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## Age determination in the European otter *Lutra lutra lutra*

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### Abstract

Aged carcasses of free living specimens of European otters *Lutra lutra lutra* from Norway by different methods. The numbers of dark-staining, incremental cementum lines observed in histological sections of teeth, the degree of cranial development, and the stage of development of the permanent dentition were compared. Based on the above information and the previously reported developmental stages of captive European, North American (*Lutra canadensis*) and African (*Aonyx* sp.) otters of approximately known age, the conclusion is that the number of dark-staining incremental cementum lines are likely to yield the approximate age in years. One source of variation in the relationship between an animal's age and the number of cementum lines in its teeth is the correlation of line formation with season of year, combined with the lack of a specific season of birth, at least over part of the natural distributional range of the European otter. Cranial development however, may be used to yield a more accurate estimate of age for otters up to 2 years-old. Dentitional changes are likely to have been completed before an age of 6 months and may thus yield additional information about the age of very young cubs.

### Introduction

Reliable age determination techniques are essential for many aspects of biological studies. In the European otter *Lutra l. lutra* the possibilities for ageing specimens from their external characteristics are very limited, because of their rapid growth. Studies of captive otters show that one year-old animals are difficult to distinguish from older ones on size grounds. The fur difference between young and adult otters is slight and the fur has already attained its adult appearance at an age of 1½-2 years (REUTHER, pers. comm.). When carcasses are available for study a different range of morphological traits can be considered. BREE et al. (1966) devised a skull index and a baculum index for separating adult male European otters from subadults. STUBBE (1969) classified his otter carcasses as "first year of life", "second year of life" and "older than two years" on a basis of skull characters, the ischiopubic junction and, for males, the os baculum. This assessment was made intuitively, on the basis of knowledge of other mustelid species (STUBBE, pers. comm.).

Development of the method of using the dark-staining incremental lines visible in histological sections of teeth and bone for age determination has greatly improved the possibilities of making relatively reliable and accurate estimates of age, at all stages of the life-cycle, for many mammalian species. For several species of animals living in the Arctic

and Temperate regions, examination of specimens of known age has shown that one line is formed each year, starting at a species specific age which depends on the age at which the permanent teeth erupt. This method and its application to a great number of species has been described and reviewed by KLEVEZAL' and KLEINENBERG (1967) and by GRUE and JENSEN (1979).

Unfortunately, the European otter does not belong to the species for which animals of known age have been available for testing the above age determination method. GRUE and JENSEN (1979) however, found that dark-staining lines were formed in the cementum of otters from Denmark during a single restricted period per year, indicating that in this species, too, a single cementum line is formed per year. For the closely-related North American river otter, *L. canadensis*, STEPHENSON (1977) states: "The number of annuli in the known-aged otters showed complete agreement with age; however, some annuli were faint and additional discontinuous lines were observed." The river otter has a fixed breeding season in the wild (HAMILTON and EADIE 1964; HARRIS 1968), whereas the European otter may breed at any time of the year, at least over part of its natural distributional range (STEPHENS 1957; JENSEN 1964; HARRIS 1968; STUBBE 1969; JOHNSEN and MYRBERGET 1980). This affects the accuracy of any attempts to age European otters from supposedly seasonal incremental lines in the cementum.

So far no studies exist which have managed to establish the true age classes over the whole age spectre of a wild population of European otter. Elucidation of the properties of indices which may provide a key to the true age of any age class represents a first step in this direction and has formed the aim of the work reported in the present paper.

## Material and methods

Carcasses of the European otter *Lutra l. lutra* obtained from Norway north of 61° N, sent to the Directorate for Wildlife and Freshwater Fish, Game Research Division, during the period 1968–1981 have been studied in the present investigation. Some of these otters had died accidentally and the whole animal was available. In most cases, however, skinned carcasses were bought from otter hunters. Whenever possible, each animal was aged by two different methods;

1. according to STUBBE's (1969) skull criteria,
2. by making counts of the cementum lines.

