Estimated Equilibrated Dietary Intakes for Nine Minerals (Na, K, Ca, Mg, P, Fe, Zn, Cu, and Mn) Adjusted by Mineral Balance Medians in Young Japanese Females

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Summary The present study sought to determine estimated equilibrated dietary intakes (EEDIs) for nine essential minerals: sodium (Na), potassium (K), calcium (Ca), magnesium (Mg), phosphorus (P), iron (Fe), zinc (Zn), copper (Cu), and manganese (Mn), using data from 17 human mineral balance studies conducted from 1986 to 2007 (subjects=178). Among these studies, two used male subjects, two subjected some or all subjects to sodium restriction, and one study utilized a low protein diet; these subjects were not included in the present analysis. Consequently, data from 13 studies of young female subjects (n=131) consuming a standard diet were selected. Balance distribution medians for six of the minerals (Na, K, Mg, Fe, Zn and Cu) were positive, so the data were adjusted to set the medians of the balances to zero. Medians for the other minerals (Ca, P and Mn) were close to zero and were not adjusted. Intake and balance for each mineral were divided by body weight (BW), lean body mass (LBM), and standard body weight (SBW), which was calculated using height and standard body mass index (BMI=22), and EEDIs were calculated as the intercept of a simple regression equation. When relationships between intake and balance of a mineral were not significant in the regression equation, a significant regression equation comparing intake and balance of another mineral was used to calculate the intercept. Significant simple regression equations were not obtained from any of the three parameters of Na or Zn, or for two of the parameters of P; thus, K, Fe and Ca balances were used to determine the intercepts for Na, Zn and P, respectively. EEDIs for the minerals were: Na (67.9, 89.0, 62.5), K (39.5, 53.5, 37.4), Ca (11.0, 14.4, 10.1), Mg (4.18, 5.51, 3.86), P (18.7, 24.6, 17.3) (mg/kg BW/d, mg/kg LBM/d, mg/kg SBW/d), Fe (180, 237, 165), Zn (168, 241, 166), Cu (30.9, 42.6, 29.7), Mn (55.1, 72.1, 50.7) (μg/kg BW/d, μg/kg LBM/d, μg/kg SBW/d), respectively. These values are nearly identical to the mean dietary intakes.

Key Words essential minerals, intake, balance, estimated equilibrated dietary intake

Estimated equilibrated dietary intake (EEDI) is defined as the intercept of a linear regression equation between intake (Y) and balance (X) in a nutrient. EEDI is also one of the major indicators obtained experimentally from human balance studies to determine the estimated average requirement (EAR) (1).

The purpose of the present study was to determine the each of EEDIs for nine essential minerals: sodium (Na), potassium (K), calcium (Ca), magnesium (Mg), phosphorus (P), iron (Fe), zinc (Zn), copper (Cu), and manganese (Mn), using data from 17 human mineral balance studies conducted from 1986 to 2007 (subjects=178).

Previously, the authors reported equilibrated dietary intake data for Ca, Mg, and P (2), and Na and K (3) using mineral balance data obtained from 1986 to 2000 (11 experiments, 109 subjects). However, among these studies were one study restriction sodium for all subjects and another imposing sodium restriction some subjects (4, 5), and two more studies using male subjects (6–8).

The authors also conducted six more studies. One of these studies utilized a low protein diet with vitamin, mineral, and dietary fiber supplementation (9).

Subjects in the remaining twelve and a half studies (n=131) were all young females who consumed a standard diet without supplementation. Only these data were included in the present analysis to ensure homogeneous experimental conditions (Table 1). Some findings about the mineral balance studies included in this analysis were published previously (2–11).

SUBJECTS AND METHODS

Subject characteristics are shown in Table 2. In this study, the relationship between dietary intake and balance for each mineral was analyzed using three param-
eters: body weight (BW), lean body mass (LBM), and standard body weight (SBW); thus, our data may be extrapolated to obese or lean populations. BW is the average fasted morning body weight measured throughout the balance period. LBM was calculated based on BW and body fat percentage, which was estimated from body density (BD), as determined by the method of Keys and Brozek (12). BD was determined in advance based on the formula of Nagamine (13), based on skin-fold thickness (upper arm back/sub-scapula) of the less skillful hand by EIKEN skin fold calipers (14).

