# Ossification Pattern in Forelimbs of the Siamese Crocodile (*Crocodylus siamensis*): Similarity in Ontogeny of Carpus Among Crocodylian Species

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## ABSTRACT

Crocodylians have highly derived elongated carpus, which is related to their use of forelimbs in many types of gaits as well as in burrowing. The objective of present study was to describe the ossification of the forelimb in five stages of Siamese crocodile (*Crocodylus siamensis*). The ossification begins approximately at stage 20 in arm and forearm bones moving sequentially to the metacarpal elements. The first carpal elements with ossification centers are radiale + intermedium and ulnare (stage 22–23), and their ossification mode is typical of long bones. Between stages 22 and 24 distal carpals 3, 4, and 5 fuse together to a single formation. In the stage 25, the ossification proceeds to the pisiform, which starts ossifying late during the embryogenesis. The phalangeal formula of the digits is 2,3,4,5,3. Although there are some interspecific differences, it appears that all crocodylians have similarly uniform skeletal pattern, the process of ossification, number of carpal elements and phalangeal formulas probably due to their similar lifestyles. Anat Rec, 301:1159–1168, 2018.   © 2018 Wiley Periodicals, Inc.

Key words: embryo; hand; limb development; morphology; skeleton

The forelimbs in tetrapods all share common features of structure, organization, and function (Rieppel, 1993a; Hinchliffe, 2002; Kardong, 2012). However, each animal species has differences in body type, locomotion or arrangement of structure. The individual roles and positions of the bones reflect also the habitat where the animal lives, and the types of gait, which it uses. Biomechanics of quadrupedal walking has broadly similar features (Griffin et al., 2004). Hence, it is necessary to study also the ontogeny of the limb, because the tetrapod limb "Bauplan" is homologous (Rieppel, 1993a; Kardong, 2012). However, this "Bauplan" is also dynamic and not rigidly fixed (Hinchliffe, 2002).

The archosaurian forelimb presents probably the most diverse structure in tetrapod body (Meers, 2003). Living archosaurs cover two clades—avians and crocodylians (Nesbitt et al., 2013; Xu et al., 2014; Nesbitt et al., 2017). Both clades have a very modified carpus, homology of which is still hotly debated (Xu and Mackem, 2013). Avian carpus may be considered as a product of flight adaptation (Xu et al., 2014) but question is why extant crocodylians have reduced carpus. It could be linked to an ancestral plasticity in archosaurs, which involves a potential natural selection of typical pattern as well as a diversity of secondary adaptations. These adaptations, as flight in birds, could cover various types of gaits and also burrowing in extant crocodylians, which are unique in the number of gaits among all sauropsids.

Development of the crocodylian carpus has been the object of interest for more than one century (Kükenthal, 1893; Holmgren, 1933; Müller and Alberch, 1990; Rieppel, 1993b; Buscalioni et al., 1997; Lima et al., 2011; Vieira et al., 2016). The unique feature of the crocodylian carpus is its highly derived status (Müller and Alberch, 1990; Rieppel, 1993b; Lima et al., 2011; Vieira et al., 2016). Its reductions are unique among sauropsids and have not been observed even in turtles, sister group of archosaurs (Zardoya and Meyer, 1998; Lyson et al., 2010; Chiari et al., 2012). The carpus of the alligator consists of three proximal and four distal carpal elements (Müller and Alberch, 1990). In carpus of *Caiman yacare* only four elements—radiale, ulnare, pisiform, and distal carpal $4+5$— are ossified (Lima et al., 2011). In *Melanosuchus niger* even the distal carpals $3+4+5$ fuse together and ossify with one ossification center (Vieira et al., 2016). However, in the hand of *Melanosuchus niger* are still four elements ossifying according to sequence described for *Caiman yacare* (Lima et al., 2011; Vieira et al., 2016). Similarly, in the adult hand of *Alligator mississippiensis* there are four ossified elements present and the rest are cartilaginous (Müller and Alberch, 1990). Therefore, it appears that chondrogenesis as well as the ossification processes have similarly uniform pattern among crocodylian species. Of note, the crocodylian forelimb exhibits a higher number of the digits (five) compared to the webfooted hindlimb (four). Only the first three digits have claws in both forelimb and hindlimb, which is typical for all crocodylians (Grigg and Kirshner, 2015).

The objective of the study was to describe the pattern of ossification in crocodylian carpus and discuss the results with recent knowledge. For our examination we used a unique source of eggs from an established breeding pair of Siamese crocodile (*Crocodylus siamensis*) held in captivity (Crocodile Zoo Protivin, Czech Republic, www.krokodylizoo.cz) as a species of true crocodile from family Crocodylidae (Oaks, 2011). Although the ontogeny of the limbs in crocodylians is fairly well described (e.g., in alligator or black caiman), there are still several open questions about the exact process of ossification in other species from family Crocodylidae, specifically: (i) when the ossification of the hand elements begins, (ii) what is the appearance of ossification, and (iii) the specific phalangeal formula.

