

# **Dietary Reference Intakes for Japanese (2015)**

**Ministry of Health, Labour and Welfare**

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March 2018

This translation work was realized with the great help of Dr. Satoshi SASAKI (Toyko University) and Dr. Aki SAITO (National Institutes of Biomedical Innovation, Health and Nutrition).

# Contents

<b>I Development and Application of Dietary Reference Intakes for Japanese</b>	<b>1</b>		
1 Introduction	2		
2 Basics of Development	6		
3 Considerations for Development	13		
4 Application of the Dietary Reference Intakes	19		
<b>II Energy and Nutrients</b>	<b>32</b>		
Energy	33		
Protein	66		
Dietary Fat	81		
Carbohydrate	104		
Energy Providing Nutrients' Balance	115		
Vitamins			
(1) Fat-soluble Vitamins	(2) Water-soluble Vitamins		
Vitamin A	126	Vitamin B <sub>1</sub>	154
Vitamin D	131	Vitamin B <sub>2</sub>	157
Vitamin E	137	Niacin	160
Vitamin K	140	Vitamin B <sub>6</sub>	163
		Vitamin B <sub>12</sub>	167
		Folate	170
		Pantothenic acid	174
		Biotin	177
		Vitamin C	180
Minerals			
(1) Macrominerals	(2) Microminerals		
Sodium	201	Iron	236
Potassium	206	Zinc	246
Calcium	210	Copper	249
Magnesium	216	Manganese	251
Phosphorus	218	Iodine	253
		Selenium	257
		Chromium	260
		Molybdenum	262

**Development and Application  
of Dietary Reference Intakes for Japanese**

## 1. Introduction

*The Dietary Reference Intakes for Japanese* proposes reference values for the intake of energy and nutrients, in the Japanese population, comprising both healthy individuals and groups, for the promotion and maintenance of health, and to prevent the occurrence of lifestyle-related diseases (LRDs).

The objectives behind the development of the *Dietary Reference Intakes for Japanese (2015)* are shown in Figure 1. The formulation of the dietary reference intakes (DRIs) employed the basic concepts of Health Japan 21 (second term)--a national health program initiated in 2013--based on the progression of aging and increase in the prevalence of diabetes and other diseases. The DRIs were developed to prevent the onset and progression of LRDs, as well as to maintain and promote health. To achieve these goals, the DRIs were determined in coordination with the guidelines for various other related diseases too.

The DRIs were determined based on scientific findings, the data of which were available. If some issues were deemed important, and yet no sufficient corresponding scientific data were available, these research topics were summarized and organized.

### 1-1. Target Individuals and Groups

The DRIs are intended for use by healthy individuals and groups, including individuals leading independent daily lives, despite having a risk of hypertension, dyslipidemia, hyperglycemia, and chronic kidney disease. They are aimed, specifically, at those who are able to participate in normal physical activities, such as walking or performing household tasks, and those with a body mass index (BMI\*) that does not deviate markedly from the standard. The inclusion range of individuals with the risk of the above-stated diseases are the level of “receiving health guidance.” For the treatment of individuals and groups with diseases, or who are at a high risk for a certain disease, nutritional management is implemented under the treatment guidelines for the disease in question, on the basis of an understanding of the basic concept of energy and nutrient intake, in the DRIs.

$$*BMI = \text{body weight (kg)} \div (\text{height [m]})^2$$

### 1-2. Targeted Energy and Nutrients for the Development of the DRIs

The DRIs for energy and nutrients, as presented in Figure 2, were determined on the basis of the Health Promotion Act. Additionally, the nutrient intake, essential to health maintenance and promotion, was examined quantitatively, and the presence of nutrients, which can scientifically be deemed sufficiently reliable, was also investigated.

**Figure 1. Dietary Reference Intakes Determined on the Basis of the Health Promotion Act**

1. Matters regarding the desired amount of calories citizens should consume to maintain and promote their health.

2. Matters regarding the optimal consumption of the following nutrients, for the maintenance and promotion of health:

(a) Nutrients, the deficiency of which, as stipulated by an ordinance of the Ministry of Health, Labour and Welfare, affects citizens' health maintenance and promotion, from the standpoint of the current nutrient intake status

- Proteins
- n-6 fatty acids and n-3 fatty acids
- Carbohydrates and dietary fiber
- Vitamin A, vitamin D, vitamin E, vitamin K, vitamin B<sub>1</sub>, vitamin B<sub>2</sub>, niacin, vitamin B<sub>6</sub>, vitamin B<sub>12</sub>, folic acid, pantothenic acid, biotin, and vitamin C
- Potassium, calcium, magnesium, phosphorus, iron, zinc, copper, manganese, iodine, selenium, chromium, and molybdenum

(b) Nutrients, the deficiency of which, as stipulated by an ordinance of the Ministry of Health, Labour and Welfare, affects citizens' health maintenance and promotion, from the standpoint of the nutrient intake status

- Fat, saturated fatty acids, and cholesterol
- Sugars (limited to monosaccharides or disaccharides and non-sugar alcohols)
- Sodium

### 1-3. Purposes and Types of Reference Values

#### • Energy

For energy, reference values were set to avoid excessive or deficient intakes.

