



AGE DETERMINATION OF THE WILDCAT (*FELIS SILVESTRIS*): A CASE STUDY OF A SAMPLE FROM THE NORTH-WESTERN BLACK SEA REGION (UKRAINE)

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Abstract

In recent decades, the wildcat (*Felis silvestris*) population has been increasing along the eastern edge of its range in Ukraine, associated with the recolonisation of territories previously inhabited by the species. An essential aspect of analysing this process is studying population dynamics, including sex and age structures. Knowing the individual age of animals is a crucial prerequisite for investigating many aspects of their life cycle and population dynamics. This information is particularly important for *Felis silvestris*, a rare species included in international and national lists of protected animals, for the conservation and management of its populations. This necessitates the verification of age determination methods in specific *Felis silvestris* populations, particularly in the south-western regions of Ukraine. To determine the age of wildcats, we employed both traditional, non-invasive methods—such as analysing suture obliteration and dental morphometric characters—and methods requiring partial tooth destruction, such as studying changes in dentin volume and cementum in the root of the canines. Cementochronology was found to be the only method allowing the wildcat population to be divided into seven year classes. Cement deposition primarily occurs at the apical tip of the canine root, allowing the use of only the lower third of the tooth for analysis. This is particularly important when working with collection materials of rare species. In the first year of life (0+), dentin fills no more than 20% of the pulp cavity width. In subsequent year classes, dentin deposition occupies at least 70% of the pulp cavity volume, enabling a clear division of the wildcat population into two groups: young (0+) and older individuals. The distance between the enamel at the neck of the canine and the edge of its alveolus is an effective tool for distinguishing young and adult individuals. The analysis of alveolar recession relative to the neck enamel can be considered a promising method for determining age groups in wildcats, including both deceased and living individuals. The use of basicranial sutures of the skull was found to be less informative: the sutures at the skull base ossify late, while the obliteration between the maxilla and premaxilla in the nasal area is only partially expressed.

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Визначення віку кішки лісової (*Felis silvestris*): дослідження вибірки з Північно-західного Причорномор'я (Україна)

Юрій Олійник

Резюме. Останніми десятиліттями спостерігається зростання чисельності лісового kota на східному кордоні його ареалу в Україні, що пов'язано з реколонізацією раніше окупованих ним територій. Важливим елементом аналізу цього процесу є вивчення динаміки популяції, включаючи статево-вікову структуру. Знання індивідуального віку є важливою передумовою для вивчення багатьох аспектів життєвого циклу тварини та її популяції. Така інформація може мати вирішальне значення, коли це стосується *Felis silvestris*, рідкісного виду, включеного до міжнародних та національних списків тварин, які знаходяться під охороною, про збереження та управління популяціями цього виду. Разом з тим, існує потреба у верифікації методів визначення віку у *Felis silvestris* в конкретних популяціях, зокрема у південно-західних регіонах України. Для визначення віку ми використовували як традиційні, неруйнівні методи — аналіз облітерації швів, морфометричних характеристик зубів, так і ті, що вимагають руйнування частини зуба — аналіз змін обсягу дентину і цементу в кореневій частині іклів. Встановлено, що цементохронологія є єдиним методом, що дозволяє розділити популяцію лісового kota на 7 річних класів. Відкладення цементу в основному на верхівці кореня іклів дозволяє використовувати для аналізу тільки нижню третину зуба, що важливо при роботі з колекційним матеріалом особливо рідкісних видів. У перший рік життя (0+) дентин заповнює не більше ніж на 20% ширини порожнини пульпи. В інших річних класах відкладення дентину в порожнині пульпи кореня ікла займають не менше 70% його об'єму. Це дозволяє розділити популяцію лісового kota на дві вікові групи: молоді (0+) і більш дорослі особини. Відстань між межею емалі в області шиї ікла і краєм його альвеоли є ефективним інструментом для поділу популяції лісового kota на групу молодих і дорослих особин. Аналіз рецесії альвеолярного краю емалі шийки зуба можна розглядати як перспективний метод визначення вікових груп не тільки мертвих, але і живих тварин у лісового kota. Використання швів базікраніальної частини черепа є малоінформативним. Шви основи черепа заживають пізно, а в носовій області облітерація між верхньощелепною і передщелепною кістками виражена лише частково.

Ключові слова: *Felis silvestris*, цементохронологія, ширина порожнини пульпи, шви черепа, морфометрія іклів.

Introduction

In recent decades, an increase in the presence of the wildcat (*Felis silvestris*) has been observed along the eastern edge of its range, in the forest-steppe and steppe regions of Ukraine. This trend is attributed to the recolonisation of territories previously inhabited by the species [Oleinik, 2020; Zagorodniuk *et al.* 2014]. An essential component of analysing this process involves studying the population dynamics of wildcats in these regions, particularly their sex and age structures. While information about an animal's sex is typically obtained during capture or collection, age estimation requires specialised methods. For felids, as with many other carnivorous mammals, age determination has traditionally relied on non-invasive methods. These approaches include analysing elements of the postcranial skeleton [Schmidt *et al.* 2019], monitoring changes in skull size, examining suture obliteration [Anderson & Wiig 1984; Schmidly & Read 1986; Garcia-Perea 1996], and assessing dental characters such as eruption patterns [Schauenberg 1969; Condé & Schauenberg 1978; Smuts *et al.* 1978] and tooth morphology changes [Smuts *et al.* 1978; Stander 1997].

More precise individual age assessments can be achieved by analysing structural changes in the root components of teeth, such as the width of the pulp cavity in canines [Meachen-Samuels & Binder 2010; Stefen 2015] and cementum deposition [Garcia-Perea & Baquero 1999; Nakanishi *et al.* 2009]. However, these methods simultaneously lead to tooth destruction.

Among the latest methods, cementochronology, which analyses annual cementum layers in the roots of teeth, despite a few unresolved issues [Perrone *et al.* 2022], is a widely applied method in carnivorans and is generally recognised as an accurate way to determine age in years [Klevezal &

Kleinenberg 1969; Morris 1972; Grue & Jensen 1979; Johnson *et al.* 1981]. This method has been successfully used in wild species, such as the wildcat (*Felis silvestris*) [Garcia-Perea & Baquero 1999] and *Lynx* [Crowe 1972; Kvam 1983; Zapata *et al.* 1997], as well as domestic cats [Grue & Jensen 1979]. A modification of this method, which simplifies the preparation of the tooth for subsequent analysis and reduces the time required to count annual cementum layers [Roulichová & Anděra 2007], makes it possible to analyse significantly larger volumes of material from different populations of the same species, causing minimal damage compared to the classic version. This is especially important in relation to specimens of rare and small species stored in museum collections.

