

CONVENTION ON INTERNATIONAL TRADE IN ENDANGERED SPECIES  
OF WILD FAUNA AND FLORA

---



Sixteenth meeting of the Conference of the Parties  
Bangkok (Thailand), 3-14 March 2013

Implementation of the African Elephant Action Plan

RESEARCH PROJECT ON THE DETERMINATION OF AGE AND  
THE GEOGRAPHICAL ORIGIN OF AFRICAN ELEPHANT IVORY

1. This document has been submitted by Germany\* in relation to agenda item 53.2.1 on *Monitoring of illegal trade in ivory and other elephant specimens – Report of the Secretariat*.
2. Germany considers this research project as a significant contribution to the implementation of the African elephant action plan (CoP15 Inf. 68), particularly to Objective 1: Reduce illegal killing of elephants and illegal trade in elephant products (see also SC 61 Inf. Doc.4).
3. The current status of the research project and its major findings will be presented to the CITES community during a side event at CoP16.
4. A practical application to the determination of origin of seized ivory is attached in **ANNEX 1**. Brief descriptions of the scientific background are attached to this information document as **ANNEX 2** (Methodology of the project part 'Determination of the origin of ivory' and Methodology of the project part 'Determination of the age of ivory').
5. Germany is particularly thankful to the continuous support of Botswana, Burkina Faso, Malawi, Mozambique and South Africa which supported this project with the provision of samples from their ivory stockpiles. Several European museums and collections as well as numerous trophy hunters gratefully provided ivory samples. The BMU appreciates the cooperation with the International Council for Game and Wildlife Conservation (CIC) and TRAFFIC which actively promoted this project.

---

\* *The geographical designations employed in this document do not imply the expression of any opinion whatsoever on the part of the CITES Secretariat or the United Nations Environment Programme concerning the legal status of any country, territory, or area, or concerning the delimitation of its frontiers or boundaries. The responsibility for the contents of the document rests exclusively with its author.*

# Isotope databases for the determination of age and provenance of elephant ivory

Stefan Ziegler<sup>a</sup>, Claudia Denkl<sup>b</sup>, Karin Hornig<sup>b</sup>, Robert Schupfner<sup>c</sup>

<sup>a</sup> WWF Germany, Reinhardtstraße 14, 10117 Berlin, Germany

<sup>b</sup> Federal Agency for Nature Conservation, Konstantinstraße 110, 53179 Bonn, Germany

<sup>c</sup> University of Regensburg, Faculty of Chemistry, Universitätsstraße 31, 93053 Regensburg, Germany

## Necessity and background

Poaching of African elephants and illicit trade in ivory has accelerated in some African sub regions during the recent years. Trafficking in this illegal wildlife products today can be considered professionalized as never been seen before. Well-organized and heavy-armed criminal bands do not only endanger elephant populations but also constitute a threat to regional stability, territorial integrity and sustainable social and economic developments of the countries concerned. International law enforcement, cross-border cooperation and effective forensic methods to uncover the structures and pathways of ivory smuggling and to differentiate illegal from legal ivory in trade are urgently needed.

Nowadays, *Loxodonta africana* can still be found in 37 range states in Sub-Saharan Africa, but certain populations, mostly in West and Central Africa, hardly exceed a few hundred individuals which are highly threatened by increased poaching. At the time of the last continent wide assessment in 2007, the African elephant population was calculated to be at least 472,000 individuals, with possible numbers exceeding 690,000 elephants. One of the key findings in the latest African Elephant Status Report (BLANC ET AL., 2007) is that elephant numbers in East and Southern Africa are increasing by 4% annually. These two sub regions hold 88% of all of the 'definite' and 'probable' elephant numbers in the African Elephant Database.

Exact methods for the determination of age and geographical origin are essential to meet the unsolved problem of ivory smuggling and can help to avoid the intermixing of legal and illegal ivory, if decisions for a restricted legal trade will be taken in future. Long-term preservation of the constantly declining elephant population of Western and Central Africa will only be possible with a control mechanism that helps to identify the age and geographical provenance of confiscated ivory. Therefore, the African Elephant Action Plan by the African Elephant range states (CoP15 inf. 68) highlights the need for improved law enforcement and management by identifying the origin of seized ivory by using relevant analytic techniques (Activity 1.4.3. of Objective 1).

At present a supporting instrument that meets court standards is not available for the CITES member states. Therefore, the development of a very exact method for age determination and the set-up of a database for the identification of the origin of ivory will help to better focus enforcement and conservation measures on an international level.

The German Federal Agency for Nature Conservation responded to this need and initiated the above mentioned project, in co-operation with its executing partners, the WWF Germany and two German Universities (University of Regensburg, University of Mainz). The project was funded by the German Federal Ministry of Environment, Nature Conservation and Nuclear Safety (BMU) and started in July 2010 and has two parts:

1. The determination of the origin of ivory
2. The determination of the age of ivory.

The second part was completed in 2012, whereas the first part of the project has been extended until December 2014.

