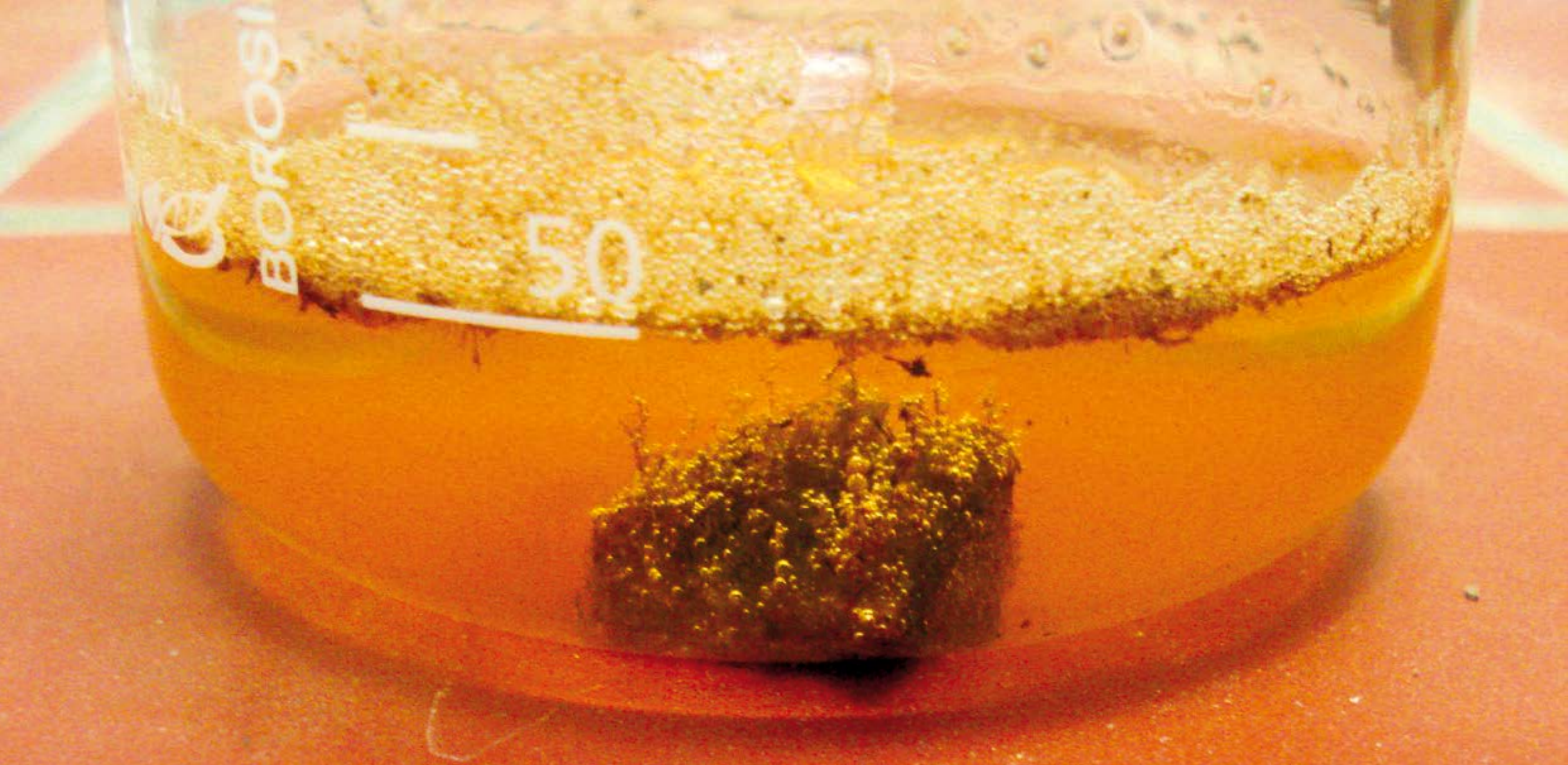

Fossil bones from the North Sea: stable isotopes

FIGURE. | *Chemical preparation of a piece of mammoth bone.*

U vindt een Nederlandse samenvatting aan het eind van de tekst.

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Abstract | **The North Sea is a unique heritage site yielding a large amount of palaeontological and archaeological data. Many fossil bones have been Radiocarbon dated to provide a time parameter to these finds. Here we discuss additional data obtained for these samples: the stable isotopes ^{13}C and ^{15}N of the dated bone collagen. These provide information on the palaeo-environment and diet of the individuals (fauna and humans) and changes therein through time.**

Introduction

In the last few decades, thousands of Pleistocene and Holocene animal fossils and archaeological finds have been retrieved from the North Sea bottom. These remains witness of the presence of humans and mammals within this area throughout different episodes in the past, when the sea level was dramatically lower than today. As a result of natural erosion and sedimentation processes most of the North Sea finds are no longer *in situ*. Moreover, contextual information is mostly lacking or very scarce due to the methods in which the archaeological material was retrieved. In order to place the fossils in a chronological context, many of these have been radiocarbon dated. This resulted in an extensive dataset, consisting of different species and varying in age from

Pleistocene up to modern. An overview of radiocarbon dated fossils from the North Sea (more than 300 samples) is published in our accompanying article in this issue. Here we focus on the stable isotopes (^{13}C and ^{15}N) of the ^{14}C dated fossils.

Stable isotopes

For ^{14}C dating, collagen is prepared from bone samples as the datable fraction. From the same collagen also the contents of the stable isotopes ^{13}C and ^{15}N is measured (the latter not for all samples). This content is expressed in delta (δ) values, a measure of enrichment (δ>0) or depletion (δ<0) relative to a reference. The δ value is defined as the deviation (expressed in permil) of the rare to abundant isotope ratio from that of the reference material:

$\delta^{13}C = [(^{13}Rsample/^{13}Rreference) - 1]$ (x1000‰) where $^{13}R = {}^{13}C/^{12}C$
and
$\delta^{15}N = [(^{15}Rsample/^{15}Rreference) - 1]$ (x1000‰) where $^{15}R = {}^{15}N/^{14}N$

The absolute isotope contents of the reference materials have been measured very accurately. For carbon, the reference material is belemnite carbonate (known as V-PDB); for nitrogen, the reference is ambient air (see Mook, 2006 and references therein). The stable isotope ratios are measured by Isotope Ratio Mass Spectrometry (IRMS). The analytical error is 0.1‰ and 0.2‰ for $\delta^{13}C$ and $\delta^{15}N$, respectively.

