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Stable Isotopes Reveal Nitrogen Loading to Lake Tanganyika from Remote Shoreline Villages

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Abstract Access to safe water is an ongoing challenge in rural areas in Tanzania where communities often lack access to improved sanitation. Methods to detect contamination of surface water bodies, such as monitoring nutrient concentrations and bacterial counts, are time consuming and results can be highly variable in space and time. On the northeast shore of Lake Tanganyika, Tanzania, the low population density coupled with the high potential for dilution in the lake necessitates the development of a sensitive method for detecting contamination in order to avoid human health concerns. We investigated the potential use of nitrogen and carbon stable isotopes of snail tissues to detect anthropogenic nutrient loading along the northeast shore of Lake Tanganyika. $\delta^{15}N$ of snails was positively related to human population size in the nearest village, but only for villages with >4000 inhabitants. The areal footprint of villages within their watershed was also significantly correlated with snail $\delta^{15}N$, while agricultural land use and natural vegetation were not. Dissolved nutrient concentrations were not significantly different between village and reference sites. Our results indicate that nitrogen isotopes provide a sensitive index of local nutrient loading that can be used to monitor contamination of oligotrophic aquatic environments with low surrounding population densities.

Keywords $\delta^{15}N$ · Anthropogenic nutrient loading · Oligotrophic · Lake Tanganyika

Introduction

Anthropogenic contamination of surface water negatively affects human health (WHO 2013) and local ecosystems (Tewfik et al. 2005). In East Africa, less than 50 % of the population has access to sanitized drinking water, and the problem is most extensive in rural areas (WHO 2013). In rural areas of Tanzania, people often extract water directly from natural water bodies, usually for want of alternative water sources. Moreover, waste water is rarely processed before being returned to the ground or nearby water bodies, due to a lack of financial resources and infrastructure (Mayo and Mubarak 2015). A method for the quick and efficient detection of surface water contamination in rural areas in Tanzania is needed to safeguard the most accessible supplies of potable water.

Lake Tanganyika in East Africa is a primary source of water for nearby villages, towns, and cities, but the nearshore environment is also heavily used for fishing, shipping, transportation, and bathing. Maintaining Lake Tanganyika's water quality is fundamental to the health and livelihood of coastal communities. Preserving the nearshore zone as a source of clean water, while allowing a diversity of other uses, is particularly important for the many widely dispersed villages along the shoreline. Small villages rarely have the capacity to process human waste or convey it away from areas where people draw water. Nor do they have the infrastructure for consistent water quality monitoring. The development of sensitive, integrative indicators of

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contamination is a first step in designing local strategies for avoiding and remediating contamination of coastal water supplies.

Standard metrics of water quality include inorganic nutrient concentrations (Munda 1993) and bacterial counts (McLellan and Salmore 2003). However, both can be highly variable in space and time, and routine monitoring is required to detect levels that are detrimental to human health. Lack of laboratory facilities and reliable refrigeration precludes the use of these approaches in remote areas. Moreover, when the receiving ecosystem is large relative to the input rates, anthropogenic nutrients can be incorporated into the food web (Karube et al. 2010; Vermuellen et al. 2011) long before changes in nutrient concentrations become detectable through bulk water sampling (Cloern 2001). Taxonomic composition of algae, fish or invertebrates can sometimes provide reliable integrated metrics of anthropogenic contamination (Munda 1993). However, these analyses are time consuming, depend upon access to taxonomic experts, and require either pre-degradation surveys or identification of appropriate reference sites (Costanza et al. 2001). The ideal tool for early detection of impacts of human nutrient loading in remote areas would require minimal field expertise and training, could be easily processed or preserved without specialized equipment, and would provide an index of anthropogenic impact that is sensitive, time-integrated, and spatially synoptic.

Nitrogen stable isotopes are widely used as a time-integrated tracer of nutrient loading (Karube et al. 2010; Vermuellen et al. 2011; Xu and Zhang 2012). Stable isotope values of aquatic food webs are substantially altered by sewage contamination (Costanza et al. 2001; Savage 2005; Tewfik et al. 2005; Vander Zanden et al. 2005; Vermuellen et al. 2011), sometimes allowing detection of loading even without long-term sampling. Human sewage has an elevated δ^{15}N value compared to industrial fertilizers and biologically-fixed nitrogen (Costanza et al. 2001), but high loading rates from any source can encourage bacterial transformations of nitrogen compounds that yield elevated δ^{15}N. The incorporation of anthropogenic nitrogen by primary producers results in a higher δ^{15}N value, which is then transferred to all subsequent trophic levels (McClelland et al. 1997). This elevation of δ^{15}N is detectable in aquatic ecosystems even at relatively low sewage loading rates (McClelland et al. 1997) and at fine spatial scales (Vermuellen et al. 2011). Monitoring primary consumer δ^{15}N modulates some short-term temporal variation in the primary producer signal and provides a reliable time-integrated index of baseline δ^{15}N (Vander Zanden and Rasmussen 1999; Vermuellen et al. 2011; Post 2002). While carbon stable isotopes (δ^{13}C) are not regularly used to detect human sewage contamination, they are used to trace the incorporation of different sources of organic carbon into aquatic food webs (Fry 1991) and as an index of ecosystem metabolism (Bade et al. 2004; O'Reilly et al. 2005; Verburg 2007). Thus, δ^{13}C values may reflect shifts in energy flow associated with nutrient enrichment (Vadeboncoeur et al. 2003).

Previous studies that have used stable isotopes to detect anthropogenic nitrogen in aquatic environments have focused on densely populated areas (Rogers 1999; Savage and Elmgren 2004; Lapointe et al. 2005; Cole et al. 2005), watersheds with high inputs of nitrogen from agricultural activities (Fry et al. 2003; Diebel and Vander Zanden 2009) or on areas with known inputs from septic systems (Lapointe et al. 2005). The efficacy of stable isotopes as a tool to detect sewage impacts in areas with low population density and high dilution rates is untested, and both of these factors could reduce the pollution signal to the point of being undetectable relative to background variation. Nonetheless, the stable isotope analysis of primary consumers has the potential to be an effective monitoring tool in Tanzania where contamination resulting from a lack of waste water treatment can lead to serious human health issues.