## 1. The determination of the origin of ivory

### 1.1 The use of isotopic fingerprinting for tracking the provenance of ivory

Forensics can play an important role in the investigation of wildlife crime through identification and profiling of ivory (CITES, 2012). Stable isotope analysis is a technique that is based on the fact that stable isotope signatures in animal tissues reflect those of local food webs and geology. Therefore it can be used for tracing the origin or migration of wildlife (PETERSON and FRY, 1987; TIESZEN and BOUTTON, 1988; TIESZEN, 1991; MICHENER and SCHELL, 1994; HOBSON, 1999). This principle has also been applied to determine the source of ivory (VAN DER MERWE ET AL., 1988; VOGEL ET AL., 1990; CERLING ET AL., 1999, 2003, 2004, 2007).

In an attempt to elaborate the predictive ability of stable isotope signatures in ivory, WWF Germany, in cooperation with the International Centre of Ivory Studies (INCENTIVS) at the University of Mainz, Germany, had been contracted by the Federal Agency for Nature Conservation (BfN) to develop a methodology with which the determination of the geographical origin can be tested.

## 1.2 Reference database for the geographical origin of elephant ivory

In total, 606 ivory samples from 24 African and six Asian elephant range states were provided between 2009 and 2012 by protected areas and CITES Management Authorities in African elephant range states, European museums and collections, trophy hunters and with the assistance of the International Council for Game and Wildlife Conservation (CIC). A large part of the samples (360 pcs.) comes from Botswana, Burkina Faso, Democratic Republic of the Congo, Malawi, Mozambique and South Africa.

Ivory fragments of at least 30 mg were taken from different positions at the very proximal end of the tusk by using a small hand saw or a pincer. Thus, it was assured that the isotopic signal reflects the environment where the animal died. The methodology, measuring five different stable isotope ratios (Carbon  $\delta^{13}\text{C}$ , Nitrogen  $\delta^{15}\text{N}$ , Oxygen  $\delta^{18}\text{O}$ , Hydrogen  $\delta^2\text{H}$ , Sulphur  $\delta^{34}\text{S}$ ), is described in ANNEX 2 of this document.

The results suggest that the combination of several isotopic parameters has the potential to provide predictable and complementary markers for estimating the origin of seized elephant ivory. Additionally, the predictive power of stable isotope signatures in ivory was improved through multiple isotope testing and multivariate statistics. The database for ivory, which in the near future will be made publicly available via the internet ([www.ivoryid.org](http://www.ivoryid.org)), can be used as a reference to predict the provenance of ivory of unknown origin. With this new approach it is also possible to distinguish between ivory from elephant populations listed in Appendix I and Appendix II. The reference database was cross-validated and test runs with seized ivory were carried out. Up to 92.2% of all ivory reference samples deriving from elephant populations listed in Appendix I of CITES were correctly assigned to their region of origin (see ANNEX 2, Table 1). Approximately 13.5% of all ivory samples from Botswana, Namibia, South Africa and Zimbabwe, whose elephant populations are listed in Appendix II of CITES, were misclassified as Appendix I populations. However, we are confident that this so-called false positive rate can be reduced if more reference samples are made available, particularly from elephant range states which are still under-represented in the database<sup>1</sup>. Furthermore, it was found out that the accuracy of the methodology is still given if only three (instead of five) isotopic markers Carbon  $\delta^{13}\text{C}$ , Nitrogen  $\delta^{15}\text{N}$  and Sulphur  $\delta^{34}\text{S}$  were measured, which may serve as a cost efficient alternative (see ANNEX 2, Table 1).

It was also possible to identify alleged poaching hot spots at country level (see ANNEX 1). With this approach wildlife authorities will be in a better position to direct law enforcement efforts more specifically and distinguish legally derived ivory from illegally sourced ivory along the production and marketing chain.

## 2. The determination of the age of ivory

### 2.1 General information about the research methods

The purpose of this part of the project is to validate a new method for the determination of age, based on isotope analysis. Additional to the existing methods using the radiocarbon ( $^{14}\text{C}/\text{C}$ ) testing the new method will combine it with the analysis of a variety of nuclides ( $^{90}\text{Sr}/\text{Ca}$ ,  $^{228}\text{Th}$ ,  $^{232}\text{Th}$  and others). Even a very precise dating method like  $^{14}\text{C}$  dating shows certain limitations. Though the content of  $^{14}\text{C}$  can be determined precisely consuming low amounts of material dating is not unambiguous at certain periods of time. This is due to the shape of the so called „bomb curves“. With the conventional method the results can be ambiguous, with the year of death not clearly defined (e. g. 1962 and 1980 for the same sample). Combining this method with the determination of strontium and thorium the result is more accurate.  $^{90}\text{Sr}$  is produced at the nuclear fission and was distributed during the global nuclear fallout within the food chains in the sixties of the last century. As alkaline earth element  $^{90}\text{Sr}$  behave very similar to calcium and is therefore transferred to calcium containing tissues like ivory.  $^{90}\text{Sr}$  can be determined until presence due to its long half life time of about 29 years. The reason is that due to nuclear testing a significantly increased value of  $^{90}\text{Sr}/\text{Ca}$  is typical for a death between 1960 and 1970. Lower values indicate a death before 1960 or after 1980. A value below the detection limit