#### • Nutrients

For nutrients, the DRIs have five types of values designed for three purposes: avoiding inadequacy, avoiding adverse health effects due to excessive intake, and preventing LRDs (Figure 2).

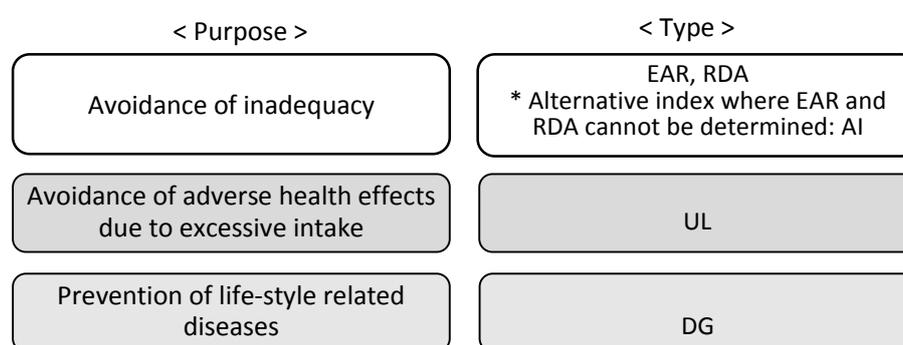
To prevent inadequate intake, the estimated average requirement (EAR) was determined. The EAR is the intake amount that would meet the nutrient requirements of 50% of the population. The recommended dietary allowance (RDA) was determined to supplement the EAR. The RDA is the intake amount that would meet the requirements of most of the population.

The adequate intake (AI) was developed for cases in which the EAR and RDA could not be set, due to insufficient scientific evidence. The AI indicates the intake amount that is

adequate to maintain a certain nutritional status. Having dietary intakes that are no less than the AI minimize the risk of inadequacy.

To avoid adverse health effects due to excessive intake, the tolerable upper intake level (UL) was determined. No ULs were set for the nutrients for which there was insufficient scientific evidence.

The DRIs for several nutrients need to be established, for the prevention of LRDs. However, the research focusing on those nutrients is not sufficient, both quantitatively and qualitatively<sup>(1)</sup>. Therefore, tentative dietary goals (DGs) for the prevention of LRDs, were determined as “the nutrient intake amount Japanese people should aim for, to prevent LRDs, in the foreseeable future.”



**Figure 2. Purposes and types of nutrition indices**

#### 1-4. Classification of Age

Age was classified based on the *Dietary Reference Intakes for Japanese (2010)*. Infants were divided into two groups: 0–5 months and 6–11 months. When the inclusion of more detailed age group settings was deemed necessary, particularly in accordance with growth, three age groups were set: 0–5 months, 6–8 months, and 9–11 months.

Individuals aged 1–17 years were considered children, and those over 18 years of age were considered adults. When it was deemed necessary for elderly individuals to be differentiated from other adults, those above the age of 70 years were termed “elderly”. Cases in which the setting of more detailed age groups was necessary, for elderly adults, were also examined.

## **2. Basics of Development**

### **2-1. Overview of the Indices**

#### **2-1-1. Energy**

BMI was adopted as an index to indicate the state of maintenance of the balance between energy intake and consumption (energy balance). The target BMI range was, therefore, presented after the comprehensive verification of the BMI range, with the lowest all-cause mortality reported in epidemiological observational studies in adults, and the current BMI status of Japanese people. However, BMI should be treated merely as a factor associated with the maintenance and promotion of health, prevention of LRDs, and avoidance of physical weakness due to old age.

Since there are several non-negligible interindividual differences that influence energy requirement, it is difficult to present energy requirement as a single value, according to sex, age group, and level of physical activity. However, the concept of energy requirement is important, and the target BMI is limited to adults. Moreover, an approximated energy requirement would be necessary to calculate the EAR of the nutrients that are known to depend on energy requirement. Therefore, the estimated energy requirement (EER) was set as a reference value, describing energy requirements and estimation methods.

#### **2-1-2. Nutrients**

##### **• Estimated Average Requirement (EAR)**

The EAR is the average daily nutrient intake level required in a population (e.g., 30–49-year-old men), calculated on the basis of the distribution of the measured requirements in a study population. In other words, the EAR is effectively defined as the estimated intake amount that meets the requirements of 50% of the individuals belonging to an age or sex group; in the remaining 50% of the population, the intake requirement is not met.

The EAR is the primary reference point for the avoidance of inadequacy, but “inadequacy” here does not just mean deficiency in the conventional sense; its definition differs depending on the nutrients.

