

THE GENUS *TESTUDO* (TESTUDINES: TESTUDINIDAE): PHYLOGENETIC INFERENCES

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Abstract

Phylogenetic analysis based on morphological evidence and recent taxa suggests that the genus *Testudo*, in the sense of most neontologists, is a paraphyletic taxon containing several major monophyletic lineages. *Testudo hermanni* group is most primitive with respect to all remaining taxa. *Agrionemys* is a valid genus for conventional *T. horsfieldii*. Taxa characterized by xiphiplastral kinesis have evolved into a primitive Middle Eastern clade, and additionally an African-Eurasian clade with two major monophyletic branches. *Testudo graeca* is paraphyletic and comprises several species. The cladistic hypothesis agrees with a phenetic analysis, and the results have good bootstrap support.

Key words: Testudines; Testudinidae; *Testudo*; Phylogenetic analysis; Unnatural taxon

Introduction

Studies dealing with phylogenetic relationships of Mediterranean and Central Asian tortoises of the genus *Testudo* Linnaeus, 1758 sensu lato (reviewed in Lapparent de Broin, 2001), i.e. inclusive of *Agrionemys* Khozatsky & Mlynarski, 1966 – are comparatively rare. Only Gmira (1993a, 1993b, 1995) has published results based on a cladistic analysis. Taxa commonly classified into *Testudo* s. l. are presented in Iverson (1992), and others have been recently described (Bour, 1995; Perälä, 1996, 2001a in press, 2002 this volume; Pieh, 2001; Pieh & Perälä, 2002 in press). Taxa or groups included in this analysis are listed in Materials and Methods.

Agrionemys, created for *T. horsfieldii* Gray, 1844 (currently with three subspecies), and which genus is characterized among other features by an immobile posterior plastron lobe (Khozatsky & Mlynarski, 1966; Mlynarski, 1966), is regarded valid by many palaeontologists (e.g., Chkhikvadze, 1970; Lapparent de Broin, 2000a, 2000b, 2001; Hervet, 2000), and it includes *Testudo hermanni hermanni* Gmelin, 1789 and *T. h. boettgeri* Mojsisovics, 1889 – according to Gmira (1993a, 1993b, 1995). The *T. hermanni* group was earlier classified in the otherwise fossil genus *Protestudo* Chkhikvadze, 1970 (Chkhikvadze, 1970). However, the type of *Protestudo* is a representative of *Agrionemys*, according to Lapparent de Broin (2000a, and pers. comm.), making *Protestudo* essentially a junior synonym of *Agrionemys*. Systematists dealing with modern species are hesitant to recognize *Agrionemys*. It is sometimes listed as a subgenus of *Testudo* (Auffenberg, 1974; Iverson, 1992).

Results from a previous (and only) cladistic analysis on *Testudo* suggest essentially that, *T. horsfieldii* and *T. hermanni* constitute a monophyletic group (genus

Agrionemys), as mentioned above, and that the remaining taxa showing xiphiplastral kinesis form the genus *Testudo* sensu stricto, inclusive of *T. kenitrensis* Gmira, 1993 – a fossil taxon from Morocco (Gmira, 1993a, and reproduced 1993b, 1995). Gmira (1993a) elevated *T. graeca graeca* Linnaeus, 1758, *T. g. ibera* Pallas, 1814 and *T. g. terrestris* Forsskål, 1775 onto species rank, but regarded *T. g. zarudnyi* Nikolski, 1896 as a subspecies of *ibera*. Most reviewers retain the concept of *T. graeca* being a single species with numerous subspecies from W Africa to E Iran, as suggested by Mertens (1946). However, Gmira's taxonomy with respect to the *T. graeca* complex is favoured here, except that *T. zarudnyi* and *T. floweri* Bodenheimer, 1935 are also considered to be full species by morphological and geographical criteria (Perälä, 2002 in this volume). Among *Testudo* s. s., *T. terrestris* is basal, followed by (from primitive to derived) *T. ibera*, *T. graeca*, and a clade comprising *T. kleinmanni* Lortet, 1883 and *T. marginata* Schoepff 1792 – whereas the position of *T. kenitrensis* could not be determined unambiguously (Gmira, 1993a). In her second hypothesis, the primitive position of *Agrionemys* is retained, but *T. ibera* is basal within *Testudo* s. s., and followed by a *terrestris-graeca* clade, and a derived monophyletic group comprising *marginata* and *kleinmanni* (either in- or exclusive of *kenitrensis*) (Gmira, 1993a). A subgeneric status was suggested for the *T. marginata* and *T. kleinmanni* lineage (Gmira, 1993a), taxa considered close relatives already by Boulenger (1889), but this subject did not rise again later (Gmira, 1995).

Other accounts of phylogenetic interest, but which will not be reviewed here, include Siebenrock (1913), Mertens (1946), Obst & Ambrosius (1971), Crumly (1984), Crumly (in: Iverson, 1992), and Sqalli-Houssaini

& Blanc (1990).

