Total Body Phosphorus in Healthy Women and Ethnic Variations

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Total body phosphorus (TBP) levels were measured in 90 black and 143 white healthy women to determine ethnic differences. The measurements were performed by in vivo delayed gamma neutron activation (DGNA) analysis at Brookhaven National Laboratory (BNL). Mean value of TBP in whites was 10.4% lower as compared with the black women (mean TBP in white women 401.4 ± 57.5 g *v* 447.7 ± 57.7 g in black women). Both subgroups have a decrease in TBP with age with a rapid phase after the onset of menopause, which corresponds to bone loss. The decrease in TBP is similar in both ethnic groups with black women losing –1.59 g/yr (-0.33%/yr) and white women losing –2.08 g/yr (-0.45%/yr).

THE QUANTITATIVE assessment of the chemical composition of the human body as a function of age is of basic interest, as well as of clinical value in diagnosing metabolic disorders. Our knowledge of the elemental composition of the human body has increased significantly since the development of in vivo neutron activation analysis (IVNAA). This technique has enabled a direct quantitative chemical profile of the human being. Evaluation of body composition in the general population by this technique has provided valuable information about the various genetic factors (eg, gender, age, ethnicity, etc) or diseases (malnutrition, critical illness, malignancies, renal failure, etc) that can influence assessment of elemental content.[1]

The details of IVNAA, which is the state of the art technique for body composition, have been described extensively by other investigators.[1-5] However, the fundamental principles include that the body is exposed to neutrons from an external source. These neutrons interact with the constituent elements of the body by collision (may or may not emit radiation), neutron capture (may or may not emit radiation), and, most important, radiative capture in which a neutron is captured by a target nucleus and a gamma ray is then emitted by the new isotope. All neutron activation techniques depend ultimately on the emission of a gamma ray characteristic of the product nucleus. This emission can occur almost immediately (within 10 to 15 seconds), in which case the technique used to measure this emission is usually referred to as "prompt γ analysis" or over a long period (on the order of minutes), in which the technique is called "delayed γ analysis" or delayed gamma neutron activation (DGNA) analysis. Total body phosphorus (TBP) is measured by DGNA.

Phosphorus makes up about 0.65% to 1.1% of the adult body.[6] Eighty-five percent of adult body phosphorus is in bone. The remaining 15% is distributed through the soft tissues, such as muscle, liver, adipose tissue, and blood.[7] Total phosphorus in whole blood (approximately 40 mg/dL) is distributed in phospholipids of red blood cells and plasma lipoproteins. A tiny fraction of TBP is present as inorganic phosphate (approximately 3.1 mg/dL) in the blood and extracellular fluid and has a critical role in metabolism of carbohydrates, lipids, and proteins.

A few reports on body composition in a healthy population have provided TBP reference ranges in a small number of subjects with no differences between ethnic groups.[8,9] We studied a large number of pre- and postmenopausal healthy women who had body composition measurements with in vivo DGNA performed at Brookhaven National Laboratory (BNL), NY, which was updated in 1987. This report analyzes the distribution of TBP in a large number of healthy women to study (1) the range of TBP in normal healthy women, (2) ethnic differences, (3) effect of aging, and (4) effect of menopause on TBP levels.

MATERIALS AND METHODS

There were 233 participants (90 black and 143 white healthy women) who volunteered for studies at BNL and were part of a larger study on body composition in black and white women. The age ranged between 20 and 70 years. All subjects were active and in good health, with body mass index (BMI) ranging between 18 and 32 kg/m^2. A cut off of 32 kg/m^2 was chosen because of the influence of body thickness on elemental analyses and because BMI misclassifies 12% of black women as obese.[10] None of the subjects had a previous history or current symptoms of metabolic, renal, cardiovascular disease, or chronic illnesses. None of the postmenopausal women were receiving estrogen, bisphosphonates, calcitonin, or any medication known to alter calcium and phosphorus metabolism. Participants were screened on site by physical exam and blood chemistries and were excluded for any abnormalities found during evaluation. A recall activity questionnaire was completed by patients and the Compendium of Physical Activity was used to evaluate habitual physical activity.[11] The study was approved by the Institutional Review Board of Winthrop-University Hospital, and written informed consent was obtained from each participant.

