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TO THE STUDY OF FOSSIL BONE REMAINS OF TORTOISES (TESTUDINES, TESTUDINIDAE) FROM THE BINAGADI BITUMEN DEPOSIT (AZERBAIJAN)

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Summary. The objects of the study were fossilised bone remains of tortoises extracted from the soil during excavations in the Binagadi bitumen deposit and suburban summer cottages of Absheron. The material was collected and processed during the period from 2020 to 2023 during expeditionary and laboratory researches. The study of the material revealed a large number of bone fragments. The generic and species affiliation of the discovered bone fragments of tortoises was established. The study of the skeletal remains of tortoises found in the Binagadi bitumen deposit in the Late Pleistocene layer (the Late Dryas) and its comparison with the bones of tortoises found in the Holocene layer (the Middle Subatlantic period) on Absheron allowed assuming that the climate at the end of the Pleistocene was cooler and more humid than in the Holocene. This was indicated by some changes in the morphology of the bony shell of tortoises: a decrease in the indices of the ratio of shell length to its width and height, as well as the width of the shell to its height; changes in the size proportions of some bone plates of the carapace and plastron. All these changes presumably reflect some intensification of the adaptation processes associated with significant climate changes in Azerbaijan in general and in its eastern part particularly during the specified geological period (more than 0.012 million years ago).

Introduction

Bitumen deposits with ancient remains of flora and fauna are confined to areas where oil comes to the Earth's surface and are most often of Quaternary age. In deposits of this type, organic remains accumulate over a long period of time and have a fairly diverse species composition, with the remains preserved to a satisfactory level for study. Natural bitumen deposits in Azerbaijan are concentrated mainly in the Absheron, Shemakha-Gobustan, Nizhne-Kura, Gabala and Guba regions. Several bitumen deposits are known on the Absheron Peninsula: Gyrmaki, Binagady, Khirdalan, Babazanan and Pirallahi Island (Artem). Only two of them - Gyrmaki and Binagady have been studied for paleofauna (Halilov, 2003; Alizadeh, 2024).

The Binagadi bitumen deposit is a unique and the largest Upper Pleistocene cemetery of flora and fauna of global significance. More than 300 species of animals and plants have been identified here (43 species of mammals, 110 species of birds, two species of reptiles, one species of amphibians, 107 species of insects, one species of mollusk, and 22 species of plants). Only the Californian deposit Rancho La Brea in the USA is comparable to it.

The Binagadi bitumen deposit was discovered in 1938 by geologist A.S.Mastan-zade, who, having discovered animal bones in bitumen, handed over the unique paleontological finds to V.V.Bogachev, who was able to appreciate the scientific significance of these finds (Jafarov, 1966). Since then, employees of the Natural History Museum named after G.Zardabi (Natural History Museum), and since 2020, the staff of the paleozoology laboratory of the Institute of Zoology of Azerbaijan have been excavating and studying the fossil remains of flora and fauna there. The location of the remains of flora and fauna coincided with the site of the most ancient oil and bitumen production on the Absheron Peninsula, where wells from which oil products were previously extracted manually have survived to this day.

The study of the Binagadi remains of animals and plants is very important for the reconstruction of the fauna of the Quaternary period of Eurasia and the history of the formation of the modern fauna of Absheron (Eybatov et al., 2015). Macrostructural markers played a leading role in determining the Binagadi osteological material, which have proven their priority significance for determining the taxonomic status of many groups of animals (Burchak-Abramovich, 1966).

The Binagadi burial site of the Quaternary fauna and flora is located on the Absheron Peninsula in the village of Binagadi. On the site of the Binagadi burial site 150-40 thousand years ago there was an ancient freshwater or slightly salty lake, around which there were natural oil spills that were destructive to animals. Oil spills also penetrated into the lake itself, increasing the area of the territory posing a danger to representatives of the local fauna. As a result of many years of work by the employees of the G.Zardabi Natural History Museum: V.V.Bogachev (1938;1939), P.V.Serebrovsky (1945;1948), N.I.Burchak-Abramovich (1949-1962), R.D.Jafarov (1954-1966), D.V.Gadzhiev (1968-1978), T.M.Eybatov (2022), N.E.Novruzov, K.A.Taptygova, T.M.Eybatov (2024) et al. identified many species of insects, birds and mammals by collecting and studying the fossil remains of the Binagadi fauna.

Since 2012, the third stage of research and excavations in the territory of the Binagadi site of fossil Quaternary fauna and flora began. During these works, old and new layers rich in samples of fossil fauna and flora were studied, in the study of which, since 2020, employees of the Paleozoology Laboratory of the Institute of Zoology of Azerbaijan have taken part together with the employees of the Natural History Museum. Currently, all collected fossil plant, entomological and osteological material is stored in the Natural History Museum of Baku.

Material and methods

The fossil bone material of tortoises was collected from 2020 to 2023 on the Absheron Peninsula in the southeastern part of the Binagadi settlement (40.06°N; 47.45°E), where the Binagadi burial site of Quaternary fauna and flora is located. During excavations, the soil mixed with bitumen containing bone and plant remains was removed layer by layer in large blocks and packed into large plastic containers. Subsequently, in the laboratory, the blocks were carefully divided into small parts, sorted and their composition was studied visually and using magnifying devices (magnifying glasses; MBI-6 and Nikon SMZ 1270 binoculars). The found objects were photographed in different projections with a scale ruler superimposed and sorted according to taxonomic classification and morphological nomenclature. When identifying the Binagadi fossil bone remains, the main consideration was given to the structural features of the macro- and microstructure of the bones, taking into account the geochronological scale, paleoclimatic and paleoecological conditions of the Upper Pleistocene (Velichko, 2009). According to the stratigraphic scale, the studied horizons corresponded to the Late Pleistocene (50-70 thousand years ago).

