

Supporting Information

Phylogeography of the Asian softshell turtle *Amyda cartilaginea* (BODDAERT, 1770): evidence for a species complex

UWE FRITZ, RICHARD GEMEL, CHRISTIAN KEHLMAIER,
MELITA VAMBERGER, PETER PRASCHAG

Vertebrate Zoology, **64**(2): 229–243.

ERRATUM

Page 238, right column, sentence starting with line 4 must read:
“However, we wish to point out that some specimens of *A. o. phayrei*, ...”

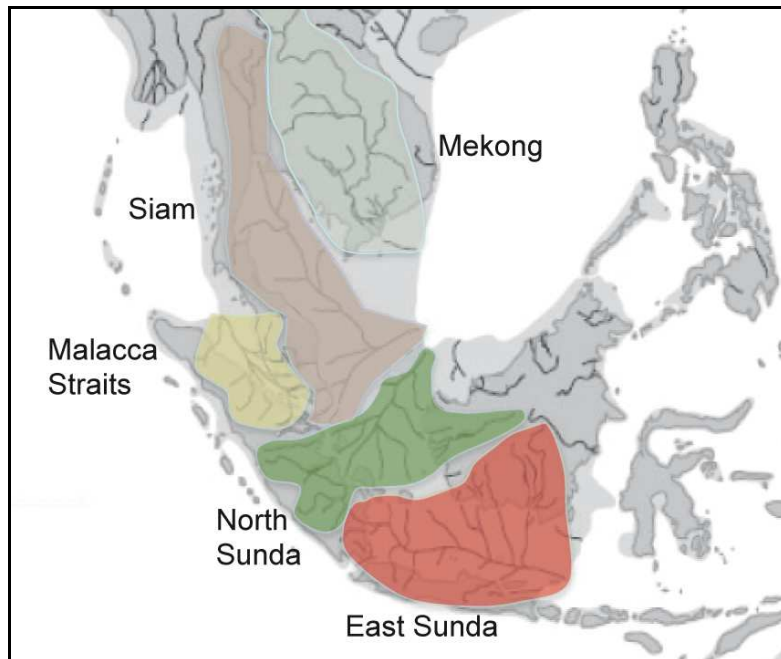


Fig. S1. Quaternary palaeodrainage systems and shorelines in Southeast Asia according to VORIS (2000). Modified from DE BRUYN et al. (2013). Shown is the lowest sea level stand (–120 m).

Table S1. Used samples, GenBank sequences and their accession numbers. For fresh samples, the sequences labelled as *cyt b* and ND4 comprised also adjacent DNA coding for tRNAs. Abbreviations: MTD D – Museum of Zoology, Senckenberg Dresden, Herpetological Collection; MTD T – Museum of Zoology, Senckenberg Dresden, Tissue Collection; NMW – Natural History Museum Vienna. Samples processed in the clean room bear asterisks. Collection year of museum specimens in brackets following locality data. For identification of *Amyda* clades with distinct taxa, see Figure 2.