##### **• Recommended Dietary Allowance (RDA)**

The RDA is an estimate of the daily average dietary intake that satisfies the needs of most of the individuals belonging to a population (97–98%), on the basis of the distribution of the measured requirements of a study population. The RDA is set for nutrients for which the EAR is available, and is calculated using the EAR.

The RDA is theoretically calculated as “the EAR + 2 × standard deviations (SDs)”, using the SD of the interindividual variability in the requirements, observed in the experiments, as the estimated SD of the interindividual variability in the requirements of a population. However, since it is difficult to obtain an accurate SD of the EAR from experiments, often, estimates have to be used.

The RDA is, therefore, obtained by the following equation:

$$\text{RDA} = \text{EAR} \times (1 + 2 \times \text{coefficient of variation}) = \text{EAR} \times \text{RDA calculating coefficient}$$

### ● Adequate Intake (AI)

If sufficient or adequate scientific evidence is not available to establish the EAR and, thus, the RDA, the AI is set for the nutrients. The AI is the intake level recommended for the sufficient maintenance of nutritional status in a specific population. In reality, AI is described as the presence of an adequate nutritional state in most individuals of a specific population. It is obtained from epidemiological studies that observe nutrient intake in a large number of healthy individuals.

AI is set based on any of the three following concepts:

- (1) When the AI is based on the biomarkers for health status and nutrient intake, in a specific population, the intake amount associated with the least deficiency is used. In this case, the median of the nutrient intakes is used.
- (2) When health status cannot be confirmed using biomarkers, but the distribution of the typical nutrient intake of a population, composed primarily of healthy Japanese people can be obtained, the median nutrient intake is used.
- (3) When the AI is based on the intake of healthy infants raised on breast milk, the nutrient concentration of breast milk and the suckled milk volume are used for the calculation.

The concept utilized for the determination of the AI depends on the nutrient, sex, and age group.

### ● Tolerable Upper Intake Level (UL)

The UL is the highest average daily nutrient intake level that is not likely to pose any risk of adverse health effects. As the intake exceeds the UL, the potential risk of adverse health effects may increase.

The UL theoretically exists between the maximum “average daily intake known not to cause adverse health effects” (no observed adverse effect level: NOAEL) and the minimum “average daily intake known to cause adverse health effects” (lowest observed adverse effect level: LOAEL). However, there are few reports on this subject, and those that examine specific populations are limited. Furthermore, the NOAEL and LOAEL have to be obtained on the basis of the results of experiments conducted under artificially constructed conditions, such as animal and *in vitro* experiments, in some cases. The UL was, therefore, set as the NOAEL or LOAEL, divided by uncertainty factors (UF), to ensure the safety in dietary intake considering the uncertainty of the obtained value. More specifically, the UL was calculated as follows:

- When the UL was calculated using data of the consumption of regular food in humans:  
$$\text{UL} = \text{NOAEL} \div \text{UF}$$
 (the appropriate value in the range of 1–5 was used as the UF)

- When the UL was calculated using data of the consumption of supplements, in humans, or of animal and *in vitro* experiments:

$$UL = LOAEL \div UF \quad (10 \text{ is used as the UF})$$

### • Tentative Dietary Goals for Preventing Lifestyle-Related Diseases (DG)

The DGs required for the prevention of LRDs were set as the current goals for Japanese individuals to reach the average daily intake of nutrients, and, thereby, prevent LRDs. DGs were calculated as the average daily intake required to achieve a nutritional status in a specific population in which the value of the biomarkers (proxy indicators), and risk of disease are considered low. Further, they were developed taking into account the findings of experimental nutritional studies, including findings primarily obtained from epidemiological studies. However, the association between nutrient intake and the risk of LRDs is continual, and there is often no threshold. In such cases, it is difficult to propose a value or range for the desired nutrient intake. DGs were, therefore, set with an emphasis on feasibility, taking into account the DRIs and disease prevention guidelines of other countries as well as current Japanese nutrient intakes, food compositions, preferences, etc.

The following three calculation methods were used when considering the characteristics of each nutrient:

- When the current nutrient intake of Japanese people was lower than the desired intake, only the values below the range were calculated. This applied to dietary fiber and potassium. Considering feasibility, the median of the desired nutrient intake and current nutrient intake was used for these values. The same extrapolation method as that used for AI (using reference body weight) was used for children. However, in the event that the nutrient intake calculated with this method was greater than the median of the current nutrient intake, the latter was set as the DG.
- When the current nutrient intake of Japanese people was greater than the desired nutrient intake, only the values above the range were calculated. This applied to saturated fatty acids and sodium (salt equivalent). These values were calculated taking into account recent trends in nutrient intake and feasibility in achieving the desired intake level. Sodium (salt equivalent) intake, in children, was extrapolated using EERs, and calculated taking into account the feasibility.
- As a composite indicator of the prevention of LRDs, in the present DRIs, for the energy-providing nutrients' balance (the proportion of proteins, lipids, and carbohydrates [including alcohol]), the total energy intake percentage was calculated.