During the primary office processing for sorting and identifying the material, the osteological collections of the Natural History Museum and the Institute of Zoology were used as comparative (reference) samples. Additional literature was also used: anatomical atlases, identifiers and guides to comparative animal anatomy (Gurtovoy et al., 1978; Carroll, 1992; Romer, Parsons, 1992; Kartashev et al., 2004; Dzerzhinsky, 2005; Titov et al., 2011; Zagumenov, 2021). Various methods have been proposed for preparing bone tissue sections (Eidlin, 1971; Smirina, 1989; Castanet, Smirina, 1990; Smirina, 1994; Klevezal, 2007). However, in our case, they did not fully ensure the production of thin sections (up to 0.02-0.04 mm), especially from bones stored in the soil for a long time, the surface layers and compacts of which, having undergone mineralisation processes during their stay in the soil, are easily destroyed and their value may ultimately be lost. At the same time, the degree of mineralisation of bone tissue in the soil is one of the important features determining the time of death of an animal [Rubezhansky, Nedilko, 1972). In addition, the methods proposed by the authors are quite complex in terms of technique and require a significant investment of time. We used a simplified method for preparing thin sections, which allows avoiding the abovementioned disadvantages and preserve the mineralised compacta layer on the bones for measuring its thickness using the eyepiece micrometer of the microscope. The essence of the method for preparing of thin transverse sections of long tubular bones was as follows. From the middle of the diaphyses of long tubular bones, cuts were made with a dental disk cutter and by further grinding on grindstones with different degrees of graininess (M20–M5 μm) transverse sections were prepared suitable for examination under a microscope in transmitted light. Then the thin sections (15–30 μm) were decalcified in a 3% nitric acid solution for 20 minutes, washed with running water and examined under a binocular microscope at 40–60x magnification. The thin sections were stored in glass penicillin vials in pure glycerin. The described method is characterised by the speed of preparative of the preparat, allows obtaining very thin sections, and is especially suitable for bones extracted from the ground. We used it in the study of a large number of bones of varying degrees of preservation.

The material for the comparative study of the skeletons of tortoises from the Binagadi Pleistocene bitumen deposits with the skeletons of modern tortoises were the bone remains of land tortoises obtained in the Holocene layer (at a depth of up to 1.5 m) when digging wells in old suburban summer cottages of the Absheron Peninsula (Shuvalan, Dubendy, Govsan, Turkyan). All linear measurements of the shells and separate bones were carried out with mechanical and digital calipers with an accuracy of 0.1 mm. Measurements of the microstructural elements of bone sections were carried out with an accuracy of 0.01 mm using the eyepiece micrometer of the MBI-6 microscope. To fully describe the incomplete skeletons of tortoises, a reconstruction of the shells was carried out using the typological extrapolation method (Meyen et al., 1988), i.e., completing the missing parts using analog (standard) images taken from zootomic atlases and processed in the graphic editors Paint and Corel Draw. Statistical processing of the primary digital data was carried out in Microsoft Excel and in the PAST 4.03 programs (Hammer et al., 2001).

Results and discussion

Description of the fossil material. Two incomplete skeletons of tortoises and separate bones of these animals were found in bituminous deposits at a depth of about 2.5 m (Fig. 1). These finds were of particular value, since they are extremely rare. The postcranial skeletons of tortoises included bones of the carapace and plastron (cervical, pygal, costal, neural, marginal, suprapygal, epiplastron, entoplastron, hyoplastron, hypoplastron, xiphiplastron), as well as the caracoid, pelvic bones, part of the cervical and caudal vertebrae, long tubular bones of the appendicular skeleton (humerus, radius, ulna and clavicle, femur, tibia) (Fig. 2).

All bones were in satisfactory condition. The presence of ossified epiphyses at the distal ends of long bones indicated that these skeletal specimens belonged to adults.

The material for the comparative study was the bone remains of land tortoises, obtained in the Holocene layer during the digging of wells in old summer cottages on the Absheron Peninsula. The number of claws on the limbs of tortoises extracted from a depth of up to 1.5 m at old summer cottages on the Absheron Peninsula was five on the front limbs and four on the back limbs. This feature clearly indicated that the discovered skeletons belonged to the genus *Testudo*, presumably to the species *Testudo graeca* Linnaeus, 1758 (Fig. 3).

