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Nannophya koreana sp. nov. (Odonata: Libellulidae): A new dragonfly species previously recognized in Korea as the endangered pygmy dragonfly *Nannophya pygmaea* Rambur

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A new dragonfly species, *Nannophya koreana* sp. nov., is described from Korea on the basis of morphology and mitochondrial cytochrome oxidase c subunit I (*COI*) gene sequences. *Nannophya* materials from Korea and other areas in Southeast Asia were compared. The new species was previously recognized in Korea as the endangered pygmy dragonfly *Nannophya pygmaea* Rambur, 1842, which is widely distributed in insular and peninsular Southeast Asia. However, male adults of the *Nannophya* population in Korea could be distinguished from other *N. pygmaea* populations by the presence of a thick, incomplete black stripe on the lateral synthorax that terminated at half-length (*vs*. continuous to wing base), light orange (*vs*. red) anal appendages, and 4−5 (*vs*. 2−3) black teeth on the ventral superior appendages. In addition, the body length of *N. koreana* was generally larger (1.2−1.4 times) than that of *N. pygmaea*, regardless of life stage. *COI* gene sequences from the two groups exhibited substantial genetic differences (>12%), thereby sufficiently substantiating their differentiation. The taxonomic status, distribution, and habitat of the new species are discussed.

Keywords: description, endangered species, Libellulidae, *Nannophya koreana* sp. nov., *Nannophya pygmaea*, Odonata

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Introduction

The late Professor Chang Whan Kim (Korea University, Seoul, Korea) collected adult specimens of *Nannophya* Rambur (Libellulidae) dragonflies from Songnisan (Mt.) in Boeun, Chungcheongbuk-do, Korea, in 1957, which represented the first collection of a *Nannophya* taxon in Korea, and identified the specimens as *Nannophya pygmaea* Rambur, 1842 (Cho, 1958; 1969; Bae *et al*., 1999). Unfortunately, the specimens have not been found in the Korea University Entomological Museum, where the specimens were supposed to be deposited. Since then, the distribution and ecology of the species has been intensively investigated, and the species has been documented from more than 20 localities throughout South Korea, including Gokseong in Jeollanam-do, Wangdungjae of Jirisan (Mt.) in Gyeongsangnam-do, Moojechineup (high moor) of Jeongjoksan (Mt.) in Ulsan, Mungyeong in Gyeongsanbuk-do, Boryeong in Chungcheongnam-do, and Muuido Island in Incheon (Bae *et al*., 1999; Lee *et al*., 2008; Yoon *et al*., 2010).

The genus *Nannophya* is generally recognized to include some of the world's smallest dragonflies and contains six species, with four in Australia, one in the Himalayas, and one (*N. pygmaea*) being widely distributed in the Orient and Australia, including in tropical and subtropical areas of Southeast Asia, China, Japan, India, and northern Australia (Steinmann, 1997; Theischinger, 2003). Indeed, because all *Nannophya* specimens in Korea were identified as *N. pygmaea*, South Korea was previously recognized as the northernmost limit of the distribution of this species (Bae *et al*., 1999).

Furthermore, the *Nannophya* population in Korea, which was recognized as *N. pygmaea*, was listed in the

Red Data Book of Korea (National Institute of Biological Resources, 2013) and was protected under the law as a Category II Endangered Species. Thus, the distribution and habitat (Bae *et al*., 1999; Lee, 2005), life history and development (Kim *et al*., 2006; 2009a; 2009b; 2010), and conservation and habitat restoration (Lee *et al*., 2008) of the population in Korea have been studied intensively.

However, comprehensive morphological and molecular examinations of *N. pygmaea* materials sampled from Northeast and Southeast Asia have revealed that the Korean population can be distinguished from other populations of *N. pygmaea*, which was originally described from insular Indonesia (Ambon and Borneo) and China (Kirby, 1890). Thus, the population in Korea is hereby described as a new species.

Materials and Methods

Materials and description

Adult dragonflies were collected using sweep nets from vegetation growing in wetlands or in pool areas of streams, and larvae were collected from the shallow areas of wetlands or stream pools using hand nets. The adult specimens were either pinned or preserved in 80% ethanol, and all larvae were preserved in 80% ethanol. The type specimens of *N. koreana* sp. nov. were deposited in the National Institute of Biological Resources (NIBR) in Incheon, Korea and in the Korea University Entomological Museum (KU) in Seoul, Korea. The terminology used in the present study generally follows Sugimura *et al*. (2001).

Phylogenetic analyses and genetic divergence calculations

Total genomic DNA extraction, PCR, and sequencing of the *COI* gene were conducted following by a protocol used by a recent Ephemeroptera barcoding study (Suh *et al*., 2019), and both universal and newly designed internal primer sets were used for PCR amplification from long-term preserved specimens. A total of 33 sequences were used for the phylogenetic reconstruction, including six newly obtained sequences from specimens from Korea, Taiwan, and Singapore and 26 GenBank sequences from specimens from Korea, Japan, Twain, China, Laos, Indonesia, Vietnam, Singapore, and Malaysia. A *COI* sequence from a dragonfly *Tetrathemis platyptera* Selys (Libellulidae) (KC122235) was included as an outgroup. All sequences (450 bp) were aligned using the ClustalW multiple sequences alignment package (Thompson *et al*., 1994) in BioEdit 7.1.9 (Hall, 1999), and the genetic divergence of the sequences and overall mean distance were estimated using p-distance in MEGA7.0.14 (Kumar *et al*., 2016). The GTR + I + G model was determined to be the best-fit evolutionary substitution model by AICc from jModeltest 2.1.7 (Darriba *et al*., 2012) and subsequent phylogenetic analyses using the maximum likelihood (ML) and neighbor-joining (NJ) were conducted. ML and NJ method were employed using the PhyML Web-Servers 3.0 (Guindon *et al*., 2010) and MEGA7.0.14, respectively, with 1000 bootstrap replicates.

