New evidence from China for the nature of the pterosaur evolutionary transition

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Pterosaurs are extinct flying reptiles, the first vertebrates to achieve powered flight. Our understanding of the evolutionary transition between basal, predominantly long-tailed forms to derived short-tailed pterodactyloids remained poor until the discovery of Wukongopterus and Darwinopterus in western Liaoning, China. In this paper we report on a new genus and species, Douzhanopterus zhengi, that has a reduced tail, 173% the length of the humerus, and a reduced fifth pedal digit, whose first phalange is ca. 20% the length of metatarsal III, both unique characters to Monofenestra. The morphological comparisons and phylogenetic analysis presented in this paper demonstrate that Douzhanopterus is the sister group to the 'Painten pro-pterodactyloid' and the Pterodactyloidea, reducing the evolutionary gap between long- and short-tailed pterosaurs.

Pterosaurs are extinct flying reptiles that achieved powered flight by developing an entirely distinctive anatomy comparable to any animals alive today. These reptiles are traditionally divided into two groups, short-tailed pterodactyloids and long-tailed 'rhamphorhynchoids'1. In previous phylogenetic studies, the long-tailed ones have been shown to be a paraphyletic group2–5. However, our understanding of the evolutionary transition between the two groups was severely limited until the discovery of Wukongopterus and Darwinopterus from western Liaoning, China. In addition to Kunpengopterus, these transitional forms have derived cranial characters and primitive postcranial features6–11, alongside Cuspicephalus scarfi, from Dorset, England, which is also considered to be a basal monofenestratan12. Two further specimens, informally named 'Rhamphodactylus' and the 'Painten pro-pterodactyloid' have also been collected from the southern Franconian region of Germany and are relevant to this question. Both these pterosaurs exhibit mosaic characters, considered as the last step on the road from non-pterodactyloids to pterodactyloids13,14. Recently, Allkaruen koi, found in northern central Chubut of Argentina, was also reported to be the sister group of monofenestratan (Wukongopteridae + Pterodactyloidea) pterosaurs15. Here, we report on a new fossil specimen found in western Liaoning, China, which sheds new light on the origin of the Pterodactyloidea.

The new specimen reported here is housed in the Shandong Tianyu Museum of Nature; it was bought from a local farmer, who said that it had been collected in Toudaoyingzi, Jiangchang. However, based on the matrix surrounding the fossil as well as our own field work in this area, we are sure that this specimen was collected from Tiaojishan Formation in Linglongta. The age of these strata are disputed as some early researchers regarded them as ranging between the Middle and Late Jurassic7,16,17. Recently, however, this unit, especially the beds bearing Darwinopterus, have been considered early Late Jurassic in age18–21. Thus, the geological age of the specimen discussed here is considered to be early Late Jurassic16,17.

Result

Systematic palaeontology. Pterosauria Kaup, 1834, Monofenestra Lü, Unwin, Jin et Ji, 2010, Douzhanopterus zhengi gen. et sp. nov.
**Etymology.** Douzhan, Chinese pinyin, the name of a buddha granted by Wukong, the Monkey King in the Chinese legend, indicating the relationship between this new pterosaur and other non-pterodactyloid monofenestratans, such as *Wukongopterus*, pterus, Greek, referring wings; species name in honor of Professor Xiaoting Zheng, who supported our research on this specimen.

**Holotype.** One complete postcranial skeleton housed at Shangdong Tianyu Museum of Nature, Pingyi, China (STM 19–35A & B) (Figs 1 and 2, Tables 1, 2 and 3).

**Locality and Horizon.** Linglongta, Jianchang, Liaoning, China; Daohugou Bed (or Tiaojishan Formation), Late Jurassic.

**Diagnosis.** Monofenestratan pterosaur diagnosed by autapomorphies including a reduced tail that is 173% the length of the humerus as well as a reduced fifth pedal digit, whose first phalange is ca. 20% length of metatarsal III. It can be further distinguished from other monofenestratan pterosaurs on the basis of the following combination of characters: the length of the mid-cervical 2.5–3.5 times of width; 22 caudal vertebrae with elongated zygapophyses and chevrons; pteroid over half length of ulna; tibia ca. 180% length of femur, and; fifth pedal digit V having two phalanges.

**Description.** This new specimen is preserved in part and counterpart, and nearly all the postcranial elements are complete and articulated. Most of the bones are broken horizontally through the middle, and nearly all shafts of long bones are hollow inside, with thin cortices. Some cervical vertebrae are argillic, and only the outlines can be distinguished. The description below is mainly based on the part of the specimen (STM 19–35A, Figs 1 and 2). Elements only preserved on the counterpart (STM 19–35B) will be mentioned in the following text.

