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## Helminth fauna of the Ryukyu Archipelago, Japan. 2. Four *Kalicephalus* species parasitic to snakes in Okinawa Island (Nematoda: Diaphanocephalidae)

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*Kalicephalus* nematodes are the hookworm-like parasites of reptiles and very characteristic in the cephalic structure. From Japan proper, Yamaguti (1935) described *K. natrixis* from *Natrix tigrina* and *Elaphe quadrivirgata*, and later, Fukui (1963) recorded this species from the same host species. However, the validity of *K. natrixis* was doubted by Schad (1962). Kamegai *et al.* (1962) detected *Kalicephalus* sp. from *Agkistrodon halys* in Honshu and Kyushu.

From the Ryukyu Archipelago, Yamaguti (1935) described *K. laticaudae* from a sea snake, *Laticauda laticaudata*, in Ishigaki Isl. This species was also collected from *L. semifasciata* in Amami-oshima Isl. by Telford (1967). Hori and Kaneko (1969) and Kagei (1973) detected *Kalicephalus* sp. from *Trimeresurus flavoviridis* in Amami-oshima Isl. Schad (1962) reported some morphological characteristics of *K. viperae chungkingensis* collected from an unidentified snake in Okinawa Isl.

Since 1980, surveys on zoonotic parasites in the Ryukyu Archipelago have been carried out, and various animals have been examined for parasites. During those surveys, the authors detected four *Kalicephalus* species from the snakes in Okinawa Isl., recently. This paper deals with the morphology and taxonomy of these nematodes.

### Materials and Methods

Most of the specimens were collected from the viscera of snakes killed with chloroform, fixed in hot 70 % ethanol. Worms from *Ophedrys semicarinatus* were recovered from the host viscera preserved in 10 % formalin solution. For microscopic observation, the parasites were cleared in glycerin-alcohol solution and mounted with 50 % glycerin jelly. Figures of worms were made with the aid of a drawing tube, Olympus BH-DA-LB. All specimens were deposited in the Department of Parasitology, School of Medicine, University of the Ryukyus.

### Description and Discussion

1. *Kalicephalus viperae chungkingensis* Hsü, 1934

(Fig. 1)

Materials studied: 5 males and 4 females.

Host: *Trimeresurus okinavensis*

Habitat: Esophagus.

Locality: Ada, Kunigami Village, Okinawa Prefecture.