## 2-2. Review Methods

The present DRIs were developed on the basis of scientific evidence, wherever possible. Scientific papers, both from Japan and other countries, and other available academic material, were systematically reviewed.

In a basic review of the issues pertaining to energy and nutrients, emphasis was placed on the problems associated with the development of the *Dietary Reference Intakes (2010)*. At the same time, the target characteristics of the elderly individuals and infants were intensively reviewed. The Patient-Intervention-Comparison-Outcome (PICO) format was used to review the relationship between energy/nutrients, and the prevention of LRD onset and progression, in order to formulate research questions on hypertension, dyslipidemia, hyperglycemia, and chronic kidney diseases. In addition to these diseases, a limited review was conducted on other diseases, if the quantitative relationship with other nutrient intakes was elucidated by a number of studies, and the disease was considered important for Japanese people. On this occasion, the review paid attention to the health status and severity classification of the research participants. These reviews were primarily conducted under the “Study on Nutritional Assessments of Metabolic Disorders that Contribute to Dietary Reference Intakes for Japanese,” funded by a 2013 Ministry of Health, Labour and Welfare Grant-in-Aid for Scientific Research (Comprehensive Project to Prevent Lifestyle-related Diseases such as Cardiovascular Disease and Diabetes). This review method will need to be standardized in the future.

Moreover, the papers and materials used in the previous development of DRIs were also reviewed as needed. However, unlike in other medical fields, the methods of determining and proving the evidence level have not been sufficiently established in the fields of Human Nutrition, Public Nutrition, and Preventive Nutrition. Furthermore, variations can occur in the obtained evidence level between nutrients.

Considering the above circumstances, when information had been quantitatively integrated, such as in the case of meta-analyses, the DRIs preferentially referred to this quantitatively integrated information. In reality, the content of each study was thoroughly examined, and the most reliable information available at that time was used.

### **2-3. Adoption Policies for the Revisions of the DRIs**

#### **● Estimated Average Requirement**

- When sufficient scientific evidence was obtained for nutrients, the EARs of which were not available, the EAR was newly determined.
- When the physical endpoints were changed in the EAR calculations, the value for the EAR was changed in accordance with the evidence.
- The EAR value was changed, as needed, with changes in the reference body size.

#### **● Recommended Dietary Allowance**

- When the EAR was newly set or changed, the RDA was newly set or changed, accordingly.
- When the coefficient of variation was changed, the RDA was changed.

<Condition necessary to change the coefficient of variation>

When clear evidence, deemed necessary to change the coefficient of variation, can be obtained.

- **Adequate Intake**

- When the distribution of the typical nutrient intakes of Japanese people was obtained for a population, which comprised very few individuals with nutrient inadequacy, the median was set as the AI. In such cases, the use of the median of the population with the lowest intake, as obtained from several reports, was recommended.

Moreover, when developing the AI, it is necessary to pay attention to the extent of “sufficient amount,” that does not indicate nutrient inadequacy. Therefore, it was handled as follows:

(1) When the AI could be determined based on the DRIs of other countries, international guidelines, survey data, etc., the appropriate value was selected regardless of the median.

(2) When the extent of “sufficient amount” was difficult to determine, it was permissible to select the median of the obtained data after describing the sufficient amount.

- **Tolerable Upper Intake Level**

- When sufficient scientific evidence was available, the UL was newly set.
- When the need to review the incidence of adverse health effects arose, as a result of new knowledge, the UL was changed.
- When new knowledge requiring a change of the UL was obtained in the process of determining the UFs, the UFs were changed.

- **Tentative Dietary Goals for Preventing Lifestyle-related Diseases**

- The presence of sufficient scientific evidence for the setting of values, combined with the higher priority given to the relationship between dietary intake and LRDs in the current Japanese population, required the DGs to be newly set.
- When the values derived from sufficient scientific evidence deviated greatly from the actual dietary intakes of citizens, the DGs were set with the current dietary intake as the target amount.

## 2-4. Age Classification

The age classification was similar to that used in the previous DRIs (Table 1.). Infants were divided into two groups: 0–5 months, and 6–11 months. Energy and proteins, which were thought to require a more detailed categorization of age groups, particularly in accordance with growth, were presented in three age groups: 0–5 months, 6–8 months, and 9–11 months.

From ages 1–17 years, individuals were considered children, and from the age of 18 years, adults. When there was a need for elderly individuals to be differentiated from other adults, those aged 70 years or older were described as “elderly”. Furthermore, for those aged 70 years or older, attention was paid to the age range in the literature that served as evidence for the development of the DRIs, and the age range was specified as needed. In light of the increase in the proportion of the elderly population, in Japan, a detailed age classification may need to be formulated for the elderly; however, this should be the topic of future study, as enough evidence has not been obtained at this time.