Lake Tanganyika is a large tropical lake where permanent stratification of the water column creates chronic nutrient scarcity (Kilham and Kilham 1990). Localized upwelling events are the main source of nutrients to the nearshore zone (Plisnier et al. 1999, Langenberg et al. 2003a; Corman et al. 2010) because inputs from riverine flow and atmospheric deposition are low (Langenberg et al. 2003b). Available nutrients are quickly taken up by algae (Corman et al. 2010) and incorporated into the food web (O'Reilly et al. 2002). The naturally low nutrient levels in this lake, the patchy distribution of human settlements along the shoreline, and the rapid incorporation of added nutrients into the food web make the Lake Tanganyika littoral zone a novel system for testing the applicability of stable isotopes to detect anthropogenic nutrient loading.

In this study, we test the efficacy of the stable isotope method for detecting anthropogenic contamination in rural areas with a low population density and no waste processing facilities. We measured dissolved water column nutrient concentrations and δ^{15}N values of benthic primary consumers to compare their value as indicators of anthropogenic nutrient loading along the northeast shore of Lake Tanganyika. We also conducted a land use analysis, as δ^{15}N values may be correlated with human activity in aquatic environments, reflecting anthropogenic nitrogen loading patterns (e.g. Peterson et al. 2007). Given the oligotrophic nature of Lake Tanganyika, and the potential for the rapid uptake of nutrients by aquatic biota, we expected that dissolved nutrient concentrations would not be correlated with village population size. Yet we expected that δ^{15}N values of primary consumers, as temporal integrators of nutrient dynamics, would be positively correlated with village population size and village land use footprint. Our aim was to detect low levels of anthropogenic nutrient loading, as this may be correlated with increased levels of bacteria and viruses in areas lacking improved sanitation, thus posing a human health risk. If successful, this method can be applied as a simple proxy to identify and monitor sites where sewage contamination may be a health concern. Our survey sites included two lakeside villages where the Jane Goodall Institute (JGI) has initiated human health improvement programs that include the construction of beach latrines accompanied by education programs. We used samples from both of these villages to assess whether beach latrines effectively reduce the detectable human influence on the $\delta^{15}N$ value of benthic primary consumers.

Methods

Land Use Analysis

The study area along the northeast shore of Lake Tanganyika was bounded to the south by the Malagarasi River and to the north by the border with Burundi (Fig. 1). Land use analysis was conducted in ArcGIS 10.2 (ESRI, USA) using the world imagery base map. A watershed map supplied by The Nature Conservancy was created from an SRTM 90 m digital elevation model using the hydrology tools within ArcGIS. The satellite imagery used to categorize land use was captured on 2 October 2009 and 1 February 2010, and had a resolution of 0.5 m with an accuracy of 10.2 m. The watershed encompassing each village was classified into three classes of land cover: 'village', 'agricultural' or 'natural vegetation'. The village category included buildings, compounds, roads, and their environs; the agricultural category comprised land that had been cleared and planted with crops; and the natural vegetation category included land with grasses, shrubs and trees. Shoreline areas were classified as village where evidence of human activity (boats, buildings, etc.) was visible; otherwise they were included in the 'natural vegetation' category. Land use within the watershed of each reference site was uniformly natural vegetation.

Dissolved Nutrient Concentrations

Sampling for dissolved water column nutrient concentrations and primary benthic consumers was conducted on July 21st and August 5–6, 2010. We sampled 11 village sites, including the two JGI beach latrine and education villages. We matched each village site with a similar nearby (<2 km distance) reference site that lacked evidence of human activity near the shoreline. Water samples were collected at 1 m depth from each sampling site using a syringe, then filtered (0.45 µm, Whatman GFX) directly into new, rinsed HDPE bottles and then kept frozen until analysis. Nitrate (NO_3^-) and soluble reactive phosphate (SRP) were quantified using standard colorimetric methods (cadmium-reduction and molybdate-blue, respectively) on an autoanalyzer (detection limit of 1 µg L^{-1}). Ammonium (NH_4^+) was analyzed with a Turner Aquaflor following Taylor et al. (2007).

Stable Isotope Analysis

Samples for stable isotope analysis were collected from rocky substrates directly in front of villages as close to the center of human activity (washing areas, boat landings) as possible. For stable isotope analysis, we collected six snails between 0–3 m depth at each site. Where possible, we collected the same species of snail (*Lavigeria nassa*). However, only other *Lavigeria* species were present at some sites, and at a few sites with little rocky habitat we could find only *Paramelania* species. Previous work has shown that all *Lavigeria* and *Paramelania* species collected in the same site have comparable $\delta^{15}N$ values (McIntyre et al. unpublished data). Snails from each site were paired by similarity in shell length, and their muscle tissue was combined to yield three replicate analyses per site. This allowed us to control for potential effects of size-based variation, while at the same time averaging results across multiple individuals to achieve representative data and minimize analytical costs. For each snail, muscle tissue was separated from the shell, operculum, and brood pouch, placed in a glass vial, and oven-dried (60°C, 48 h). Each replicate was then homogenized and subsampled (1.2 mg) for analysis. Stable isotope ratios of carbon (C) and nitrogen (N) were quantified using a Finnigan Delta-Plus isotope ratio mass spectrometer at the Cornell University Stable Isotope Laboratory. Isotope ratios are expressed using the standard $\delta^{13}C$ and $\delta^{15}N$ notation relative to certified standards (PDB carbonate, N_2 gas). High analytical precision ($SD < 0.2$ % for both $\delta^{13}C$ and $\delta^{15}N$; $n = 28$) was demonstrated using an in-house standard (trout muscle tissue) interspersed throughout the sample run.