Taxonomic Accounts

Family Libellulidae Selys, 1840
Genus *Nannophya* Rambur, 1842

***Nannophya koreana* Bae, sp. nov.**
한국꼬마잠자리 (신칭) (Fig. 1)
Nannophya pygmaea: Cho, 1958: 45 (male adult); Cho, 1969: 885 (adult, nymph); Bae *et al*., 1999: 288 (male and female adults, nymph); Lee, 2001: 123 (male & female adult figures); Jung, 2011: 285 (nymph), not Rambur (misidentification).

Type material examined. Holotype: male adult (No. NIBR IN 0000618157, pinned), SOUTH KOREA, Jeollanam-do, Haenam-gun, Songji-myeon, Mabong-ri, 28 V 2012, J.A. Jeon, deposited at NIBR. **Paratypes:** 2 male adults (No. NIBR IN 0000618159, 0000618160, pinned), same data and deposition as holotype; 1 male adult (No. NIBR IN 0000482500, pinned), SOUTH KOREA, Incheon, Jung-gu, Muui-dong, Muui Is., 14 VIII 2006, K.G. Kim, NIBR; 1 female adult (No. NIBR IN 0000812695, pinned), SOUTH KOREA, Jeollanam-do, Hampyeong-gun, Haebo-myeon, Geumgye-ri, 20 VI 2016, H.C. Jeong, NIBR; 2 male adults (No. KUEM 0000003728, 0000003729, pinned) & 5 female adults (No. 0000003704, 0000003705, 0000003730 (**Allotype**), 0000003731, 0000003732, pinned), SOUTH KOREA, Gyeongsangbuk-do (mislabeled as Gyeongnam), Mungyeong-si, Nongam-myeon, Yulsu-ri, 7 VII 2006, D.G. Kim, J.M. Hwang & H.H. Song, KU; 12 male adults & 3 female adults (in 80% Ethanol), SOUTH KOREA, Gyeongsangbuk-do, Mungyeong-si, Nongam-myeon, Yulsu-ri, at abandoned paddy, 17 VI 2006, D.G. Kim, KU; 7 female adults (in 80% Ethanol), SOUTH KOREA, Gyeongsangbuk-do, Mungyeong-si, Nongam-myeon, Yulsu-ri, at abandoned peddy, 19 VI 2006, D.G. Kim, KU; 5 male adults & 4 female adults (pinned), SOUTH KOREA, Chungcheongnam-do, Buyeo-gun, Jangam-myeon, 14 VI 2007, D.G. Kim, KU.
Other material examined. *Nannophya koreana*: SOUTH KOREA: 9 nymphs (small to large-sized, in 80% Ethanol), Incheon, Jung-gu, Muui-dong, Muui Is., at hill wetland, 7 V 2006, Y.J. Bae, T.J. Yoon, J.H. Yum &

D.G. Kim, KU; 31 male adults (in 80% Ethanol), Chungcheongnam-do, Buyeo-gun, Jangam-myeon, behind Jangam Elementary School, 29 VI 2010, D.G. Kim, KU; 169 nymphs (mid- to large-sized, in 80% Ethanol), Gyeongsangbuk-do, Mungyeong-si, Nongam-myeon, Yulsu-ri, at abandoned paddy, 17 VI 2006, 3 VIII 2006, 20 X 2006, 16 III 2007, 17 IV 2007, 4 VI 2007, 19 VI 2007 & 29 VII 2007, D.G. Kim, KU; 1 nymph (large-sized, in 80% Ethanol), Jeollanam-do, Gokseong-gun, Wolbong-ri, at abandoned paddy, 28 X 1999, Y.J. Bae, KU. ***Nannophya pygmaea*:** SINGAPORE: 2 male adults (in 80% Ethanol), Sime Road, Sime Track, Central Catchment Nature Reserve, MacRitchie Reservoir, 14 VII 2009, Y.J. Bae & V.V. Nguyen, KU. CAMBODIA: 1 male adult (in 80% Ethanol) & 34 nymphs (mid- to large-sized, in 80% Ethanol), Kampong Tamon Province, Sam Tuk district, Okampop Amgyl, E105°28′34.7″ N12°39′01.7″, 15 XII 2009, KU; 1 male adult & 6 nymphs (in 80% Ethanol), Siem Reap Province, Siem Reap distract, Phum Angluag Thom village, Thma chruonh, alt. 332 m, E104°10′12.6″ N13°32′28.4″, 24 IV 2010, D.G. Kim, M.J. Baek, J.S. Kim, S. Polin & H. Narith, KU.