The stage of tooth development was also noted for those animals whose permanent dentition was still incomplete. Teeth were considered as being fully developed when the neck of the tooth had reached the edge of the alveole.

Skulls were cleaned by boiling. Histological sections, usually of the upper canines, were generally made according to the method described by REIMERS and NORDBY (1968). The teeth sections were initially 30 microns thick, but later on this was reduced to 16 microns. For some specimens sections were also made from other teeth than the canines. The dental formula was assumed to be

$$\frac{3 \ 1 \ 4 \ 1}{3 \ 1 \ 3 \ 2} = 36 \text{ (HARRIS 1968).}$$

## Results

### Development of permanent teeth

A total of 10 otter cubs, whose permanent teeth were still developing, were available for an investigation of the sequence in which the teeth of this species develop. The process of dentitional change was fairly advanced in all cases. Incisors and P<sup>1</sup> were fully grown. Thus the sequence of development of these particular teeth could not be established. The following consistent pattern of permanent tooth development was finally obtained:

The incisors and first premolars develop first. Development of the molars is completed next, then that of the premolars P<sup>2</sup>, P<sup>4</sup>, and the canines, although the sequence within this group varies. The upper canines are a little delayed compared to the lower ones. The last

permanent teeth to be developed are the third premolars of both jaws, replacing carnassial-like deciduous premolars which are retained until after the permanent carnassials have become functional.

Sections of upper canines were made from 3 cubs whose canines were not quite fully developed, and from 4 cubs whose skull development was very similar to that of the former, except that all their teeth were fully developed. In the sections from the cubs with still developing canines no cementum was present and only an extremely thin dentine, about 0.1 mm thick near the apex. In those from the cubs with fully-developed teeth a thin layer of light-staining cementum was present. Thus deposition of cementum does not commence until about the time when the dentition change is complete.

### Configuration of the incremental cementum lines in older animals

The cementum layer deposited before the formation of the first incremental line was always relatively broad near the apical end of the root. In animals with more than one incremental line, this cementum layer was much broader than the light-staining layers between the lines (Fig. 1a). In all the otters investigated the cementum deposit was concentrated to the apical part of the root and quickly tapered off to a very thin layer towards the cementum/enamel interface (Fig. 1b). The maximum number of primary lines observed was nine. Lines which were judged to be secondary were sometimes seen, but resorption lines were uncommon. Usually, different teeth from the same animal showed the same number of lines, but the clarity of the lines differed.