---

<sup>1</sup> Priority African elephant range states from which samples are required to increase the accuracy of the database are: Chad, Kenya, Tanzania, and Zambia (in alphabetical order). The selection follows criteria, such as area of elephant habitat, no. of subpopulations and no. of climate classes (Ziegler et al., 2011).

indicates a death before 1955.  $^{228}\text{Th}$  and  $^{232}\text{Th}$  are naturally occurring radionuclides. If the time of death is dated back before 1960 the ratio of the two nuclides is 1:1; if the death was approximately 1990 the ratio is much higher than 1:1. It could be shown, that the additional determination of  $^{90}\text{Sr}$  and  $^{228/232}\text{Th}$  increases the certainty and the validity of the resulting time period. Therefore by combining these analyses the time of death of the elephant can be determined with a high degree of certainty. This makes the method very precise and extremely reliable.

The University of Regensburg had been contracted by the Federal Agency for Nature Conservation (BfN) to develop a more precise methodology for the determination of the age of ivory.

All results of this project can be found in the citations in the reference list.

## ANNEX 1: Seizures

### 1. Application of the method to seized ivory

By coincidence, German and Chinese customs seized illegal ivory consignments and made samples available to the project. In order to test the new methodology and to determine the provenance of the seized ivory, the isotopic signature of the samples were analysed and assignment simulations run.

For the testing purpose, the ivory reference database was partitioned into regional training categories: (i) Central, (ii) East, (iii) Southern, and (iv) West, based on the regional structure of the African Elephant Database (Blanc et al., 2007). Isotopic data deriving from Asian elephant ivory formed a fifth class: (v) Asia. In a second run, the reference database was partitioned into 24 African and 6 Asian elephant range states from which at least one sample is stored in the database. During the testing process, the class of each test sample was individually predicted and assigned one regional and country class label. In applying the  $k$ -NN rule (see Annex 2), the predicted class of the test sample is set equal to the most frequent true class among the  $k$  nearest training samples.

#### 1.1 Case 1: Seizure in Leipzig, Germany

In spring 2011, the customs department at the DHL freight centre in Leipzig, Germany seized a shipment of total 35 kg ivory (674 pieces), declared as plastic goods, which was posted in Nigeria and designated for import into Hong Kong. Out of this consignment, 40 ivory bracelets were chosen for isotope analysis to find out from which geographical area the carved ivory might derive from. Each ivory bracelet was defined as one test sample and treated individually.

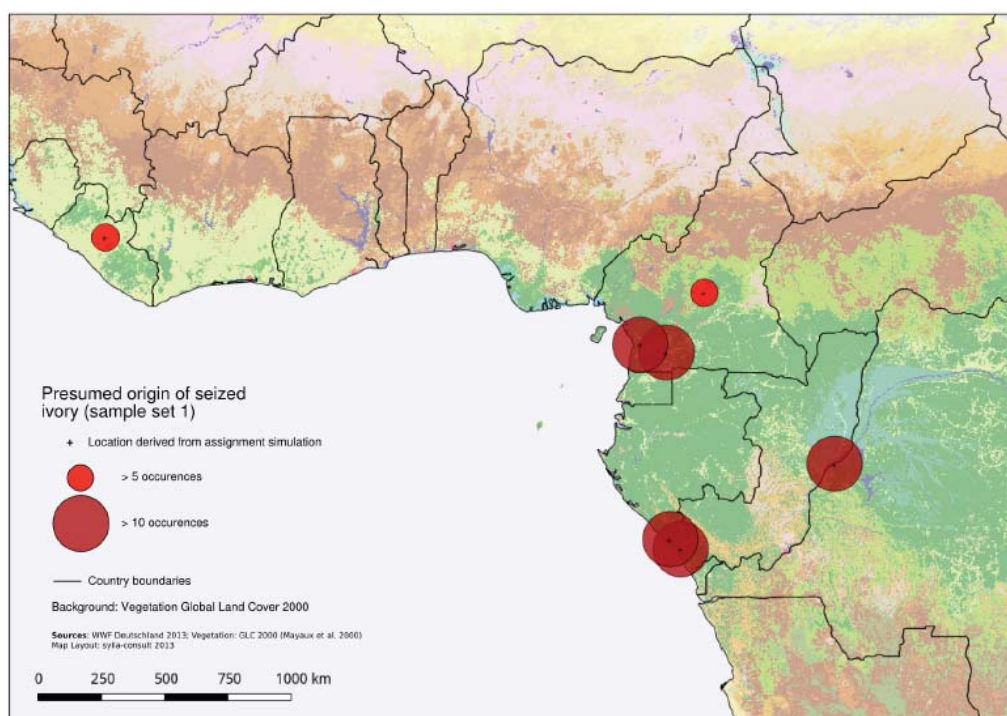


Figure 1: Occurrence of reference sites from 40 assignment simulations from seized ivory in Leipzig, Germany in early 2011.