Male adult (holotype). *Dimensions*. Body length 18.2 mm; forewing length 14.8 mm; forewing width 4.0 mm; hindwing length 14.0 mm; hindwing width 5.5 mm; abdomen length 11.7 mm; superior appendage 0.94 mm. *Coloration*. Body generally brownish red with light orange pigmentation basally in fore and hindwings (Fig. 1A, 1B). *Head*. Vertex, occiput, frons, postclypeus, and anteclypeus light orange, densely covered with reddish black hair-like setae; mouthparts reddish black, densely covered with reddish black hair-like setae. Compound eyes connected at medial line; upper part reddish brown; lower part reddish black. Ocelli reddish black with light yellow ring. Antennae reddish black. *Thorax*. Pronotum black, densely covered with reddish black hair-like setae. Synthorax generally brownish red with small black markings on upper part of humeral suture and metepisternal suture, covered with dense reddish black hair-like setae; lateral synthorax brownish red with 1 thick black stripe terminating at 1/2 height above spiracle (Fig. 1B); ventral synthorax black. Wings transparent, tinged with light orange pigmentation in basal area; veins black and venation reduced, with tiny black spines sparsely on longitudinal and cross veins and with rowed tiny black spines along wing margins; costa with 2 rows of tiny black spines. Forewings (Fig. 1A) with light orange pigmentation in basal 1/4 area reaching to basal area of triangle, with thickened nodus and black pterostigma; antenodals 5; postnodals 7; arculus slightly contorted and situated between 1st and 2nd antnodals; Rs and MA arising near posterior margin of arculus and merging distally; R_{4+5} situated along line of Rs; 1st radial cell with 1 crossvein; triangle somewhat quadrilateral with arched anterior margin; triangle, subtriangle, and supratriangle without divided cells. Hindwings (Fig. 1A) with light orange pigmentation in basal 1/3 area reaching to distal area of triangle, with thickened nodus and black pterostigma; antenodals 4; postnodals 7; supratriangle without crossvein; anal loop not developed. All legs black, with hair-like setae dorsally and ventrally, with row of long spines on anteroventral and posteroventral margins of apical femora, tibiae, and tarsi; claws paired; each claw bifurcate, with much smaller ventral branch. *Abdomen*. Abdominal segments I and anterior 1/3 of segment II reddish black, with dense reddish black hair-like setae; posterior 2/3 of segment II−segment X brownish red, dorsally with relatively sparse hair-like setae, ventrally with dense reddish black hair-like setae; each posterodorsal, lateral, and paired ventral margins with rowed black teeth. Accessory appendage black. Anal appendages light orange (Fig. 1B), covered with reddish black hair-like setae; superior appendages (Fig. 1E, 1F) almost straight laterally from dorsal view, slightly curved downward from lateral view, posteromedially with expanded round margin with 4−5 distinct black teeth on round margin, apically pointed with single black tooth; inferior appendage (Fig. 1E, 1F) reaching 0.85 × length of superior appendage, basally broadly lobed and apically bifurcate, terminally with paired black teeth.

Female adult (allotype). *Dimensions*. Body length 17.8 mm; forewing length 16.0 mm; forewing width 4.5 mm; hindwing length 14.5 mm; hindwing width 6.0 mm; abdomen length 10.6 mm. *Coloration*. Body generally reddish brown, with black and light orange markings and stripes, with light orange pigmentation basally in fore and hindwings (Fig. 1C, 1D). *Head*. Vertex, occiput, and frons black, densely covered with black hair-like setae; postclypeus and anteclypeus light yellow, with reddish brown markings centrally, densely covered with black hair-like setae. Compound eyes reddish brown, connected at medial line. Ocelli reddish brown with light yellow ring. Antennae black. Mouthparts reddish black, densely covered with black hair-like setae. *Thorax*. Pronotum dark brown, covered with dense light yellow hair-like setae. Synthorax generally reddish black; anterior synthorax reddish black, with paired large triangular light yellow markings at mid-length, with paired transverse light yellow markings at wing base (Fig. 1C). Lateral synthorax light yellow with 1 thick black stripe completely connected to wing base (Fig. 1D), with small black marking near wing base, with sparse light yellow hair-like setae. Ventral synthorax black, with small light yellow markings diversely on suture, with dense light yellow hair-like setae. Wings transparent, tinged with light orange pigmentation in basal area (Fig. 1C); veins black and venation reduced; venation and coloration similar to male. Legs generally black; trochanter with light yellow markings. *Abdomen*. Abdominal segments (Fig. 1C, 1D) generally reddish brown with