SBW was calculated based on average morning height measured throughout the balance period based on the formula proposed by the Japan Society for the Study of Obesity (JASSO) (15): SBW = 22.2 × [Height (m)]² (kg SBW).

For each experiment, all subjects gave written informed consent. All studies in the present analysis were carried out according to the rules of the Helsinki Declaration. The ethical committee of the National Institute of Health and Nutrition, established in 1990, approved all studies. All studies were carried out in the

### Table 1. Subjects, balance study duration and daily energy and nutrient intake.

| Sex | Sub. (n) | Balance study duration (d) | Ex. | Ref. | Calculated values | Measured values |
|-----|---------|---------------------------|-----|------|------------------|-----------------|
|     |         |                           |     |      | Energy (kcal/d)  |    | Protein (g/d)  |    | Lipid (mg/d)  |    | Carbo. (mg/d)  |    | Na (mg/d)      | K (mg/d) | Ca (mg/d) | Mg (mg/d) | P (mg/d) | Zn (mg/d) | Fe (mg/d) | Cu (mg/d) | Mn (mg/d) |
| 1   | F       | 12                        | 8   |      | 1.557           | 74.5 | 65.7           | 166.8 | 3.894           | 2.550 | 672 | 261 | —   | 8.86 | 9.42 | 1.47 | 3.64 |
| 2   | F       | 8                         | 10  |      | 1.729           | 77.8 | 48.5           | 243.9 | 3.693           | 2.472 | 719 | 279 | 1.116 | 9.31 | 12.93 | 1.46 | 3.30 |
| 3   | F       | 12                        | 8   |      | 1.636           | 65.3 | 64.0           | 194.8 | 3.462           | 2.085 | 502 | 196 | 891 | 7.58 | 10.64 | 1.28 | 2.37 |
| 4   | F       | 8                         | 12  |      | *1 1.930         | 86.8 | 57.0           | 263.2 | 4.055           | 2.679 | 629 | 261 | 1.259 | 24.31 | 12.62 | 2.54 | 3.09 |
| 5   | F       | 7                         | 2   |      | *1 1.815         | 75.8 | 52.8           | 253.2 | 2.488           | 2.194 | 653 | 214 | 1.182 | 13.23 | 11.24 | 2.20 | 2.92 |
| 6   | F       | 8                         | 8   |      | 1.712           | 68.8 | 47.5           | 251.0 | 3.077           | 2.204 | 671 | 244 | 1.158 | 9.32 | 10.77 | 2.37 | 3.22 |
| 7   | F       | 12                        | 8   |      | *2 1.922         | 66.0 | 68.0           | 257.7 | 3.445           | 1.829 | 294 | 188 | 833 | 7.01 | 10.25 | 1.38 | 2.75 |
| 8   | F       | 11                        | 8   |      | *2 1.906         | 65.6 | 67.9           | 254.5 | 3.398           | 1.864 | 347 | 186 | 807 | 7.83 | 8.74 | 2.07 | 2.63 |
| 9   | F       | 12                        | 8   |      | 1.633           | 59.0 | 41.1           | 252.7 | 3.298           | 1.719 | 659 | 185 | 882 | 7.09 | 8.43 | 2.39 | 2.35 |
| 10  | F       | 11                        | 8   |      | 1.647           | 61.2 | 45.2           | 244.4 | 3.308           | 1.813 | 510 | 160 | 837 | 7.64 | 5.52 | 1.05 | 2.37 |
| 11  | F       | 10                        | 8   |      | 1.638           | 54.5 | 45.6           | 249.1 | 3.315           | 2.009 | 661 | 288 | 895 | 7.16 | 6.38 | 1.25 | 3.53 |
| 12  | F       | 6                         | 12  |      | 1.906           | 73.2 | 42.9           | 303.5 | 4.840           | 2.400 | 646 | 232 | 1.083 | 9.21 | 12.26 | 2.01 | 3.24 |
| 13  | F       | 12                        | 8   |      | 1.659           | 62.4 | 45.3           | 245.3 | 3.489           | 1.936 | 534 | 181 | 851 | 8.24 | 5.61 | 1.10 | 2.94 |
| Total | 131    |                           |     |      | Max 1.930       | 86.8 | 68.0           | 303.5 | 4.840           | 2.679 | 719 | 288 | 1.259 | 24.31 | 12.93 | 2.54 | 3.64 |
|      |        | Min 1.557                  |      |      | 54.5           | 41.1 | 166.8          | 4.488 | 1.719           | 294 | 160 | 807 | 7.01 | 5.52 | 1.05 | 2.35 |

