Biomed Environ Sci, 2026; 39(4): 482-485

Letter

Total Energy Expenditure in Healthy Middle-aged Chinese Adults with Light-intensity Physical Activity: A Doubly Labeled Water Study

Minghang Guo[1,&], Jie Feng[1,&], Guangbo Tong[2], Weidong Li[1], Xinghua Tong[3],
Zhibin Ren[1], Deqian Mao[1,#], and Lichen Yang[1,#]

Overweight and obesity are well-recognized risk factors for a wide variety of chronic diseases and have become a significant public health issue worldwide[1,2]. The report on Nutrition and Chronic Disease Status of Chinese Residents (2020) demonstrated that over half of Chinese adults aged 18 years and older were overweight or obese, with prevalence rates of 34.3% for overweight and 16.4% for obesity. Recent studies have reported that both men and women have experienced an increase in the prevalence of overweight and obesity after middle age. This notable age-related disparity in obesity prevalence suggests that middle age represents a critical window for obesity prevention and intervention in the Chinese population. Accurate determination of total energy expenditure (TEE) is essential for establishing precise estimated energy requirements (EER) for specific populations. The doubly labeled water (DLW) method represents the gold standard for measuring free-living TEE[3,4]. However, DLW-measured TEE data for middle-aged Chinese adults are currently lacking. Consequently, in the 2023 Chinese Dietary Reference Intakes (DRIs), the EER for Chinese adults aged 50–60 years was derived by applying an empirical 5% downward adjustment to values for young adults (18–45 years old)[5]. The direct measurement of TEE using DLW in this demographic is essential for filling this gap. In this study, we aimed to provide the first DLW-measured TEE data for healthy, normal-weight Chinese adults aged 50–60 years with light-intensity physical activity and to preliminarily evaluate age-related changes in TEE. Eighteen healthy participants (eight males and ten females) aged 50–60 years were recruited from communities in Jizhou District, Tianjin, North China, between June 2022 and January 2023. Inclusion criteria included normal body mass index (BMI 18.5–24 kg/m^2), stable body weight (BW) (fluctuations ≤ ± 2 kg over the last 3 months), and a self-reported light habitual physical activity level, verified by self-report and interview preceding enrollment. The exclusion criteria comprised thyroid dysfunction, hepatic/renal impairment, metabolic disorders, actively attempting weight loss or following a weight-loss diet within the last 3 months prior to enrollment, and menstruating females. All participants exhibited normal clinical biomarker levels within the reference ranges (Supplementary Table S1). Body composition was assessed by bioelectrical impedance analysis (BIA; MC-980MA, TANITA). Physical activity was objectively monitored using waist-worn ActiGraph accelerometers for ≥ 4 consecutive days (≥ 10 hours valid wear-time/day) and analyzed with ActiLife 6.0 software. The data showed that our recruited subjects maintained a uniform light physical activity level (LPA).

The DLW experimental protocol for the measurement of TEE was in accordance with the International Atomic Energy Agency (IAEA) guidelines[6]. A 7-day testing period (following a 3-day adaptation period for dietary stabilization) was used to balance measurement precision and participant compliance in this community-based study. On the morning of day 4, after collecting a baseline urine sample, the participants received an accurately calibrated oral dose of DLW (1 g $^{2}H_2{}^{18}O$ per kilogram BW; ^{2}H: 10.3 atom%, ^{18}O: 8.3 atom%) with careful supervision to ensure complete ingestion. The dose was optimized for our 7-day protocol and instrument sensitivity. Post-dose urine samples were collected at 1, 2, 3, 4, and 5 h on the day of administration and on the mornings of the

