

## Short Communication

# Hair mineral analysis by X-ray fluorescence spectrometry: associations with body fat

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Hair mineral analysis using an X-ray fluorescence spectrometer as a method of estimating body fat percentage (BF%) was investigated. Body fat percentage of 24 healthy Japanese, aged 20-27 years, was measured using a hand-held impedance analyzer (BF%<sub>IMP</sub>). X-ray (K $\alpha$ -ray) intensities of sulfur, chlorine, potassium, calcium, titanium, and iron (Fe) in hair were measured using an X-ray fluorescence spectrometer. Body fat percentage was also measured using a Hologic whole body dual energy X-ray absorptiometer (BF%<sub>DXA</sub>) in nine subjects selected from the above 24 subjects based on their BF%<sub>IMP</sub>. Correlations of the two BF%s with Fe-K $\alpha$  were significant (BF%<sub>IMP</sub>:  $r = 0.60$   $n = 24$ ,  $p < 0.01$ ; BF%<sub>DXA</sub>:  $r = 0.67$   $n = 9$ ,  $p < 0.05$ ). The mean ( $\pm$ SD) biases (measured minus estimated using multiple regression equations by Fe-K $\alpha$ ) for BF%<sub>IMP</sub> and BF%<sub>DXA</sub> were  $2.97 \pm 2.25\%$  and  $1.77 \pm 1.33\%$ , respectively. The SEEs for the two equations for BF%<sub>IMP</sub> and for BF%<sub>DXA</sub> were less than 4%. These results suggest that Fe-K $\alpha$  may be a predictor of body fat percentage. However, the subjects were few and only Japanese in their twenties, so that further investigation is needed for methodological generalization.

**Key Words:** body fat percentage, hair mineral content, Japanese, prediction equations, X-ray fluorescence spectrometer

## INTRODUCTION

Obesity is a serious condition associated with health risks such as cardiovascular disease.<sup>1</sup> For this reason, easy-to-use body fat measures have been developed as health indices and appliances. At present, BMI, skinfold measurements, and bioelectrical impedance are considered suitable for epidemiological estimation of body fat percentage.<sup>2</sup> In particular various impedance analyzers are already on the market worldwide.

Although underwater weighing (UWW) is traditionally considered the standard method of estimating body fat percentage (BF%), measuring using a Hologic whole body dual energy X-ray absorptiometer (DXA) is becoming a new standard method.<sup>3</sup> Hologic whole body dual energy X-ray absorptiometer measures BF% with greater precision than UWW.<sup>4</sup>

Recently, hair analysis has received a large amount of academic and commercial interest for wide-ranging applications, but the degree of success of analytical interpretation has been minimal.<sup>5</sup> However, the possibility of hair analysis for epidemiology and etiology studies was reviewed.<sup>5</sup> In addition, though it is limited to several minerals, an X-ray fluorescence spectrometry is available for simple measurement of hair mineral concentrations without complicated preparation including acid dissolution.<sup>6</sup> Using the relationship between BF% and mineral concentration in the body, BF% is determined by measuring the whole body concentration of potassium (K) using a human counter.<sup>3</sup>

The aim of the present study was to examine the possibility of hair mineral analysis using an X-ray fluorescence spectrometer as a method of estimation of BF%. If avail-

able for body fat estimation, it is usable in disabled and even the deceased as well as the general population.

## MATERIALS AND METHODS

The participants were 24 (14 women and 10 men) healthy Japanese university students aged 20-27 years (Table 1). After obtaining oral informed consent, hair was sampled and body fat was measured. One hair from the top of the head was sampled for mineral analysis using an X-ray fluorescence spectrometer (OURSTEX200tX, Ourstex Corp, Osaka, Japan). X-ray (K $\alpha$ -ray) intensities (cps/mA) of sulfur (S), chlorine (Cl), potassium (K), calcium (Ca), titanium (Ti), and iron (Fe) in hair were able to be measured. Then, the body fat percentage (BF%<sub>IMP</sub>) was estimated using a hand-held impedance analyzer, following the instructions in the manufacturer's manual (Omron HBF-306-W, Omron Healthcare, Kyoto, Japan). Measurement was done at least 2 hours after breakfast (including beverage).

