2. Human diet

"Tell me what you eat and I will tell you what you are"

Anthelme Brillat-Savarin (1826)
Physiologie du Gout, ou Meditations de Gastronomie Transcendante

An archaeological site can provide various lines of evidence on how prehistoric humans utilized their environment. Archaeobotanical analysis of charred plant remains or pollen grains allows a better understanding of which plants were gathered or cultivated, and calculations on the timing and efficiency of ancient cropping systems can be made (Jones and Colledge 2001; Bogaard 2004). On the other hand, archaeozoological studies of faunal assemblages can provide information on which animal species were selected for hunting or domestication (Lyman 1994). The mortality profile in domestic species may indicate whether animals were raised for meat or secondary products like milk (Halstead 1998). Artefacts may also indicate what ancient people ate; the best examples of which are fish hooks and so called milk strainers (Hedges 2009). Also, residue analysis of fats and proteins on potsherds indicates which types of food were produced or stored (Craig 2002; Evershed *et al.* 2008). While these approaches make the presence or absence of food items evident, they may not provide a full picture on ancient human dietary choice and behaviour, which may also be affected by age, sex or dietary taboos. Here, direct measures of the consumer's body tissue can determine which primary food sources were utilized by the individual. What remains of a human individual in the archaeological context is, in the best case, a buried skeleton. Depending on the burial conditions and environment, skeletal remains have the potential to persist over millennia and retain a lifetime dietary signal which is preserved in the bones' chemical components. In archaeological science this individual dietary signal is traced with the stable isotope ratios of carbon, nitrogen and sulphur. Following the basic principle of "you are what you eat", the isotopic compositions of food sources are incorporated into body tissues and are preserved in the organic fraction of bone or more specifically the bone collagen (Ambrose 1993; Kohn 1999). On the basis of modern collagen, several reliable criteria have been identified to test the chemical integrity of archaeological bone (DeNiro 1985; Ambrose 1990; van Klinken 1999; Nehlich and Richards 2009). Measures of collagen stable isotope ratios are therefore regarded to truly reflect the *in vivo* dietary signature. As each isotope system is driven

by different chemical and ecological mechanisms, different types of food may have different ranges of isotope values. This forms the basis of palaeodietary reconstruction.

2.1. Carbon & Nitrogen

Carbon stable isotopes are expressed as the ratios between the heavy (^{13}C) and the light (^{12}C) isotope of the element carbon as $\delta^{13}C$, and are measured in ‰ units relative to the international standard Pee Dee Belemnite (PDB), a carbonate rock in South Carolina, USA. In terrestrial ecosystems, variation in $\delta^{13}C$ is mainly a result of different photosynthetic carbon reduction pathways (C_3, C_4 or CAM) in green plants. The underlying mechanism is the fractionation in isotope ratio which occurs during chemical processes, e.g. within a plant or animal tissue and is due to the slightly different chemical properties of the two isotopes (Peterson and Fry 1987; Ambrose 1993). C_4-plants produce significantly higher $\delta^{13}C$ values than C_3-plants, providing a useful method to trace the domestication and production of C_4-plants; one example is maize from the New World (Vogel and van der Merwe 1977). While temperate Europe is clearly dominated by C_3-plants, solely the C_4-plant millet (*Panicum miliaceum)* has been introduced with Neolithic expansion. According to $\delta^{13}C$ data, millet was an important staple food in the Bronze and Iron Ages in some parts of Europe (Murray and Schoeninger 1988; Tafuri *et al.* 2009). Apart from plants, $\delta^{13}C$ values correspond to certain positions in the food web, as a specific isotope fractionation (+1-2‰) occurs with every step in the food chain (DeNiro and Epstein 1978). Moreover, variation in $\delta^{13}C$ within terrestrial C_3-dominated ecosystems can correspond to the density and position within a forest canopy (Tieszen 1991; van der Merwe and Medina 1991) or can be affected by latitude and altitude (Körner *et al.* 1991). The $\delta^{13}C$ values of marine ecosystems are relatively uniform and mainly determined by atmospheric carbon, planktonic photosynthesis and respiration as well as by the underlying sedimentation. The $\delta^{13}C$ ratios in freshwater systems can vary largely due to the various potential sources of carbon (Boutton 1991). Measurements of $\delta^{13}C$ in archaeological bone collagen reflect the carbon fraction from the dietary protein component, but do not encompass the main component of dietary carbon, the carbohydrates and fats. While the analysis of the mineral phase of bone (apatite) does represent these components (Ambrose and Norr 1993; Kellner and Schoeninger 2007), its validity remains controversial due to largely unsolved contamination and alteration issues (Koch *et al.* 1997).