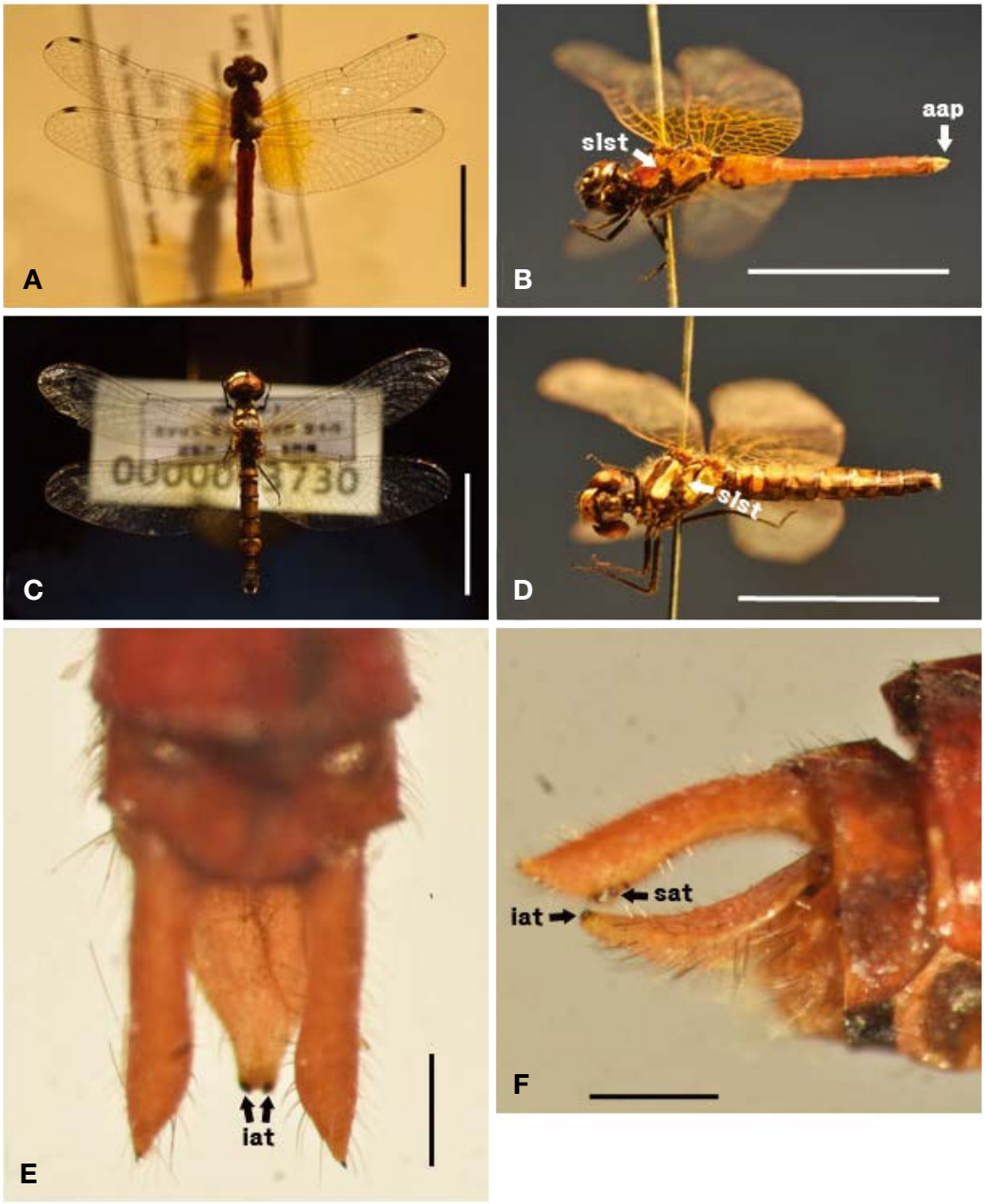

Fig. 1. *Nannophya koreana*: A. male adult, dorsal; B. male adult, lateral; C. female adult, dorsal; D. female adult, lateral; E. male anal appendages, dorsal; F. male anal appendages, lateral. A–D: bar = 10 mm; E, F: bar = 0.3 mm; aap = anal appendage; iat = inferior apical teeth; sat = superior apical teeth; slst = synthorax lateral stripe.

transverse light markings; posterior margins black, with row of black teeth; segments I and anterior 1/3 of segment II black, with dense light yellow hair-like setae; segment III reddish brown, with large anterolaterial light yellow areas, with relatively sparse hair-like setae; segment IV reddish brown with darker lateral areas; segments V–VIII reddish brown, with large anterolateral light yellow areas, with sparse light yellow hair-like setae; segments VIII–X reddish brown, with dense light yellow hair-like setae; segment VIII with small anterolateral light yellow markings. Ventral abdomen black, with dense light yellow hair-like setae. Abdominal appendages light yellow, covered with dense light yellow hair-like setae; cerci terminally pointed; paraproct ca. 0.5 × length of cerci.

Mature nymph. *Dimensions*. Body length 8.6–9.8 mm; head width 2.9–3.1 mm; hind wing sheath length 3.5–3.6 mm; hindfemur 2.3–2.5 mm; hindtibia 2.6–2.8 mm. Body (Fig. 3F) relatively small; body color pale brown to