---

doi: 10.3967/bes2026.028

1. National Institute for Nutrition and Health, Chinese Center for Disease Control and Prevention, Key Laboratory of Public Nutrition and Health, National Health Commission of the People's Republic of China, Beijing 100050, China; 2. The Yinliu Town Health Center of Jizhou District, Tianjin 301900, China; 3. Lohas Lemon Farm, Jizhou District, Tianjin 301903, China

next 6 days. Tap water used for cooking was also collected to establish the local isotope baseline. All samples were immediately separated and frozen at −20 °C until analysis. Isotopic analysis was performed using a Nu Perspective isotope ratio mass spectrometer (IRMS). TEE measured by DLW (TEE-DLW) was calculated as follows: TEE-DLW (kcal/day) = rCO_2(L/day) × (1.106 + 3.94/FQ)[7]. In the above formula, the rate of CO_2 production (rCO_2) was calculated from the deuterium and oxygen-18 turnover rates obtained via the DLW technique for each participant. The duplicate diet method directly measured the proportions of three macronutrients to obtain an objective, individual-specific value for the Food Quotient (FQ). The mean FQ was determined to be 0.88 ± 0.01 (mean ± SD). The quality control criteria and calculation procedure for TEE-DLW were detailed in the supplemental materials, with final day isotope enrichment > 50 ppm above baseline.

The average age of the participants was (55.6 ± 2.7) years. The mean BMI was 23.3 kg/m^2. The mean TEE-DLW was (2,266.8 ± 412.9) kcal/d for males and (1,823.0 ± 180.5) kcal/d for females (*P* = 0.007) (Table 1). Multiple linear regression analysis was used to identify primary determinants of TEE (Table 2). The explanatory variables of age and sex explained only 36.4% of the variance in TEE (Model 1). The inclusion of BW or fat-free mass (FFM) improved the adjusted R^2 to 65.6% (Model 2) and 55.3% (Model 3). This finding is consistent with the established understanding that BW, which comprises both fat mass and FFM, is a key determinant of energy metabolism. The FFM represents the metabolically active tissue compartment encompassing the skeletal muscle, organs, and other non-fat tissues that collectively drive basal metabolic processes. Notably, after adjusting for either BW or FFM in our regression models, the initially significant effect of sex on TEE-DLW was no longer significant, indicating that sex differences in TEE were largely mediated by variations in BW or FFM, rather than representing intrinsic metabolic differences between sexes. Interestingly, age failed to emerge as a significant predictor in Model 3, suggesting that within

Table 1. Characteristics and TEE-DLW of the study participants

Characteristics	**Total**	**Male**	**Female**	***P*-value**[a]
Sample size	18	8	10	–
Age (year)	55.6 ± 2.7	55.3 ± 2.5	55.8 ± 3.0	0.686#
Height (cm)	161.4 ± 7.3	168.3 ± 5.1	155.9 ± 2.4	0.000#
Weight (kg)	57.8 (56.1, 61.1)	61.3 (58.6, 69.8)	56.4 (54.1, 57.4)	0.000*
BMI (kg/m^2)	23.3 (21.7, 23.7)	22.1 (21.1, 23.6)	23.5 (22.5, 23.7)	0.408*
FFM (kg)	38.2 (36.3, 47.9)	48.3 ± 4.0	36.4 ± 1.3	0.000#
TEE-DLW (kcal/d)	2,025.9 (1,687.1, 2,193.1)	2,266.8 ± 412.9	1,823.0 ± 180.5	0.007#

Note. Values were expressed as mean ± SD or median (P_{25}, P_{75}) according to data distribution. [a]Comparison between male and female: *Mann-Whitney *U* test; #*t*-test. BMI, body mass index; FFM, fat-free mass.

