We have applied techniques similar to those we used to successfully determine the area in which an elephant lived, to rhino horn. But in the case of rhino we are dealing with two species with very different dietary habits, and in addition the sample material is not a calcified tissue like bone or tooth, but a dense hair-like material. We found that we could easily distinguish between black and white rhinoceros from their carbon isotope compositions, and a combination of nitrogen isotopes and various heavy isotopes served to pinpoint the source area very well.

ISOTOPES AND RHINO HORN

Stable isotopic "fingerprinting" which was earlier used to pinpoint the origin of elephant ivory has since been applied to rhino horn.

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Stable isotope analysis is a research tool originally developed in geochemistry, but because it is so useful for investigating all sorts of natural processes, it is now often used in the life sciences as well, for instance in botany and zoology. *Stable isotopes* are different forms of the same chemical element with slightly different atomic masses; they are not radioactive, hence the name "stable". For convenience we commonly distinguish two kinds, "light" and "radiogenic" isotopes. The former include elements with low molecular weight, and the latter are so named because they are the *products* of radioactive decay.

The isotopes of carbon and nitrogen, the basic building blocks of all living organisms, are examples of light isotopes which are incorporated into animal tissues from food. Because the heavier form is incorporated more slowly, the relationship, or ratio, between the two forms changes subtly as the element passes along the foodweb. This process is called *fractionation*. Most plants, such as trees, shrubs, forbs and cool-habitat grasses discriminate strongly against heavier carbon atoms (^{13}C) during photosynthesis. But savanna grasses are different – they use another kind of photosynthesis. The result is that grasses are isotopically very distinct from other plants in African savanna environments. Animals in turn reflect these differences,

ISOTOPES AND RHINO HORN

so that grazers are distinct from browsers (Fig 2). The heavier isotope of nitrogen (^{15}N) tends to become more concentrated as it passes along the foodweb, especially in dry habitats. Therefore the nitrogen isotope ratio in animals tissues reflects moisture or aridity of the local habitat.

In contrast radiogenic isotopes pass unchanged along the foodweb. ie. they are not fractionated. This is because the mass difference between two isotopes is too small to matter to plants or animals. Trace elements such as strontium. lead and neodymium fall into this group. Since they originate in the parent rocks of the region. the ratios of the various radiogenic isotopes tell us directly about the geochemical composition and age of the local geology.

We reasoned that carbon isotope ratios would effectively reveal the species of rhinoceros, since *Diceros bicornis* (black rhino) is a browser and *Ceratotherium simum* (white rhino) is almost exclusively a grazer. But it also meant that carbon could tell us little about local vegetation, since rhinos do not vary their diets according to the proportions of trees and grasses in the landscape as elephants do. Nitrogen isotopes should reveal aspects about the rainfall of the area, as before. And, although we were very uncertain about the concentration of trace elements in the hairlike horn, we again expected that the radiogenic isotopes would reflect the local geology. Because we had lost carbon for source tracing, we added two more elements, lead and neodymium, in addition to strontium, to our "toolkit".

Then we proceeded to analyse about 150 small samples of rhinoceros horn from known areas in the southern African countries, South Africa, Namibia and Zimbabwe (Fig 1). Most of our samples came from horns in Parks Board holdings. but we need so little material that mere shavings of horn from animals which had been tranquilised for other purposes such as veterinary inspections or translocations were also used. Happily, the results conformed mostly to our expectations. Carbon isotope ratios of horn proved to be an excellent indicator of the species. and in some cases we turned up mistakes in the original labelling of the animal's horn! This aspect of the study had immediate forensic repercussions.

