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Diversity and distribution of Brachionidae (Rotifera) in Thailand, with a key to the species

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ABSTRACT

We investigated the diversity of brachionid rotifers in Thailand. Rotifers were collected from a wide range of habitat types (294 sites, 508 samples) including canals, lakes, peat swamps, ponds, reservoirs, rice fields, rivers, swamps and temporary ponds from April 1998 to January 2004. Twenty-eight species including 11 infrasubspecific forms belonging to five genera were identified. The most frequently encountered species was Brachionus falcatus *(50.7% of the sampling sites), followed by* Keratella cochlearis *(50%),* B. angularis *(49.7%),* K. tropica *(46.3%) and* B. forficula *(45.9%). Less frequently encountered species were* B. caudatus*,* B. durgae *and* B. urceolaris*. These species were found in low abundances and only in a single site. Two sites in the Northeast – Nong Changpeuak and Bueng Srithat – recorded the greatest richness with 15 taxa, whereas no brachionid rotifers were found in 31 sites. On average, the species richness was five taxa per sampling site. A key to the genera and species of Thai brachionid rotifers is provided.*

Key words: biodiversity, Brachionus, Keratella*, rotifers, Southeast Asia*.

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INTRODUCTION

Rotifera of the family Brachionidae are amongst the most well-studied rotifer groups. This is largely due to their ecological and economic relevance (Segers, 2008). The most well known are the brackish water species of the *Brachionus plicatilis* complex, which are important as a primary live food source for commercial fish and shrimp larvae, and are therefore extensively used in aquaculture (Lubzens *et al.*, 2001). Moreover, three freshwater species, *B. calyciflorus*, *B. rubens* and *B. urceolaris*, are particularly suited as test organisms for ecotoxicological studies (Snell and Janssen, 1995). The common occurrence of phenotypic plasticity resulting in polymorphism has confounded the taxonomy of brachionid rotifers (Nogrady *et al*., 1993). The most striking examples of phenotypic plasticity occurs in *Brachionus* and *Keratella*. For example, *Keratella cochlearis* and *K.tropica* are notoriously variable morphologically (Hofmann, 1980; Marinone and Zagarese, 1991). There are several factors that affect rotifer phenotypic plasticity. These include temperature (Bielanska-Grajner, 1995) and, especially, presence of predators (Marinone and Zagarese, 1991). Different morphological variants of species have been treated as infrasubspecific varieties or forms (Nogrady *et al*., 1993). However, there is cryptic diversity in the genus *Brachionus*. For instance, a re-examination of existing available names proposed *B. plicatilis* and *B. rotundiformis* as the correct names for the *L-* and *S-types* of *B. plicatilis*, respectively (Segers, 1995), and more species are being distinguished in the *B. plicatilis* complex (Ciros-Pérez *et al*., 2001) as well as in other taxa in the genus *Brachionus* (Gilbert and Walsh, 2005; Xiang *et al*., 2010, 2011a, 2011b).

During the past decades, there has been a steady increase in interest in rotifer fauna in Thailand. However, information on Thai rotifers is still incomplete and there is a strong need to increase our knowledge on the subject. This also holds for the family Brachionidae. Recently, 169 species belonging to seven genera: *Anuraeopsis, Brachionus, Kellicottia, Keratella, Notholca, Plationus* and *Platyias* have been listed as making up the Brachionidae (Segers, 2007, 2008). Among these genera, *Brachionus* is the most diverse genus, containing 64 species and contributing the second most number of species to rotifer diversity in Southeast (SE) Asia (Segers, 2001). Thai brachionid rotifers have been recorded in a number of publications (Boonsom, 1984; Sanoamuang *et al*., 1995; Chittapun and Pholpunthin, 2001; Sanoamuang and Savatenalinton, 2001; Segers *et al*., 2004; Chittapun *et al*., 2007). Most studies have focused on the northeastern and southern parts of the country while few records are available from other regions. Considering the economic and ecological importance of the family Brachionidae in tropical and SE Asian waters, we provide a review of the taxonomy and distribution of this family in Thai waters. In addition, keys to the genera and species of Thai bra-

chionid rotifers were constructed based on external morphology, and illustrations were provided.

METHODS

Sample collection and examination

Rotifers were collected from a wide range of habitats (294 sites, 508 samples) including canals, lakes, peat swamps, ponds, reservoirs, rice fields, rivers, swamps and temporary ponds. Horizontal tows were made at a depth of 2 m with an approximate linear distance of 5 m in open water using a 30-µm mesh size plankton net. In each site, sampling was performed in both dry and rainy seasons from April 1998 to January 2004. However, sampling was done only once in 80 sites. Samples were preserved in 5% formaldehyde-solution. Locations of the sampling sites are shown in Fig. 1. Water temperature, conductivity, pH, dissolved oxygen (DO), turbidity and salinity were measured at each locality using a Horiba Water Quality Checker (U-10; Horiba Ltd., Fukuoka, Japan). Brachionid rotifers were examined under an Olympus CH-10 compound microscope at 400-1000× (Olympus, Tokyo, Japan). The rotifers were identified to species level using the keys and publications from several taxonomists (Ahlstrom, 1940; 1943; Koste, 1978; Koste and Shiel, 1987). Drawings were made using an Olympus CH-10 microscope (Olympus) equipped with a drawing tube. The specimens were prepared for observation with a scanning electron microscopy (SEM) (SEM-LEO 1450) using the critical point drying technique described in earlier publications (Sanoamuang and McKenzie, 1993; Segers, 1993; Segers and Dumont, 1993). In order to analyse the relationship between the environmental variables and species data, canonical correspondence analysis (CCA) was performed with the PC-ORD statistical software programme (version 4; MjM Software Design, Geleneden Beach, OR, USA).The CCA procedure produces an ordination diagram in which species are represented by points and environmental variables by vectors. A set of environmental variables and species composition were tested for significance of relevant trends by the Monte Carlo permutation test.

RESULTS AND DISCUSSION

Species composition of Thai brachionid rotifers

A total of 39 taxa, 28 species and 11 infrasubspecific forms of the family Brachionidae were recorded. A list of the identified brachionid rotifers, their distribution and characteristics of their habitats is shown in Tab. 1. Description of sampling sites and variables measured at the sampling sites are presented in Supplementary Tab. 1. The most diverse genus of Thai Brachionidae is *Brachionus*, comprising 18 species and 10 infrasubspecific forms, followed by *Keratella* (6 species and 1 infrasubspecific form), *Anuraeopsis* (2 species), *Plationus* and *Platyias*. The two last genera contained only a single species each.This concurs with Segers (2008), who reported that *Brachionus* is the most diverse genus of the family Brachionidae in the world (64 species) (Segers 2007, Silva-Briano *et al*., 2007). Most of the species recorded from Thailand are common and widely distributed. On average, species richness was five taxa per site. Two sites in the northeastern area (Nong Changpeuak and Bueng Srithat) had the greatest richness with 15 taxa; whereas, no brachionid rotifers were found in 31 sites. The most frequently encountered species were *Brachionus falcatus* (in 50.7% of samplings), followed by *Keratella cochlearis* (50%), *B. angularis* (49.7%), *K. tropica* (46.3%) and *B. forficula* (45.9%). Species with low frequencies were *B. caudatus*, *B. durgae* and *B. urceolaris*. These species were only found, as a few specimens, in a single site.

Distribution of Thai brachionid rotifers

Thirteen of the Thai brachionid taxa are widely distributed throughout Thailand (northern, northeastern, central and southern regions) as shown in Tab. 1. Six taxa are found to have a restricted distribution: *B. angularis* f. *bidens* and *B. forficula* f. *minor* are recorded only from the North; *B. caudatus*, *B. durgae* and *B. urceolaris* occur only in Central Thailand; and *K. edmondsoni* is recorded only from the Northeast. It is noteworthy that the following eight species, reported by previous authors (Sanoamuang *et al*., 1995; Chittapun and Pholpunthin, 2001; Segers *et al*., 2004) were not recorded here, probably due to their rarity and restricted distribution: *Anuraeopsis navicula*, *Brachionus kostei*, *B. lyratus*, *B. sessilis*, *B. srisumonae*, *B. variabilis*, *Keratella javana* and *K. taksinensis*. These include the two SE Asian brachionid endemics: *B. srisumonae*, known from the northeastern region and Laos, and *K. taksinensis,* from the South.