Date: July, 1981

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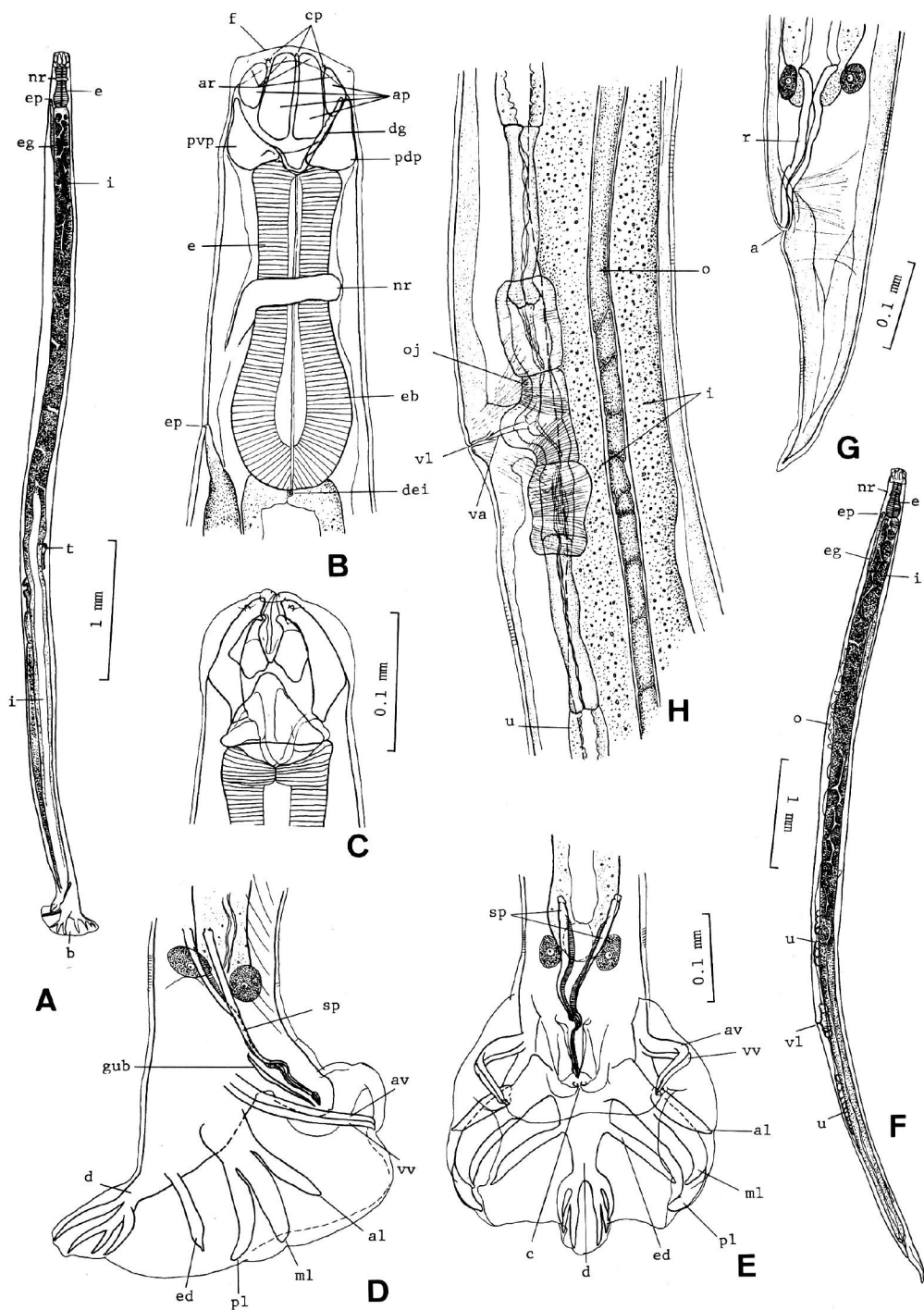


Fig. 1. *Kalicephalus viperae chungkingensis* Hsü, 1934

- A. Male, general view; B. Anterior extremity of male, lateral view; C. Anterior extremity of male, ventral view; D. Bursa copulatrix, lateral view; E. Bursa copulatrix, ventral view; F. Female, general view; G. Posterior extremity of female, lateral view; H. Vulval region, lateral view.

Body slim, almost straight in fixed condition. Face rounded, with cuticle expanded medially and dorsoventrally. Anterior chitinoid ridge also rounded. Posterior ventral and dorsal pieces almost triangular and with smooth posterior edges. Dorsal gutter developed and beaded. Esophagus with long neck and elongated bulb. Excretory pore positioned posterior to middle of esophageal bulb or posterior to esophagus. Cervical papillae present at level of posterior end of esophagus. Excretory glands not overlapping base of esophageal bulb.

Male: Body tapered to both extremities. Posterior margin of bursa copulatrix oblique in lateral view. Ventral rays thin, run parallel close together in their total length. Lateral rays derived from a common trunk. Antero-lateral shortest and more divergent than postero-lateral from medio-lateral ray. Externo-dorsal ray derived from the dorsal ray near its base and has thin distal end. Dorsal ray divided into two inner and two outer branches at same level, former branches are redivided into inner longer and outer shorter offshoots. Spicules equal, having short strongly shouldered tips. Gubernaculum present. Genital cone prominent. Dorsal lip of genital cone has two papillae.

Female: Body tapered to both extremities. Reproductive system didelphic. Vulva not protruded. Tail conical, slightly bent ventrad, and lacking terminal spike.

Measurements of the worms are shown in Table 1.