The assignment simulations provided overwhelming evidence that the majority of the seized ivory derives from elephant populations in Central Africa. 38 out of 40 test samples were assigned to Central Africa. Simulations at country level assigned 25 ivory bracelets to Cameroon. This is in so far remarkable because the training data set only contains 11 references samples from Cameroon. Figure 1 depicts those localities which occur most frequently in all 40 assignment simulations. We only display reference sites which occur at least six times. The frequency of locations in Cameroon and Republic of Congo is evident, and we conclude with high probability that the bulk of ivory that was seized in Leipzig does originally derive from this part of Africa.

#### 1.2 Case 2: Seizure in Tianjin, China

This case refers to a shipment exported illegally from Dar es Salaam, Tanzania. It was seized in China in early 2012, when the customs department in Tianjin discovered 931.7 kg (363 pieces) of ivory. Out of this seizure 10

samples were selected and analysed to find out from which geographical area the carved ivory might derive from. Each ivory piece was defined as one test sample.

The simulations assigned five samples to Tanzania, four samples to Malawi and one sample to Mozambique with the conditional probability of at least 60 percent. According to our simulations the provenance area covers a vast geographical range which stretches more than 1,500 km throughout East and eastern Southern Africa (Figure 2). This is, however, not surprising for a relatively large consignment of illegal ivory. The assignment of 50 percent of the Chinese samples to Tanzania is remarkable because the ivory database only contains 15 reference samples from this elephant range state, which has one of Africa's largest elephant range areas.

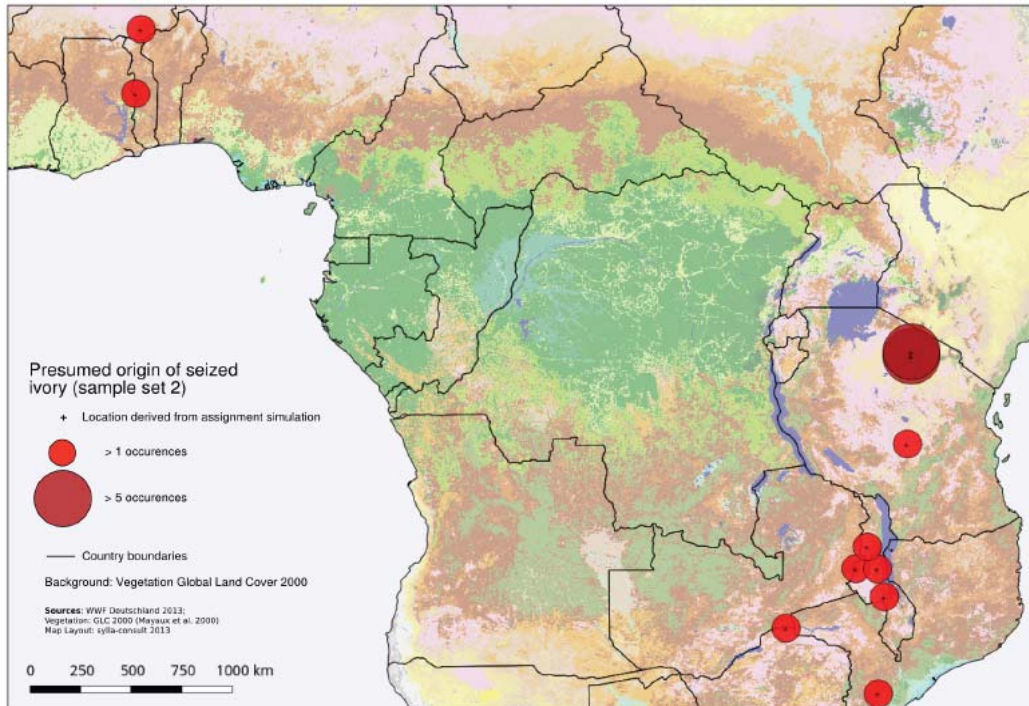


Figure 2: Occurrence of reference sites from 10 assignment simulations from seized ivory in Tianjin, China in early 2012.

## ANNEX 2: Scientific background

### 1. Methodology of the project part 'Determination of the origin of ivory'

#### 1.1 Isotope analysis

All samples were analysed at the Agroisolab Facility for Stable Isotope Research in Jülich, Germany. The samples were pulverised in a ball mill with the grinding jar continually cooled with liquid nitrogen at  $-96^{\circ}\text{C}$ . Afterwards the samples were cleaned with dichloromethane for six hours to extract non-polar substances, such as tissue fat, and then allowed to air dry at  $60^{\circ}\text{C}$  for 36 hours. Samples were stored in an exsiccator to avoid humidification. From each sample between 1 mg and 4.5 mg of ivory was sub-sampled for stable isotope analyses. Powdered ivory was loaded into 4 x 6 mm tin capsules for  $^{13}\text{C}/^{12}\text{C}$ ,  $^{15}\text{N}/^{14}\text{N}$  and  $^{34}\text{S}/^{32}\text{S}$  measurement. Silver capsules (3.3 x 5 mm) were used for the  $^{18}\text{O}/^{16}\text{O}$  and  $^2\text{H}/\text{H}$  analysis. We used continuous flow isotope ratio mass spectrometers (IRMS) and measured five different stable isotope ratios ( $\delta^{13}\text{C}$ ,  $\delta^{15}\text{N}$ ,  $\delta^{18}\text{O}$ ,  $\delta^2\text{H}$ ,  $\delta^{34}\text{S}$ ).