*1: Semi-dried oyster was used in dietary menu to increase zinc (Zn) intake.
*2: Low calcium study.
—: Not determined.
Sub.: subjects, Ex.: exercise using a bicycle ergometer was performed to estimate sweat mineral losses, Carbo.: carbohydrate.
Metabolic studies for the evaluation of nutritional balance, in which subjects have to eat all food supplied and maintain a set schedule, have the potential to be stressful for subjects. However, the results of one experiment (No. 12, Table 1) showed that participation in the metabolic study had almost no effect on subjective fatigue level (16) or immunoglobulin levels (17).

The foods used in each study were selected from those commercially available. Some foods were avoided because of heterogeneous nutrient content revealed through chemical measurements taken prior to the studies. Both processed and nonperishable foods were purchased at the same time from the same lot before the experiments so as to ensure consistent nutrient content. Fresh foods were obtained from the same district and the same market.

Dietary menus were designed by a registered dietitian so as to meet dietary allowances in Japan (17), except for the experiments involving special diets, for which food composition tables were used (18, 19). All the menus calculated using old food composition tables (19) were re-calculated by the new tables (20) for this analysis.

All foods were washed with ion-free water passed through an ion-exchange resin, if necessary, weighed, cooked separately, and distributed uniformly among dishes for the subjects and diet samples.

The subjects were required to consume all of the diet. They were allowed no other food, but could drink as much ion-free water as they wanted. The weight of the water consumed was measured and recorded.

Duplicated diet samples were preserved throughout the experiments and kept in a refrigerator for 1 d until blending. For blending, refrigerated diet samples were weighed and put into a mixer (MX150S, National, Japan). An adequate volume of ion-free water was added, and then the mixture was gradually homogenized for about 30 min by a slide transmission (RIKO-SLIDETRANS RSA-5, Tokyo-Rikosha, Japan) attached to the mixer (21).

The homogenized diet samples were prepared in triplicate and weighed in polypropylene bottles. Five milliliters of nitric acid (UGR grade, Kanto Chemical Co., Inc, Japan) was then added and the mixture was kept at room temperature until digestion.

Digestion was performed in heat-resistant glass beakers (Pyrex, Iwaki Glass, Japan) on a hot plate at temperatures below 140˚C, with nitric acid and hydrogen peroxide (for trace analysis, Wako Pure Chemical Industries, Ltd., Japan). The interior of the bottle was washed with nitric acid added to the sample in the beaker (21).

After digestion, an adequate volume of pure water (Milli-Q, Millipore, Japan) was added to the samples, which were then put on a hot plate at a temperature below 90˚C for one night to determine phosphorus content. Then, 0.5 M nitric acid was added to attain a fixed volume (21).

Eight minerals (Na, K, Ca, Mg, Fe, Zn, Cu, and Mn) were measured by an atomic absorption spectrophotometer (AAS, Varian AA-5, Australia) after the mixture was diluted to an appropriate concentration with
When measuring Ca, a final concentration of 2,500 ppm strontium (strontium chloride hexahydrate for AAS analysis, Wako Pure Chemical Industries, Ltd.) was added to the samples to avoid interference due to P. The height of the burner was adjusted to create the same absorption factor for the same Ca concentration at different pH values, thereby avoiding interference due to pH (2, 5). Phosphorus was measured by a colorimeter (Molybdenum blue method) (5, 21).

Fecal specimens were collected throughout the experiment and were separately sampled for the balance periods based on the appearance of the ingested marker in the feces (5). Fecal samples were measured in the same way as dietary samples.

Urine samples were directly diluted in 0.5 M nitric acid, and measured in the same way as the other samples. Urine Fe, Cu, and Mn content was low and relatively consistent among the first three experiments, and thereafter, urine output of Fe, Cu, and Mn was presumed to be 0.1 mg/d, 0.05 mg/d, and 0.01 mg/d, respectively, without measurement.