On another day, body fat percentage (BF%<sub>DXA</sub>) of nine of 24 participants chosen for their BF%<sub>IMP</sub> was measured using a Hologic whole body dual energy X-ray absorptiometer (QDR4500W, software version 11.2.5, Hologic, Waltham, MA, USA) at a medical center. Measurement was carried out at least 2 hours after lunch (in-

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Manuscript received 6 November 2012. Initial review completed 17 December 2013. Revision accepted 2 February 2013.

doi:10.6133/apjcn.2013.22.2.14

**Table 1.** Physical characteristics of subjects

	Women (n = 14)	Men (n = 10)	All subjects (n = 24)
Age (years)	21.4 ± 1.8	22.8 ± 2.0	22.0 ± 2.0
Weight (kg)	50.0 ± 5.4	65.0 ± 12.4	56.3 ± 11.6
Height (m)	1.60 ± 0.05	1.72 ± 0.08	1.65 ± 0.09
BMI (kg/m <sup>2</sup> )	19.6 ± 1.8	21.9 ± 3.2	20.6 ± 2.7

Each value is the mean ± s.d.

cluding beverage). Mean values (±SD) for the physical characteristics of the nine subjects were 22.9 (2.9) years for age, 58.8 (13.6) kg for weight, 1.65 (0.09) m for height, 21.4 (3.5) kg/m<sup>2</sup> for BMI, and 23.4 (6.2) % for BF%<sub>IMP</sub>.

Statistical analyses were performed with pooled data for both sexes because the number of subjects was small. The relationships between body fat percentages determined by BMI (BF%<sub>IMP</sub>) and X-ray intensity (BF%<sub>DXA</sub>) of each mineral were investigated using Pearson's correlation coefficients. Pearson's correlation coefficient between BF%<sub>IMP</sub> and BF%<sub>DXA</sub> was also determined. Models for predicting body fat percentages (BF%<sub>IMP</sub> and BF%<sub>DXA</sub>) were developed by using multiple regression analysis with X-ray intensity of minerals as potential independent variables. Potential interaction terms were explored in model development and a forward-backward stepwise selection procedure was applied for the derivation of prediction equation models. All analyses were carried out with the statistical software program Statcel3 (version 3, 2011; OMS Publishing Inc, Tokyo).

This research was reviewed and approved by the Azabu University Research Service Division.

## RESULTS AND DISCUSSION

X-ray intensity (cps/mA) of each mineral was as follows: S 1801 ± 498 (range 900 to 2757), Cl 71.1 ± 44.1 (15.9 to 152), K 8.7 ± 6.4 (1.0 to 27.2), Ca 193 ± 200 (66.8 to 982), Ti 172 ± 86.9 (86.6 to 462), and Fe 67.9 ± 15.3 (44.6 to 103).

There were significant correlations between BF%<sub>IMP</sub> with BMI ( $r = 0.53$ ,  $p < 0.01$ ) and Fe-K $\alpha$  ( $r = 0.60$ ,  $p < 0.01$ ) (Table 2). Multiple regression analysis produced the following equation for BF%<sub>IMP</sub>:

$$\text{BF\%}_{\text{IMP}} = 9.93 + 0.18 \times \text{Fe-K}\alpha \quad (1)$$

where multiple correlation coefficient ( $R$ ) = 0.60 ( $p < 0.01$ ) and SEE = 3.86% body fat, Fe-K $\alpha$  is expressed in cps/mA. The mean bias of estimated BF%<sub>IMP</sub> using this model from actual measured values was 2.97 ± 2.25% (range -8.4% to 6.2%).

The correlation of BF%<sub>IMP</sub> and BF%<sub>DXA</sub> was high ( $r = 0.95$ ,  $p < 0.001$ ) (Table 3). The BF%<sub>IMP</sub> showed a between-method difference of 2.77 ± 1.44% (range -3.6% to 5.0%) compared with the BF%<sub>DXA</sub>. The correlation of BF%<sub>DXA</sub> was significant with only Fe-K $\alpha$  among minerals studied ( $r = 0.67$ ,  $p < 0.05$ ) (Table 3). Multiple regression analysis produced the following equation for BF%<sub>DXA</sub>:

$$\text{BF\%}_{\text{DXA}} = -21.24 + 0.46 \times \text{Fe-K}\alpha + 0.01 \times \text{S-K}\alpha - 0.02 \times \text{Ca-K}\alpha + 0.03 \times \text{Ti-K}\alpha \quad (2)$$

where  $R = 0.95$  ( $p < 0.05$ ) and SEE = 3.25% body fat, mineral-K $\alpha$  is expressed in cps/mA. The mean bias of estimated BF%<sub>DXA</sub> using this model from actual measured values was 1.77 ± 1.33% (range -4.2% to 2.1%) (Figure 1).