## MATERIAL AND METHODS

Fertilized surplus eggs of Siamese crocodile (*Crocodylus siamensis*) were obtained from The Crocodile Zoo Protivín between years 2013 and 2014. Eggs were opened at weekly intervals. Series of six embryos at age 21–54 dpo [days post oviposition; stages 16–25 (Ferguson, 1985)] was obtained. Normal incubation period in *Crocodylus siamensis* is around 68–80 days between temperature 28° and 33°C. Eggs were incubated without rotation at $33° \pm 1°C$, which usually results in hatching at 68 days (Lang and Andrews, 1994). According to the Czech law, studies on embryos still contained in the eggs are exempt from approval of the Institutional Animal Care and Use Committee. Whole embryos after removal from the eggs were photographed for the record using a Nikon D7000 DSLR camera. After 48 hr fixation in 4% paraformaldehyde in phosphate buffer saline, isolated forelimbs were skinned if possible and subjected to whole mount Alcian Blue—Alizarin Red staining (Potthoff, 1984). After clearing in 1% KOH and glycerol, photos of the limbs were taken using an Olympus BX51 and Olympus SZX12 microscope fitted with an Olympus DP71 and DP50 CCD camera, respectively. The images were assembled and labeled in Adobe Photoshop (Adobe Systems, Palo Alto, CA).

## RESULTS

### Stage 16 (21 dpo)

At stage 16 (Fig. 1A) chondrogenetic condensations of the long bones (humerus, ulna, and radius) were present as well as the mesenchymal condensations of the radiale + intermedium, ulnare, distal carpal 4 and metacarpals III and IV. The digital arch between digits II and III was clearly visible (Fig. 1B).

### Stage 20 (30 dpo)

The centers of ossification were present in the long bones—humerus, ulna, and radius (Fig. 2A). Polydactyly–bifurcation of the distal phalanx of the thumb (digit I) was present in one specimen. The following elements were observed in the carpus: radiale + intermedium, ulnare, pisiform, and centrale. The distal carpal 4 and 5 were still distinct but very close to each other.

### Stage 22 (46 dpo)

Centers of ossification appeared in the metacarpals I–III. Digits I–III were ungual (possessed claws). The distal carpals 4 and 5 fused together as distal carpal $4+5$ (Fig. 2B).

### Stage 22–23 (54 dpo)

Ossification continued in the humerus, ulna, and radius, as well as in the metacarpals I–III. The centers of ossification newly appeared in metacarpals IV–V and in the digits. The centers of ossification were visible also in the largest carpal elements, radiale + intermedium and ulnare (Fig. 2C). Phalangeal ossification pattern was the following: digit I: 1, 2 (centers of ossification appeared); digit II: 1, 2, 3 (centers of ossification appeared); digit III: 1, 2, 3, 4 (centers of ossification appeared); digit IV: 1, 2 (centers of ossification appeared), 3, 4, 5 (cartilaginous); digit V: 1 (center of ossification appeared), 2, 3 (cartilaginous).
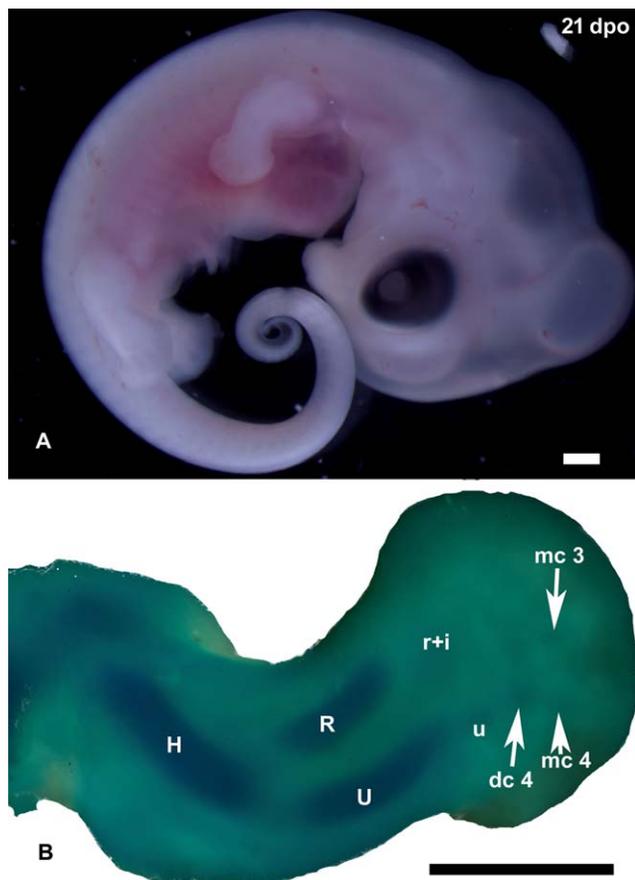
Fig. 1. **A**. Right lateral view of the embryo of *Crocodylus siamensis* in stage 16 (the youngest stage analyzed). Scale bar = 1 mm. **B**. Pattern of chondrogenesis in stage 16 of *Crocodylus siamensis*: dc 4—distal carpal 4, H—humerus, mc 3—metacarpal 3, mc 4—metacarpal 4, R—radius, r + i—radiale + intermedium, U—ulna, u—ulnare. Scale bar = 1 mm.