Taxon	Voucher	Clade	Geographical Provenance	12S	<i>cyt b</i>	ND4	Cmos	ODC	R35
<i>Amyda cartilaginea</i> s.l.	MTD T 6122	4	Bangladesh: hills of Chittagong	LM537429	LM537462	LM537495	LM537528	LM537547	LM537565
<i>Amyda cartilaginea</i> s.l.	MTD T 6123	4	Bangladesh: hills of Chittagong	LM537430	LM537463	LM537496	LM537529	LM537548	LM537566
<i>Amyda cartilaginea</i> s.l.	MTD T 6124	4	Bangladesh: hills of Chittagong	LM537431	LM537464	LM537497	LM537530	LM537549	LM537567
<i>Amyda cartilaginea</i> s.l.	MTD D 42530	6	Cambodia	LM537432	LM537465	LM537498	LM537531	LM537550	LM537568
<i>Amyda cartilaginea</i> s.l.	MTD T 10672	6	Cambodia	LM537433	LM537466	LM537499	LM537532	LM537551	LM537569
<i>Amyda cartilaginea</i> s.l.	MTD D 42529	6	Cambodia: Ratanakiri: Lumphat	LM537434	LM537467	LM537500	LM537533	—	LM537570
<i>Amyda cartilaginea</i> s.l.	NMW 32943*	5	China: Yunnan: Ruili (1993)	LM537435	LM537468	LM537501	—	—	—
<i>Amyda cartilaginea</i> s.l.	NMW 30203:4*	2	Indonesia: East Java: Jombang (1898)	LM537436	LM537469	LM537502	—	—	—
<i>Amyda cartilaginea</i> s.l.	NMW 30206:2*	2	Indonesia: West Java: Garut, Tji Maunk (1928)	LM537437	LM537470	LM537503	—	—	—
<i>Amyda cartilaginea</i> s.l.	NMW 30209:2*	2	Indonesia: West Java: Sukabumi (c. 1900)	LM537438	LM537471	LM537504	—	—	—
<i>Amyda cartilaginea</i> s.l.	NMW 30208:1*	2	Indonesia: West Java: Tasikmalaya (1926)	LM537439	LM537472	LM537505	—	—	—
<i>Amyda cartilaginea</i> s.l.	NMW 30205:2*	2	Indonesia: Kalimantan (Borneo): Liang Buaya (1885)	LM537440	LM537473	LM537506	—	—	—
<i>Amyda cartilaginea</i> s.l.	NMW 14909*	2	Indonesia: Kalimantan (Borneo): Muara Teweh (1883)	LM537441	LM537474	LM537507	—	—	—
<i>Amyda cartilaginea</i> s.l.	NMW 30204:4*	3	Indonesia: Kalimantan (Borneo): Nanga Badau (“Nanga Bandang”; 1874)	LM537442	LM537475	LM537508	—	—	—
<i>Amyda cartilaginea</i> s.l.	MTD T 10673	2	Indonesia: Sulawesi	LM537443	LM537476	LM537509	LM537534	LM537552	LM537571
<i>Amyda cartilaginea</i> s.l.	NMW 18249*	3	Indonesia: Sumatra (1942)	LM537444	LM537477	LM537510	—	—	—
<i>Amyda cartilaginea</i> s.l.	MTD T 5312	6	Laos: Vang Vieng	LM537445	LM537478	LM537511	LM537535	LM537553	LM537572
<i>Amyda cartilaginea</i> s.l.	NMW 30205:3*	1	Malaysia: Sarawak (Borneo): Baram river (1901)	LM537446	LM537479	LM537512	—	—	—
<i>Amyda cartilaginea</i> s.l.	MTD T 11234	3	Malaysia: Sarawak (Borneo): Batang Ai dam	LM537447	LM537480	LM537513	LM537536	LM537554	LM537573
<i>Amyda cartilaginea</i> s.l.	MTD T 11233	3	Malaysia: Sarawak (Borneo): Engkari river near Ukum Longhouse	LM537448	LM537481	LM537514	LM537537	LM537555	LM537574
<i>Amyda cartilaginea</i> s.l.	MTD D 40366	5	Myanmar: Lake Inlé	LM537449	LM537482	LM537515	LM537538	LM537556	LM537575
<i>Amyda cartilaginea</i> s.l.	MTD T 9858	5	Thailand	LM537450	LM537483	LM537516	LM537539	LM537557	LM537576
<i>Amyda cartilaginea</i> s.l.	MTD T 9859	5	Thailand	LM537451	LM537484	LM537517	LM537540	LM537558	LM537577
<i>Amyda cartilaginea</i> s.l.	MTD T 9860	5	Thailand	LM537452	LM537485	LM537518	LM537541	LM537559	LM537578
<i>Amyda cartilaginea</i> s.l.	NMW 30204:1*	5	Thailand (1903)	LM537453	LM537486	LM537519	—	—	—