In some cases, age determination based on tooth structure (e.g. *in vivo*) is performed using complex instrumental methods such as tomography and radiography [Park *et al.* 2014; White & Belant 2016; Schmidt *et al.* 2019].

Combinations of all these methods, as shown by P. Fleming *et al.* [2021] in the example of domestic cats, are applicable for determining the age of deceased cats. At the same time, the degree of informativeness can be related to several factors, including competition, dietary variability, diet characteristics, and growth patterns. In individuals from populations occupying territories with less continental climates, the clarity and uniformity of growth layers in recording structures are less pronounced than in areas with sharply continental climates [Klevežal 1996]. To increase the reliability of age determination methods, it is considered necessary to adapt verification protocols, sometimes even for different populations of the same species [Grue & Jensen 1979].

Thus, the verification of age determination methods of *Felis silvestris* in the south-western regions of Ukraine, primarily focusing on simple, fast methods with minimal destructive impact on the examined specimens, was the main goal of our study.

Materials and Methods

The material for the study included samples from 48 cat specimens collected between 2001 and 2018 in the territory of Odesa Oblast, Ukraine. Among these, 38 specimens were identified as wildcats (*Felis silvestris*) and 10 specimens were classified as *Felis catus* (domestic cats), collected from settlements and adjacent territories. Species identification was based on morphological traits (body size, fur colouration) [Ragni & Possenti 1996; Kitchener *et al.* 2005] and anatomical characteristics (cranial volume, morphometric parameters) [Schauenberg 1969; Daniels *et al.* 1998; Kruger *et al.* 2009], which are considered sufficient to distinguish between domestic and wild cats both in captivity and in the field [Devillard *et al.* 2014]. According to the index expressing the ratio of total skull length to skull volume, specimens with values equal to or less than 2.75 were classified as *Felis silvestris*, while skulls with values greater than 3.0 were classified as *F. catus*. Individuals displaying signs of ‘hybridisation’ (4 individuals, with indices in the range of 2.75–3.0) were included in the wildcat group. Taking into account that morphologically ‘pure’ wildcats can, with a certain degree of probability, be genetically classified as ‘hybrids’ with the domestic form and vice versa [Oliveira *et al.* 2008; Lecis *et al.* 2006], and the impossibility of conducting genetic analysis of the available material, the samples were divided into only two groups: wildcat and domestic cats.

The acquisition of material was non-systematic, and in approximately 30% of cases the collecting site was identified only at the level of the region (e.g. Odesa Oblast) without precise details regarding the date of the animal’s death. Sex was determined for 26 specimens of *Felis silvestris*.

In the literature, three sutures are commonly noted as criteria for age-related changes in carnivorans based on the degree of obliteration: BB—sutura basioccipito-basisphenoidalis; PB—sutura presphenoid-basisphenoidalis; and MP—sutura maxillo-premaxillaris (Fig. 1a) [Harris 1978; Roulichová & Anděra 2007]. The sequence and extent of suture fusion were analysed in 30 wildcat skulls.

The process of suture obliteration was divided, following A. Goswami *et al.* [2013], into four categories: Open—The suture remains open with little or no contact between the bones along most of its length (Fig. 1b). Partially closed—Contact between bones occurs in some parts of the suture (Fig. 1c). Closed—The suture is completely fused (Fig. 1d). Closed and nearly invisible—The suture is fully overgrown and barely detectable (Fig. 1f).

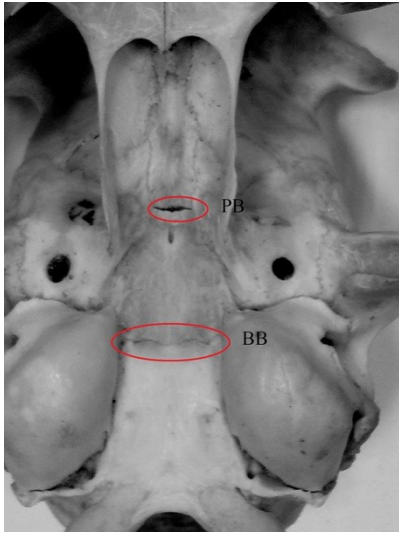
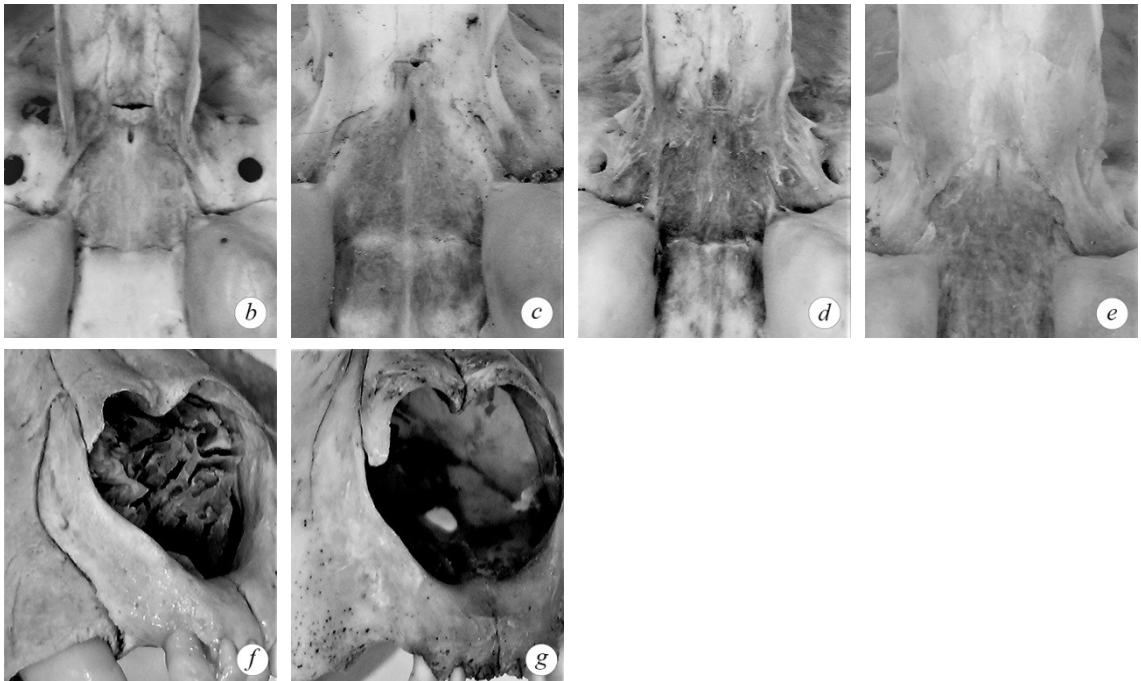