DGNA analysis was performed on all of the subjects at the BNL facility, which was upgraded in 1987.[12] The neutron exposure was provided by an array of 14 ^{238}Pu, Be neutron sources of 1.55 TBq (42 Ci) each. The neutron sources are arranged in 2 rows above and below the midline of the subject. Fast neutrons are moderated by surrounding the subject with a 2-cm layer of polyethylene. The exposure time is fixed at 5 minutes and delivers a total neutron (QF = 10) and gamma dose measured at less than 2.8 mSv (278 mrem). The induced activity is measured in the whole body counter (WBC) for 5 minutes, starting 3 minutes postirradiation. The detectors used in the WBC are 32 rectangular NaI detectors, each with a volume of 10.2 cm × 10.2 cm ×

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Table 1. Mean Values for Clinical Characteristics, TBP, TBCa, and TBK

	Blacks (n = 90)	Whites (n = 143)	*P* Value
Age (yr)	44.1 ± 11.9	48.6 ± 12.5	<.011
Age at menopause (yr)	49.8 ± 2.2	51.3 ± 2.7	<.02
Height (cm)	163.4 ± 6.1	164.3 ± 6.0	NS (*P* = .18)
Weight (kg)	69.0 ± 10.2	64.3 ± 9.2	<.0004
BMI (kg/m^2)	25.9 ± 3.6	23.8 ± 3.3	<.0001
TBP (kg)	0.448 ± 0.058	0.401 ± 0.057	<.0001
TBCa (kg)	0.776 ± 0.083	0.720 ± 0.095	<.0001
TBK (kg)	0.103 ± 0.014	0.096 ± 0.013	<.0001
Physical activity (kcal)	6,709.0 ± 6,279.7	7,807.9 ± 8,052.4	NS (*P* = .79)

NOTE. Values are means ± SD.

Abbreviations: n, no. in groups; BMI, body mass index; TBP, total body phosphorus; TBCa, total body calcium; TBK, total body potassium; NS, not significant.

45.7 cm. These detectors are positioned as 16 above the patient and 16 below the subject. The 5-minute count is optimized for the rapid decay ($t_{1/2}$ = 2.8 minutes) of the induced signal from body phosphorus generated by the fast reaction $^{31}P(n,\alpha)^{28}Al$. In this facility, the WBC is placed in a well-shielded room, which reduces the background activity and improves accuracy. The in vivo activation technique provides precision of ± 2.6% for TBP as measured in an anthropometric phantom.[12]

The patients in our study also had total body calcium (TBCa) and total body potassium (TBK) measurements by DGNA and WBC at the same facility.[13] We have incorporated part of the data on TBCa and TBK for subanalysis of TBP in our study.

Statistical Analysis

The data are expressed in grams for TBP. The average values are expressed as the mean ± standard deviation. Because the dependent variables (TBCa, TBK, TBP) were normally distributed, differences between blacks and whites were assessed by the unpaired *t* test. However, age, weight, height, and BMI depart from normality, so a rank-sum test was used instead of a *t* test. Means adjusted for height, weight, and age were obtained by analysis of covariance, in which height, weight, and age were the covariates. Multiple linear regression was used to determine what variables were significant predictors of TBP. We also used spline (segmented) regression models, in which 2 straight lines are fitted to join at a common point; with our data, we chose age = 50 years (average at menopause) as the common point or "knot" of the spline. The spline model would illustrate the sharp effect of menopause. All analyses were performed using version 6.12 of SAS (SAS Institute, Cary, NC).