Arm sweat during exercise was collected after cleaning the skin surface on one side of the hand and arm with pure water and ion-free gauze treated with ethylene-diamine-tetra-acetic acid di-ammonium salt (EDTA: Wako Pure Chemical Industries, Ltd.), covering the whole hand and arm with a long polyethylene bag wrapped with tape. The collected sweat was filtered through a 0.10 μm pore size Teflon filter (Fluoro pore, Sumitomo Electric Industries, Ltd., Japan) and ethanol (for trace analysis, Wako Pure Chemical Industries, Ltd.) to remove any solids. Sweat samples were measured in the same way as the other samples. Sweat volume was calculated from weight loss during exercise as estimated by a balance (sensitivity 10 g) (5). Total sweat loss of minerals (Na, K, Ca, Mg and Zn) during exercise throughout the balance period was divided by the number of days in the balance period, and calculated as sweat loss per day. Sweat P, Fe, Cu, and Mn content during exercise was so low and under the limit of measurement, that dermal output of Fe, Cu, and Mn was not taken into consideration when calculating the balance. Dermal losses of minerals except through sweat in the physical exercise were not measured in any subjects, but considered when medians of the balance distributions in minerals were positive.

Additional indicators in this paper are defined as follows:

\[
\text{Apparent absorption} = \frac{\text{Intake} - \text{Fecal output}}{\text{Intake}} \times 100\%
\]

\[
\text{Balance} = \frac{\text{Intake} - \left[\frac{\text{Fecal output}}{\text{Intake}} \times \text{Sweat loss}}\right]}{\text{Urine output}} + \text{Sweat loss} \times 100\%
\]

*Only when sweat loss during exercise was estimated.

Intake and balance for each mineral were divided by body weight (BW), lean body mass (LBM), and standard body weight (SBW). Then, estimated equilibrated dietary intakes (EEDIs) were calculated as the intercept
of a simple regression equation.

Statistical analysis was performed using Stat View-J 5.0.

The authors found the medians of the balance distributions for six minerals (Na, K, Mg, Fe, Zn, Cu) were positive, so the data were adjusted to set the medians for the balances to zero. Medians for the other minerals analyzed (Ca, P, and Mn) were not altered.

Adjusted regression equation is calculated as follows:

Original regression equation:

\[ y = ax + b \]

\[ x_1: \text{median of balance distribution} \]

Adjusted regression equation:

\[ y = a(x - x_1) + b + ax_1 \]
Estimated Equilibrated Dietary Intakes for Minerals (Na, K, Cu, Mg, P, Fe, Zn, Ca, and Mn)

Table 3–1.  Equilibrated intake of 9 minerals (n=131).

1) Estimated by mg (µg)/kg body weight (BW)/d

| Element      | Units          | Intake | Balance | Equilibrated intake | r²   | p        |
|--------------|----------------|--------|---------|---------------------|------|----------|
|              | Mean ±SD       | Median | Mean ±SD | Mean ±SD (% of Intake) | Intercept | 95% CI   |
| Sodium (Na)  | mg/kg BW/d     | 68.0±9.7 | 67.0    | 6.07±4.06  | 6.04  | (9.0)    | 67.9 | 66.4–69.4 | 0.207 | <0.0001 |
|              |                |        |         |         |       |          | 62.0 | 59.4–64.5 |        |          |
| Potassium (K)| mg/kg BW/d     | 40.7±6.8 | 39.2    | 4.79±3.47  | 4.68  | (11.9)   | 40.7 | 39.5–41.8 | 0.055 | 0.0071 |
|              |                |        |         |         |       |          | 38.5 | 36.6–40.5 |        |          |
| Calcium (Ca)| mg/kg BW/d     | 11.0±2.8 | 11.5    | 0.04±1.52  | 0.00  | (0.0)    | 11.0 | 10.5–11.4 | 0.058 | 0.0057 |
| Magnesium (Mg)| mg/kg BW/d | 4.20±0.82 | 4.08    | 0.18±0.39  | 0.16  | (3.9)    | 4.18 | 4.05–4.32 | 0.142 | <0.0001 |
|              |                |        |         |         |       |          | 4.06 | 3.91–4.20 |        |          |
| Phosphorus (P)| mg/kg BW/d | 18.6±3.5 | 17.6    | −0.18±1.45 | −0.21 | (−1.2)   | 18.7 | 18.1–19.3 | 0.035 | 0.0412 |
| Iron (Fe)    | µg/kg BW/d     | 180±49 | 179     | 28.9±28.7 | 28.1  | (15.7)   | 180  | 172–188   | 0.094 | 0.0004 |
|              |                |        |         |         |       |          | 165  | 154–177   |        |          |
| Zinc (Zn)    | µg/kg BW/d     | 182±82 | 160     | 14.5±24.1 | 15.4  | (9.6)    | 181  | 168–195   | 0.059 | 0.0053 |
|              |                |        |         |         |       |          | 162  | 142–182   |        |          |
| Copper (Cu)  | µg/kg BW/d     | 32.8±10.7 | 28.7   | 4.78±10.5 | 4.14  | (14.4)   | 32.3 | 30.9–33.8 | 0.416 | <0.0001 |
|              |                |        |         |         |       |          | 29.6 | 28.0–31.2 |        |          |
| Manganese (Mn)| µg/kg BW/d | 55.0±8.1 | 53.6    | −0.48±8.11 | 0.89  | (1.7)    | 55.1 | 53.4–56.7 | 0.063 | 0.0165 |

