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Energy expenditure and intake comparisons in Chilean children 4-5 years attending day-care centres

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Abstract

Introduction: **the doubly labelled water (DLW) method has an accuracy of 1% and within-subject precision of 5-8%, depending on subject's age and environments issues. Energy intake assessment is prone to errors (>15-20%) depending in the method utilized.**

Objective: **to quantify DLW methodology errors in four to five year olds that could affect the comparison with energy intake.**

Methods: **energy expenditure (TEE, by DLW), was assessed during 14 days in 18 preschool children, who attended eight hours daily to day-care centres. Energy intake was determined by a combined method: food weighing during weekdays and recall after leaving the Centre (17h to sleep time) plus 24 h recall, during the weekend. Several assumptions affecting DLW total error were assessed to determine their influence in the comparison to energy intake (i.e. background variability, space ratio, proportion of water subject to fractionation, food quotient value).**

Results: **the individual mean energy expenditure was 1 373 ± 177 kcal and the energy intake (1 409 ± 161 kcal). The overall difference between intake and expenditure was 42.9 kcal/day (limits of agreement + 259.1 to -112.3 kcal/day). TEE measurement error only explained a minor quantity (2.4%), between both measurements, and the observed mean isotope dilution space was 1.030 ± 0.010 confirming the value utilized in adults studies.**

Conclusions: **energy expenditure data is similar to other studies in preschool children. The small difference found between energy intake and expenditure may be attributed to the applied energy intake methodology, the homogeneous diet at care centres during the week-days and the lower DLW methodology error.**

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Key words: *Doubly labelled water. Energy expenditure. Energy intake. Preschool children. Day-care centres.*

COMPARACIÓN ENTRE EL GASTO Y EL CONSUMO DE ENERGÍA EN NIÑOS CHILENOS DE 4-5 AÑOS ASISTENTES A JARDINES INFANTILES

Resumen

Introducción: **el método del agua doblemente marcada (ADM) tiene una precisión del 1% y en un mismo sujeto es de 5-8%, dependiendo de la edad y el entorno del sujeto. La evaluación de la ingesta energética es propensa a errores (> 15-20%), dependiendo del método utilizado.**

Objetivo: **cuantificar los errores metodológicos del ADM en niños de 4-5 años que podrían afectar la comparación con la ingesta de energía.**

Métodos: **el gasto de energía (GTE, por ADM), se evaluó durante 14 días en 18 preescolares, asistentes a guarderías infantiles. La ingesta energética se determinó mediante un método combinado: pesaje de alimentos durante los días de la semana y registro después de salir del centro (17 horas en adelante), además de un recordatorio de 24 horas, durante un día del fin de semana.**

Resultados: **el promedio individual del gasto energético total fue 1373 ± 177 kcal y la ingesta de energía (1409 ± 161 kcal). La diferencia global entre la ingesta y el gasto fue 42,9 kcal/día. El error de medición del GET explicó una variación del 2,4%, entre ambas mediciones, y el espacio de dilución de isótopos fue 1030 ± 0.010, confirmando el valor utilizado en los estudios de adultos.**

Conclusiones: **los datos de GET fueron similares a otros estudios realizados en niños en edad preescolar. La pequeña diferencia encontrada entre la ingesta y el gasto energético se puede atribuir a la metodología de la ingesta de energía aplicada, la dieta homogénea en los centros de atención, durante los días de la semana, y el bajo error metodológico del ADM.**

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Palabras claves: *Agua doblemente marcada. Gasto energético. Ingesta de energía. Preescolares. Guarderías infantiles.*

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Abbreviations

DLW: doubly labelled water.
TEE: total energy expenditure.
FQ: Food Quotient.

Introduction

Near five hundred thousand children receive food at school (twice) or day-care centre (three times), in Chile. The doubly labelled water method (DLW) has been extensively used for the measurement of total energy expenditure (TEE) since the 1980's[1-6] and much of the data is now incorporated into the formulation of human energy requirements[7]. In Chile, DLW methodology has been used to measure energy requirements in both infants[8] and preschool children[9,10,11].