## Stage 24 (42 dpo)

Ossification progressed in the long bones of the stylopod and zeugopod as well as in the metacarpals (I–IV). Ossification started in the metacarpal V. The centers of ossification in the radiale and ulnare were now clearly distinguishable. Ossification pattern of the radiale and ulnare was similar to the ossification of the long bones—diaphyseal centers of ossification were located in the middle part of the bone (Fig. 2D,E; Fig. 3). The fused distal carpals 3 + 4 + 5 were clearly visible as a single formation. The pattern of phalangeal ossification was the following: digit I: 1 (beginning of ossification), 2 (center of ossification appeared); digit II: 1 (beginning of ossification), 2, 3 (centers of ossification appeared); digit III: 1 (beginning of ossification), 2, 3, 4 (centers of ossification appeared); digit IV: 1, 2 (centers of ossification appeared), 3, 4, 5 (cartilaginous); digit V: 1, (center of ossification appeared), 2, 3 (cartilaginous).

## Stage 25 (54 dpo)

Ossification of the metacarpals, digits, radiale + intermedium, and ulnare further progressed. The

center of ossification appeared in the pisiform (Fig. 2F). At this stage, the centrale and distal carpals were poorly distinguishable. The phalangeal formula for the hand was 2,3,4,5,3. Ossification of the phalanges had the following pattern: digit I: 1, 2, (ungual, ossification in progress); digit II: 1, 2, 3 (ungual, ossification in progress); digit III: 1, 2, 3, 4 (ungual, phalanx 4—beginning of ossification); digit IV: 1, 2 (centers of ossification appeared), 3, 4, 5 (cartilaginous); digit V: 1 (beginning of ossification), 2 (center of ossification appeared), 3 (cartilaginous).

Comparison of ossification pattern in the forelimb among stages is summarized in a schematic diagram (Fig. 4).

## DISCUSSION

Carpal structure could be linked to functional abilities (Fontanarrosa and Abdala, 2014). Among living sauropsids there are particularly two clades—avians and crocodylians, which possess greatly derived carpus. The number of skeletal elements is highly reduced in both clades. The reason why is still questionable in avians (Xu and Mackem, 2013) as well as in crocodylians (this work). However, it seems that in extant crocodylians both chondrogenesis (Müller and Alberch, 1990) as well as ossification (Rieppel, 1993b; Lima et al., 2011; Vieira et al., 2016) have the similar patterns.

### Ontogenesis of the Forelimb in Extant Crocodylians

Ontogenesis of the forelimb in extant crocodylians has typical similar features for all species examined (Müller and Alberch, 1990; Rieppel, 1993b; Lima et al., 2011; Vieira et al., 2016), and this work. In *Alligator mississippiensis, Melanosuchus niger*, and *Crocodylus siamensis* a characteristic digital arch is present in embryos around age 20 dpo [(Müller and Alberch, 1990; Vieira et al., 2016); this work]. From this digital arch distal carpals 2 and 3 arise (Müller and Alberch, 1990). In more advanced embryos—around stage 20 (30 dpo) in *A. mississippiensis* and *C. siamensis*, and stage 12 (26 dpo) in *M. niger*, most of the carpal elements are present in the cartilaginous form (Müller and Alberch, 1990; Vieira et al., 2016).