1 Equilibrated intake of Na was obtained from the relationship between Na intake and adjusted K balance.
2 Equilibrated intake of P was obtained from the relationship between P intake and Ca balance (only in Table 3–2 and 3–3) (n=119).
3 Equilibrated intake of Zn was obtained from the relationship between Zn intake and adjusted Fe balance.
4 n=90.

Then, EEDIs were calculated as the intercept of a simple regression equation between dietary intake and the (adjusted) balance. When there was no significance in the regression equation between the intake and (adjusted) balance of a mineral, a significant regression equation for the intake and (adjusted) balance of another mineral was used to calculate the intercept. Ninety-five percent confidence intervals (95% CI) for the intercepts were also calculated.

RESULTS

Among the 131 subjects in the 13 balance studies, 12 subjects (No. 1, Table 1) did not undergo phosphorus measurement (P). So, P balance data is from 119 subjects; 40 subjects (Nos. 1–4, Table 1) did not have reliable fecal manganese (Mn) measurements as revealed by variation among triplicate samples, so Mn balance was measured in only 91 subjects.

Urine output of Fe, Cu, and Mn was low and relatively consistent among the first three experiments, and thereafter, urine output of Fe, Cu, and Mn was presumed to be 0.1 mg/d, 0.05 mg/d, and 0.01 mg/d, respectively.

The distributions of the balances of the nine minerals were analyzed. Histograms of mineral content divided by kg BW/d are shown in Fig. 2. The medians of the balance distributions for six minerals (Na, K, Mg, Fe, Zn, and Cu) were positive, so the data were adjusted to set the medians of the balances to zero. Medians for the other minerals analyzed (Ca, P, and Mn) were not altered. Means of the balances ranged from −0.97 to 16.03%, and SDs from 5.97 to 31.95% of the corresponding mineral intake.

Estimated equilibrated dietary intakes (EEDIs) and corresponding 95% CIs were calculated as the intercept of a simple regression equation between dietary intake and (adjusted) balance for each mineral. EEDIs divided by kg BW/d are shown in Fig. 3.

The scales of the two axes in the figures are the same. EEDIs divided by kg LBM/d and by kg SBW/d are not shown as figures.

Significant simple regression equations were not obtained from any of the three estimations of Na and Zn, or from two of the estimations for P. In these cases, adjusted K, adjusted Fe, and Ca balances were used to determine the intercepts for Na, Zn, and P, respectively. Those data divided by kg BW/d are shown in Fig. 4. Those data divided by kg LBM/d or by kg SBW/d are not...
shown as figures.

Intakes, balances, and estimated equilibrated dietary intake adjusted and pre-adjusted balances for the nine minerals divided by the three parameters are shown in Table 3.

**DISCUSSION**

This is the first mineral balance study to analyze nine minerals at the same time in 131 human subjects.

**Experimental protocol and duration of balance periods**

This experiment consisted of three phases: the pre-balance (2–4 d), balance (8–12 d), and post-balance (3–4 d) periods, each consisting of a special diet with a 4- or 5-d rotating menu. The results of the study confirmed that the duration of the experiments and the balance periods were sufficient to estimate the relationship between the intake and balance of the minerals measured.