BF% as estimated by a hand-held impedance analyser (BF%<sub>IMP</sub>) was very close to the measured value given by a Hologic whole body dual energy X-ray absorptiometer (BF%<sub>DXA</sub>). Correlations of the two BF%s (BF%<sub>IMP</sub> and BF%<sub>DXA</sub>) are significant with only Fe-K $\alpha$  among the minerals measured in this study. This indicates that Fe-K $\alpha$  may be a estimator of body fat percentage. The SEEs for equations (1) for BF%<sub>IMP</sub> and (2) for BF%<sub>DXA</sub> produced in this study are less than 4%. This value is comparable to prediction errors for various estimation methods found in other studies.<sup>2</sup>

As for the relationship of body Fe to fat accumulation, some iron-binding proteins such as transferrin and ferritin are known to contribute to antioxidant defense by chelating transition iron and preventing it from catalyzing the production of free radicals in the cell.<sup>7</sup> It is reported that increased oxidative stress in accumulated fat is an important pathogenic mechanism of obesity-associated metabolic syndrome.<sup>8</sup> Hair Fe concentrations might reflect an inner state of a dynamic phenomenon changing on the body fat.

For budgetary limitation, the number of subjects that we could measure body fat % by DXA was limited in particular. The subjects in this study were few and only Japanese in their twenties. And all within a normal weight range. It would be interesting to see how this works on people with higher BMIs. Previous investigators have documented that the relationship between anthropometric

**Table 2.** Pearson's correlation coefficients ( $r$ ) of estimated body fat percentage (BF%<sub>IMP</sub>) with BMI and X-ray intensity of each mineral

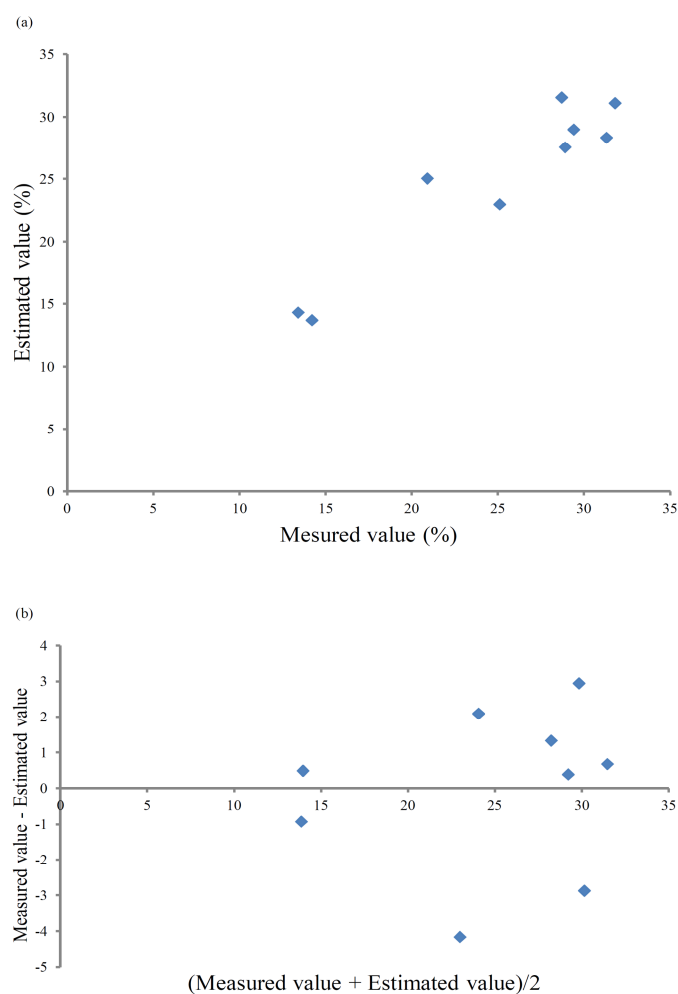
Measurement	$r$	Significance
BMI	0.53	0.008
S-K $\alpha$	0.16	0.454
Cl-K $\alpha$	-0.12	0.572
K-K $\alpha$	0.26	0.221
Ca-K $\alpha$	0.39	0.060
Ti-K $\alpha$	0.37	0.072
Fe-K $\alpha$	0.60	0.002

$n = 24$ . BF%<sub>IMP</sub>, body fat percentage estimated from impedance.