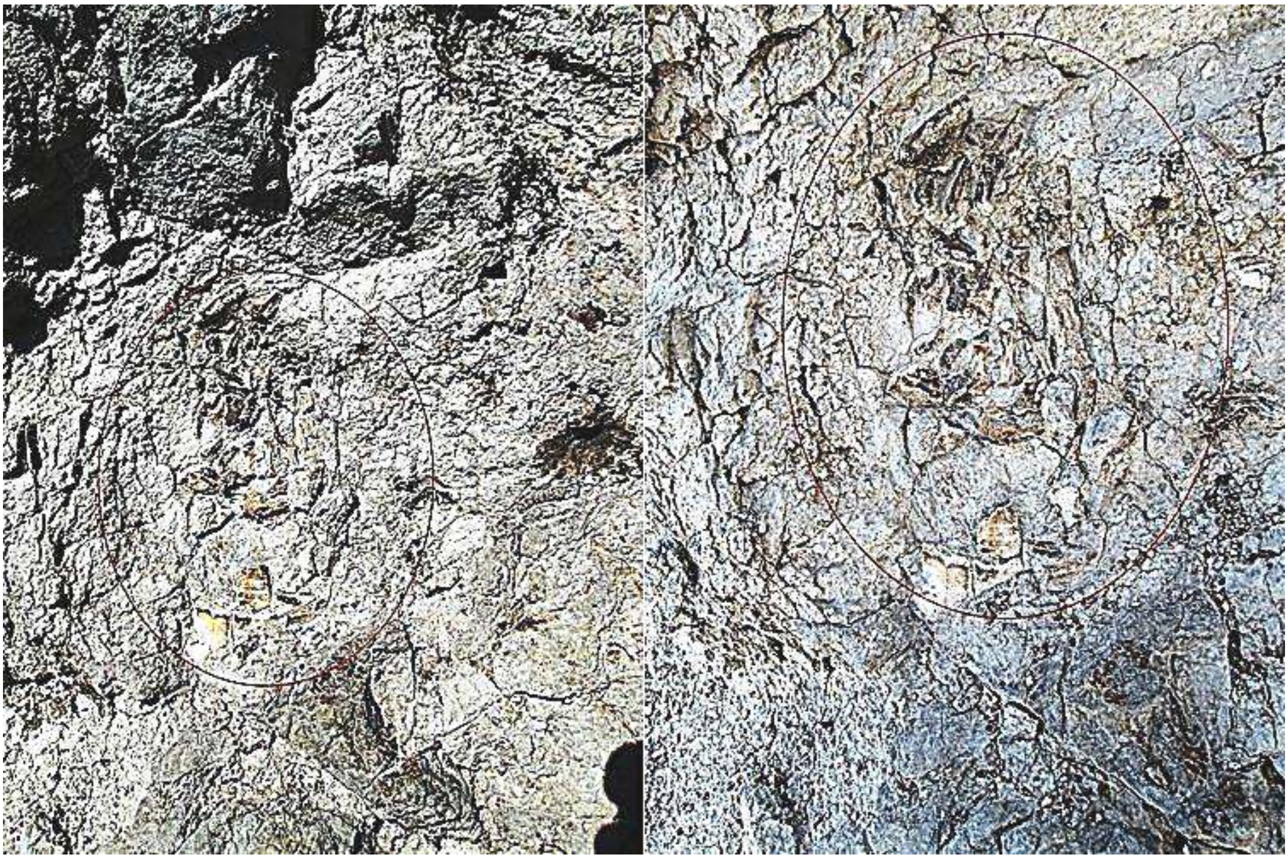

Fig. 1. Location of tortoise bone fragments in bitumen layers in a section of the formation at a depth of 2.5 m (Binagady, 2020)

Fig. 2. Bone material of tortoises recovered during excavations at the Binagadi bitumen deposit

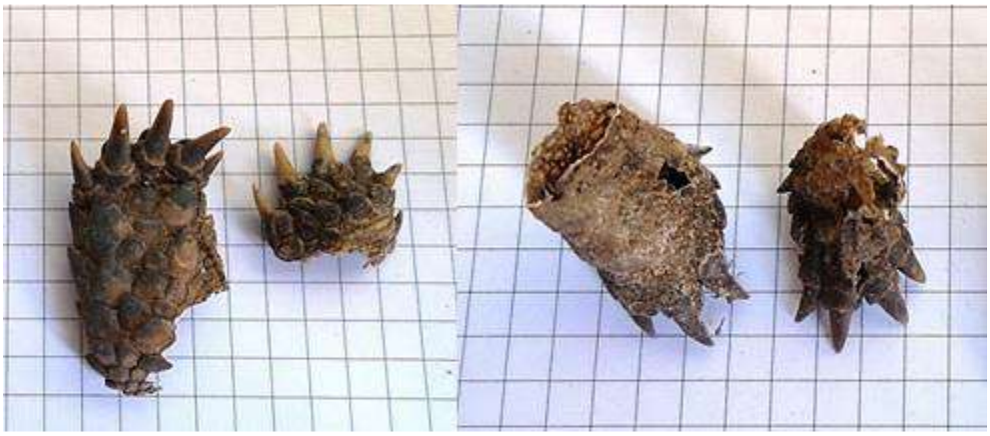

Fig. 3. Fragments of the front five-fingered and back four-fingered limbs of the tortoise *Testudo graeca* extracted from a depth of up to 1.5 m in the summer cottages of the Absheron Peninsula (paws with claws)

Due to the complex and confusing taxonomy of this group, we did not carry out subspecific identification of tortoise bones. Usually, it is carried out based on a set of key morphological features (Vasilyev et al., 2014). However, different geographic populations of *Testudo graeca* are characterised by morphological heterogeneity in a number of diagnostic features, which complicates the unambiguous recognition of subspecies. Therefore, mitochondrial genome markers were used by different researchers for the taxonomy of this group. Different authors distinguish from 4 to 20 subspecies within this taxon (Ananyeva et al., 1998; Fritz et al., 1996; Pieh, Perälä, 2002). Currently, it is customary to consider all populations of this species as a difficult polytypic *T. graeca* complex, consisting of 11 morphological species and six subspecies (Fritz et al., 2007).

Taxonomic position of tortoises of the genus Testudo in the Caucasus. Until the mid-1980s, it was believed that only one subspecies of the spur-thighed tortoise *T. graeca ibera* Pallas, 1814 inhabits the Caucasus (Bannikov et al., 1977). It was later discovered that it is polymorphic and, based on the morphological differences of individual populations, three new subspecies with isolated ranges were described: *T. g. nikolskii* Chkhikvadze et Tuniev, 1986 (the western Caucasus); *T. g. armeniaca* Chkhikvadze, 1991 (Armenia: Araks River valley to the Zangezur Gate) and *T. g. pallasi* Chkhikvadze et Bakradze, 2002 (foothills of Dagestan) (Parham et al., 2006). Later, based on the variability of the mtDNA cytb gene and ISSR markers, it was concluded that the taxon *T. g. pallasi* Chkhikvadze et Bakradze, 2002 is a junior synonym of the subspecies *T. g. armeniaca* Chkhikvadze, 1991. It is believed that the habitat of this subspecies covers the central part of the western coast of the Caspian Sea, the eastern and central parts of the Caucasus region, including the Araks River valley in Armenia and Turkey (Fritz et al., 2007). It has been shown that allopatric populations of tortoises belonging to the subspecies *T. g. armeniaca* live on the territory of Dagestan (Russia), Azerbaijan, Armenia and part of Turkey, and *T. g. ibera*, living in Transcaucasia, represent two separate evolutionary lineages. One of them unites tortoises *T. g. ibera* from Georgia and the Mediterranean (ibera sensu lato), as well as the West Caucasian tortoises *T. g. nikolskii*, and the other – the tortoises *T. g. ibera* from Azerbaijan, *T. g. armeniaca* and *T. g. pallasi*. According to paleontological data, *T. g. nikolskii* and *T. g. pallasi* descended from a single ancestor and could have diverged in the late Pliocene or early Pleistocene (Chkhikvadze, Tuniev, 1986).

