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ARTICLE

FIRST CONFIRMED FOSSIL TURTLE EGGSHELLS (OOGENUS *TESTUDOOLITHUS*) FROM THE LOWER CRETACEOUS OF THAILAND

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ABSTRACT

In contrast to the nearly global occurrence of fossil turtle eggs and eggshells, Southeast Asia had long been void of such fossil record. Consequently, the study of the turtle reproductive paleobiology was difficult in the region, despite that it helps to understand the evolutionary success of turtles through the Mesozoic. Here, we describe the first fossil turtle eggshells from the Lower Cretaceous of Thailand, and discuss their paleobiogeographical and paleoenvironmental implications. The specimens include two eggshell fragments (BHK-D585 and BHK-D586) of different types, which are from the Berriasian to the lower Barremian Sao Khua Formation, the Khorat Group exposed in Chaiyaphum Province, Thailand. The Sao Khua Formation represents a floodplain and yields numerous terrestrial vertebrate fossils. BHK-D585 and BHK-D586 are contained in the reddish-brown conglomerate. These eggshells share a smooth external surface and tightly-interlocking acicular crystal. BHK-D585 and BHK-D586 have the shell thicknesses of 0.62 – 0.64 mm and 0.55 – 0.68 mm, and 1.5:1 and 2:1 height-to-width ratio of an eggshell unit, respectively. A non-branching, sinuous pore is present in BHK-D586. These microstructural characters are consistent with those of the oogenus *Testudoolithus*, and suggest both of the fragments belong to non-marine turtles. Compared to other known turtle ootaxa with rigid eggshell, BHK-D585 and BHK-D586 have unique combinations of the shell thicknesses and height-to-width ratio of an eggshell unit. We conclude that BHK-D585 and BHK-D586 in the Sao Khua Formation are the first reproductive evidence of non-marine turtles in Southeast Asia. They increase the morphological diversity of known turtle ootaxa and imply the presence of a suitable paleoenvironment for preservation of their eggshells in northeastern Thailand during the Early Cretaceous.

Key words : turtle, fossil egg, fossil eggshell, vertebrate paleontology, the Sao Khua Formation, Thailand, Early Cretaceous

今井拓哉・Pratueng JINTASAKUL・東　洋一・野田芳和・Duangsuda CHOKCHALOEMWONG（2016）タイの下部白亜系から初めて見つかったカメ類卵殻化石（卵属*Testudoolithus*）について．福井県立恐竜博物館紀要 15：1–6.

これまで東南アジアからは，化石カメ類の卵や卵殻化石の記録はなかった．本論では，タイの下部白亜系から初めて発見されたカメ類卵殻化石を記載する．標本は2点の卵殻（BHK-D585，BHK-D586）で，タイ北東部に露出する河川成のサオ・クア層（ベリアシアン～下部バレミアン）から産出した．両標本は，凹凸のない表面と隙間なく連なる針状結晶という共通の特徴を持つ．卵殻の厚さはそれぞれ 0.62–0.64 mm，0.55–0.68 mmで，卵殻単位の偏平比はそれぞれ1.5：1と2：1である．これらの微細構造は卵属*Testudoolithus*のものと一致し，非海生カメ類のものと考えられる．既知のカメ類卵タクサと比較すると，BHK-D585 とBHK-D586 はそれぞれ特徴的な厚さと卵殻単位比の組み合わせを持つ．このことから，両標本は東南アジアからの初めての非海生カメ類繁殖の証拠であり，前期白亜紀のタイ北東部には，卵殻化石が保存されうる環境があったことを意味する．