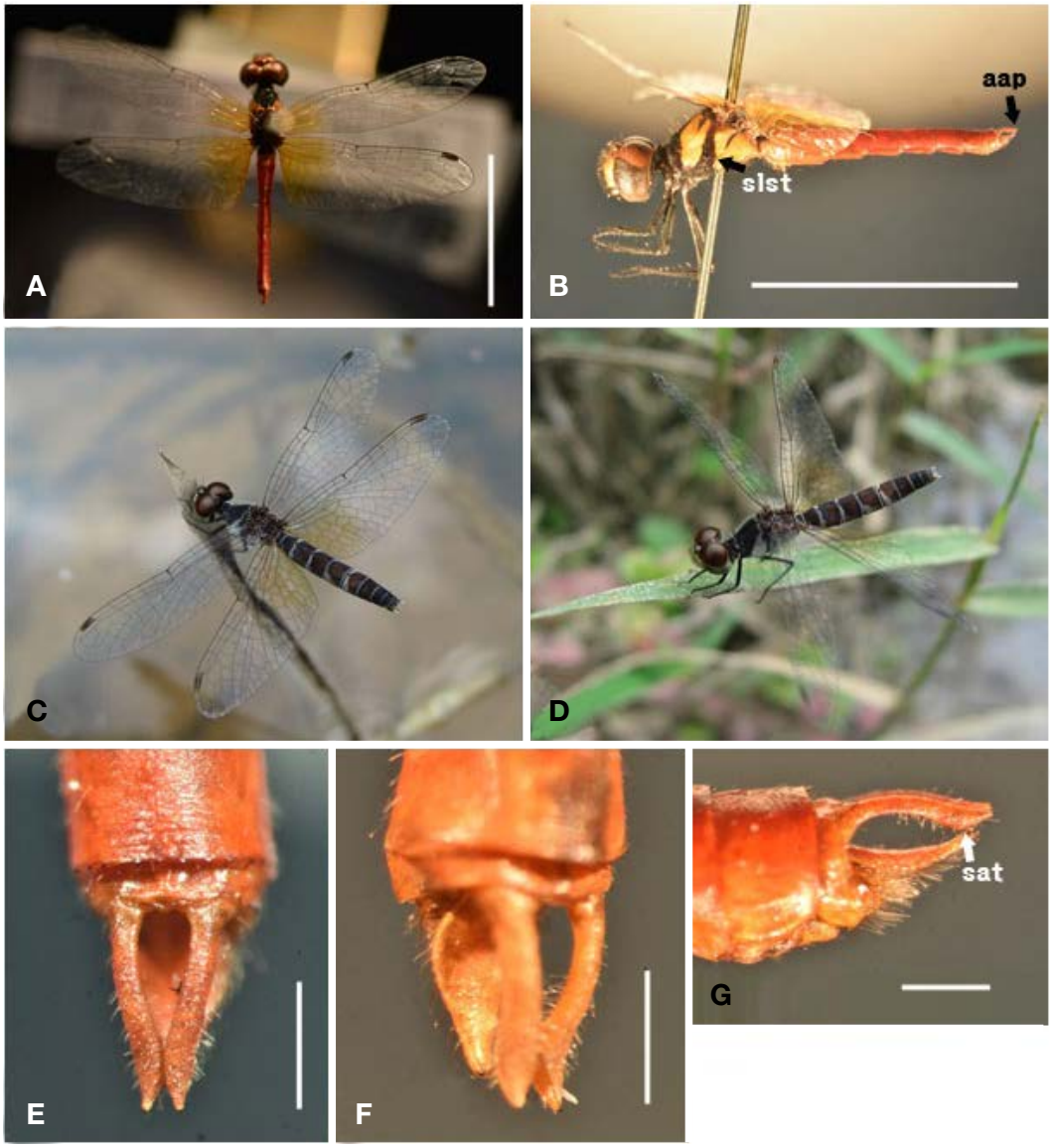

Fig. 2. *Nannophya pygmaea*: A. male adult, dorsal; B. male adult, lateral; C. female adult, dorsal; D. female adult, dorsolateral; E. male anal appendages, dorsal; F. male anal appendages, dorsolateral; G. male anal appendages, lateral. A, B: bar = 10 mm; E–G: bar = 0.5 mm; aap = anal appendage; sat = superior apical teeth; slst = synthorax lateral stripe.

dark brown; body surface covered with tiny dark brown grains and sparse long hair-like setae. *Head*. Head ca. 1.5 × broader than pronotum, inverted trapezoidal, with prominent occipital lobes. Clypeus with dense stout hair-like setae. Compound eyes anterolaterally located. Antennae short, filamentous; flagella 5-segmented. Submentum short, slightly exceeding posterior margin of fore coxae; anterior margin of medial lobe prominent and pointed, with minute serrations and sparsely with ca. 5–6 short stout setae; mental setae 9 (outer 3–5 longest and others gradually shorter). Lateral lobe large, triangular, and fringed with short bristles, with a distinct movable hook and 6 long lateral setae; anterior margin with 10–12 short stout setae; lateral margin with row of short hair-like setae. *Thorax*. Pronotum with row of long hair-like setae on lateral margin. Mesosternal ridge with row of long hair-like setae. Hind wing sheath large, reaching mid-length of abdominal segment VII. Legs relatively short; hindfemora slightly shorter than head width; femora with sparse long hair-like setae on posterior margin; tibiae with long hair-like setae on posterior and anterior margins; tarsi 3-segmented, with row of short stout setae basally; claws double, without denticles. *Abdomen*. Abdomen short, broad, and oval (widest at segment VI); each abdominal dorsum with tiny denticles on posterior margin; segments VIII–IX with very acute lateral spines and long hair-like setae