RESULTS

The data for TBP was normally distributed in the 2 ethnic groups. The TBP values for the 2 groups and the anthropometric data are given in Table 1. The black women were heavier than the white women, and BMI was significantly higher in blacks than whites. Sixty percent of black women and 57% of white women were premenopausal in our study. The average age at menopause was 50.9 years. The estimation of physical activity did not show any interracial differences (Table 1). TBP makes up approximately 0.66% of the total body weight in black women versus 0.63% in white women (P = .16). The mean value of TBP in whites is lower by 10.4% when compared with blacks in all age groups combined (mean value for white women is 401.39 ± 57.48 g and for blacks, 447.75 ± 57.69 g, P = .0001). When TBP values are adjusted for weight and height, black women still had significantly higher TBP levels as compared with whites for decades 30 to 39, 40 to 49, 50 to 59, 60 to 69, and for all decades combined (Table 2).

Stepwise-multiple-regression analyses were performed to determine the significance of various demographic parameters,

Table 2. TBP Levels by Decades in Black and White Women

Age Group	Race	No.	Height (cm)	Weight (kg)	BMI (kg/m^2)	TBP (g)	Adjusted TBP* (g)
20-29	B	13	167.5 ± 7.7	67.4 ± 8.8	24.0 ± 3.0	476.1 ± 58.4	469.7
	W	10	165.6 ± 5.5	61.1 ± 9.3	22.2 ± 2.4	418.4 ± 58.5	426.7
	P value		.59	.07	.13	.03	.102
30-39	B	19	161.8 ± 6.9	67.0 ± 12.6	25.5 ± 4.0	459.9 ± 51.0	464.6
	W	27	164.8 ± 6.4	61.1 ± 7.8	22.6 ± 3.4	434.3 ± 47.3	431.0
	P value		.12	.09	.006	.088	.012
40-49	B	19	163.1 ± 4.1	69.8 ± 10.4	26.2 ± 3.8	459.4 ± 54.3	465.2
	W	27	164.9 ± 5.8	66.1 ± 8.9	24.2 ± 3.1	427.4 ± 55.3	423.9
	P value		.14	.24	.04	.028	.003
50-59	B	23	162.9 ± 5.7	69.7 ± 9.2	26.3 ± 3.4	422.2 ± 58.4	422.6
	W	25	164.2 ± 6.0	63.7 ± 8.9	23.7 ± 3.5	384.5 ± 47.0	384.1
	P value		.56	.03	.01	.017	.018
60-69	B	10	162.8 ± 6.1	71.3 ± 9.3	26.9 ± 3.3	418.6 ± 49.4	422.9
	W	38	163.1 ± 6.2	65.9 ± 10.0	24.7 ± 3.2	357.9 ± 40.3	356.8
	P value		.80	.05	.03	.0002	.0001

*Mean TBP adjusted for height and weight.

such as age, race, height, and weight. White race, increasing age, and decreasing height are significant predictors of decreased TBP ($P < .0001$), whereas weight is not a significant predictor ($P = .24$) for estimating TBP (Table 3). In terms of percent variation of TBP explained R^2, age is a somewhat stronger predictor of TBP compared with race (Table 3). From the regression, adjusting for age, height, and weight, white women, on average, are about 38.4 g lower than blacks for TBP.

Using separate linear regression models for the 2 races with age, height, and weight as independent variables, the decrease in TBP in black women is –1.59 g/yr (-0.33%/yr) and for white women, -2.08 g/yr (-0.45%/yr). The decrease of TBP with age was significant for both blacks and whites ($P = .0011$ and $P = .0001$, respectively), but there was no significant difference in the decrease of TBP between the 2 ethnic groups, ie, -1.59 versus –2.08 g/yr ($P = .31$).

