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Short communication

Testing the Protein Leverage Hypothesis in a free-living human population ☆

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ABSTRACT

The Protein Leverage Hypothesis (PLH) predicts that humans prioritize protein when regulating food intake. We tested a central prediction of PLH: protein intake will remain more constant than fat or carbohydrate in the face of dietary changes in a free-living population. Data come from a large sample of adult Filipino women participating in the Cebu Longitudinal Health and Nutrition Survey (CLHNS) located in Philippines. Longitudinal data analysis showed that, as predicted, calories of dietary protein remained more constant over time than calories of dietary carbohydrates or fat, even if corrected for the low proportional contribution of protein to dietary energy.

Introduction

Dramatic changes in diet and lifestyle, referred to as the nutrition transition, are occurring in low and middle income countries where economic development and urbanization are proceeding rapidly (Bray & Popkin, 2007; Colin Bell & Bishai, 2002; Eckhardt et al., 2003). Despite rising rates of obesity globally, how individuals regulate the quantity and types of food items that they consume remains only partially understood. Some studies indicate that food choices are associated with taste, cost and convenience (Mela, 2006). In addition, there is growing evidence that specific macronutrients have different impacts on appetite and the regulation of energy intake (Jebb, 2007). While all three macronutrients – carbohydrates, fat and protein – exert some degree of influence on total energy intake, protein is the most satiating and tightly regulated (Gosby et al., 2011; Griffioen-Roose, Mars, Finlayson, Blundell, & De Graaf, 2011; Halton & Hu, 2004; Soenen & Westerterp-Plantenga, 2008; Weigle et al., 2005).

Simpson and Raubenheimer developed the Protein Leverage Hypothesis (PLH) which postulates that, as a consequence of relatively strong protein appetite compared with the other two macronutrients, humans adjust their food intake to maintain a relatively constant dietary protein intake and consequently will have higher energy intakes on diets with low protein density (Griffioen-Roose et al., 2012; Simpson, Batley, & Raubenheimer, 2003; Simpson & Raubenheimer, 2005). There is growing support for PLH from experimental studies and meta-analyses of macronutrient intake regulation (Gosby et al., 2011; Simpson & Raubenheimer, 1997, 2005; Simpson et al., 2003; Sørensen, Mayntz, Raubenheimer, & Simpson, 2008). However, PLH has yet to be tested outside of experimental settings in free-living humans. In this study, we tested a central prediction of PLH that calories of dietary protein will remain more constant over time than calories of dietary carbohydrates or fat in the face of dietary changes in a free-living population over a two decade period (from 1986 to 2005).

Materials and methods

Data come from the Cebu Longitudinal Health and Nutrition Survey (CLHNS), which began in 1983 and follows a cohort of women residing in Metropolitan Cebu City in the central Philippines (Adair, Popkin, et al., 2011). Cebu, with a population nearing 2 million, is one of the fastest growing and most rapidly developing regions of the Philippines and shares many characteristics with other large cities in transitional Asian countries. For instance, the mean real household income in Cebu nearly doubled among study participants from the 1980s to 1990s, which was also accompanied by shifts in diet and physical activity patterns (Colchero, Caballero, & Bishai, 2008). The CLHNS has been reviewed and approved by the

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University of North Carolina Institutional Review Board, Office of Human Research Ethics (Adair, Gultiano, & Suchindran, 2011).

We used these longitudinal data to test the hypothesis that protein intake will remain more constant than fat or carbohydrates. We analysed 2031 women's 24-h dietary recall data from 1986, 1994, 1998, 2002, and 2005. We also tested the relationship between socioeconomic factors, such as household income and urbanization, on the pattern of macronutrient intakes in this population undergoing shifts in diet and lifestyle.

Variables

At each sampling time, dietary intake was measured by trained interviewers using two 24-h recalls, and their mean was used in the analyses. Food intakes were converted to macronutrient and energy intakes using the Philippines Food Composition Tables produced by the Food and Nutrition Research Institute of the Philippines (FNRI, 1997). Calories of dietary protein (PRO), carbohydrates (CHO), and fat (FAT) were calculated by multiplying grams intake by four (calories per gram) for PRO and CHO, and by nine for FAT.

Household income was derived from the sum of all household members' cash-income and the value of in-kind earnings; for compatibility over time, income values were deflated using year-appropriate Philippines consumer price indices. Urbanization was derived from an urbanicity index score based on seven criteria: population size, population density, communication, transportation, healthcare services, education, and market availability (Colchero & Bishai, 2008; Dahly & Adair, 2007; Victora et al., 2008).