Relationship between rotifer diversity and habitat characteristics

The greatest diversity of brachionid rotifers was in ponds and rivers (30 taxa), followed by canals (27 taxa) and swamps (25 taxa). All the habitats having higher rotifer diversity were permanent water bodies, whereas temporary water bodies (*i.e.* rice fields and temporary ponds) were inhabited by relatively few (14) taxa. This result concurs with the report on the brachionid fauna of Northeastern Thailand, where 32 brachionid rotifers were encountered in permanent waters bodies and only five species were recorded from temporary ponds (basins) (Sanoamuang *et al*., 1995). In peat swamps, characterised by acidic and brownish water, only a few species could be found. For example, Chittapun and Pholpunthin (2001)

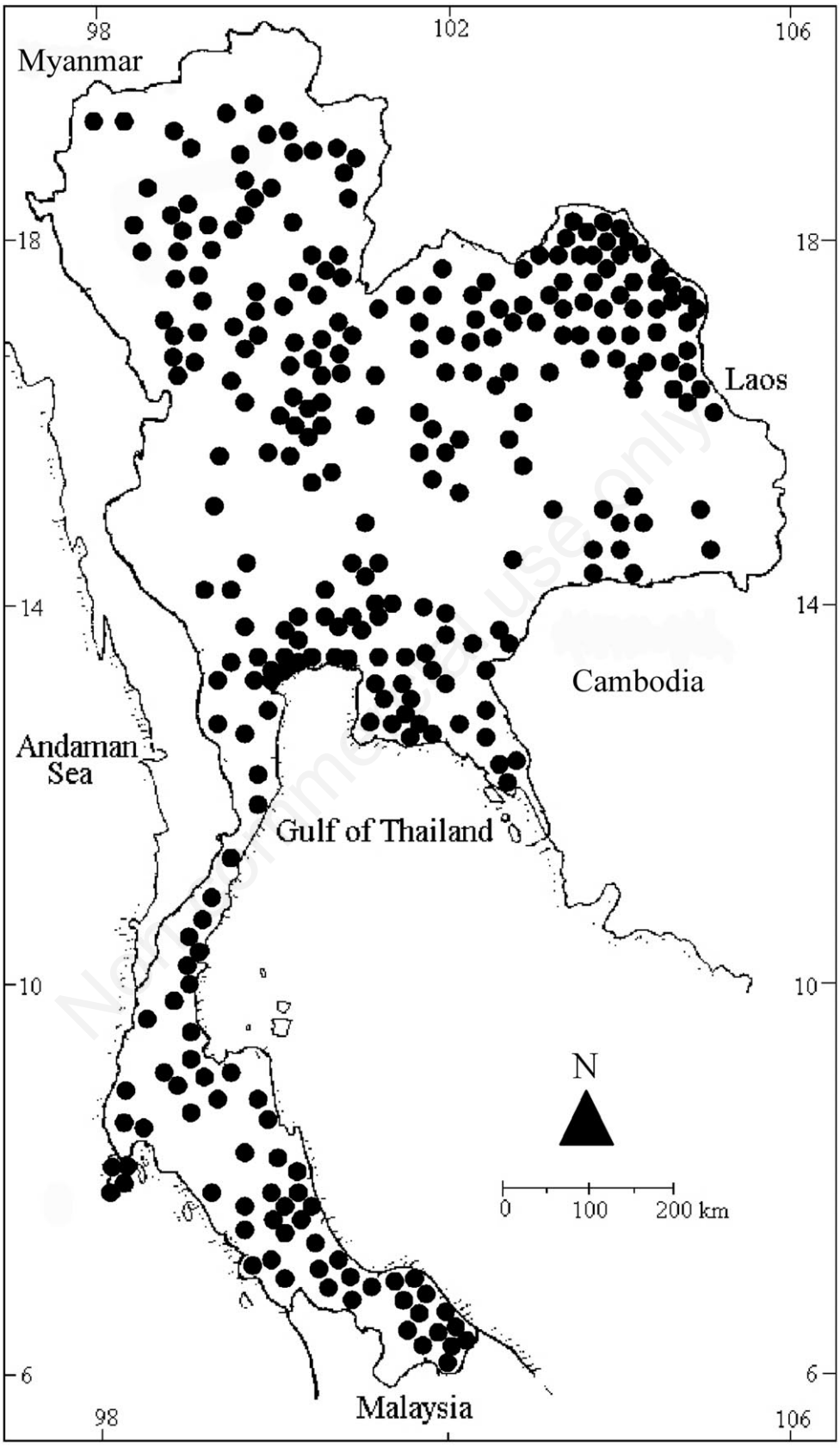

Fig. 1. Map of Thailand showing location of sampling sites (black spots).

investigated the rotifer fauna in three acidic peat swamps (pH 4.30-6.37) in Southern Thailand. A total of 93 rotifer species were recorded, only four of which were brachionids: *Anuraeopsis fissa, Brachionus quadridentatus* f. *mirabilis, Keratella javana* and *Plationus patulus*. These results agree with the observation that *Brachionus* rotifers are generally absent from acidic waters (Ahlstrom, 1940). Another environmental variable that influences the distribution of rotifers is salinity: no brachionid rotifers were found in estuaries. Only one species, *B. rotundiformis*, has been found in salinities ranging from 0.04 to 2.73 practical salinity units (PSU). This result agrees with previous work that indicated that rotifer communities are limited by salinity (Ruttner-Kolisko, 1974) and that rotifer biodiversity decreases with increasing water salinity (Sladecek, 1983). Attayde and Bozelli (1998) also reported that the species diversity of rotifers decreased with increasing marine influence. On the other hand, species richness of rotifers in Turkish continental waters was significantly increased with salinity in sub-saline waters between 1000 and 6000 µS cm^{-1} (Kaya *et al.*, 2010). In addition, the family does contain an important number of brackish-

Tab. 1. List of brachionid rotifers recorded in Thailand during this study.

Species	Habitat	Distribution area
Anuraeopsiscoelata (De Beauchamp, 1932)	Ca, L, P, Re, Ri, Sw	N, NE, C, S
A. fissa (Gosse, 1851)	Ca, P, Ps, Re, Rf, Ri, Sw, T	N, NE, C, S
Brachionus angularis Gosse, 1851	Ca, L, P, Re, Rf, Ri, Sw, T	N, NE, C, S
B. angularis f. *bidens* Plate, 1886	Ri	N
B. bennini (Leissling, 1924)	P, Ri	N, NE
B. bidentatus f. *inermis* (Rousselet, 1906)	P, Re, Ri, Sw	N, NE, C
B. budapestinensis Daday, 1885	P, Re	NE, C
B. calyciflorus Pallas, 1766	Ca, P, Ps, Re, Ri, Sw	N, NE, C, S
B. calyciflorus f. *amphiceros* Ehrenberg, 1838	Ca, L, P, Ri	N, NE, C
B. calyciflorus f. *anuraeiformis* Brehm, 1909	Ca, L, P, Re, Rf, Ri, Sw	N, NE, C, S
B. caudatus Barrois and Daday, 1894	Re	C
B. caudatus f. *aculeatus* Hauer, 1937	Ri, Sw	N, NE, C
B. caudatus f. *apsteini* Fadeew, 1925	Ca, P, Re, Ri, Sw	N, NE, C
B. dichotomus f. *reductus* Koste and Shiel, 1980	Ca, P, Re, Sw	N, NE, C
B. diversicornis (Daday, 1883)	Ca, L, P, Re, Ri, Sw	N, NE, C
B. donneri Brehm, 1951	Ca, L, P, Re, Ri, Sw	N, NE, C
B. durgae Dhanapathi, 1974	P	C
B. falcatus Zacharias, 1898	Ca, L, P, Ps, Re, Rf, Ri, Sw, T	N, NE, C, S
B. forficula Wierzejski, 1891	Ca, L, P, Ps, Re, Ri, Sw, T	N, NE, C, S
B. forficula f. *minor* Voronkov, 1913	Ri, Sw	N
B. murphyi Sudzuki, 1989	Ca, Rf	N, NE
B. nilsoni Ahlstrom, 1940	L, Ri	N, C
B. quadridentatus Hermann, 1783	Ca, L, P, Re, Rf, Ri, Sw	N, NE, C, S
B. quadridentatus f. *brevispinus* Ehrenberg, 1832	Ca, P, Re, Ri	N, NE, C
B. quadridentatus f. *cluniorbicularis* Skorikov, 1894	Rf, Ri	N, NE, C
B. quadridentatus f. *melhemi* (Barrois and Daday, 1894)	P, Ri	N, C, S
B. quadridentatus f. *mirabilis* (Daday, 1897)	Ca, P, Rf	N, S
B. rotundiformis Tschugunoff, 1921	Ca, P, Ps, Ri	NE, C, S
B. rubens Ehrenberg, 1838	Ca, P, Re, Rf, Ri	N, NE, C
B. urceolaris (Müller, 1773)	Ca	C
Keratella cochlearis (Gosse, 1851)	Ca, L, P, Re, Rf, Ri, Sw	N, NE, C, S
K. edmondsoni (Ahlstrom, 1943)	Ca, L, P, Re, Sw	NE
K. lenzi Hauer, 1953	Ca, L, P, Re, Ri, Sw	N, NE, C, S
K. lenzi f. *heliaca* Bērziņš, 1955	L, P, Re, Ri, Sw	N, NE, C
K. procurva (Thorpe, 1891)	Ca, Sw	N, NE, C
K. tecta (Gosse, 1851)	Ca, L, P, Re, Ri, Sw	N, NE, C
K. tropica (Apstein, 1907)	Ca, L, P, Ps, Re, Rf, Ri, Sw, T	N, NE, C, S
Plationus patulus (Müller, 1786)	Ca, L, P, Ps, Re, Rf, Ri, Sw	N, NE, C, S
Platyias quadricornis (Ehrenberg, 1832)	Ca, P, Ps, Re, Rf, Ri, Sw	N, NE, C, S

Ca, canal; L, lake; P, pond; Re, reservoir; Ri, river; Sw, swamp; N, North; NE, Northeast; C, Central; S, South; Ps, peat swamp; Rf, rice field; T, temporary pond.

water or even marine taxa, especially in *Keratella* (Segers and De Smet, 2008; Fontaneto *et al*., 2008).