*K. v. chungkingensis* is parasitic to various snakes including *Trimeresurus* spp. in the Southeast Asia. Schad (1962) examined specimens of this species collected from an unidentified snake in Okinawa Isl. and noticed that they had shorter stouter tails than did the Chinese and Formosan specimens. The same feature is also observed in the present worms. *Trimeresurus okinavensis* is first recorded as a host of *K. v. chungkingensis*.

## 2. *Kalicephalus costatus indicus* Ortlepp, 1923

(Fig. 2)

Materials studied: 6 males and 7 females.

Host: *Opheodrys semicarinatus*

Habitat: Small intestine.

Locality: Yona, Kunigami Village, Okinawa Prefecture.

Date: July, 1981.

Body relatively stout, somewhat contracted probably due to inadequate fixation. Cervical region expanded. Face rounded, with inflated cuticle, directing almost anteriorly. Anterior chitinoid ridge wide. Posterior ventral and dorsal pieces large and a thick superficial welt connects them. Esophagus short, thick and with bulb. Excretory pore positioned anterior to middle of esophageal bulb. Cervical papillae located behind level of excretory pore.

Male: Ventral rays of bursa copulatrix almost same in length, run together with each other. Antero-lateral ray shortest among laterals. Medio-lateral and postero-laterals almost equal, diverging distally. Externo-dorsal ray originated near base of dorsal ray. Dorsal ray divided into two branches which immediately redivided into outer and inner offshoots. Inner offshoots divided distally. Spicules equal, alate, with spatulated tips. Gubernaculum prominent. Genital cone contracted in present specimens.

Female: Body tapered to posterior end. Uteri prodelphic. Vulva slightly elevated. Tail conical,