Fig. 1. Overgrowth of sutures in the wildcat *Felis sylvestris*: (a) BB, sutura basioccipito-basisphenoidalis; PB, sutura presphenoid-basisphenoidalis; (b) suture open; (c) partial overgrowth; (d) closed; (e) suture invisible; (f) sutura maxillo-premaxillaris open; (g) sutura maxillo-premaxillaris partial overgrowth.

Рис. 1. Заростання швів у лісової кішки *Felis sylvestris*: (a) BB, sutura basioccipito-basisphenoidalis; PB, sutura presphenoid-basisphenoidalis; (b) шов відкритий; (c) часткове заростання; (d) закритий; (e) шов невидимий; (f) sutura maxillo-premaxillaris відкритий; (g) sutura maxillo-premaxillaris — часткове заростання.



Throughout life, enamel wear occurs due to the influence of food, while cementum accumulates in the root part of the tooth. This process is accompanied with changes in the morphological parameters of teeth. In this study, isolated canines extracted from the right upper jaw were analysed. Each tooth was measured in two planes: mesio-distal (thickness) and buccal-lingual (width). Additionally, the total tooth length was measured (Fig. 2). The selected parameters included: total tooth height, width and thickness of the tooth neck in the crown area (measured along the enamel), width and thickness of the root under the enamel, maximum width and thickness of the root.

All measurements were taken using callipers with an accuracy of 0.1 mm.

The area of the open canine canal at the apical part of the root was calculated as the area of an ellipse using the formula:

$$S = (0.5 \times D) \times (0.5 \times d) \times \pi,$$

where 'D' is the larger diameter of the canal, and 'd' is the smaller diameter of the canal.

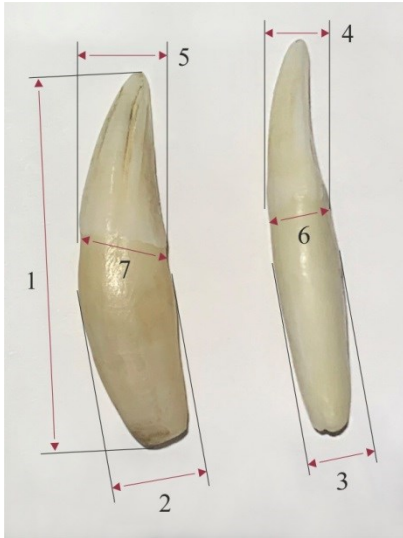


Fig. 2. Morphometric characteristics of the canine: 1, total tooth height; 2, maximum root thickness; 3, greatest root width; 4, tooth neck width along the enamel; 5, tooth neck thickness along the enamel; 6, tooth neck width below the enamel; 7, tooth neck thickness below the enamel.

Рис. 2. Морфометричні характеристики ікла: 1, загальна висота (довжина) зуба; 2, максимальна товщина кореня; 3, найбільша ширина зуба; 4, ширина шийки зуба уздовж емалі; 5, товщина шийки зуба по емалі; 6, ширина шийки зуба під емаллю; 7, товщина шийки під емаллю.

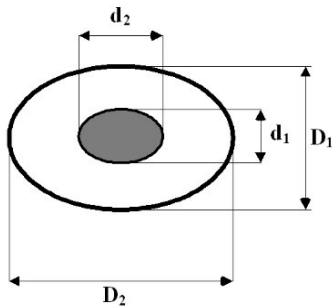


Fig. 3. Measurement scheme of the tooth root and its canal:

d_1 and D_1 —respectively, the smallest diameter of the canine canal and the root of the tooth; d_2 and D_2 —respectively, the largest diameter of the canine canal and the root of the tooth.

Рис. 3. Схема вимірювання кореня зуба і його каналу:

d_1 і D_1 — відповідно найменший діаметр каналу ікла і кореня зуба, d_2 і D_2 — відповідно найбільший діаметр каналу ікла і кореня зуба.

Before extracting the tooth from the alveolus, the distance from the edge of the enamel at the tooth neck to the edge of the alveolus socket along the mesial and distal edges of the canine was measured. The root outcrop length was determined from photographs using PhotoM software (version 1.21).

Age estimation was conducted based on the rate of dentin overgrowth in the canine pulp cavity ('canal') using two methods: by V. Smirnov [1960] and J. Meachen-Samuels & W. Binder [2010].

In the first method [Smirnov 1960], a formula utilising four measurements at the widest part of the canine root was applied (Fig. 3):

$$I = s_1/s_2 \times 100\% = (d_1 \times d_2) / (D_1 \times D_2) \times 100\%,$$

where d_1 and d_2 are the smallest diameters of the canine canal, and D_1 and D_2 are the largest diameters of the tooth root; s_1 and s_2 represent the cross-sectional areas of the tooth cavity and root, respectively.

In the second method [Meachen-Samuels & Binder 2010], a formula utilising four measurements and the percentage was calculated as:

$$\% I = 100\% \times ((D_2 - d_2)/D_2),$$

where d_2 and D_2 denote the largest diameters of the canine canal and tooth root, respectively.

In 30 wildcat specimens, cementum deposits were analysed at the root of permanent canines with overgrown apical sections. Each annual layer consisted of two zones, referred to as light ('summer') and dark ('winter') lines.

