

Author's response to reviews

Title: Relative Fat Oxidation is Higher in Children than Adults

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Version: 2 Date: 9 May 2007

Author's response to reviews:

Cover Letter

Below you will find our responses to the reviewer's comments. We have addressed all comments. In particular we have reanalyzed the data to determine whether gender differences(Dullo) explained our findings and whether using the ratio method for expressing RMR affected our results(Piers). Thank you for those valid concerns and as you will see they made a difference in the way we interpreted the data as well as the basic results.

Manuscript

Relative Fat Oxidation is Higher in Children than Adults, Kostyak et al
MS # 1841826004124376

Responses to review by Dr. Leonard S. Piers
First, thank you for your valuable input.

Major Compulsory Revisions

Comment 1

In accordance with your suggestion, we reanalyzed the data according to the recommendations by Poehlman and Toth, AJCN 1995. The crux of that paper was that the intercept of the relationship between resting metabolic rate and fat free mass is not zero. An important distinction between our protocol and that of the referenced paper is that most of our data were collected at a time which was not truly resting. After the first energy expenditure determination our subjects had a meal and were allowed to amulate at a low level. However, we did go back and use the referenced protocol for that first measurement. For this analysis, and to be consistent with the Poehlman and Toth paper, we converted our energy expenditure to kJ/min. The non-zero intercepts we found for adults (1.3) and children (1.73) are similar to the intercepts reported by Poehlman of 1.51 for young men aged 25 yrs. The other important consideration from the Poehlman paper is whether covarying FFM alters the findings from ANOVA that RMR is different between children and adults. When we co-varied LBM it showed that RMR in kcals/lbm was not different between adults and children. The revised manuscript provides this analysis as follows

"The majority of data collected and presented in this study represent non-resting conditions however, differences in resting metabolic rate, i.e. the starting metabolic rate before a perturbation, could influence the interpretation of our results. Differences in body size require caution in interpreting resting energy expenditure when children and adults are compared. As an example, when fat free mass (x axis, kg) is regressed against resting metabolic rate (RMR, kJ/min), the y intercept is not zero. This non-zero intercept can lead to a statistical bias when comparing data from groups that differ greatly in metabolic body size. Our data in this respect for adults are similar to those of Poehlman and Toth who found Y intercepts of 1.35 and 1.51 for women and men respectively. Our intercept for adults was 1.3. One way to eliminate this bias is to use analysis of covariance with LBM as the co-variate which removes the effect of LBM on RMR. When we preformed this analysis the F ratio ($F=7.7$, $p=.08$) showing that the differences between adults and children for RMR was not significant. Thus the greater rate of fat oxidation in children is not due to a greater metabolic rate. "

Comment 2

The reviewer suggest that we consider the information contained in two other references, Elia or Livesey AJCN 1988(two consecutive papers). While these two excellent papers provide exceeding important

information, we suggest that they are not directly relevant to our work for the following reasons:

Livesey and Elia, AJCN 47:608-628,1988.

The overall thesis of this paper is that the use of the non-protein respiratory quotients to estimate energy utilization without consideration of the fuels being utilized can be hazardous. The paper evaluates the effect of conventional and artificial proteins, fats and carbohydrates on heat of combustion, heat equivalent of oxygen and RQ's for a very large number of substrates. Such data would be relevant if our different groups had ingested meals which contained different components. All subjects (children and adults) ingested Ensure thus the types of fat, carbohydrate and protein ingested were identical while the amount of each meal ingested depended on the weight of the subject. We did not feel it is necessary to alter our discussion based on the data from this reference. The author does note that among many interesting observations, an interesting fact is presented in this paper. Combustion of a certain triglyceride from adipose tissue from children and adults results in an RQ which is virtually identical between children and adults (Table 2 page 594).

The second paper Elia and Livesey AJCN 47:591-607 provides important information but again we do not feel it is relevant to our work. That reference presents a wonderful analysis, both theoretical and data-based to show the problems encountered when the nonprotein respiratory quotients exceed 1.0. This situation occurs in several conditions none of which existed in our study. The one condition upon which Elia and Livesey concentrate is net lipid synthesis. The average RQ values we present in the manuscript clearly show that non protein RQ never exceeds 1.0 and most are between 0.8-0.88. We did not feel it is necessary to alter our discussion based on the data from this reference.

Comment 3

The values provided in figures 1 and 2 are the respiratory quotients not adjusted for urinary nitrogen excretion. We chose to present the data in this way to be consistent with most other papers on this topic. Because there were no differences in urinary nitrogen excretion when expressed relative to body weight we do not feel that changing all the data to non-protein respiratory quotients will increase the understanding of our data.

Comment 4

The reviewer asks that we present data in Figure 2 using only two groups, adults and children. The other reviewer asks that we delve more deeply into a discussion about whether the differences we see are valid based on gender as well as age and we have accommodated that request. Specifically " The potential importance of gender in this analysis deserves greater attention". We would like to keep figure 2 as a four group presentation because we have done additional analyses requested by the other reviewer.

Comment 5

We have dropped this sentence as you suggest.

Comments for reviewer Dr. Abdul Dullo

First, Thank you for your comments

Comment 1

According to your suggestion, we reanalyzed the data using a 2 factor ANOVA. The following section was added to the paper based on those results.

"In an attempt to determine the contribution of fat oxidation to daily calorie expenditure, we calculated the grams of fat oxidized per kcal of energy expenditure. This value was higher in children (0.047 \pm 0.01 g/kcal) compared to adults (0.032 \pm 0.01, $p < 0.02$). When groups were analyzed according to gender and age, a 2 factor ANOVA showed fat oxidation for females, both children (0.052 \pm 0.016 g/kcal) and adults (0.042 \pm 0.014) were significantly higher ($F = 6.07$, $p = 0.025$) than values obtained for males of a comparable age (boys 0.041 \pm 0.006 and men 0.023 \pm 0.013 g/kcal). Children, both girls and boys, had higher fat oxidation rates relative to adults ($F = 5.6$, $p = 0.03$). The F ratio for gender by age interactions was not significant, ($F = 0.54$, $p = 0.47$).

Comment 2

We recruited our subjects based on their not being highly unusual in terms of physical activity. Those (adults or children) engaging in significant training would have been excluded although no such individuals volunteered for the study. Children and adults were normally active. Maximal oxygen consumption (ml/kg/min) was a little higher in children but the standard deviations around both group means were large. In response to your inquiry we performed a regression of VO₂max (ml/kg/min) vs fat oxidation (relative to energy expenditure). No correlation existed. We would rather not address this issue to any great extent in the paper because it was not our intention to determine whether exercise training (assuming it and genetics have a large influence on VO₂max) alters fat oxidation. We have added a couple of sentences in the discussion about this issue. Such an inquiry would require selective recruitment based on training status and we did not do this.

Comment 3

We have dropped the expression of metabolic body size.

Comment 4

According to your comment, we have added the following section to the methods section.

"The experimental diet is representative of a typical American diet based on NHANES dietary data. For example, the average intake of SFA, MUFA, and PUFA are: 11%, 15%, and 7%, respectively. In addition, fiber intake is consistent with current average intakes of adults and children in the U.S. Furthermore, dietary cholesterol in the experimental diet is representative of cholesterol intake in males, females and children."

References: MMWR Weekly (CDC). Trends in intake of energy and macronutrients -- United States, 1971-2000. 53:80-82, 2004. Ervin R.B., J.D. Wright, C.Y. Wang, and J. Kennedy-Stephenson. Dietary intake of fats and fatty acids for the United States population: 1999-2000. Advance Data 348:1-8, 2004