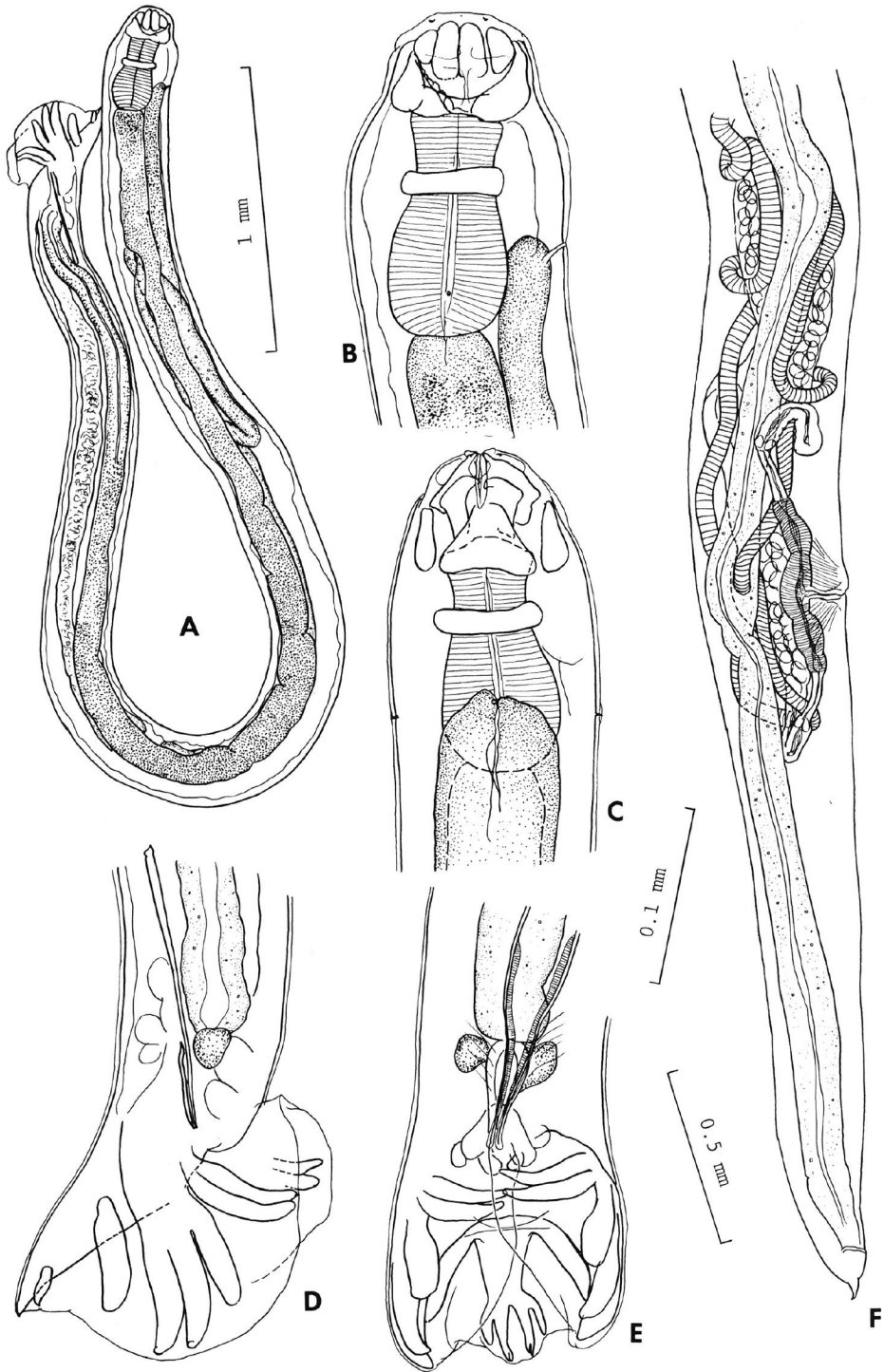


Fig. 2. *Kalicephalus costatus indicus* Ortlepp, 1923

A. Male, general view; B. Anterior extremity of male, lateral view; C. Anterior extremity of male, ventral view; D. Bursa copulatrix, lateral view; E. Bursa copulatrix, ventral view; F. Posterior part of female, lateral view.

with terminal spike.

Morphometric data are stated in Table 1.

*K. c. indicus* is widely distributed in Oriental and Australian areas. This species has been found not only from the snakes, but also from the lizards and even from a wild cat. The present worms have deeper buccal capsule and longer distance between the cephalic apex and the excretory pore than do the specimens from other countries (Hsü, 1934; Baylis, 1936; Schad, 1962). The inadequate fixation might cause these discrepancies but it may not be denied that these are due to the host or geographical variations. The snake, *Ophedrys semicarinatus*, is recorded as a new host of *K. c. indicus*. Although there has been no reliable report on *K. c. indicus* from Japan, Schad (1962) suspected that *K. natrixis* Yamaguti, 1935 was this species in part.

### 3. *Kalicephalus posterovulvus* Schad, 1962

(Fig. 3)

Materials studied: 10 males and 11 females.

Host: *Natrix pryeri pryeri*

Habitat: Esophagus.

Locality: Ada, Kunigami Village, Okinawa Prefecture.

Date: July, 1981.

Body slim, almost straight in fixed condition. Face rounded, directing anteriorly or slightly tilted dorsally, with cuticle expanded medially and strongly inflated in corners. Anterior chitinous ridge rounded. Posterior ventral chitinous piece almost triangular. External edge of the posterior ventral piece rounded, without protrusions. Posterior dorsal piece narrow. Dorsal gutter long and with prominent internal ridge beaded weakly. Esophagus composed of relatively long neck and elongated bulb. Excretory pore positioned anterior to middle of esophageal bulb. Cervical papillae present behind level of excretory pore. Excretory glands large and overlapping base of esophageal bulb.

Male: Body slender, tapered to anterior and posterior extremities. Antero-ventral and ventro-ventral rays of bursa copulatrix fused basally but separated distinctly through distal two-thirds. Lateral rays sharing a common base, divergent at same level, forming almost equal angles. Antero-lateral ray slightly shorter than other laterals. Externo-dorsal ray stout basally, curved and pointed distally. Dorsal ray branched two times: inner termination slightly distal to outer termination. Spicules equal, alate and pointed. Gubernaculum present. Genital cone short and thick. A pair of papillae on distal end of dorsal lip of cloaca.