Reliable remains of Pliocene land tortoises belonging to the *Testudo graeca* complex are known from a number of locations in the North Caucasus. Fossil remains of *T. g. ibera* tortoises are known in Western Georgia from the Paleolithic and Neolithic, i.e. this tortoise was widespread in Western Transcaucasia much later, at the end of the Holocene (Chkhikvadze, 2010). A special place among the *Testudo graeca* species complex is occupied by *T. g. zarudnyi* tortoises, which are found in a few refugia of the ancient Mediterranean on the Iranian Plateau. They probably survived there since the end of the Miocene or the beginning of the Pliocene, i.e. they are the most ancient forms of land tortoises in the region and belong to the relics of the Eastern Mediterranean (Pereshkolnik, Leontyeva, 2014). In addition, another extinct subspecies *Testudo graeca binagadensis* Khozatsky, 1978 existed in the Pleistocene of Apsheron (Alekperov, Khozatsky, 1977). The form *T. g. binagadensis*, described by L.I.Khozatsky as a special subspecies of the Mediterranean tortoise, differs from the typical modern one in a number of details of the structure of the shell and postcranial skeleton. The most important feature of the Binagadi subspecies is the relatively greater thickness of all bones, which is revealed both while comparing the same parts of the shell of this form with the modern one, and the proportions of the bones of their postcranial skeleton. This is especially noticeable on such bones as the humerus, femur and other tubular bones of the appendicular skeleton. The structural features of the Binagadi land tortoise allow, according to L.I.Khozatsky, to suggest that "it, hiding in shallow burrows, inhabited more or less open biotopes and adhered to shrub and sparse woody vegetation." It should be noted that the descendants of the Binagadi tortoises in eastern Transcaucasia now live in an environment somewhat similar to that which existed during the late Pleistocene (Alekperov, 1978).

Taxonomy of tortoises based on the bones of the postcranial skeleton. The taxonomic position of the tortoises which remains were found in the Binagadi bitumen deposit is presented below:

Class: Reptilia
Order: Testudines
Suborder: Cryptodira
Superfamily: Testudinoidea
Family: Testudinidae Gray, 1825
Genus: *Testudo* Linnaeus, 1758
Testudo graeca Linnaeus, 1758
Testudo graeca ssp. indet.

The tortoises which remains of incomplete skeletons and separate bones were found in the Binagadi bitumen deposit should presumably be considered subspecies of *Testudo graeca*: *Testudo graeca ibera* Pallas, 1814 (lives to this day) or the extinct *Testudo graeca binagadensis* Khozatsky, 1978 (which lived in the Middle and Upper Pleistocene of the Absheron Peninsula) (Alekperov, Khozatsky, 1977) until their molecular (DNA) studies are conducted.

Material: incomplete postcranial skeletons of two specimens of land tortoise, including carapace and plastron bones, two or three posterior cervical vertebrae, forelimb bones (two humeri), hindlimb bones, and anterior caudal vertebrae. It was decided to temporarily retain the third specimen from the bitumen layers of the horizon at the excavation site until further stratigraphic studies could be conducted.

The shells (carapaces and plastrons) were reconstructed using the available fragments was carried out using the typological extrapolation method (Material and Methods) (Fig. 4).

Comparative study of osteological material of tortoises. At the first stage of the work on the study of faunal remains of the Binagadi Pleistocene bitumen deposits, as a result of excavations, bitumen material was removed from a new bone-bearing horizon located at a depth of 1.5-2.5 m, which was a dense accumulation of plant fragments and bones of various animals enclosed in bitumen. According to the stratigraphic scale, the removed material corresponded to the Upper Pleistocene. A detailed study of the material revealed fragments of the skeleton, specimens of the humerus, radius, femur, tibia and other tubular and flat bones belonging to various animals: birds, small mammals, as well as fragments of the shell and long tubular and flat bones of tortoises. In addition to a visual study of the shape of the shell bones and tubular bones of tortoises, a reconstruction of the shells was carried out (Material and Methods), and the main morphometric parameters were measured: 1) maximum bone length; 2) width of the proximal epiphysis; 3) thickness of the articular head; 4) minimum width of the diaphysis; 5) maximum width of the diaphysis; 6) width of the distal epiphysis; 7) anteroposterior diameter of the proximal articular complex; 8) anteroposterior size of the distal articular complex.

At the second stage of the work, a comparative study of the osteological material of tortoises obtained during the digging of wells in the old summer cottages of the Absheron Peninsula was carried out. The material of the comparative study of fossil and modern osteological materials were all the bones of land tortoises of the Binagadi Pleistocene bituminous deposits discovered by us at the first stage of excavations, their reconstructed shells (Fig. 4); bones of the internal skeleton, as well as whole shells of modern land tortoises found during the second stage of excavations in the Holocene layer on the Absheron Peninsula (Shuvalan, Dubendy, Govsan, Turkan) (Fig. 5).

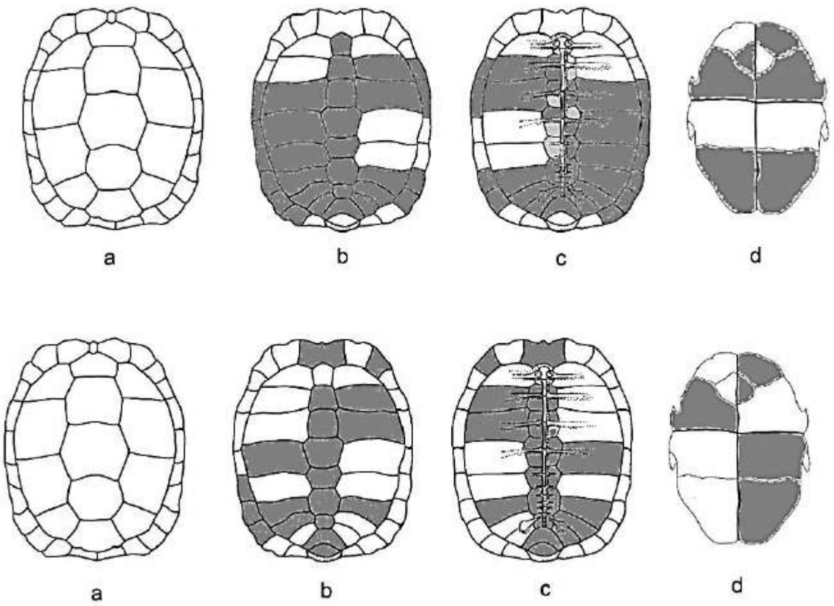

Fig. 4. Schematic figures of reconstructed tortoise shells found in bitumen deposits: a – general view of the nominal bony-horny carapace (dorsal surface); b – bony carapace (dorsal surface); c – bony carapace (ventral surface); d – bony plastron (ventral surface). The present skeletal fragments are highlighted in dark gray in the drawings