Statistical Analysis

Population data for the shoreline villages were obtained from censuses conducted by JGI in 2007. Population growth rates in this area are high (often >4 % annually), but more recent data were not available. Total and percent village and agricultural land use were tested for a linear relationship with the $\delta^{15}N$ of primary consumers collected along the watershed shoreline. Dissolved nutrient concentrations and stable isotope values were compared

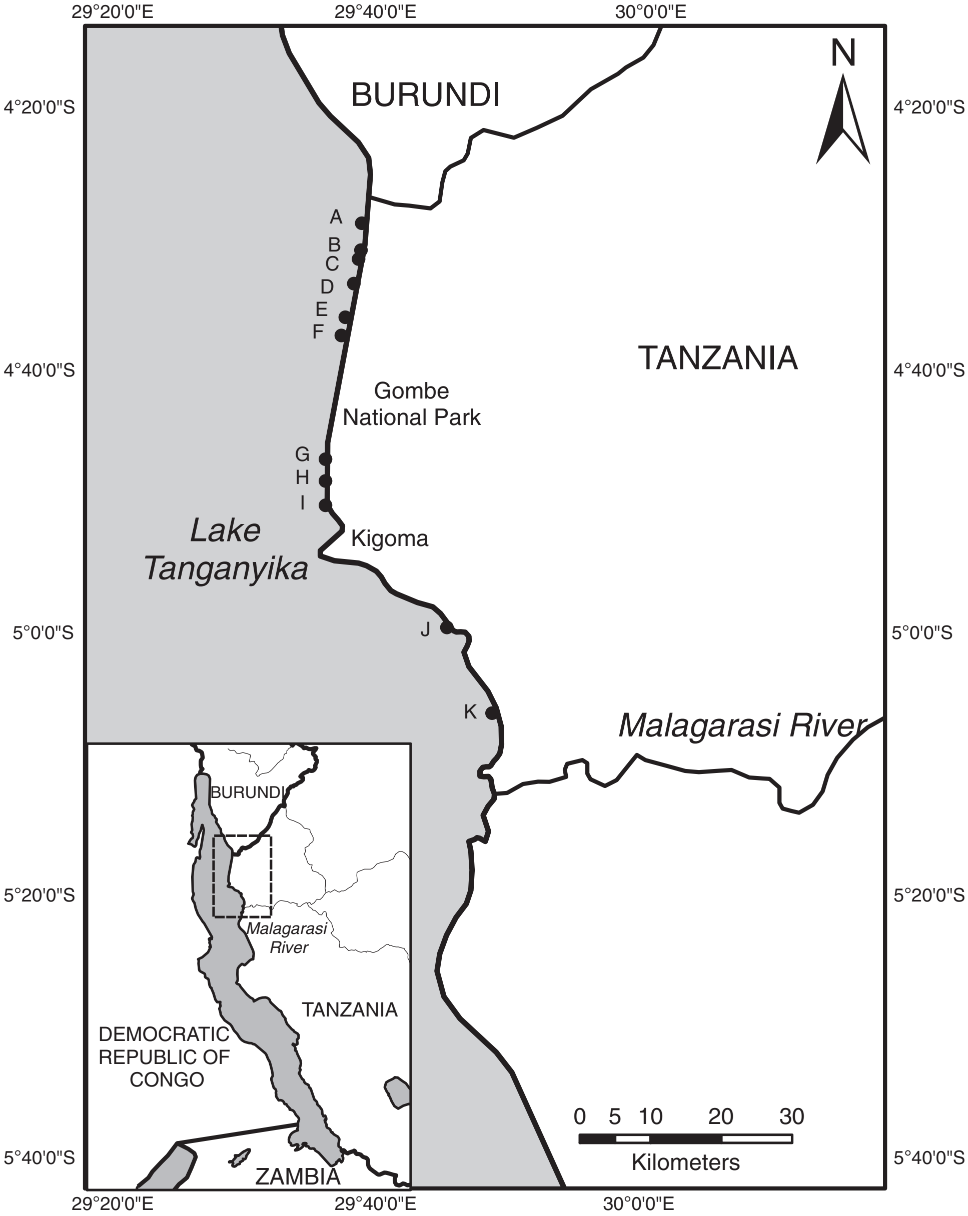

Fig. 1 Map of Northeastern coast of Lake Tanganyika including sampling sites and village locations

between all reference and village sites using a paired student's *t*-test when assumptions were satisfied (SRP, $\delta^{13}C$), or a Mann-Whitney *U* test otherwise (NO_3, NH_4^+, $\delta^{15}N$) (Zar 2010). A Welch's *t*-test was used to test for a significant difference in $\delta^{15}N$ and $\delta^{13}C$ between each village-reference pair. We used analysis of covariance (ANCOVA) to test whether the slopes of $\delta^{15}N$ against population differed between village and reference sites, where reference sites were assigned the population of the nearest village. The relationships between population and $\delta^{15}N$, $\delta^{13}C$, NO_3–N, SRP and NH_4^+ were determined using linear regressions. All analyses were performed in R (R Core Team 2014).

Results

Land Use Analysis

Within watersheds, village areas were most commonly located near the shoreline, buffered by a stretch of beach and/or vegetation. Agricultural areas were rarely located close to the shore. Rather, natural vegetation and/or village areas separated the shoreline from agricultural areas. Percent land use ranged from 0 to 57.0 for the agricultural category, from 11.1 to 92.9 for the natural vegetation category, and from 3.4 to 58.5 for the village category. There was no significant correlation between $\delta^{15}N$ and log total agricultural or natural vegetation area (m^2) within village watersheds ($p > 0.05$). However, there was a significant correlation between log total village land use and δ^{15}N ($r^2 = 0.52$, $p < 0.05$) (Fig. 2). There was no significant correlation between δ^{15}N and percent agricultural land use, while the percent village land use within a watershed was positively correlated with δ^{15}N ($r^2 = 0.51$, $p < 0.05$).

Dissolved Nutrient Analysis

Nutrient concentrations in nearshore waters were uniformly low (Table 1). The range in concentrations for NO_3^-, SRP and NH_4^+ overlapped widely between reference sites and village sites, and there were no significant differences between nutrient concentrations at reference and village sites. Mean concentrations for villages were 10.0, 2.0, and 6.3µg L^{-1}, for NO_3^-, SRP and NH_4^+, respectively, while mean concentrations for reference sites were 9.8, 1.3, and 4.2 µg L^{-1}. Since nutrient sampling was not replicated, it was not possible to test for differences between specific village-reference pairs. However, a regression analysis of reference site nutrient concentrations on village site nutrient concentrations for village-reference pairs showed no significant correlation for NO_3^- ($F_{1,7} = 0.77$, $p > 0.05$, $r^2 = -0.04$), SRP ($F_{1,9} = 1.71$, $p > 0.05$, $r^2 = 0.07$) or NH_4^+ ($F_{1,8} = 0.76$, $p > 0.05$, $r^2 = 0.02$). Thus, reference locations close to villages with relatively high nutrient concentrations did not necessarily have high nutrient concentrations, and vice versa.