**Table 1. Age Classification**

Age
0-5 (months)*
6-11 (months)*
1-2 (years)
3-5 (years)
6-7 (years)
8-9 (years)
10-11 (years)
12-14 (years)
15-17 (years)
18-29 (years)
30-49 (years)
50-69 (years)
70 years or older

\* For energy and protein, these age categories were classified into 0-5, 6-8, and 9-11 months old.

## 2-5. Reference Body Size (Reference Height and Reference Weight)

### 2-5-1. Purpose

The body size (height and body weight) referenced in the development of the present DRIs was assumed to be the average Japanese body size, according to sex and age. This was referred to as the reference body size (reference height and body weight; Table 2). Previously, this value was referred to as the “standard body size”; however, the expression was revised to “reference body size” as it refers to the average Japanese body size, but not the desired body size.

**Table 2. Reference Body Size (Reference Height and Reference Weight)**

Gender	Male		Female	
Age (years)	Reference Height (cm)	Reference Weight (kg)	Reference Height (cm)	Reference Weight (kg)
0-5(months)	61.5	6.3	60.1	5.9
6-11(months)	71.6	8.8	70.2	8.1
6-8(months)	69.8	8.4	68.3	7.8
9-11(months)	73.2	9.1	71.9	8.4
1-2 (years)	85.8	11.5	84.6	11.0
3-5 (years)	103.6	16.5	103.2	16.1
6-7 (years)	119.5	22.2	118.3	21.9
8-9 (years)	130.4	28.0	130.4	27.4
10-11 (years)	142.0	35.6	144.0	36.3
12-14 (years)	160.5	49.0	155.1	47.5
15-17 (years)	170.1	59.7	157.7	51.9
18-29 (years)	170.3	63.2	158.0	50.0
30-49 (years)	170.7	68.5	158.0	53.1
50-69 (years)	166.6	65.3	153.5	53.0
70 years or older	160.8	60.0	148.0	49.5

### 2-5-2. Basic Concept

The standard height and body weight values, used in the assessment of children's body sizes, by the joint committee on growth reference value, The Japanese Society for Pediatric Endocrinology and the Japanese Association for Human Auxology, were used as the reference body sizes of infants and children<sup>(2)</sup>.

Meanwhile, the ideal standard body size in adults, according to sex and age group, is yet to be revealed. Therefore, based on the policies of the Dietary Reference Intakes for Japanese (2005 and 2010), the most recent data available were used as the current values for the calculation of a representative value for each sex and age group.

Currently, the prevalence of overweight for Japanese men is approximately 30%, while that of underweight for Japanese women aged 20–30 years is approximately 20%. Furthermore, there are problems associated with height and weight measurements in elderly individuals. The ideal body size needs to be verified on the basis of these facts in the future.

### 2-5-3. Calculations Methods

#### ● Infants and Children

The median values of the relevant age groups, in months and years, were cited on the basis of the standard values for height and weight used in the assessment of children's body

sizes by the joint committee on growth reference value, The Japanese Society for Pediatric Endocrinology and the Japanese Association for Human Auxology<sup>(2)</sup>.

● **Adults (18 Years and Older)**

The median for the height and weight of each relevant sex and age group in the 2010 and 2011 National Health and Nutrition Survey was used. Pregnant and lactating women were excluded from the calculation. The following statistics, showing the distribution, were used as reference material (Supplemental Tables 1 and 2).

**Supplemental Table 1. The distribution of body height (25, 50, 75 percentile)**

Age (years)		Percentile		
		25	50	75
Male	18-29	167.0	170.3	175.0
	30-49	167.0	170.7	175.0
	50-69	162.7	166.6	170.5
	70 years or older	157.2	160.8	165.2
Female	18-29	154.4	158.0	161.5
	30-49	154.5	158.0	161.3
	50-69	150.0	153.5	157.0
	70 years or older	143.3	148.0	152.0

**Supplemental Table 1. The distribution of body weight (25, 50, 75 percentile)**

Age (years)		Percentile		
		25	50	75
Male	18-29	57.0	63.2	70.8
	30-49	62.0	68.5	76.2
	50-69	60.0	65.3	72.2
	70 years or older	53.9	60.0	66.2
Female	18-29	46.1	50.0	55.0
	30-49	48.0	53.1	59.3
	50-69	48.0	53.0	58.6
	70 years or older	43.8	49.5	55.1

### **3. Considerations for Development**

#### **3-1. Intake Sources**

The energy and nutrients in consumed foods were examined. Intake from the diet was used for calculation; however, apart from regular food, the energy and nutrients contained in foods consumed for health promotion and not intended for the treatment of disease were also examined. These include so-called health drinks, nutritional supplements, foods with fortified nutrients (fortified food), foods for specified health uses, foods with nutrient function claims, and so-called health foods and supplements. However, the UL of folic acid was set for intake only from sources other than regular food.