Fig. 5. Fragments of the internal skeleton and whole shells of tortoises extracted from the Holocene layer of Absheron on old summer cottages (up to 1.5 m)

The compared osteological material was represented by the humerus, radius, femur, tibia long tubular bones and flat bones of the carapace and plastron (Tables 3-8). The fossil bones were visually and morphologically almost indistinguishable from the bones of modern representatives of the species, except that the metric indicators of the fossil tortoises for separate bones differed slightly from the bone sizes of modern individuals (Tables 4, 6, 8). The compared osteological material was extracted from a depth of up to 1.5 m.

The humerus, radius, femur, tibia, as well as the bones of the carapace and plastron (98 in total) were examined. The following morphometric parameters were used:

1) long bones: maximum bone length; width of the proximal epiphysis; thickness of the articular head; minimum width of the diaphysis; maximum width of the diaphysis; width of the distal epiphysis; anteroposterior diameter of the proximal joint complex; anteroposterior size of the distal joint complex;

2) carapace: bone plates of the vertebral row, including the supracaudal and caudal parts (measurements – length along the midline and greatest width); all costal plates (measurements – proximal width, distal width, maximum length); all marginal plates (measured in the same way as the costal plates);

3) plastron: length and greatest width of all plates along the midline.

In addition, the general dimensions of whole and reconstructed shells were used: length of the carapace (L. car.), greatest width of the carapace (Lt. car.), height of the shell (Al. t), length of the plastron (L. pl.) and its greatest width (S. pl.) (Table 1).

The age of fossil tortoises from the late Pleistocene and Holocene layers was estimated using two methods: an empirically developed formula (L.car(mm)/12) (Novruzov, 2016) and thin sections from the middle of the diaphysis of long tubular bones (Table 2). The number of annual layers found in the thin sections was supplemented by 2-3 layers that underwent resorption during the growth of the animal.

Table 1

Metric parameters of reconstructed late Pleistocene (No. 1, 2) and whole Holocene (No. 3-7) bony shells of tortoises of the genus *Testudo* (mm)

Character/Indice	№ 1	№ 2	№ 3	№ 4	№ 5	№ 6	№ 7
L. car.	123.5	108.0	93.0	96.4	86.4	71.6	73.7
Lt. car.	102.9	85.6	77.3	75.8	73.2	62.7	59.2
Al.t	63.3	59.0	47.5	49.6	47.9	39.9	40.3
L. pl.	110.5	93.7	78.1	85.8	76.4	66.6	63.6
S. pl.	90.5	81.5	69.8	70.5	66.3	57.5	57.0
L. car. / Lt. car.	1.20	1.26	1.20	1.27	1.18	1.14	1.24
L. car. / Al.t	1.95	1.83	1.95	1.94	1.80	1.79	1.82
Lt. car. /Al.t	1.62	1.45	1.62	1.52	1.52	1.57	1.46

Table 2

Data on the age of tortoises obtained empirically (by formula) and from bone sections

№	L. car.(мм)	Age (years)		
		By formula	By thin sections	Average
1	123.5	10.29	6(+3)	9.6
2	108.0	9.00	5(+3)	8.5
3	93.0	7.75	4(+3)	7.3
4	96.4	8.03	4(+3)	7.5
5	86.4	7.20	4(+2)	6.6
6	71.6	5.96	3(+2)	5.4
7	73.7	6.14	3(+2)	5.5

The correlation analysis of morphometric parameters of bone elements of the carapace and plastron and the overall dimensions of the reconstructed shell of land tortoises of the genus *Testudo* found during excavations at the Absheron summer cottages and in the Binagadi bitumens was carried out. Comparison of the correlation coefficients showed that the area of the greatest correlation of bone elements of the shell falls on the middle (dome-shaped) part, where the vertebral (I-IV) and costal (I-V) plates come into contact. At the junction of the first spine and the first marginal plate to the cervical plate, the area of the carapace is characterised by a reliable correlation. Some parameters of the plates are also significantly related: supracaudal, caudal and the last marginal. The largest correlation zone between the studied parameters in the plastron falls on the area of the epiplastral and endoplastral plates.

Table 3

Morphometric indices of long tubular bones of fossil tortoises (No. 1, 2)

№	Material	Dimensions, mm							
		1	2	3	4	5	6	7	8
1	Humerus	32.4	13.1	5.1	4.1	7.8	8.6	11.6	8.5
	Radius	20.4	6.6	5.1	4.2	3.4	5.1	7.2	6.8
	Femur	28.3	9.7	5.8	3.2	5.5	7.9	9.6	7.2
	Tibia	21.1	6.8	5.0	4.3	6.7	4.9	7.1	4.6
2	Humerus	28.5	11.3	6.6	3.9	6.1	8.9	10.9	8.1
	Radius	22.2	7.9	5.1	2.3	4.3	5.8	7.9	5.8
	Femur	23.2	6.7	5.0	2.6	3.6	6.9	8.3	6.1
	Tibia	18.0	5.3	4.1	2.0	3.5	3.7	5.1	3.4

Table 4

Morphometric indices of long tubular bones of the modern tortoise

Material	Dimensions, mm							
	1	2	3	4	5	6	7	8
Humerus	29.1	12.0	4.6	2.3	5.3	6.1	9.0	7.1
Radius	20.8	4.1	2.2	1.4	2.4	2.1	4.0	2.2
Femur	26.1	14.6	4.1	0.23	1.09	7.1	9.5	3.6
Tibia	20.8	5.0	3.8	1.7	3.1	3.4	4.1	3.3

Note (for tables 3, 4): 1 – maximum bone length; 2 – width of proximal epiphysis; 3 – thickness of articular head; 4 – minimum width of diaphysis; 5 – maximum width of diaphysis; 6 – width of distal epiphysis; 7 – anteroposterior diameter of proximal articular complex; 8 – anteroposterior size of distal articular complex.