The diversity of brachionid rotifers in lentic ecosystems (lakes, peat swamps, ponds, reservoirs, rice fields, swamps and temporary ponds, n=37 taxa) was higher than in lotic ecosystems (canals and rivers, n=32 taxa). Two taxa, *Brachionus angularis* f. *bidens* and *B. caudatus*, were found only in lotic ecosystems, while *B. durgae*, *B. murphyi*, *B. urceolaris*, *Keratella edmondsoni* and *K. procurva* were recorded only from lentic ecosystems. This result supports earlier studies indicating that *Brachionus* are planktonic rather than periphytic or benthic animals, although there are notorious exceptions, *e.g. B. quadridentatus* (Pejler and Bērziņš, 1989). The most eurytopic brachionid rotifers were *B. angularis* (9 habitat types), *K. tropica* (9 habitat types) and *B. forficula* and *B. falcatus* (8 habitat types). Four taxa were found in distinct habitat types: *B. angularis* f. *bidens* occurred in rivers, *B. caudatus* inhabited a canal, *B. durgae* and *B. urceolaris* were restricted to ponds.

During the rainy season, diversity of the brachionid rotifers was higher than during the dry season. Two species (*Brachionus budapestinensis* and *B. murphyi*) were found only during the dry season whereas four species and two infrasubspecific forms (*B. angularis* f. *bidens*, *B. bennini*, *B. caudatus*, *B. durgae*, *B. forficula* f. *minor* and *B. urceolaris*) were recorded only during the rainy season.

Results of CCA indicate that most of the variation in environmental variables (11.9%) is explained in the first axis (eigen value=0.479). Correlation between the first axis and species-environmental variables was 0.857. The second axis (eigen value=0.067) accounted for 1.7% of the variation in the data set. Correlation between the second axis and species-environmental variables was 0.419. The species and their abundance were significantly correlated with three environmental variables: temperature (inter-set correlation=0.42), conductivity (inter-set correlation=0.76) and salinity (inter-set correlation=0.85) (Monte Carlo test, P= 0.01). The two latter variables seem

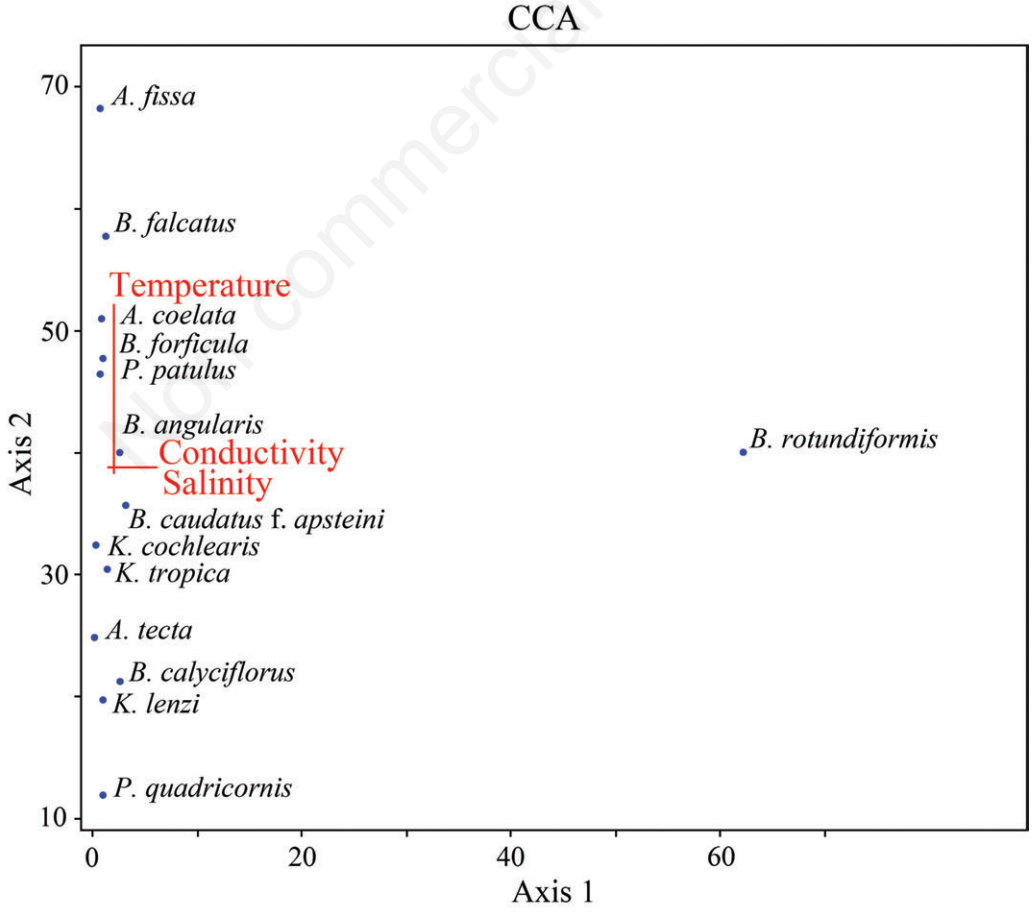

Fig. 2. Canonical correspondence analysis ordination of rotifer species and environmental variables. Species are represented by circles and environmental variables by lines.

to have the same weight in their correlations with species data. Generally, salinity reflects the concentration of ions in the water, which can be measured in terms of electrical conductivity. Taxa that showed different affinities or tolerance to the three environmental variables were also determined (Fig. 2). Two species (*Anuraeopsis fissa* and *B. falcatus*) were positively correlated with temperature, while two species (*Platyias quadricornis* and *K. lenzi*) were negatively correlated with it. This result is in accordance with another report, in which *A. fissa* has been found in warmer water in New Zealand water bodies (Duggan *et al*., 2001). Also, Segers (2007) indicated that all *Anuraeopsis* species are warm water animals. Only one species, *B. rotundiformis*, was strongly positively correlated with conductivity and salinity while the other species were negatively correlated with these variables. Regarding habitat, low salinity values (0.04-2.73 PSU) were measured in some canals and a river locating in the central part of Thailand (*i.e.* Amphawa, Nakwang, and Sapandam canals, and Tha Chin river). This is a consequence of these canals and river being influenced by their closeness to the gulf of Thailand.

Key to the recorded Thai brachionid rotifers

Previous publications (Sanoamuang *et al*., 1995; Chittapun and Pholpunthin, 2001; Sanoamuang and Savatenalinton, 2001; Chittapun *et al*., 2002, 2007; Segers *et al*., 2004) and the present study of original material reveal that the Thai brachionid fauna consists of 37 species belonging to 5 genera. This includes *Anuraeopsis* (3 species), *Brachionus* (23 species), *Keratella* (9 species), *Plationus* (1 species) and *Platyias* (1 species). Keys to the genera and species of the Thai Brachionidae are provided below.