NO_3^- was negatively related to population ($F_{1,8} = 7.06$, $p < 0.05$, $r^2 = 0.40$) while SRP ($F_{1,9} = 2.24$, $p > 0.05$, $r^2 = 0.11$) and NH_4^+ ($F_{1,8} = 0.27$, $p > 0.05$, $r^2 = -0.08$) were not related significantly to population (Fig. 3). NO_3^- ($F_{1,25} = 2.71$, $p > 0.05$, $r^2 = 0.06$), SRP ($F_{1,28} = 2.29$, $p > 0.05$, $r^2 = 0.04$), and NH_4^+ ($F_{1,27} = 0.18$, $p > 0.05$, $r^2 = 0.03$) concentrations were not significantly related to snail δ^{15}N. SRP

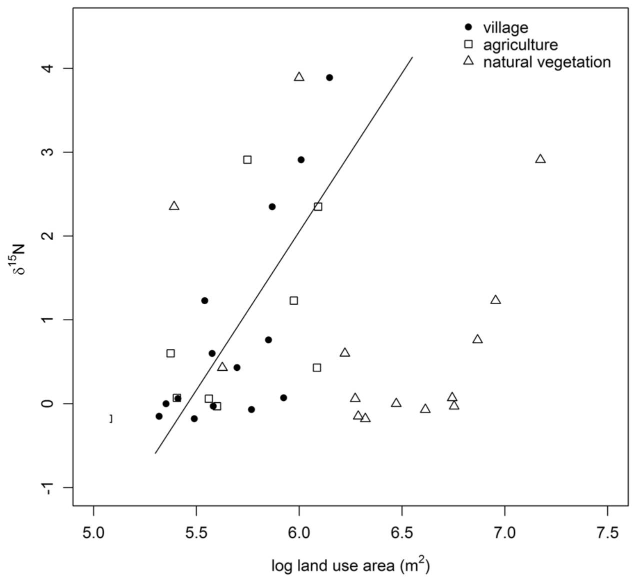

Fig. 2 δ^{15}N values in snail tissue and land use area (log m^2) of village, agricultural, and natural vegetation. The correlation between δ^{15}N and land use was significant for village area only, as depicted by the line ($r^2 = 0.52$, $p < 0.05$)

Table 1 Nutrient concentration range and means for village and reference sites

Nutrient	Type	Range µg/L	Mean µg/L
NO_3^-	Reference	0.29–26.79	9.8
	Village	0–36.13	10.0
SRP	Reference	0.3–3.08	1.3
	Village	0.26–5.55	2.0
NH_4^+	Reference	0.68–12.84	4.2
	Village	1.33–13.89	6.3

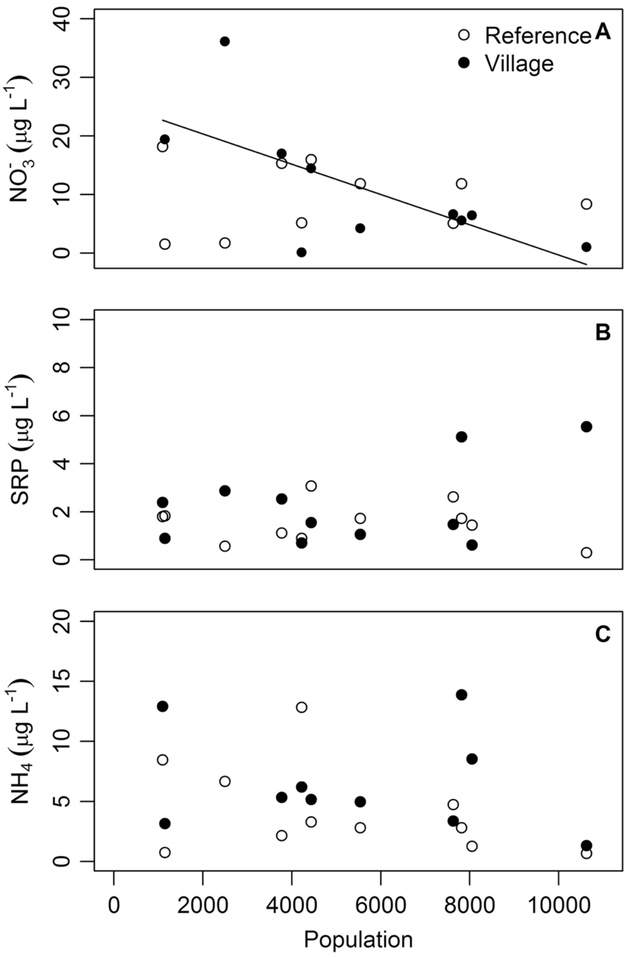

Fig. 3 Nutrient concentrations with site population for NO_3^- (**a**); SRP (**b**) and; NH_4^+ (**c**). Reference sites were assigned the population of the nearest village. A significant correlation was found for village nitrate concentrations with population only ($p < 0.05$, $r^2 = 0.40$). Note the change in the y axis

($F_{1,28} = 6.71$, $p < 0.05$, $r^2 = 0.16$) and NH_4^+ ($F = {}_{1,27} = 7.62$, $p > 0.05$, $r^2 = 0.19$) were negatively related to snail $\delta^{13}C$, while NO_3^- ($F_{1,25} = 0.23$, $p > 0.05$, $r^2 = 0.03$) was not significantly related to snail $\delta^{13}C$.