#### 1.2 Statistical analysis

The five stable isotopic measurements of the data can be described as a vector in a five-dimensional space. The Euclidian distance between vectors was calculated by using the open source software packet 'R' and formed the basis for the statistical model. We applied the nearest neighbor (NN) rule, which was first developed by Fix and Hodges (1951, 1989) as the pattern classification algorithm. The basic rationale for the NN rule is that samples with low Euclidian distance belong to the same class meaning that those samples are likely derived from the same place of origin. Due to the huge spatial range and the ecological heterogeneity of the African and Asian elephant habitat we assumed that the pattern classes do overlap to some extent, rendering the NN rule sub-optimal. Thus, we also performed the  $k$ -nearest neighbor ( $k$ -NN) rule that classifies the vector to the class that appears most frequently among its  $k$  nearest neighbors and performed the rule with  $k=5$  (STONE, 1977).

In order to evaluate the performance of the predictive model we applied cross-validation across all reference samples, which involved the determination of classification accuracy for multiple partitions of the input samples used in training. The partition of training categories followed the inclusion of both recent elephant species in the CITES Appendices. We built two classifiers, representing elephant populations, which are currently listed on Appendix I and Appendix II in CITES and performed the leave-one-out cross-validation based on the  $k$ -NN rule. That way we evaluated to what extent the combination of different stable isotopes increases the conditional probability with which reference samples were correctly assigned. The combination with the best performance in terms of correct assignments was used to calculate sensitivity and the false positive rate of the ivory reference database as a predictor to distinguish the provenance of ivory. A perfect predictor would be described as 100% sensitivity (i.e. all ivory samples from Appendix I populations are assigned to Appendix I populations) and a false positive rate of zero (i.e. no samples from Appendix II populations are assigned to Appendix I populations).

#### 1.3 Results

Stable isotope values of elephant ivory showed a considerable range of variation. We also found that combining isotopes increased the conditional probability with which reference samples were correctly assigned. The exception from this rule is the isotopic ratio of  $\delta^{34}\text{S}$ , which performs well in distinguishing Appendix I from Appendix II elephant populations. However, the false positive rate of  $\delta^{34}\text{S}$  is fairly high which makes this isotope marker less practical if no additional markers are applied. The false positive rate can be reduced through the combination of stable isotopes which has direct cost implications (see Table 1). The lowest false positive rate in our cross-validation exercise yields the combination of all five stable isotopes ratios. However, though the false positive rate for the combination of carbon, nitrogen and sulphur is one percent higher, focusing on these isotopic markers might be a cost efficient alternative without compromising the accuracy of the methodology.

*Table 1:* Results of isotope combinations with cross-validation across all reference samples which involved the determination of classification (populations of Appendix I vs. Appendix II).

**Sensitivity:** number of correctly assigned Appendix I samples / total Appendix I samples;

**False positive rate:** 1 – number of correctly assigned Appendix II samples / total Appendix II samples;

**Accuracy:** [number of correctly assigned Appendix I samples + number of correctly assigned Appendix II samples] / number of reference samples.

Costs are estimated prices for the analysis at a certified commercial laboratory within the European Union.

Isotope combination	Sensitivity (%)	False positive rate	Accuracy (%)	Estimated costs (US\$)
$\delta^{13}\text{C}$	74.3	0.253	74.5	65
$\delta^{15}\text{N}$	75.0	0.354	71.1	65
$\delta^{34}\text{S}$	92.2	0.275	84.8	130
$\delta^{18}\text{O}$	68.2	0.315	68.4	130
$\delta^2\text{H}$	80.1	0.556	66.7	130
$\delta^{13}\text{C}, \delta^{15}\text{N}$	80.7	0.213	80.0	110
$\delta^{18}\text{O}, \delta^2\text{H}$	77.4	0.545	65.4	225
$\delta^{13}\text{C}, \delta^{15}\text{N}, \delta^{18}\text{O}, \delta^2\text{H}$	82.1	0.202	81.2	360
$\delta^{13}\text{C}, \delta^{15}\text{N}, \delta^{34}\text{S}$	89.5	0.146	88.0	240
$\delta^{18}\text{O}, \delta^2\text{H}, \delta^{34}\text{S}$	91.9	0.236	86.1	360
$\delta^{13}\text{C}, \delta^{15}\text{N}, \delta^{18}\text{O}, \delta^2\text{H}, \delta^{34}\text{S}$	90.5	0.135	89.0	450