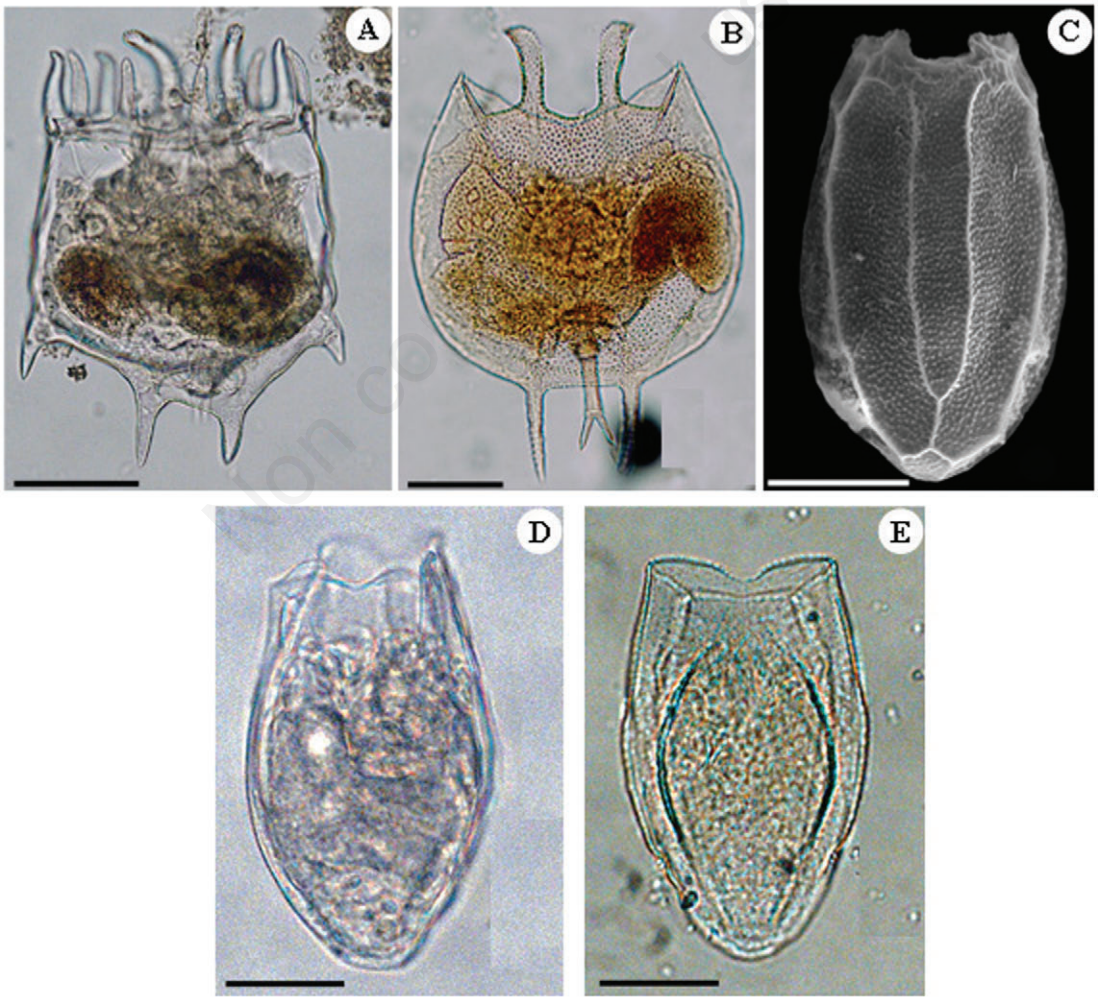

Fig. 3. A) *Plationus patulus* (Müller); B) *Platyias quadricornis* (Ehrenberg); C) *Anuraeopsis coelata* (De Beauchamp); D) *A. fissa* (Gosse); and E) *A. navicula* (Rousselet). Scale bars: A,B) 50 µm; C-E) 20 µm.

Key to the genera of Brachionidae in Thailand

1. Foot present (in contracted material the foot may be completely retracted within the lorica. However, presence of a foot can always be ascertained by the relatively large foot opening in such specimens) 2
   Foot absent.....4
2. Foot jointed (pseudosegmented), two toes 3
   Foot annulated, retractile, two toes..... ***Brachionus* Pallas**
3. Ventral head aperture with 4 spines, dorsal head aperture with six spines. Lorica asymmetrical ..... ***Plationus* Segers, Murugan and Dumont**
   [(One species in Thailand, *Plationus patulus* (Müller)] (Fig. 3A)
   Ventral head aperture without spines, dorsal with two spines .....***Platyias* Harring**
   [(One species in Thailand, *Platyias quadricornis* (Ehrenberg)](Fig. 3B)
4. Dorsal plate with 6 anterior spines; 1, 2 or no posterior spines .....***Keratella* Bory de St Vincent**
   Lorica without spines; species relatively small ..... ***Anuraeopsis* Lauterborn**

Genus *Anuraeopsis* Lauterborn

This genus contains 10species, 3 of which (*Anuraeopsis coelata*, *A. fissa* and *A. navicula*) have been recorded from Thailand. The former two were found during the present study, the latter was recorded during previous studies only. In general, species are identified by means of body shape, shape of occipital edge of the lorica, surface pattern of lorica and body length. The description of the basic morphological structures of *Anuraeopsis* spp. was presented in Koste and Shiel (1987).

Key to the known species of genus *Anuraeopsis* Lauterborn in Thailand

1. Dorsal plate with double ridges connected in posterior third, lateral margin with facets (Fig. 3C) ....***A. coelata* (De Beauchamp)**
   Dorsal plate without ridges and facets.....2
2. Head aperture margin smooth, lorica oval shaped (Fig. 3D) .....***A. fissa* (Gosse)**
   Head aperture margin with fine serration, lorica navicula-shaped (Fig. 3E) .....***A. navicula* Rousselet**

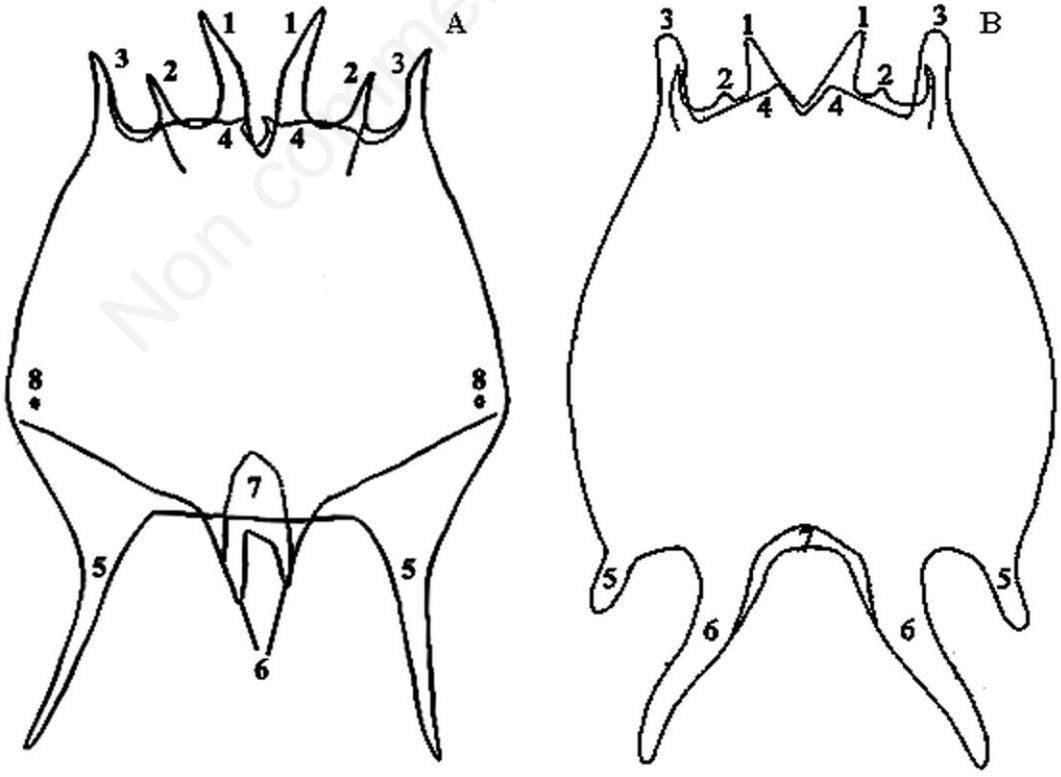

Fig. 4. Lorica of genus *Brachionus*: A) *Brachionus quadridentatus* Hermann and B) *B. srisumonae* Segers, Kotethip and Sanoamuang. 1, anterior median spines; 2, anterior intermediate spines; 3, anterior lateral spines; 4, ventral anterior margin; 5, posterior lateral spines; 6, spines of foot opening; 7, foot opening; 8, lateral antennae.