Since the dark accessory line forms during periods of stunted growth (typically in winter) [Grue & Jensen 1979], animals whose canines displayed a light ('summer') line at the periphery during the autumn–winter period were designated as 'n+' (incomplete year), where n represents the number of

dark ('winter') lines formed. These individuals, along with specimens exhibiting either a forming or fully formed cementum line at the root periphery ('n') (complete year), were assigned to the same year class. According to R. Garcia-Perea & R. Baquero [1999], discontinuous lines were observed between the first continuous line and the dentin–cementum boundary.

Cementum layers were determined on upper canine root sections prepared in accordance with the recommendations of J. Roulichová & M. Anděra [2007].

The tooth, extracted from the skull, was affixed to a wooden cube with PVA glue and successively ground on abrasive stones with progressively finer grain sizes (80, 180, 320, and 600). The upper canine root sections were examined at 16x magnification under reflected light using an MBS-9 binocular microscope and an E-Trek digital camera (DCM 900, 9-megapixel resolution).

Results

The examined sutures in the wildcat are overgrown in different sequences. Most often, the sutura basioccipito-basisphenoidalis begins to undergo obliteration first (Table 1; Fig. 1b). By the age of 3–4 years, most individuals exhibit a completely obliterated suture (Fig. 1d). Subsequently, after 4 years of age, the proportion of wildcats in which the suture is not only closed but also completely invisible increases (Fig. 1f).

The obliteration of the sutura presphenoid-basisphenoidalis begins later, around 2–3 years of age, and demonstrates significant chronological variability. More than half of the individuals retain an open or partially open suture between 2 and 7 years of age (Table 1). Complete overgrowth typically occurs starting from the age of three.

The sutura maxillo-premaxillaris is the least prone to obliteration. Partial overgrowth of this suture was observed in approximately one-third of individuals starting at an age of 4 years (Table 1; Fig. 1g). However, complete overgrowth was not noted in any of the studied age groups. In domestic cats, the pattern of suture overgrowth follows a similar trend.

After a tooth erupts, its root canal remains open apically for a certain period as the tooth continues to grow in length. The presence of an open apical canal in the canine root is a definitive indicator of an animal in its first year of life. Among the examined canines, five exhibited an unclosed apical canal. The canal shape was ellipsoidal, with the largest diameters ranging from 2.6 mm to 1.8 mm. The canal area varied between 1.23 mm² and 3.47 mm². The percentage index of pulp overgrowth (% I), calculated according to J. Meachen-Samuels & W. Binder [2010], ranged from 30.8% to 5.5%. This variation indicates the prolonged and highly variable rate of canine root overgrowth in wildcats (Table 2). By the end of January, the apical canal of the wildcat canine remains unclosed. In individuals aged two years and older, the canal in the apical part is completely closed.

With the closure of the apical foramen of the canine, the formation of the tooth is completed. Subsequently, changes in its parameters are associated with the nature of nutrition and the growth of cement in the root part. In young (0+) wildcats, the teeth are sharp.

Table 1. Degree of obliteration of wildcat cranial sutures (n = 30)

Таблиця 1. Ступінь облітерації швів черепа кішки лісової (n = 30)

Age	Suture BB				Suture PB				Suture MP	
	Opened	Partly opened	Closed	Closed invisible	Opened	Partially open	Closed	Closed, invisible	Opened	Partly opened
0+	1	2			3				3	
3		1	1			1	1		2	
4	1	2	2	2	2	2	2	1	5	2
5			3	1	2	2			4	
6			2	6	3	3	2	1	5	4
7		1		3		1	1	2		2
8				1				1	1	

Table 2. Area of the open apical opening of wildcat canines

Таблиця 2. Площа відкритого апікального отвору в іклах лісової кішки

№	Date	Canine root area in the apical region (mm ²)	Canine apical foramen area (mm ²)	Coefficient % I [Meachen-Samuels & Binder 2010] (%)
63	autumn	4.08	1.91	30.8
37	30.10	2.47	1.23	17.8
15	28.01	3.34	2.20	20.0
10	??	4.21	3.47	5.5

Among other year groups, three-quarters had slightly rounded teeth, and 5 specimens (aged 4–7 years) displayed noticeable signs of wear (ranging from 0.5 to 1.5 mm).

As a result, no direct correlation between the selected parameters of the canine and the specific age of the animal was established. The morphological characters of canines in the studied sample of wildcat teeth indicate only a tendency for their values to decrease over time (Fig. 4, 5). However, significant variability in these parameters exists within each year class (see: Fig. 5). The statistical significance of this trend, tested using Spearman's correlation, was confirmed only for two parameters related to the tooth neck area: thickness under the enamel and thickness along the enamel ($r = -0.82$; $p < 0.023$).

With age, as cement accumulates, the root appears to shift out of the alveolus. This is reflected in the increasing distance between the edge of the alveolus and the neck of the tooth (the visible part of the root). This feature, measured along the mesial and distal edges of the canine, shows a clear tendency to increase. Specifically, the parameter measuring the distance from the enamel edge to the alveolus, assessed along the distal surface of the canine, demonstrates a statistically significant correlation with age ($r = -0.58$; $p < 0.001$).

This allows to for the differentiation of wildcat yearlings in the autumn–winter period, when this parameter does not exceed 1.2 mm (Fig. 6). Due to the high individual variability in adults, this character cannot be used to discriminate older year classes.