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INTRODUCTION

Fossilized hard-shelled eggs and eggshells of turtles (order Testudines) occur in every continent except Antarctica, and in as old as Early Jurassic sediments (Lawver and Jackson, 2014). This observation indicates that turtles which lay hard-shelled eggs were present at least in the Early Jurassic, and gained world-wide distributions through the Mesozoic. Knowledge of the evolution of reproduction in turtles during the Mesozoic is important because this era is most likely the time where turtles first appeared, dispersed, and evolved substantially toward the modern forms (Li et al., 2008; Schoch and Sues, 2015). Thus, among oviparous amniotes, the processes through which turtles acquired their reproductive strategies, and the contribution of these strategies to their evolutionary success are key to better understand the evolutionary history of turtles. In addition, turtle eggshell is characteristically composed of aragonite, in contrast to calcitic eggshells of other amniotes (Hirsch, 1983). Therefore, the fossil record of turtle eggs and eggshells helps to address such questions as: when turtles acquired hard-shelled eggs, in what ways turtles with hard-shelled eggs radiated, and what selective pressure may have caused the evolution of unique aragonite eggshell only among turtles.

The Mesozoic fossil record of turtle eggs and eggshells is biased, which limits our understanding of the evolution of turtle reproduction through the time. According to Lawver and Jackson (2014), while the fossil record of Mesozoic turtle eggs and eggshells is ubiquitous in Europe, East Asia, North America, other parts of the world often lack such a fossil record. The lack of a record in some regions may be due to a lack of fossil-bearing Mesozoic strata or paleontological expertise. In other areas where fossil turtle skeletons are present, it is more likely that their absence is biological (e.g., reproduction with soft-shelled eggs) or taphonomic (e.g., sediment ideal for preservation of skeletal elements but not eggshells).

Southeast Asia is one of such regions where there is no fossil record of turtle eggs and eggshells. Particularly in Thailand, numerous skeletal remains of turtles had been discovered, while there has been an apparent lack of their eggs and eggshells in Mesozoic strata, without plausible explanation.

Here, we report turtle eggshells from the Berriasian to the lower Barremian Sao Khua Formation of the Khorat Group, cropping out in Chaiyaphum Province, northeastern Thailand. These eggshells add to the known morphological diversity of the Mesozoic non-marine turtle eggshell. In addition, as the first Mesozoic turtle eggshells from Thailand, they fill the paleobiogeographical gap of the Mesozoic turtle eggshells of Southeast Asia, and suggest the possible presence of the better-preserved Mesozoic turtle eggs and nests in Thailand, aiding our understanding of turtle reproduction during the Mesozoic in the region.

GEOLOGY

The Khorat Group is mostly composed of the Mesozoic sediments. It is widely exposed in the northeastern part of Thailand, namely the Khorat Plateau. Some of the formations belonging to the Khorat Group, including the Sao Khua Formation, have yielded a variety of the Mesozoic terrestrial vertebrates (Department of Mineral Resources, 2014).

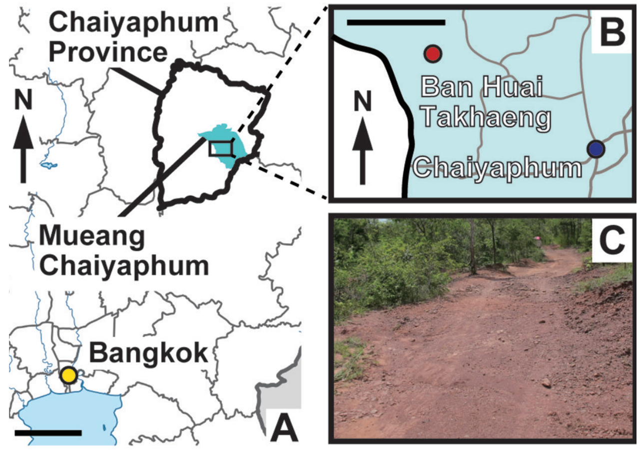

FIGURE 1. Maps and a photo of the locality. **A,** location map of Chaiyaphum Province, scale bar = 100 km; **B,** location map of Ban Huai Takhaeng (square in A), scale bar = 10 km; **C,** overview of the locality.