Our model improves significantly in terms of R^2 if TBP decreases in a nonlinear fashion with age. The curve is shown in Fig 1. We chose age 50 as the "knot" in the linear spline model ($R^2 = .456$); there was minimal decrease in TBP until age 50 in both races combined followed by a rapid decrease. For both races combined, the decrease was –0.98 g/yr until age 50 ($P = .0273$) and –3.19 g/yr after age 50 ($P < .0001$). With the linear models, we have seen that weight is not a significant predictor of TBP, nor is the interaction of age and race (age × race), ie, there is no significant difference in the decrease of TBP between the 2 ethnic groups. This turns out to be also true for the spline model. Consequently, in the model depicted in Fig 1, we have ignored the insignificant weight and age × race factors; this explains why the curves are parallel for each race and are adjusted only for height and not weight.

We performed a multiple regression in which TBP was taken as a dependent variable, and TBCa and TBK were independent variables. Other independent variables, such as age, race, menopausal status, height, and weight were also included as candidates in the analysis. TBCa and TBK were found to be positively correlated with TBP ($r = .83$, $P < .0001$ and $r = .57$, $P < .0001$, respectively). TBCa was the major predictor of TBP ($R^2 = .68$) followed by TBK ($R^2 = .018$) and race ($R^2 = .012$). Although TBP and TBK are highly correlated ($r = .57$), TBK plays a relatively small role in the model predicting TBP ($R^2 = .018$) once TBCa is in the model. TBCa and TBP are very highly correlated ($r = .83$), and this high correlation generally holds for the various race and pre- or postmenopausal status subgroups. Virtually all of the variability of TBP can be explained by TBCa; TBK only contributes an R^2 of about 2%. Once TBCa is used as a predictor, TBK plays a relatively minor role, because it is well correlated with TBCa.

Table 3. Regression Coefficients (Mean ± SE) for TBP Using a Linear Model

Parameter	Coefficient ± SE	*P* Value	R^2
Intercept	−85.5 ± 88.1	.3330	
Age (yr)	−1.91 ± 0.26	.0001	.227
Height (cm)	3.60 ± 0.56	.0001	.116
Race*	−38.4 ± 6.7	.0001	.100
Weight (kg)	0.406 ± 0.346	.2420	.003
		Total R^2 = .446	

*Race is a categorical variable defined as race = 0 (black), race = 1 (white).

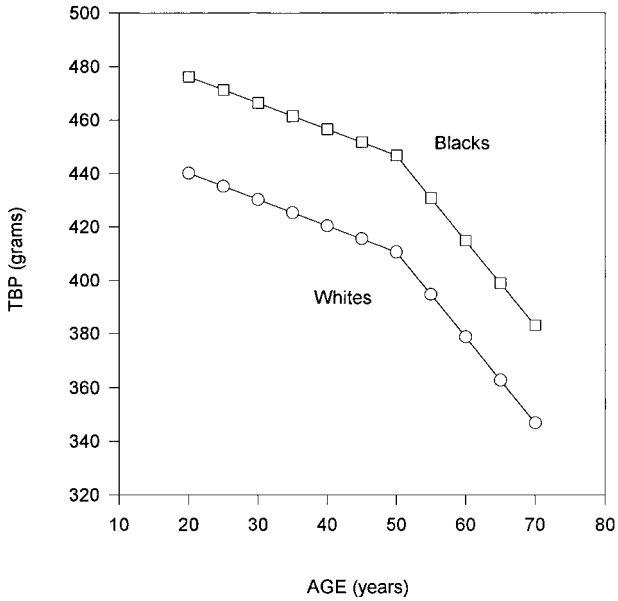

Fig 1. The change in TBP with age in black and white women using a spline model, adjusted to mean height for each race. The change at the time of menopause is apparent. For blacks younger than than 50, TBP = 495.76 – 0.98 × age ($P = .0273$); for blacks older than 50, TBP = 606.03 – 3.19 × age ($P < .0001$); for whites younger than 50, TBP = 456.02 – 0.98 × age ($P = .0273$); for whites older than 50, TBP = 566.29 – 3.19 × age ($P < .0001$).