This article re-evaluates *Testudo* s. l. on the grounds of cladistic methodology, i.e. relationships are inferred based on shared derived characters (synapomorphies). Only recent species are examined. All generally recognized taxa are taken into account, as well as additional populations representing natural groups. The analysis is inclusive of very recent data, some of which were not presented at the Hyères congress. Nevertheless, much of the available material is still incompletely analysed. Thus, this study is to be taken as provisional. The discussion is kept accordingly brief.

Materials and Methods

Specimens examined. – The material includes all *Testudo* s. l. specimens, some 2000 in total, from the following collections: The Natural History Museum, London; California Academy of Sciences, San Francisco; Field Museum of Natural History, Chicago; Museum of Zoology, Hebrew University of Jerusalem; Museum of Comparative Zoology, Harvard University, Cambridge; Muséum d'Histoire Naturelle Lyon; Muséum National d'Histoire Naturelle, Paris; Zoology and Palaeontology departments; Staatliches Museum für Tierkunde, Dresden; Museum of Vertebrate Zoology, University of California, Berkeley; Naturhistorisches Museum, Vienna; Chelonian Research Institute, Oviedo; Staatliches Museum für Naturkunde, Stuttgart; Zoological Museum, Tel-Aviv University; Florida Natural History Museum, University of Florida, Gainesville; Zoological Museum, University of Jordan, Amman; The Finnish Museum of Natural History, Helsinki; The National Museum of Natural History, Washington D.C., and the collection of the author at the University of Bristol. Additionally, representatives of *Indotestudo* Lindholm, 1929 were examined in London and Paris.

Ingroup taxa / groups, with rough geographical distributions. – (1) *Testudo graeca* s. s. (Algeria); (2) *Testudo graeca anamurensis* Weissinger, 1987 (ancient Cilicia, and vicinity in S Turkey); (3) *Testudo graeca armeniaca* Chkhikvadze & Bakradze, 1991 (Araks River valley: E Turkey, Armenia); (4) *Testudo ibera* s. s. (Kura River valley, Caucasus); (5) *Testudo graeca nikolskii* Chkhikvadze & Tuniyev, 1986 (isolated on N Black Sea coast in NW Caucasus); (6) *Testudo terrestris* (Middle East): detailed distribution in Perälä (2002) in this volume; (7) *Testudo zarudnyi* (Khorāsān province, E Iran); (8) *Testudo antakyensis* Perälä, 1996 (N Levant): detailed distribution in Perälä (2002) in this volume; (9) *Testudo graeca soussensis* Pieh, 2001 (Souss Valley, Morocco); (10) *Testudo floweri* (SW Levant) detailed distribution in Perälä (2002) in this volume; (11) *T. nabeulensis* (Highfield, 1990) (Tunisia-NW Libya): this species, but not its original genus *Furculachelys* Highfield, 1990, is considered valid by Pieh & Perälä (2002 in press), see Highfield (1990) for further details; (12) Group "Highland" (South-Central Levant): detailed distribution in Perälä (2002) in this volume; (13) Group "Libya", a species from Cyrenaica, Libya, to be described in Pieh & Perälä (2002); (14) Group "Zagros" (Zagros mountains in the Middle East), a species described elsewhere in this volume (Perälä, 2002); (15) *Testudo hermanni hermanni* (SW Europe); (16) *Testudo hermanni*

boettgeri (SE Europe); (17) *Testudo kleinmanni* (N Africa); (18) Group "Negev" (N Sinai, Egypt - Nege desert, Israel): a species described in Perälä (2001a, in press); (19) *Testudo marginata* (Greece, S Albania, Italy, Sardinia); (20) *Testudo weissingeri* Bour, 1999 (SW Peloponnese, Greece); (21) *Testudo horsfieldi horsfieldii* (at least southern Afghanistan, Pakistan); (22) *Testudo horsfieldii kazakhstanica* (Chkhikvadze, 1988) (at least Kazakhstan and parts of Uzbekistan); (23) *Testudo horsfieldii rustamovi* (Chkhikvadze, Amiranashvili & Ataev, 1990) (at least SW Turkmenistan).

Taxa listed here as subspecies are likely to be full species, but conventional taxonomy is retained until the matter is tested.

Phylogenetic inference. – Phylogenetic analyses were performed using PAUP* Version 4.0b8 for Windows (Swofford, 2001). Trees were edited using TreeView Version 1.5.2 (Page, 1996), and Micrografx Window Draw 5.0 (Micrografx, Inc.). The character matrix contains 24 taxa and 61 characters. 54 characters are strictly morphological. As for the remaining morphometric characters, character states were determined by statistical significance from ANOVA and the non-overlap of 95% confidence intervals. A morphometric character was standardized for maximum carapace length following procedures in Perälä (2001; 2002 in this volume). Character polarities were determined using *Indotestudo* from South Asia as outgroup, thus codings found in the data matrix do not indicate a priori assumptions of primitive or derived character states. Another analysis was run with the most primitive taxon from the first analysis as outgroup, to check whether this would have any function on the initial topology. If not, this would maximize confidence about the selection of the initial outgroup. The single multistate character was treated as ordered (Wagner character). All characters have equal weights. Missing characters were coded with question marks. Phylogenetic hypotheses were reconstructed using maximum parsimony and branch-and-bound search, guaranteed to find the most parsimonious tree (MPT), or trees. Results were subjected to bootstrap analysis using default settings. The character data were also subjected to an Archie-Faith-Cranston randomization test for phylogenetic structure. Additionally using the random trees option, one million trees were sampled randomly from the set of all possible trees to test the robustness of the phylogenetic result. Though phylogenetic hypotheses based on overall similarity are not considered sound from a theoretical point of view, a phenogram was constructed using neighbor-joining distance for comparative reasons, and to cheer up those readers attracted to a phenetic approach. The character matrix with taxon assignments is presented in Fig. 1.