### Ossification of the Carpus in Crocodylians

Ossification of the carpus has similarities among all extant crocodylians. In *Crocodylus siamensis* ossification starts in stage 20, when most of the carpal elements are present as cartilaginous anlagens. The first centers of ossification appear in the humerus, ulna, radius, and in the metacarpals I–III. The ossification then moves to the phalanges of digits and the centers also appear in the carpal elements—radiale + intermedium and ulnare (stage 22–23), which ossify as typical long bones—from the diaphysis in the middle part of the bones similar to radius and ulna (Reno et al., 2007; Romão et al., 2012; Drake et al., 2014). Such ossification pattern is highly unique among vertebrates. In stage 24 (Fig. 3) distal carpals 3 + 4 + 5 fuse and they are clearly visible as a single structure similar to stage 15 in *Melanosuchus niger* (Vieira et al., 2016). In stage 25, the center of
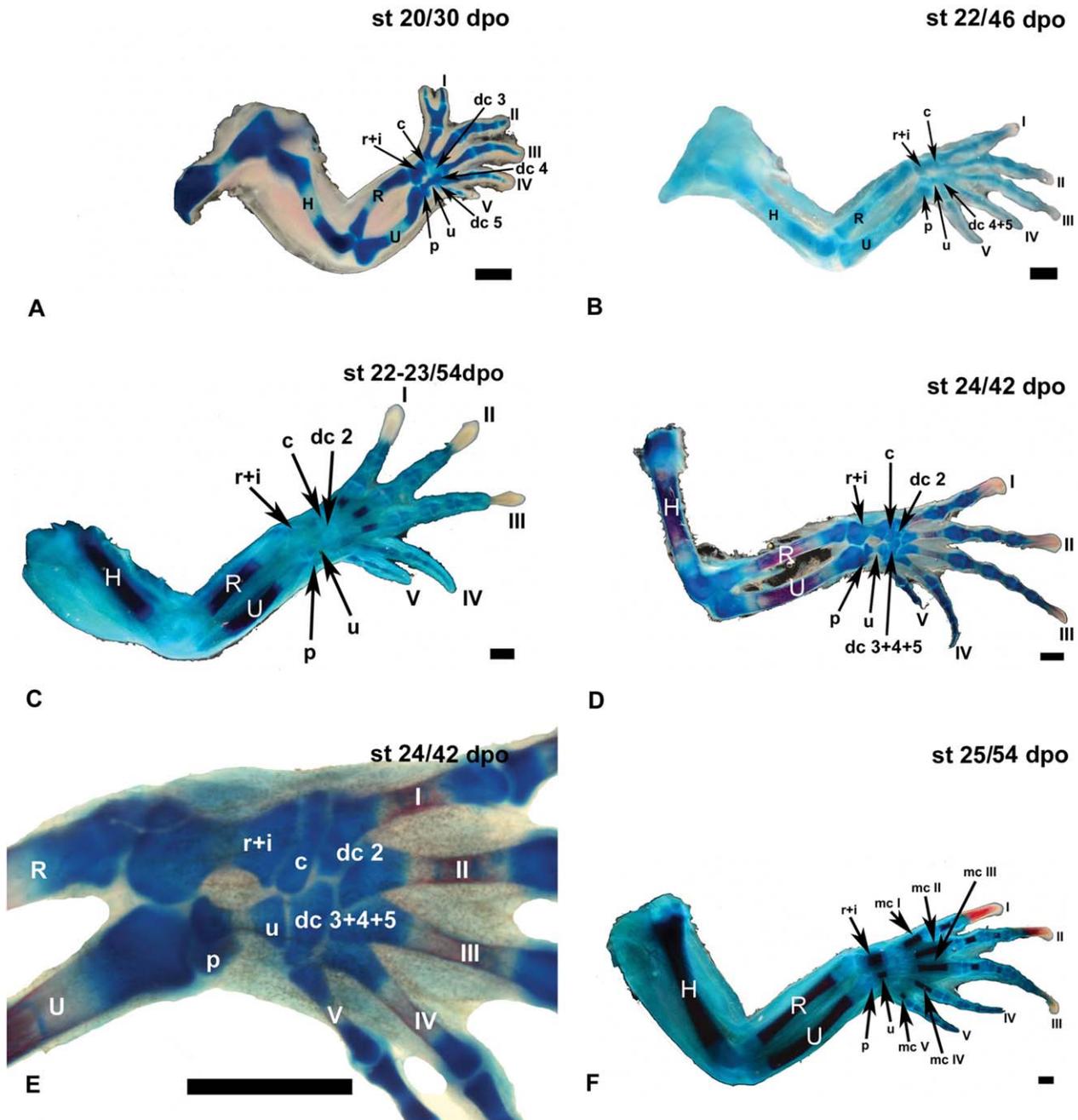
Fig. 2. Pattern of ossification of the forelimbs of *Crocodylus siamensis* embryos, dorsal views: (**A**) stage 20, (**B**) stage 22, (**C**) stage 22–23, (**D**) stage 24, (**E**) detail of the carpus in stage 24, (**F**) stage 25. Abbreviations: H—humerus, I–V—digits, c—centrale, dc 2—distal carpal 2, dc 3 + 4 + 5—distal carpal 3 + 4 + 5, H—humerus, mc I–V—metacarpals I–V, p—pisiform, R—radius, r + i—radiale + intermedium, U—ulna, u—ulnare. Scale bars = 1 mm. Note the polydactyly in stage 20. Digits I–III bear claws (visible from stage 22). Figure 2D note missing phalanges 2 and 3 in digit V. Bone labels are positioned over the ossification centers, wherever possible.