Analysis

Longitudinal analyses were used to characterize the changes in macronutrient intakes during the 20-year period (from 1986 to 2005). All analyses were performed using SAS V9.2. To investigate differences in the change in energy intake from PRO, CHO and FAT over time, a general linear mixed model was fitted to the data with both time and macronutrient (PRO, CHO or FAT) as repeated measures. A spatial power covariance structure was fitted to the measurements within a subject over time, with different variance estimates for each macronutrient; subject was included as a random variable. Age at time 1, macronutrient, time (measured in years) and the macronutrient-time interaction were included as explanatory variables. The interaction, testing the difference in slopes over time for the different macronutrients, was the test statistic of interest. As the distribution of energy intake was skewed, the log of energy intake was used as the dependent variable.

To investigate whether household income and urbanization influenced the difference in change in macronutrient intake over time, general linear mixed models were fitted to the data with both time and macronutrient (PRO, FAT or CHO) as repeated measures. A spatial power covariance structure was fitted to the measurements within a subject over time, with different variance estimates for each macronutrient and subject included as a random variable. Age at time 1, macronutrient, time (measured in years) and household income or urbanicity index, and the 2 and 3 way interactions of time and macronutrient with household income or urbanicity index were included as explanatory variables with the 3 way interaction testing whether the difference in slopes over time for the different macronutrients differed depending on household income or urbanicity index. As the distribution of calorie intake was highly right skewed, the log of calorie intake was used as the dependent variable. Where the 3 way interaction was found to be significant random coefficients general linear mixed models were fitted separately for calories from PRO, FAT and CHO. Time was a repeated measure with individual subject intercepts and slopes across time being modelled as random effects. Age at time 1, macronutrient, time (measured in years), household income, urbanicity index, and the interactions of time with income and urbanicity index were included as explanatory variables. Initially, income and urbanicity index were included as continuous variables, but in order to investigate the interactions they were categorised: urbanicity index into six equally space levels, and household income into 12 mainly equally spaced levels (tail categories grouped because of small numbers). As the distribution of energy intake was skewed, the log of energy intake was used as the dependent variable.

Results

Macronutrient intake, household income and urbanicity index varied over time (Table 1). The slopes of the relationship between time and caloric intake from each macronutrient differed significantly ($p < 0.0001$). PRO kcal intake increased slightly over time [slope (SE) = 0.005 (0.0009)], but at a slower rate than FAT kcal intake [slope (SE) = 0.02 (0.002)] while CHO kcal intake decreased slightly [slope (SE) = −0.008 (0.0007)] (Fig. 1).

We next tested the effect of both household income and urbanicity index on change in individual macronutrient intake over time. There was strong evidence for differences in the rate of change in macronutrient intake over time by income and urbanicity (both $p < 0.0001$); therefore, the macronutrients were analysed separately. In relation to PRO, there was evidence of a difference in the change of PRO intake over time depending on income ($p = 0.003$) and urbanicity ($p = 0.006$). Although the pattern was not very clear especially across income, the estimates of the slopes were negative and were more negative as income or urbanicity level increased. Also, there was evidence of an effect of both urbanicity ($p < 0.0001$) and income ($p = 0.02$) on change in CHO intake over time, again with the slope becoming more steeply negative as income or urbanicity level increased. Finally, there was strong evidence for an effect of both urbanicity and income (both $p < 0.0001$) on the change in FAT intake over time, again with the

Table 1
Characteristics of CLHNS women across survey years[a].

Survey	1986	1994	1998	2002	2005
N	2031	1880	1831	1656	1380
Age, y	28.5 (20.7–40.5)	37.8 (30–50.1)	42 (34–53.8)	44.9 (37.4–57.2)	48 (40–59.6)
BMI, kg/m^2	20.1 (16.7–25.8)	22.8 (17.6–30.2)	23.3 (17.2–30.7)	24.3 (17.6–31.6)	24.3 (17.7–31.9)
Energy intake, kcal	1205.9 (494.2–2546.7)	1283.9 (563.5–2436.8)	1278.9 (568–2433.6)	1171 (503.2–2479.8)	1088.3 (485.6–2118.2)
Protein intake, kcal	141.4 (57.7–335)	164.0 (68.6–363.2)	160 (64–366.4)	163.7 (58–394.2)	158 (60.2–363.6)
Carbohydrates intake, kcal	900.8 (394.2–1800.1)	889.6 (418.7–1622.3)	857.5 (422.2–1600.6)	790.7 (363.9–1481.3)	741.3 (356.2–1312.1)
Fat intake, kcal	89.8 (15.1–649.4)	148.9 (23–853.2)	163.5 (25–865.7)	131.8 (19.1–887.7)	137.8 (20.5–648.7)
Household income, pesos	164.4 (34.2–635.9)	381.8 (114.4–1266)	425.9 (157.8–1320.8)	443.4 (150.3–1555.1)	440.1 (139.4–1672.2)
Urbanicity index	30 (8–47)	39 (12–51)	41 (14–56)	45 (16–59)	43 (16–57)

[a] Values are medians (5th–95th percentile).