This new material comprises a small-sized pterosaur, with a wingspan of 0.74 m, twice the total length of coracoid, humerus, ulna, metacarpal IV, and four wing phalanges. Based on the fusion of syncarpals, scapulocoracoid, tibia and fibula, as well as the fact that the extensor tendon process is fused with the first wing phalange, this specimen is an osteologically mature individual. The first seven cervical vertebrae are preserved in STM 19–35, while additional elements preserved on the counterpart might include a fused atlas-axis. This section of the series has a distinct centrum condyle and posterior odorsal directed neural spine, while the other part is obscured. Each mid-cervical vertebra has a pair of developed pre- and post-zygapophyses. The vertebrae are elongated, and the ratio of length to width is 2.5–3.5 (Table 1). There are nine dorsal vertebrae preserved, suggesting a total of 13 due to the distance between the last preserved dorsal vertebra and the sacrum. Six sacral vertebrae form a sacrum, and their transverse processes are directed lateroposteriorly, thicker than those of the of dorsal vertebrae. The caudal vertebrae of STM 19–35 are complete, even the distal most caudal, which was buried in the part before preparation. There are 22 caudal vertebrae, with...
a total length of 83.86 mm (Fig. 1b). These approximately elongate from the first to the sixth, and shorten to the last (Table 2). The zygapophyses and chevrons of the anterior caudal vertebrae are extremely elongated, at least three times length of centra (Fig. 1b), while the last elongated zygapophysis or chevron is present at the position of the sixteenth caudal. The last caudal vertebrae only have tiny centra. The length of tail is moderately reduced relative to the size of this animal (Fig. 3a; Supplementary Table 1), intermediate between long-tailed forms and short-tailed pterodactyloids.

The sternum of STM 19–35 is not complete, broken into two pieces. The cristospine is not preserved, but the left part of the sternal plate is nearly complete and no keel can be seen. The length of the sternum is similar to the width of the plate; the anterior margin is inclined posteriorly, and the posterior margin is nearly straight.

The scapula is slender. It is elongated and approximately 40% longer than coracoid (Table 3). No ventral expansion is present on the coracoid. The humerus is straight. It has a short deltopectoral crest, which is trapezoid and placed proximally, and it has a distinct ulnar crest, which directs posteriorly. No pneumatic foramen can be observed because of preservation, and the ulna is slightly thicker in mid-shaft than the radius. The pteroid is elongated, reaching half the length of ulna, and the outlines of the right carpals can be distinguished. The proximal
and distal syncarpals formed and one small sesamoid attaches to the distal lateral carpal. The metacarpals are moderately elongated, around 65% the length of humerus and 53% the length of ulna, comprising a ratio between those basal and derived pterosaurs (Fig. 3b; Supplementary Table 2). Metacarpals I-III are articulated with the carpal as well as the wing metacarpal, and manual digits I-III bear large claws. The second wing phalange is the longest one; the first and third phalanges have similar length; the fourth one is slightly shorter than the others, but still over 80% length of the longest one (Table 3).

The pelvic girdles are fused with the sacrum; the preacetabular process is obscured but the postacetabular process is complete and short. Anterior to the acetabular fossa, two pieces of bones are present which we interpret as prepubes. These are longer than wide, and not fused in the middle. The femur of STM 19–35 is slightly curved and has a distinct head and constricted neck, forming a 150° angle with its shaft. The tibia is straight, and fused with the proximal and distal ends of the fibula, which reach up to 45% the length of the tibia (Table 3). Rectangular lateral and medial tarsals are preserved in each hind limb; metatarsal II is the longest, while metatarsal IV is shorter than metatarsals I-III. Metatarsals III and IV are 31% the length of the tibia, while metatarsal V is the shortest (Table 3). The fifth pedal digit still has two phalanges; the first of these is straight while the second is slightly curved (Fig. 1c).

The first pedal phalange is only 20% to 23% the length of metatarsal III, larger than those of pterodactyloids, but shorter than non-pterodactyloids (Fig. 3c; Supplementary Table 3).