Female: Body tapered to posterior end. Uteri prodelphic. Vulva far to posterior. Postvulval body lacks ovarian coils. Tail conical, simple and without terminal spike.

Measurements of worms are presented in Table 1.

*K. posterovulvus* was first described on the materials mainly from Formosan snakes (Schad, 1962). The morphological and morphometrical features of the present worms are identical with those of original description. This is the first record of *K. posterovulvus* from the snake of the genus *Natrix*.

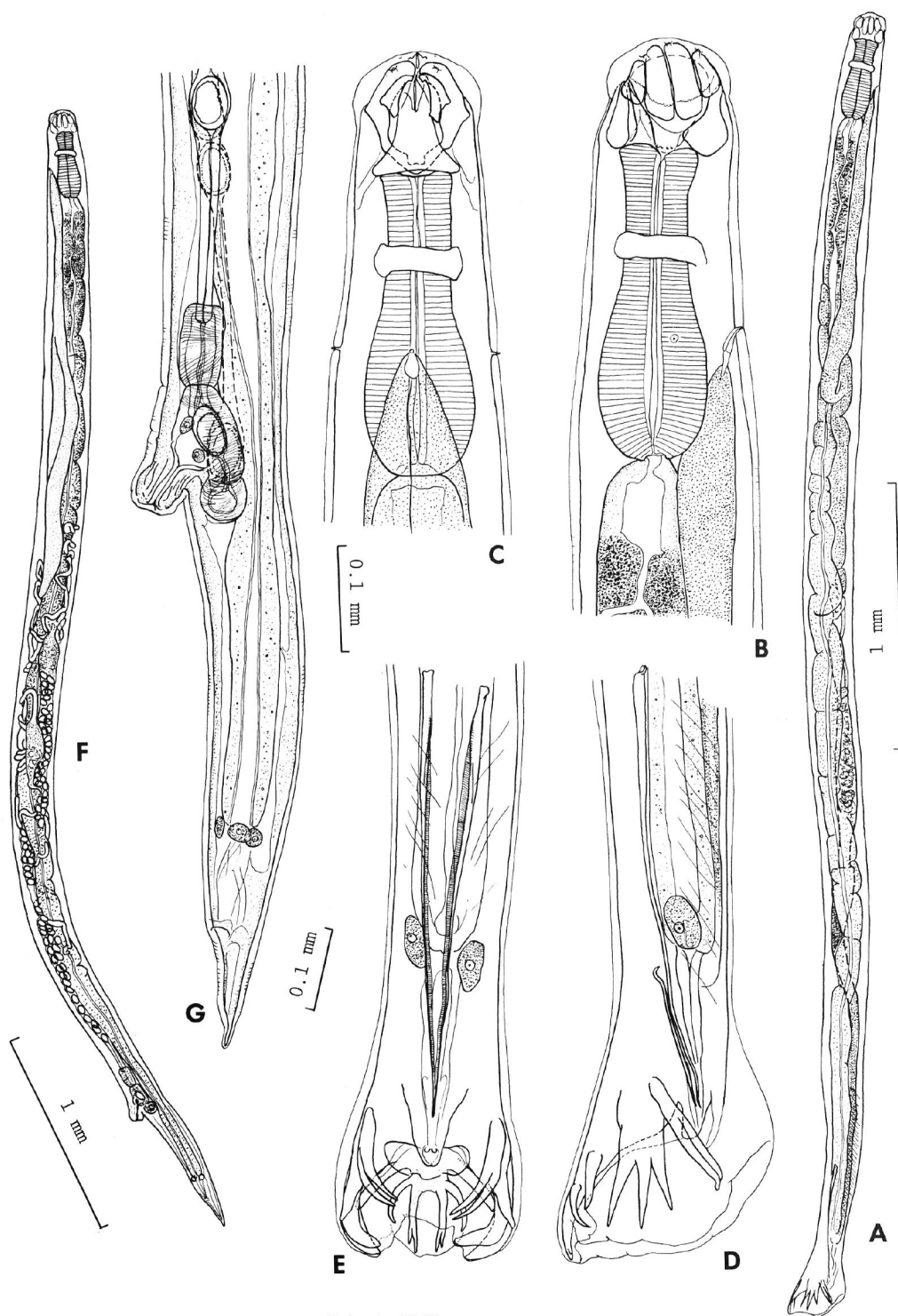


Fig. 3. *Klicecephalus posterovulvus* Schad, 1962

A. Male, general view; B. Anterior extremity of male, lateral view; C. Anterior extremity of male, ventral view; D. Bursa copulatrix, lateral view; E. Bursa copulatrix, ventral view; F. Female, general view; G. Posterior part of female, lateral view.

Table 1 Morphometric data for *Kalicephalus* spp. recovered from snakes in Okinawa Island

| Species                                   | <i>K. viperae chungkingensis</i> | <i>K. costatus indicus</i>     | <i>K. postero-vulvus</i>    | <i>K. brachycephalus</i>    |
|---|----------------------------------|--------------------------------|-----------------------------|-----------------------------|
| Host                                      | <i>Trimeresurus okinavensis</i>  | <i>Ophiodrys semicarinatus</i> | <i>Natrix pryeri pryeri</i> | <i>Natrix pryeri pryeri</i> |
| Male:                                     |                                  |                                |                             |                             |
| No. worms measured                        | 5                                | 6                              | 10                          | 3                           |
| Body length                               | 5.7-6.6                          | 4.7-5.7                        | 3.3-4.8                     | 3.7-5.8                     |
| Maximum width of body                     | 0.20-0.26                        | 0.25-0.27                      | 0.13-0.20                   | 0.22-0.28                   |
| Head diameter                             | 0.13-0.16                        | 0.19-0.21                      | 0.12-0.13                   | 0.16-0.29                   |
| Depth of buccal capsule                   | 0.12-0.14                        | 0.16-0.17                      | 0.11-0.12                   | 0.16-0.28                   |
| Distance from cephalic apex to nerve ring | 0.21-0.23                        | 0.20-0.23                      | 0.17-0.19                   | 0.20-0.32                   |
| to excretory pore                         | 0.35-0.38                        | 0.27-0.29                      | 0.24-0.28                   | 0.32-0.41                   |