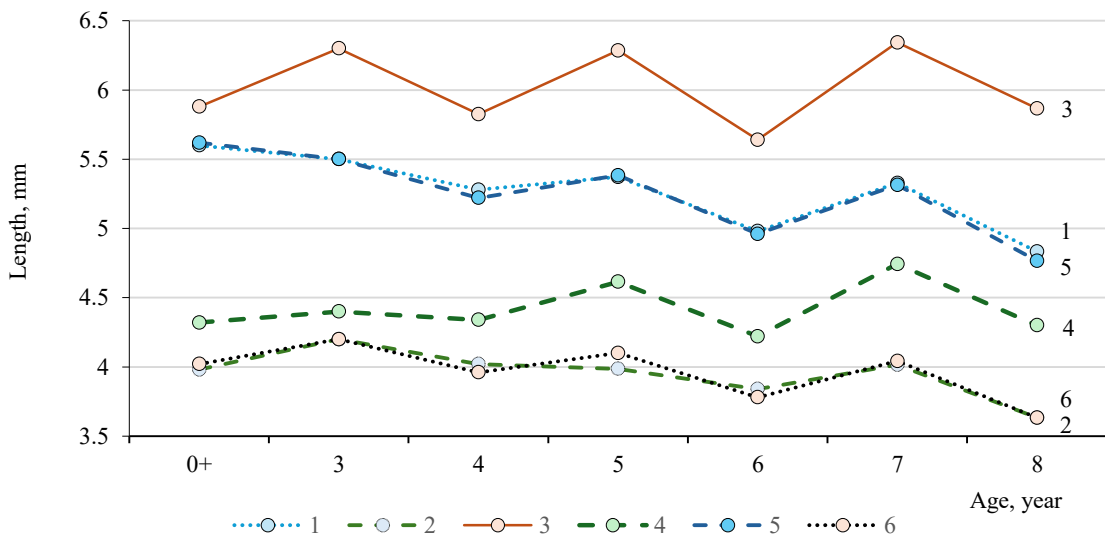


Fig. 4. Age-related changes in the average values of canine parameters. 1, tooth neck thickness below the enamel; 2, tooth neck width below the enamel; 3, maximum root thickness; 4, maximum root width; 5, tooth neck thickness along the enamel; 6, tooth neck width along the enamel.

Рис. 4. Зміна з віком середніх значень параметрів іклів: 1, товщина шийки під емаллю; 2, ширина шийки зуба під емаллю; 3, максимальна товщина кореня; 4, максимальна ширина зуба; 5, товщина шийки зуба по емалі; 6, ширина шийки зуба по емалі.

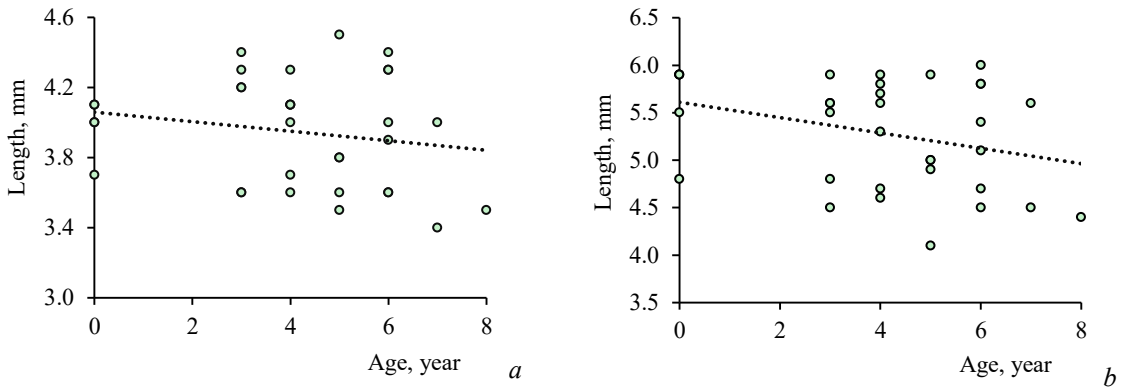


Fig. 5. Distribution of the tooth neck thickness under the enamel (a) and tooth neck thickness along the enamel (b).
 Рис. 5. Розподіл товщини шийки зуба під емаллю (a) і товщини шийки зуба по емалі (b).

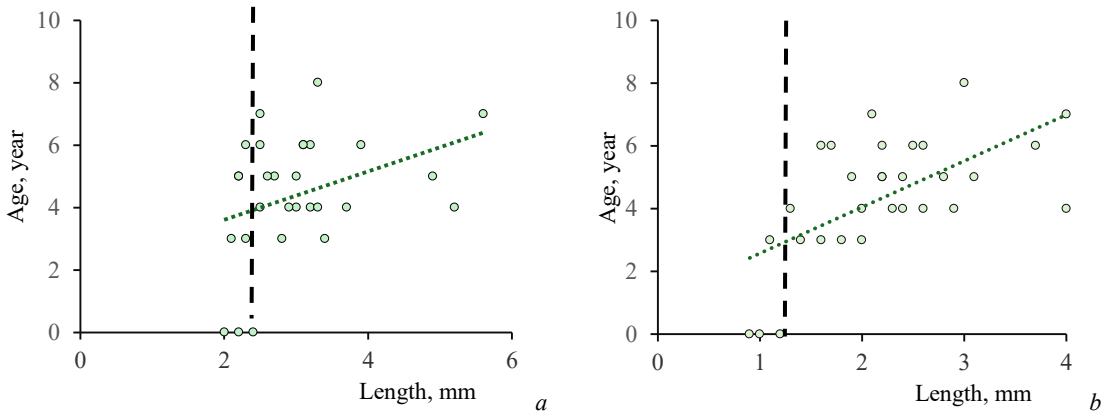


Fig. 6. Distance from the edge of the alveolus to the neck of the tooth (canine): (a) mesial side; (b) distal side.
 Рис. 6. Відстань від краю альвеоли до шийки зуба (ікла): (a) мезіальна сторона; (b) дистальна сторона.

During the first year of life, overgrowth of the canine canal with dentin does not exceed 20% (Fig. 7). In older year classes (>2 years), a significant range of individual variation in the overgrowth of pulp cavity (root canal) was observed, ranging from 71.0% to 86.17%. In these cases, inter-individual differences significantly overlapped age-related differences. The coefficient of determination (R^2) in the model examining the dependence of pulp cavity width on age was 0.6344.

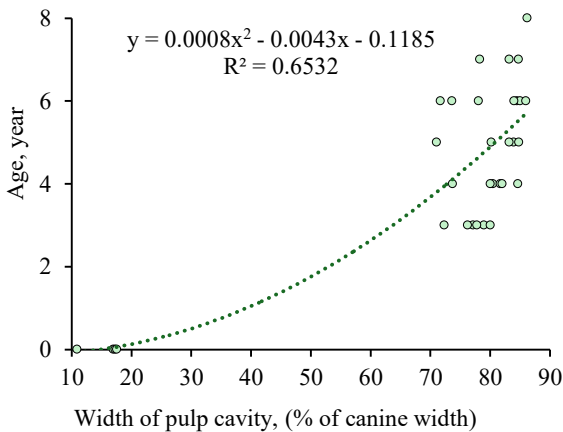


Fig. 7. Changes in the width of the canine pulp cavity in % of the maximum width of the canine in the wildcat.