Table 5

Morphometric indices of carapace bones of fossil tortoises (No. 1, 2)

№	Material	Dimensions, mm			
		1	2	3	4
1	Marginale	15.7	18.2	21.0	22.5
	Neurale	18.7	13.3	19.4	19.1
	Costale	41.2	20.3	44.4	44.2
	Nuchale	31.1	24.3	27.7	27.3
	Pygale	19.4	15.3	21.7	22.3
2	Marginale	18.8	16.3	22.9	21.6
	Neurale	12.1	20.0	19.1	19.6
	Costale	45.1	44.1	46.9	47.1
	Nuchale	24.8	30.1	27.1	27.3
	Pygale	15.1	18.9	21.6	22.1

Table 6

Morphometric indices of carapace bones of modern tortoise

Material	Dimensions, mm			
	1	2	3	4
Marginale	15.1	12.6	15.4	17.7
Neurale	18.7	10.8	18.2	18.6
Costale	33.2	13.4	34.2	36.1
Nuchale	22.0	22.4	20.1	20.2
Pygale	20.6	13.5	22.7	23.1

Table 7

Morphometric indices of plastron bones of fossil tortoises (No. 1, 2)

№	Material	Dimensions, mm			
		1	2	3	4
1	Epiplastron	20.8	20.5	18.2	19.7
	Entoplastron	23.4	23.0	22.2	23.1
	Hyoplastron	36.5	45.6	50.1	47.2
	Hypoplastron	31.5	44.4	52.3	52.8
	Xiphiplastron	33.6	29.8	39.6	36.1
2	Epiplastron	21.0	20.1	15.6	20.2
	Entoplastron	23.1	23.3	22.7	23.0
	Hyoplastron	50.9	36.5	49.6	46.6
	Hypoplastron	45.1	30.8	51.7	52.8
	Xiphiplastron	29.3	34.1	39.4	36.4

Table 8

Morphometric indices of the plastron bones of the modern tortoise

Material	Dimensions, mm			
	1	2	3	4
Epiplastron	19.8	21.0	15.1	23.2
Entoplastron	22.2	23.1	23.1	21.7
Hyoplastron	36.1	45.2	49.8	46.1
Hypoplastron	31.9	44.6	51.8	46.2
Xiphiplastron	33.9	29.7	39.1	36.6

Note (for tables 5–8): 1 – transverse length of bone; 2 – transverse width of bone; 3 – transverse diagonal of bone to the left; 4 – transverse diagonal of bone to the right

Correlation and regression analysis of the measurements of the tortoise bones (Tables 9-11) showed a strong positive correlation between the long tubular bones (r = 0.925-0.998) with the bones of the carapace (r = 0.612-0.996) and the bones of the plastron (r = 0.606-0.998). The exceptions were the nuchale (r = - 0.097) and (r = 0.203) on the carapace and the entoplastron (r = - 0.062) and hyoplastron (r = - 0.245) on the plastron (Tables 10-11).

Microstructure of long tubular bones of tortoises in transverse sections. Thin-sectioned bone crossections (0.03 mm) suitable for examination under a microscope in transmitted light were made after taking the main measurements from the middle of the diaphysis of the long tubular bones. In total, three series of thin sections were made: thin sections from one bone from several sections of the diaphysis and the proximal and distal epiphyses of tortoises; thin sections from the middle of the diaphysis of different bones.

A study of the microstructure of the long tubular bones of tortoises found in the Binagadi bituminous deposits showed that the periosteum is significantly damaged or virtually absent in most bones (Fig. 6). The bones have a clearly defined growth cycle, demonstrating all stages of postembryonic ontogenesis. The narrow dark layers visible in the sections can be considered as winter layers (the period of growth cessation), and the wide transparent layers as spring-summer, i.e., a continuous growth cycle. Closer to the opening of the medullary canal, the process of resorption of bone tissue previously replaced by endosteum occurs, which leads to complete or partial disappearance of the first layers. The layers are better expressed in the diaphyseal part of the long tubular bones.

Table 9

Results of correlation and regression analysis of morphometric parameters of long tubular bones of fossils (No. 1, 2) and modern tortoises

№	Material	*r*	*t*	*p*	*m*
1	Humerus	0.996	28.05	0.0001	10.3%
	Radius	0.983	13.27	0.0001	28.7%
	Femur	0.942	6.85	0.001	127.0%
	Tibia	0.989	16.05	0.0001	22.1%
2	Humerus	0.993	20.60	0.0005	10.6%
	Radius	0.984	13.31	0.0004	40.6%
	Femur	0.925	5.96	0.001	135.1%
	Tibia	0.998	35.87	0.00001	8.7%

Note: r – Pearson correlation coefficient; t – Student's t-test; p – significance of difference; m – average approximation error (%)

Table 10

Results of correlation and regression analysis of morphometric parameters of carapace bones of fossil (No. 1, 2) and modern tortoises

№	Material	*r*	*t*	*p*	*m*
1	Marginale	0.612	1.09	0.471	9.0%
	Neurale	0.988	9.22	0.0687	2.2%
	Costale	0.996	15.17	0.0403	2.1%
	Nuchale	-0.097	0.138	0.9126	4.9%
	Pygale	0.988	8.94	0.0708	2.6%
2	Marginale	0.776	1.74	0.3318	6.6%
	Neurale	-0.439	0.691	0.6149	18.5%
	Costale	0.836	2.15	0.2766	18.4%
	Nuchale	0.203	0.294	0.8181	4.8%
	Pygale	0.350	0.528	0.6908	18.6%