ossification arises already in the last element of the carpus—the pisiform. Ossification of the distal carpal 3 + 4 + 5 is supposed to start after the beginning of ossification in the pisiform, as was described for the *Caiman yacare* (Lima et al., 2011; Vieira et al., 2016). The rest of distal carpals and centrale should be cartilaginous (Müller and Alberch, 1990). Generally, ossification starts earlier in metacarpals than in the carpus itself similarly

to *Alligator mississippiensis* (Rieppel, 1993b), *Caiman yacare* (Lima et al., 2011), and *Melanosuchus niger* (Vieira et al., 2016). After stage 25, ossification occurs in the entire carpus as well as in all the digits and long bones and continues until the adulthood. The dominant digits are I–III. These digits (I–III) start to ossify earlier than the remaining ones and they also bear claws typical for all crocodylians (Müller and Alberch, 1990;

Fig. 3. Foetus of *Crocodylus siamensis* in stage 24 (42 dpo). Note the claws on digits I–III in both forelimbs and hindlimbs.

Rieppel, 1993b; Lima et al., 2011; Grigg and Kirshner, 2015; Vieira et al., 2016). Phalangeal formula of *Crocodylus siamensis* is 2,3,4,5,3. In *Crocodylus palustris* we found the same phalangeal formula as we describe here for *C. siamensis*—2,3,4,5,3 (M. Gregorovicova, unpubl. data, 2015), which could be due to the fact that *C. siamensis* and *C. palustris* are probably sister species (Man et al., 2011; Oaks, 2011). Phalangeal formula was established in the hand early in reptilian evolution, and it has strong tendency for the persistence of its primitiveness form—2,3,4,5,3. This count is adhered to in many later types, even in the varied specialization/reduction in the hand (Romer, 1956). However, in phalangeal formulas the crocodylian species can differ from each other as was shown for *Caiman yacare*—2,3,4,3,2 (Lima et al., 2011) —similar to the formula of *Melanosuchus niger*—2,3,4,4,2 (Vieira et al., 2016) —probably due to their relatively close relationship (Hrbek et al., 2008; Grigg and Kirshner, 2015). In *Alligator mississippiensis* the phalangeal formula is also slightly different—2,3,4,5,4 (Müller and Alberch, 1990).

## Carpal Region of the Sauropsids

***Comparison with extinct crocodylians.*** In archosaurs the limbs have tendency for reduction in ossification of limb bone ends, carpus, or tarsus (Romer, 1956). This is frequently found in known aquatic species. There is also a great trend toward an aquatic life, where the major structures may be strongly affected. The group Metriorhynchidae from the Jurassic can serve as an example of reduction in carpal region. Metriorhynchids were thallatosuchian, the most extremely adaptated marine group of archosaurians (Wilberg, 2015).

They had a specialized paddle-like forelimbs with the loss of the pisiform, centrale and intermedium (Romer, 1956; Young et al., 2010). Radiale and ulnare were secondarily reduced in length and the metacarpal I was expanded (Romer, 1956).

The taxa closer to Crocodyliformes had more compact carpal region and metapodials and it seems that they were digitigrade in both fore and hind limbs (Irmis et al., 2013). This type of carpus was described also in the extinct genus *Protosuchus* (Mook, 1951), which was terrestrial (Grigg and Kirshner, 2015). Genus *Protosuchus* is an early crocodyliform (Pol et al., 2013) and eusuchian crocodiles as the surviving crocodiles are all within this clade (Sereno et al., 2001; Grigg and Kirshner, 2015).

Although it seems that the number of distal carpals is invariant since the group arose during the Upper Triassic (Romer, 1956), there could be found differences among fossil finds (Romer, 1956; Buscalioni et al., 1997) and embryonic development of extant crocodylians (Müller and Alberch, 1990; Lima et al., 2011; Vieira et al., 2016). As an example serves the presence of pisiform, which is absent in some fossils but it arises in extant crocodylian embryos.