Photosynthesis causes isotopic fractionation: plants have a lower ^{13}C content than the CO_2 of the atmosphere they grow in. There are two main photosynthesis processes, known as C3 and C4. C3 plants have $\delta^{13}C$ values ranging from -23 to -29‰; for C4 plants that is from -10 to -15‰. For the North Sea (and temperate climates in general) only C3 plants are relevant.
For atmospheric CO_2 the $\delta^{13}C$ value is around -8‰. Thus, the fractionation for C3 plants is around 16‰; the plants are depleted in ^{13}C by 16‰ or 1.6%.
Also the ^{14}C isotope is depleted, by 3.2%. Theoretically this number corresponds to an effect of about 250 years (for detailed calculations see e.g. Mook, 2006 and Fry, 2008). This effect is normalized for by the Radiocarbon convention.

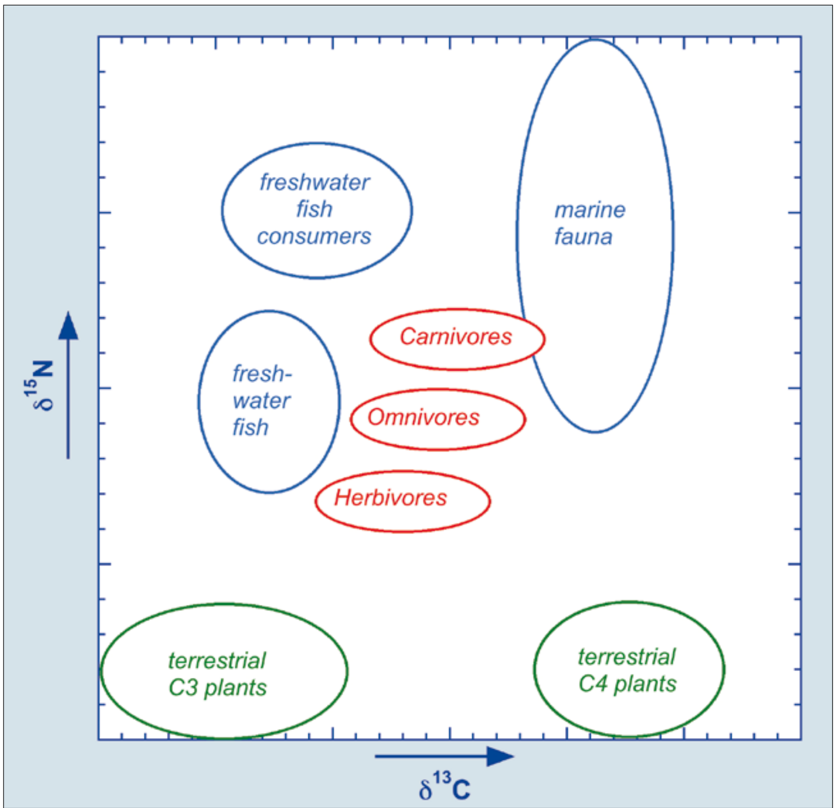

FIGURE 1. | *Schematic representation of (part of) the food web, indicating the relative ranges of $\delta^{13}C$ and $\delta^{15}N$ values.*

The stable isotopes ^{13}C and ^{15}N are proxies for the diet of an individual, for bone collagen for roughly the last decade of its lifetime (Hedges & Reynard, 2007). Consuming C3 or C4 plants is a clear example; corn and millet are C4 plants, and significant consumption of these will show in the $\delta^{13}C$ values of the bone collagen.

For the North Sea context, two effects which are most relevant are (1) the trophic chain, and (2) aquatic food sources. Only C3 vegetation is relevant here.

(1) As the food source that constitutes the main component of a diet passes on to the next trophic level, the $\delta^{13}C$ and $\delta^{15}N$ values are both raised (Kohn, 1999). This leads to the much used phrase "you are what you eat (plus a few permil)". The increase in δ value per trophic step is ca. 1‰ for $\delta^{13}C$, and ca. 3–5‰ for $\delta^{15}N$ (Bocherens & Drucker, 2003).

(2) Organisms living in aquatic reservoirs show specific isotopic ratios, depending on the specific reservoir. Marine organisms show $\delta^{13}C$ around -14‰; for freshwater, this is about -24‰ typical for the region. Their $\delta^{15}N$ values vary widely and can reach values up to 18‰ for high-trophic level fish and mammals. These are general values, showing variations of a few permil.

Aquatic isotope effects also show in the bone collagen of terrestrial organisms (including humans), when a significant part of their diet originates from aquatic sources; in particular fish, but perhaps also other fauna like waterfowl, otters etc. The $\delta^{15}N$ values are then elevated; the $\delta^{13}C$ values enable a distinction between the sea and rivers.