## 2. Methodology and Results of the project part ‘Determination of the age of ivory’

### 2.1 Isotope Analysis

First the well suited analysis methods were adapted to the sample material and the analytical boundaries. Ivory of elephants like *Loxodonta africana*, *Loxodonta cyclotis* oder *Elephas maximus* are suited rather well because of the sufficient high contents of carbon and calcium. But the analysis methods must be executed as efficient and undisturbed as possible realizing very low limits of detection. It is necessary to separate and concentrate these elements without significant losses first and purify them from possibly interfering radionuclides like  $^{40}\text{K}$  and  $^{137}\text{Cs}$ . Previously the sample must be incinerated and after the radiochemical purification it must be prepared to the most suited method to detect the nuclear radiation. In addition to this as few sample as possible should be destroyed. Therefore the analysis methods have been connected. Afterwards analyses of certified standard materials demonstrated that the coupled determination methods realize the limits of detection being necessary to determine very low values of activities. During a further step the correlation of radionuclide content versus time after death assumed has been certified experimentally. Further findings are the bombing curve of  $^{14}\text{C}/\text{C}$  is nearly the same no matter analysing ivory or wood. As expected the bombing curve of  $^{90}\text{Sr}/\text{Ca}$  of ivory is of similar shape as of human bone tissue. It is remarkable that the specific activity of  $^{90}\text{Sr}/\text{Ca}$  is much higher in ivory compared to human bone tissue. The shape of his curve is similar to that of  $^{14}\text{C}/\text{C}$  resulting in ambiguously interpreting  $^{90}\text{Sr}/\text{Ca}$  values. An unambiguous dating is enabled at following findings. If  $^{90}\text{Sr}/\text{Ca}$  is lower than 0,003 Bq/g Ca the death occurred before about 1958. At values above 0, 4 Bq/g Ca the time of death can be assumed to be occurred during about 1960 and 1970. If the interpretation is ambiguous analysing thorium is a further possibility. With increasing time from death to analysis the ratio of activity of  $^{228}\text{Th}/^{232}\text{Th}$  decreases from about 40 to about 1 if death occurred before 1968. The radionuclide analyses of a complete ivory tusk resulted in the determination of a representative sample site at the stump. The determination of further radionuclides like  $^{228}\text{Ra}$  is able to support the dating. Dating tissues from tetrapod vertebrates with Calcium content similar to bone tissues or ivory or antlers the determination of isotope profiles can be well suited. In contrast to this no further information is obtained analysing materials as horn of rhinoceros or tortoise shell or coat or claw, because of their low calcium content. At last, the dating method of determining isotope profiles has been applied to undated ivory samples to prove its suitability for daily use. To fulfil the qualification a well-appointed radio analytical laboratory must be available in addition to a well trained staff and low-level nuclear detection devices like Liquid Scintillation Counting (LSC) or Acceleration Mass Spectrometry (AMS) and low-level- gas-flow-beta-counter and alpha-spectrometry for example with silicon surface barrier junction detector. The time required to perform a complete isotope profile is about one day per sample for the chemical digestion one further day per element for radiochemical purification procedures including sample preparation and from about three days up to about ten days per sample and element for counting. Altogether the time required is estimated to about four to five weeks. The time for processing may be shortened if the results of  $^{14}\text{C}/\text{C}$  are unambiguous. In this case further radionuclide analyses may be unnecessary. The sample amount needed to determine the complete isotope profile is about ten grams and must not decrease below eight grams. Determining  $^{14}\text{C}/\text{C}$  only few grams are required applying LSC and even much less applying AMS. The costs thereby incurred are about 550 € per sample (without value added tax) determining the complete isotope profile and about 350 € per sample (without value added tax) determining  $^{14}\text{C}/\text{C}$  only.



## 2.2 Results

The developed and optimized method was applied to ivory samples of unknown age to gain experience and data. Up to now 34 ivory samples of unknown age were already analysed. An unambiguous result was obtained for 32 samples by analysing  $^{14}\text{C}$ ,  $^{90}\text{Sr}$  and  $^{228}\text{Th}/^{232}\text{Th}$ . Only for 2 samples the results of the three single parameters are not in agreement with each other. Therefore it was not possible to conclude an unambiguous result. Table 1.1 shows the data of two samples (as an example), for which unambiguous results were obtained.

Table 2.1: Results of ivory samples of unknown age.

sample	periodofdeath $^{14}\text{C}$	a ( $^{90}\text{Sr}$ ) / Bq/g Ca	ratio $^{228}\text{Th}/^{232}\text{Th}$	resultofdating
8000027	1963 – 1964 1973 – 1978	0.063 ± 0.003	1.6 ± 0.5	1963 – 1964
8000043	1963 – 1964 1973 – 1978	0.016 ± 0.002	7.3 ± 1.6	1973 – 1978

For sample 8000027 the earlier time period (1963 – 1964) of the both possibilities according to the  $^{14}\text{C}$  analysis is the correct one, because the value of the ratio of  $^{228}\text{Th}/^{232}\text{Th}$  is close to one and the concentration activity of  $^{90}\text{Sr}$  a( $^{90}\text{Sr}$ ) relatively high (cf. data of samples of known age in the 3<sup>rd</sup> progress report). For sample 8000043 the later time period (1973 – 1978) of both possibilities represents the result of dating. The time of death of this animal cannot be so far in the past, according to the high ratio of  $^{228}\text{Th}/^{232}\text{Th}$  of 7.1. The value of the concentration activity of  $^{90}\text{Sr}$  a( $^{90}\text{Sr}$ ) is also in agreement with the chosen time period of death (cf. 3<sup>rd</sup> progress report).