Stable Isotope Analysis

Mean $\delta^{15}N$ was 0.06 % ± 0.35 (SD) at reference sites, which was significantly different from the mean for village sites of 2.13 % ± 2.98 (Mann Whitney *U*, $W = 941.5$, $p < 0.05$) (Table 2). Mean $\delta^{13}C$ was −12.77 % ± 1.08 at reference sites, and −13.71 % ± 1.48 at village sites, and a paired *t*-test indicated no significant difference between values at reference and village sites ($t = 1.51$, df = 10, $p > 0.05$). There was a significant positive relationship between snail $\delta^{15}N$ and village population ($F_{1,9} = 16.93$, $p < 0.05$, $r^2 = 0.61$) (Fig. 4), but not snail $\delta^{13}C$ and village population ($F_{1,9} = 0.90$, $p > 0.05$, $r^2 = 0.01$). The ANCOVA analysis resulted in a significant effect of site type (village vs. reference) on $\delta^{15}N$ ($F_{3,17} = 11.77$, $p < 0.05$) (Fig. 4) but not $\delta^{13}C$ ($F_{3,17} = 2.54$, $p > 0.05$) when controlling for population. Welch's *t* test results for village-reference pairs are

Table 2 Location, population and mean stable isotope values ± (SD) for village sites and their paired reference sites

Site	Latitude (S)	Longitude (E)	Population	$\delta^{13}C$		$\delta^{15}N$	
				Village	Reference	Village	Reference
A	4 28.946	29 39.260	7817	−16.76 (0.43)	−11.84 (0.28)	2.91 (0.24)	−0.32 (0.21)
B	4 31.000	29 38.999	5540	−12.61 (0.18)	−11.84 (0.28)	0.00 (0.39)	−0.32 (0.21)
C	4 31.678	29 39.172	1145	−11.78 (0.05)	−11.97 (0.05)	−0.15 (0.11)	0.02 (0.36)
D	4 33.537	29 38.789	7626	−11.88 (0.24)	−11.57 (0.21)	−0.03 (0.32)	−0.13 (0.10)
E	4 36.110	29 38.444	3773	−12.39 (0.08)	−11.98 (0.39)	−0.07 (0.05)	0.02 (0.04)
F	4 37.495	29 38.167	4434	−13.80 (0.29)	−11.98 (0.39)	0.76 (0.21)	0.02 (0.04)
G	4 46.909	29 36.159	4219	−12.32 (0.19)	−12.48 (0.24)	1.23 (0.35)	−0.15 (0.14)
H	4 48.573	29 36.481	2496	−12.76 (0.37)	−14.47 (0.54)	−0.18 (0.07)	0.20 (0.13)
I	4 50.423	29 36.660	1094	−15.00 (0.99)	−14.91 (0.36)	0.07 (0.14)	−0.08 (0.25)
*J	4 59.878	29 46.048	8049	−13.71 (0.81)	−13.44 (0.33)	2.35 (1.60)	0.41 (0.17)
*K	5 06.252	29 48.679	10623	−14.64 (1.63)	−11.77 (0.16)	3.89 (1.07)	−0.14 (0.13)

Some village sites share a reference site, when the reference site was located equidistant between them.

* indicates involvement in the JGI education and beach latrine program

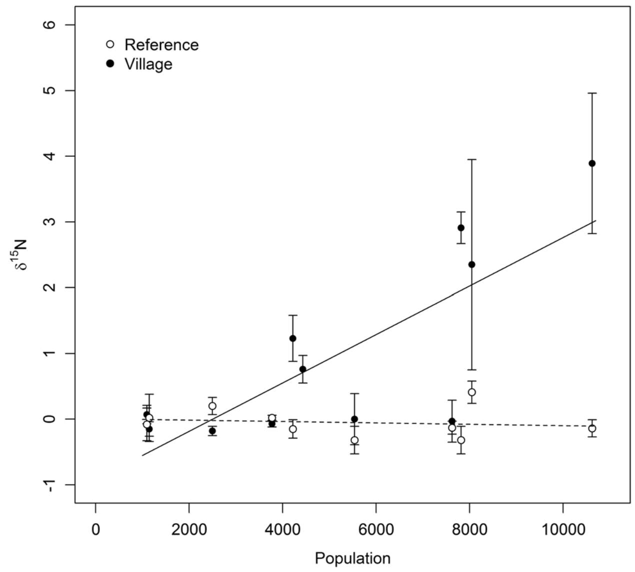

Fig. 4 Linear regression of $\delta^{15}N$ values against population. The solid line represents the linear relationship for the village sites while the dashed line represents the linear relationship for the reference sites

Table 3 Welch's *t* test results for village-reference comparisons of $\delta^{13}C$ and $\delta^{15}N$

Site	$\delta^{13}C$			$\delta^{15}N$		
	t value	degrees of freedom	*p* value	*t* value	degrees of freedom	*p* value
A	16.73	3.47	<0.001	17.38	3.92	<0.001
B	3.99	3.41	0.022	1.25	3.09	0.298
C	3.86	3.57	0.022	0.77	2.42	0.508
D	1.51	2.49	0.245	0.5	2.5	0.658
E	1.79	2.15	0.205	2.26	3.45	0.098
F	6.48	3.72	0.004	5.77	2.12	0.025
G	0.91	3.78	0.417	6.35	2.66	0.011
H	4.53	3.54	0.014	4.59	2.99	0.019
I	0.16	2.5	0.886	0.91	3.12	0.429
J	0.68	6.97	0.515	2.93	5.22	0.031
K	4.28	5.19	0.007	8.94	5.27	<0.001

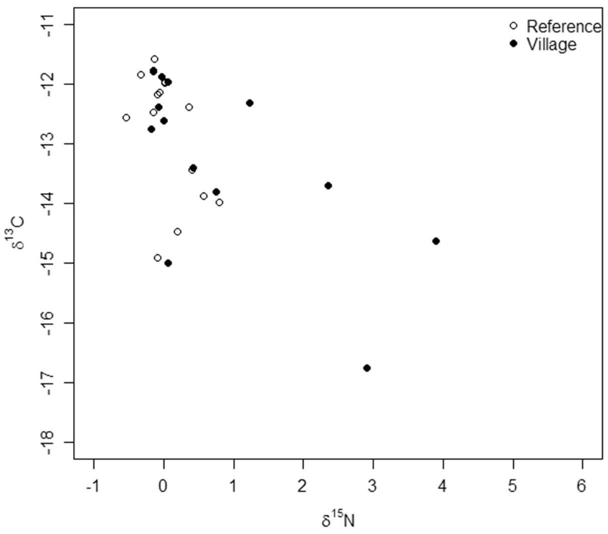

Fig. 5 $\delta^{15}N$ and $\delta^{13}C$ values from snail tissue collected at reference and village sites

reported in Table 3. Across all sites, snail $\delta^{15}N$ and $\delta^{13}C$ were significantly negatively correlated ($p < 0.05$, $r^2 = 0.31$) (Fig. 5). At both villages where JGI had constructed a beach latrine, $\delta^{15}N$ was significantly higher than the nearest reference site.