| Esophagus length                          | 0.29-0.30                        | 0.26-0.28                      | 0.27-0.30                   | 0.28-0.45                   |
| Maximum width of esophagus                | 0.10-0.13                        | 0.14-0.15                      | 0.08-0.10                   | 0.12-0.15                   |
| Spicule length                            | 0.25-0.28                        | 0.34-0.38                      | 0.35-0.42                   | 0.29-0.37                   |
| Gubernaculum length                       | 0.09-0.12                        | 0.10-0.15                      | 0.13-0.15                   | 0.14-0.15                   |
| Female:                                   |                                  |                                |                             |                             |
| No. worms measured                        | 4                                | 7                              | 11                          |                             |
| Body length                               | 7.2-7.7                          | 4.7-6.7                        | 3.7-6.2                     |                             |
| Maximum width of body                     | 0.26-0.29                        | 0.21-0.39                      | 0.15-0.26                   |                             |
| Head diameter                             | 0.17                             | 0.19-0.24                      | 0.13-0.16                   |                             |
| Depth of buccal capsule                   | 0.14-0.16                        | 0.16-0.20                      | 0.11-0.13                   |                             |
| Distance from cephalic apex to nerve ring | 0.22-0.25                        | 0.21-0.27                      | 0.18-0.21                   |                             |
| to excretory pore                         | 0.37-0.52                        | 0.27-0.29                      | 0.24-0.31                   |                             |
| Esophagus length                          | 0.33-0.34                        | 0.29-0.32                      | 0.29-0.32                   |                             |
| Maximum width of esophagus                | 0.13-0.15                        | 0.13-0.18                      | 0.08-0.12                   |                             |
| Vulval ratio                              | 1.83-2.00 : 1                    | 2.22-3.33 : 1                  | 6.12-8.06 : 1               |                             |
| Tail length                               | 0.27-0.30                        | 0.17-0.19                      | 0.11-0.16                   |                             |
| Egg dimensions, $\mu\text{m}$             | 75-85 x 43-66                    | 69-85 x 33-40                  | 58-63 x 33-40               |                             |

Measurements in mm unless otherwise stated.