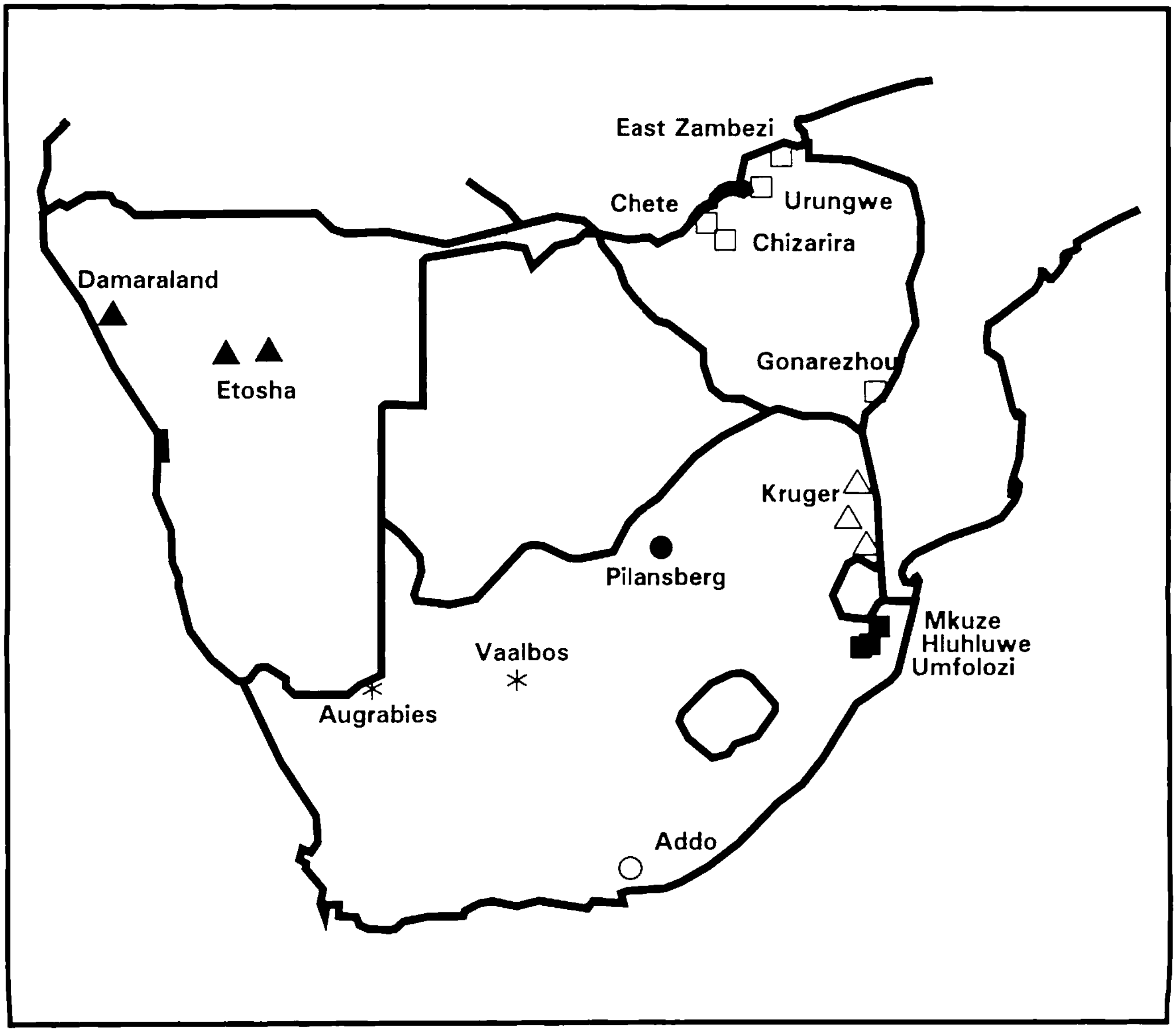
We used the remaining isotopes to check whether they gave clear differences between the various areas we had sampled. In combination the ratios varied significantly from area to area with good clustering in any one place. We did not have enough neodymium and lead results to do multi-element statistical tests, but simple plots of two isotopes at a time illustrate the point. We found that nitrogen and strontium isotopes alone separated many of the sites. Although nitrogen isotopes usually only indicate broad rainfall zones, in combination with a radiogenic isotope the signature is usually quite distinctive. This is particularly so for separating rhinos from the very arid parts of the subcontinent, such as parts of Namibia and Addo, from much wetter habitats in Zimbabwe for instance. Horns from Addo stood out even from other arid areas.

To our consternation we obtained very peculiar results from a male Addo rhino rather whimsically called "Pixie", which had entirely different values for the tip and base of its horn. To our relief the mystery was solved when it was rather impishly explained that Pixie had been moved from Hluhluwe to Addo at a tender age!