Table 2. Multiple linear regression models for predicting TEE-DLW (*N* = 18)

Variable	**Model 1**		**Model 2**		**Model 3**	
	β (95% *CI*)	*P*-value	*β* (95% *CI*)	*P*-value	*β* (95% *CI*)	*P*-value
(Intercept)	3809.48 (667.17, 6951.78)	0.021	1721.45 (−897.32, 4340.21)	0.180	629.35 (−3,028.88, 4,287.58)	0.718
Age (year)	−35.60 (−91.80, 20.60)	0.197	−43.55 (−85.36, −1.73)	0.042	−19.03 (−68.24, 30.19)	0.421
Sex (male = 1)	424.20 (122.08, 726.32)	0.009	64.73 (−240.40, 369.87)	0.656	−303.97 (−934.80, 326.87)	0.319
BW (kg)	–	–	45.63 (19.25, 72.00)	0.002	–	–
FFM (kg)	–	–	–	–	61.94 (12.86, 111.03)	0.017

Note. *N*, sample size; BW, body weight; FFM, fat-free mass. Model 1 was adjusted for age, and sex; adjusted R^2 = 0.364, *P* = 0.013. Model 2 was adjusted for age, sex, and BW; adjusted R^2 = 0.656, *P* < 0.001. Model 3 was adjusted for age, sex, and FFM; adjusted R^2 = 0.553, *P* = 0.002. Sex: 1 = Male, 0 = Female.

the 50–60 year age range examined, aging per se may not substantially influence TEE when differences in FFM were properly accounted for.

To further explore potential age-related changes in energy metabolism, the current TEE-DLW data from middle-aged adults were compared with the original TEE-DLW data from previously published studies conducted in our laboratory on younger healthy Chinese adults aged 20–26 years, who were also classified as having a LPA according to the 2023 Chinese DRIs criteria[8,9]. In the unadjusted direct comparisons, no significant reduction in TEE-DLW with age was observed in either the male or female participants (Supplementary Table S2). Moreover, in pooled regression analysis (N = 50), the age group (young vs. middle-aged) was not a significant predictor of TEE after adjusting for BW (β = 5.65, P = 0.927) or FFM (β = 127.80, P = 0.132) (Table 3). This statistical finding suggested that the differences in TEE may primarily reflect underlying variations in body composition rather than intrinsic metabolic changes associated with the aging process itself. In addition to our study, a large-scale study by Pontzer et al. (2021), published in Science, also reported no significant decline in TEE after FFM adjustment in adults aged 20–60 years[10]. Therefore, our findings highlight the importance of considering BW or FFM as primary determinants when evaluating energy metabolism across the lifespan.

The 2023 Chinese DRIs proposed EERs of 1,950 kcal/d for males and 1,600 kcal/d for females (aged 50–60 years, LPA). When normalized to the standard BWs of the DRIs, our DLW-measured TEE values were substantially higher: 19% for males (2,330 *vs.* 1,950 kcal/d) and 11% for females (1,778 *vs.* 1,600 kcal/d). This discrepancy likely stems from methodological differences. The EERs were derived from a factorial approach (BMR × PAL) that relied on population-averaged estimates. However, our findings, based on a relatively small sample size, should be interpreted with caution and warrant further verification in larger studies.

In conclusion, this study provides the first DLW-based TEE data for healthy Chinese adults aged 50–60 years who engage in light-intensity physical activity. After adjusting for BW or FFM, no significant sex-related differences in TEE were observed, highlighting BW and FFM as primary determinants of TEE in this population. The small and homogeneous sample and cross-sectional design limit the generalizability of our findings. Future studies with larger and more diverse samples (e.g., overweight/obese individuals and broader age ranges) are warranted to validate these results and better characterize energy expenditure across key demographic groups.

Funding This work was supported by the National Key R&D Program of China (grant numbers 2020YFC2006300 and 2020YFC2006302).

Competing Interests The authors declare no competing interests.

Ethics This study was approved by the Ethical Committee of the National Institute for Nutrition and Health, Chinese Center for Disease Control and Prevention (Approval No. 2022-016). All participants provided written informed consent after receiving a comprehensive explanation of the study procedures.