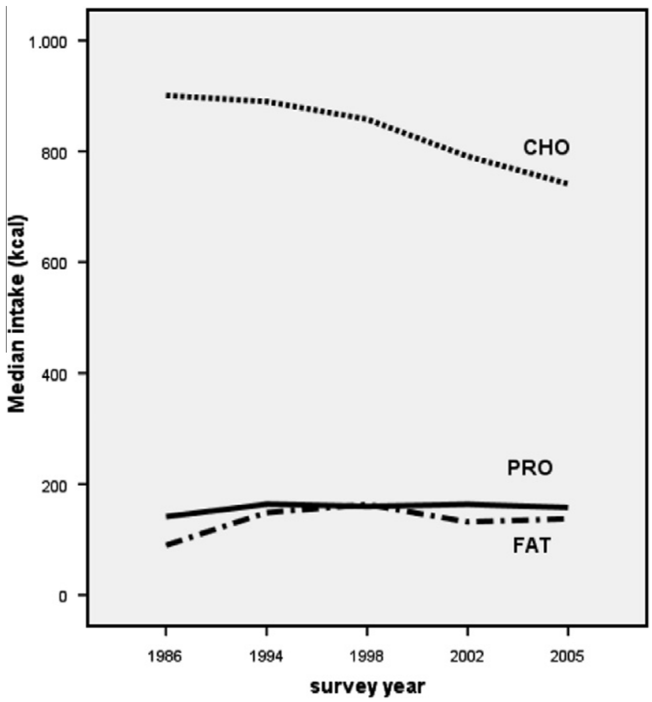

Fig. 1. Macronutrient intakes of CLHNS women across survey years. The slopes of the relationship between time and log of kcal intake from each macronutrient differed significantly ($p < 0.0001$). PRO intake increased slightly over time [slope (SE) = 0.005 (0.0009)], but at a slower rate than FAT intake [slope (SE) = 0.02 (0.002)] while CHO intake decreased slightly [slope (SE) = −0.008 (0.0007)].

slope becoming more steeply negative as urbanicity or income level increased. In summary, the effect of income and urbanicity on the change in macronutrient intake over time showed the same pattern: a larger decrease over time for increasing household income or urbanicity categories; however, the effect on PRO was very small, and that on both FAT and CHO more marked, particularly FAT.

Discussion

In the study population, macro nutrient intake changed during the decades of observation. We hypothesized that protein intake would be most conserved as these changes unfolded. In support of this prediction, our analysis showed that calories of dietary protein remained more constant over time than calories of dietary carbohydrates or fat, even when controlled for the absolute level of each macronutrient in the diet. Although calories of dietary protein increased slightly over time, the increase was at a slower rate than that for fat, while carbohydrates decreased slightly. These findings are consistent with the interpretation that the total amount of protein consumed is more tightly regulated than intake of carbohydrates or fat (Gerbens-Leenes, Nonhebel, & Krol, 2010; Kelles & Adair, 2009; Popkin, 2006; Sheehy & Sharma, 2010).

The increase that we observed in protein intake over the study period, albeit considerably smaller than the decrease in carbohydrates, may reflect an interaction between regulatory biology and socioeconomic changes that affect the relative affordability of different macronutrients (Brooks, Simpson, & Raubenheimer, 2010). The dietary shift we observed is consistent with patterns observed in other low or middle income countries. The socioeconomic structure of the study population changed over time with some female participants moving into higher income and urbanicity categories over the course of the study. Despite the fact that calories of dietary fat and protein increased slightly over time, calorie intake decreased among women who remained in the same income and urbanicity category. As urbanicity or income scores increased over time, the change of macronutrient became more steeply negative: the macronutrient intake of women in the lowest income or urbanicity category showed the smallest decrease while women in the higher income or urbanicity categories showed the highest decrease.

Although previous studies using the CLHNS data have found minimal evidence of selection bias due to attrition of subjects during the course of the study, such problems are worth bearing in mind (Adair, Gultiano, et al., 2011). Another possible limitation of this study is that dietary recall systematically underestimates energy intake; furthermore, two 24-h diet recalls were used when three 24-h recalls are viewed as more reliable. Because all three macronutrients are likely to be influenced similarly by these factors, they are not likely to contribute to the differences in macronutrient intake over time that we observe in the sample.

Balancing these potential limitations, the studýs strengths lie in the detailed environmental, socioeconomic, and demographic information of the CLHNS from individual, household and community-levels that provides an opportunity to explore how changes of macronutrient patterns relate to changing community and household level conditions over a 20-years period. More longitudinal studies need to be conducted to explore dietary shifts in developing countries where the obesity epidemic and its related chronic diseases are the most serious challenge to long-term health. In conclusion, our findings indicate that energy from protein intake remained more constant than that from carbohydrates or fat intake in a human population undergoing shifts in diet and lifestyle over a two decade period, even in different income and urbanization categories. This is consistent with the idea that recent changes in the protein density of the human diet have played a causal role in the developing obesity epidemic – the Protein Leverage Hypothesis (Simpson & Raubenheimer, 2005).

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