Good separation between areas was also observed for radiogenic isotopes alone, strontium and neodymium in particular. This is especially true for areas with distinctive geological features such as Pilanesberg which is an extinct volcanic crater with rocks of uniform age and very distinctive radiogenic isotope patterns. Lead isotopes do not appear to be such effective discriminators. although they helped separate the Namibian samples. We could distinguish samples even from very close sources, such as Mkuze and Umfolozi, and sub-areas of the larger parks, such as Kruger. Horns from the south-east regions, which are underlain by Mesozoic volcanic rocks, are very different to those from south-west regions, where extremely old (more than 2500 million years) Archaean rocks predominate.

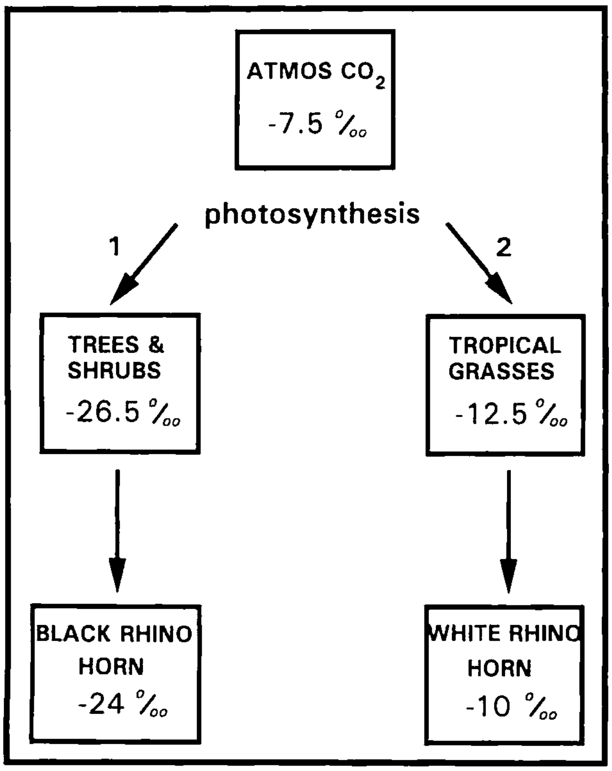
One question remained – how much variation is there in the isotopic signature of one horn? In order to answer this we analysed two horns from Hluluwe and Mkuze, at 5cm intervals. Just over 5cm growth per year appears to be the norm, although it seems that horn of dehorned rhinos grows faster. We found small but patterned variations in the carbon and nitrogen isotopes corresponding to seasonal and annual changes in rainfall and vegetation, but no variation for radiogenic isotopes. From this we concluded that the animals had never moved sufficiently far to change the geological isotopic signature. The light isotope changes are relatively small and do not affect species identification at all, but must be borne in mind when nitrogen isotopes are used for sourcing. Interestingly, the patterns inherent in the light isotope data of horns could tell us something about the dietary habits and drought stresses to which individuals were subjected during their lifetimes.

FIG 1



We have shown that the species and origin of individual rhinos can be verified from isotopic analysis of its horn. Where do we go from here? Quite obviously any answer to this question would *a priori* have to be in the best interest of rhino conservation in Africa. But our results do point to at least two possibilities. The first and perhaps simplest immediate application is that it provides a definitive method for telling whether poached horns came from black or white rhinos. Mistakes can be made when using simple visual observations, and our results confirmed this in some cases. Another application in the forensic field is to pinpoint the origins of poached or smuggled material, so that criminal investigations can be pointed in the right direction, geographically speaking.

FIG 2



We have now shown that there is a natural means for distinguishing various sources of horn, and this introduces the potential for an altogether more radical approach – the introduction of a legal trading system. For instance horn derived from natural mortalities in certain Parks could be isotopically distinguished from those emanating (illegally) from whatever other regions. The advantages would be that the demand for horn which undoubtedly exists in the East however much it is railed against, could be satisfied from natural mortalities, and the various Parks would benefit from the revenue. It would require that horns from all countries with rhino populations be isotopically fingerprinted, which is not really a huge undertaking, because rhinos are regrettably rare in most parts of Africa, except for small pockets in Kenya, Tanzania, Zambia and Zimbabwe, and the more stable populations in South Africa and Namibia. We have already begun to tackle this task with the help of various African conservation authorities.

Given the failure of the total ban approach, it seems obvious that the rhino problem requires critical and radical review. We hope that our results will assist the current debate about the best way to conserving rhinos on the continent.

Notes

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