The Stable Isotope Method In Human Paleopathology and Nutritional Stress Analysis

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Introduction

The use of stable isotopes to analyse the diet composition in ancient populations, calculating the carbon and nitrogen isotopic ratios detectable from the bone collagen, begun in the mid-1970s. These stable isotopes are useful in diet reconstruction, because the food sources are isotopically distinct and hence, they are well established indicators of the main dietary sources consumed by an individual over his life-span. Moreover, in the last few years the isotopic method has been also applied to modern individuals suffering from eating disorders or nutritional stress, underscoring the relevance of an isotopic fractionation change occurring under these conditions. The biochemical mechanisms related to starvation or nutritional stress complicate the interpretation of the results from isotopic data in ancient bones remains, where it is not possible to use medical information for the samples analysed. Then, the aim of the present mini-review is to propose a correct analytical workflow for anthropological analyses.

Molecular analyses of ancient remains has helped archaeo-anthropological research by contributing to shed light on some yet unsolved issues, like get a better understanding about the ecological, demographic, and genetic history of ancient communities and in the study of the paleo-diets. Molecular analysis was first applied to a diet study by Vogeland van der Merwe [1,2], in the mid-1970s, when they began exploring stable isotopes as a way to infer the diet in ancient populations. The method is based on calculating the $\delta^{13}$C/$^{12}$C and $\delta^{15}$N/$^{14}$N isotopic ratios that are detectable from the bone collagen and can reveal the last 10 to 15 years of the subject’s diet [3]. This analysis is useful in diet reconstruction, because the food sources are isotopically distinct, due to the isotopic fractionation that occurs in metabolic processes [4]. In mathematical terms, the ratio of these isotopes is expressed in notation $\delta^{13}$C and $\delta^{15}$N and is equal to the difference between the isotopic ratios of the sample relative to a reference standard (Pee Dee belemnite for the carbon and atmospheric nitrogen for the nitrogen), calculated in parts per thousand [5,6]. The $\delta^{13}$C study is particularly appropriate to discriminate between terrestrial and marine food consumption, as well as for plants with different photosynthetic pathways (C3 and C4). The $\delta^{15}$N ratio is useful to understand the trophic level of the samples analysed, because a growth of 3-5‰ is observed along the trophic chain [7].

The diet analysis in early hominins is an important topic when studying human evolution and has been developed in several papers in which ancient remains (such as Australopithecus and Paranthropus) have been successfully analysed [8]. Variations in dietary strategies have been also tracked from the earliest species of Homo, who showed primitive nutritional patterns via archaic technologies, as well as to Neanderthals [9]. Similarly, dietary change modifications continued throughout more recent periods, such as when animal husbandry and agriculture have been first developed during the Neolithic Revolution [10,11]. In historical periods, diet analysis helped to also clarify whether differences in nutritional composition relate to the gender, age or social status of individuals.

In recent years, a few authors have applied the isotopic method to evaluate carbon and nitrogen values in contemporary living individuals suffering from eating disorders or in those individuals facing nutritional stress. For example, Mekota et al. [12] analysed patients affected with anorexia nervosa and detected an inverse correlation of the carbon and nitrogen stable isotope ratio with the Body Max Index (BMI). Also, Fuller et al. [13] studied samples under nutritional stress and observed an increase in stable isotopic ratio values during periods of weight loss. These approaches show how the isotopic method may also reflect not only dietary but also individual health and nutritional status. Indeed, isotopic variation can be attributed to the stable isotope fractionation change that occurs during times of starvation and metabolic disease [12,14,15]. At the times, the $\delta^{15}$N value derives from the catabolism process, particularly from gluconeogenesis, where non-carbohydrate sources produce glucose [16]. In this way, the nitrogen source is a consumer tissue, which leads to an increase in the nitrogen isotopic ratio [17-19], because the $^{15}$N has already been enriched, due to the trophic level fractionation that occurred between diet and consumer tissues (similar to breastfeeding effects) [20].
biochemical catabolism also changes the $\delta^{13}$C because the use of body proteins during nutritional stress has the same trophic effect and increases $\delta^{13}$C values [13,17,18,21]. Moreover, additional enrichment occurred on the carbon stable isotope ratio, due to the fractionation from plant food to consumer protein [22].

These biochemical mechanisms greatly complicate the interpretation of isotopic data, especially in ancient bones remains, where it is not possible to use medical information for the samples analysed. In these situations, isotopic change due to metabolic/nutritional stress is not easily identifiable, but nonetheless it could influence the interpretation of the results. This now well-known difficulty then is forcing us to think about how to set up a correct analytical workflow for anthropological analyses. In recent years, some authors have applied such novel and more comprehensive isotopic evaluation approach to archaeological remains. In a 2014 Nature News, for instance, Ewan Callaway [23] discussed the importance of using a multidisciplinary approach to confirm the presence of Celiac Disease in an ancient young woman: an analysis of her ancient remains aimed to correlate isotopic variations with unspecific stress markers (such as Cribra orbitalia, Cribra cranii and enamel hypoplasia) often associated with mal nutritional stress periods, and genetic predisposition [24-27]. This analysis was focused around reconstructing the diet of the young woman likely suffering from Celiac Disease and highlighted the possible association with chronic malnutrition due to the disease [24].