It is reported that in general, the DLW method has a relative accuracy of 1% and within-subject precision of 5-8%[12] but errors of the method should be assessed in the age groups and environments in which it is applied. In Chile, TEE has been measured in infants and preschool age children[8-10] as these age group are relevant in public health nutrition due to the increasing prevalence of obesity in children[13-15]. Validated measurement of energy intake in preschool children is also clearly important for the development of policy recommendations to help prevent obesity from early age[16]. As known TEE must equal energy intake in individuals to achieve energy balance, thus the technique may be used to validate the assessment of energy intake by food weighing or questionnaires[17].

In most reports, energy intake is underestimated whatever the method used for intake measurements[18,19]; Kipnis *et al*. found that the 24 hours food recall method may underestimate energy intake by 12-14% in adult population[20,21]. However, in studies where a person, other than the subject, is responsible for recording dietary intake, energy intake may often correspond to DLW determined energy expenditure[22]. In the present study, we analyze some of the errors derived from the approximations and assumptions in the DLW method (involving isotopic background variation, goodness of fit of the data to the model, isotope dilution spaces, fractionation and respiratory quotient), in the context of a comparison between energy intake and energy expenditure in Chilean preschool children, who attend day-care centres.

Materials and methods

Subjects

The study was carried out on 15 children of low socio-economic status aged 3-5 years. They were selected from public day-care centres in Santiago, Chile, according to the following criteria: birth weight 3000-4000 g., normal nutritional status, no congenital disease and without medication or illness at the time of the study. Children entered the study when their parents had been informed about the experimental protocol and had given written consent. The Ethical Committee of the Institute of Nutrition and Food Technology approved the study. A description of the subjects is provided in table I.

DLW technique

To test the effect of isotope dose two different doses were administered: dose H (high), 0.2 g/kg deuterium oxide (99.9%) and 2.0 g/kg 10% $H_2^{18}O$ for nine children, and dose L (low), 0.08 g/kg deuterium oxide (99.9%) and 1.55 g/kg 10% $H_2^{18}O$ for six children. To assess the significance of baseline variation, seven urine samples of 6 ml were collected daily for a week, after dosing. One urine sample was taken every day, for eight consecutive days. Urine samples were immediately sealed in individual sterile tubes and refrigerated at -18 to -22 ºC.

Hydrogen was equilibrated with water in the presence of a Pt-Al catalyst at 20 ºC for the deuterium analysis[23]. Oxigen-18 labelled samples were equilibrated with 4% CO_2 in He and placed in a thermo-regulated bath at 25 ºC for 15 hours[24]. After equilibration, isotopic content of the samples was measured in a HYDRA IRMS mass spectrometer (Europa Scientific, Crewe, United Kingdom). Measurement precision was 0.4-0.6 ppm for deuterium analysis and 0.2-0.3 ppm for oxygen-18. Slopes and intercepts of the regressions of log enrichment above base line versus time were calculated with a computer program adjusting least minimal squares (LMS) to the data. Propagation of errors in the measurement of CO_2 production was calculated from Cole TJ *et al*. and Ritz P *et al*.[25,26].

Uncorrected CO_2 production rate was corrected by isotope fraction, assuming that a constant proportion of total water losses were fractionated[27]. After this correction, the CO_2 production rate was converted into energy expenditure by assuming a fixed respiratory quotient or a calculate one[28].

Anthropometry

The children were weighed, at the day care-centres, the day that that the DLW protocol started, then at mid time (4-5 days) and finally on the 13-14 days using a standardised SECA scale (model H W 60 with scale up to 60 kg and precision of 0.01 g, Vogel and Halke, Hamburg, FRG).