DISCUSSION

TBP is significantly higher in black than white women. The white women have a mean TBP 10.4% lower than the black women. Because phosphorus is distributed mainly in the skeleton and muscle mass, it was anticipated that black women with higher BMI would have higher lean body mass and muscle mass leading to higher values of TBP, but our data suggests that BMI is not a major predictor of TBP. In fact, values of TBP in black women are significantly higher than white women when controlled for BMI and age. Multiple regression leads to the conclusion that TBP measurements depend significantly on age and race, in which age is a somewhat stronger predictor of TBP.

The involutional change in the TBP levels in black and white women in this study showed that the loss of TBP with aging is similar in both the ethnic groups (about 2 g/yr from the linear model). The rapid decrease of TBP after age 50 corresponds to the decrease in bone mass after the onset of menopause. Decrease in muscle mass with aging, as determined by TBK levels, has insignificant consequences on the levels of TBP. Thus, one would hypothesize that strategies, which could preserve skeletal mass with aging, would also maintain the TBP levels.

The TBCa data, which were analyzed previously on the same set of patients, showed that black women had about 8% higher TBCa values on average as compared with white women.[13] The

regression analysis showed that both black women and white women lost TBCa at similar rates with a gradual decrease in the premenopausal phase followed by a rapid decrease after menopause. The trend of TBK in the same subjects showed that the black women had higher TBK measurements than white women in general. The lifetime decrease of TBK was barely significant (8.2 g, $P < .04$) in black women, whereas the white women lost TBK markedly (23.6 g, $P < .0001$) with no evidence for an effect of menopause on decrease of TBK.[13] The close correlation between TBP and TBCa seems to be merely related to the fact that both of these elements are abundantly found in the skeletal compartment. There is no overlap in measuring these 2 elements by DGNA.

The earlier studies, which have reported TBP values in the population, were performed at the older facility for DGNA at BNL. In 1987, the WBC was upgraded and in 1991, a series of new new phantoms were used to calibrate the existing delayed gamma neutron activation systems at BNL.[14] This improved the reproducibility of measurement of TBP (coefficient of variation [CV] prior ± 4% *v* CV of ± 2.6% after the system upgrade). Ellis[8] has reported TBP values on the same system in 1989. He studied TBP in 1,134 white women (mean, 397 ± 64 g) and 35 black women (mean, 402 ± 57 g) between the ages of 25 to 75 years with no statistical difference between the groups.[8] In a previous study in 1976 at BNL, TBP values were measured in 45 women and 39 men ages 30 to 90 years.[9] This study showed that TBP values decreased with age in both men and women. In women, there was an accelerated rate of decrease of TBP corresponding to menopause similar to our study.

Measurement of TBP by DGNA not only gives an estimate of the bone mass, but can also be used to evaluate total body muscle mass in vivo in humans by applying Elemental Partition Analysis.[7] In this technique, TBP was first measured by IVNAA. The main contributors to TBP are the bone and skeletal muscle. Adipose tissue and the liver contribute to less than 3%. Dual x-ray absorptiometry (DXA) was then performed to evaluate the total bone mineral and therefore bone phosphorus. Soft tissue (nonbone) phosphorus is then derived by subtraction from TBP. Corrections were applied for the small contributions of the liver and adipose tissue to TBP to derive muscle phosphorus. This technique is anticipated to be used clinically in managing sarcopenia by evaluating interventions intended to maintain or increase muscle mass, such as exercise and growth hormone treatment. Another clinical use has been described in patients on dialysis or nondialysis patients with chronic renal insufficiency, in which TBP levels have been found to be altered, indicating increase in skeletal tissue and/or soft tissue calcification.[15] However, to study changes in disease states, it is essential to know the accurate reference values in a normal healthy population.

Our present data is the first to provide ethnic specific reference values for TBP in healthy women. This study also suggests that the decrease in TBP is similar in both ethnic groups. Loss of skeletal mass after menopause seems to be the major contributor to decrease in TBP levels. The proposed models of involutional loss should be confirmed by a longitudinal study in the various ethnic groups.

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