#### **3-2. Intake Period**

The DRIs provide references for habitual intake, and are expressed in units *per day*; they do not present references for short-term diet (e.g., for 1 day). This is because nutrient intake varies greatly from day to day<sup>(3-6)</sup>, and the adverse health effects discussed in the DRIs occur as a result of the habitual excess or inadequate intake of energy and nutrients.

The time required for the adverse health effects associated with the inadequate or excess intake of nutrients to manifest varies greatly, depending on the type of nutrient and adverse health effect. For example, consuming a diet almost completely devoid of vitamin B<sub>1</sub> can result in a large decrease in serum vitamin B<sub>1</sub> levels after 2 weeks, with various vitamin B<sub>1</sub> deficiency symptoms manifesting within 4 weeks<sup>(7)</sup>, indicating the need for nutritional management within 1 month. In contrast, one report noted a relationship between the excessive intake of sodium (salt) and age-related increases in blood pressure<sup>(8)</sup>, suggesting the importance of nutritional management over several decades. The time required for adverse health effects to develop or improve varies greatly depending on the type of nutrient and adverse health effect.

It is difficult to specifically show a certain habitual intake period, from the viewpoint of the intake characteristics of energy and nutrients, i.e., day-to-day variations. According to results of studies that very broadly observed the day-to-day variations in energy and nutrient intake<sup>6-8</sup>, the time required to understand or manage habitual intake is approximately “1 month,” excluding some nutrients with very large day-to-day variations, allowing for some degree of measurement error and interindividual differences.

#### **3-3. Intake Frequency and Ratio, and Speed of Eating**

The frequency of consuming meals, throughout the day, particularly pertaining to whether or not breakfast is consumed, has been reported to contribute to the incidence of diseases such as obesity and cardiovascular disease<sup>(9)</sup>. Moreover, differences in the intake ratios of energy and nutrients between meals throughout the day have been reported to have an effect on the development of metabolic syndrome<sup>(10)</sup>. Some studies have also reported the association between the differences in time zones and nutrient intake<sup>(11)</sup>. These reports suggest the likely involvement of energy, nutrient intake, and metabolism in human biological circadian rhythms,

as well as an association of deviations between circadian and daily life rhythms with energy and nutrient metabolism<sup>(12)</sup>. Furthermore, some reports claim that intake speed is involved in the development of obesity, metabolic syndrome, and diabetes<sup>(13)(14)(15)(16)(17)</sup>. These reports focus more on the physiological effects of intake timing and speed on the body rather than habitual energy and nutrient intake. However, dietary intake in daily life is also influenced by external factors, in addition to biological circadian rhythms, suggesting the need for further basic research and epidemiological studies.

### 3-4. Handling Research Data

#### • Data on the Nutrient Intake Status of Japanese People

Exemplary scientific papers describing the nutrient intake status of Japanese people were cited, and when there was a lack of appropriate data, values based on data from the most recent National Health and Nutrition Survey were cited.

Importantly, it has been revealed that there are underreported findings in most dietary survey methods, including dietary records. However, it remains unclear to what extent the National Health and Nutrition Survey underestimates nutrient intake. This issue needs to be verified in the future.

#### • Methods of Integrating Research Results

To integrate research results, the policy presented in Table 3 was determined.

**Table 3. Basic policy to integrate research results**

Quality of the studies	Are there any studies examining Japanese people?	To integrate the studies' results:
Relatively homogeneous	Yes	Use the results of the Japanese studies preferentially
	No	Use the average value of the total results of the studies
Largely different between studies	Yes; the study's quality is high	Use the results of the Japanese studies preferentially
	Yes; but the study's quality is relatively low	Chose the studies with high-quality and use the average value of these studies' results
	No	

#### • Handling Interventional Studies Using Supplements and Other Food Sources

A few nutrients are expected to prevent the occurrence of some LRDs if consumed in quantities that markedly exceed the amount that can be consumed from regular foods. In order to verify those effects, interventional studies using supplements are sometimes conducted. However, it was reported that undesirable health effects might arise after a certain favorable effect<sup>(18)</sup>. A prudent stance should, therefore, be taken in validating the consumption of large quantities of specific nutrients from sources other than regular food (such as supplements).

Therefore, the present DRIs did not include data from studies using quantities of specific nutrients that were deemed clearly impossible to consume in combination with regular food (excluding supplements). However, these studies were also reviewed for use as reference material, in the determination of the DRIs.

### 3-5. Extrapolation Methods

#### ● Basic Concept

The values employed in calculating the five types of reference values (EAR, RDA, AI, UL, and DG) used in the DRIs were observed in individuals limited to a certain sex and age. Values, therefore, need to be extrapolated from these reference values to determine the DRIs, according to sex and age group.