Dietary intake

Food weighing

Food intake was measured twice during the TEE study period by standard procedures: by weighing all

Table I
Description of subjects participating in the study

Subjects	Sex[1]	Age (y)	Weight (Kg)	Height (cm)	BMI[2] (Kg/m^2)	ZWH[3]	ZHA[4]	Birth Weight (g)	Birth Height (cm)
Higher Dose									
1	M	5.2	20.2	110.3	16.7	0.8	-0.2	3.5	50.5
2	M	4.8	18.6	106.6	16.4	0.6	-0.6	3.9	53.0
3	F	5.4	19.1	111.3	15.4	0.2	0.1	3.2	51.0
4	F	5.1	15.8	102.1	15.1	-0.1	-1.7	3.2	47.0
5	F	4.1	16.0	102.3	15.3	0.0	-0.1	3.8	49.0
6	M	3.8	14.8	96.1	16.0	0.1	-1.0	3.7	52.0
7	F	4.2	17.1	103.0	16.1	-0.1	0.1	3.1	48.5
8	M	3.3	15.2	99.3	15.4	-0.3	0.6	3.2	54.0
9	F	3.3	13.4	91.7	15.9	0.1	-1.1	3.5	50.0
Mean		4.4	16.7	102.5	15.8	0.1	-0.4	3.5	50.6
SD		0.8	2.2	6.4	0.5	0.4	0.7	0.3	2.2
Lower dose									
10	M	5.2	19.4	109.8	16.1	0.4	-0.5	3.1	45.6
11	M	4.9	18.3	106.3	16.2	0.4	-0.7	3.3	51.0
12	M	5.0	17.3	105.6	15.6	0.0	-1.0	3.2	49.0
13	M	5.1	16.7	106.5	14.7	-0.6	-1.1	3.4	51.0
14	M	3.6	18.0	104.8	16.4	0.6	1.4	3.2	50.0
15	M	3.9	15.9	97.5	16.7	0.5	-0.9	3.5	52.0
Mean		4.6	17.6	105.1	16.0	0.2	-0.5	3.3	49.8
SD		0.7	1.2	4.1	0.7	0.4	0.9	0.2	2.3

[1]Sex: M: Males. F: Females. [2]BMI: body mass index. [3]ZWH: Z-score Weight for Height. [4]ZHA: Z-score Height for Age.

the food ingested by the child at the day-care centres (breakfast, lunch and snack). All food was weighed before being eaten by the child, and any left overs were weighed again. A trained nutritionist using a Scaltec scale (Hamburg, Germany) with a precision of 0.1 g performed the weighing.

Recall data

On the same days that food was weighed at the day care-centres, a dietary recall was used for breakfast and the remaining periods (after 17:00 hours). The mother recorded in a notebook all the food ingested by the children at home. Next morning, the nutritionist in charge interviewed the mother to determine the type of food, portion sizes and preparation of the food. The same procedure was repeated during one day of the weekend (either Saturday or Sunday) and then on the next Monday morning. The data obtained from the food intake recall at home, plus the food ingested at the day care-centre and determined by food weighing was combined in a single week weighed average daily value. Finally, the weekly food energy intake was calculated from the dietary data using in house food intake software with typical Chilean foods values[29].

Respiratory quotient

The respiratory quotient of the diet (FQ) was calculated from the measured dietary intake data as described by Black AE *et al*.[30].

Statistical Methods

Descriptive statistics including minimum, maximum, and frequency distribution were derived for all variables. The Shapiro Wilk goodness-of-fit test and homogeneity of variance test were performed for continuous variables. Normal variables were expressed as mean ± standard deviation. Student's t test was used to test for significance of differences.

Subsequently, the results were compared using CO_2 production fractionation and TEE (kcal) and per kg of weigh, using individual FQ (TEEind) versus TEEfix using RQ value = 0.85, with repeated measures (parametric analysis). The significance level was set at $p < 0.05$. Data was analysed using STATA 12.0 (StataCorp. 2011. Stata Statistical Software: Release 12. College Station, TX: StataCorp LP.).