***Comparison with modern birds.*** The extant crocodylians are closely related to the avians and together are the only surviving archosaurs (Nesbitt et al., 2013, 2017). The bird hand is investigated with great interest. Although there is probably pentadactyl ground state of hand in modern birds (Kundrát et al., 2002; Larsson and Wagner, 2002) similarly to the crocodylians (Larsson et al., 2010), there are still open questions about avian carpal region (Kundrát, 2009; Botelho et al., 2014). The avians have highly reduced carpal region fused as carpometacarpus in the adulthood (Vazquez, 1992). Avian wrist has reductions in the number of ossifications (Botelho et al., 2014). It seems that typical features in avian embryonic development are radiale + intermedium as a single ossification, reappearance of the pisiform, and loss of ulnare (Botelho et al., 2014), which is in contrast well developed in crocodylians (Müller and Alberch, 1990; Rieppel, 1993b; Lima et al., 2011; Vieira et al., 2016). The avian pisiform has the same embryonic origin as in other reptiles (Botelho et al., 2014) and it is involved in locomotion (Haines, 1946). In birds the pisiform is widely used for flight (Vazquez, 1992). Another feature, which the avian embryos share with crocodylians, is radiale + intermedium single ossification (Rieppel, 1993b; Lima et al., 2011; Botelho et al., 2014; Vieira et al., 2016).

***Comparison with turtles.*** It comes then as no surprise that crocodylians share similar features in the structure of the carpus, which is significantly different from other clades including turtles, a sister group of archosaurs (Zardoya and Meyer, 1998; Lyson et al., 2010; Chiari et al., 2012). Turtles have more plesiomorphic limbs with more carpal elements than crocodylians (Burke and Alberch, 1985; Sheil, 2003, 2005; Sheil and Greenbaum, 2005; Hitschfeld et al., 2008; Sheil and Portik, 2008). The ossification of the hand in turtles starts first in the metacarpal elements (Sheil and Portik, 2008) similarly to crocodylians (Rieppel, 1993b; Lima et al.,
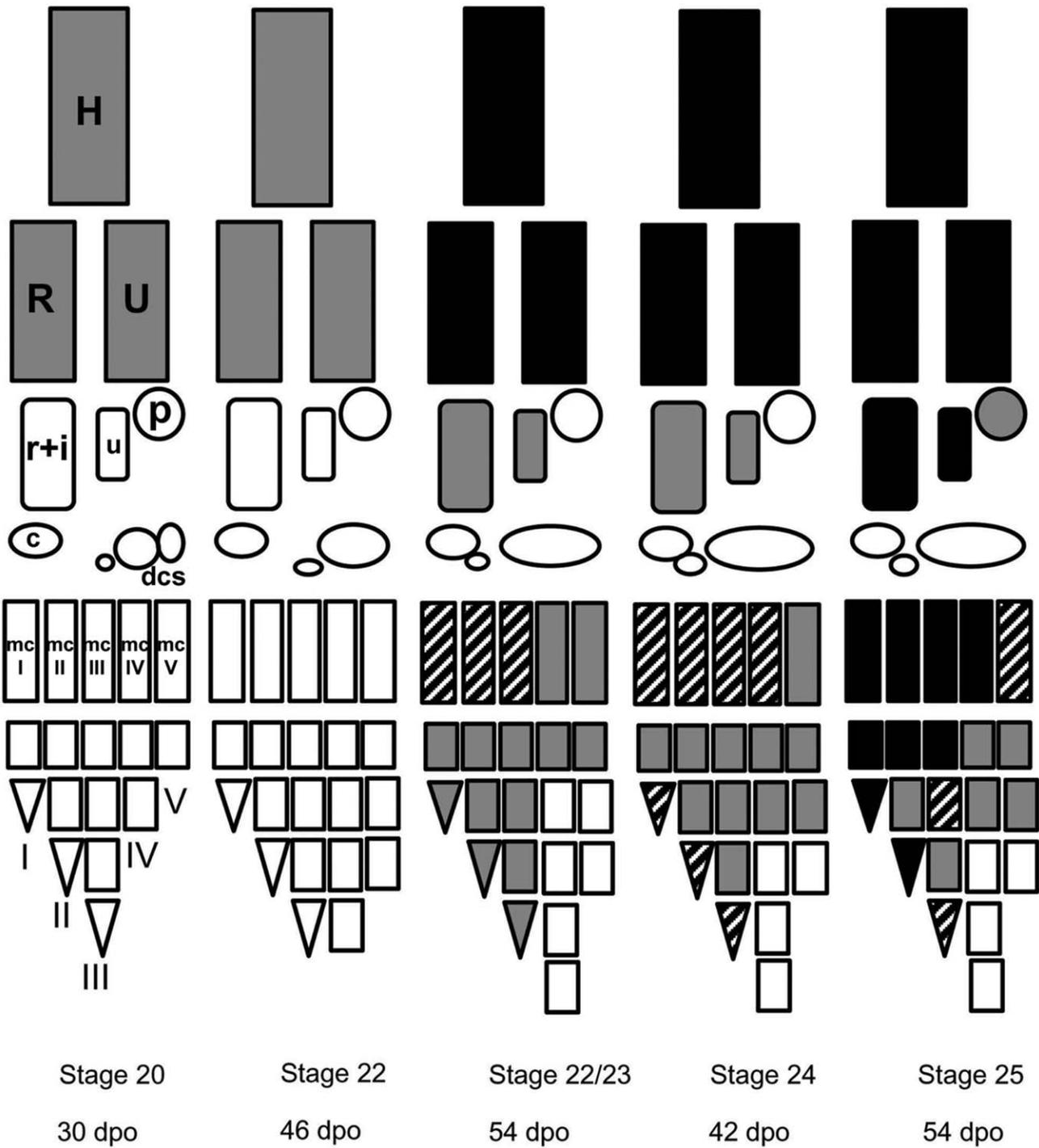
Fig. 4. Schematic diagram comparing the ossification process in forelimb of *Crocodylus siamensis* embryos. Abbreviations: c—centrale, dcs—distal carpals, H—humerus, mc I–V—metacarpals I–V, p—pisiform, R—radius, r + i—radiale + intermedium, U—ulna, u—ulnare. Roman numerals indicate digit numbers. Degree of ossification (cartilage only with no ossification, weak with initiation of ossification, weak retention of red stain, strong retention of stain) is indicated by white < gray < diagonal white hashing on black background < black.