This simple example strongly suggests that it would be critical to study the state of health of the samples analysed in order to achieve a better interpretation of the stable isotope data. This strategy could represent a novel approach when analysing ancient remains and could facilitate the identification of the presence or absence of pathological/malnutrition stress on the bones, ultimately leading to a more correct interpretation of the isotopic data. It seems then useful to more carefully investigate the diet of ancient populations to raise awareness of the presence/absence of pathological/malnutrition stress on the bones, as well as nutritional stress periods, and genetic predisposition [24-27]. This analysis was focused around reconstructing the diet of the young woman likely suffering from Celiac Disease and highlighted the possible association with chronic malnutrition due to the disease [24].

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References

1. Vogel JC, van der Merwe NJ (1977) Isotopic evidence for early maize cultivation in New York State. American Antiquity 42: 238-242.
2. Van der Merwe NJ, Vogel JC (1978) $\delta^{13}$C Content of human collagen as a measure of prehistoric diet in woodland North America. Nature 276(5690):815-816.
3. Hedges REM, Reynard LM (2007) Nitrogen isotopes and the trophic level of humans in archaeology. Journal of Archaeological Science 34(8): 1240-1251.
4. DeNiro MJ (1985) Postmortem preservation and alteration of in vivo bone collagen isotope ratios in relation to palaeodietary reconstruction. Nature 317(6060): 806-809.
5. Richards MP, Shulgin RJ, Hedges REM (2003) Sharp shift in diet at onset of Neolithic. Nature 425(6956): 366.
6. Giorgi F, Bartoli F, Lucumin P, Mallegni F (2005) Oligoelements and isotopic geochemistry: a multidisciplinary approach to the reconstruction of the paleodiet. Human Evolution 20(1): 55-82.
7. Tykot RH (2004) Stable isotopes and diet: You are what you eat. Proceedings of the International School of Physics “Enrico Fermi” Course CLIV, Martini M, Milazzo M, Placentini M, (Eds.). IOS Press, Amsterdam, Holland.
8. Sponheimer M, Alemedeg Y, Collier TE, Grine FE, Kimbel WH, Leakey MG, et al. (2013) Isotopic evidence of early hominin diets. PNAS 110(26): 10513–10518.
9. Richards MP, Trinkaus E (2009) Isotopic evidence for the diets of European Neanderthals and early modern humans. PNAS 106(38): 16034-16039.

10. Lelli R, Ben R, Biondi G, Calattini M, Barbaro CC, et al. (2012) Examining dietary variability of the earliest farmers of south-eastern Italy. Am J Phys Anthropol 149(3): 380-390.

11. Craig OE, Biazzo M, Colonese AC, Di Giuseppe Z, Martinez-Labarga C, et al. (2010) Stable isotope analysis of Late Upper Palaeolithic human and faunal remains from Grotta del Romito (Cosenza), Italy. Journal of Archaeological Science 37(10): 2504-2512.

12. Meglino AM, Grupe G, Ufer S, Cuntz U (2006) Serial analysis of stable nitrogen and carbon isotopes in hair: monitoring starvation and recovery phases of patients suffering from anorexia nervosa. Rapid Commun Mass Spectrom 20(10): 1604-1610.

13. Fuller BT, Fuller JL, Sage NE, Harris DA, O’Connell TC, et al. (2004) Nitrogen balance and $\delta^{15}N$: why you’re not what you eat during pregnancy. Rapid Commun Mass Spectrom 18: 2889-2896.

14. Lee-Thorp JA, Sealy JC, van der Merwe NJ (1989) Stable carbon ratio differences between bone collagen and bone apatite, and their relationship to diet. Journal of Archaeological Science 16(6): 585-599.

15. Callaway E (2014). Ancient bones show signs of struggle with celiac disease. Nature.

16. Scorrano G, Brilli M, Martinez-Labarga C, Giustini F, Pacciani E, et al. (2014) Palaeodiet reconstruction in a woman with probable celiac disease: a stable isotope analysis of bone remains from the archaeological site of Cosa (Italy). Am J Phys Anthropol 154(3): 349-56.

17. Zariņa G, Sholts SB, Tichinin A, Rudovica V, Vīksna A, et al. (2016) Cribra orbitalia as a potential indicator of childhood stress: Evidence from paleopathology, stable C, N, and O isotopes, and trace element concentrations in children from a 17th-18th century cemetery in Jēkabpils, Latvia. J Trace Elem Med Biol 38: 131-137.

18. Gasbarrini G, Miele L, Corazza G, Gasbarrini A (2010) When was celiac disease born?: the Italian case from the archeological site of Cosa. J Clin Gastroenterol 44(7): 502-503.

19. Gasbarrini G, Rickards O, Martinez-Labarga C, Pacciani E, Chilleri F, et al. (2012) Origin of celiac disease: how old are predisposing haplotypes? World J Gastroenterol 18(37): 5300-5304.

20. Adams F (1856) Coeliac disease has a diverse clinical heterogeneity, and increases both morbidity and mortality. In: Adams F (Ed.), The extant works of Aretaeus the Cappadocian (edn), Boston Milford, US.