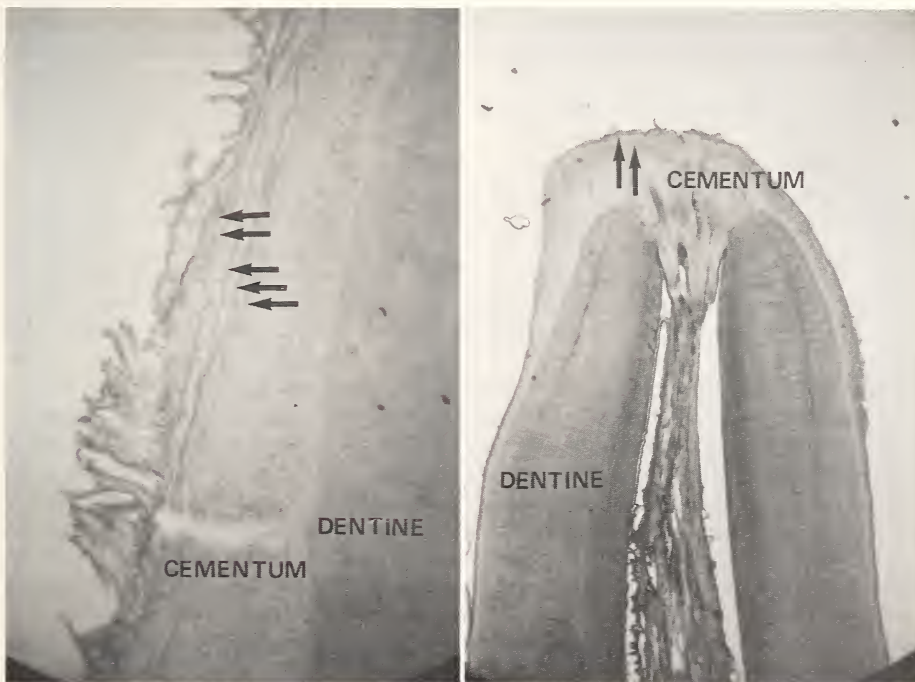


Fig. 1. Longitudinal sections from roots of permanent teeth from European otters *Lutra lutra lutra*. a (left): Cementum and part of the dentine in an upper canine showing 5 dark-staining incremental cementum lines (at point of arrows). The first layer of light-staining cementum is much broader than the consecutive layers. – b (right): The root of a third upper incisor with 2 dark-staining incremental lines (at point of arrows). The cementum is concentrated to the apical part of the tooth



Line configuration seemed to be quite similar to that of the river otter *L. canadensis* (STEPHENSON 1977). The darkstaining lines often lay close together, were occasionally wavy, fusing, diverging and becoming diffuse in some tooth sections.

### Time of formation of incremental lines

In the European otter the process of deposition of dark-staining incremental lines can only be identified if the technical quality of tooth section is optimal. Also, the morphological distinctness of lines must attain a certain standard which was not the case for all the samples. Altogether, the section from 45 animals, whose date of death was known, reached the desired standard. The dates were distributed over all the months of the year, varying from 1 to 8 animals per month. Lines in process of deposition were found in all months from February to July, inclusive, excepting June (Fig. 2).

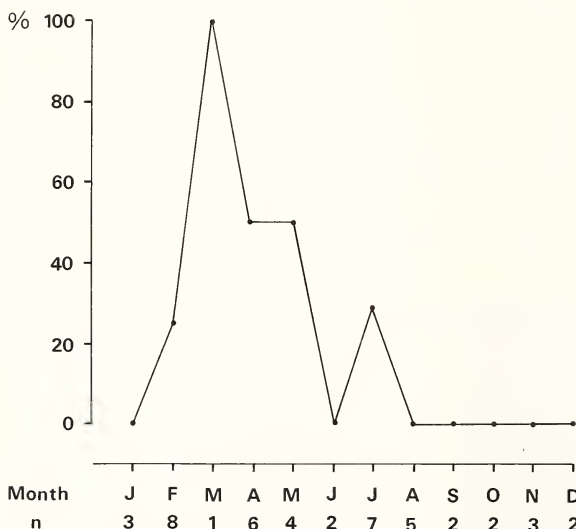


Fig. 2. Proportion of otters *Lutra lutra lutra* from Norway in which dark-staining cementum incremental lines were in the process of deposition. Otters with very thin cementum were not included (cf. GRUE and JENSEN 1979)

### Comparison of age classification based on incremental line number and on skull criteria

STUBBE'S (1969) age criteria comprise "animals in their first year of life", "animals in their second year of life", "animals more than two years old". In the present study the categories "approximately one year old" and "approximately two years old" were added, to take account of those skulls which possessed intermediate characteristics and which might equally well have been included in either of two consecutive age groups.