Discussion

Nitrogen stable isotope values in primary consumers provided a sensitive assay of local nutrient loading despite the large volume into which nutrients are diluted in Lake Tanganyika. Significant differences in nitrogen stable isotope values were often detected between village sites and the closest reference site, while there was no significant difference between village and reference site nutrient concentrations. The positive relationship between snail $\delta^{15}N$ and village population size, and village area within the watershed, is strong evidence that nitrogen stable isotopes can detect nutrient loading from villages.

Water column nutrient concentrations showed no indication of nutrient loading along the coast of Lake Tanganyika. Indeed, there were no differences in nitrogen and phosphorus concentrations between village and reference sites, nor was there a positive correlation between dissolved nutrients and village population size. We infer that nutrients from anthropogenic sources have a low residence time in nearshore waters, either because nutrients are rapidly sequestered by primary producers (McClelland et al. 1997; Corman et al. 2010) or because water movement along the shoreline dilutes and transports the local load. Previous studies indicate that in Lake Tanganyika, nutrient concentrations in the littoral and pelagic zones are similar (Corman et al. 2010) and that nutrients are taken up quickly by biota (O'Reilly et al. 2002; Corman et al. 2010). Rapid incorporation of nutrients into the food web may have prevented the detection of anthropogenic nutrient loading using dissolved nutrient concentrations, even at sites where contamination was indicated by stable isotope data.

The relationships between nitrogen stable isotope values and village population sizes ($r^2 = 0.61$), and village land use area ($r^2 = 0.51$), coupled with significant differences between $\delta^{15}N$ at multiple village-reference pairs, indicates that anthropogenic nutrients are affecting water quality in the nearshore area of Lake Tanganyika. This could represent contamination from human waste, but other anthropogenic sources of nitrogen could also be involved in elevating $\delta^{15}N$ values of benthic primary consumers. Agriculture is practiced around most villages, and both animal manure fertilizers and synthetic agricultural fertilizers can lead to enriched $\delta^{15}N$ in aquatic food webs (Diebel and Vander Zanden 2009). However, no livestock were observed during sampling, and our land cover analysis suggests that agricultural land area is not a predictor of $\delta^{15}N$ values. Furthermore, substantial agricultural contributions to nutrient loads are unlikely during our dry-season sampling period, when lack of rainfall precludes substantial runoff from the land surface.

Some villages were used for docking fishing boats, and the adjacent beaches were used for drying fish catches, resulting in high volumes of fish being transported through the sampling area. Most studies examining fish effects on $\delta^{15}N$ in aquatic and terrestrial systems focus on spawning migrations rather than the effects of fisheries on nearshore environments (Bilby et al. 1996; Childress et al. 2014). Fish from Lake Tanganyika have higher $\delta^{15}N$ values than our snail samples (Campbell et al. 2008; Wagner et al. 2009), but the impact of cleaning or drying harvested fishes at landing beaches on snail $\delta^{15}N$ is unknown. Overall, we consider human wastes to be the most likely cause of elevated $\delta^{15}N$ in snails, because $\delta^{15}N$ is significantly linearly related to the local population size of villages, as well as to the total land area within the village watershed categorized as village. However, population size could also correlate with fishery inputs, livestock density, or other input pathways that we could not quantify. In any case, our findings provide compelling evidence of the efficacy of using nitrogen stable isotopes as a time-integrated index of localized anthropogenic nutrient loading in oligotrophic, low population density environments.

Some village sites inhabited by thousands of people exhibited $\delta^{15}N$ values that were just as low as nearby reference sites. It is possible that in these areas, the nearshore environment may be more efficiently flushed by currents or wave action, exporting anthropogenic inputs before they enter the food web. Using satellite imagery, we did not find any obvious association between shoreline orientation or shape which would influence wave and current energy and the magnitude of $\delta^{15}N$ enrichment. Alternatively, villages with populations of around 4000 or more which do not exhibit significantly higher snail $\delta^{15}N$ values relative to reference sites may have devised solutions for dealing with wastes that could be transferable to other locations. It would be worthwhile to engage in dialog with these villages to determine their waste disposal practices and whether the practices could be implemented at other villages.

Although much of the variability in $\delta^{15}N$ can be explained by population, it was difficult to determine the factors affecting $\delta^{13}C$ of snails. We expected any correlation between $\delta^{13}C$ and nutrient concentrations (and therefore nutrient inputs) to be positive because high primary productivity can lead to enriched carbon isotope values (Meyers and Ishiwatari 1993; O'Reilly et al. 2005; Verburg 2007). However, $\delta^{13}C$ values were weakly but negatively correlated with SRP and NH_4^+ as well as $\delta^{15}N$. Therefore, it is unlikely that increased benthic primary productivity is driving the $\delta^{13}C$ values in this study. Other potential factors, such as the presence of the Malagarasi River delta to the south of the sampling region also fail to explain $\delta^{13}C$ values. Dissolved inorganic carbon (DIC) in the Malagarasi River may be depleted in $\delta\ ^{13}C$ as a result of respiration of organic matter from the upstream wetlands, relative to the rest of the lake where carbon isotope values may be dominated by atmospheric exchange (Bade et al. 2004). However, in this study, $\delta^{13}C$ was not correlated with the distance from the delta. Moreover, Verburg (2007) suggested that the impact of the Malagarasi River on Lake Tanganyika would be minimal and localized because river water is cold relative to lake water, thereby forcing it to sink to the bottom rather than mixing into the nearshore waters. Measuring the $\delta^{13}C$ of DIC was beyond the scope of this study, but could help to elucidate the factors responsible for the $\delta^{13}C$ variation that we observed. It is possible that nutrient loading from villages increased phytoplankton biomass (Vadeboncoeur et al. 2014), thereby decreasing benthic productivity due to shading. This could have produced more negative snail $\delta^{13}C$ either by depressing benthic algal $\delta^{13}C$ or by supplementing snail diets with sedimenting phytoplankton (Vadeboncoeur et al. 2003; Devlin et al. 2013). Unfortunately, we cannot test these potential explanations with the available data.