Results

Assumptions effects

Natural abundance variation in the correlation between ^2H and ^{18}O and its relation to the relative amounts of the isotope doses given, contribute to imprecision in the final CO_2 production estimate[31,32]. In this study, seven background samples collected on successive days prior to the DLW study were measured and errors arising from this source were calculated related to the goodness of fit of the data to the model[26]. The percentage contribution of each of these errors to CO_2 production rate and their combined values are shown in table II.

The component of error associated with background variation tended to be relatively larger (3.76%) for the low dose group compared to 2.99% in the high dose. But neither this, the goodness of fit error or the total error was significantly different between groups (Table II).

It is now usual in calculations to use values for the isotope dilution spaces that are normalised to population means. This is justified if analytical error contributes most to this component of overall variance[25] and of course the mean value used, typically 1,0297, needs to correspond to the true value for the study population. The effect of deviations in the values in the ratio of measured space from deuterium dilution to oxygen-18 dilution, compared to an externally derived, assumed population mean (1,0297) is shown in table III. The observed mean isotope dilution space was 1,030 ± 0.010, no different from the customarily assumed population, thus mean CO_2 production values

Table II
Background variability error covariance of both isotopic background values. Total error of the fit and overall error in 16 subjects

Subject	*CO_2 (moles)*	*Background variability error (%)*	*Goodness of fit error (%)*	*Total error (%)*
		Higher Dose		
1	10,76	1,11	2,97	3,18
2	13,34	1,45	1,55	2,13
3	10,74	2,64	3,69	4,54
4	10,01	1,35	3,76	4
5	9,79	1,53	2,48	2,92
6	13,02	4,43	6,62	7,97
7	10,97	2,98	4,83	5,68
8	11,09	5,05	3,43	6,1
9	9,89	3,71	4,07	5,51
Mean	11,1	2,7	3,7	4,7
SD	1,3	1,5	1,4	1,8
		Lower dose		
10	15,87	2,26	1,21	2,56
11	14,05	6,61	2,47	7,05
12	12,49	1,18	3,81	3,99
13	12,1	1,87	3,22	3,72
14	12,3	5,09	2,68	5,75
15	9,68	5,97	2,85	6,62
Mean	12,7	3,8	2,7	4,9
SD	2,1	2,3	0,9	1,8

calculated by each of these procedures produces no relative bias (Table III).

The effects of smaller (0,20) or greater (0,40) proportion of water assumed to be subject to fractionation as compared to the value of 0,30 applied in this and other studies on children[27] are shown in table IV. The effect of the magnitude of fractionation assumptions largely depends upon the relative difference between the slopes of the isotope disappearance curves (the smaller the difference the greater the effect). In practice, larger differences will be found when water turnover rates are high. Thus, the magnitude of effects recorded in one set of circumstances (country, climate etc.) cannot be applied in others. In this study, the effects were small. An overestimate of 1,8% in CO_2 was found when a factor of 0,20 is utilized, and a lower value of CO_2 (-1,9%), if the opposite situation is assumed (0,40).

Assessment of subject's diet produced a different value from the typical utilized in many studies[30]. For example, in Europe, a factor of 0,85 has been obtained from individual diet results[18,19]. This present study, mean FQ value was also 0,85. Thus when mean energy expenditures were calculated using the fixed value of 0,85 (TEE_{fix}) for FQ and the individually determined FQ (TEE_{ind}), the values were the same (Table V).

Comparison of TEE and energy intake

The total energy intake was calculated as a weighted average of weekday and weekend energy intake. The individual mean energy expenditure (1373+177 kcal) and energy intake (1409+161 kcal) are shown in table VI.

Discussion

DLW methodology has been used as a gold standard reference method to assess the validity of energy intake. In general, the measurement of energy intake underestimates the real needs, although the difference found in children under six years of age, is lower compared to older children. In fact, this difference increases over age and sex, affecting the outcome and it is worse in girls[33-35].