### Systematics of *Douzhanopterus zhengi* sp. et gen. nov.

In order to access the position of *Douzhanopterus zhengi* gen. et sp. nov. within the Pterosauria, we carried out a new phylogenetic analysis (see Methods and Supplementary Methods). This analysis was mainly based on the previous work of Wang et al., consisting of 64 species (two outgroups and 62 ingroups) and 89 characters (including two continuous characters,
modified from Andres et al.5). We also added Darwinopterus, Kunpengopterus sinensis, Cuspicephalus scarfi, the 'Painten pro-pterodactyloid', Allkaruen koi, Changchengopterus pani, Douzhanopterus zhengi, and Kryptodrakon progenitor, used TNT for the analysis, and generated 114 most parsimonious trees (MPTs), 253.383 steps in length (CI = 0.600; RI = 0.852). The strict consensus of these MPTs shows that the Monofenetrata and Pterodactyloidea are monophyletic, with the support of 12 and 18 characters, respectively (see the synapomorphies listed in the Supplementary Information). However, the relationship of non-pterodactyloid monofenestratans in this group are still not well-resolved. Through the comparison of 114 MPTs, two unstable taxa were identified, Cuspicephalus scarfi and Allkaruen koi. Both of them lie in basal positions of Monofenestrata. The reduced strict consensus tree without these two taxa is shown in Fig. 4. In this reduced tree, the 'Painten pro-pterodactyloid' is the sister group to the Pterodactyloidea, while Douzhanopterus occupies a more basal position, the sister group to the clade including the 'Painten pro-pterodactyloid' and the Pterodactyloidea. The 'Wukongopteridae' is not resolved as a monophyletic group in this analysis and Wukongopterus lii is the most basal monofenestran.

Discussion
Relationships among basal monofenestratans. Some basal monofenestratans have been considered transitional forms between non-pterodactyloids and pterodactyloids, including Wukongopterus6, Darwinopterus7–9, Kunpengopterus8, Cuspicephalus10, Allkaruen11, Rhamphodactylus12, and the 'Painten pro-pterodactyloid'13,14. However, just the upper jaw of Cuspicephalus are preserved13, and it is absent in Douzhanopterus. The overlap with Allkaruen and Douzhanopterus is limited to cervical vertebrae8, which means that too limited information is available to distinguish these taxa from one another.
Figure 4. The reduced strict consensus tree of 114 MPTs each 253.393 steps in length. 1. Monofenestra; 2. Pterodactyloidea.
Comparing the new Chinese taxon to 'Rhamphodactylus', the 'Painten pro-pterodactyloid' has been discussed in more detail. Both animals are young juvenile individuals because no fusion of postcranial elements can be found. They are in the same size and have quite similar ratios of postcranial elements, although the measurements of 'Rhamphodactylus' were calculated in this study based on figures. Indeed, these two were not treated as the same taxon because of differences in their geological ages. Compared with the new Chinese material, both these enigmatic taxa share the presence of an elongated metacarpal IV (62–66% the length of the humerus, and 46–53% the length of the ulna), reduced first (20% to 31% the length of metatarsal III) and second phalanges of pedal digit V, as well as a reduced tail (0.8–1.7 the length of the humerus) with elongated zygapophyses and chevrons, demonstrating the transitional position of these three specimens between other non-pterodactyloid monofenestratans and pterodactyloids. However, 'Douzhanopterus' varies compared to the two German specimens in a number of postcranial ratios (Table 4). For example, the new Chinese taxon has a larger pteroid/ulna length ratio than that of the two German specimens (Table 4), while its femur length ratio is much larger than that of the other two non-pterodactyloid monofenestratans (Table 4). 'Douzhanopterus' and the 'Painten pro-pterodactyloid' have two reduced phalanges of the fifth pedal digit relative to other non-pterodactyloids, while our new Chinese taxon boasts a smaller ratio between the first phalange of pedal digit V to metatarsal III (20% and 23%) compared to the 'Painten pro-pterodactyloid' (31%). The tails of the two German pterosaurs are also incomplete, lacking their distal-most ends. Thus, assuming that these distal-most caudal vertebrae exhibit the same average lengths as the anterior parts of the 'Painten pro-pterodactyloid', the whole tail of the 'Painten pro-pterodactyloid' would have a maximum length of 29.6 mm, shorter than its humerus. 'Douzhanopterus' also has 22 caudal vertebrae, more than the 'Painten pro-pterodactyloid' which has an estimated 17, the former also has much larger tail/humerus length ratio, nearly twice than that of the latter, but still less than half that of other non-pterodactyloid monofenestratans (Table 4). The centra, rod-like zygapophyses, and chevrons of the caudal vertebrae in 'Douzhanopterus' are slightly more elongated than those of the German specimens, but not so much as in other non-pterodactyloids. Based on our as yet unpublished study of young basal monofenestratan specimens, tail length relative to body size changes, but the number of caudal vertebrae and the elongation of fifth pedal digits remain stable during ontogeny. Hence, 'Douzhanopterus' has a primitive tail but a slightly derived foot relative to the German specimens; thus, as the result of our phylogenetic analysis, 'Douzhanopterus' lies in a more basal position than the 'Painten pro-pterodactyloid'.