The Sao Khua Formation represents the middle part of the Khorat Group, conformably underlying the Phra Wihan Formation and overlaying the Phu Phan Formation (Department of Mineral Resources, 2014). It consists of reddish-brown, calcrete-bearing sedimentary rocks, ranging from claystone to conglomerates (Racey, 2009), likely representing a meandering fluvial system under semi-arid paleoclimatic conditions (Meesook, 2000). A palynological analysis suggests that the sediments were deposited in the Berriassian to the early Barremian (Racey and Goodall, 2009). The formation is known to produce abundant vertebrate remains (Department of Mineral Resources, 2014). Notably, Buffetaut et al. (2005) described a small fossil amniote egg containing embryonic skeletons, reporting that the embryo belongs to Aves. However, a more detailed observation utilizing the micro-computed-tomography technique by Fernandez et al. (2015) identifies the embryo as Anguimorpha.

MATERIALS AND METHODS

Studied materials include two eggshell fragments, BHK-D585 and BHK-D586, which are currently housed in the Northeastern Research Institute of Petrified Wood and Mineral Resources, Nakhon Ratchasima Rajabhat University, Thailand. The locality is situated in a community forest of Ban Huai Takhaeng, Khok Sung Subdistrict, Mueang Chaiyaphum District, Chaiyaphum Province, Thailand (Fig. 1), where the Sao Khua Formation predominantly crops out. The matrices of BHK-D585 and BHK-D586 are composed of reddish-brown conglomerates consistent with the general lithology of the Sao Khua Formation.

Each eggshell fragment was broken into half, and each half fragment was prepared for a standard 30 μm-thick petrographic