In the present study, energy intake in children was evaluated by weighing all food ingested by the child at the day-care centre and a dietary recall at home. The overall difference between intake and expenditure was 42,9 kcal/day (limits of agreement + 259,1 to -112,3 kcal/day).

At the individual level, absolute agreement between intake and expenditure would not be expected from

Table III
Effects of fractionated water proportion (0,2, 0,3 and 0,4)[a] on CO_2 production

Subject	*$CO_{2\ 0.2}$ (mols)*	*$CO_{2\ 0.3}$ (mols)*	*$CO_{2\ 0.4}$ (mols)*
1	10.01	9.82	9.63
2	12.55	12.31	12.12
3	10.68	10.44	10.22
4	9.31	9.13	8.96
5	8.99	8.76	8.53
6	12.27	12.01	11.83
7	10.11	9.87	9.63
8	10.16	9.89	9.62
9	9.14	8.94	8.74
10	14.91	14.70	14.49
11	13.18	13.00	12.81
12	11.67	11.48	11.29
13	11.35	11.11	10.92
14	11.45	11.16	10.92
15	8.96	8.77	8.57
Mean	10.98	10.76	10.55
SD	1.73	1.73	1.73

[a] $P < 0,05$ Repeated measures ANOVA.

Table IV
Observed dilution spaces ratio and values of CO_2 production for observed (o) ratio and normalized ratio (n)

Subject	*Observed Ratio*	*$CO_{2\,o}$ mole/d*	*$CO_{2\,n}$ mole/d*	*$CO_{2\,o}$/ $CO_{2\,n}$*
1	1.029	10.8	10.79	1.001
2	1.038	12.31	12.66	0.972
3	1.046	9.65	10.44	0.924
4	1.047	8.99	10.01	0.90
5	1.040	8.76	9.24	0.948
6	1.029	13.21	12.71	1.039
7	1.013	12.30	11.56	1.064
8	1.032	11.48	11.59	0.991
9	1.017	11.11	10.58	1.050
10	1.024	11.42	11.17	1.022
11	1.028	11.27	11.17	1.009
12	1.033	9.89	10.06	0.983
13	1.026	8.31	8.16	1.018
14	1.021	12.24	11.79	1.038
15	1.023	9.7	9.43	1.029
Mean	1.030	10.76	10.76	0.999
SD	0.010	1.48	1.26	0.047

Table V
Effects of fractionated water proportion (0,2, 0,3 and 0,4)[a] on normalized CO_2 production

Subject	*$CO_{2\ 0,2}$ (mole/d)*	*$CO_{2\ 0,3}$ (mole/d)*	*$CO_{2\ 0,4}$ (mole/d)*
1	11.14	10.79	10.76
2	12.84	12.66	12.47
3	10.68	10.44	10.20
4	10.79	10.62	10.45
5	9.47	9.24	9.01
6	12.99	12.78	12.56
7	11.75	11.56	11.36
8	11.78	11.59	11.39
9	10.76	10.58	10.39
10	11.36	11.17	10.98
11	11.41	11.17	10.94
12	10.33	10.06	9.79
13	8.36	8.16	7.96
14	12.02	11.79	11.56
15	9.62	9.43	9.24
Mean	11.02	10.80	10.60
SD	1.25	1.25	1.25

[a] $P < 0,05$ Repeated measures ANOVA.

a single measurement; but at a group level, the measurement of TEE should validate the measurement of habitual energy intake[18]. In a recent review, a similar comparisons in adult studies reported biases ranging from -59 to +24% but in most cases the intake measurement underestimated expenditure[36].

In infants and small children, data is less consistent. Lanigan *et al.*, observed a non-significant bias (+7,3%) for weighed intake compared to DLW of in infants of 6-24 months old[37,38]; Davies in 1,5-4,5 year old children found a non-significant bias of 4% in the comparison of energy intake and expenditure[39]. Livingstone found that utilising diet history in children of 3-5 years old, produced a 14% significant overestimation at 3 years of age and 11% at 5 years, compared to energy expenditure[27].