**Data selection**

In this analysis, data from four and a half experiments were excluded because of heterogeneous environmental and dietary conditions. In addition, the authors did not include data from experiments in which subjects underwent sodium restriction, because sodium restriction also decreases the absorption of Ca and Mg (22, 23). However, in this analysis, the authors included both data from experiments in which subjects underwent physical exercise and in which subjects were sedentary, because physical activity levels of a population group were not taken into consideration in determination of EAR for minerals (1). Further analysis is required to clarify the effects of physical exercise on mineral balance in humans.

**Adjustment of the balance medians to zero**

Mineral balances in this study followed a normal distribution. However, the medians of the balances for six minerals (Na, K, Mg, Fe, Zn, and Cu) were positive, ranging from 3.9 to 16.1% of the median intake of the corresponding mineral, while the others (Ca, P, Mn) were close to zero. The authors believe these results reflect the fact that some minerals are lost through other pathways such as through the skin and menstruation, rather than in urine and feces, and that regulatory mechanisms including intestinal absorption, intestinal excretion, renal excretion, and exchange to and from bone and cells successfully maintain mineral balances at zero. Thus, all the medians were set to zero to determine EEDIs.

| Table 3–2. Equilibrated intake of 9 minerals (n=131). |
|---|---|---|---|---|---|---|---|---|
| **Element** | **Units** | **Intake** | **Balance** | **Equilibrated intake** | **r^2** | **p** |
| | | Mean±SD | Median | Mean±SD | Median | Intercept | 95% CI |
| Sodium (Na) | mg/kg LBM/d | 89.4±12.7 | 86.7 | 7.90±5.32 | 7.48 (8.6) | 89.0 | 87.1–91.0 (before adjustment) |
| Potassium (K) | mg/kg LBM/d | 53.7±9.8 | 51.8 | 6.31±4.59 | 6.03 (11.6) | 53.5 | 51.9–55.2 (before adjustment) |
| Calcium (Ca) | mg/kg LBM/d | 14.4±3.8 | 15.2 | 0.06±1.94 | 0.00 (0.0) | 14.4 | 13.8–15.0 (before adjustment) |
| Magnesium (Mg) | mg/kg LBM/d | 5.54±1.17 | 5.27 | 0.24±0.52 | 0.21 (4.0) | 5.51 | 5.32–5.70 (before adjustment) |
| Phosphorus (P) | mg/kg LBM/d | 24.5±5.0 | 22.8 | −0.24±1.88 | −0.28 (−1.2) | 24.6 | 23.7–25.5 (before adjustment) |
| Iron (Fe) | μg/kg LBM/d | 237±67 | 235 | 38.1±37.3 | 37.9 (16.1) | 237 | 226–248 (before adjustment) |
| Zinc (Zn) | μg/kg LBM/d | 241±118 | 204 | 19.2±31.1 | 19.3 (9.5) | 241 | 221–261 (before adjustment) |
| Copper (Cu) | μg/kg LBM/d | 43.1±14.1 | 38.6 | 6.24±13.50 | 5.47 (14.2) | 42.6 | 40.6–44.5 (before adjustment) |
| Manganese (Mn) | μg/kg LBM/d | 71.9±11.5 | 70.7 | −0.53±10.19 | 1.12 (1.6) | 72.1 | 69.8–74.4 (before adjustment) |

1 Equilibrated intake of Na was obtained from the relationship between Na intake and adjusted K balance.
2 Equilibrated intake of P was obtained from the relationship between P intake and Ca balance (only in Table 3–2 and 3–3 (n=119).
3 Equilibrated intake of Zn was obtained from the relationship between Zn intake and adjusted Fe balance.
4 n=90.
Determination of EEDIs using balance data for other minerals

Mineral balance of Na and Zn divided by kg BW/d was not significantly increased by increased dietary intake, and there was no significant increase in Na, P, or Zn balance divided by kg LBM/d or by kg SBW/d. For these minerals, the authors used a significant relationship between the intake and balance of a non-corresponding mineral instead of the non-significant balance of the corresponding mineral to determine the intercepts of the intake.

Significant relationships between the intakes and balances of non-corresponding minerals suggested the presence of unknown mutual relationships among the minerals used for the determination of the intercepts. That is, some unknown mechanisms must play essential roles in mineral metabolism by controlling intestinal absorption, renal excretion, and transportation to and from cell and bone across extra cellular space at the same time affecting plural minerals. In our previous studies, metabolism of Na and K (4, 5, 24), Ca, Mg, and P (2, 8, 25, 26) and fecal minerals (27) affected each other.