Figure 1 shows an example of ranges for C3 and C4 plants, terrestrial fauna (herbivores, omnivores and carnivores), aquatic fauna and humans consuming terrestrial food, freshwater food and marine food. The $\delta^{13}C$ and $\delta^{15}N$ values are general numbers. Key parameters are the baselines (starting values for the stable isotope ratios) of the food chain. The exact values are determined by context. For a general reference, see Fry (2009).

species (English)	species (Latin)	environment & diet	n	$\delta^{13}C$ (‰)	n	$\delta^{15}N$ (‰)
Straight tusked elephant	*Elephas antiquus*	terrestrial herbivore	7	-20.7	7	9.9
Woolly mammoth	*Mammuthus primigenius*/sp.	terrestrial herbivore	29	-22.0	25	6.9
Cave lion	*Panthera spelaea*	terrestrial carnivore	2	-19.3	2	8.5
Arctic fox	*Alopex lagopus*	terrestrial carnivore	1	-20.7	1	8.5
Hyena	*Crocuta crocuta spelaea*	terrestrial carnivore	1	-20.1	0	
Dog/wolf	*Canis lupus*/sp.	terrestrial carnivore	2	-22.5	2	9.0
Bear	*Ursus arctos*/sp.	terrestrial omnivore	2	-21.3	2	5.6
Otter	*Lutra lutra*	freshwater carnivore	2	-25.5	1	8.6
Wolverine	*Gulo gulo*	terrestrial carnivore	1	-21.2	0	
Woolly rhinoceros	*Coelodonta antiquitatis*	terrestrial herbivore	5	-20.7	3	4.4
Horse	*Equus caballus*/sp.	terrestrial herbivore	7	-21.2	7	4.8
Roe deer	*Capreolus capreolus*	terrestrial herbivore	2	-21.8	2	2.7
Moose	*Alces alces*	terrestrial herbivore	5	-21.0	5	3.2
Red deer	*Cervus elaphus*	terrestrial herbivore	14	-21.6	9	3.5
Giant deer	*Megaloceros giganteus*	terrestrial herbivore	4	-20.0	3	4.9
Reindeer	*Rangifer tarandus*	terrestrial herbivore	16	-19.9	12	3.8
Balaenidae	Balaenidae	marine carnivore	1	-16.7	1	9.3
Bowhead whale	*Balaena mysticetus*	marine carnivore	1	-14.8	1	14.3
Common rorqual	*Balaenoptera physalus*	marine carnivore	1	-12.6	0	
Beluga whale	*Delphinapterus leucas*	marine carnivore	4	-15.5	3	15.2
White-beaked dolphin	*Lagenorhynchus albirostris*	marine carnivore	2	-12.2	2	15.9
Grey whale	*Eschrichtius robustus*	marine carnivore	14	-14.3	12	14.2
Killer whale	*Orcinus orca*	marine carnivore	2	-12.0	2	16.4
Grey seal	*Halichoerus grypus*	marine carnivore	2	-13.6	1	14.5
Walrus	*Odobenus rosmarus*	marine carnivore	5	-13.3	4	12.1
Bottlenose dolphin	*Tursiops truncatus*	marine carnivore	2	-11.9	2	15.4
Harp seal	*Pagophilus groenlandica*	marine carnivore	2	-15.4	0	
Wild boar	*Sus scrofa*	terrestrial omnivore	3	-21.7	2	5.1
Musk ox	*Ovibos moschatus*	terrestrial herbivore	1	-20.1	0	
Bison	*Bison priscus*/sp.	terrestrial herbivore	3	-20.4	1	3.9
Aurochs	*Bos primigenius*	terrestrial herbivore	5	-22.1	5	5.3
Caprinae	Caprinae	terrestrial herbivore	1	-19.2	1	7.5
Bovid	Bovidae	terrestrial herbivore	5	-22.0	5	5.6
Hare	*Lepus* sp.	terrestrial herbivore	1	-21.3	0	
Beaver	*Castor fiber*	terrestrial herbivore	1	-22.1	1	4.8
Human	*Homo sapiens*	terrestrial omnivore	124	-20.0	123	12.1
Great auk	*Pinguinus impennis*	marine carnivore	3	-14.5	3	17.1
Unknown	unknown	unknown	6	-21.7	6	5.2

TABLE 1. | *Overview of analysed species, environment and average values of the stable isotope ratios $\delta^{13}C$ and $\delta^{15}N$.*

This article presents an extensive and unique dataset of stable isotope signals of fossil remains from the North Sea area. The majority consists of data from mammal fossils, which can be used as a way to increase our knowledge about ecological aspects of the North Sea area in the past. Also, the dataset consists of data on numerous human remains, which can shed light on human dietary patterns and lifestyle within the North Sea area through time.

The current dataset consists of data of terrestrial and aquatic organisms. For the latter, the ^{14}C dates need correction for so-called reservoir effects in order to derive absolute ages. These are caused by lower natural ^{14}C concentrations in aquatic environments compared with the atmosphere, leading to apparent ages: contemporaneous organisms from rivers and oceans contain less ^{14}C and thus date older. For the marine environment, the apparent age is generally 400 years for the North Atlantic during the Holocene era. For rivers, the effect is larger; the typical value for the Rhine is 1300 years.
For estuaries like the Dutch delta, there is a mixture of seawater and riverwater, leading to varying values of the reservoir effect.
Reservoir effects also apply to terrestrial organisms with a significant aquatic food subsistence, including humans. The stable isotopes $\delta^{13}C$ and $\delta^{15}N$ of fossil

bones are a necessary tool to quantify these reservoir effects. The present tables consist for a major part of unpublished ^{14}C, ^{13}C and ^{15}N data, and can be used in the future to further zoom in on this question. In all figures and tables, the ^{14}C ages (BP) are given instead of calibrated dates (calBP) to avoid reservoir effect ambiguities. The BP numbers are not corrected for reservoir effects, whereas the calibrated dates are corrected.

Material and methods

The dataset consists of stable carbon and nitrogen data of directly radiocarbon dated samples of numerous mammalian species, representing marine carnivores, terrestrial carnivores, omnivores (incl. humans), herbivores, and one bird species (Table 1). A grand total of 289 samples is analysed: 165 for animals (104 Pleistocene and 61 Holocene), and 124 for humans.
The data include at least 25 artefacts/worked items made of antler and bone.

The majority of the animal samples correspond to the Pleistocene. Indeed, many samples belong to typical Late Pleistocene mammoth steppe fauna, including woolly mammoth, cave lion, arctic fox, hyena, woolly rhinoceros, horse, giant deer, reindeer, and steppe bison. In addition, the dataset includes 19 marine animals with a Pleistocene age, such as bowhead whale, common rorqual, beluga whale, grey whale, grey seal, harp seal, walrus, and great auk.