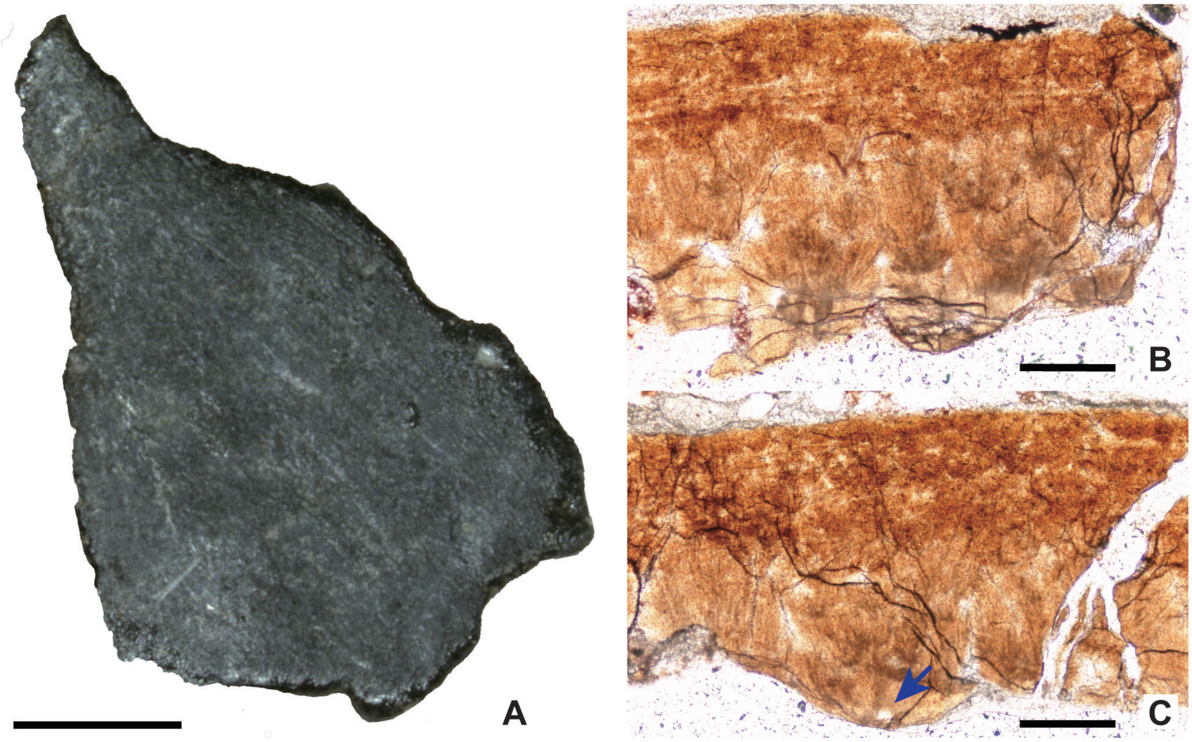

FIGURE 2. External surface and a radial thin-section of BHK-D585. **A,** smooth external surface, scale bar = 1 mm; **B–C,** radial-thin section under a petrographic microscope in unpolarized light. Note radiating acicular crystals typical of turtle eggshells, and a possible nucleation center (blue arrow in C), scale bars = 0.2 mm.

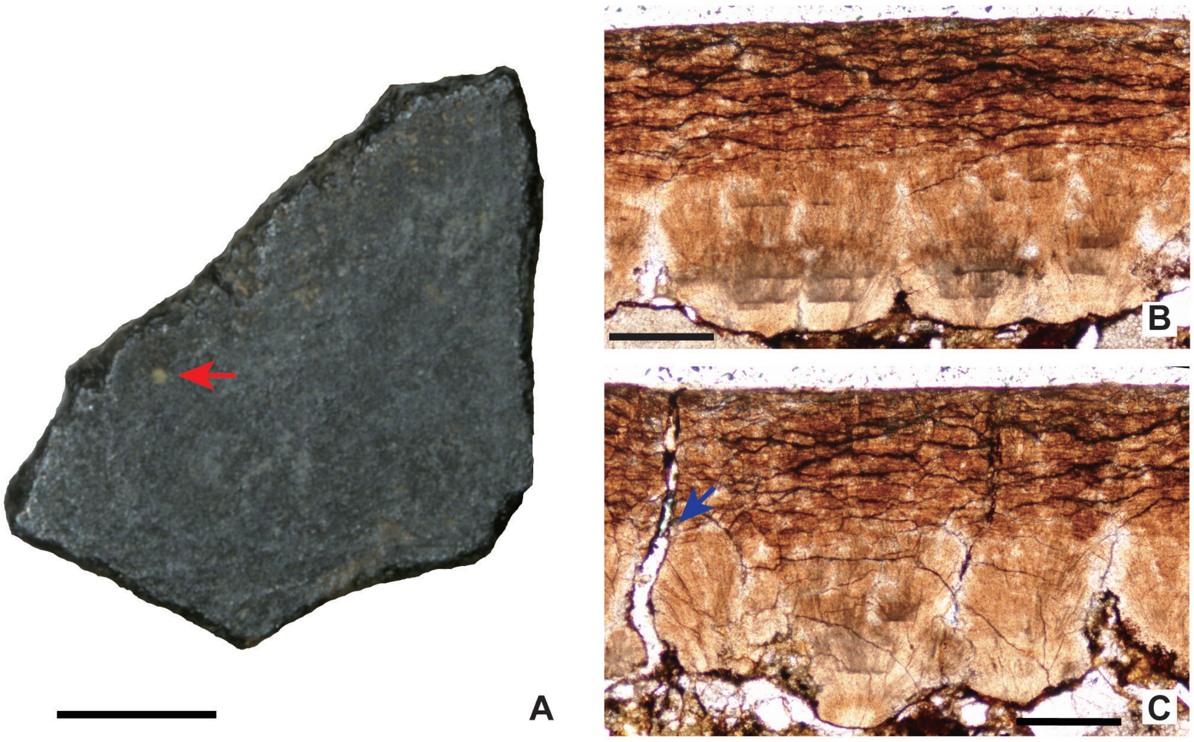

FIGURE 3. External surface and a radial thin-section of BHK-D586. **A,** smooth external surface, note a round pore opening (red arrow), scale bar = 1 mm; **B–C,** radial-thin section under a petrographic microscope in unpolarized light. Note radiating acicular crystals typical of turtle eggshells, and a pore canal (blue arrow in C), scale bars = 0.2 mm.

thin-section and examined under a Nikon E600 POL polarized light microscope. Although microstructural observation on gold- or platinum-coated specimens by a scanning electron microscope (SEM) is desirable in the studies of fossil eggshells, each fragment in this study was too small and we chose not to perform the analysis by SEM to preserve unaltered materials. The macro- and micro-structural characters are compared with other Mesozoic eggshells.