In addition, the authors previously reported significant correlations between Na intake and Ca and Mg balance (22), suggesting that there are relationships between the intake of one mineral and the balance of another one, although the relationships among minerals have not been fully demonstrated.

Comparision of percent of the standard deviations of the balances to intakes

In this study, comparison of percent standard deviations (SDs) of the balances to intakes of the corresponding minerals ranged widely, from 5.97 to 31.92%. SDs for Na, K, Mg, and P were within 10% of intake, while SDs for Ca, Fe, Zn, and Mn ranged from 10 to 20% of intake. SD for Cu was more than 30% of intake. These findings demonstrate that regulation of mineral balance differs among minerals, but the reasons for these differences are not clear. Further analysis and experiments are needed.

EEDI values are close to mean intakes of the corresponding minerals

The three intercepts of the regression equations comparing Na intake and balance of Na (not significant), K (significant), and Mg (significant) were 68.033, 67.895, and 67.890 mg/kg BW/d, respectively, and all three intercepts were close to the mean value for Na.

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**Table 3–3. Equilibrated intake of 9 minerals (n=131).**

| Element          | Units          | Intake Mean ±SD | Median | Balance Mean ±SD | Median (of Intake) | Equilibrated intake | r² | p         |
|------------------|----------------|-----------------|--------|------------------|-------------------|---------------------|----|-----------|
| Sodium (Na)³     | mg/kg SBW/d    | 62.5±8.2        | 61.0   | 5.58±3.81        | 5.5               | 62.5                | 0.221 | <0.0001   |
|                  |                | 57.3            | 55.1–59.4 |                  |                   |                     | (before adjustment) |
| Potassium (K)    | mg/kg SBW/d    | 37.5±6.0        | 36.3   | 4.40±3.24        | 4.37              | 37.4                | 0.049 | 0.0115    |
|                  |                | 35.7            | 33.9–37.4 |                  |                   |                     | (before adjustment) |
| Calcium (Ca)     | mg/kg SBW/d    | 10.1±2.5        | 10.9   | 0.04±1.36        | 0.00              | 10.1                | 0.107 | 0.001    |
| Magnesium (Mg)   | mg/kg SBW/d    | 3.87±0.78       | 3.63   | 0.17±0.36        | 0.15              | 3.86                | 0.123 | <0.0001   |
| Phosphorus (P)²  | mg/kg SBW/d    | 17.2±3.1        | 16.1   | -0.14±1.31       | -0.18             | 17.2                | 0.048 | 0.0169    |
| Iron (Fe)        | μg/kg SBW/d    | 166±44          | 170    | 26.5±25.9        | 24.3              | 165                 | 0.093 | 0.0082    |
|                  |                | 152             | 142–163 |                  |                   |                     | (before adjustment) |
| Zinc (Zn)³       | μg/kg SBW/d    | 167±76          | 144    | 13.2±21.7        | 14.1              | 166                 | 0.048 | 0.0116    |
|                  |                | 150             | 132–169 |                  |                   |                     | (before adjustment) |
| Copper (Cu)      | μg/kg SBW/d    | 30.1±9.6        | 26.1   | 4.33±9.43        | 3.78              | 29.7                | 0.395 | <0.0001   |
|                  |                | 27.3            | 25.9–28.7 |                 |                   |                     | (before adjustment) |
| Manganese (Mn)⁴  | μg/kg SBW/d    | 50.6±7.7        | 49.1   | -0.40±7.21       | 0.81              | 50.7                | 0.078 | 0.0072    |
|                  |                | 49.1            | 49.1–52.2 |                 |                   |                     | (before adjustment) |

¹ Equilibrated intake of Na was obtained from the relationship between Na intake and adjusted K balance.
² Equilibrated intake of P was obtained from the relationship between P intake and Ca balance (only in Table 3–2 and 3–3) (n=119).
³ Equilibrated intake of Zn was obtained from the relationship between Zn intake and adjusted Fe balance.
⁴ n=90.
intake (68.0 mg/kg BW/d) (Figs. 3 and 4). In most cases, the intercepts of regression equations comparing intake of one mineral and balances of both corresponding and non-corresponding minerals were close to the mean intake. This may be due to having mineral intake levels near the required levels.