The numbers of animals in each age category based on skull criteria and on incremental line counts are shown in Table 1. For this purpose animals with more than 2 incremental lines were combined to form a single category, which also includes those animals for which the exact number of lines was difficult to determine, providing there were definitely more than 2 present. Assuming that grouping by STUBBE'S criteria assigns the right age to every animal, and that one dark-staining incremental line is deposited every year, starting when the animals are between  $\frac{1}{2}$  and  $1\frac{1}{2}$  years old as suggested by GRUE and JENSEN (1979), then the postulated age relationships of the different skull and cementum line categories should be as follows: Animals "in their first year of life" ( $< 1$  in Table 1) and animals "approximately one year old" ( $\approx 1$ ) may have 0 or 1 incremental line. Animals "in their second year of life" ( $1-2$ ) may have 0, 1 or 2 incremental lines. Animals "approximately 2 years old" ( $\approx 2$ ) may have 1 or 2 incremental lines. Animals "more than two years old" ( $> 2$ ) may have 1, 2 or  $> 2$  lines.

In only one case was a deviation found from the expected pattern: viz. an animal with 2 incremental lines was classified as "approximately one year old" on its skull criteria. This was a pregnant female. The age classification was based on the fact that the nasal sutures

Table 1

The age distributions (numbers of animals in the different age groups) given by the two different estimations

Estimated age in years <sup>1</sup>	Number of dark-staining incremental cementum lines seen in tooth sections			
	0	1	2	> 2
< 1 year	19	1	—	—
≈ 1 year	1	5	1	—
1–2 years	4	4	—	—
≈ 2 years	—	5	4	—
> 2 years	—	6	12	36

<sup>1</sup> based on Stubbe's (1969) skull criteria.

were very clearly visible, as they should be in the category “first year of life”, whereas the remaining skull characteristics were typical of those of the age category “second year of life”.

## Discussion

### Age in relation to developmental stage of permanent teeth

It is important to find a means of ageing of juvenile European otters individually on morphological grounds since the possibility of assigning an approximate age based on the sampling date is ruled out for a species which has no particular season of birth. If the age at which the dentitional change takes place can be determined, and if the sequence of tooth replacement is known, then jaws, and even jaw fragments in which deciduous or still-developing permanent teeth are present, may allow a relatively accurate estimate of the age of the animal to be made.

Only scant information is ascertainable from previous reports about the different stages of tooth development in relation to age. The few observations made also derive from otter cubs which were born in the wild and then kept in captivity from a very early age, judging by their stage of development compared to that of cubs born in captivity. From these reports (COCKS 1881, 1877 in HARRIS 1968; STEPHENS 1957; NEAL 1962 in HARRIS 1968) the age at which the permanent teeth start to emerge is estimated to be about 2 months. For three cubs of an African species of otter, *Aonyx* sp., reared in captivity after they were about 2 weeks-old, the permanent dentition started to appear in the ninth or tenth week, and all the deciduous teeth had been shed by the end of the eighteenth week.

In the Mustelidae the permanent teeth generally develop early in life. In Sweden, the permanent dentition of young badgers, *Meles meles*, born that year, was usually complete in the middle of July (AHNLUND 1976). Judging by the usual season of birth of badgers they must then have been approximately 4 months old.

It seems reasonable to assume that the permanent teeth of the European otter become fully grown (as defined in this paper) at some time during the first 6 months of life. In the present investigation the otter cubs which had reached an advanced stage of dentitional change were probably between 3 and 6 months old. Most likely they were all approximately the same age when killed, perhaps in their fourth month of life.

### Age in relation to the numbers of incremental cementum lines and to cranial development

Because, in the European otter, the dark-staining cementum layers are situated close together and are wavy, it is particularly important that the tooth sections made are thin to

prevent line overlap. Probably sections less than 16 micron in thickness would be preferable (see GRUE and JENSEN 1979). For the same reason, it is most important to choose sections made along the centre-line of the tooth where the sectioning plane is at right-angles to the dark staining layers.

The observed season of formation of the cementum lines in the Norwegian otters, largely corresponds with that recorded for Danish otters (GRUE and JENSEN 1979), starting in February. As pointed out by KLEVEZAL' and KLEINENBERG (1969) the first stage of deposition of dark-staining or of light-staining cementum is not easily observed. Probably the observed period is somewhat delayed in relation to the true period of line formation.