Many finds come from a relatively limited number of areas from the North Sea, specifically locations that are frequently exploited by fishermen and that are suitable for sand extraction purposes. Indeed, about a third of the samples come from the important fishing areas Bruine Bank (Brown Bank, n=53) and Eurogeul (Eurogully, n=43). Also, a large part (n=38) comes from the Dutch province Zuid-Holland, where many large sand suppletion projects were realised in recent years.

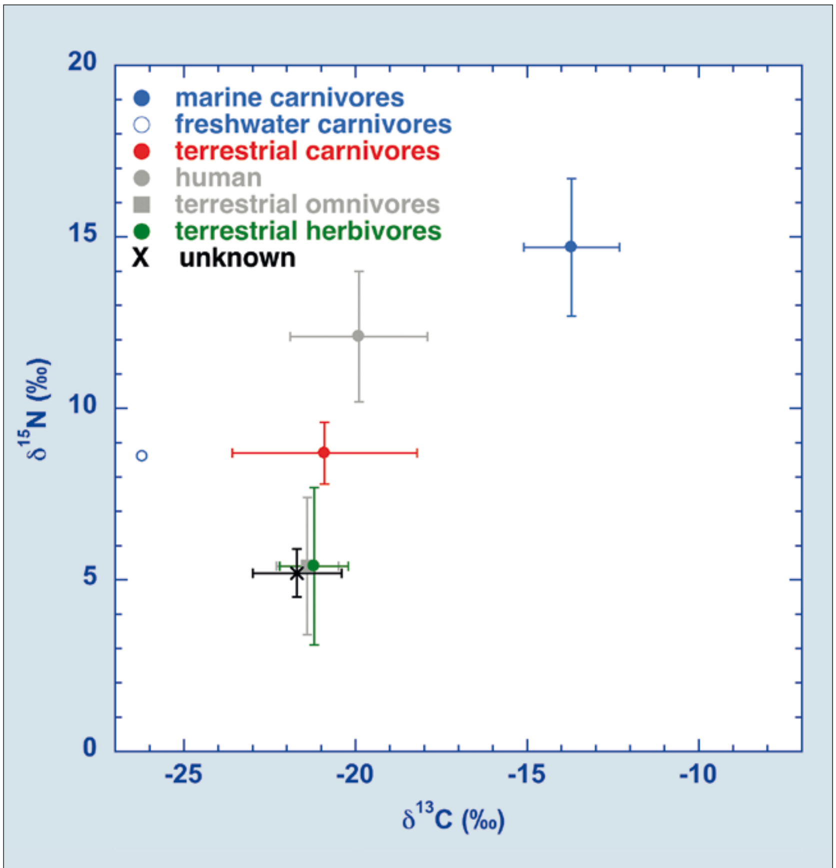

Figure 2. | *Mean stable isotope ($\delta^{13}C$ and $\delta^{15}N$) values with one sigma standard deviation for all animal samples, ordered per trophic level (herbivore, carnivore, omnivore) and habitat (marine, terrestrial). All samples are from mammals.*

Results

Table 1 shows the analysed species, number of samples, class, environment (marine or terrestrial) and trophic level, with the (averaged) value of $\delta^{13}C$ and $\delta^{15}N$.

Figure 2 shows the mean $\delta^{13}C$ and $\delta^{15}N$ values for the samples, roughly ordered per trophic niche. In general, the isotope signals of most species fit into broad groups as shown in Figure 1. As expected, the marine carnivores show the highest $\delta^{13}C$ and $\delta^{15}N$ values. Their $\delta^{13}C$ values range between -16.7 and -11.4‰, and the $\delta^{15}N$ values between +9.3 and +17.7‰. The terrestrial herbivores show $\delta^{13}C$ values ranging from -23.3 to -18.9‰. The $\delta^{15}N$ values cover a large range of 9.9‰, the lowest being +1.7‰ and the highest $\delta^{15}N$ value (+11.6‰) being higher than those of the terrestrial omnivorous animals and terrestrial carnivores in the current dataset.

Apart from the humans, the terrestrial omnivores data fall within the range of these of terrestrial herbivores; the $\delta^{13}C$ values range between -22.3 and -20.3‰, and $\delta^{15}N$ between +3.5 and +7.7‰.

The humans show huge ranges of both isotope values. Their $\delta^{13}C$ values range between -24.7 and -13.4 ‰ and the $\delta^{15}N$ values range all except one (=4.5‰) between +8.0 and +17.4‰. The terrestrial carnivores have $\delta^{13}C$ and $\delta^{15}N$ values between -25.6‰ and -19.2‰ and between +7.7 and +10.2‰, respectively. The dataset consists of two samples of a freshwater carnivore, an otter, with $\delta^{13}C$ values of -26.2 and -24.8‰, and a $\delta^{15}N$ value of +8.6‰ (the $\delta^{15}N$ of only one sample has been measured).

These categories of trophic level/habitat are composed of a variety of species from different time periods. In the following, results of a number of species will be discussed in more detail. This selection is primarily based on the number of available samples per species.

Discussion

Pleistocene and Holocene mammal fossils from the North Sea area

The data for the Pleistocene samples

are shown in Figure 3. As can be seen, the δ¹⁵N values of straight-tusked elephant (+7.5 to +11.6‰) and many of woolly mammoth (+5.0 to +9.1‰) exceed these of other herbivores. Enrichment of ¹⁵N (high δ¹⁵N values) is commonly observed for woolly mammoths (Bocherens, 2003) and has also been observed for Middle-Pleistocene straight-tusked elephants from Germany (Kuitems, 2020). The current dataset also includes woolly mammoth samples with lower δ¹⁵N values. This has also been observed for Late-Glacial woolly mammoths in the Ukraine (Drucker *et al.*, 2014), and has been associated with changing environments and/or climates and changes of niche occupation after the Last Glacial Maximum (LGM). Pre-LGM values of mammoths from Europe (including samples from the Ukraine) show higher δ¹⁵N values, ranging between 6.7‰ and 11.1‰. The current dataset of the North Sea contains only mammoth samples dating pre-LGM, the majority being older than the ¹⁴C background (>45,000 BP). The woolly mammoth samples with the lowest δ¹⁵N values (between +5.0 and +6.0‰) are all older than 45,000 years BP, the limit of the ¹⁴C dating range for bone.