The validity of the average agreement between the measures in the present study is supported by our calculations on the relative importance of RQ and fractionation assumptions. By extension of the calculations used for table V it can be shown that to increase the mean TEE by 2,4% to match the mean intake would require the use of RQ=0,874 rather than 0,85. Similarly, the amount of water fractionated would need to decrease to 16% in order to explain a 2,4% of the difference. Reported data on children this age elsewhere does not substantiate this value[27].

The correlation of intake and expenditure is adequate (r=0,68) which agrees with other authors[37,39,40].

Table VI
TEE (kcal) and per kg of weight using individual FQ (TEE_{ind}) versus $TEE_{fix,}$ using fixed RQ value = 0,85

Subject	*Calc FQ*	*TEE_{ind}[a] (kcal/d)*	*TEE_{ind}[b] kcal/kg/d*	*TEE_{fix}[a] (kcal/d)*	*TEE_{fix}[b] kcal/kg/d*	*Energy intake[c] week*	*Energy intake[c] weekend*	*Energy intake weighed*
1	0.88	1337	66.3	1373	68.1	1235	1214	1272
2	0.87	1539	79.8	1566	81.2	1513	1678	1560
3	0.80	1195	61.2	1227	62.9	1604	1080	1454
4	0.87	1247	79.9	1269	78.3	1311	1359	1324
5	0.82	1146	70.3	1114	69.0	1395	1099	1310
6	0.87	1650	83.1	1680	84.6	1685	1420	1609
7	0.87	1536	84.8	1564	86.3	1485	1274	1424
8	0.87	1434	80.5	1460	81.9	1244	1903	1432
9	0.86	1401	82.4	1413	83.2	1255	1449	1310
10	0.77	1568	105.5	1452	97.7	1750	1441	1662
11	0.86	1420	82.9	1433	83.7	1678	1163	1531
12	0.86	1247	81.8	1258	82.6	1377	1129	1306
13	0.85	1057	79.2	1057	79.2	1142	1241	1170
14	0.85	1557	86.5	1557	86.5	1565	1739	1615
15	0.83	1256	78.9	1234	77.5	1337	1050	1255
Mean	0.85	1373	80.2	1377	80.2	1438	1349	1416
SD	0.03	177	9.9	180	8.5	189	258	152

[a,b,c] $P < 0,05$ Repeated measures ANOVA.

Although the difference range between individual differences can vary from -7% to +10% from the real value, the adequacy of these results may be attributed to energy intake method and the homogeneous diet that preschool children ate during the week-days at the day care-centres.

Irrespective of this potential bias, we also examined the relationship between the individual errors predicted in table II. TEE measurement error only explains a minor quantity of the 2,4% value between both measurements, thus TEE error would appear to have a small effect on this difference.

In summary, the main factor influencing the comparison between TEE and energy intake could be strongly related to energy intake measurement factors. In this study the errors in intake measurement could only come from the home recordings, where food intake is more variable, particularly at weekends[33]. In the Framingham Study the association between energy intake in the 3-4 year old age group, predicted the intake in the subsequent period (5-6 year olds) where 57,1% to 85,7% of the children remained in the energy intake top two quintiles. Sixty percent of the 93,3% children with the highest intake at baseline (3-4 year olds) were still in the top two quintiles at 7-8 years of age[35]. Thus, the validity of energy intake assessment methods is a very important tool for the study and prevention of childhood weight excess. In this paper, the comparison to DLW to a mixed energy intake methodology prove to be successful and may be applicable to similar places where subjects eat near 75% of their daily energy intake. As a general conclusion, energy intake determination is crucial and requires to be valid especially at preschool age, due to its significant association to the later presence of obesity and future interventions[41,42].

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Conflict of Interest

None the authors has a conflict of interest.

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