Рис. 7. Зміни ширини порожнини пульпи (каналу) ікла у % від максимальної ширини ікла у кішки лісової.

Table 3. Number of year classes (according to cementochronology), n = 38

Таблиця 3. Кількість річних класів (за цементахронологією), n = 38

	Year class, years						
	0+	3	4	5	6	7	8
Number of individuals	5	2	7	6	12	4	3

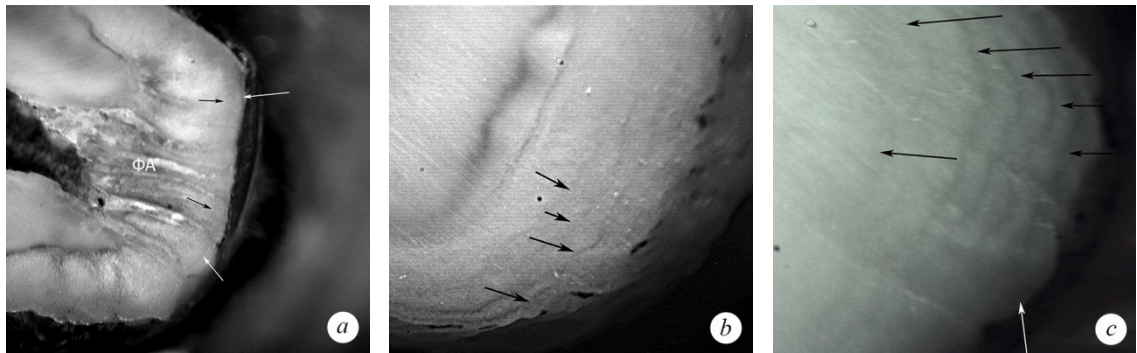


Fig. 8. Cement deposition in the root of the upper canine of the wildcat (apical part of the canine root). (a) 3+ (May) (arrows—two dark ‘winter’ layers of cement; the formation of a ‘light’ layer of cement is noticeable); (b) 4+ (four clearly visible, fully developed dark lines of cement); (c) 7+ (January) (six developed dark lines, the seventh is formed on the outer edge of the root, light arrow). AD, apical delta.

Рис. 8. Відкладення цементу в корені верхнього ікла лісової кішки (апикальна частина зуба). (a) 3+ (травень) (стрілки — дві темні «зимові» шари вторинного цементу; помітно утворення «світлого» шару цементу); (b) 4+ (чотири добре помітні, повністю розвинені темні лінії вторинного цементу); (c) 7+ (шість розвинених темних ліній, сьома утворюється на зовнішньому краї кореня — світла стрілка). AD, apical delta.

The results of cementochronology conducted on canines are presented in Table 3. The dark lines, representing the deposition of cement layers, are most clearly visible in the apical part of the root and the adjacent lateral surface (Fig. 8). In 12 individuals, the formation of incomplete or broken lines was noted. The remaining specimens exhibited only well-defined lines corresponding to the second and subsequent autumn–winter periods of life.

Discussion

Ossification of sutures

The pattern of fusion of the cranial sutures has been used for many decades as a method for assessing age. However, this method is rather unreliable and requires considerable improvements through the implementation of new techniques for studying cranial sutures, methods for assessing their degree of obliteration, and shifting focus to the examination of cranial base and facial sutures [Ruengit *et al.* 2020]. The basicranial sutures are of particular interest [Cendekiawan *et al.* 2010]. Experimental evidence has shown that the length of the facial part of the cranium is significantly affected by the timing of sutural fusion at the base of the skull [Rosenberg *et al.* 1997]. Functionally, the fusion of basicranial sutures may reduce variability and probability of deformations occurring in the skull during mastication [Herring & Teng 2000].

The obliteration of sutures in the basal part of the cranium is characterised by heterochrony in the rate and timing of development, which varies depending on ecological specialisation in placental mammals and marsupials [Rager *et al.* 2014; Sánchez-Villagra *et al.* 2008], primates [Rengasamy Venugopalan & Van Otterloo 2021], and carnivorans [Goswami *et al.* 2013]. Goswami *et al.* [2013], in their analysis of timing of skull suture closure concluded that canids exhibit a greater degree of heterochrony than felines, with food specialisation exerting a significant influence on suture obliteration. In felines, the percentage of fully closed sutures is generally lower compared to canids.

Furthermore, larger felines have higher percentages of obliterated sutures (e.g. 54.7% in *Panthera tigris*, ~60% in *Panthera leo*) than smaller species like the wildcat (*Felis silvestris*, 33.6%) [Goswami *et al.* 2013]. Among terrestrial predators, species closest to felines (e.g. wildcats) in terms of cranial suture closure are *Vulpes vulpes* and *Canis lupus*, which show 1.5–2 times higher maximum percentages of suture closure. The weak overgrowth of the studied sutures (BB, PB, and MP) at the skull base in *Felis silvestris* renders this tool largely uninformative for age determination, unlike its applicability in foxes.

The basioccipito-basisphenoidalis (BB) synchondrosis is the first among the sutures we studied to distinguish two age groups: individuals younger than 3–4 years and older individuals. A similar pattern of obliteration was observed by R. Perea-García [1996] in four species of *Lynx*.

According to T. Kvam [1983], in *L. lynx*, this suture closes at 5–6 years of age and is completely obliterated in older individuals. Meanwhile, other sutures, such as PB and especially MP, on the outer skull surface remain open or partially open even in very old specimens (7–8 years old) [Kvam 1983; Perea-García 1996]. The weak obliteration of the latter two sutures makes them unsuitable as independent tools for age determination. This conclusion is supported by our limited findings regarding wild and mongrel *Felis catus* specimens.

Previously, for domestic cats, Fleming *et al.* [2021] established that the complete closure of basal sutures occurs over a long-time interval: BB from 2 to 7 years, and PB from 3 to 13 years. In Persian cats and domestic shorthair cats, the final overgrowth of these sutures can take up to 181 to 240 months (15 to 20 years) [Schmidt *et al.* 2019].