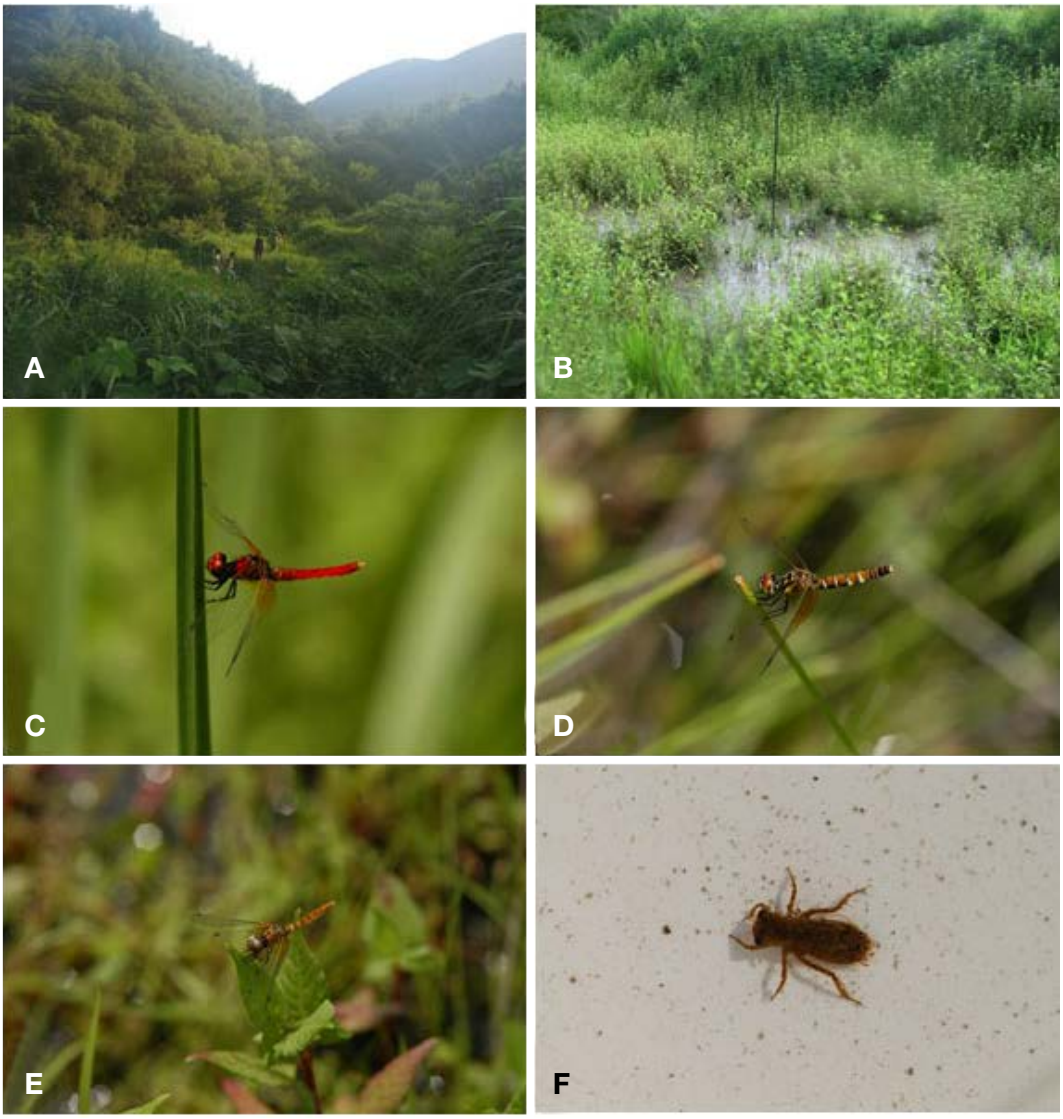

Fig. 3. *Nannophya koreana* habitat in Mungyeong, Gyeongsangbuk-do, Korea (A−E): A. landscape of abandoned rice field; B. habitat with abundant aquatic plants (*Persicaria thunbergii*); C. male adult at habitat; D. female adult at habitat; E. immature male adult at habitat; F. nymph (from Muuido, Incheon, Korea).

on lateral margins; segment IX posteroventrally with relatively dense long hair-like setae. Anal pyramid short and thick; epiproct triangular, apically sharply pointed; paraprocts slightly longer than epiproct; cerci slightly shorter than epiproct.

Diagnosis. Male adults of *N. koreana* (Fig. 1A, 1B, 1E, 1F) can be easily distinguished from those of *N. pygmaea* (Fig. 2A, 2B, 2E−2G) by the presence of a thick, incomplete black stripe on the lateral synthorax that terminates at half-length (Fig. 1B) (*vs*. continuous to wing base; Fig. 2B), light orange (Fig. 1B) (*vs*. red; Fig. 2B) anal appendages, and 4−5 (Fig. 1F) (*vs*. 2−3; Fig. 2G) black teeth on the ventral superior appendages. Meanwhile, female adults of *N. koreana* (Fig. 1C, 1D) can be easily distinguished from those of *N. pygmaea* (Fig. 2C, 2D) by the presence of large, paired, triangular, light-yellow markings on the anterior synthorax (Fig. 1C) (*vs*. lacking such markings; Fig. 2C, 2D) and less prominent basal wing pigmentation (Fig. 1C, 1D) (*vs*. more prominent wing pigmentation; Fig. 2C, 2D). Furthermore, the body length of *N. koreana* specimens is generally larger (1.2−1.4 times) than those of *N. pygmaea* specimens, regardless of life stages (male adult, female adult, or nymph).

Etymology. The species epithet '*koreana*' refers to the country of origin.

Distribution. Korea.

Habitat and ecology. Nymphs of *N. koreana* inhabit shallow wetlands (5−15 cm in depth) with clean water and

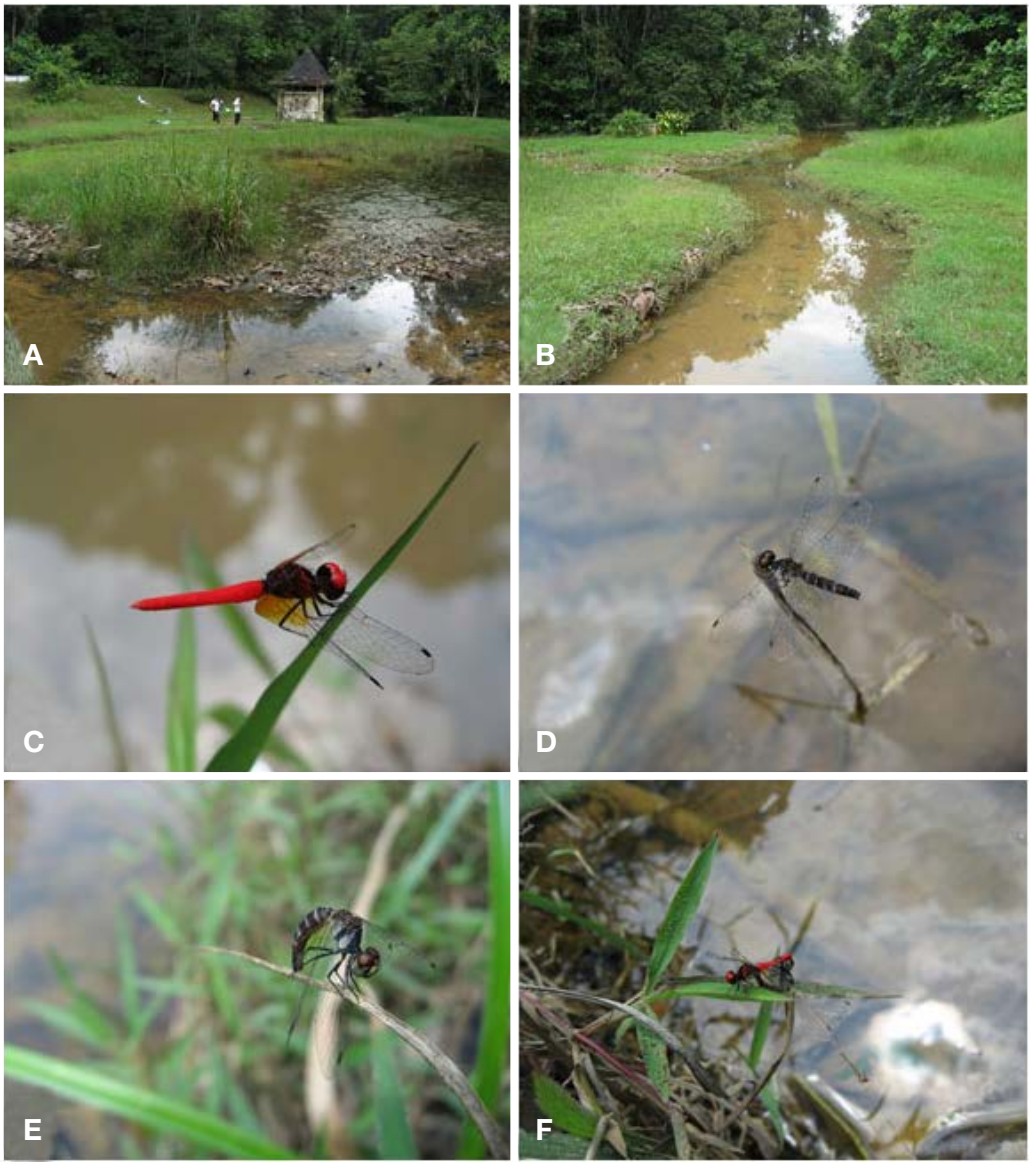

Fig. 4. *Nannophya pygmaea* habitat in Central Catchment Nature Reserve, Singapore: A. lentic habitat; B. lotic habitat; C. male adult at habitat; D & E. female adult at habitat; F. male and female adults in copulation at habitat.