Table 11

Results of correlation and regression analysis of morphometric parameters of plastron bones of fossil (No. 1, 2) and modern tortoises

№	Material	*r*	*t*	*p*	*m*
1	Epiplastron	0.672	1.28	0.42145	9.1%
	Entoplastron	-0.630	1.14	0.45650	1.6%
	Hyoplastron	0.998	22.54	0.02821	0.6%
	Hypoplastron	0.947	4.18	0.14924	4.6%
	Xiphiplastron	0.994	13.33	0.04763	1.0%
2	Epiplastron	0.851	2.29	0.26149	5.9%
	Entoplastron	-0.062	0.08	0.94387	2.5%
	Hyoplastron	-0.245	0.35	0.78173	9.8%
	Hypoplastron	0.234	0.34	0.79070	15.2%
	Xiphiplastron	0.606	1.07	0.47597	7.3%

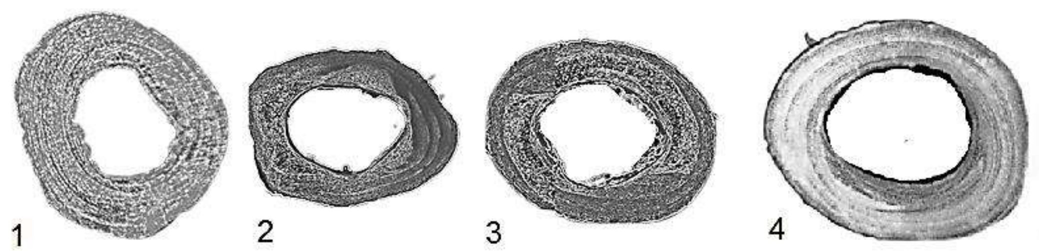

Fig. 6. Microstructure of long tubular bones of tortoises from Binagadi bitumens: 1 – femur; 2 – tibia; 3, 4 – humerus (×64)

Microstructural elements were also noted in the thin sections of long tubular bones: vascular and Haversian canals, osteons, lacunae, insertions of coarse fibrous bone tissue in the middle and endosteal zones. The thin sections of the bones of the skeletons of identified tortoises (Pleistocene horizon) were compared with the thin sections made from the bones of identified tortoises (Holocene horizon) by the number of microstructural elements. To count the microstructural elements (lacunae, osteons, Haversian and vascular canals), we used the method of Golubovich (1991) with some modification. In the space of the bone thin section, using the eyepiece of a microscope with an inserted lens with a grid (grid square = 10,000 sq. μm) at 64x magnification, the microstructural elements were counted (Table 12).

Correlation and regression analysis of the number of microstructural elements in the bone sections of the compared tortoises showed a strong positive correlation only for the metatarsus (r = 0.987), a moderate negative correlation for the fibula (r = −0.562), and a weak correlation for the radius and femur (r = 0.200 and 0.261, respectively). The number of adhesion lines in the sections made from one bone and from all the studied long tubular bones taken from one skeletal sample was the same. A study of a series of sections made from the middle of the diaphysis of the long tubular bones of turtles showed that the process of endosteal bone tissue resorption, which caused the complete or partial disappearance of the first two or three layers in the growth zone, makes it difficult to determine the exact stage of reptile ontogenesis. Comparative study of the microstructure of long tubular bones of tortoises discovered during the second stage of excavations in the Binagadi bituminous deposits and tortoises found in the Holocene layer showed: 1) all bones have a clearly expressed growth cycle; 2) narrow dark layers visible in thin sections should be considered winter layers (growth cessation period); 3) wide light (transparent) layers should be considered spring-summer continuous growth cycle; 4) the number of gluing lines in thin sections of all studied long tubular bones taken from one sample is the same; 5) a clear pattern of resorption of endosteal bone tissue and complete or partial destruction of the first two or three layers of the growth zone is observed. The process of resorption of bone tissue previously replaced by endosteum leading to complete or partial disappearance of the first layers in both cases makes it difficult to determine the exact stage of ontogenesis of these reptiles. Consequently, in each specific case of determining the duration of ontogenesis, 2-3 layers that underwent resorption during the growth of the animal should be added to the number of annual layers found in the sections.

Conclusion

The study of bitumen material extracted from the Binagadi burial site of the Quaternary fauna and flora allowed identifing two incomplete skeletons and separate bones of land tortoises of the genus *Testudo*. The study of the bone skeleton of tortoises found in the Binagadi burial site in the Late Pleistocene layer (Late Dryas) in comparison with the bones of tortoises found on Absheron in the Holocene layer (Middle Subatlantic period) leads to the assumption that the climate at the end of the Pleistocene was cooler and more humid than in the Holocene. This is indicated by some changes in the morphology of the bony shell of the Mediterranean tortoise: a decrease in the indices of the ratio of the length of the shell to its width and height (L.car/L.t.car.; L.car/Al.t.); the width of the shell to its height (L.t.car/Al.t.); changes in the size proportions of some bone plates of the carapace and plastron.

Table 12

Number of microstructural elements in thin sections of different bones of identified and already identified tortoises (per 10,000 sq. μm)

Microstructural element	Name of the bone													
	Humerus		Ulna		Radius		Femur		Fibula		Tibia		Metatarsus	
	I	II	I	II	I	II	I	II	I	II	I	II	I	II
Osteon	–	–	–	–	+	+	+	+	+	+++	–	–	++	++
Haversian Canal	+	+	+	+	+	+	+	++	+	++	+	+	++	+++
Vascular canal	+	–	+	+	+	+	+	+	+	+	+	–	+	+
Lacuna	+	+	+	–	+	+	+	+	+	+	–	–	+	+

Note: I – material being identified; II – already identified material. Number of microstructural elements: (+++) – 20-30; (++) – 10-20; (+) – less than 10; (−) – absent

All these changes, confirmed by correlation-regression analysis, presumably reflect some intensification of adaptation processes associated with significant climate changes in Azerbaijan in general and in its eastern part in particular during the specified geological period. They are expressed in a greater need of the species for protective and thermoregulatory-compensatory burrowing in the ground during periods of night pause, winter hibernation and summer aestivation, ensuring its successful survival in the resulting arid climatic conditions. In the microstructure of tortoise bones removed from bituminous layers, resorption processes are more pronounced and affect 3-4 adhesion lines and the endosteum region, whereas in the comparative material of Holocene tortoises, resorption affects 1-2, less often 3 adhesion lines, without affecting the endosteum. The ontogenetic age of the fossil remains of tortoises was determined to be within 10-12 years.