Table S1 continued									
Taxon	Voucher	Clade	Geographical Provenance	12S	cyt b	ND4	Cmos	ODC	R35
<i>Amyda cartilaginea</i> s.l.	NMW 30204:2*	5	Thailand (1903)	LM537454	LM537487	LM537520	—	—	—
<i>Amyda cartilaginea</i> s.l.	NMW 30205:1*	5	Thailand: Bangkok (1870)	LM537455	LM537488	LM537521	—	—	—
<i>Amyda cartilaginea</i> s.l.	NMW 30206:1*	5	Thailand: vicinity of Bangkok (1972)	LM537456	LM537489	LM537522	—	—	—
<i>Amyda cartilaginea</i> s.l.	MTD T 11232	5	Unknown	LM537457	LM537490	LM537523	LM537542	LM537560	LM537579
<i>Amyda cartilaginea</i> s.l.	MTD T 7074	2	Unknown	LM537458	LM537491	LM537524	LM537543	LM537561	LM537580
<i>Amyda cartilaginea</i> s.l.	MTD T 7075	2	Unknown	LM537459	LM537492	LM537525	LM537544	LM537562	LM537581
<i>Amyda cartilaginea</i> s.l.	MTD T 7076	2	Unknown	LM537460	LM537493	LM537526	LM537545	LM537563	LM537582
<i>Amyda cartilaginea</i> s.l.	MTD T 7077	2	Unknown	LM537461	LM537494	LM537527	LM537546	LM537564	LM537583
<i>Dogania subplana</i>	—	—	Unknown	AF366350	AF366350	AF366350	—	HE801842	—
<i>Nilssonina formosa</i>	MTD T 6068	—	China: Yunnan: Shuangbai	HE801637	HE801688	HE801740	HE801763	HE801806, HE801844	HE801869
<i>Nilssonina gangetica</i>	MTD T 6062	—	Bangladesh: Patuakhali district	HE801642	HE801693	HE801742	HE801768	HE801810, HE801846	HE801874
<i>Nilssonina hurum</i>	MTD T 3421	—	Bangladesh: Khulna	HE801657	HE801709	AM495218	HE801778	HE801819, HE801855	HE801884
<i>Nilssonina leithii</i>	MTD T 3099	—	India: Maharashtra: Purna river	HE801668	HE801721	AM495225	HE801788	HE801828, HE801864	HE801894
<i>Nilssonina nigricans</i>	MTD T 3415	—	Bangladesh: Chittagong: Nasirabad: shrine pond	HE801670	HE801723	AM495227	HE801790	HE801830	HE801896
<i>Palea steindachneri</i>	—	—	Unknown	FJ541030	FJ541030	FJ541030	HE801804	HE801843, HE801868	HE801910
<i>Pelodiscus maackii</i>	MTD T 4235, MTD T 4236	—	Russia: Primorsky Territory: Lake Khanka	FM999003	FM999019	FM999011	HE801805	—	HE801911

Table S2. Primer sequences and lengths of PCR products for historical samples. Nucleotides in brackets represent variable positions.

DNA fragment	Primer	Primer sequence (5'-3')	Length of obtained DNA fragment [bp] ¹
12S rRNA short	AmCa_12s_For1	TAC AAT AGA AAT AAC TAC GG	33
	AmCa_12s_Rev1	TTC TTA ATT TAC TAC TAA ATC C	
12S rRNA long	AmCa_12s_For1b	CAA CTA AGA TGG AAG AAA TGG G	63
	AmCa_12s_Rev1	TTC TTA ATT TAC TAC TAA ATC C	
cyt <i>b</i>	AmCa_cytb_For1	GTT TCT CCG TAG ATA ATG CC	108
	AmCa_cytb_Rev1	TCG GTG TTT GAG TTT AAT CC	
ND4	AmCa_ND4_For1	GTC ACA CCA TCA CAC ACA CG	70
	AmCa_ND4_Rev1	TAT AAG CAG TAT A(AC)T GGG CC	

¹ Length after primers were trimmed.

Table S3. PCR conditions for historical and fresh samples.

DNA fragment		ID	C	Thermocycling conditions			
				D	A	E	FE
12S rRNA short	Historical samples	94°C, 5 min	40	94°C, 30 s	54°C, 30 s	72°C, 30 s	72°C, 10 min
12S rRNA long	Historical samples	94°C, 5 min	40	94°C, 30 s	52°C, 30 s	72°C, 30 s	72°C, 10 min
cyt <i>b</i>	Historical samples	94°C, 5 min	40	94°C, 30 s	46–54°C, 30 s	72°C, 30 s	72°C, 10 min
ND4	Historical samples	94°C, 5 min	40	94°C, 30 s	45–49°C, 30 s	72°C, 30 s	72°C, 10 min
12S rRNA	Fresh samples	94°C, 5 min	30–35	94°C, 30 s	50°C, 30 s	72°C, 60 s	72°C, 10 min
cyt <i>b</i> parts 1 & 2	Fresh samples	94°C, 5 min	30–35	94°C, 30 s	50°C, 30 s	72°C, 60 s	72°C, 10 min
ND4	Fresh samples	94°C, 5 min	35	94°C, 30 s	53°C, 30 s	72°C, 60 s	72°C, 10 min
Cmos	Fresh samples	94°C, 5 min	35	94°C, 30 s	58°C, 30 s	72°C, 60 s	72°C, 10 min
ODC	Fresh samples	94°C, 5 min	30–40	94°C, 30 s	60°C, 30 s	72°C, 60 s	72°C, 10 min
ODC alternative	Fresh samples	94°C, 5 min	40	94°C, 30 s	55°C, 30 s	72°C, 60 s	72°C, 10 min
R35	Fresh samples	94°C, 5 min	35	94°C, 30 s	53°C, 30 s	72°C, 60 s	72°C, 10 min

Abbreviations: ID = initial denaturing, C = number of cycles, D = denaturing, A = annealing, E = extension, FE = final extension.