abundant macrophytes, such as *Persicaria thunbergii* and *Juncus effusus* var. *decipiens*, and seem to prefer abandoned rice fields in rural and remote hilly or mountainous areas (Fig. 3A–3E). Adults are found around wetlands (e.g., abandoned rice fields), where they move slowly above the wetland surface, and are found in other areas when the wetlands become dry. The habitat, ecology, life history, and behavior of this species have been well studied in Korea (Bae *et al*., 1999; Lee, 2005; Kim *et al*., 2010).

The habitat of *N. pygmaea* (Fig. 4) is generally similar to that of *N. koreana*. Adults of *N. pygmaea* (Fig. 4C–4F) are often found around small streams in open areas of tropical forest (Fig. 4A, 4B).

Discussion

Nannophya pygmaea Rambur was originally described from a female specimen in the M. Serville collection of the Paris Museum, but its sampling locality was not available (Rambur, 1842). Later, Kirby (1890) reported that the species was distributed in Ambonia (Ambon), China, Borneo, and other places. However, the type specimen was eventually lost (Steinmann, 1997). Therefore, the *Nannophya* specimens from Korea were compared to the original description of *N. pygmaea*, to photos of the *Fylla exigua* Kirby, 1889 (=*Nannophya exigua*, a junior synonym of *N. pygmaea*) lectotype, which was collected from Gilolo (=Halmahera Island, Indonesia) and deposit-

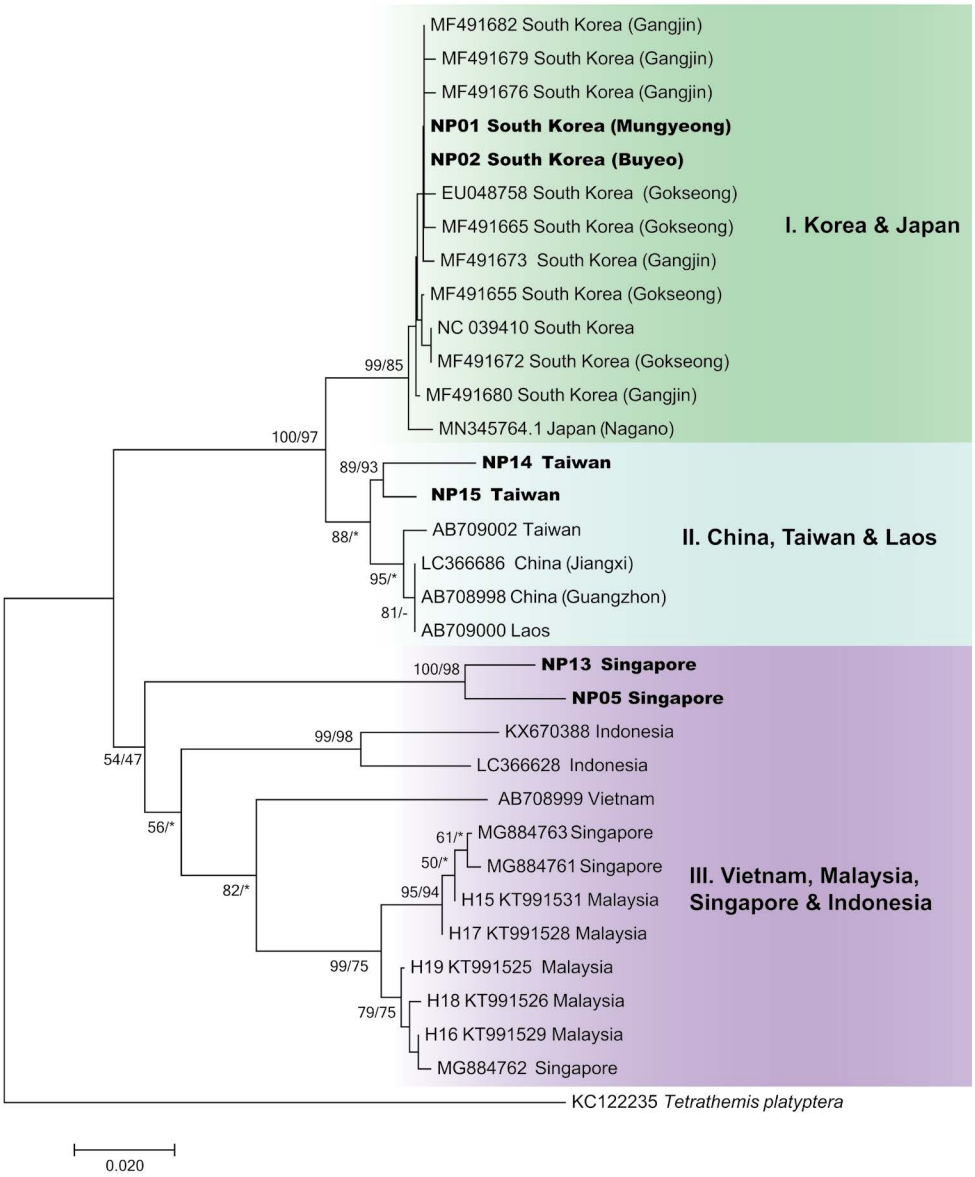

Fig. 5. Phylogenetic tree based on 32 mitochondrial *COI* gene sequences of the *Nannophya pygmaea* species group from nine regions in Northeast and Southeast Asia. Sequences generated by the present study are shown in bold. Branch values indicate neighbor-joining (NJ) and maximum likelihood (ML) bootstrap support values, respectively. Tree topology and branch lengths reflect the results of NJ analysis. Asterisks (*) indicate branches not supported by ML analysis, and dashes (-) indicate support values of less than 50.