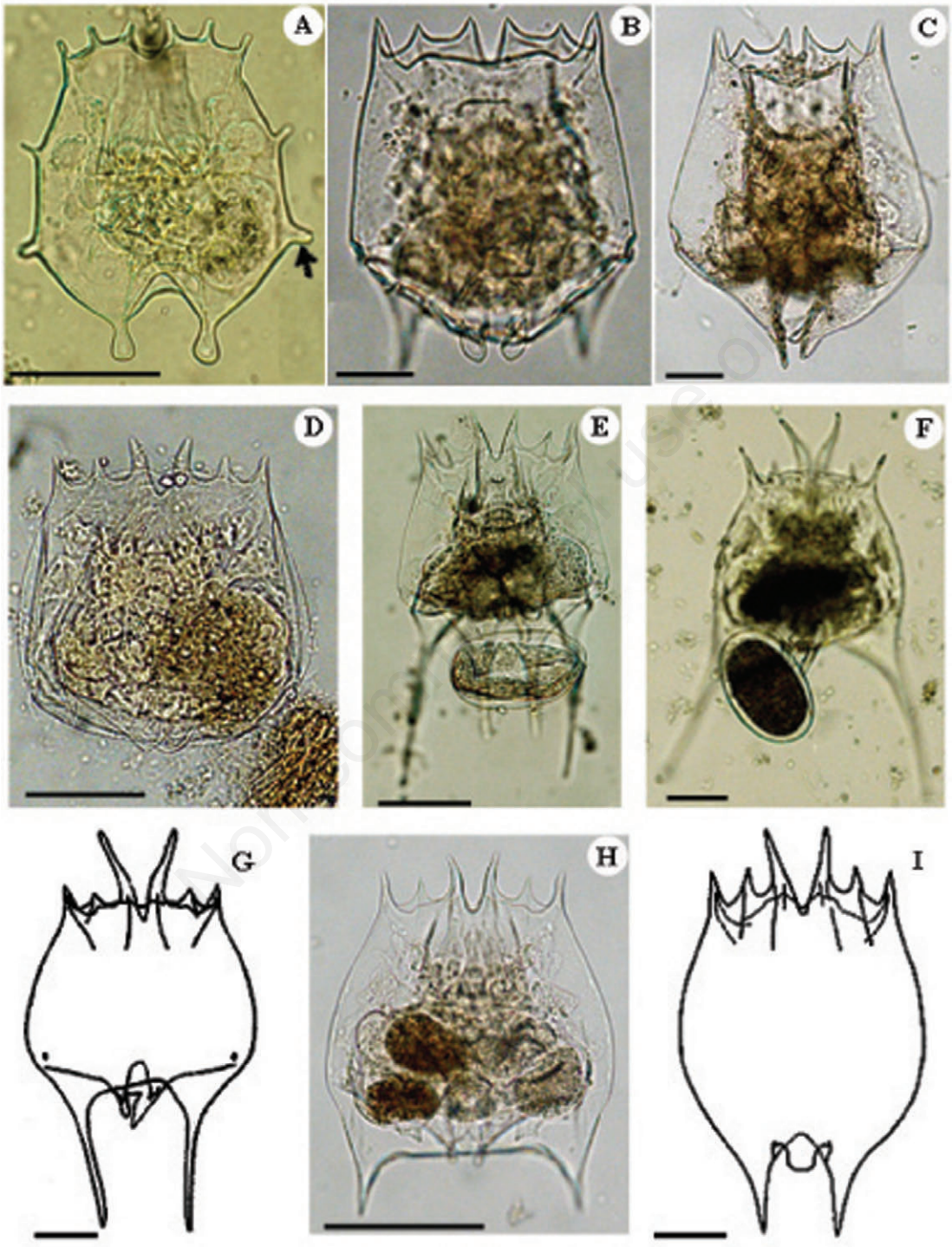

Fig. 5. A) *Brachionus donneri* Brehm; B) *B. bidentatus* f. *bidentatus* Anderson; C) *B. bidentatus* f. *inermis* (Rousselet); D) *B. quadridentatus* f. *cluniorbicularis* Skorikov; E) *B. quadridentatus* f. *mirabilis* (Daday); F) *B. quadridentatus* f. *melhemi* (Barrois and Daday); G) *B. quadridentatus* f. *quadridentatus* Hermann; H) *B. quadridentatus* f. *brevispinus* Ehrenberg; and I) *B. variabilis* Hempel. Scale bar: 50 µm. I) Modified after Ahlstrom (1940).

Genus *Brachionus* Pallas

Brachionus is one of the first described genera of monogonont rotifers. The genus is considered cosmopolitan since its members can be found widely throughout all geographical regions of the world (Segers, 2007, 2008). It is a diverse group of brachionid rotifers spread both globally as well as in Thai water bodies. The various species can be identified by the number of anterior dorsal spines (0, 2, 4 and 6), relative size and shape of anterior ventral spines, shape of ventral head aperture margin, position of foot aperture (terminal or ventral), shape of dorsal projection over foot aperture and special features. An overview of these features is presented in Fig. 4.

For the key to species, the publication presenting its description is given after the scientific name within parenthesis.

Key to the known species of genus *Brachionus* Pallas in Thailand

1. Dorsal anterior margin with six spines, median, intermediate and lateral spines present 2
   Dorsal anterior margin with two or four spines, only median or median and lateral spines present 20
2. Lateral antennae on conical protuberance, ventral anterior margin with four rod-shaped spines (Fig. 5A).. .....***B. donneri*** **Brehm** (Koste and Shiel, 1987)
   Lateral antennae not on conical protuberance, ventral anterior margin without spines 3
3. Sheath or tubular sheath surrounding foot opening..4
   No definite foot sheath; foot aperture in same plane as lorica 10
4. Antero-lateral lorica spines longer than, or at least as long as, antero-middorsal spines 5
   Antero-middorsal lorica spines longer than antero-lateral spines 6
5. Postero-lateral spines present (Fig. 5B).….***B. bidentatus*** **f.** ***Bidentatus*** **Anderson** (*Plationus patulus* will key out here if the retracted foot was misinterpreted as being annulated rather than pseudosegmented) (Kuczynski, 1991)
   Postero-lateral spines absent (Fig. 5C) ....***B. bidentatus*** **f.** ***inermis*** **(Rousselet)** .....(Kuczynski, 1991)
6. Postero-lateral spines developed 7
   Postero-lateral spines absent, posterior corners rounded (Fig. 5D).....***B. quadridentatus*** .....**f.** ***cluniorbicularis*** **Skorikov** .....(Koste and Shiel, 1987)
7. Foot opening spines rather short.....8
   Foot opening spines rather long, at least half as long as postero-lateral spines extending backward from the lorica, postero-lateral spines long, mostly divergent (Fig. 5E) .....***B. quadridentatus*** **f.** ***mirabilis*** .....**(Daday)** (Ahlstrom, 1940)
8. Postero-lateral spines parallel or convergent.....9
   Postero-lateral spines rather long and divergent (Fig. 5F).....***B. quadridentatus*** **f.** ***melhemi*** .....**(Barrois and Daday)** (Koste and Shiel, 1987)
9. Median spines longest, 2-3 times longer than lateral spines; lorica relatively large (Fig. 5G) ....***B. quadridentatus*** **f.** ***quadridentatus*** .....**Hermann** (Ahlstrom, 1940)
   Median spines slightly longer than lateral spines, postero-lateral spines of moderate length; lorica relatively small (Fig. 5H).....***B. quadridentatus*** **f.** ***brevispinus*** .....**Ehrenberg** (Koste and Shiel, 1987)
10. Sub-squarish or tongue-shaped extension of dorsal plate overhanging foot opening (Fig. 5I)..... .....***B. variabilis*** **Hempel** (Ahlstrom, 1940)
    Sub-square extension over foot aperture absent 11
11. Antero-lateral spines rod shaped, postero-lateral spines short and terminally bifid (Fig. 6A)..... .....***B. srisumonae*** **Segers, Kotethip and** .....**Sanoamuang** (Segers *et al*., 2004)
    Antero-lateral spines acute at tip 12
12. Antero-intermediate spines longest, 3-5 times longer than median spines, spines of foot opening long (Fig. 6B).....***B. falcatus*** **Zacharias** (Kuczynski, 1991)
    Antero-intermediate spines not longest 13
13. Ventral anterior margin separated into four lobes or four blunt projections, living in saline water (Fig. 6C) .....***B. rotundiformis*** **Tschugunoff** .....(Ciros-Pérez *et al.*, 2001
    Ventral anterior margin not separated into four lobes or projections 14
14. Foot opening somewhat ventral, large and more or less round, intermediate spines rudimentary, sometimes epizoic on Cladocera (Fig. 6D) .....***B. sessilis*** **Varga** .....(Koste and Shiel, 1987)
    Foot opening terminal, free-swimming 15
15. Ventral head aperture margin smoothly undulated, with a median sinus, and two pairs of lobes set closely together, lorica relatively large (Fig. 6E) ...***B. durgae*** .....**Dhanapathi** (Dhanapathi, 1974)
    Ventral head aperture margin undulated, or with a single pair of projections and a central sinus 16
16. Lateral projections of the foot aperture diverging, convoluted, dorsal lorica with facets (Fig. 6F) ..***B. kostei*** .....**Shiel** (Koste and Shiel, 1987)
    Lateral projections of the foot aperture absent or rudimentary 17
17. Lorica with a pattern of wavy lines 18
    Lorica usually lightly stippled 19
18. Lorica truncate posteriorly in lateral view (Fig. 6G-H)…….…***B. bennini*** **(Leissling)** (Ahlstrom, 1940)
    Lorica pointed posteriorly in lateral view (Fig. 6I-J) . .....***B. urceolaris*** **(Müller)** (Ahlstrom, 1940)