2011; Vieira et al., 2016). However, the dominant digits in turtles are II–IV (Sheil and Portik, 2008) differing slightly from those in crocodylians (I–III). Ossification of the carpal elements (such as ulnare, intermedia etc.) was not observed in prehatchling specimens of

*Trachemys* but in *Apalone*, slight ossification of these elements was observed (Sheil and Portik, 2008). However, the ossification of the carpal elements before hatching is a constant observation for extant crocodylians (Rieppel, 1993b; Lima et al., 2011; Vieira et al., 2016).

The reason for reinforced carpus lies probably in the burrowing lifestyle in e.g., *Testudo horsfieldii* or exposed walking lifestyle typical for e.g., *Testudo hermanni* (Hitschfeld et al., 2008). In turtles (such as in *Apalone*) the reinforced carpus is not necessary in contrast to tortoises (*Testudo*), because the majority of the turtles are swimmers. However, locomotion among turtles differs substantially (Pace et al., 2001). It is possible that the reinforced unique carpus of the crocodylians is related to their posture on the land and in the number of gaits coupled with their significant weight.

*Comparison with lepidosauria.* In comparison to Lepidosauria, the differences in carpal bones are less pronounced among crocodylians (Abdala et al., 1997; Shapiro, 2002; Fabrezi et al., 2007; Leal et al., 2010; Diaz and Trainor, 2015). The other feature distinguishing the lepidosaurian carpus from the crocodylians is the frequent presence of the sesamoids (Fabrezi et al., 2007), which was also observed in non-squamate lepidosaurian *Sphenodon punctatus* (Regnault et al., 2017). It is therefore relatively complicated to compare the crocodylian carpus and the carpus of Lepidosauria. Several features, however, could be compared between crocodylian and squamate carpal region. The ossification starts in metacarpals (Rieppel, 1994; Abdala et al., 1997; Fabrezi et al., 2007) earlier than in the rest of the hand as was observed in crocodylians. However, there is no or very weak ossification in carpus itself before hatching in many squamates (Fabrezi et al., 2007) thus differing from crocodylians ((Lima et al., 2011; Vieira et al., 2016); this work). In squamates as well as in crocodylians there is a strong dominant position of the digit III (Rieppel, 1993b, 1994). Last but not least, the crocodylians share with the squamates one significant feature—development of a distinct and single radiale condensation (Fabrezi et al., 2007). When comparing *Sphenodon* (tuatara), the only surviving taxon of order Sphenodontia (Rest et al., 2003; Cree, 2014), and crocodylian carpal region there can be found the differences not only in the presence of sesamoids in tuatara but also in number and character of centrale. *Sphenodon* possesses two ossified centralia (Carroll, 1988), whereas centrale in extant crocodylians should be a single cartilaginous structure (Müller and Alberch, 1990).