Author's Contributions Writing the original draft, Methodology, Data analysis: Minghang Guo; Conducting the experiments, Writing the original draft, Methodology, Validation: Jie Feng; Recruiting

Table 3. Comparison of regression models in young and middle-aged adults for predicting TEE-DLW (N = 50)

Characteristic	Model 1		Model 2	
	β (95% *CI*)	*P*-value	β (95% *CI*)	*P*-value
(Intercept)	1084.50 (341.48, 1827.52)	0.005	855.32 (−26.98, 1737.62)	0.057
Age group(middle-aged adults = 1)	5.65 (−118.36, 129.67)	0.927	127.80 (−39.75, 295.36)	0.132
Sex (male = 1)	307.61 (133.44, 481.78)	0.001	146.89 (−139.54, 433.32)	0.307
BW (kg)	13.46 (0.06, 26.86)	0.049	—	—
FFM (kg)	—	—	23.31 (2.22, 44.39)	0.031

Note. For comparison, young healthy adult TEE data were obtained from two of our laboratory's previous publications[8,9]. N, sample size; BW, body weight; FFM, fat-free mass. Model 1 was adjusted for BW, age group, and sex; adjusted R^2 = 0.530, P < 0.001. Model 2 was adjusted for FFM, age group, and sex, adjusted R^2 = 0.538, P < 0.001. Sex: 1 = male, 0 = Female; age group: 0 = young adults, 1 = middle-aged adults.

volunteers: Guangbo Tong and Xinghua Tong; Conducting the experiments: Zhibin Ren and Weidong Li. Methodology, Supervision: Deqian Mao; Writing, Review, Supervision: Lichen Yang. All authors reviewed and approved the final version of the manuscript and agree to be accountable for all aspects of the work.

Acknowledgements The authors gratefully acknowledge the editor and reviewers for their critical and constructive comments, whose valuable input has been essential to the revision and overall improvement of this manuscript.

Data Sharing The supplementary materials will be available in www.besjournal.com.

[&]These authors contributed equally to this work.

[#]Correspondence should be addressed to Deqian Mao, Professor, E-mail: maodq@ninh.chinacdc.cn; Lichen Yang, PhD, Professor, E-mail: yanglc@ninh.chinacdc.cn

Biographical notes of the first authors: Minghang Guo, Ph.D., majoring in nutrition and food hygiene, E-mail: gmh699@whu.edu.cn; Jie Feng, Ph.D., Associate Professor, majoring in nutrition and analytical chemistry, E-mail: fengjie@ninh.chinacdc.cn

Received: November 12, 2025;

Accepted: March 2, 2026

REFERENCES

1. Bray GA, Kim KK, Wilding JPH. Obesity: a chronic relapsing progressive disease process. A position statement of the World Obesity Federation. Obes Rev, 2017; 18, 715−23.
2. Chen K, Shen ZW, Gu WJ, et al. Prevalence of obesity and associated complications in China: a cross-sectional, real-world study in 15.8 million adults. Diabetes Obes Metab, 2023; 25, 3390−9.
3. John RS. The history and theory of the doubly labeled water technique. Am J Clin Nutr, 1998; 68, 932S−8S.
4. Westerterp KR. Doubly labelled water assessment of energy expenditure: principle, practice, and promise. Eur J Appl Physiol, 2017; 117, 1277−85.
5. Chinese Nutrition Society. Dietary reference intakes for China. People’s Medical Publishing House. 2023. (In Chinese)
6. IAEA. Assessment of body composition and total energy expenditure in humans using stable isotope techniques. Vienna: International Atomic Energy Agency, 2009.
7. Speakman JR, Yamada Y, Sagayama H, et al. A standard calculation methodology for human doubly labeled water studies. Cell Rep Med, 2021; 2, 100203.
8. Zhuo Q, Sun R, Gou LY, et al. Total energy expenditure of 16 Chinese young men measured by the doubly labeled water method. Biomed Environ Sci, 2013; 26, 413−20.
9. Liu JM, Yang XG, Piao JH, et al. Dietary energy requirements of young adult women in China by the doubly labeled water method. Asia Pac J Clin Nutr, 2010; 19, 520−5.
10. Pontzer H, Yamada Y, Sagayama H, et al. Daily energy expenditure through the human life course. Science, 2021; 373, 808−12.