The reference values for EAR and AI are often obtained from daily intakes (weight/day), whereas the reference values for UL are often obtained from intake per kg of body weight (weight/kg, body weight/day). Therefore, extrapolation methods were determined for each type of values.

For the RDA, the EAR was extrapolated from the reference EAR, according to sex and age group, and then each extrapolated EAR was multiplied by the RDA calculation coefficient. For DG, the AI was first extrapolated from the AI reference values, according to sex and age group, and then each extrapolated AI and the median nutrient intake of each sex and age group was used to set the DG for each sex and age group.

#### ● Estimate Average Requirement and Adequate Intake

It is difficult to determine extrapolation methods that take into account the characteristics of nutrients. Therefore, focusing on the strong relationship between energy metabolic efficiency and body surface area, the body surface area estimated from height and/or body weight is widely used in extrapolation<sup>(19)</sup>. Several formulas, for the estimation of body surface area from height and/or body weight, have been proposed. In the present DRIs, a method using a body weight ratio to the 0.75 power, proposed in 1947, was adopted<sup>(20)</sup>. This method has recently been examined in greater detail, and is reportedly useful in estimating the organ weight of various organisms, including mammalian cardiovascular and respiratory organs<sup>(21)</sup>.

The following approaches were used for adults and children:

When the reference value for the EAR or AI was given as the amount of intake per day (weight/day), and the representing value of body weight of the target population is clear in the study from which the reference value was obtained, the reference value was extrapolated as follows:

$$X: X0 \times (W/W0)^{0.75} \times (1 + G)$$

X: The sought EAR or AI of the age group (intake per day)

X0: The EAR or AI reference value (intake per day)

W: The sought reference body weight of the age group

W0: Typical body weight of the participants in the study from which the reference value for the EAR or AI was obtained (mean or median)

G: Growth factor (value determined based on Table 4)

Depending on the studies, the EAR or AI reference value may sometimes be given per kg of body weight. In such instances, the reference value was extrapolated as follows:

$$X = X0 \times W \times (1 + G)$$

X: The sought EAR or AI of the age group (intake per day)

X0: The EAR or AI reference value (intake per kg of body weight)

W: The sought reference body weight of the age group

G: Growth factor (value determined based on Table 4)

In the case of children, it is necessary to take into account the amount utilized for growth, and the amount accumulated in the body, in association with growth. Thus, the values adopted by the FAO/WHO/UNU<sup>(22)</sup> and the *US–Canada Dietary Reference Intakes*<sup>(19)</sup>, as growth factors, were modified to suit Japanese age groups and used in the present DRIs (Table 4).

**Table 4. Growth factors used to estimate EAR or AI**

Age	Growth Factor
6-11 (months)	0.30
1-2 (years)	0.30
3-14 (years)	0.15
15-17 (years) (male)	0.15
15-17 (years) (female)	0
18 years or older	0

In 6–11-month-old infants, there are two ways in which values can be extrapolated: 1) extrapolating from the values of 0–5-month-old infants; and 2) adopting the median of 0–5-month-old infants and the median of 1–2-year-old infants.

When extrapolating values from the DRIs of 0–5-month-old infants, the following formula has been proposed<sup>(19)</sup>.

**(Body weight of the reference body size of 6–11-month-old infants ÷ body weight of the reference body size of 0–5-month-old infants)<sup>0.75</sup>**

However, this formula does not take into account growth factors, because 0–5-month-old infants are still undergoing growth, and components attributable to growth factors are included in their DRIs. If the reference body weight is substituted, the formulas for men and women are  $(8.8 / 6.4)^{0.75}$  and  $(8.2 / 5.9)^{0.75}$ , respectively, producing respective reference values of 1.27 and 1.28. This formula provides an extrapolated value that varies slightly between men and women, so the mean of the extrapolated values for men and women is used as the AI for both men and women.

Some nutrients are extrapolated using other methods, taking into account nutrient characteristics and available data, such as the following:

- Calculations based on nutrient intake from breast milk and intake from sources other than breast milk

The following formula was used:

**Nutrient concentration of breast milk × average milk intake + intake from sources other than breast milk**

- Calculations from values extrapolated from the DRIs for 0–5-month-old infants and 18–29-

year-old adults

This is how the mean of the values extrapolated using these two methods was set as the AI, and this method was used for water-soluble vitamins. Specifically, the reference values for the AI of 0–6-month-old infants was calculated independently from those of the AI of 0–5-month-old infants and the EAR (or AI) of 18–29-year-old adults. The values obtained for men and women were then averaged, and the same value was set for both. This value was rounded to obtain a common AI for men and women. Extrapolation was performed using the following method:

- Extrapolation from the AI of 0–5-month-old infants

**(AI of 0–5-month-old infants) × (reference body weight of 6–11-month-old infants ÷ reference body weight of 0–5-month-old infants)<sup>0.75</sup>**

- Extrapolation from the EAR (or AI) of 18–29-year-old adults

**(EAR [or AI] of 18–29-year-old adults) × (reference body weight of 6–11-month-old infants ÷ reference body weight of 18–29-year-old adults)<sup>0.75</sup> × (1 + growth factor)**

A value of 0.30 was used for growth factors, based on the values adopted by the FAO/WHO/UNU and the *US–Canada Dietary Reference Intakes* (Table 4).