List of characters and character states. – (1) Typically a fixed and rigid xiphiplastron (no plastral kinesis): 0 = absent, 1 = present; (2) Epiplastral region heavily flattened: 0 = absent, 1 = present; (3) Typically humero-pectoral sulcus crosses the entoplastron: 0 = absent, 1 = present; (4) Anterior plastron lobe heavily tilted relative to horizontal level: 0 = absent, 1 = present; (5) Gular pouch / epiplastral excavation very small, 1

present at all: 0 = absent, 1 = present; (6) Gular pouch / epiplastral excavation very small (and spheroid): 0 = absent, 1 = present; (7) Gular pouch / epiplastral excavation moderately deep and deltoid: 0 = absent, 1 = present; (8) Gular pouch / epiplastral excavation very well developed and deep: 0 = absent, 1 = present; (9) Entoplastron typically not concealed anteriorly by gulars / epiplastral lip in dorsal view: 0 = absent, 1 = present; (10) Neural series consists typically of 8 neurals: 0 = absent, 1 = present; (11) Last two or three neurals typically hexagonal, with short lateral sides facing anteriorly: 0 = absent, 1 = present; (12) Typically one (fused) suprapygale bone only: 0 = absent, 1 = present; (13) Bony carapace rim typically without excessive spiking (character retained into adulthood): 0 = absent, 1 = present; (14) Supracaudal sulcus crosses onto suprapygale: 0 = absent, 1 = present; (15) Except for the anterior notch, gulars more or less edged (rectangular) in dorsal view, especially in males: 0 = absent, 1 = present; (16) Anterior proximal part of epiplastron rounded, indented towards rear in ventral view: 0 = absent, 1 = present; (17) Gular impression (of single scute) on epiplastron dorsally shorter than wide (at least in males): 0 = absent, 1 = present (Note: This character is much more pronounced in *hermanni* and *horsfieldii* than in any other group, but it was decided not to use multistates.); (18) Anterior rim of epiplastron frequently rounded (domed outwards) proximally, in dorsal / ventral view: 0 = absent, 1 = present; (19) Gulars dorsally very long: 0 = absent, 1 = present; (20) Gulars excessively cuneiform in dorsal aspect: 0 = absent, 1 = present; (21) Females exceptionally high-domed (very high shell: up to over 56% of length): 0 = absent, 1 = present; (22) Central median carapacial keel retained into adulthood (at least in first vertebral): 0 = absent, 1 = present; (23) Dimensions of anterior shell opening very small: 0 = absent, 1 = present; (24) Anterior shell opening close to being tent-shaped in anterior view (overturned gentle U-shape): 0 = absent, 1 = present; (25) Anterior proximal border of nuchal bone rounded and protruding heavily towards the anterior: 0 = absent, 1 = present; (26) Supracaudal typically divided centrally (in longitudinal section): 0 = absent, 1 = present; (27) Supracaudal flared heavily to the rear in both sexes: 0 = absent, 1 = present; (28) Supracaudal more or less V-shaped in dorsal and ventral view: 0 = absent, 1 = present; (29) Shell elongate and trapezoidal in dorsal view, especially in males: 0 = absent, 1 = present; (30) Mid-body very narrow at bridge (at marginals 5-6): 0 = absent, 1 = present; (31) Slender mid-body considerably rounded (double-S-shaped outline) in dorsal view: 0 = absent, 1 = present; (32) Anterior carapace rim strongly inverted centrally (deep cervical notch): 0 = absent, 1 = present; (33) Very short ventral diagonal length of first costal: 0 = absent, 1 = present; (34) Anterior and posterior marginals strongly flared in both sexes: 0 = absent, 1 = present; (35) Height apparently always less than half the length of plastron: 0 = absent, 1 = present; (36) Horny tail claw (retained in skeleton): 0 = absent, 1 = present; (37) Tail tubercle very prominent: 0 = absent, 1 = present; (38) Aggregate of enlarged round or diamond-shaped tubercles found typically on upper thigh region: 0 = absent, 1 = present; (39) Thigh-spurs: 0 = absent, 1 = absent or present, 2 =

