First data on reproduction and hatchling morphology of *Mesoclemmys heliostemma* (McCord, Joseph-Ouni & Lamar, 2001)

The Amazon Toad-headed Turtle *Mesoclemmys heliostemma* (McCord, Joseph-Ouni & Lamar, 2001), represents one of the least known Amazonian turtles. Despite the species' vast distribution area in the Amazonian lowland of Brazil, Ecuador, Perú and Venezuela (and probably also Colombia) (McCord et al. 2001; Cisneros-Heredia 2006; Molina et al. 2012; Forero-Medina et al. 2014; Morcatty & Cobra 2015), very little was published about its biology.

During a herpetological survey conducted in the area of Iquitos, in the framework of a scientific cooperation between the National Museum, Prague, the Czech Republic, and the Universidad Nacional de la Amazonia Peruana (UNAP), Iquitos, Perú, two juvenile specimens of *M. heliostemma* lacking the characteristic juvenile head pattern were collected in temporal water bodies in a seasonally flooded forest on the right bank of Rio Nanay near the village of Anguilla (03°54'45" S, 73°39'39" W, Region Loreto, Perú) in March, 2002. With the aim to confirm the determination of the collected specimens and obtain additional data on morphology and biology of this then practically unknown turtle species, both individuals were taken to UNAP and later to the National Museum, Prague. Here, the turtles were kept under laboratory conditions and underwent a parasitological examination, which resulted in description of a new species of intestinal parasite of the genus *Eimeria* (Široký et al. 2006). The successive development of secondary sexual characters and sexual activity in one of the specimens indicated that the animals represent a pair potentially capable of reproduction. Therefore, the turtles were placed in a plastic container (115 cm x 80 cm x 80 cm high) shallowly filled with water (depth 18 cm, temperature 24-28 °C), which was equipped with a shelter (a partly submerged plastic box of 56 cm x 39 cm x 22 cm) and a place suitable for egg deposition (a plastic box of the same size as the shelter, filled with a mixture of peat and leaf litter). Information obtained on oviposition and egg incubation as well as growth and coloration of juvenile *M. heliostemma* is summarized below.

First sporadic copulation attempts were recorded in October 2011 (at the turtles' age of ca. 10-11 years). More frequent copulation attempts seen in November 2011, resulted in an obvious scar in the central part of the male's plastron at the place of frequent contact with the female's carapace keel. The cloaca of the female became swollen at this time. A real copulation was observed on January 25, 2012, and the first oviposition took place on February 22, 2012. Data of following ovipositions obtained until the end of 2015 are summarized in Table 1.

The complete process of egg deposition (including digging and covering the nest) observed in February 2012, (5 eggs) and July 2014 (6 eggs) lasted 125 and 65 minutes, respectively. The oviposition proper lasted 26 and 6 minutes, respectively, and the intervals between depositions of individual eggs varied from 0.45 to 5.0 min-

Table 1: Reproduction parameters observed in captive *Mesoclemmys heliostemma* (McCord, Joseph-Ouni & Lamar, 2001).

Date of oviposition	Clutch size	Date of hatching	Number of hatchlings	Duration of incubation period (Days)
22 February, 2012	5	-	-	-
27 June, 2012	2	-	-	-
12 February, 2013	6	01-03 October, 2013	2	231-233
16 April, 2013	2	-	-	-
10 January, 2014	6	09 August, 2014	1	211
29 July, 2014	6	17 March, 2015	2	231
01 January, 2015	9	16 November, 2015	2	229
23 September, 2015	11	?	?	?

Fig. 1: *Mesoclemmys heliostemma* (McCord, Joseph-Ouni & Lamar, 2001). A – female laying eggs; B – hatchling opening the eggshell; C – fresh hatchling, dorsal view; D – the same individual, ventral view; E – fresh hatchling with silver-whitish cephalic mark; F – the same individual after three months; G – hatchling with bright yellow cephalic mark; H – the same individual after three months.

Table 2: Growth of the straight carapace length (SCL; mm) of two parental specimens and seven captive-bred juveniles of *Mesoclemmys heliostemma* (McCord, Joseph-Ouni & Lamar, 2001) (the first measurement of the parental specimens corresponds to the first month of captivity).