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К ИЗУЧЕНИЮ ИСКОПАЕМЫХ КОСТНЫХ ОСТАНКОВ ЧЕРЕПАХ (TESTUDINES, TESTUDINIDAE) ИЗ БИНАГАДИНСКОГО БИТУМНОГО МЕСТОРОЖДЕНИЯ (АЗЕРБАЙДЖАН)

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Резюме. Объекты исследования – ископаемые костные останки черепах, извлеченные из грунта в процессе раскопок в Бинагадинском битумном месторождении и старых дачных участках Абшерона. Материал собран и обработан за период с 2020 по 2023 гг. в процессе экспедиционных и лабораторных исследований. Собранный остеологический материал хранится в Естественноисторическом музее города Баку и в лаборатории палеозоологии Института зоологии Азербайджана. Изучение материала позволило выявить большое количество костных фрагментов. Установлена родовая и видовая принадлежность обнаруженных костных фрагментов черепах. Изучение костных останков скелетов черепах найденных в Бинагадинском битумном месторождении в позднеплейстоценовом слое (поздний дриас) и сравнение его с костями черепах найденных на Абшероне в голоценовом слое (средний субатлантический период) позволило сделать предположение, что климат в конце плейстоцена был более прохладным и влажным, чем в голоцене. На это указывали некоторые изменения в морфологии костного панциря черепах: уменьшение индексов соотношения длины панциря к его ширине и высоте, а также ширины панциря к его высоте; изменения размерных пропорций некоторых костных пластинок карапакса и пластрона. Все эти изменения, предположительно, отражают некоторое усиление адаптационных процессов, связанных со значительными изменениями климата в Азербайджане вообще и в его восточной части в частности за указанный геологический период (более 0.012 млн. лет назад). Совокупность идентифицированных останков бинагадинского тафоценоза весьма важна для реконструкции фауны четвертичного периода Евразии и истории формирования современной фауны Абшерона. Обнаруженные микро- и макроструктурные маркеры Бинагадинского остеологического материала могут иметь приоритетную значимость для установления таксономического статуса будущих ископаемых находок. Цикличность роста костной ткани в микроструктуре длинных трубчатых костей черепах позднеплейстоценового слоя позволяет корректировать данные о климатических условиях четвертичного периода.

Ключевые слова*: битумное месторождение, ископаемые останки, черепахи, морфология, кости, микроструктура*

AZƏRBAYCANIN BİNƏQƏDİ QIR YATAĞINDA TAPILAN TISBAĞALARIN (TESTUDİNES, TESTUDINIDAE) FOSİL SÜMÜK QALIQLARININ ÖYRƏNİLMƏSİNƏ DAYİR

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Xülasə. Tədqiqat obyektləri Binəqədi qır yatağında və Abşeron baqlarında su quyularının qazıntı zamanı torpaqdan çıxarılan tısbağaların sümük qalıqlarıdır. Material 2020-2023-cü illərdə toplanmışdır. Toplanmış osteoloji material Bakı şəhərinin Təbiət Tarixi Muzeyində və Azərbaycan Zoologiya İnstitutunun paleozoologiya laboratoriyasında saxlanılır. Tədqiqat nəticəsində çoxlu sayda sümük fraqmentləri aşkar edilmişdir. Tısbağaların aşkar edilmiş sümük parçalarının cinsi və növ mənsubiyyəti müəyyən edilmişdir. Binəqədi bitum yatağında gec Pleistosen qatında (gec drias) tapılan tısbağaların skeletlərinin sümük qalıqlarının öyrənilməsi və Abşeronda Holosen qatında (orta subatlantik dövr) tapılan tısbağaların sümükləri ilə müqayisə edilməsi pleistosenin sonundakı iqlimin holosenə nisbətən daha sərin və rütubətli olduğunu irəli sürdü. Bunu tısbağaların sümük qabığının morfologiyasındakı bəzi dəyişikliklər göstərdi: qabığın uzunluğunun eninə və hündürlüyünə, həmçinin qabığın genişliyinin hündürlüyünə nisbətinin indekslərində azalma; bəzi karapaks və plastron sümük lövhələrinin ölçü nisbətlərində dəyişikliklər. Bütün bu dəyişikliklər, ehtimal ki, ümumilikdə Azərbaycanda və onun şərq hissəsində, xüsusən də göstərilən geoloji dövrdə (0.012 milyon ildən əvvəl) iqlimdəki əhəmiyyətli dəyişikliklərlə əlaqəli uyğunlaşma proseslərinin bir qədər güclənməsini əks etdirir. Tısbağa skeletləri haqqında əldə edilən morfoloji məlumatlar Testudo cinsinin mürəkkəb politipik kompleksinin fərqləndirilməsində müqayisəli morfoloji tədqiqatlarda, filogenetik və taksonomik növ fərqlərində istifadə edilə bilər. Binəqədi tafosenozunun müəyyən edilmiş qalıqlarının məcmusu Avrasiyanın dördüncü dövrünün faunasının yenidən qurulması və Abşeronun müasir faunasının yaranma tarixi üçün çox vacibdir. Binəqədi osteoloji materialının aşkar edilmiş mikro və makrostruktur markerləri gələcək fosil tapıntılarının taksonomik statusunun yaradılması üçün prioritet ola bilər. Gec Pleistosen təbəqəsinin tısbağalarının uzun borulu sümüklərinin mikrostrukturunda sümük toxumasının dövri böyüməsi dördüncü dövrün iqlim şəraiti haqqında məlumatları tənzimləməyə imkan verir.

Açar sözlər: *bitum yatağı, fosil qalıqları, tısbağalar, morfologiya, sümüklər, mikrostrukturu*