Teeth

An open pulp cavity at the apical part of the canine root is universally recognised as a sign of an age range from one to two years. Wilson [1999], based on results of X-ray examinations of *Felis catus* teeth, found that the upper canines are the last to close in 80% of cases at 7–11 months of age. Similar conclusions were drawn by Goraya *et al.* [2024], who showed that in intact females and castrated males, the tips of the upper canines close between 12 and 14 months, while the tips of the lower canines close between 10 and 12 months.

In the Iberian wildcat (*Felis silvestris*), the roots of the canines remain open between 6 and 18 months of age [García-Perea & Baquero 1999]. Piechocki & Stiefel [1988] reported sporadic closure of the canine canal at 6–8 and 10 months, with mass closure beginning at 11 months.

For Bengal cats, Nakanishi *et al.* [2009] observed that the apical foramen of the canines gradually closes from October to March, fully closing at one year of age. Later, at 1.5 to 2 years, the apical part of the root is completely closed in caracals [Stuart & Stuart 1985] and in *Lynx pardinus* [Zapata *et al.* 1997].

Information about the diameter or area of the pulp cavity opening in the canine root remains insufficient to fully understand the dynamics of closure. According to Stuart & Stuart [1985], during the 10 to 15 months of a kitten's development, the diameter of the pulp cavity opening in the root decreased by half: from 3.4 mm to 1.65 mm. Piechocki & Stiefel [1988], studying *Felis silvestris*, noted differences in this parameter between males and females with high individual variability. For males, they reported 0.3 mm in October (10 months) and 0.5 mm in December (8 months), while for females, the diameter was 1.6 mm in December at 6 months of age.

Furthermore, in mid-February, these authors observed an unhealed canal opening of up to 3 mm in one individual (8 months old). This highlights the need to consider not only the correlation between pulp cavity closure and age but also indirect factors such as the time of birth of kittens. For kittens born earlier in the year, the canine canal closure in the apical root region occurs earlier.

In the wildcat population of the Black Sea region, a female with blind kittens was observed as early as mid-January [Potapov 2000]. Although late winter to early spring is considered the typical start of mating season [Tokar & Matveev 2010], such early births may account for the delayed closure of the canine canal in the apical root observed in our studies.

The formation of the set of permanent teeth in wildcats is completed at the age of 175–180 days in females and 190–195 days in males [Condé & Schauenberg 1978]. After the pulp cavity at the apex of the canine root becomes overgrown, by approximately one and a half years of age, the length of the canines reaches, as shown by Piechocki & Stiefel [1988], 21 to 28 mm (in a single case up to 30 mm). These values represent the final (true) size of the canines, after which their length remains almost constant. Over time, the parameters of the tooth, particularly the canines, are influenced by opposing processes: abrasion of the enamel crown and the build-up of cementum in the root part of the tooth.

As a result of cementum deposition on the apical surface of the root, the tooth is gradually pushed out of the alveolus, compensating for the abrasion of the tooth crown [Piechocki & Stiefel 1988]. This process maintains the consistent height of the tooth above the alveolus and the total length (height) of the tooth. The size of the canines in individuals older than 20 months remains within the parameters typical of young wildcats during their first year of life [Piechocki & Stiefel 1988]. Consequently, using total tooth length as a marker of a specific age (or year class) is problematic. Other quantitative characters of the canines (e.g. thickness, root and crown width) for age assessment are not well-documented in the literature and, as in our study, may prove uninformative for estimating the age of wildcats.

More attention has been paid to qualitative characters of tooth crown wear. Numerous criteria have been described in the literature for assessing tooth crown wear in felines [Nakanishi *et al.* 2009; Stander 1997; White & Belant 2016]. Most of these criteria are subjective and difficult to standardise. Age classifications based on them typically group animals into broad age ranges rather than precise year classes. For example, Marti & Ryser-Degiorgis [2018] proposed a promising scheme for determining the lifetime age of Eurasian lynxes, dividing individuals into six age groups using qualitative characteristics such as tooth colour, tartar deposits, canine morphology, and tooth wear. In Bengal cats, Nakanishi *et al.* [2009], based on canine abrasion, distinguished only three groups: 1 year, 2–6 years, and over 7 years. In lions, Smuts *et al.* [1978] used a conventional visual classification: barely noticeable signs of wear at the tips of the teeth appear at 3–4 years of age, moderate wear at 7–9 years, and significant wear only in individuals older than 10 years. An analysis of PM2 abrasion [White & Belant 2016] yielded a different age group scale for lions, emphasising the varying roles of different tooth groups in food processing.

Diet composition significantly influences tooth wear. A diet including harder foods, such as bones, leads to faster tooth wear [Binder *et al.* 2002]. Similarly, sand ingestion in certain regions accelerates tooth abrasion [Smuts *et al.* 1978]. The small-sized prey of wildcats in the Black Sea region (e.g. mice, voles) has a negligible impact on tooth wear. As a result, fewer than 20% of wildcats in older age categories show visible canine wear.

In the practice of field research, methods for determining the age of living individuals are particularly attractive and valuable. These methods are based not only on measuring tooth abrasion but also on assessing tooth colour [Smuts *et al.* 1978; Marti & Ryser-Degiorgis 2018], tartar deposition on canines [Marti & Ryser-Degiorgis 2018], or gum line recession [Currier 1979; Laundré *et al.* 2000; Fàbregas & Garcés-Narro 2014]. The latter is considered a promising method [Laundré *et al.* 2000] but requires further development. It should be applied cautiously to hunted animals, as the gums of deceased specimens may lose turgor and recede, affecting the accuracy of age assessment. Its equivalent for working with collection material can be the measurement of the distance from the edge of the enamel to the edge of the alveolus.

Smuts *et al.* [1978] previously suggested considering the height between the enamel margin and the alveolus edge, though without providing clear justification. Later studies demonstrated that this distance changes with age due to cement build-up in the apical part of the root of feline canines [Kvam 1983; Piechocki & Stiefel 1988]. The gradual protrusion of the tooth from the alveolus, resulting from this process, corresponds to the growth of cement deposits with age.