The δ¹³C values of woolly mammoths (-22.4 to -21.4‰) are rather negative (depletion of ¹³C) compared to these of other herbivores, including straight-tusked elephants (-21.2 to -20.2‰). Also, this picture (woolly mammoths are depleted in δ¹³C compared to other herbivores) is known for other localities, and has tentatively been linked to seasonally metabolism of stored fat (Bocherens, 2003).

All horses (n=6) in this dataset originate from the (Late) Pleistocene. Their δ¹⁵N and δ¹³C values range from +2.3 to +6.4‰ and from -22.0 to -19.9‰, respectively. The general view of the horse diet is that it consists predominantly of grasses. However, dental wear studies on molars from Middle Pleistocene horses from Europe, with considerably low δ¹⁵N values, indicated that these specimens were mainly browsing (i.e. eating foliage). The lower δ¹⁵N values have also been interpreted as the result of a browsing diet for these specimens (Kuitems *et al.*, 2020), since usually browsers tend to have lower δ¹⁵N values than grazers and also lower δ¹³C values. Another recent study on a Middle Pleistocene site from France (Richards *et al.*, 2017) revealed that stable isotope values from horse dentine samples in a temporal sequence followed environmental and possibly climatic changes through time. Here, the lowest δ¹⁵N values (about +2 to +3‰) are linked to low temperatures. The lowest δ¹⁵N values of horse samples in the current dataset from the North Sea can either be caused by a specific diet (i.e., browsing) or low temperatures and/or high amounts of precipitation (i.e., affecting the δ¹⁵N values of plants they are feeding on), or by a combination of these factors.

Well-defined temporal changes in the distribution of δ¹³C and δ¹⁵N values have been observed for several species within North-western Europe, related to the climatic and environmental differences during the Late-Glacial and/or around the Pleistocene-Holocene boundary. These include changes in atmospheric CO_2 concentrations (Richards & Hedges, 2003), climatic variation on a local-scale including soil moisture conditions (Stevens and Hedges, 2004), and forest development after the LGM (Kuitems, 2020), leading to depleted δ¹³C values in the Holocene.

Indeed, the Pleistocene-Holocene boundary is characterised by severe climatic and environmental changes for the area of the current North Sea. In general, the onset of the Holocene is characterised by wetter and warmer climatic circumstances, sea level rise and forest development. The current stable isotope record for the North Sea does not allow for statements on temporal trends: none of the species is represented (in appropriate numbers) in both Pleistocene and Holocene. Moreover, in general, samples from the last part of the Late-Pleistocene, i.e. the LGM and the Late Glacial, are lacking.

In contrast to the characteristic Pleistocene 'mammoth steppe fauna', generally thriving in open landscapes, the Holocene samples represent specimens from temperate species and forest dwellers. This is illustrated by the North Sea

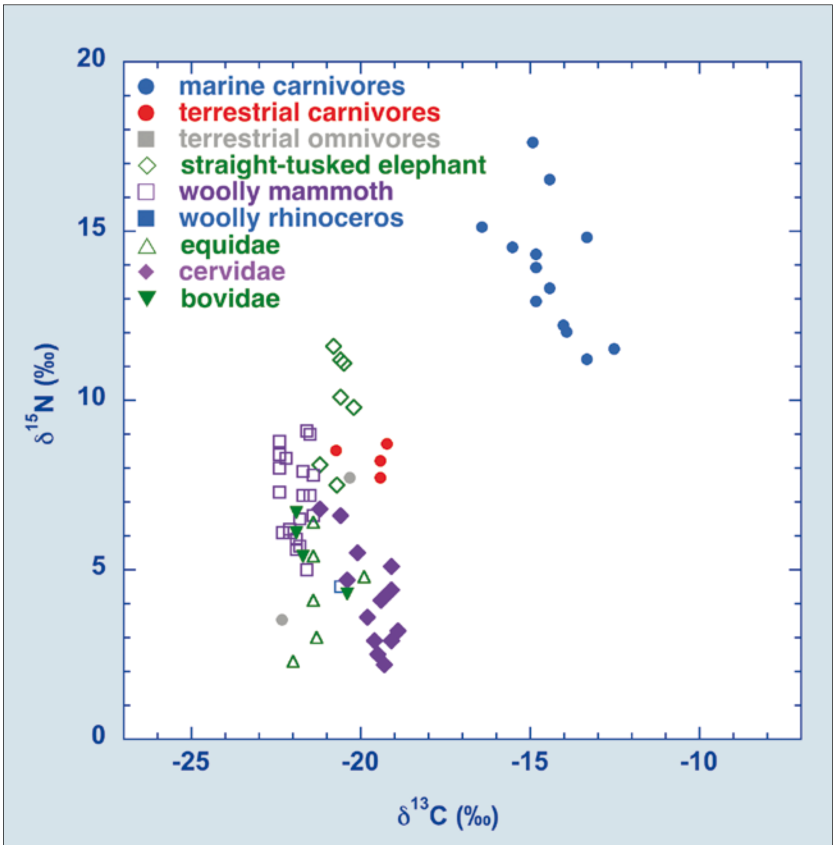

Figure 3. | *Stable isotope (δ¹³C and δ¹⁵N) values for Pleistocene mammal samples from the North Sea.*