In Table 2.2. the data of two samples are shown, for which the resulting values of  $^{14}\text{C}$ ,  $^{90}\text{Sr}$  and  $^{228}\text{Th}/^{232}\text{Th}$  are not in agreement with each other.

Table 2.2: Results of ivory samples of unknown age

sample	periodofdeath $^{14}\text{C}$	a ( $^{90}\text{Sr}$ ) / Bq/g Ca	ratio $^{228}\text{Th}/^{232}\text{Th}$	resultofdating
8000053	1963 – 1964 1971 – 1975	0.020 ± 0.002	11.4 ± 2.8	not unambiguous
8000062	1964 – 1965 1966 – 1970	0.057 ± 0.006	6.2 ± 0.9	not unambiguous

Two different time periods are obtained from the values of the  $^{14}\text{C}$  analysis of both samples. The time periods are between 1963 and 1975 for sample 8000053 and between 1964 and 1970 for sample 800062. The ratio of  $^{228}\text{Th}/^{232}\text{Th}$  of sample 8000053 has a high value of 11.4 which is an evidence that the possible time of death is later than 1990 (3<sup>rd</sup> progress report). Further it is noticeably, that the value of the concentration activity of  $^{232}\text{Th}$  is located at the lower range (95% confidence level, N = 27 values without outliers). The value of the concentration activity of  $^{90}\text{Sr}$  in this sample speaks in favour for a time period about 1955 or a time period after 1990 (cf. 3<sup>rd</sup> progress report). So the earlier time period of the  $^{14}\text{C}$  analysis could be possible, but also the time period which resulted from the thorium ratio. No time period can be chosen which is in agreement with all single results. The value of the thorium ratio of sample 8000062 results in a time period after 1985 (cf. 3<sup>rd</sup> progress report) and is not in agreement with the two time periods obtained from the  $^{14}\text{C}$  analysis. The value of the concentration activity of  $^{90}\text{Sr}$  is in agreement with the time periods of  $^{14}\text{C}$  and also with the time period of the thorium ratio. So also in this case no unambiguous result is possible regarding to the three different parameters. Overall it was possible to achieve an unambiguous result using this method for 32 of 34 samples. Only for two samples it was not possible to get an unambiguous result using the data of all three single parameters. These are possible reasons:

- The different uptake of radionuclides with food, especially the alkaline earth metals  $^{90}\text{Sr}$  and  $^{228}\text{Ra}$ . This can be neglected for  $^{14}\text{C}$ , because the determined value considers the  $^{14}\text{C}$  activity related to the mass of carbon. The concentration activity of  $^{90}\text{Sr}$  and the ratio of  $^{228}\text{Th}/^{232}\text{Th}$  are considerably affected by the uptake of the regarding nuclides, so bigger differences seem to be feasibly. Meanwhile the indication is ensured, that the activity concentration of  $^{90}\text{Sr}$  related to the mass of calcium is significantly higher than corresponding values for human bone tissue.
- The blank or rather statistical effects of the nuclides  $^{232}\text{Th}$  and  $^{228}\text{Th}$  could also have caused increased values for the ratio of  $^{228}\text{Th}/^{232}\text{Th}$ .

At the moment these factors are investigated. The concentration activity of  $^{228}\text{Ra}$  related to the site of sampling is determined spectrometrically for the second half of a whole tusk (cf. 3<sup>rd</sup> progress report). The results are compared to the values of the thorium isotopes and  $^{90}\text{Sr}$ .

