The other two samples aligned to the White Rhino (Ceratotherium simum) with the genetic distance being 0.0176. The Holstein Cow sample showed a genetic distance of 0.2295 when compared to the closest previously unknown sample. The results confirmed that the unknown samples were from Rhino, with four being of Black Rhino and two being of White Rhino. The results do not determine from which part of the animal the powdered material originated; probably either horn or bone. Elemental analysis using SEM EDX could differentiate material high in sulphur (horn) from material high in calcium (bone). The results indicated that all the six samples were more likely to be from horn than bone. High power microscopy was also able to produce images of scale patterns found in horn, thus again supporting the material coming from horn and not bone. The DNA sequence from the cytochrome B gene was used to compare all the five extant species of Rhino. The DNA data support the Java and Indian Rhino being closely related. The White and Black Rhino are closely related, both being from Africa. The Sumatra Rhino is in between the two groupings, surprisingly more related to the African Rhino than the Java Rhino.

Conclusion: The use of a partial DNA sequence from the cytochrome B gene has been found to be highly effective at the identification of animal species. The DNA locus could differentiate samples originating from any of the five extant species of Rhino. The combination of elemental analysis and SEM allows the body part from where the sample was taken to be determined.

Keywords: Rhino, DNA, Cytochrome B

WIL-FOM-03 Source Location of African Elephant Ivory and Rhinoceros Horn by Stable Isotope Ratio Analysis

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We have explored the use of stable isotope ratio measurements for the purpose of tracing African elephant ivory and rhinoceros horn to the wildlife refuges from which they were obtained. This procedure is based on differences in plant cover, rainfall and geological history. Three different isotope ratios are usually sufficient to identify a source. Savanna grasses follow the C₄ photosynthetic pathway, producing plant material with $^{13}C/^{12}C$ ratios ($\delta^{13}C$ values, relative to PDB) of about -12.5 % (per mil), while trees and shrubs follow the C₃ pathway and produce foliage with $\delta^{13}C$ values of about – 26.5%; dense forests have plants with $\delta^{13}C$ values more negative than -22%. Nitrogen isotopes ratios are related to rainfall or water stress: animals from areas with rainfall >400mm/year have $\delta^{15}N$ values of about 5-10%, while the values are higher in arid regions. These isotope ratios are recorded in the tissues of herbivores, with known fractionations during the synthesis of different tissues. The age of the geological substrate can be determined by stable isotope ratios in which the parent is radioactive: we have used ⁸⁷Sr/⁸⁶Sr, but various isotope ratios of, e.g., neodymium and lead are also effective.

For elephants we have compared the isotope ratios for 20 refuges in 10 countries (van der Merwe *et al.* 1990) and we continue to add to this database. Since elephants prefer to browse, their δ^{13} C values track the availability of trees and shrubs in their feeding range, while the nitrogen and strontium isotope ratios are patterned as predicted.

For rhinos, source location is less complicated, due to the scarcity of rhinos in Africa. The δ^{13} C values of rhino horn distinguish browsing black rhinos from grazing white rhinos (which are essentially confined to South Africa), while a combination of nitrogen, strontium and lead isotopes can identify the source (Hall-Martin *et al.* 1993). Trace element data have also been successfully integrated with isotopic ratios in exploratory studies.

It is possible, in theory, to determine the source of ANY wildlife

product with the right combination of isotopic and trace element data, but an extensive database is required to do so. It is easier to confirm whether a wildlife product comes from a specified source, e.g., the stockpile of ivory in Kruger National Park, South Africa, which is about to be sold with CITES approval. It is also possible to use "isotopic tagging" to monitor such shipments and the items that are made from the raw material (Kruger 1996).

Finally, where only specimens from captive breeding programs may be legally sold, e.g., various species of birds, their diet can be chosen to have an isotopic label that is significantly different from natural populations in the wild; similarly, cultivated endangered plants (e.g., cycads) can also be traced (using the technique for animals) or labeled through fertilizer or water of a specific isotopic or trace element composition.

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Keywords: stable isotopes, ivory, rhino horn

WIL-FOM-04

Ivory Sourcing Using Stable Isotopes

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Stable isotopes indicate aspects of diet and of water sources. Ratios of carbon and oxygen isotopes in ivory $({}^{13}C/{}^{12}C$ and ${}^{18}O/{}^{16}O$, respectively) differ significantly between different parts of Africa, and even within different African countries. In particular, the ${}^{13}C/{}^{12}C$ ratios of savanna elephants differs from that of forest elephants, so that ivory from the forest elephants of central Africa differs from those from east or southern Africa. This difference is due primarily to differences of ¹³C/¹²C ratios within C3 plants. Regionally in eastern and southern Africa, elephants from certain regions incorporate C4 plants into their diet, allowing further differentiation. The total range in d13C in dentine from ivory, including hippopotamus ivory, is from +1 to -19 permil. The ${}^{18}O/{}^{16}O$ ratio of ivory is determined by the ¹⁸O/¹⁶O ratio of local precipitation, which varies somewhat across the continent. The total range in d18O of African ivory is from +3 to -7 permil. Thus, crossplots of ${}^{18}\text{O}/{}^{16}\text{O}$ and ${}^{13}\text{C}/{}^{12}\text{C}$ ratios can often differentiate elephant ivory source regions. This method has promise to be used in conjunction with other methods to help determine possible source regions of confiscated ivory. In this presentation, we discuss differences between ivory samples from East Africa and those from Central Africa.

Keywords: elephants, isotopes, diet

WIL-FOM-05

Natural Stable Isotope Variation as a Tool to Trace Back the Origin of Organic Material

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Our environment is the sum of atoms that differ in their composition of neutrons and protons. This results in different chemical