No baseline data were collected prior to the introduction of the current JGI latrine and education programs, hence we cannot draw strong inferences about its overall impact except to say that nitrogen loading was still evident at both sites that have received these interventions. Indeed, it is possible that latrines may serve as a source of nutrients if latrine effluent reaches the lake (e.g. Knappett et al. 2011). Given that the two villages with the highest population also had elevated $\delta^{15}N$ and were participants in the JGI program, the presence of beach latrines may be a confounding factor in our study. However, several other villages which were not participants in the JGI program also displayed elevated $\delta^{15}N$ values, suggesting that the stable isotope method is a sensitive method for detection of nutrient loading.

Nitrogen isotopes clearly indicated anthropogenic inputs from villages with more than 4000 people, but may be less effective at detecting contamination from smaller villages. Our sample size for the snail analysis was low (3 replicates), potentially preventing us from detecting subtle changes in contamination. As well, it is still necessary to investigate the potential link between detection of contamination via $\delta^{15}N$ values and human health, by conducting parallel measurements of coliform bacteria, incidences of schistosome parasites (liver flukes, which have an aquatic life stage), diarrheal disease incidence, or other health metrics. Indeed it is possible that parallel methods would reveal potential health risks at villages where $\delta^{15}N$ values were indistinguishable from reference sites, and vice versa. In addition, the applicability of this method between seasons should be confirmed. The significant differences we found between village and reference site $\delta^{15}N$ were large, yet variation between wet and dry seasons has been noted in tropical aquatic environments and may confound the interpretation of stable isotope data (e.g. Lau et al. 2009).

In addition to being used as a detection tool for anthropogenic nutrient loading of surface water along the shoreline of Lake Tanganyika and other oligotrophic, low population density areas, this stable isotope method could also be applied towards achieving water quality objectives. For example, this method could be used to help optimize design and placement of beach latrines in rural areas. We found fine-scale variation in $\delta^{15}N$ when we sampled snails at multiple sites along the shoreline of individual villages; the apparent localized variation in $\delta^{15}N$ of slow moving primary consumers (Michel et al. 2007) could be helpful to identify specific pathways by which nutrients reach the lake near each village, and point to optimal latrine locations or other nutrient diversion strategies. As well, this method may be sensitive enough to help identify nutrient diversion strategies that are most effective. Spatially explicit stable isotope sampling within a site before and after implementing waste management strategies would enable rigorous evaluation of the efficacy of any interventions. In addition, the straightforward nature of sampling snails provides an opportunity for the involvement of citizen scientists. Given the importance of community input and participation for the success of sanitation programs (Mayo and Mubarak 2015), the potential for the monitoring program to be driven by community members may encourage program uptake and effectiveness.

The significant differences in $\delta^{15}N$ between many villages and reference sites suggest that nitrogen stable isotope ratios are a viable tool for detecting and monitoring anthropogenic nutrient loading along the shoreline of this large tropical lake with low population densities and high dilution potential. Analysis of nitrogen isotope ratios of snails could provide a straightforward way to rapidly assess anthropogenic nutrient loading provided that there is funding for sample shipment and laboratory analysis. The overall costs of collecting and analyzing stable isotope samples are reasonable, given the high spatial resolution, time-integration, and sensitivity of this method to detect nutrient loading. Additionally, programs aiming at reducing nutrient loading can use $\delta^{15}N$ to identify sites where further testing for human health risks is necessary, prioritize remediation sites and assess effectiveness of interventions over time. Thus, this method can be pursued in rural areas to further environmental management and human health.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no competing interests.

References

Bade DL, Carpenter SR, Cole JJ, Hanson PC, Hesslein RH (2004) Controls of δ^{13}C-DIC in lakes: geochemistry, lake metabolism, and morphometry. Limnol Oceanogr 49:1160–1172

Bilby RE, Fransen BR, Bisson PA (1996) Incorporation of nitrogen and carbon from spawning coho salmon into the trophic system of small streams: evidence from stable isotopes. Can J Fish Aquat Sci 53:163–173

Campbell L, Verburg P, Dixon DG, Hecky RE (2008) Mercury biomagnification in the food web of Lake Tanganyika (Tanzania, East Africa). Sci Total Environ 402:184–191

Childress ES, Allan JD, McIntyre PB (2014) Nutrient subsidies from iteroparous fish migrations can enhance stream productivity. Ecosystems 17:522–534

Cloern JE (2001) Our evolving conceptual model of the coastal eutrophication problem. Mar Ecol-Prog Ser 210:223–253

Cole ML, Kroeger KD, McClelland JW, Valiela I. (2005) Macrophytes as indicators of land-derived wastewater: application of a δ^{15}N method in aquatic systems. Water Resour Res 41. doi:10.1029/2004WR003269

Corman JR, McIntyre PB, Kujoba B, Mbemba W, Fink D, Wheeler CW, Gans C, Michel E, Flecker AS (2010) Upwelling couples chemical and biological dynamics across the littoral and pelagic zones of Lake Tanganyika, East Africa. Limnol Oceanogr 55:214–234

Costanza SD, O'Donohue MJ, Dennison WC, Loneragan NR, Thomas M (2001) A new approach for detecting and mapping sewage impacts. Mar Pollut Bul 42:149–156

Devlin SP, Vander Zanden MJ, Vadeboncoeur Y (2013) Depth-specific variation in carbon isotopes demonstrates resource partitioning among the littoral zoobenthos. Freshwater Biol 58:2389–2400

Diebel MW, Vander Zanden MJ (2009) Nitrogen stable isotopes in streams: effects of agricultural sources and transformations. Ecol Appl 19:1127–1134

Fry B (1991) Stable isotope diagrams of freshwater food webs. Ecology 72:2293–2297

Fry B, Gace A, McClelland JW (2003) Chemical indicators of anthropogenic nitrogen-loading in four Pacific estuaries. Pac Sci 57:77–101

Karube Z, Sakai Y, Takeyama T, Okuda N, Kohzu A, Yoshimizu C, Nagata T, Tayasu I (2010) Carbon and nitrogen stable isotope ratios of macroinvertebrates in the littoral zone of Lake Biwa as indicators of anthropogenic activities in the watershed. Ecol Res 25:847–855

Kilham P, Kilham SS (1990) Endless summer: internal loading processes dominate nutrient cycling in tropical lakes. Freshw Biol 23:79–89