In *Lynx lynx*, part of the canine root becomes visible 36 months after birth [Kvam 1983]. Analysis of the dynamics of canine protrusion from the alveolus allows the division of wildcat populations

into two age groups: 1–2 years (0+) and >2 years. However, this variability complicates establishing precise age stratification criteria for *Felis silvestris*. At the same time, a more accurate age estimation was obtained by measuring the distance between the enamel edge and the alveolus on the distal surface of the canine. The mesial edge, compared to the distal edge, is characterised by greater curvature, which complicates the determination of age stratification criteria in *Felis silvestris*. Nevertheless, such a non-destructive method of age determination can be useful when working with collection materials of such rare species as the wildcat in Ukraine.

Dentin/cementum annule

Non-destructive approaches can also include a method of measuring the progressive accumulation of dentin in the pulp cavity, which tends to narrow with age in the canines, performed using X-ray equipment on living [Lee *et al.*, 2020] or collection materials [Binder *et al.* 2002; Park *et al.* 2014; Stefen 2015; Zapata *et al.* 1997]. A simpler, more accessible, and cost-effective alternative involves mechanical tooth treatment, first proposed by Smirnov [1960] and later mentioned by Spinage [1973]. Closure of the pulp cavity is regarded as ‘...a reasonable indicator of the age of predators’ [Binder *et al.* 2002], as it does not depend on diet and does not correlate with the degree of enamel wear.

According to Park *et al.* [2014], in *Felis catus*, a progressive decrease in the pulp cavity width occurs up to 12–24 months, followed by high variability in values ranging from 2 to 9 years. This variability limits the ability to assess age beyond approximately 10 months. For *Lynx pardinus*, Zapata *et al.* [1997] identified three age groups based on pulp cavity width: 1 year, 1–2 years, and older. In lions, pulp cavity overgrowth ceases after about 8–9 years [Smuts *et al.* 1978], possibly due to their larger tooth sizes. Similarly, delayed closure of pulp cavities has been observed in cheetahs.

Thus, existing data on age-related changes in pulp cavity width highlight its limited value. Our results align with these findings, indicating that age determination is possible only for two groups: 1–2 years and older.

A widely used and accurate method for determining age is currently the counting of cementum lines in the tooth root. A detailed review of research directions on cement deposition in teeth is provided by Perrone *et al.* [2022]. The advantage of this method is that, unlike dentin, which is deposited in the tooth for a limited time, the formation of cement lines continues until the end of the animal’s life.

Dark and light lines of cement are directly fixed when they are examined. In contrast, changes in the pulp cavity, when filled with dentin, require additional calculations and are assessed using calculation coefficients [Binder *et al.* 2002]. However, the formation of a dark line in the cementum does not represent the calendar year of the animal’s life but rather the time of its appearance. Cement line deposits are most pronounced in regions where food resources and breeding seasons exhibit strong seasonality [Hiller *et al.* 2022]. It typically occurs during winter when cementum growth slows down [Garcia-Perea & Baquero 1999].

The timing of the appearance of the first cement line remains a subject of debate. In the Bengal cat, Nakanishi *et al.* [2009] observed the emergence of a short, indistinct dark line on the root side even before the closure of the apical foramen. The early onset of cementogenesis in wildcats, starting at around 4 months of age, was previously reported by Piechocki & Stiefel [1988]. This process, however, does not always occur during the early stages of development. In the first year of life, a continuous (‘winter’) line is formed in only a part of *Lynx pardinus* individuals [Zapata *et al.* 1997]. According to the findings of Garcia-Perea & Baquero [1999], the first dark line does not form in about half of Iberian wildcats during the autumn–winter period of their first year. These authors also noted the presence of discontinuous lines between the first visible line and the cementum–dentin boundary, likely associated with the closure period of the apical foramen. The open apical root region may inhibit the formation of a complete, continuous line during the animal’s first year.

The varying proportions of broken lines, ‘kitten lines’, and continuous lines led Garcia-Perea & Baquero [1999] to propose adding one to the count of continuous lines to estimate the age of Iberian

wildcats. Continuous lines begin to form from the second autumn–winter period, typically at an average age of 21 months in wildcats and 18–24 months in *Lynx pardinus* [Zapata *et al.* 1997]. A similar approach has been deemed appropriate for estimating the age of wildcats in the north-western Black Sea region and was applied in our study.

Our data indicate that the period of overgrowth of the apical foramen is prolonged. By late October and January, the apical foramen was still open. This corresponds to an age of at least 8–11 months, assuming April as the time of birth. Under these conditions, the formation of a continuous line is not possible. We observed discontinuous lines in only about one-third of individuals, likely forming during this first autumn–winter period of life when the apical foramen closes. During this time, either a discontinuous line may form or no line may be present, as noted by other researchers [Zapata *et al.* 1997; Garcia-Perea & Baquero 1999].

A discontinuous line on the mesial or distal surface of the canine is located very close to the dentin line. The distance between the first continuous line and the discontinuous line is significantly greater. The spacing between the first continuous and discontinuous lines on the lateral surfaces of the canine root is relatively dense, which can sometimes make their identification challenging in the apical foramen). Frequently, these and subsequent cement lines are located outside the apical delta (Fig. 8a), through which nerve endings penetrate the pulp. Most of the canines we examined showed only continuous lines. Their formation occurs after the apical foramen overgrows, which, under Black Sea region conditions, happens during the second year of life.

Based on this, the following rule was applied for age determination: the first clearly identifiable continuous dark line located after dentin formation represents an age of 2 years (corresponding to the second winter). This approach helps to avoid subjectivity in age assessments by different researchers employing various methods to identify the presence of the first-year line in tooth sections. The number of continuous lines counted using this method is increased by one, which will give the desired age estimate. This estimate can be refined if the birth time of the animal is known.

Conclusions

1. Cementochronology effectively divides the wildcat population into distinct year classes.
2. The volume of dentin overgrowth in the canine canal is no more than 20% in wildcats during their first year of life. In older age groups, this overgrowth increases to 3–4 times higher, exhibiting high individual variability.
3. A distance of 1.25 mm between the enamel edge at the tooth neck and the alveolus edge serves as a clear marker distinguishing young cats (0+) from individuals over 2 years old.
4. The sutura basioccipito-basisphenoidalis begins to close predominantly in individuals aged 5 years and older.

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