

**Simple method of age determination in red fox, *Vulpes vulpes***Jana ROULICHOVÁ<sup>1</sup> and Miloš ANDĚRA<sup>2</sup>*Department of Zoology, National Museum (Natural History), Václavské nám. 68, CZ-115 79 Praha 1, Czech Republic; e-mail: <sup>1</sup>vulpesvulpes@centrum.cz, <sup>2</sup>milos\_andera@nm.cz*

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**Abstract.** A simple method of age determination in red fox (*Vulpes vulpes*) is described. It is based on counting dentine layers on longitudinally sanded canine roots without the tedious procedure of preparing histological sections. The method makes it possible to estimate rather reliably the age of red fox specimens from one year onwards.

**Key words:** sanding of dental roots, counting dentine layers

Exact individual age determination is an important part of population studies in various animal groups. In the case of red fox (*Vulpes vulpes*), several methods have been used, based on body weight, body length, cranial dimensions, dry weight of eye lenses, degree of fusion of cranial sutures or development of the sagittal crest, tooth wear, occlusion of the dental pulp, or increments in dentine layers (Klevezal & Klejnenberg 1967, Bree et al. 1974, Harris 1978, Nelson & Chapman 1982, Driscoll et al. 1975, Goszczyński 1989, Wanderer & Lüps 1993, Ansoerge 1994, Cavallini & Santini 1995, Zapata et al. 1995 etc.). Counting the increment layers of secondary dental cement is considered relatively the most exact method. In sections through the tooth roots, they appear as characteristic annual rings consisting of paler (summer) opaque and darker (winter) transparent areas (e.g. Klejnenberg & Klevezal 1966, Grue & Jensen 1973, Harris 1978, Nelson & Chapman 1982, Cavallini & Santini 1995, Zapata et al. 1995). Longitudinal sagittal sections are more satisfactory, as in the technically simpler transverse sections the interpretation of the number of increment zones may be problematical since the cement layers are not uniformly spaced on the outside of the root and, hence, it is significant to select a suitable level of the section. While the reliability of this latter method is up to 90% (Grue & Jensen 1973), its notable drawback is in being considerably time-consuming. The tooth must be decalcified with 5% nitric acid (36–48 hours) and subsequently rinsed with water (24 hours), then the transverse section 20 µm thick must be made with a freezing microtome (at -20 °C) and stained using standard histological techniques (hematoxylin) and finally examined under an optical microscope at 80x magnification.