The discovery of 'Douzhanopterus' enhances our understanding of the origin of the Pterodactyloidea. Indeed, 'Wukongopterus', 'Darwingopterus', and 'Kunpengopterus' were first proposed by Lü et al.7,8,9,10. Building on this, the intermediate characters seen in 'Douzhanopterus' can also be divided into two modules, an anterior part encompassing the postcranium, mainly cervical vertebrae and metacarpals, and a posterior part, mainly encompassing caudal vertebra and feet. In the anterior part, elongation of cervical is seen, as was the case in 'Wukongopterus' and 'Darwingopterus', whose cervical length/diameter ratios are no more than 2.5. In
**Methods**

The phylogenetic analysis was conducted with the software Tree Analysis Using New Technology (TNT) version 1.1. The ‘Max trees’ setting was set to 10,000 and zero-length branches were collapsed using the rule ‘min. length = 0’. Two ‘Traditional search’ rounds were used, first starting with Wagner trees and duplicating 1,000 times, while the second round was built on trees from memory. The command ‘best’ was used to filter the trees for the best score, and all others were set as default. As discussed in the text, we added *Darwinopterus* zhengi gen. et sp. nov., the ‘Painten pro-pterodactyloid; Allkaruen koi, Kryptodrakon progenitor, Caspicepsalus scarpfi, Changchengopterus pani, Darwinopterus modularis, D. linglongtaensis, D. robustens, and Kunpengopterus sinensis to the matrix of Wang et al.\(^3\), as well as two continuous characters, the ratio between the metacarpal/humerus length and caudal length/diameter (modified from Andres et al.\(^3\)). Another new character relating to the length of the first phalanx of pedal digit V compared to metatarsal III was added as character 88, while characters that relate to the humerus/metacarpal-IV, femur/metacarpal-IV, and ulna/metacarpal-IV length ratios as used by Wang et al.\(^6\) were deleted, to avoid artificially upweighting the ratio of metacarpal IV to other elements. We also re-coded *Wukongopterus* lii for character 50; this character relates to peg-like teeth. In the holotype and the only specimen of *Wukongopterus* lii, the skull is incomplete, and there are less than 15 peg-like teeth preserved on either side. Thus, the inferred number of teeth for each side in upper and lower jaws are 16 and 12, respectively\(^8\). If this is correct, 16 teeth should be scored as state 2 and 12 teeth is state 1; we consider that it is better to choose the two states, but not just state 2. The complete characters and synapomorphies of nodes present in the strict consensus tree are listed in Supplementary Methods. The file with complete matrix, which can be run directly in TNT, is provided as Supplementary Dataset.