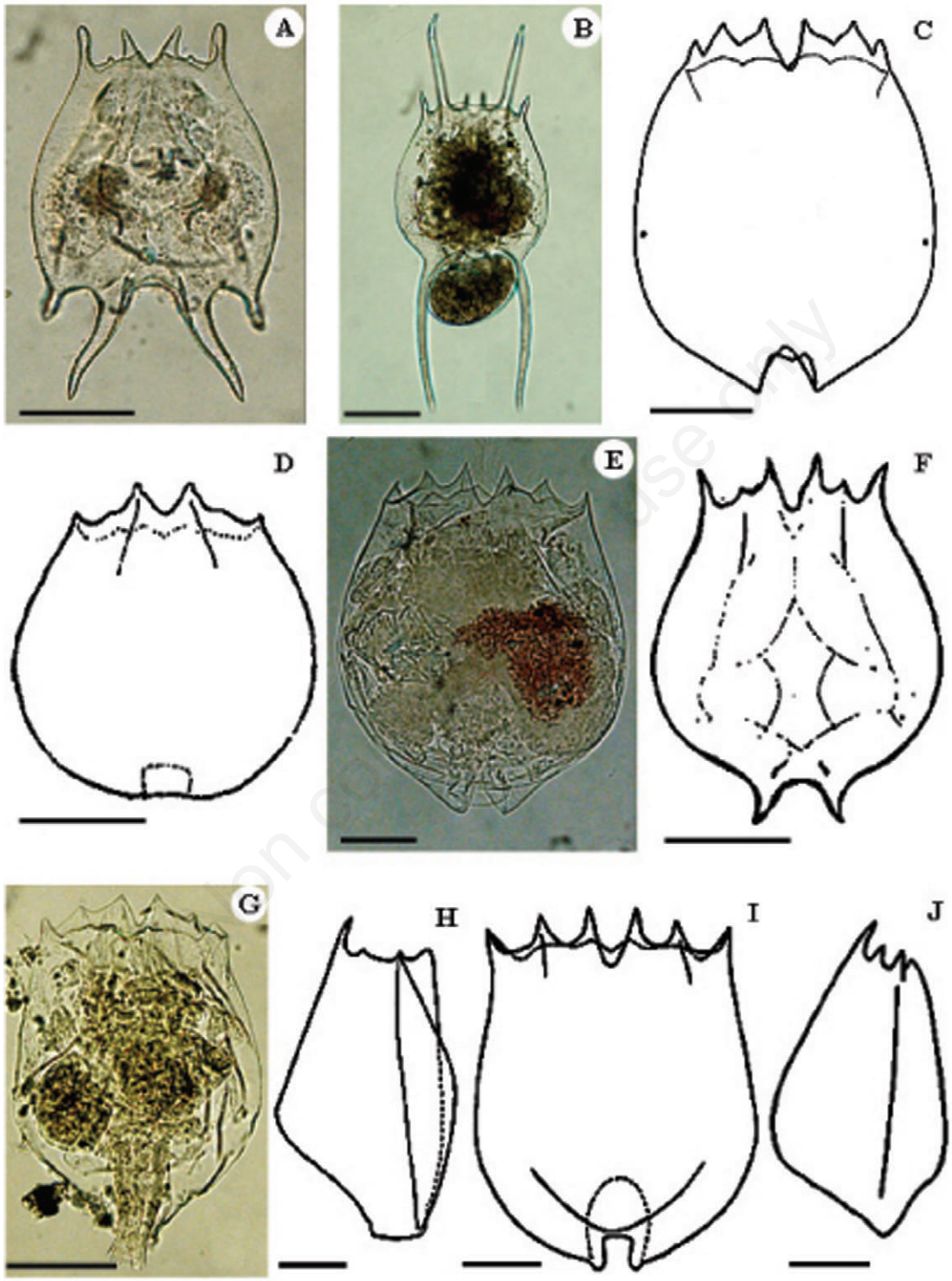

Fig. 6. A) *Brachionus srisumonae* Segers, Kotethip and Sanoamuang; B) *B. falcatus* Zacharias; C) *B. rotundiformis* Tschugunoff; D) *B. sessilis* Varga; E) *B. durgae* Dhanapathi; F) *B. kostei* Shiel; G,H) *B. bennini* (Leissling); G) ventral view; H) lateral view; I,J) *B. urceolaris* (Müller), I) dorsal view, and J) lateral view. Scale bar: 50 µm. D-F) Modified after Koste and Shiel (1987) and H-J) after Ahlstrom (1940).

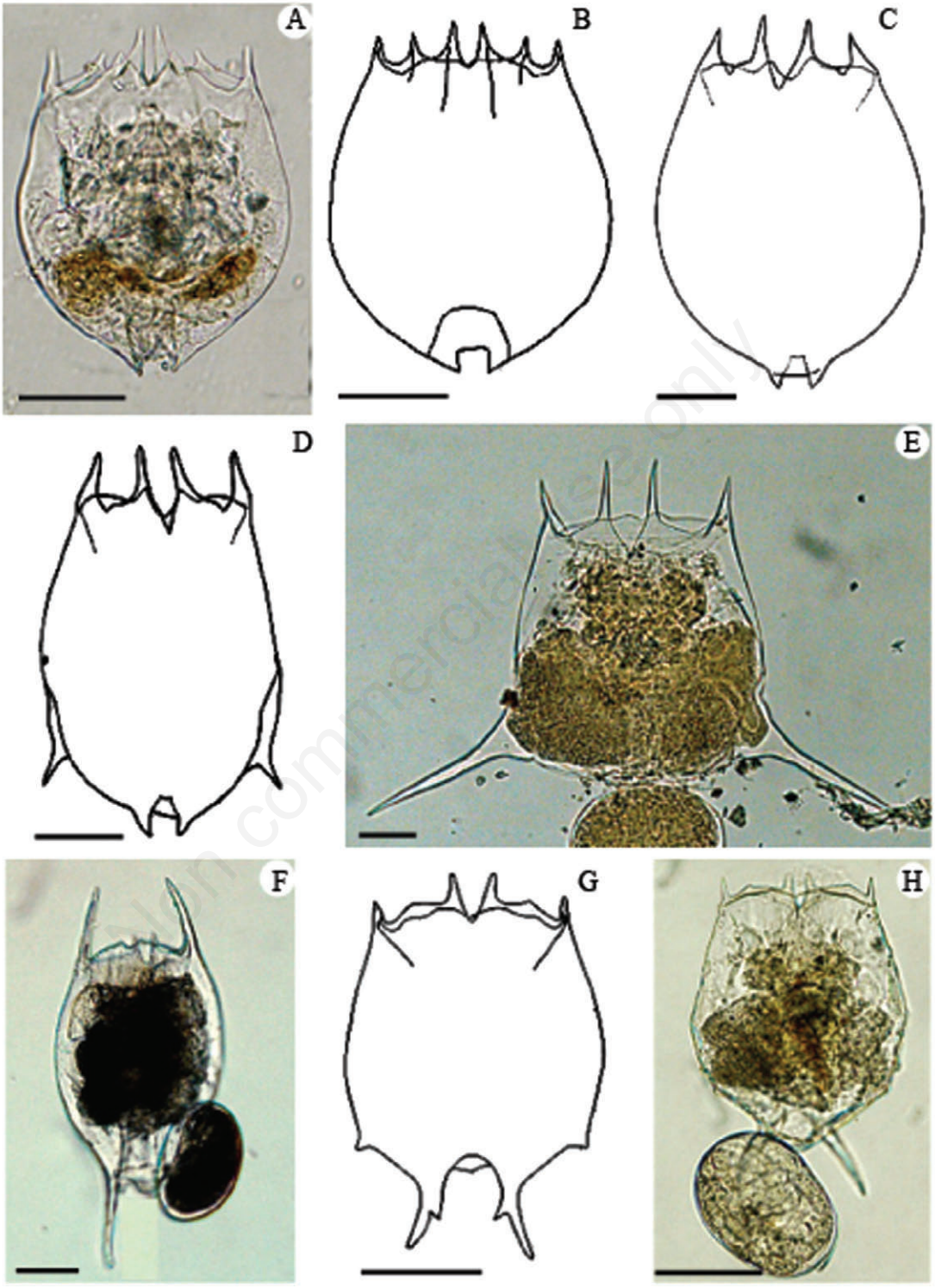

Fig. 7. A) *Brachionus rubens* Ehrenberg; B) *B. nilsoni* Ahlstrom; C) *B. calyciflorus* f. *calyciflorus* Pallas; D) *B. calyciflorus* f. *anuraeiformis* Brehm; E) *B. calyciflorus* f. *amphiceros* Ehrenberg; F) *B. diversicornis* (Daday); G) *B. caudatus* f. *aculeatus* Hauer; H) *B. caudatus* f. *apsteini* Fadeew. Scale bars: 50 µm.