Knappett PS, Escamilla V, Layton A, McKay LD, Emch M, Williams DE, Huq R, Alam J, Farhana L, Mailloux BJ, Ferguson A (2011) Impact of population and latrines on fecal contamination of ponds in rural Bangladesh. Sci Total Environ 409:3174–3182

Langenberg VT, Sarvala J, Roijackers R (2003a) Effect of wind induced water movements on nutrients, chlorophyll-a, and primary production in Lake Tanganyika. Aquat. Ecosyst Health 6:45–58

Langenberg VT, Nyamushashu S, Roijackers R, Koelmans A-A (2003b) External nutrient sources for Lake Tanganyika. J Great Lakes Res 29:169–180

Lapointe BE, Barile PJ, Littler MM, Littler DS (2005) Microalgal blooms on southeast Florida coral reefs II. Cross-shelf discrimination of nitrogen sources indicates widespread assimilation of sewage nitrogen. Harmful Algae 4:1106–1122

Lau DCP, Leung KMY, Dudgeon D (2009) What does stable isotope analysis reveal about trophic relationships and the relative importance of allochthonous resources in tropical stream? A synthetic study from Hong Kong. Freshw Biol 54:127–141

Mayo AW, Mubarak T (2015) Challenges of adoption of urine-diversion dry toilets technology as sanitation option by coastal communities of Mkuranga District in Tanzania. Afr. J Environ Sci Technol 9:482–492

McClelland JW, Valiela I, Michener RH (1997) Nitrogen stable isotope values in estuarine food webs: A record of increasing urbanization in coastal watersheds. Limnol Oceanogr 42:930–937

McLellan SL, Salmore AK (2003) Evidence for localized bacterial loading as the cause of chronic beach closings in a freshwater marina. Water Res 37:2700–2708

Michel E, Mcintyre PB, Chan J (2007) A snail's space sets a snail's pace: movement rates of *Lavigeria* gastropods in Lake Tanganyika, East Africa. J Molluscan Stud 73:195–198

Meyers PA, Ishiwatari R (1993) Lacustrine organic geochemistry–an overview of indicators of organic matter and diagenesis in lake sediments. Org Geochem 20:867–900

Munda IM (1993) Changes and degradation of seaweed stands in the northern Adriatic. Hydrobiologia 260-261:239–253

O'Reilly CM, Hecky RE, Cohen AS, Plisnier PD (2002) Interpreting stable isotopes in food webs: recognizing the role of time averaging at different trophic levels. Limnol Oceanogr 47:306–309

O'Reilly CM, Dettman DL, Cohen AS (2005) Paleolimnological investigations of anthropogenic environmental change in Lake Tanganyika: VI. Geochemical indicators. J Paleolimnol 34:85–91

Peterson GS, Sierszen ME, Yurista PM, Kelly JR (2007) Stable nitrogen isotopes of plankton and benthos reflect a landscape-level influence on Great Lakes coastal ecosystems. J Great Lakes Res 33:27–41

Plisnier PD, Chitamwebwa D, Mwape L, Tshibangu K, Langenberg V, Coenen E (1999) Limnological annual cycle inferred from physical-chemical fluctuations at three stations of Lake Tanganyika. Hydrobiologia 407:45–58

Post DM (2002) Using stable isotopes to estimate trophic position: models, methods, and assumptions. Ecology 83:703–718

R Core Team (2014) R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, http://www.R-project.org/

Rogers KM (1999) Effects of sewage contamination on macro-algae and shellfish at Moa Point, New Zealand using stable carbon and nitrogen isotopes. New Zeal J Mar Fresh 33:181–188

Savage C (2005) Tracing the influence of sewage nitrogen in a coastal ecosystem using stable nitrogen isotopes. Ambio 43:145–150

Savage C, Elmgren R (2004) Macroalgal (*Fucus vesiculosus*) δ^{15}N values trace decrease in sewage influence. Ecol Appl 14:517–526

Taylor BW, Keep CF, Hall Jr RO, Koch BJ, Tronstad LM, Flecker AS, Ulseth AJ (2007) Improving the fluorometric ammonium method: matrix effects, background fluorescence, and standard additions. J N Am Benthol Soc 26:167–177

Tewfik A, Rasmussen JB, McCann KS (2005) Anthropogenic enrichment alters a marine benthic food web. Ecology 86:2726–2736

Vadeboncoeur Y, Jeppesen E, Vander Zanden JM, Schierup HH, Christoffersen K, Lodge DM (2003) From Greenland to green lakes: cultural eutrophication and the loss of benthic pathways in lakes. Limnol Oceanogr 48:1408–1418

Vadeboncoeur Y, Devlin SP, McIntyre PB, Vander Zanden MJ (2014) Is there light after depth? Distribution of periphyton chlorophyll and productivity in lake littoral zones. Freshw Sci 33:524–536

Vander Zanden MJ, Rasmussen JB (1999) Primary consumer δ^{13}C and δ^{15}N and the trophic position of aquatic consumers. Ecology 80:1395–1404

Vander Zanden MJ, Vadeboncoeur Y, Diebel MW, Jeppesen E (2005) Primary consumer stable nitrogen isotopes as indicators of nutrient source. Environ Sci Technol 39:7509–7515

Verburg P (2007) The need to correct for the Suess effect in the application of δ^{13}C in sediment of autotrophic Lake Tanganyika, as a productivity proxy in the Anthropocene. J1 Paleolimnol 37:591–602

Vermuellen S, Sturaro N, Gobert S, Bouquegneau JM, Lepoint G (2011) Potential early indicators of anthropogenically derived nutrients: a multiscale stable isotope analysis. Mar Ecol-Prog Ser 422:9–22

Wagner CE, McIntyre PB, Smith K, Michel E, Gilbert D (2009) Diet predicts intestine length in Lake Tanganyika's cichlid fishes. Funct Ecol 23:1122–1131

World Health Organization. (2013) Progress on sanitation and drinking-water 2013 update.

Xu J, Zhang M (2012) Primary consumers as bioindicator of nitrogen pollution in lake planktonic and benthic food webs. Ecol Indic 14:189–196

Zar JH (2010) Biostatistical analysis, 5th edn. Prentice Hall, Upper Saddle River, NJ