SYSTEMATIC PALEONTOLOGY

Oofamily TESTUDOOLITHIDAE Hirsch, 1996

Type oogenus.—*Testudoolithus* Hirsch, 1996.

Diagnosis.—Eggshell of the oofamily Testudoolithidae is diagnosed by the following unique combination of characters: testudoid basic type, the spherurigidis morphotype, a calcareous layer with interlocking shell units, simple and widely spaced pores between shell units, and spheroidal to ellipsoidal egg shape.

TESTUDOOLITHUS Hirsch, 1996
(Figs. 2 and 3)

Type oospecies.—*Testudoolithus rigidus* Hirsch, 1996.

Diagnosis.—Eggshell of the oogenus *Testudoolithus* is diagnosed by the following unique combination of characters: spherical egg shape, shell unit height greater than width, and shell thickness of 0.2–1.0 mm.

Oospecies *TESTUDOOLITHUS* oospp.

Referred specimens.—BHK-D585 and BHK-D586, eggshell fragments of different types.

Distribution.—Ban Huai Takhaeng, Khok Sung Subdistrict, Mueang Chaiyaphum District, Chaiyaphum Province, Thailand; the Sao Khua Formation (Berriasian-early Barremian) (Figs. 2 and 3).

Descriptions.—BHK-D585 has smooth external surface and has a thickness of 0.62–0.64 mm. In the thin-section, it exhibits a shell microstructure of a single layer of tightly interlocking, radiating acicular crystals. In one of the mammillary cones appears a colorless circular structure, likely a nucleation center. Individual shell units have approximately 1.5:1 height-to-width ratio (Fig. 2C).

BHK-D586 has smooth external surface and its thickness ranges 0.55–0.68 mm. A circular pore opening, which is approximately 0.05 mm in diameter, is present on the external surface. In the thin-section, a non-branching, sinuous pore canal runs from the inner surface to the external surface of the shell, narrowing toward the external side (Fig.3C). The lower half of the shell is characterized by tightly-interlocking, radiating acicular crystals, whereas the upper half exhibits a blocky texture with an extinction pattern different from the lower half under polarized light. A possible nucleation center is observable in one of the mammillary cones. A shell unit has approximately 2:1 height-to-width ratio (Fig. 3C).

DISCUSSION

The microstructural characters of the specimens are consistent with those seen in rigid eggshells of extant turtles, suggesting their affinity to extinct species of non-marine turtles. Turtle taxa from the Sao Khua Formation include the adocid *Isanemys srisuki* Tong, Buffetaut and Suteethorn, 2006, and the carettochelyid *Kizylkumemys* sp., whose remains frequently occur in the formation (Tong et al., 2009). It is unknown whether BHK-D585 and BHK-D586 were laid by any of these species, because they were not associated with skeletal remains of turtles. Both adocids (Zelenitsky et al., 2008) and carettochelyids (Ewert, 1979) are known to have rigid eggshell with shell units taller than wide.

BHK-D585 differs from other previously known valid ootaxa of Testudoolithidae in terms of the thickness (Table 1). The height-to-width ratio of a shell unit of BHK-D585 resembles that of *Haininchelys curiosa* Schleich, Kästle and Groessens-van Dyck, 1988, although *H. curiosa* is much thinner.