present. Note: in *T. marginata* and *T. weissingeri* the frequency of individuals with or without spurs is not significantly different (Perälä, 2001b, in press). Whether this is true for *T. g. soussensis*, allegedly a spurless species, has not been tested – however, individuals with spurs are encountered frequently in museum samples; (40) Typically 4 claws (and digits) on fore feet: 0 = absent, 1 = present; (41) Parietal exhibits strong downward curvature posteriorly: 0 = absent, 1 = present; (42) Dorsal skull outline modestly convex throughout: 0 = absent, 1 = present; (43) Long parietal-supraoccipital complex ca. straight, or slightly convex: 0 = absent, 1 = present; (44) Edge of upper jaw may be slightly serrated (dentate): 0 = absent, 1 = present; (45) Edge of lower jaw (dentary) may be slightly serrated (dentate): 0 = absent, 1 = present; (46) Prootic not concealed by the parietal anteriorly and dorsally: 0 = absent, 1 = present; (47) Prefrontal bone projecting anteriorly (deep V-shaped maxillary-prefrontal notch present in lateral view): 0 = absent, 1 = present; (48) Crista supraoccipitalis projects posteriorly far beyond condylus occipitalis: 0 = absent, 1 = present; (49) Tip of short crista supraoccipitalis typically round, blunt and paddle-like: 0 = absent, 1 = present; (50) Tip of short and slender crista supraoccipitalis typically moderately sharp and bent slightly downwards: 0 = absent, 1 = present; (51) Males with very short relative plastron length (typically less than 85% of CL): 0 = absent, 1 = present; (52) Entoplastron usually hexagonal: 0 = absent, 1 = present; (53) Entoplastron usually heptagonal (with transversely straight anterior border): 0 = absent, 1 = present; (54) Extremely large anterior and posterior space between plastron surface and ventral carapace rim: 0 = absent, 1 = present; (55) Thigh-spurs broad-based cornets: 0 = absent, 1 = present; (56) Gulars typically projecting anteriorly beyond first and second marginals: 0 = absent, 1 = present; (57) Gulars ventrally wide and short (high gular width/length ratio, around 1.5): 0 = absent, 1 = present; (58) Very strong reversion of posterior marginals in males: 0 = absent, 1 = present; (59) Male supracaudal double-S-shaped in dorsal view: 0 = absent, 1 = present; (60) Tendency for first vertebral to develop lyre-shaped: 0 = absent, 1 = present; (61) Vertebrae 2-4 form a relatively streamline series, especially in males (maximum width not much greater than width of transverse seams separating vertebrae): 0 = absent, 1 = present. All characters are subject to individual variation.

Results

Using *Indotestudo* as outgroup, one most parsimonious tree is retained. Length = 87 steps; consistency index (CI) = 0.7126. The phylogeny of *Testudo* s. l. according to this hypothesis is as follows (Fig. 2). *T. h. hermanni* and *T. h. boettgeri* form a basal monophyletic lineage. A *Testudo horsfieldii* clade including all three taxa is derived with respect to the *hermanni*, but primitive with regard to the remaining taxa. *T. h. rustamovi* is basal within this lineage. *Testudo* s. s. comprises three major geographical clades. These are: A monophyletic Middle Eastern group with *T. terrestris* and Highland, *T. antakiensis* and *T. floweri* as sister clades, respectively. *Testudo ibera* s. s. alone constitutes a monophyletic lineage between the strictly

Middle Eastern group and the remaining major clades. Secondly, there is essentially an African group with a basal monophyletic *T. graeca* s. s. – *T. nabeulensis* lineage, and a more derived clade comprising *T. g. soussensis* (basal), Libya, *T. kleinmanni*, and Negev. The remaining major clade, a sister group of the previous one, is formed by Asian and southern European populations: *T. g. armeniaca* (basal), Zagros, *T. g. nikolskii*, *T. zarudnyi*, *T. g. anamurensis*, and finally *T. marginata* and *T. weissingeri*, which species are unsurprisingly each other's sisters. A bootstrap provides good support for all major clades (75–99% support for the major lineages). The *hermanni* clade, like the monophyletic *horsfieldii* complex, have good bootstrap support (91% and 99% respectively). Taxa characterized by xiphiplastral kinesis form a robust clade (94%). The branch leading to *T. ibera* s. s., as well as the one supporting the *terrestris*–Highland–*antakyensis*–*floweri* clade have been collapsed, but *floweri* and *antakyensis* are each other's sister species with a high degree of confidence (99%). The hypothesis from bootstrap analysis is depicted in Fig. 3. Results from a neighbor-joining analysis are fully identical with those retained from the initial cladistic analysis. The evaluation of 1 million trees sampled equiprobably from the set of all possible trees resulted in a mean tree length of 257.27 steps (± 13.80 standard deviation), and a shortest tree of 170 steps (one tree). This suggests the data contain a strong phylogenetic signal, and that the original phylogenetic hypothesis was not by-product of chance. The Archie-Faith-Cranston test yielded a highly significant result for phylogenetic structure ($p < 0.01$).