**References**

1. Wellnhofer, P. *The Illustrated Encyclopedia of Pterosaurs*. (Salamannder Books, 1991).
2. Bennett, S. C. Taxonomy and systematics of the late Cretaceous pterosaur *Pteranodon* (*Pterasauria*, *Pterodactyloidea*). *Occas. Pap. Mus. Nat. Hist. Univ. Kansas* **169**, 1–70 (1994).
3. Kellner, A. W. A. In: *Evolution and Palaeobiology of Pterosaurs* (eds Buffetaut, E. & Mazin, J. M.) 105–137 (Geological Society, 2003).
4. Unwin, D. M. In: *Evolution and Palaeobiology of Pterosaurs* (eds Buffetaut, E. & Mazin, J. M.) 139–190 (Geological Society, 2003).
5. Andres, B., Clark, J. & Xu, X. *The earliest pterodactyloid and the origin of the group*. *Curr. Biol.* **24**, 1011–1016 (2014).
6. Wang, X., Kellner, A. W. A., Jiang, S. & Meng, X. An unusual long-tailed pterosaur with elongated neck from western Liaoning of China. *An. Acad. Bras. Cienc.* **81**, 793–812 (2009).
7. Lü, J., Unwin, D. M., Jin, X., Liu, Y. & Ji, Q. Evidence for modular evolution in a long-tailed pterosaur with a pterodactyloid skull. *Proc. Roy. Soc. B Biol. Sci.* **277**, 383–389 (2010).
8. Wang, X. et al. New long-tailed pterosaurs (*Wukongopteridae*) from western Liaoning. *China. An. Acad. Bras. Cienc.* **82**, 1045–1062 (2010).
9. Lü, J., Xu, L., Chang, H. & Zhang, X. A new darwinopterid pterosaur from the Middle Jurassic of western Liaoning, northeastern China and its ecological implications. *Acta Geol. Sin.* **85**, 507–514 (2011).
10. Liu, J. et al. An egg-adult association, gender, and reproduction in pterosaurs. *Science*, **331**, 321–324 (2011).
11. Wang, X. et al. Eggshell and histology provide insight on the life history of a pterosaur with two functional ovaries. *An. Acad. Bras. Cienc.* **87**, 1599–1609 (2015).
12. Martill, D. M. & Etches, S. A new monofenestratan pterosaur from the Kimmeridgian Clay Formation (Kimmeridgian, Upper Jurassic) of Dorset, England. *Act. Pal. Pol.* **58**, 285–294 (2012).
13. Rauhut, O. W. M. Ein “Rhamphodactylus” aus der Mörnsheim-Formation von Mühlheim. *Jahresbericht 2011 und Mitteilungen der Freunde der Bayerischen Staatsammlung für Paläontologie und Historische Geologie München e.V.* **40**, 69–74 (2012).
14. Tischlinger, H. & Frey, E. A new pterosaur with mosaic characters of basal and pterodactyloid pterosauria from the Upper Kimmeridgian of Pinaten (Upper Palatinate, Germany). *Acta Palaeontol. Pol.* **52**, 1–13 (in German with English abstract) (2013).
15. Codorniú, L., Carabajal, A. P., Pol, D., Unwin, D. & Rauhut, O. W. M. A Jurassic pterosaur from Patagonia and the origin of the pterodactyloid neurocranium. *PeerJ* **4**, e2311 (2016).
16. Liu, Y. Q. et al. U-Pb zircon age for the Daohugou Biota at Ningcheng of Inner Mongolia and comments on related issues. *Chin. Sci. Bull.* **51**, 2634–2644 (2006).
17. Zhang, H., Wang, M. & Liu, X. Constraints on the upper boundary age of the Tiaojishan Formation volcanic rocks in West Liaoning–North Hebei by LA-ICP-MS dating. *Chin. Sci. Bull.* **53**, 3574–3584 (2008).
18. Liu, Y. et al. Timing of the earliest known feathered dinosaurs and transitional pterosaurs older than the Jehol Biota. *Paleo-* **3**, 323, 1–12 (2012).
19. Sullivan, C. et al. The vertebrates of the Jurassic Daohugou Biota of northeastern China. *J. Vert. Paleont.* **34**, 243–280 (2014).
20. Wang, X. et al. Timing of Linglongta Pterosaur Fauna from Western Liaoning and *Zhejiangopterus*: a summary of geochronology and stratigraphic sequence of pterosaur fossil-bearing beds in China. *Earth. Sci. Front.* **21**, 137–184 (in Chinese with English abstract) (2014).
21. Chu et al. High-precision U-Pb geochronology of the Jurassic Yanliao Biota from Jianshang (western Liaoning Province, China): Age constraints on the rise of feathered dinosaurs and eutherian mammals. Geochim. Geophys. Geosyst. 17, doi: 10.1002/2016GC006529 (2016).
22. Bennett, S. C. The ontogeny of Pteranodon and other pterosaurs. Paleobiology 19, 92–106 (1993).
23. Kellner, A. W. A. Comments on Triassic pterosaurs with discussion about ontogeny and description of new taxa. An. Acad. Bras. Cienc. 87, 667–689 (2015).
24. Goloboff, P. A., Farris, J. S. & Nixon, K. C. TNT, a free program for phylogenetic analysis. Cladistics 24, 774–786 (2008).
25. Wellnhofer, P. Die Pterodactyloidea (Pterosauria) der Oberjura-Plattenkalke Suddentschlands, Bayer. Akad. Wiss. Math. Nat. Klasse Nf. H. 141, 1–133 (in German) (1970).
26. Wang, X. & Zhou, Z. A new pterosaur (Pterodactyloidea, Tapejaridae) from the Early Cretaceous Jiufotang Formation of western Liaoning, China and its implications for biostratigraphy. Chinese Sci. Bull. 48, 16–23 (2003).

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Author Contributions
X.-L.W. and S.-X.J. designed the research plan. X.-L.W., S.-X.J., J.-Q.Z., and X.C. performed analysis. X.-L.W., S.-X.J., and X.C. wrote the manuscript. All authors discussed and commented on the manuscript.

Additional Information
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