Vulval ratio is expressed as prevulval body length : postvulval body length = x : 1



4. *Kalicephalus brachycephalus* Maplestone, 1931

(Fig. 4)

Materials studied: 3 males.

Host: *Natrix pryeri pryeri*

Habitat: Rectum (?)

Locality: Ada, Kunigami Village, Okinawa Prefecture.

Date: July, 1981.

Body slim. Head very large. Face tilted slightly dorsally. Anterior ridge curved, narrow in width. Posterior dorsal piece crescent in shape. Posterior ventral piece elongated longitudinally. Weakly-developed superficial welt present between both posterior pieces. Dorsal gutter beaded. Anterior part of head strongly compressed laterally. Esophagus with long neck and elongated bulb. Excretory pore present in areas between nerve ring and middle of esophageal bulb. Cervical papillae present at level of esophageal bulb.

Male: Posterior margin of bursa copulatrix almost transverse. Bursal rays thin and fine. Ventral rays diverging distally. Lateral rays arising from a common trunk. Antero-lateral ray shortest among laterals. Dorsal ray relatively short. Dorsal ray divided into two inner and two outer branches at same level. Inner branches shorter than outer ones, redivided distally. Spicules almost equal, with spatulated tips. Gubernaculum relatively large. Genital cone protruded. A pair of small papillae present on dorsal lip of cloaca and one papilla on the ventral lip of it.

Morphological data are shown in Table 1.

*K. brachycephalus* is characterized by a very large buccal capsule. The morphological appearances of the present materials are identical with the previous descriptions (Hsü, 1934; Schad, 1962), although the distance from the cephalic apex to the nerve ring and the excretory pore are slightly shorter. There has been no record of *K. brachycephalus* from Japan but Schad (1962) suspected that *K. naticis* Yamaguti, 1935 was this species in part.

#### Summary

Four *Kalicephalus* species (Nematoda: Diaphanocephalidae), i. e., *K. viperæ chungkingensis* Hsü, 1934 from *Trimeresurus okinavensis*, *K. costatus indicus* Ortlepp, 1923, from *Opheodrys semicarinatus*, *K. posteroovulvus* Schad, 1962 and *K. brachycephalus* Maplestone, 1931 from *Natrix pryeri pryeri*, were collected in Okinawa Island, Japan. *K. c. indicus*, *K. posteroovulvus* and *K. brachycephalus* were first recorded from Japan. *T. okinavensis*, *O. semicarinatus* and *N. p. pryeri* were recorded as the new hosts for *K. v. chungkingensis*, *K. c. indicus*, and *K. posteroovulvus* and *K. brachycephalus*, respectively. The morphological characteristics of the worms were described and figured.