In a subsequent analysis with *Indotestudo* deleted and *T. hermanni* complex as outgroup, the cladistic hypothesis is exactly the same as previously, with no change in tree topology. The only possible MPT has a length of 80 steps. The consistency index is higher than previously (CI = 0.7750) because overall homoplasy is reduced due to an optimal outgroup. The fact that topology does not change with the change of outgroup gives added confidence for the results. The optimization of the outgroup is also reflected in better bootstrap support (Fig. 4). The separation of the combined *horsfieldii*–*Testudo* s. s. clade from the *hermanni* complex is very well supported (98% bootstrap confidence). The monophyly of the *T. horsfieldii* complex (99%), a group lacking xiphiplastral kinesis (among *hermanni* taxa), as well as the monophyly of *Testudo* s. s. (100%), the clade comprising groups with a hinged plastron, is unambiguous. *T. ibera* s. s. is very likely a member of the Middle Eastern clade (or clades), and not a basal sister lineage in an African-Eurasian clade, as indicated by a weak bootstrap value (56%). The relative uncertainty of the phylogenetic position of *T. ibera* s. s. is most likely due to a complete lack of skull data from the Kura River valley. Actually, mutual relationships among the initial Middle Eastern clade and *ibera* s. s. are largely unresolved except for the sister group relationship of *antakyensis* and *floweri* (99%). The combined African-Eurasian clade (98%), as well as the dichotomy within it into monophyletic Eurasian (93%) and African (91%) lineages is well supported, as are by and large phylogenies within the Eurasian lineage (relative phylogenies of *anamurensis* and *zarudnyi* remain shaky).

Although the monophyly of the African clade has good support the mutual relationships within the clade have not, with the exception of the sister group relationship of *T. kleinmanni* and the Negev tortoise. A neighbor-joining phenogram is otherwise identical to the most parsimonious cladistic hypothesis, but separate clusters of *T. terrestris* plus Highland, and *T. antakyensis* and *T. floweri*, respectively, form independent lineages (Fig. 5).

Discussion

The phylogenetic hypotheses presented herein are generally well supported, and they make geographical sense. The fact that two testable methods, cladistics and a phenetics, produce essentially the same hypotheses provides added confidence for the results. According to the present hypothesis, *Testudo* s. l. consist of four important monophyletic groups (including one with a major dichotomy). The concept of a monophyletic *T. graeca* (Mertens, 1946) is unbearable.

Agrionemys sensu Gmira (1993a, 1993b, 1995) is not a natural group even though *hermanni* and *horsfieldii* share many primitive characters relative to the other taxa. Otherwise, phylogenies correspond well with the basic structure of Gmira's first hypothesis (Gmira, 1993a, 1993b, 1995). On the other hand, the old inference that the *T. hermanni* complex is most primitive of all *Testudo* (Chkhikvadze, 1970), is well supported. This monophyletic lineage comprising *T. h. hermanni* and *T. h. boettgeri* should clearly be classified as a separate genus. Because *Protestudo* is synonymous with *Agrionemys* (Lapparent de Broin, 2000a), and no other name is available, a new genus should be created for the *hermanni* group. This would be sensible on the basis of a unique neural configuration alone. In *hermanni*, and excluding individual abnormalities, there are typically 8 neurals, in other taxa typically 7; see also Pritchard (1988). The last 2–3 neurals are typically hexagonal (primitive) in *hermanni*. In other *Testudo* s. l. neurals are typically quadrilateral and octagonal, and only the last (7th) may be hexagonal in a normally developed series without deformities. Even when the last neural is hexagonal, the bone is typically not of the same shape, i.e. with short lateral sides anteriorly, as found in *hermanni*. Other unique characters for the *hermanni* complex (relative to the remaining *Testudo* s. l.) are a longitudinally divided supracaudal, the relative vertical flattening of the epiplastral region or anterior plastron lobe, and many characters not taken into consideration in this analysis, like the (large) number of scales on anterior extremities, the presence of "bald" regions on the same, skull characters, and characters pertaining to the manus.

Though the name is not applicable for the *hermanni* lineage, *Agrionemys* is a valid genus for the *horsfieldii* complex only. Characters unique for *Agrionemys* include the presence of only 4 claws and digits in the anterior extremities (5 digits in all other groups though the number of claws may vary between 4 and 5 in *hermanni*), a vertebral keel retained into adulthood (at least on the first vertebral), the dorsal supracaudal sulcus which crosses onto the suprapygial bone, the heavily flattened upper carapace, the aggregate of enlarged round or diamond-shaped tubercles on the upper thigh region, and other characters not discussed herein, but associated

especially with the skull, and manus. The three recent *Agrionemys horsfieldii* subspecies (*horsfieldii*, *kazachstanica*, *rustamovi*) clearly deserve full species status, as already hinted by Chkhikvadze et al. (1990). The mutual combinations of characters 52, 53, and 54 alone support such a classification, in addition to other shell features, but also manus and skull characters.