### 3. References

#### Project part ,Determination of origin of ivory‘

- BLANC, J.J., BARNES, R.F.W., CRAIG, G.C., DUBLIN, H.T., THOULESS, C.R., DOUGLAS-HAMILTON, I. AND HART, J.A. (2007). African Elephant Status Report 2007: an update from the Elephant Database. - Occasional Paper Series of the IUCN Species Survival Commission, No. 33.; IUCN/SSC African Elephant Specialist Group; IUCN, Gland, Switzerland. vi + 276 pp.
- CERLING, T.E., HARRIS, J.M. AND LEAKEY, M.G. (1999). Browsing and grazing in elephants: the isotope record of modern and fossil proboscideans. *Oecologia* 120: 364-374.
- CERLING, T.E., HARRIS, J.M. AND PASSEY, B.H. (2003). Diets of East African bovidae based on stable isotope analysis. *Journal of Mammalogy* 84: 456-470.
- CERLING, T.E., HART, J.H. AND HART, T.B. (2004). Stable isotope ecology in the Ituri Forest. *Oecologia* 138: 5-12.
- CERLING, T.E., OMONDI, P. AND MACHARIA, A.N. (2007). Diets of Kenyan elephants from stable isotopes and the origin of confiscated ivory in Kenya. *African Journal of Ecology* 45: 614-623.
- CITES (2012). Elephant Conservation, illegal killing and ivory trade. 62<sup>nd</sup> Meeting of the Standing Committee, Geneva (Switzerland), 23-27 July 2012.
- FIX, E. AND HODGES, J.L. (1951). Discriminatory analysis – nonparametric discrimination: Consistency properties. Tech. Rep. 4, Project no. 21-29-004. USAF School of Aviation Medicine, Randolph Field, Texas.
- FIX, E. AND HODGES, J.L. (1989). Discriminatory analysis – nonparametric discrimination: Consistency properties. *Internat. Statist. Rev.* 57: 238-247.
- HOBSON, K.A. (1999). Tracing origins and migration of wildlife using stable isotopes: a review. *Oecologia* 120: 314-326.
- MICHENER, R.H. AND SCHELL, D.M. (1994). Stable isotope ratios as tracers in marine aquatic food webs. In: Lajtha, K. and Michener, R.H. (Eds.) *Stable isotopes in ecology and environmental science*. Blackwell Scientific Publications. Pp 138–157.
- PETERSON, B.J. AND FRY, B. (1987). Stable isotopes in ecosystem studies. *Ann. Rev. Ecol. Syst.* 18: 293-320.
- STONE, C.J. (1977). Consistent nonparametric regression. *Ann. Statist.* 5: 595-645.
- TIESZEN, L.L., AND BOUTTON, T.W. (1988). Stable carbon isotopes in terrestrial ecosystems research. In: Rundel, P.W., J.R. Ehleringer and K.A. Nagy (Eds.) *Stable Isotopes in Ecological Research*. Springer Verlag. Pp 167/195
- TIESZEN L.L. (1991). Natural variations in the carbon isotope values of plants: implications for archaeology, ecology, and paleoecology. *Journal of Archaeological Science* 18: 227-248.
- VAN DER MERWE, N.J., LEE-THORP, J.A. AND BELL, R.H.V. (1988). Carbon isotopes as indicators of elephant diets and African environments. *African Journal of Ecology* 26: 163-172.
- VOGEL, J.C, EGLINGTON, B. AND AURET, J.N. (1990). Isotope fingerprints in elephant bone and ivory. *Nature* 346: 747-749.
- ZIEGLER, S., JACOB, D. AND HORNIG, K. (2011). Determination of age and geographical origin of African elephant ivory. Research project supported by the German Federal Agency for Nature Conservation and funded by the Federal Ministry for the Environment, Nature Conservation and Nuclear Safety. 2<sup>nd</sup> progress report for the project part “Geographical origin”, 01 Jan. – 30 June 2011.

#### Project part ,Determination of age of ivory‘

- SCHMIED, S.A.K. (2012): Entwicklung und Validierung einer Analysenmethode zur Bestimmung von <sup>90</sup>Sr im Rahmen der Datierung von Elfenbein mittels der Radionuklide <sup>14</sup>C, <sup>90</sup>Sr und <sup>228/232</sup>Th. Dissertation, Universität Regensburg.
- BRUNNERMEIER, M. (2012): Entwicklung und Validierung einer Methode zur Bestimmung des Todeszeitpunkts von Elefanten durch Bestimmung von <sup>14</sup>C und <sup>228</sup>Th/<sup>232</sup>Th in Elfenbein. Dissertation, Universität Regensburg.
- SCHMIED, S.A.K., BRUNNERMEIER, M.J., SCHUPFNER, R., WOLFBEIS, O.S. (2011): Age Assessment of ivory by analysis of <sup>14</sup>C and <sup>90</sup>Sr whether there is an antique on hand. *Forensic Science International* 207(1-3) e1-e4.
- BRUNNERMEIER, M.J., SCHMIED, S.A.K., MÜLLER-BOGE, M., SCHUPFNER, R. (2011): Dating of ivory from 20<sup>th</sup> century by determination of <sup>14</sup>C by the direct absorption method. *Applied Radiation and Isotopes*, DOI: 10.1016/j.apradiso.2011.10.006.
- SCHMIED, S.A.K., BRUNNERMEIER, M.J., SCHUPFNER, R., WOLFBEIS, O.S. (2012): Dating ivory by determination of <sup>14</sup>C, <sup>90</sup>Sr and <sup>228/232</sup>Th. *Forensic Science International*, accepted.
- BRUNNERMEIER, M.J., SCHMIED, S.A.K., SCHUPFNER, R. (2012): Distribution of <sup>14</sup>C, <sup>90</sup>Sr and <sup>228</sup>Th in an elephant tusk. *Journal of Radioanalytical and Nuclear Chemistry*, DOI: 10.1007/s10967-011-1587-8
- HUA, Q., BERBETTI, M.(2004): Review of tropospheric bomb <sup>14</sup>C data for carbon cycle modelling and age calibration purposes *Radiocarbon*, 46(3), 1273-1298.
- HUA, Q., BERBETTI, M., ZOPPI, U., CHAPMAN, D.M., THOMSON, B. (2003): Bomb radiocarbon in tree rings from northern New South Wales, Australia: Implications for dendrochronology, atmospheric transport, and air-sea exchange of CO<sub>2</sub>. *Radiocarbon*, 45(3), 431-447.

MERILLEISENBUD (1987): Environmental Radioactivity, Academic Press.

BUNDESMINISTERIUM DES INNERN.(1977):Bericht der Bundesregierung: Umweltradioaktivität und Strahlenbelastung.