**Table 3.** Pearson's correlation coefficients ( $r$ ) of body fat percentage (BF%<sub>DXA</sub>) with BMI and X-ray intensity of each mineral

Measurement	$r$	Significance
BF% <sub>IMP</sub>	0.95	0.000
BMI	0.58	0.106
S-K $\alpha$	0.25	0.515
Cl-K $\alpha$	0.23	0.548
K-K $\alpha$	0.34	0.365
Ca-K $\alpha$	0.39	0.305
Ti-K $\alpha$	0.63	0.070
Fe-K $\alpha$	0.67	0.049

$n = 9$ . BF%<sub>DXA</sub>, body fat percentage measured using a Hologic whole body dual energy X-ray absorptiometer. BF%<sub>IMP</sub>, body fat percentage estimated from impedance.



**Figure 1.** Scatter plots (a) and Bland-Altman different plot (b) between estimated body fat % by the equation (2) and body fat % measured by DXA.

measures (BMI and skinfold fat) and measured fat percentage were not independent of age and gender or ethnicity.<sup>9</sup> In addition, variables such as age and gender are reported to contribute to hair mineral concentrations.<sup>5</sup> Thus, further epidemiological investigation is needed for methodological generalization. However, even considering these points, hair mineral analysis by X-ray fluorescence spectrometry as a method of estimation of BF% may have some degree of applicability.

#### ACKNOWLEDGMENTS

This research was supported by a project grant (Intellectual Property Utilization Research) awarded by the Azabu University Research Service Division.

#### AUTHOR DISCLOSURES

The authors declare that they have no conflict of interest.

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## Short Communication

**Hair mineral analysis by X-ray fluorescence spectrometry: associations with body fat**

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*School of Veterinary Medicine, Azabu University, Sagamihara, Japan***X-光螢光分析儀分析之頭髮礦物質及其與體脂肪之相關性**

探討利用 X 光螢光分析儀分析之頭髮礦物質是否可用來估測體脂肪百分比。以手握式阻抗分析儀測量 24 位 20-27 歲健康日本人之體脂肪百分比(BF%<sub>IMP</sub>)，並利用 X 光螢光分析儀檢測頭髮中硫、氯、鉀、鈣、鈦和鐵之 X 光(K $\alpha$  射線)強度。此外，根據 BF%<sub>IMP</sub>之結果，從 24 位中選出 9 位受試者再利用全身性雙能 X 光吸光儀來檢測其體脂肪百分比(BF%<sub>DXA</sub>)。兩種體脂肪百分比與鐵之 K $\alpha$  射線強度(Fe-K $\alpha$ )有顯著相關(BF<sub>IMP</sub>:  $r=0.60$ ,  $n=24$ ,  $p<0.01$ ; BF<sub>DXA</sub>:  $r=0.67$ ,  $n=9$ ,  $p<0.05$ )。BF%<sub>IMP</sub> 和 BF%<sub>DXA</sub> 的平均( $\pm$ 標準差)偏差(測量值減去利用 Fe-K $\alpha$  多元迴歸方程式求出之估計值)分別為  $2.97\pm 2.25\%$  和  $1.77\pm 1.33\%$ ；且 BF%<sub>IMP</sub> 和 BF%<sub>DXA</sub> 方程式之估計值的標準誤差(SEE)皆小於 4%。以上結果顯示 Fe-K $\alpha$  可能當做體脂肪百分比之預測因子。然而，本研究受試者人數過少，且主要為 20 餘歲日本人，因此為了使此估測方式更為普及，有賴更多研究證實。

**關鍵字：**體脂肪百分比、頭髮礦物質含量、日本人、預測方程式、X-光螢光分析儀