new freshwater planarian from Honolulu, Oahu Island, Hawaii, U.S.A. Zool. Sci., Tôkyô, 1: 487–500. 1984 b. A list of retrobursal triclads inhabiting freshwaters and aquatic probursal triclads regarded as marine relicts, with corrective remarks on our 1984 paper published in Zoological Science, Tôkyô. Occ. Publ., Biol. Lab. Fuji Women's College, Sapporo (Hokkaidô), Japan, (11): 1–8.

KAWAKATSU, M., OKI, I., TAMURA, S., OGREN, R. E., YAMADA, T. & MURAYAMA, H., 1990. Preprint of papers given at the Sixth International Symposium on the Turbellaria, Hirosaki, Japan (August 7-12, 1990). Occ. Publ., Biol. Lab. Fuji Women's College, Sapporo (Hokkaidô), Japan, (22): 1-16.

KAWAKATSU, M., OKI, I., TAMURA, S., TAKAI, M., TIMOSHKIN, O. A. & PORFIRJEVA, N. A., 1993. Preprint of papers given at the Seventh International Symposium on the Turbellaria, Åbo, Finland (June 17–22, 1993). Occ. Publ., Biol. Lab. Fuji Women's College, Sapporo (Hokkaidô), Japan, (25): 1–20.

KAWAKATSU, M., OKI, I., TAMURA, S. & YAMAYOSHI, T., 1985. Reexamination of freshwater planarians found in tanks of tropical fishes in Japan, with a description of a new species, *Dugesia austroasiatica* sp. nov. (Turbellaria; Tricladida; Paludicola). Bull. Biogeogr. Soc. Japan, 40: 1–19.

LANG, A., 1884. Die Polycladen (Seeplanarien) des Golfes von Neapel und der Angrenzenden Meeresabschnitte. In: Fauna und Flora des Golfes von Neapel und der angrenzenden Meeres-Abschnitte herausgegeben von der Zoologischen Station zu Neapel. XI. Monographie. I-IX + 1-688 pp. + pls. 1-39. Verlag von Wilhelm Engelmann, Leipzig.

Fig. 4 (on page 68). Reproduction in reduced size of Mr. K ISOMURA's Japanese article on the "Suffering of Tropical Fishes in the Amazon." After The Asahi Newspaper (Evening edition, October 7, 1995). For explanation, see the "Remarks on the accidental distribution" in the present paper.

MARCUS, E., 1946. Sôbre Turbellaria Brasileiros. Zoologia, Fac. Filos., Ciênc. Let. Univ. São Paulo, (11): 5–250 (+ pls. I–XXX)+ pl. XXXI.

MITCHELL, R. W. & KAWAKATSU, M., 1972. A new family, genus, and species of cave-adapted planarian from Mexico (Turbellaria, Tricladida, Maricola). Occ. Pap., Mus. Texas Tech Univ., (8): 1–16.

OKI, I., TAMURA, S. & KAWAKATSU, M., 1976. A technique for the study of planarian chromosomes. The Heredity (Iden), Tôkyô, 30 (12): 32-40. (In Japanese.)

OKI, I., TAMURA, S., OGREN, R. E. & KAWAKATSU, M., 1991. Karyology of four land-planarian species of the genus *Bipalium* from Japan. Hydrobiologia, 227: 163-167.

OKI, I., TAMURA, S., TAKAI, M. & KAWAKATSU, M., 1993. Chromosomes of *Temnocephala minor*, an ectosymbiotic turbellarian on Australian crayfish found in Kagoshima Prefecture, with karyological notes on exotic turbellarians found in Japan. VII ISBT Åbo/Turku Abstracts, p. 91. Åbo Akademi Univ., Åbo, Finland. 1995. Chromosomes of *Temnocephala minor*, an ectosymbiotic turbellarian on Australian crayfish found in Kagoshima Prefecture, with karyological notes on exotic turbellarians found in Japan. Hydrobiologia, 305: 71–77.

OKI, I., TAMURA, S., YAMAYOSHI, T. & KAWAKATSU, M., 1980. Preprint of a paper given at the Third International Symposium — The Biology of the Turbellaria in Honour of Prof. Tor G. KARLING, Hasselt (Diepenbeek), Belgium (August 10–15, 1980). Occ. Publ., Biol. Lab. Fuji Women's College, Sapporo (Hokkaidô), Japan, (2): 1–23.

SLUYS, R., 1990. A monograph of the Dimarcusidae (Platyhelminthes, Seriata, Tricladida). Zool. Scripta, 19: 13-29.

STIMPSON, W., 1857. Prodromus descriptiones animalium evertebratorum quae in Expeditione and Oceanum, Pacificum Septentrionalem a Republica Federata missa, Johanne RODGERS Duce, observavit et descripsit. Proc. Acad. Nat Sci. Philad., 9: 19-31.

TAKAI, M., 1989. A new collecting method of freshwater planarians and a system of breeding in large number. Acta Eruditiorum (General Educ., Saga Med. School), (8): 15-26. (In Japanese, with English summary.)

Addresses of the Authors:

- Dr. Masaharu KAWAKATSU, Professor of Biology, Biological Laboratory, Fuji Women's College, Kita-16, Nishi-2, Kita-ku, Sapporo (Hokkaidô) 001, Japan.
- Dr. Sachiko TAMURA, Ôsaka Prefectural Institute of Public Health, Nakamichi-1-chôme 3-69, Higashinari-ku, Ôsaka 537, Japan.
- Dr. Iwashirô OKI, Environmental Creative Engineering Laboratory Inc., Tokiwachô-1-chôme 4-12, Chû'ô-ku, Ôsaka 540, Japan.
- Dr. Masayuki TAKAI, Professor of Biology, Biological Laboratory, Saga Medical School, Nabeshima 5-1-1, Saga 849, Japan.
- Dr. Eudoxia Maria FROEHLICH, Professor of Biology, Emeritus, Instituto de Biociências, Departamento de Zoologia, Universidade de São Paulo, Rua do Matão, Travessa 14, Cidade Universitária, Caixa Postal 11461, Cep 05422-970, São Paulo, SP, Brasil.