The age at which the first layer of cementum is laid down is likely to vary with the season at which a particular cub is born, since line formation seems to be a seasonal phenomenon whereas births are not. GRUE and JENSEN (1979) have suggested that this age varies from  $\frac{1}{2}$  to  $1\frac{1}{2}$  years-old. In accordance with the findings for the North American river otter (STEPHENSON 1977) the broad inner layer of light-staining cementum is probably due to a more rapid rate of deposition rather than to a long time elapsing before the first dark-staining layer is laid down. The first line can at least be expected to have developed after the permanent teeth are fully grown and before the cub is 2 years-old.

The differences in the age estimates given by the skull criteria and the cementum line counts can, with a single exception (1 %), be explained by the expected age difference at the time when the first dark-staining incremental line is formed. Thus, the results obtained by these two methods are not contradictory. However, fairly wide limits exist within which individual variation may occur.

The age estimates based on skull development derives in the present paper from a combination of several skull criteria. This procedure has the advantage over age estimates based on single skull traits that combination of several traits tends to reduce the variance of the estimates.

Even if the age criteria used have not yet been tested on European otters of known age, there are fairly strong indications that they can provide useful information about the ages of otters. The seasonal formation of the cementum lines in the teeth of both Danish and Norwegian otters indicates that, for the European otter, one dark-staining line is usually formed annually. Thus it seems reasonable to assume that the following approximate relationship exists between the number of cementum lines and age:

Number of incremental lines	Age interval in years	Median of age interval in years
n	$(n - \frac{1}{2}, n + 1\frac{1}{2})$	$n + \frac{1}{2}$

Some difficulties in interpretation may arise due to the irregularity of the line pattern in the teeth of individual otters. Even so, the number of primary lines can usually be determined in most cases. For some studies that include young otters, line counts may be too indiscriminate to be used as the sole estimate of age. Both the dental and the cranial development may then need to be used to achieve more accurate estimates of the ages of young otters. It is an advantage that both these morphological characteristics undergo rapid changes during the early period of life. The relationship between the skull criteria and age is less influenced by the season of birth of the individual otter than can be expected to be the case for the incremental cementum lines. However, the accuracy of this ageing technique can only be known when the teeth and skulls of a sufficient number of otters of known age have been studied.

### Acknowledgements

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### Zusammenfassung

#### *Altersbestimmung bei europäischen Fischottern, Lutra lutra lutra*

Kadaver von wildlebenden europäischen Fischottern aus Norwegen wurden nach verschiedenen Methoden in ihrem Alter bestimmt. Die Anzahl der Wachstumslinien im Zahnzement, die in histologischen Zahnschnitten dunkel gefärbt wurden, das Entwicklungsstadium des Schädels und der Entwicklungsgrad der permanenten Zähne wurden verglichen.

Auf der Grundlage in dieser Weise gewonnener Information und auf der Basis von früheren Berichten über die Entwicklungsstadien von zahmen europäischen Fischottern, von nordamerikanischen Fischottern (*Lutra canadensis*) und von afrikanischen Kapottern (*Aonyx* sp.) mit annähernd bekanntem Alter, wird festgestellt, daß die Anzahl der Wachstumslinien im Zahnzement vermutlich der Anzahl der Lebensjahre entspricht.

Die jahreszeitabhängige Ausbildung von Wachstumslinien, und die Tatsache, daß Geburten – zumindest in Teilen des Verbreitungsgebietes des europäischen Fischotters – zu allen Jahreszeiten vorkommen können, verursachen vermutlich Variationen im Verhältnis zwischen Alter und Anzahl der Wachstumslinien. Die Bestimmung der Schädelentwicklung kann aber für Fischotter bis zum Alter von 2 Jahren eine genauere Altersabschätzung liefern. Der Zahnwechsel ist vermutlich schon nach 6 Monaten beendet und kann deshalb bei sehr kleinen Jungen zusätzlich Information über das Alter liefern.

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