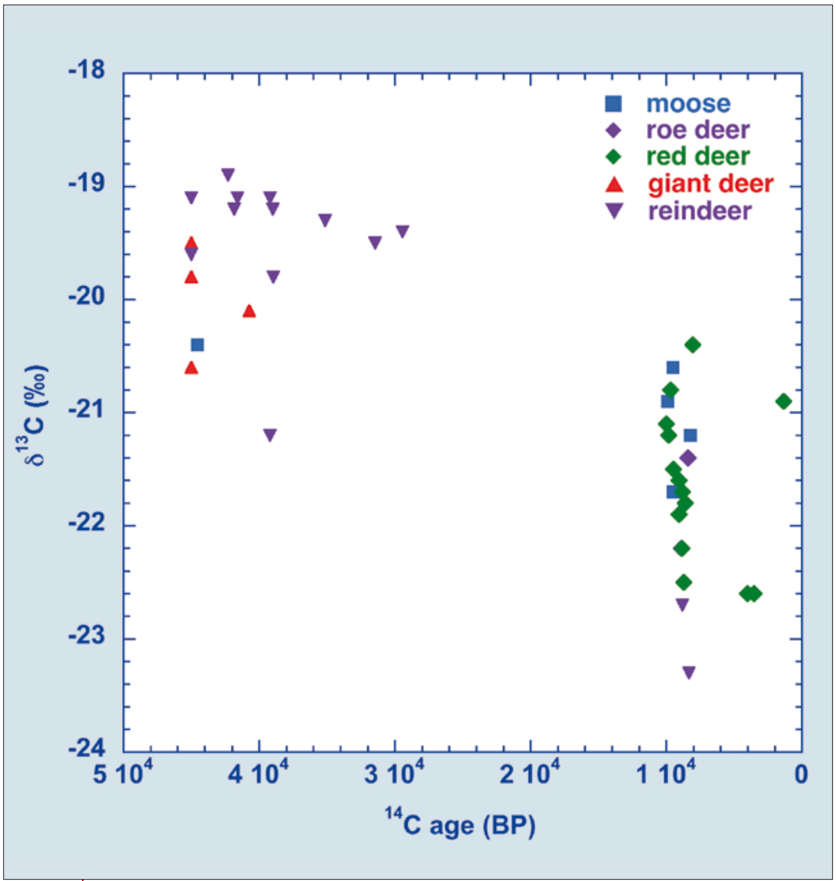

FIGURE 4. | *Stable isotope δ^{13}C values for samples of cervids from the North Sea through time.*

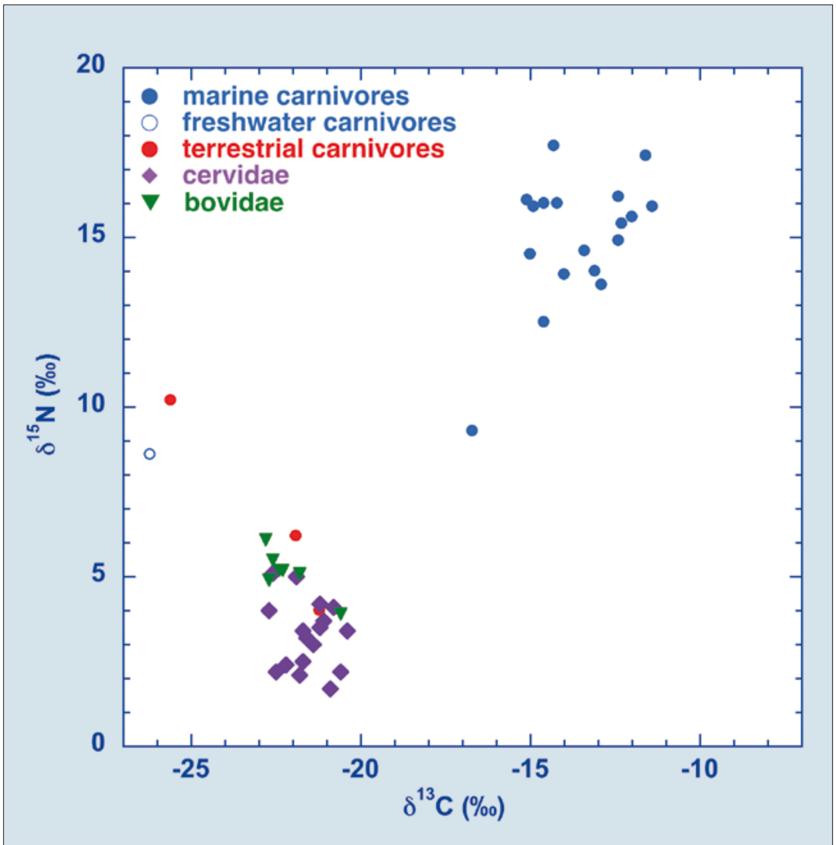

FIGURE 5. | *Stable isotope (δ^{13}C and δ^{15}N) values for Holocene animal samples from the North Sea.*

samples of cervids: Pleistocene samples are chiefly from reindeer, and consists further of giant deer and moose, whereas the Holocene samples of cervids predominantly consist of red deer, and further include moose and roe deer. As shown in Figure 4, the δ^{13}C values of samples from Holocene cervids (n=22, average = -21.7 ‰) are considerably more negative than those of the Pleistocene cervids (n=20, average = -19.7‰).

Our interpretation is that this difference can partly be ascribed to the changed δ^{13}C values of plants caused by the Pleistocene to Holocene climatic shift. Moreover, this difference can be explained by dissimilarities between the species, such as niche occupation, physiology and dietary composition. For instance, in general the diet of reindeer consists for a significant part of lichens, which commonly yield higher δ^{13}C values than vascular plants.

Considering the marine bone samples, a number of the walruses (*Odobenus rosmarus*) has a low nitrogen isotope ratio (n=4; average = +12.1‰) compared to most other marine mammals (average = +14.5‰). This might be ascribed to the feeding on food sources of a low trophic level, such as clams and other molluscs.
Also, it should be noted that due to the sea level rise, samples of marine mammals recur in the Holocene record.

Freshwater species are only represented by two samples from a Holocene otter. Also, the stable isotopes of the Holocene dog/wolf (*Canis* sp.; GrA-24209) show mainly a freshwater signal (Fig. 5). This is not surprising, since the Holocene dog lived together with humans, as is known from other sites in North-western Europe (e.g. Fischer *et al.*, 2007). In general, freshwater fish was an important part of the diet of Mesolithic humans in this area (see next paragraph). Their dogs might have consumed the remains of the human meals, resulting in a freshwater signal.