The stable isotope ratio of nitrogen ($\delta^{15}N$) is expressed as the ratio between the heavy (^{15}N) and the light (^{14}N) isotope and is described in ‰ units in relation to the standard value of atmospheric air (AIR), which is ~0‰. In terrestrial ecosystems, nitrogen enters the biosphere from the atmosphere mainly via nitrogen fixing soil bacteria which is then utilized by plants. In terrestrial plants, variation in $\delta^{15}N$ is mainly driven by climate, temperature, precipitation and salinity (Heaton *et al.* 1986; van Klinken *et al.* 2000). On the other hand, variation in $\delta^{15}N$ of consumers is dominated by the trophic level effect. Similar to carbon, $\delta^{15}N$ fractionates in every step in the food chain within the tissue of an organism, leading to an enrichment of approximately 2-5‰ in each trophic level (DeNiro and Epstein 1981; Minagawa and Wada 1984; Hedges and Reynard 2007). As aquatic environments have enriched plant baseline values and exhibit more complex and nested food webs, $\delta^{15}N$ values in marine and freshwater animals are significantly higher than in terrestrial species (Schoeninger and DeNiro 1984). The combination of both isotope systems allows for characterizing herbivorous, omnivorous and carnivorous diets and differentiating between aquatic and terrestrial food sources. As nitrogen is most abundant in the protein fraction (amino acids) of body tissues, data obtained from bone collagen mainly reflects the isotopic composition of dietary protein (Ambrose 1993). Hence, the relative dietary contribution from plant or animal protein, or a mixture of both, can be detected in archaeological bone samples. In humans, differences in the levels of animal protein consumption can be correlated to social status, assuming that meat and milk can be considered desirable foods (Le Huray *et al.* 2006). It is important to note that due to similar trophic levels of meat and milk (and milk-products), these two types of protein cannot be differentiated using stable isotopes. However, this fact is particularly useful in studying breastfeeding and weaning behaviour in ancient humans and animals. Infants living on their mother's milk are isotopically enriched compared to the nursing female, and their dietary signal appears as carnivorous (Fuller *et al.* 2006). The presence or absence of such a nursing signal can provide information on the nursing and weaning age in prehistoric populations (Katzenberg and Pfeiffer 1995; Schurr 1998).

2.2. Sulphur

Stable sulphur isotope analysis in bone collagen has been shown to be a promising new tool for dietary reconstructions. The ratio of the heavy (^{34}S) versus the light (^{32}S) isotope of sulphur is

expressed as $\delta^{34}S$ and measured in ‰ units relative to the meteorite standard Canyon Diablo Troilite (CDT). While it had been difficult to analyse sulphur in archaeological samples due to technical constraints, recent improvements allow for smaller samples sizes and lower measurement errors (Giesemann *et al.* 1994; Morrison *et al.* 2000). Furthermore, this new analytical approach was strengthened by the introduction of quantity controls for bone collagen to assure the integrity of $\delta^{34}S$ ratios in collagen (Nehlich and Richards 2009). The main application of $\delta^{34}S$ in archaeological material is to differentiate between marine, freshwater and terrestrial dietary sources (Craig *et al.* 2006; Nehlich *et al.* 2010; Nehlich and Wahl 2011). Within different ecosystems, anaerobic bacteria fractionate sulphur isotopes (Canfield 2001) and cause strong variations in $\delta^{34}S$ values in freshwater, marine and terrestrial ecosystems, ranging from -22‰ to +20‰. Organisms living in marine ecosystems have $\delta^{34}S$ values close to +20‰, whereas purely terrestrial animals have values lower than +10‰, typically between +2‰ and +6‰ (Peterson and Fry 1987; Richards *et al.* 2003). Freshwater values may fall between these two extremes, depending on the conditions for sulphur cycling in the respective aquatic biotope. The first studies on prehistoric populations consuming freshwater fish have shown the reliability of this method in detecting aquatic resource utilization (Nehlich *et al.* 2010; Nehlich and Wahl 2011).