Testudo s. s. is completely separable from the two primitive clades by an important derived character (among others): a plastron with a movable posterior lobe. Pertaining to this clade, *Testudo graeca* sensu lato (inclusive of a multitude of subspecies) is paraphyletic without question. This unnatural taxon comprises several full species by any standards. The situation highlights the relative lack of taxonomic study of the recent Old World testudinids in comparison with, e.g., taxonomic advances made in North American turtle research within a period since the publication of the Handbook of Turtles (Carr, 1952) half a century ago, until today. Though superficially more or less similar, all *T. graeca* s. l. dealt with here can be identified by unique characters, which do not vary clinally – but this subject is not within the scope of this article. *T. marginata* and *T. weissingeri*, and *T. kleinmanni* with its sister species from the Negev are not only derived members of their respective clades, according to the present hypothesis, but they are also phylogenetically closer to some *graeca* than other nominal subspecies of *graeca* are with regard to each other. An endnote by Perälä (in: Lapparent de Broin, 2001) suggested the existence of an African-Middle Eastern clade, based on an earlier (unpublished) analysis where a *graeca-nabeulensis* clade was sister to the Middle Eastern taxa. After a re-evaluation of skull characters this relationship is not supported by the present hypothesis, and *graeca-nabeulensis* is a sister to the other Africans.

Within *Testudo s. s.*, the primitive, more or less Middle Eastern clade (*terrestris*, Highland, *antakyensis*, *floweri*, and *ibera* s. s.) is characterized by the unique combination of xiphiplastral kinesis incorporated into a plastron with a very weakly developed epiplastral excavation (gular pouch), if present at all. This is the only group among *Testudo s. s.* characterized by an anteriorly projecting prefrontal, i.e. a deep V-shaped maxillary-prefrontal notch observed best in lateral view, and which character is found otherwise in the two more primitive clades only. The parietal is deeply notched posteriorly. Edges of jaws can be serrated (dentate), a character not

found in the more derived clades, except for, interestingly, in the Libyan species, in which the maxillary shows weak serration. Curiously, *floweri* and *antakyensis* feature a skull with the prootic exposed. This character has evolved in three other species in the African clade (Libya, *kleinmanni*, Negev); see also Bour (1989). The character is derived in *Testudo* s. l., and it seems to have evolved twice independently. The morphological distinction of the Middle Eastern lineage or lineages should eventually be mirrored in classifications.

The remaining *Testudo s. s.*, the African and Eurasian clades, are separable from the more primitive clades by several combinations of derived characters pertaining to skull structure (e.g., reversal of deep parietal notching relative to the Middle Eastern clade), several features pertaining to the gular and epiplastral structures – characters more primitively expressed in the African lineage. A conservative approach would be to retain the generic name *Testudo* Linnaeus, 1758 – type species *Testudo graeca* Linnaeus (with a type locality in Santa Cruz, Oran, Algeria), for this African-Eurasian clade only. However, the structural differences between both major lineages within this group are so great as to suggest at least subgeneric status for both. A subgeneric rank for *T. kleinmanni* and *T. marginata* (Gmira, 1993a) is not supported by the present hypothesis, but both species share a common ancestry at the lineage leading to the African-Eurasian clade. Therefore, it is neither surprising that the absence of thigh-spurs, a reversal in *Testudo s. s.* as pointed already by Gmira (1993a, 1993b, 1995), has occurred in both the African (*soussensis*, *kleinmanni*, Negev) and the Eurasian clades (*marginata*, *weissingeri*), but which process is seemingly not complete in either *soussensis* (pers. obs.), or *marginata* and *weissingeri* (Perälä, 2001b).

In conclusion, *Testudo* s. l. phylogenies presented here are fairly sensible from a geographical point of view, and are by and large in congruence with views expressed earlier by palaeontologists, especially as reviewed recently by Lapparent de Broin (2001). According to the present hypothesis, the genus *Testudo*, in the sense of most neontologists (reviewed in Iverson, 1992), is an unnatural taxon comprising several lineages at generic, and possibly subgeneric, level, as well as one paraphyletic taxon at species level (*T. graeca*). *T. graeca* s. l. is a composite of several species. This situation is not accurately reflected in current taxonomy.