Human fossils from the North Sea area

Besides numerous samples from human skeletal remains, abundant finds of stone tools indicate the presence of Mesolithic hominins in the North Sea area. Indeed, Mesolithic humans inhabited the higher areas, such as river dunes, in the coastal landscape of the North Sea. This was also known from archaeological sites such as Hardinxveld-Giessendam. Amongst the faunal remains that were submitted for radiocarbon dating are numerous pieces of worked bone and antler, including artefacts. These comprise long bones of bovids, equids, and cervids, and antler predominantly of red deer. The bone material from six of such objects could not be identified at species level. These are plotted in Figure 2 as 'unknown'. As is visible in this figure – and as expected from the species of which the other artefacts were made (i.e., mainly cervid) – their $\delta^{13}C$ and $\delta^{15}N$ values fall within the ranges of terrestrial herbivores. This is not surprising as artefacts were usually made from cervid bone.

Our isotopic dataset contains more than a hundred human skeletal remains with a North Sea context. These include bones from the sea floor found by fishing or dredging (e.g., Amkreutz *et al.*, 2018) as well as beach finds. Recently, an overview was published of 56 human bone finds (van der Plicht *et al.*, 2016). Of these, 33 were Mesolithic (including a few Late Palaeolithic), the finds of the remaining 23 range between Roman and recent times. The main conclusion of the stable isotope analysis was that the Mesolithic humans were predominantly consumers of freshwater protein.

Since this 2016 publication, the dataset of human finds has significantly grown in size. The fossils are either Mesolithic, or ranging between today and younger Roman times. One particular exceptional date is at 5,020 BP (GrM-10161) from the island of Texel. The average stable isotope ratios $\delta^{13}C$ and $\delta^{15}N$ of the human bones are shown in Figure 2; the $\delta^{15}N$ values are plotted against time in Figure 6A. There is no proper baseline available for isotope values of different isotope resourses in this area, and $\delta^{15}N$ values are affected by multiple factors. But many of the humans have $\delta^{15}N$ values which are indicative for aquatic dietary resources (in particular those with $\delta^{15}N$ values larger than 12‰). Also, abundant amounts of fish remains from excavated archaeological coastal sites such as at Hardinxveld-Giessendam reveal that fish may have been the most important dietary resource for Mesolithic humans, in particular pike (Louwe Kooijmans, 2005).

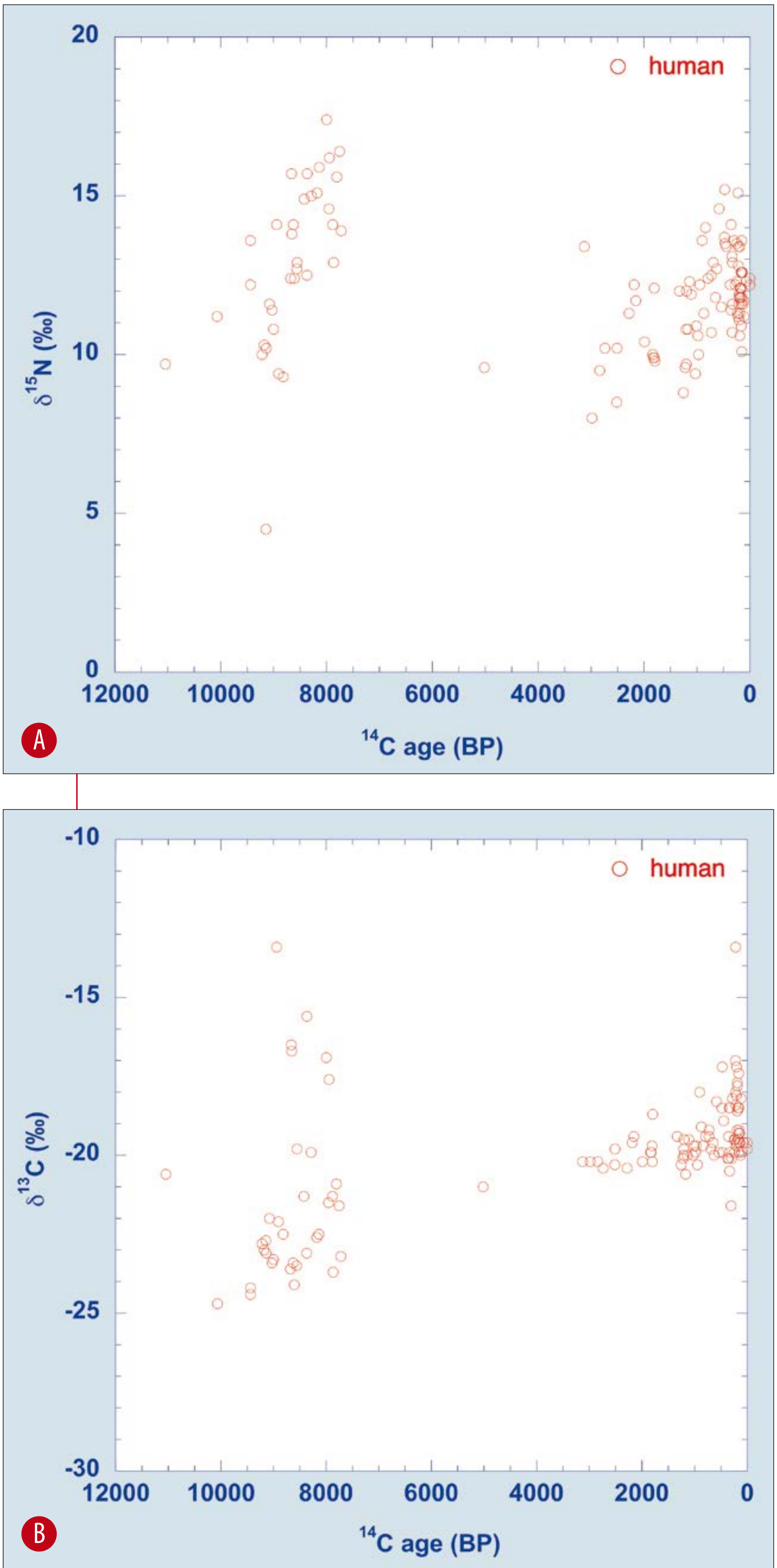

Figure 6. | *Stable carbon isotope values of human bones from the North Sea through time. Top: figure 6A, $\delta^{15}N$ vs. ^{14}C age (BP); bottom: figure 6B $\delta^{13}C$ vs. ^{14}C age (BP).*