BHK-D586 most closely resembles *Emydoolithus laiyangensis* Wang et al., 2013 in terms of the height-to-width ratio of a shell unit and the shell thickness (Table 1). However, the shell thickness slightly exceeds that of *E. laiyangensis.* While *Testudoolithus rigidus* and *Haininchelys curiosa* have the ranges of the shell unit height-to-width ratio within which that of BHK-D586 falls, it is much thinner than BHK-D586. The blocky texture observed in the radial thin section of the upper half of the shell is likely a result of recrystallization, based on the extinction pattern under polarized light. Isaji et al. (2006) reports similar pattern of recrystallization in the upper part of the fossil turtle eggshell from the Lower Cretaceous of Japan.

BHK-D585 and BHK-D586 are the first record of the Mesozoic turtle eggshells from Thailand (Lawver and Jackson, 2014), and increases the morphological diversity of the known turtle ootaxa. Their occurrence indicates that, during the Berriasian to the early Barremian, one or more turtles reproduced with hard-shelled eggs in the floodplain represented as the Sao Khua Formation today.

It is paleobiogeographically and paleobiologically expectable that turtle eggshells occur in the Sao Khua Formation, because 1) turtle eggshells are not uncommon in other Mesozoic terrestrial strata of Asia and 2) well-preserved amniote eggs were recovered from the formation previously (Buffetaut et al., 2005), suggesting that the Sao Khua Formation generally allows the preservation of eggshells composed of calcium carbonate. Nevertheless, the discovery of BHK-D585 and BHK-D586 from the region that had long been void of the record of fossil turtle eggshells advocates that more rigorous search for such eggshells are desired. The presence of BHK-D585 and BHK-D586 in the Sao Khua Formation suggests there is possibly better preserved turtle eggs and nests in the locality, as well as presently unreported eggs and eggshells of other amniotes including dinosaurs which were as thriving as turtles during the time.

TABLE 1. Comparison of BHK-D585 and BHK-D586 with known rigid fossil eggshells of the testudoid basic type. *Calculated by the authors.

Ootaxon, taxon, or specimen number	Shell thickness (mm)	Shell unit height-to-width ratio	Age	References
Chelonoolithus braemi	0.2	1:1	Early Jurassic	Kohring, 1998
Haininchelys curiosa	0.25–0.30	1.2–2.3:1	Paleocene	Schleich et al., 1988
PIN 4225-2, 3	0.3–0.4	?	Late Cretaceous	Mikhailov et al., 1994
Testudoolithus rigidus	0.22–0.24	2:1	Cretaceous to Pliocene	Hirsch, 1996
Uncatalogued	0.190–0.260	2:1	Late Cretaceous or Paleocene	Bajpai et al., 1997
127/CRP/89	0.80	3.5:1	Late Cretaceous	Mohabey, 1998
Testudoolithus hirshi	0.15	3:1	Late Jurassic	Kohring, 1999
SBEI-274, 290	0.200–0.250	1.33–1.35:1*	Early Cretaceous	Isaji et al., 2006
SBEI-285, 288, 885	0.400–0.430	2.05–2.00:1*	Early Cretaceous	Isaji et al., 2006
MOR710	0.700	?	Late Cretaceous	Jackson and Schmitt, 2008
Testudoolithus jiangi	0.70–1.0	2.5–3.0:1	Early Cretaceous	Jackson et al., 2008
Adocus sp.	0.50–0.65	2.5–3.5:1	Late Cretaceous	Zelenitsky et al., 2008
Testudoolithus oosp.	0.250–0.280	2.5:1	Late Cretaceous	Knell et al., 2011
Testudoolithus oosp.	0.40–1.04	3.5–4.2:1	Late Cretaceous	Tanaka et al., 2011
Emydoolithus laiyangensis	0.400–0.500	2–5:1	Late Cretaceous	Wang et al., 2013
DUGF/80-82	0.370–0.450	1.06–1.14:1	Late Cretaceous	Prasad et al., 2015
PIMUZ lab#2012.IW30	0.44	2:1	Late Cretaceous	Lawver et al., 2015
BHK-D585	0.62–0.64	1.5:1	Early Cretaceous	This study
BHK-D586	0.55–0.68	2:1	Early Cretaceous	This study

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