However, the intercept of the correlation equation between Zn intake and Mn balance (162.9 μg/kg BW/d) was lower than the mean intake of Zn (182 μg/kg BW/d). Data in Fig. 4 indicate that this may be because the data in rows 1–4, Table 1, including the experiment with the highest Zn intake (24.31 mg/d) were omitted in this analysis because of the absence of Mn data. Using the data from rows 5–13 in Table 1, in which Mn data are included, the mean intake of Zn was calculated to be 162.4 μg/kg BW/d, close to the intercept for Zn intake compared with Mn balance (162.9 μg/kg BW/d) or Zn balance (162.7 μg/kg BW/d, not significant). Lower mean intake of Zn in the experiments with Mn data also lowered the intercept for Zn. These data suggest that EEDIs will vary depending on the dietary intake of minerals when the balance of a mineral is within a relatively narrow range, but its intake ranges relatively widely, such as in the case of Zn.

In contrast, EEDI changes may not occur in a mineral if balance is strongly correlated with intake, such as in the case of Cu.

**EEDI for Zn**

The authors found that the EEDI for Zn fluctuated depending on levels of Zn intake. High Zn diets are not rare because certain foods, such as oysters, liver, and beef, contain relatively large amounts of Zn (20). In two studies not included in the present analysis, large quantities of Zn (38.0 and 27.9 mg/d) were ingested by subjects, and more than half of subjects (11/19) showed elevated Zn balances (more than 4 mg/d; 12–24% of the intake) (7, unpublished observation). Given these results, the authors calculated EEDI for Zn based on Fe balance, where all data (n=131) were used. The EDDIs presented in this report may not be sufficient to promote health in humans.

The present human mineral balance studies were performed on subjects maintained in comfort in a controlled environment. An experiment confirmed that participants were not subjected to excessive stress (16, 17). However, under normal living conditions, Ca and Mg were reported to be conserved in response to light exercise (27, 28), but lost in response to heavy exercise (28, 29), excess energy intake (30–32), or stress (32, 33). Relatively large amounts of Na, K, Zn are excreted in sweat in response to heat exposure (34) or physical exercise (28, 35). We are ignorant about the shortage of these minerals in cell and bone where minerals are heterogeneously distributed, because serum mineral levels are relatively constant (36, 37).

These data suggest that under normal living conditions, mineral intake greater than the EEDIs may be needed to maintain optimal human health. Further studies are needed to determine the actual mineral intake levels required to maintain health.

**CONCLUSIONS**

The authors determined the estimated equilibrated dietary intakes (EEDIs) for nine essential minerals: sodium (Na), potassium (K), calcium (Ca), magnesium (Mg), phosphorus (P), iron (Fe), zinc (Zn), copper (Cu), and manganese (Mn), using mineral balance data from 13 studies of young female subjects (n=131) consuming a standard diet.

The medians of the balance distributions for six minerals (Na, K, Mg, Fe, Zn and Cu) were positive, and were adjusted to zero. Medians for the other minerals (Ca, P and Mn) were close to zero and were not adjusted.

Intake and balance of each mineral was divided by body weight (BW), lean body mass (LBM), and standard body weight (SBW), which was calculated by height and standard body mass index (BMI=22), and EEDIs were calculated as the intercepts of a simple regression equation comparing intake and balance of a mineral.

Significant simple regression equations were not obtained in any of the three parameters for Na or Zn, or in two parameters for P; thus, the K, Fe, and Ca balances were used to determine the intercepts for Na, Zn, and P, respectively. However, the intercept of the correlation equation between Zn intake and Mn balance (162.9 μg/kg BW/d) was lower than the mean intake of Zn (182 μg/kg BW/d), suggesting that EEDIs may be altered depending on the dietary intake of a mineral when the balance of that mineral is within a relatively narrow range, but intake ranges relatively widely, such as in the case of Zn.

EEDIs for each element are as follows: Na (67.9, 89.0, 62.5), K (39.5, 53.5, 37.4), Ca (11.0, 14.4, 10.1), Mg (4.18, 5.51, 3.86), P (18.7, 24.6, 17.3) (mg/kg BW/d, mg/kg LBM/d, mg/kg SBW/d), Fe (180, 237, 165), Zn (168, 241, 166), Cu (30.9, 42.6, 29.7), Mn (55.1, 72.1, 50.7) (μg/kg BW/d, μg/kg LBM/d, μg/kg SBW/d), respectively.

These values are very similar to the mean dietary intakes.

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