As can be seen in Figure 6B, there is a discrepancy between the Mesolithic and later inhabitants of the North Sea region. The low $\delta^{13}C$ values point to a significant component of freshwater fish in the diet of Mesolithic humans. The $\delta^{13}C$ values of the non-Mesolithic human bones have intermediate $\delta^{13}C$ values, suggesting a mixed marine/freshwater/terrestrial food source. This can be expected in a delta region like the coastal Netherlands.

Compared to the other samples of the current dataset, there is one exceptionally low $\delta^{15}N$ value of 4.5‰. This is not necessarily an ingenuine result, as the human $\delta^{15}N$ ranges generally between 4 to 10‰ for terrestrial diets, depending on the trophic level of the food protein. Such low $\delta^{15}N$ values for human bone have been observed before, for example for the Mesolithic in Germany (Terberger *et al*., 2012). But it remains one single observation which would benefit from confirmation.

Conclusions

For the vast majority of animal remains from the North Sea, this is a unique dataset concerning stable isotope values. The stable isotope dataset consists of information on both extinct and extant terrestrial and aquatic species. The extinct species include straight-tusked elephant, woolly mammoth, woolly rhinoceros, and cave lion. The dataset also contains more than 100 samples from human skeletons.

The North Sea area has been archaeologically and palaeontologically extensively explored by both classic methods and modern techniques. For more than 30 species, the stable isotope data add valuable information on environmental conditions and dietary habits in this area through time.

Samenvatting

De stabiele isotopen ^{13}C en ^{15}N in fossiel bot.

Gedurende de laatste tientallen jaren zijn grote aantallen fossiele botten van de Noordzeebodem verzameld, zowel van fauna als van mensen uit de laatste ijstijd en het Holoceen. Deze zijn voor een deel gedateerd door middel van het radioactieve isotoop ^{14}C. Dit geeft de belangrijke “tijd” parameter voor de monsters, welke vrijwel allemaal zonder verdere context zijn gevonden.

Voor het berekenen van een datering is ook de concentratie van het stabiele isotoop ^{13}C nodig (voor correctie van isotopenfractionering); die wordt daarom standaard meegenomen in de metingen. Gedurende de jaren 1960 bleek dat het ^{13}C gehalte op zich ook informatie bevatte betreffende omgevingsfactoren waarin het te dateren organisme leefde, alsook informatie over het dieet. Datzelfde bleek gedurende de jaren 1990 voor het stabiele stikstof isotoop, ^{15}N. Tegenwoordig wordt dan ook standaard voor collageen van fossiel botmateriaal het gehalte van alle drie genoemde isotopen ^{14}C, ^{13}C en ^{15}N bepaald. Ook als de onderzoeksvraag alleen een datering betreft.

De bepaling van de stabiele isotopen ^{13}C en ^{15}N geeft informatie betreffende de omgeving en het voedsel van de gedateerde individuen. Voor de Noordzee is dit een uniek gegevensbestand. Voor 289 monsters wordt dit hier besproken; 165 voor fauna, waarvan 104 Pleistoceen en 61 Holoceen en 124 voor menselijke resten. De fauna omvat ook archeologische artefacten en uitgestorven dieren zoals de wolharige mammoet, wolharige neushoorn, sabeltandkat en reuzenhert en verder o.a. poolvos, paard, rendier, hertachtigen en steppewisent. Aanvullend is ook een aantal mariene dieren zoals walrus, zeehondsoorten, walvissoorten en reuzenalk.

Mammoeten zijn ten opzichte van andere herbivoren relatief verrijkt in ^{15}N. Voor de Noordzee vertonen enkele mammoeten lagere ^{15}N waarden, wat een aanwijzing is voor een veranderend landschap, leefmilieu, klimaat of dieet. De geanalyseerde paarden zijn allemaal Pleistoceen, hun dieet bestaat voornamelijk uit grassen. De ^{15}N waarden zijn een aanwijzing voor een specifiek dieet (d.w.z. grazen) dan wel lagere omgevingstemperaturen.

In tegenstelling tot de Pleistocene mammoetsteppefauna, zijn de Holocene botmonsters karakteristiek voor een bosachtig landschap en gematigde temperaturen. Dit blijkt vooral uit het spectrum van hertachtigen in de Noordzee: voor het Pleistoceen is dat vooral rendier, eland en reuzenhert, voor het Holoceen edelhert, ree en ook eland. De Holocene hertachtigen blijken verarmd in ^{13}C ten opzichte van die van het Pleistoceen. Waarschijnlijk is dat veroorzaakt door de verandering van de vegetatie (met specifieke ^{13}C waarden) na de Pleistoceen/Holoceen overgang.

Een aparte categorie stabiele isotopenwaarden betreft aquatische milieus. Die verschillen aanzienlijk voor de terrestrische, zoetwater en mariene reservoirs. Meest opvallend is dat de botmonsters van de mesolithische mens die in het huidige Noordzeegebied leefde een overwegend zoetwater signaal vertonen; het dieet moet voor een groot gedeelte uit riviervis (geen zeevis) hebben bestaan. Ditzelfde geldt ook voor een hond, die blijkbaar met de mens meeat.
De hoeveelheid menselijk botmateriaal uit de Noordzee is groot, meer dan 100 monsters; dat aantal neemt nog steeds toe. Hiervan is 33 mesolithisch gedateerd. De grote hoeveelheid is vooral te danken aan forensisch onderzoek.

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