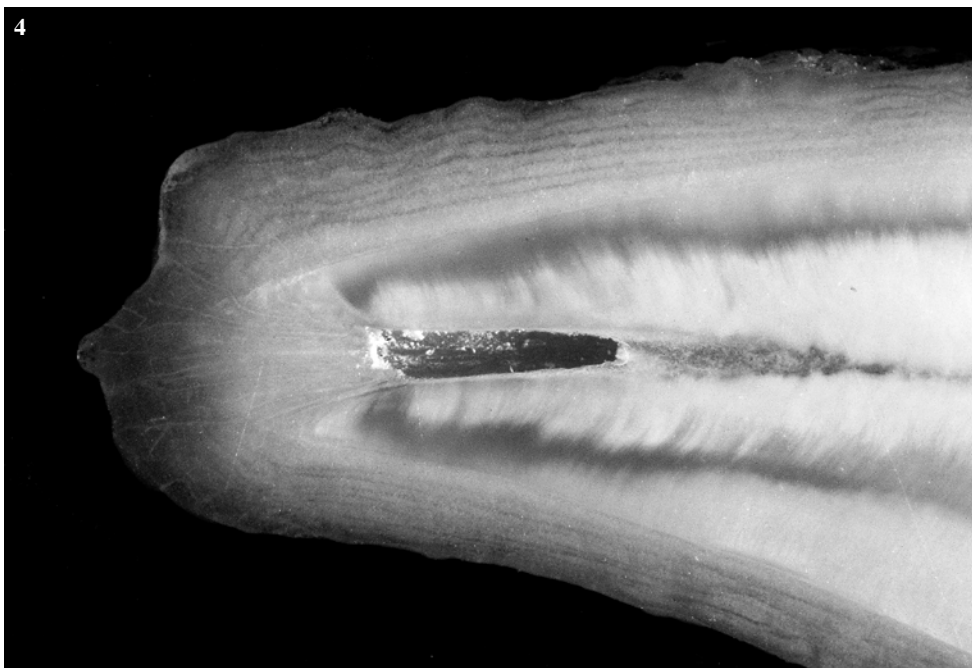
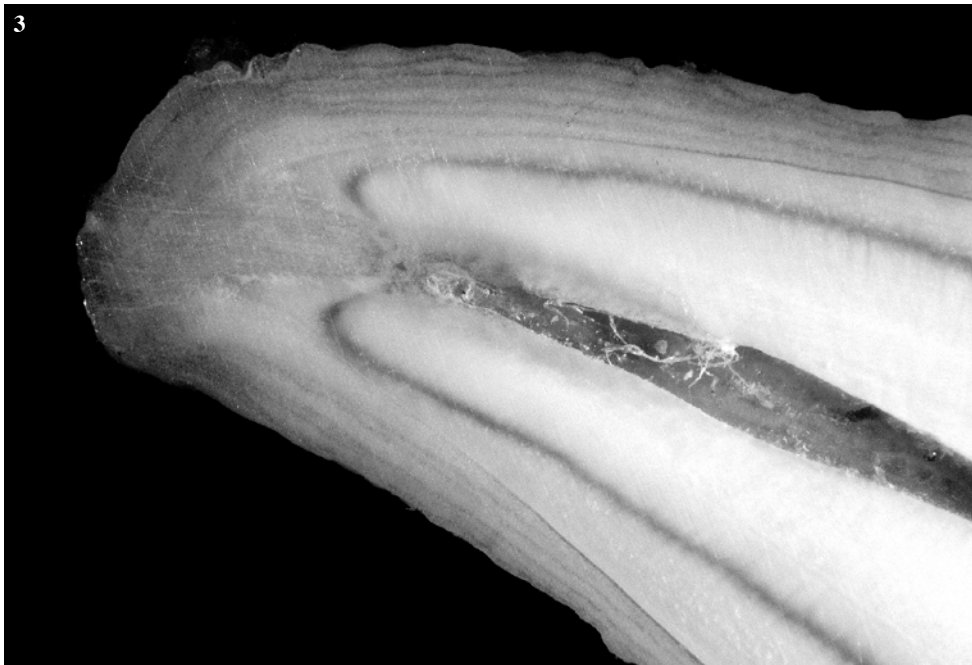
In studies of the red fox population structure in the Czech Republic we used a much simpler method of counting the increment layers of secondary dental cement, based on longitudinal sanding of dental roots. They were prepared from one or two upper canines or any of the lower ones (should, in extreme cases, any of the canines be missing, one can use any available premolar). The tooth was carefully extracted from the jaw (by hand or with dental forceps). Then its root was ground by hand down to roughly half its thickness, using sandpaper coarseness ca 80 then smoothed at coarseness ca 400 and finally polished at very fine coarseness ca 1200. The preparation can be examined under a stereoscopic microscope

1



2





**Figs 1–4.** Sandings made from the root of red fox upper canine. (1) 12 months of age (arrow: beginning formation of first dark layer of secondary cement); (2) 2 years of age (two clearly visible, fully developed dark lines of secondary cement all along the circumference of root); (3) 5 years of age (five dark lines of secondary cement visible on either side of root); (4) almost 8 years of age (seven fully developed lines, eighth line discernible on outer margin of root).

at 25x magnification, without any staining (Figs 1 – 4). The age of the specimens more than a year old is estimated by simply counting the dark lines and adding the number of months between the estimated birth date (in central Europe this is on 1<sup>st</sup> April on average; Ansorge 1990, own data) and the known date of death. The dark lines are most satisfactorily discernible at the sides of the root close to its apex. In most cases, it was impossible to count them directly on the apex due to this place being often damaged or its structure being “glassy” and the lines being indiscernible. For mutual checking and greater precision, it is recommended to make at least two preparations from each specimen.

To test the effectiveness of the method described above, we used a series of 344 red fox skulls deposited in the collections of the Zoological Department of the National Museum in Prague, shot between 1995 and 2004 in various regions of the Czech Republic (most frequently in northern and south-western Bohemia). The material was obtained from the State Veterinary Institute in Liberec-Vratislavice, comprising only specimens shown to be negative as to rabies. Data on locality, sex and date of killing were available for each specimen. The skulls were prepared from fresh materials, no fixation of tooth was performed. At the same time the age of individual specimens was determined by other methods as well, namely by examination of tooth wear (M<sup>1</sup>) and occlusion of selected cranial sutures. Accurate aging made it possible to study the ontogenetic variability and age structure of the Czech red fox populations (Sedláčková & Anděra 2006, Anděra & Sedláčková 2007).

The main advantage of the method described above is in its insignificant amount of time and material. The age of the specimens can be determined with considerable precision until they reach a great age. Another inherent advantage in this careful method where materials in a collection are concerned is that it does not require complete damage of the teeth examined; (with careful grinding the tooth can subsequently be replaced in the jaw without any visible damage). We believe that using this method in other species of carnivores is possible.

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