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GROUPS	CHARACTERS												
	1	2	3	4	5	6							
	0	0	0	0	0	0							
INDOTESTUDO	11101	00011	10011	01000	00000	00000	00000	10000	00011	10001	01000	01000	0
HERMANNI	11001	00011	11001	11000	00000	10000	00000	10000	00011	01000	01000	00000	1
BOETTGERI	11101	00011	11001	11000	00000	10000	00000	10000	00011	01000	01000	00000	1
HORSFIELDII	10101	00010	00011	11000	01000	00000	00001	10101	00011	01000	00100	10000	1
KAZACHSTANICA	10101	00010	00011	11000	01000	00000	00001	10101	00011	01000	00110	10100	1
RUSTAMOVI	10101	00010	00011	11000	01000	00000	00001	10101	00011	01000	01000	10000	1
TERRESTRIS	00001	00010	00001	01000	00000	00000	00000	00020	10011	01010	00000	00000	1
FLOWERI	00000	10010	00101	01000	00000	00000	00000	01020	11011	11000	00000	00000	1
HIGHLAND	00001	00010	00001	01000	00000	00000	00000	00020	10011	01010	00000	00000	1
ANTAKYENSIS	00000	10010	00101	01000	00000	00000	00000	01020	10011	11000	00000	00000	1
IBERA	00001	00010	00001	01000	00000	00000	00000	00020	?????	?????	00000	00000	0
ARMENIACA	00000	00100	00000	00100	00000	00000	00000	00020	?????	?????	00001	10000	0
GRAECA	00000	01000	00100	00000	00000	00000	00000	00020	00000	00001	00000	00001	0
NABEULENSIS	00000	01000	00100	00000	10000	00000	00000	00020	00000	00001	00000	01001	0
ZARUDNYI	00000	00100	00000	00111	00101	00011	00010	00020	00100	00100	00000	00000	0
ZAGROS	00000	00100	00000	00101	00001	00000	00000	00020	00100	00100	00000	00000	0
NIKOLSKII	00000	00100	00000	00111	00101	00000	00000	00020	?????	?????	00000	00000	0
ANAMURENSIS	00000	00100	00000	00111	00101	00011	00110	00020	00100	00100	00000	00000	0
SOUSSENSIS	00000	01000	00000	00000	00100	00000	00000	00010	00000	00001	00000	00001	0
LIBYA	00000	01000	00000	00000	00100	00001	10000	00020	00001	10001	00000	00111	0
MARGINATA	00000	00100	00000	00111	00101	01011	00110	00010	00100	00100	10000	00000	0
WEISSINGERI	00000	00100	00000	00111	00101	01011	00110	00010	00100	00100	00000	00000	0
KLEINMANNI	00010	01000	01100	00000	00110	00101	01000	00000	00000	10001	00000	00001	0
NEGEV	00010	01000	01100	00000	00110	00101	11000	00000	00000	10001	00000	00000	0

Fig. 1: Phylogenetic data matrix containing 61 characters. Groups (left column) are assigned to character states 0-1, or in one case, 0-2 (multistate character 39). Missing data = ?. See text for character descriptions.

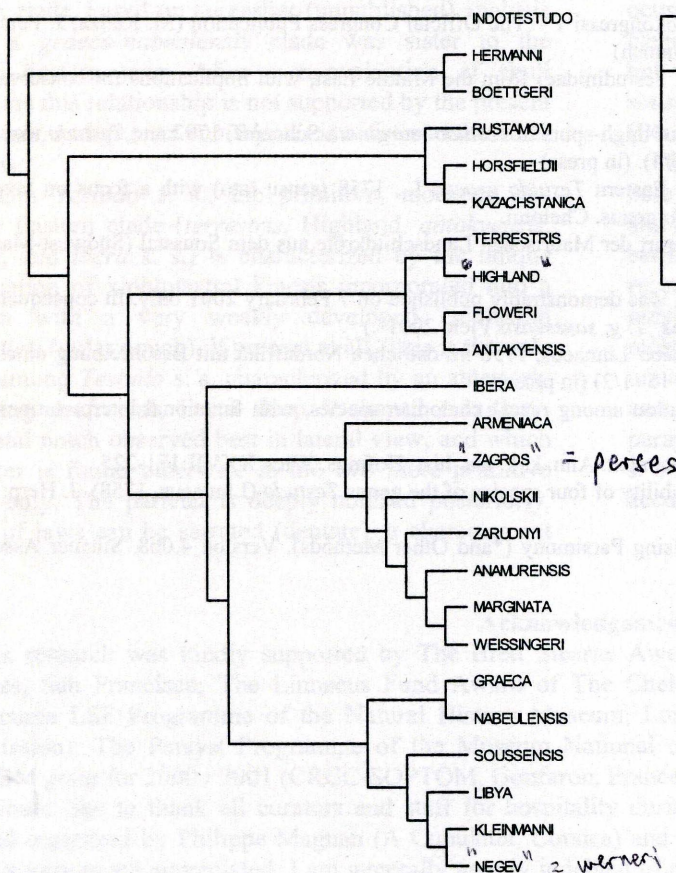


Fig. 2: Initial phylogenetic hypothesis with *Indotestudo* as outgroup. TL = 87 steps.