Parents SCL			Juveniles SCL							
Month	Male	Female	Month	Juv 1	Juv 2	Juv 3	Juv 4	Juv 5	Juv 6	Juv 7
1	112	77	1	55	59	59	61	59	-	-
3	115	85	3	78	87	-	-	-	82	85
6	125	94	5	87	94	-	-	-		
9	132	111	7	94	104	-	-	-		
12	132	115	10	103	112	-	-	-		
15	136	123	11	-	-	-	106	109		
18	142	129	18	-	-	130				
21	148	135	26	147	161					
24	153	139								
48	198	186								
120	253	272								
132	270	295								
144	285	310								
156	303	311								

utes. The female released each egg within about a second and guided them into the nesting pit with her hind legs (Fig. 1A).

Data summarizing clutch and egg size, incubation period (at 27-29 °C with short irregular fluctuations between 18-31 °C) and measurements (in mm) of hatchlings are as follows: clutch size 2-11 eggs (N = 8); egg size 41.7-50.8 mm x 32.2-41.4 mm (mean 46.7 mm x 35.0 mm; N = 25); incubation period 211-233 days (N = 4; Table 1); straight carapace length 55.3-61.0 (mean 58.5; SD 2.05; N = 5); carapace width 37.7-39.8 (mean 39.1; SD 0.83; N = 5); carapace height 17.0-19.8 (mean 18.3; SD = 1.27; N = 5); plastron length 44.0-45.5 (mean 45.0; SD 1.32; N = 5). Available data on the growth of two parental specimens and seven captive-bred juveniles are summarized in Table 2.

The coloration of seven survived and four fully developed dead hatchlings corresponded fairy well with the description of juvenile coloration given by McCord et al. (2001) and Molina et al. (2012). Despite both parental specimens having an entirely black head upon capture at the estimated age of 2-3 and 10-12 months (see Table 2), all hatchlings possessed a distinct silver-whitish (8 cases; Figs. 1C, 1E) or bright yellow (3 cases; Fig. 1G) V-shaped cephalic mark. Light facial bands originating from the tip of the nostril and passing the dorsal edge of the tympanum arrived at the cranial part of the neck. In the three specimens that had a bright yellow cephalic mark, a narrow, more or less distinct dark brown line bordered the sagittal edges of the facial bands (Fig. 1H). The arms of the marking were interrupted in the area dorsal to the tympanum and continued on the cranial part of the neck.

The light cephalic mark of all the juvenile specimens observed became indistinct relatively fast, by the age of three months (Fig. 1F). In some individuals, the bands had disappeared completely after one year from hatching, in another (the oldest) juvenile very slight traces of these bands remained visible even after 25 months. Similarly, the bright yellow cephalic mark became less distinct within three months from hatching (Fig. 1H) and disappeared completely within 25 months of life. The brightness of the faded facial bands can vary most likely dependant on circadian changes in physiological conditions, as occasionally the visibility of bands was better during the night in some individuals.

Comparison of available information on the duration of the incubation period in Amazonian *Mesoclemmys* indicates that incubation of eggs takes longer time in *M. heliostemma* (7-7.5 months) than in sympatric and closely related *M. raniceps* (Gray, 1855) (4-5 months; Böhm 2009). Data available for the widely distributed species *M. gibba* (SCHWEIGGER, 1812) show that although juveniles hatch commonly after 116-200 days at 28-29 °C, they can hatch even after 252 and 270 days when temperature fluctuates between 25 °C (night) and 27 °C (day) (MÉTRAILLER 2006). A possible explanation of the long incubation period in *M. heliostemma* can be seen in the temporal correlation of the time of oviposition (or hatching) with the seasonal floods in lowland Perú. At Iquitos, the water level of the Amazon River culminates in April and May whereas, the water level is lowest in September (e.g., GOULDING et al. 2003). Assuming that the females of *M. heliostemma* lay the eggs after the inundation maximum (in June and July), the incubation period would last for the entire low water period. If so, the hatchlings would emerge approximately between January and March when the level of the rising water matches the level of the dropping water at the time of oviposition. The estimated age of juvenile specimens of *M. heliostemma* found at Rio Nanay in March 2002 corresponds fairly well to the above idea.

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AUTHOR: Jiří MORAVEC – Department of Zoology, National Museum, Cirkusová 1740, 19300 Praha 9, Czech Republic < jiri.moravec@nm.cz >.