## Functional Morphology of the Limbs in Extant Crocodylians—Gaits and Burrowing

Extant crocodylians are also unique in posture and in the number of types of gait among all reptiles (Zug, 1974; Gatesy, 1991; Hutchinson, 2006; Allen et al., 2010; Cott, 2010). While they are on land, crocodylians have a wide variety of gaits, thanks to their specific semierect posture for better terrestrial mobility (Reilly and Elias, 1998; Hutchinson, 2006; Grigg and Kirshner, 2015). The crocodilian-specific gaits are sprawling walk, high walk, and galloping (Zug, 1974; Gatesy, 1991; Renous et al., 2002; Cott, 2010; Grigg and Kirshner, 2015). Sprawling walk is used as a quick escape downslope to the water, when the toes grip the substrate as the crocodile pulls the body along (Grigg and Kirshner, 2015). High walk is the normal gait on land and it is a symmetrical walking with at least two feet on the ground in semierect posture (Willey et al., 2004). The majority of the weight is on the

hindlimbs (Gatesy, 1991; Reilly and Elias, 1998; Willey et al., 2004), but the crocodile has to support the hindlimbs by strong wrists in order to run. In such rungalloping, which is the fastest gait, forelimbs strike the ground first, whereas hindlimbs are in the air (Zug, 1974; Renous et al., 2002). Therefore, it is convenient to have reinforced carpus to avoid breaking the wrist. Stabilization of the carpus by reducing the number of bones as well as strengthening the bones by modifying them to the type of the long bones could reinforce the carpal region for physically and mechanically demanding activities such as running in case of a heavy reptile such as crocodile. Note that in high walk and galloping the tail is a dead weight on the land (Willey et al., 2004). Moreover, the semierect posture is probably an adoption from the earliest crocodylians—crocodylomorphs (Parrish, 1987) and this implication points to the fact that extant crocodylians are secondarily aquatic. Of note, the most aquatic extant crocodylian is gharial (*Gavialis gangeticus*) (Grigg and Kirshner, 2015). Although gharial has pentadactyl forelimb like the other crocodylians (Grigg and Kirshner, 2015), this species is unable to perform high walk or galloping when it reaches the adulthood (Milàn and Hedegaard, 2010; Grigg and Kirshner, 2015). However, little is known about its embryonical development and skeletogenesis due to its rarity. On the other hand, the reason why gharial cannot perform highwalking or galloping in adulthood could be linked not primarily to the hand skeleton structure itself but rather to the loss of bracing system. There is a difference in the configuration of paravertebral shield and the length of the lateral epaxial muscles in the cranial half of the tail between gharial and the other living crocodylians. This "configuration" is less diverse in gharials. Adult gharials also cannot flex the head ventrally against the neck in degree observed in other taxa (Salisbury and Frey, 2000, 2004).

Burrowing is another physically demanding activity, where the carpus may play a role. Burrowing is listed for 16 species of extant crocodylians including gharials, which also can make hole nests in the dirt or sand (Trutnau and Sommerlad, 2006; Grigg and Kirshner, 2015). Many species are skilled burrowers such as mugger crocodile (*Crocodylus palustris*), West African crocodile (*Crocodylus suchus*), or alligators (*Alligator mississippiensis*, *Alligator sinensis*), all well known for their ability to hide themselves in the self-made burrows in order to endure the unfavorable seasons (Trutnau and Sommerlad, 2006; Grigg and Kirshner, 2015).

## Anomalies

Polydactyly of the terminal phalanx of the thumb (digit I) and also preaxial polydactyly of the hindlimbs (8 digits per limb, not shown) was observed in one embryo out of thirteen specimens at stage 20 (30 dpo). Malformations such as polydactyly, absence of digits or syndactyly are known to occur sporadically in crocodylians (Giles, 1948; Ferguson, 1985), especially if breeding females are too young or too old (Ferguson, 1985). Although the occurrence of polydactyly was described in sauropsids (Martínez-Silvestre et al., 1997; Bauer et al., 2009; Kaliontzopoulou et al., 2013), it is not as widely reported as in case of mammals (Chase, 1951; Dunaway, 1969; Pugsley, 1985; Galis et al., 2001; Chapman, 2006;

Gugolek et al., 2011), especially in marine mammals (Sedmera et al., 1997; Ortega-Ortiz et al., 2000), or amphibians (Bishop, 1947; Cooper, 1958; Lada, 1999; Vorobyeva, 1999; Machado et al., 2010).

## CONCLUSION

Development of the hand in the Siamese crocodile (*Crocodylus siamensis*) is alike to hand development in extant crocodylians—alligator (Müller and Alberch, 1990; Rieppel, 1993b) as well as in caimans (Lima et al., 2011; Vieira et al., 2016), however the differences could be found between the fossil finds of Crocodylomorpha and extant crocodylians (Romer, 1956). In comparison with avians, the carpal region of both clades (avians and crocodylians) is dramatically reduced with similar characterstics (Rieppel, 1993b; Kundrát, 2009; Botelho et al., 2014; Vieira et al., 2016) probably due to their specific functional purposes. The carpus is highly reduced also when compared to turtles (Burke and Alberch, 1985; Sheil and Portik, 2008; Fabrezi et al., 2009) and Lepidosauria (Carroll, 1988; Shapiro, 2002; Fabrezi et al., 2007; Leal et al., 2010; Diaz and Trainor, 2015; Regnault et al., 2017).

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