19. Median and intermediate spines showing a narrow anterior part, rounding outward and forming a broad base. Ventral foot aperture nearly parallel-sided (Fig. 7A).....***B. rubens*** **Ehrenberg** (Ahlstrom, 1940)
    Median and intermediate spines thin, acutely pointed. Ventral foot aperture large, with diverging lateral margins (Fig. 7B) ***B. nilsoni*** **Ahlstrom** (Ahlstrom, 1940)
20. Anterior dorsal margin with 4 spines, median and lateral 21
    Anterior dorsal margin with 2 well-developed spines, median; if present, intermediate and/or lateral rudimentary 30
21. Lorica indistinctly separated into dorsal and ventral plates, anterior dorsal spines elongate, with broad basis 22
    Lorica with clear dorsal and ventral plates, lateral margins sharp 24
22. Postero-lateral spines developed, anterior spines more or less equal 23
    Postero-lateral spines absent, antero-dorsal median spines longer than antero- lateral spines (Fig. 7C) .....
    .....***B. calyciflorus*** **f.** ***calyciflorus*** **Pallas**
    .....(Ahlstrom, 1940)
23. Postero-lateral spines short (22-46 mm) (Fig. 7D) ....
    .....***B. calyciflorus*** **f.** ***anuraeiformis*** **Brehm**
    .....(Kuczynski, 1991)
    Postero-lateral spines very long (127-195 mm) (Fig. 7E).....***B. calyciflorus*** **f.** ***amphiceros*** **Ehrenberg**
    .....(Kuczynski, 1991)
24. Antero-lateral spines much longer than median spines, foot opening spines close together at their point of origin (Fig. 7F) .....***B. diversicornis*** **(Daday)**
    .....(Ahlstrom, 1940)
    Antero-median spines longer than lateral spines or these spines are of about equal length 25
25. Foot opening spines present 26
    Foot opening spines absent.....29
26. Extension of dorsal plate overhanging foot opening, posterior spines quite close together.....27
    No extension of dorsal plate over foot opening, foot opening spines wide apart at base, and with knee-like swellings on inner side 28
27. An accessory spur-like spine arising from inner dorsal side of each foot opening spine (Fig. 7G) .....
    ....***B. caudatus*** **f.** ***aculeatus*** **Hauer** (Ahlstrom, 1940)
    Accessory spine not developed, foot opening spines smooth (Fig. 7H).....***B. caudatus*** **f.** ***apsteini*** **Fadeew**
    .....(Ahlstrom, 1940)
28. Lorica relatively large, foot opening spines equal in length (Fig. 8A) .***B. forficula*** **f.** ***forficula*** **Wierzejski**
    .....(Ahlstrom, 1940)
    Lorica relatively small, foot opening spines relatively short, sometimes asymmetrical (Fig. 8B).....
    .....***B. forficula*** **f.** ***minor*** **Voronkov**
    .....(Koste and Shiel, 1987)
29. Equal antero-dorsal spines, lateral spines straight, lorica firm with cuticular ridges (Fig. 8C) .....
    .....***B. budapestinensis*** **Daday** (Ahlstrom, 1940)
    Unequal antero-dorsal spines, lateral spines directed outwards, lorica relatively soft (Fig. 8D) .....
    .....***B. murphyi*** **Sudzuki** (Sudzuki, 1989)
30. Antero median spines long, foot opening spines long
   31
    Antero median spines short, foot opening spines short or absent.....32
31. Lorica oval-shaped antero intermediate spines absent, but antero lateral short (Fig. 8E) .....***B. caudatus***
    ....**f.** ***caudatus*** **Barrois and Daday** (Ahlstrom, 1940)
    Lorica pear-shaped antero intermediate spines short, but antero lateral absent (Fig. 8F).....***B. dichotomus***
    f. ***reductus*** **Koste and Shiel** (Koste and Shiel, 1980)
32. Foot opening with spines lyre-shaped, antero intermediate and lateral spines of lorica rudimentary (Fig. 8G)
    .....***B. lyratus*** **Shephard** (Koste and Shiel, 1980)
    Foot opening spines, if present, not lyre-shaped, antero intermediate and lateral spines of lorica absent.....33
33. Small but sharp, pointed lateral projections of the foot aperture present (Fig. 8H) .....***B. angularis***
    .....**f.** ***bidens*** **Plate** (Ahlstrom, 1940)
    Pointed lateral projections of the foot aperture absent, if present, rudimentary (Fig. 8I).....***B. angularis***
    .....**f.** ***angularis*** **Gosse** (Ahlstrom, 1940)

Genus *Keratella* Bory de St Vincent

Nine species have been recorded in Thailand. One of them (*K. taksinensis*) is endemic to the country. Species are identified based upon the following characters: number of anterior spines, position of posterior spine(s) and pattern of facets on the dorsal lorica.

For the key to species, the publication presenting its description is given after the scientific name within parenthesis.

Key to the known species of genus *Keratella* Bory de St. Vincent in Thailand

1. Dorsal plate with median ridge.....2
   Dorsal plate without median ridge.....3
2. Posterior spine always present, pointed slightly ventrally; species relatively large, (Fig. 9A) .....
   .....***K. cochlearis*** **(Gosse)** (Ahlstrom, 1943)
   Posterior spine generally absent, when present, short and sharply set-off from the lorica, held in body axis; species relatively small; anterior spines mostly relatively short, (Fig. 9B) .....***K. tecta*** **(Gosse)**
   .....(Ahlstrom, 1943)
3. Central row of facets present 4
   Central facets not in a row but as an irregular group

