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# Abstract

In forensic cases it can be challenging to differentiate between a live birth and a stillbirth. Few methods are accepted which become less precise with an increased post mortem interval. Therefore, it is aimed to evaluate the utility of stable isotope analysis and tooth histology to detect stillbirth. Up to now 8 archaeological "mother-infant-pairs" (MIP) were screened for their stable isotope composition of nitrogen ( $\delta^{15}N$ ) and carbon ( $\delta^{13}C$ ) and tooth histology was performed in 2 cases. A slight trend in the stable isotope data is visible however, further samples are required to fully evaluate their utility.

## 2 Introduction

In forensic cases that involve remains of a neonate it is necessary to established if the child was born alive (Fig. 1). Forensic experts have limited methods and with increased decomposition the evaluation becomes more difficult.

One method is the examination of a neonatal line (NNL). It can be observed in the enamel of deciduous teeth and is related to labor. To be clearly visible the child has to survive birth at least 7 to 10 days (1, 2). Stable isotopes are used in archaeological studies in the context of weaning but to date no study has addressed the identification of stillbirth. The aim is to investigate if the stable isotopes  $\delta^{15}N$  and  $\delta^{13}C$  could provide additional information about the survival of birth.





Fig. 2: Different finding situations of MIP, circled in red the fetal/ neonatal remains. A: Term fetus (CTT 2015; 10LM to 2Mt ex utero) in the pelvic cavity and between the femora; B: Preterm neonate (DOM 2011; ~7LM) lying at the right arm of the female; C: Preterm fetus (DOM 2014; ~6LM) lying within the pelvic cavity.

#### Results

- The collagen quality criteria were fulfilled in 7/8 cases.
- The deviation of  $\delta^{15}N$  and  $\delta^{13}C$  for each of the 7 MIP was calculated.
- Less deviation is seen for  $\delta^{13}$ C values than for  $\delta^{15}$ N values (Fig. 3).

Fig. 1: Classification of stillbirth depending on the time of gestation and the development of the fetus. Several factors can lead to a stillborn child including the mothers health status (based on <u>8</u>).

## ③ Materials & Methods

#### Isotope ratio mass spectrometry:

- 8 MIP from medieval burial sites where the fetus/ neonate was found in close association with a female (Fig. 2; Table 1).
- Previous study: 15 neonatal and 2 female remains from a roman series; no association between neonates and adults is given (3).

#### Tooth histology:

Tooth histology could be performed for 5 individuals (both series).

Sample ID	Bone sample	Age ± [LM]	Developmental stage	Collagen	Histology	Burial position in relation to female
DOM 2011	femur + humerus	7	preterm neonate	~	x	arm
DOM 2013	femur	10	coffin birth?/ term fetus	$\checkmark$	$\checkmark$	between femora and in pelvis
DOM 2014	femur + humerus	6	preterm fetus	~	x	pelvis
CTT 2015	femur	10-2Mt	coffin birth?/ term fetus	~	~	in pelvis and between femora
WTW-2	femur	8-9	preterm neonate	x	x	arm
WAA 84	femur	10	term neonate	$\checkmark$	x	thorax
WAK 101	ribs	9	preterm fetus	$\checkmark$	x	pelvis
WAK 104	ribs	10	term fetus	$\checkmark$	x	pelvis

- The roman series indicate the beginning of a breastfeeding signal for 14/15 neonates (Fig 4).
- No NNL was observed on the examined teeth (Fig. 5-6).



Fig. 3: Empty symbols represent preterm/ term neonates; filled symbols indicate preterm fetuses; striped symbols indicate term fetuses, possible coffin births/ stillbirths. LM = lunar months. A: Deviation of  $\delta^{13}$ C of 7 MIP. **B**: Deviation of  $\delta^{15}$ N of 7 MIP.





Table 1: Information for the fetus/ neonates of the MIP from medieval contexts. The age was estimated based on skeletal elements. LM = Lunar months; Mt = months ex utero.



Fig. 4:  $\delta^{13}$ C against  $\delta^{15}$ N values for the roman neonates & average ± SD; female average (black line) ± SD of the female values (dotted lines). Red circle = tooth histology was performed.

#### Discussion (5)

The preliminary data might indicate that  $\delta^{13}C$  is more indicative for the survival of birth than  $\delta^{15}N$ . This is contrary to the present knowledge (4-7) which is presented by the roman series. Problematic for this series is that the actual mothers are not known. An assumption about the relation of  $\delta^{13}$ C and  $\delta^{15}$ N data between some females and neonates is therefore difficult.



Fig. 5: Different micrographs and close-ups of a modern deciduous incisor with NNL indicated by arrows (a) and asterisks (b-c) (2).

Fig. 6: Roman series. Micrographs of an incisor (transmitted light (A) & phase contrast (B-C)), white asterisk = dentin-enamel-junction; B & C: Close-up of A/ B at expected site of NNL; D: SEM image at expected site of NNL. No NNL is observable.

## Conclusion

The necessity to develop additional methods to detect stillbirth is given: Stillbirth is still ten times more often recorded in medical records than sudden infant death syndrome in high income countries  $(\underline{8}, \underline{9})$ . The discovery of discharged neonates is frequently reported. With a sufficient sample size archaeological material might provide fundamental information for further in depth studies.

The fact that no NNL was observed requires further investigation, especially since no exhaustive study regarding the appearance of a NNL is present in the literature.

There are several limitations to this study: To date the sample size is small and archaeological samples have to be used as a proxy. Further studies should be done to generally evaluate the applicability of stable isotopes and tooth histology in the context of stillbirth in forensic cases.

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