ed in the British Museum (Natural History) (Kirby, 1889), and to fresh materials that were collected from Singapore, Cambodia, and Japan. Since *N. pygmaea* is primarily distributed in tropical Southeast Asia, fresh *N. pygmaea* and *N. koreana* materials from Singapore and Korea, respectively, were used for direct comparison.

Other *Nannophya* species that have been described from tropical Southeast Asia, such as *Fylla exigua* Kirby, 1889 (type locality: Borneo) and *Nannodiplax yutsehongi* Navás, 1935 (type locality: Anhui Province, China), were synonymized with *N. pygmaea* (Steinmann, 1997). Bae *et al.* (1999) commented that the *Nannophya* population of Korea could be a distinct species or subspecies of *N. pygmaea* because the northernmost distribution of the *Nannophya* population in Korea is sufficiently geographically separated from tropical Southeast Asia, from which *N. pygmaea* was originally described.

Male adults of *N. koreana* can be morphologically distinguished from its congeners, especially *N. pygmaea*, by characters such as the lateral synthorax stripe, anal appendage color, and superior appendage teeth number. Female adults can be distinguished by the lateral synthorax

Table 1. Estimates of evolutionary divergence between *Nannophya* clades in Northeast and Southeast Asia (see Fig. 5).

Comparisons between clades	Mean distance (%)	SE
Clade I *vs*. Clade II	4.08%	0.86%
Clade I *vs*. Clade III	13.29%	1.53%
Clade II *vs*. Clade III	13.38%	1.45%
Clade I *vs*. outgroup (*Tetrathemis platyptera*)	19.67%	2.61%
Clade II *vs*. outgroup (*Tetrathemis platyptera*)	19.52%	2.58%
Clade III *vs*. outgroup (*Tetrathemis platyptera*)	19.48%	2.23%

Clade I: Korea and Japan
Clade II: Taiwan, China, and Laos
Clade III: Vietnam, Malaysia, Singapore and Indonesia
SE: standard error estimate(s)

Table 2. Estimates of evolutionary divergence between *Nannophya koreana* collected from Korea and *Nannophya* populations collected from eight regions in Northeast and Southeast Asia.

Regions compared to Korea	Mean distance (%)	SE
Japan (clade I)	0.88%	0.41%
China (clade II)	3.83%	0.96%
Laos (clade II)	3.83%	0.96%
Taiwan (clade II)	4.29%	0.88%
Malaysia (clade III)	12.57%	1.91%
Indonesia (clade III)	13.50%	1.79%
Singapore (clade III)	13.92%	2.01%
Vietnam (clade III)	14.01%	1.98%
Outgroup	19.60%	2.56%

Clade I: Korea and Japan
Clade II: Taiwan, China, and Laos
Clade III: Vietnam, Malaysia, Singapore, and Indonesia
SE: standard error estimate(s)

stripes and basal wing pigmentation. In addition, the body size of *N. koreana* (e.g., body length, forewing length, and hindwing length of male and female adults and body length of nymphs) is generally 1.2−1.4 times larger than that of *N. pygmaea*.

Distinct phylogenetic relationships and level of *COI* sequence divergence also substantiate the species-level status of *N. koreana* among *Nannophya* species group populations. The *N. pygmaea* species group is separated into three distinct clades: the Korea and Japan clade (Clade I); the China, Taiwan, and Laos clade (Clade II); and the Vietnam, Malaysia, Singapore, and Indonesia clade (Clade III) (Fig. 5). The strongly supported monophyly of *N. koreana* (Clade I) with high bootstrap values in both NJ (99) and ML (85) phylogenetic inferences indicates an unlikely conspecific relationship between *N. koreana* (Clade I) and *N. pygmaea* (Clade III) (Fig. 5). Furthermore, *N. koreana* clearly lies beyond the typical intraspecific level of genetic distance among other *Nannophya* populations in tropical Southeast Asia (Clade III, mean distance, 13.29%), from which *N. pygmaea* was originally described: Malaysia (12.57%), Indonesia (13.50%), Singapore (13.92%), and Vietnam (14.01%) (Tables 1 and 2). Indeed, a barcoding study of *N. pygmaea* (Low *et al*., 2016) showed similar results, i.e., a large genetic distance (10.64−11.75%) between the populations in Korea and in other countries.

However, the genetic distance of the specimens from Japan lays within the intraspecific level (0.88%), and those of the Clade II populations were only slightly beyond the interspecific levels: China (3.83%), Laos (3.83%), and Taiwan (4.29%) (Table 2). Generally, over 2−3% of *COI* sequence divergence has been suggested to be appropriate for species delimitation in most animal taxa (Hebert *et al*., 2003). Given the level of sequence divergences (3.83−4.29%) between Clade II populations and *N. koreana* (Clade I), the formation of distinct separate clades might indicate the possibility of another regional-specific new species present in this *Nannophya* group. Further detailed taxonomical examination using more extensive sampling in those regions will be required for clarifying this issue.

Since this small dragonfly species, with taxonomic status unclear, has received copious attention from the public owing to conservational and cultural issues in Korea, the present study may resolve previous taxonomic confusion regarding *Nannophya* species in Korea. The results of the present study also indicate that further species-level revision of *Nannophya* and related genera is required.

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