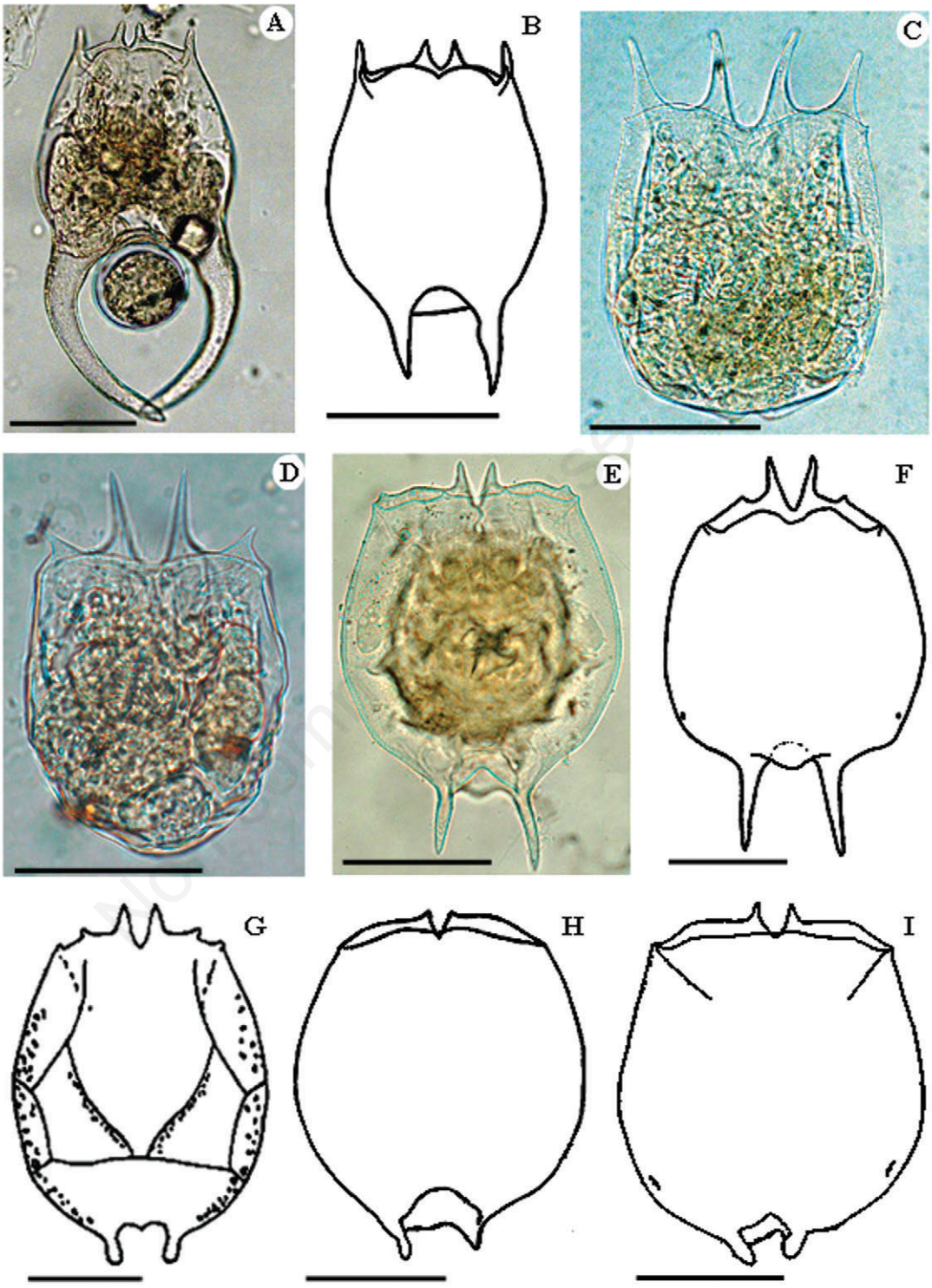

Fig. 8. A) *Brachionus forficula* f. *forficula* Wierzejski; B) *B. forficula* f. *minor* Voronkov; C) *B. budapestinensis* Daday; D) *B. murphyi* Sudzuki; E) *B. caudatus* f. *caudatus* Barrois and Daday; F) *B. dichotomus* f. *reductus* Koste and Shiel; G) *B. lyratus* Shephard; H) *B. angularis* f. *bidens* Plate; I) *B. angularis* f. *angularis* Gosse. Scale bar: 50 µm. G) Modified after Koste and Shiel (1987).

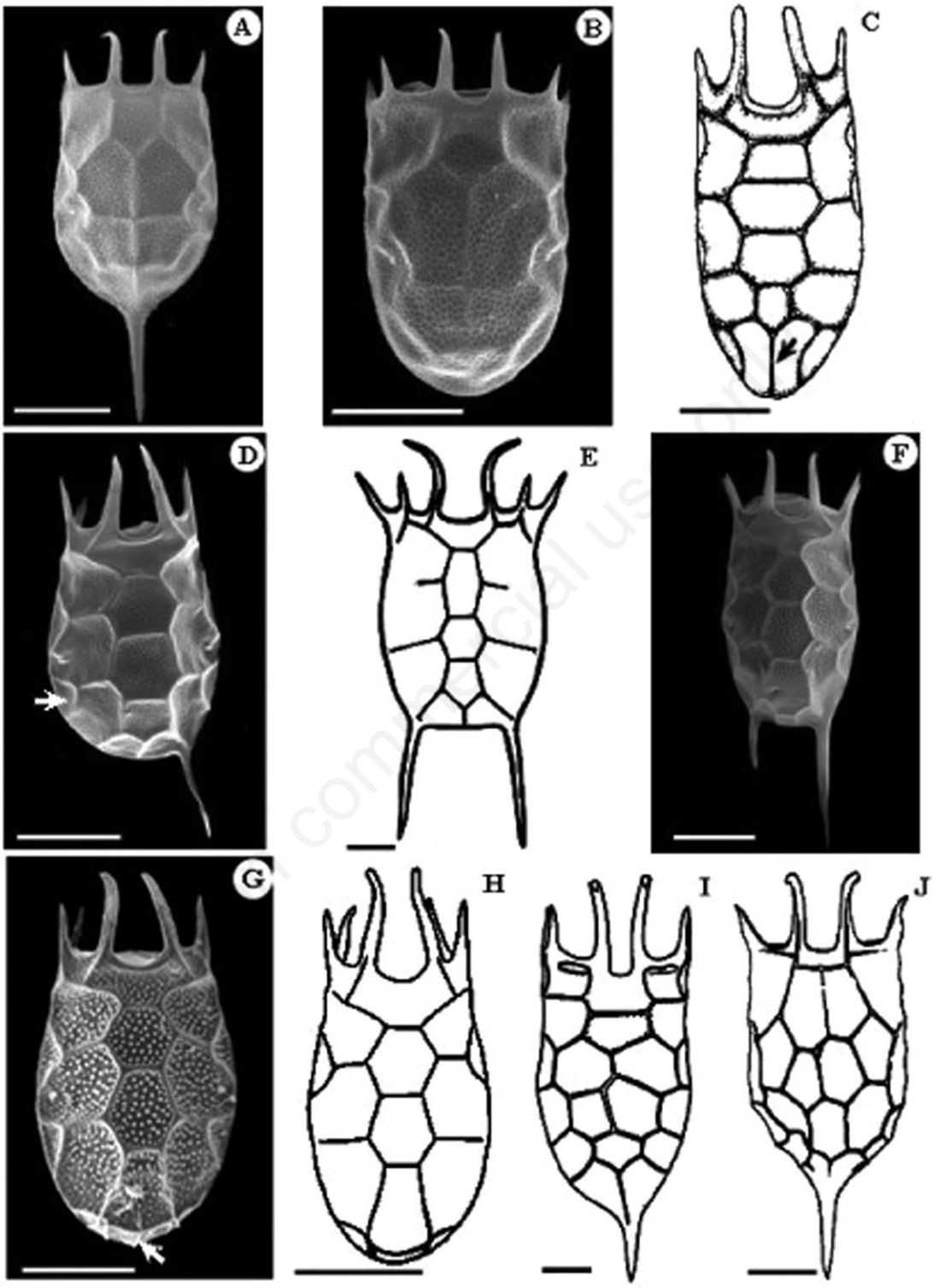

Fig. 9. A) *Keratella cochlearis* (Gosse); B) *K. tecta* (Gosse); C) *K. taksinensis* Chitapun, Pholpunthin and Segers; D) *K. procurva* (Thorpe); E) *K. edmondsoni* Ahlstrom; F) *K. tropica* (Apstein); G) *K. lenzi* f. *heliaca* Bērziņš; H) *K. lenzi* Hauer; I) *K. mixta* (Oparina-Charitonova); J) *K. javana* Hauer. Scale bar: 30 µm. C) Modified after Chittapun *et al*. (2002); I) after Ahlstrom (1943); J) after Koste and Shiel (1987). Arrowheads in C,D) and G) indicate median line, marginal plaque and short median ridge, respectively.

of facets.....9
4. Posterior median facet terminates in a short median line 5
   Posterior median facet does not terminate in a median line 7
5. Median row of facets consists of three enclosed facets 6
   Median row of facets consists of four enclosed facets (Fig. 9C).....***K. taksinensis*** **Chittapun, Pholpunthin .....and Segers** (Chittapun *et al*., 2002)
6. Marginal plaques present, posterior spines asymmetric and straight (Fig. 9D).....***K. procurva*** **(Thorpe)** .....(Ahlstrom, 1943)
   Marginal plaques absent, posterior spines symmetric and characteristically bent (Fig. 9E) ..***K. edmondsoni*** .....**Ahlstrom** (Ahlstrom, 1943)
7. Lorica posteriorly with terminal row of minute facets; no posterior spines 8
   Lorica with a small, median posterior facet; lorica mostly with asymmetrical posterior spines (Fig. 9F) . .....***K. tropica*** **(Apstein)** (Ahlstrom, 1943)
8. Short median ridge near the terminal end of the lorica present (Fig. 9G) .....***K. lenzi*** **f.** ***heliaca*** **Bērziņš** .....(Koste and Shiel, 1987)
   Short median ridge near the terminal end of the lorica absent (Fig. 9H).....***K. lenzi*** **Hauer** .....(Koste and Shiel, 1987)
9. Dorsal plate with six enclosed central facets (Fig. 9I) .....***K. mixta*** **(Oparina-Charitonova)** .....(Ahlstrom, 1943)
   Dorsal plate with nine enclosed central facets (Fig. 9J) .....***K. javana*** **Hauer** (Ahlstrom, 1943)

CONCLUSIONS

Previous publications and the present study reveal that the Thai brachionid fauna consists of 49 taxa, 37 species and 12 infrasubspecific forms. This paper developed keys to the genera and species. In addition to this, useful taxonomic characters were identified as being: type of foot, the number of occipital spines, relative size and shape of occipital spines, shape of occipital edge of the lorica, surface pattern on the dorsal lorica, general body shape, position of posterior spine, position of foot aperture and shape of dorsal projection over foot aperture. In future taxonomic works, we suggest investigating external morphology together with trophi structure, one of the most useful taxonomic characters, and using molecular tools to solve the taxonomic problems in some species-groups (*e.g. B. caudatus* and *B. quadridentatus* groups).

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