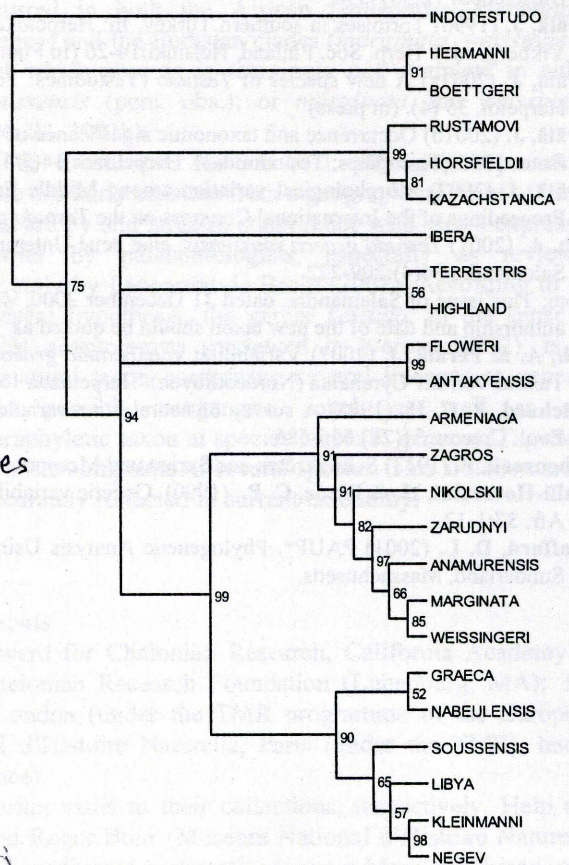


Fig. 3: Bootstrapped tree from first analysis with *Indotestudo* as outgroup

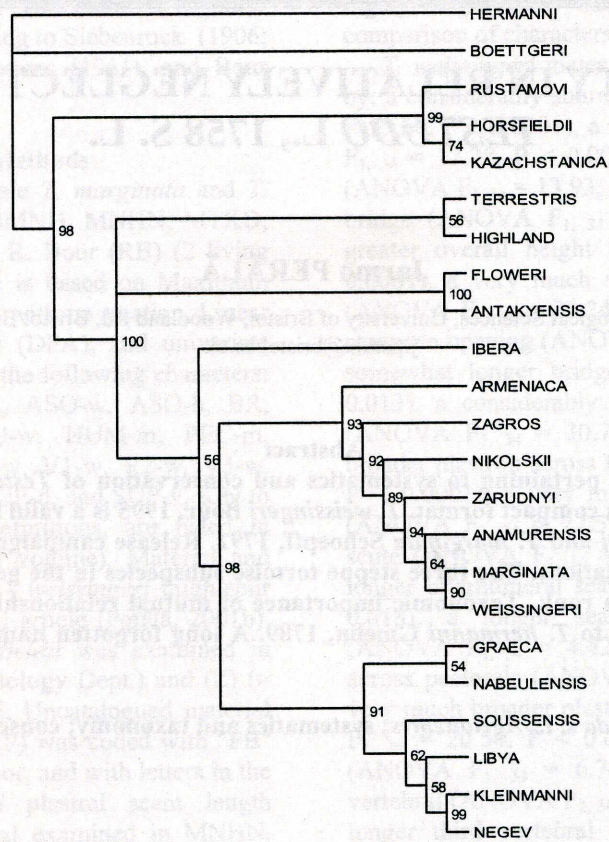


Fig. 4: Bootstrapped phylogenies with *T. h. hermanni* and *T. h. boettgeri* as outgroup.

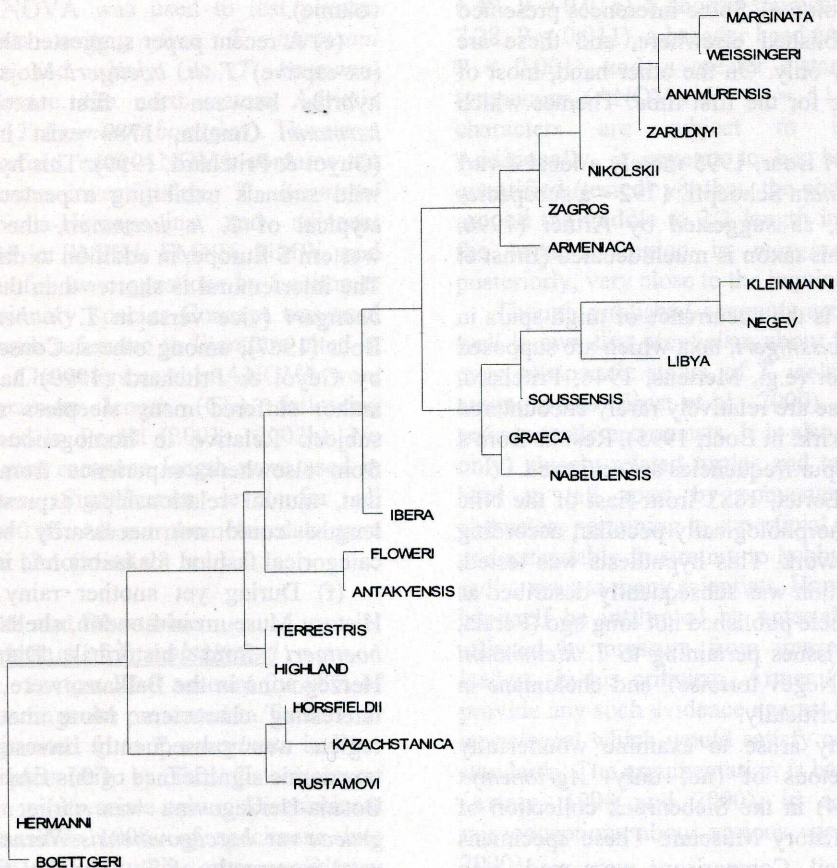


Fig. 5: Neighbour-joining phenogram depicting *Testudo* s. l. relationships