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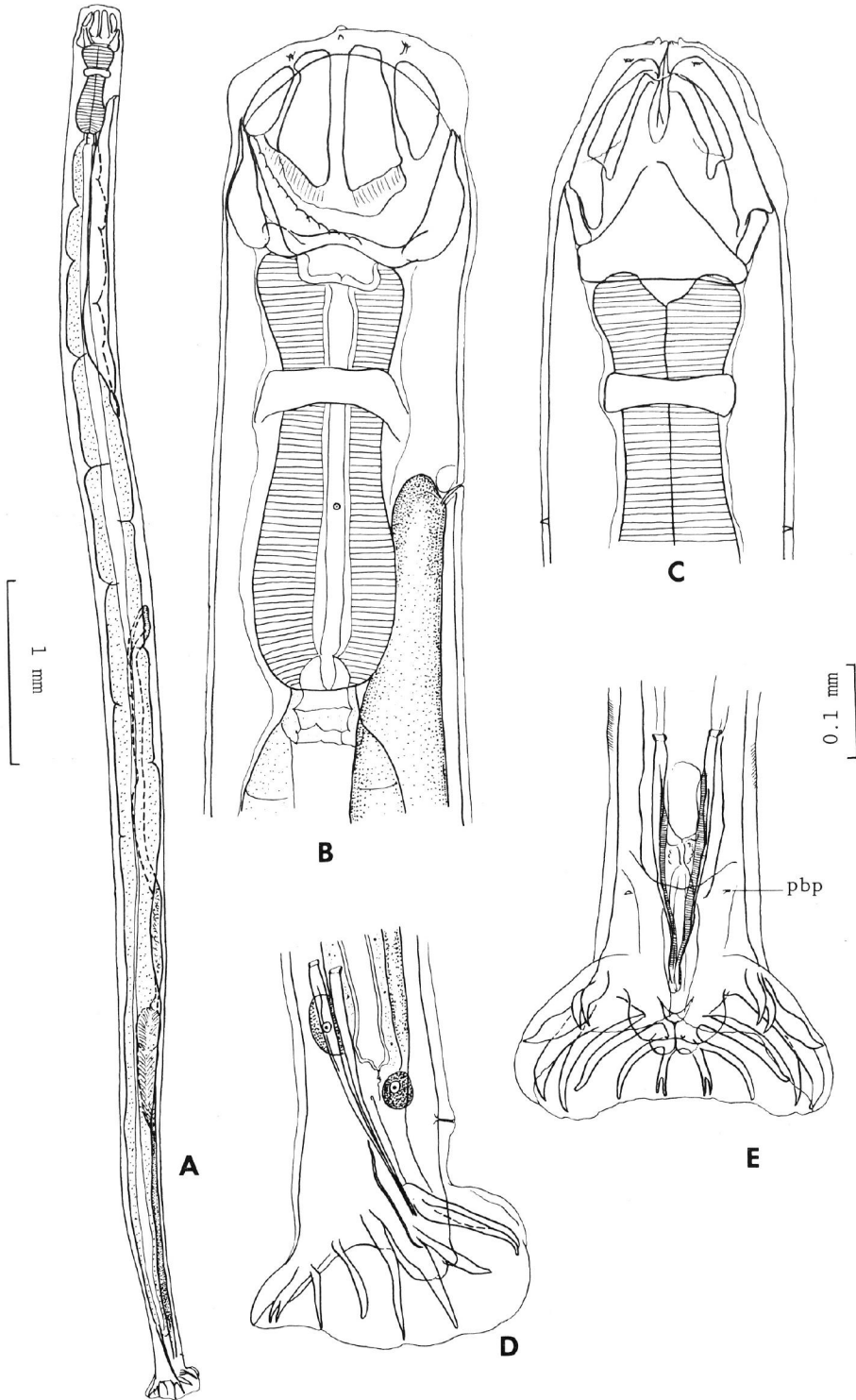


Fig. 4. *Kalicephalus brachycephalus* Maplestone, 1931

A. Male, general view; B. Anterior extremity of male, lateral view; C. Anterior extremity of male, ventral view; D. Bursa copulatrix, lateral view; E. Bursa copulatrix, ventral view.

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Abbreviations used in figures: a. anus; al. antero-lateral ray; ap. anterior plates; ar. anterior chitinous ridge; av. antero-ventral ray; b. bursa copulatrix; c. cloaca; cp. cephalic papillae; d. dorsal ray; dei. deirid; dg. dorsal gutter; e. esophagus; eb. esophageal bulb; ed. externo-dorsal ray; ag. excretory glands; ep. excretory pore; f. face; gub. gubernaculum; i. intestine; ml. medio-lateral ray; nr. nerve ring; o. ovary; oj. ovijector; pbb. prebursal papillae; pdp. posterior dorsal piece; pl. postero-lateral ray; pvp. posterior ventral piece; r. rectum; sp. spicule(s); t. testis; u. uterus; va. vagina; vl. vulva; vv. ventro-ventral ray.