#### • Tolerable Upper Intake Level

As with the EAR and AI, no theoretical and sufficiently reliable extrapolation method exists for UL. Reference values were, therefore, calculated for age groups with insufficient evidence, using one of the following methods:

When the reference value for UL was calculated, per kg of body weight, the following formula was used:

$$X = X0 \times W$$

X: The sought UL of the age group (intake per day)

X0: UL reference value (intake per kg of body weight)

W: The sought body weight of the reference body size of the age group

When the reference value for the UL was calculated, per day, the following formula was used.

$$X = X0 \times (W/W0)$$

X: The sought UL of the age group (intake per day)

X0: UL reference value (intake per kg of body weight)

W: The sought body weight of the reference body size of the age group

W0: Typical body weight of the participants in the study from which the reference value for the UL was obtained (mean or median)

### 3-6. Rounding Values

The EAR, RDA, AI, UL, and DG values were rounded, in accordance with the rules shown in Table 5, taking into account the reliability and convenience of the reference values. A single rule was applied to the reference value of each nutrient for both men and women, in the child, adult, and elderly individual age groups. The same display digit numbers were used in the additional amounts for infants, pregnant women, and lactating women as those used in

the values of the other sex and age groups. After rounding, the values were smoothed, as needed, to prevent large discrepancies between the age groups.

## 4. Application of the DRIs

### 4-1. Basic Concept

The application of the DRIs to the dietary modification of healthy individuals and groups, for the purpose of health maintenance and promotion and prevention of LRDs, is based on the concept of the PDCA cycle (Figure 3). First, an assessment of dietary intake status is conducted to determine if the energy and nutrient intake are sufficient. Based on this, intake is improved through the formulation of a dietary improvement plan. Subsequently, results are evaluated, including a dietary assessment. Lastly, the plan and its content are improved upon, on the basis of the results of these evaluations.

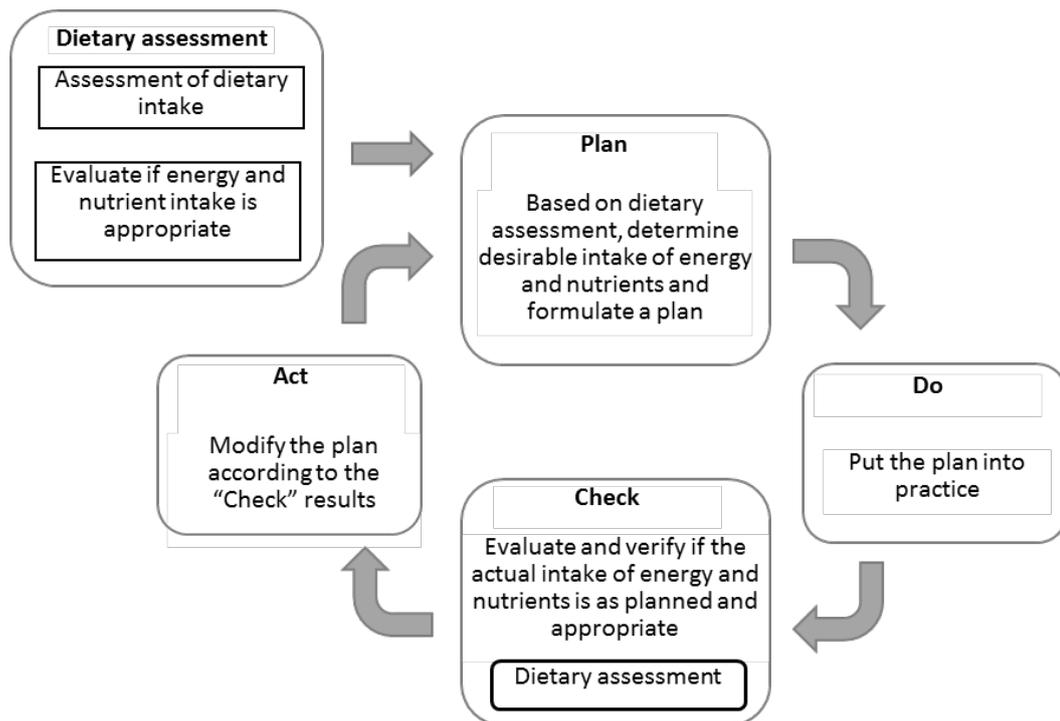


Figure 3. Application of Dietary Reference Intakes and PDCA cycle

### 4-2. Methods of Assessing Dietary Intake

#### • Applying DRIs and Assessing Dietary Intake

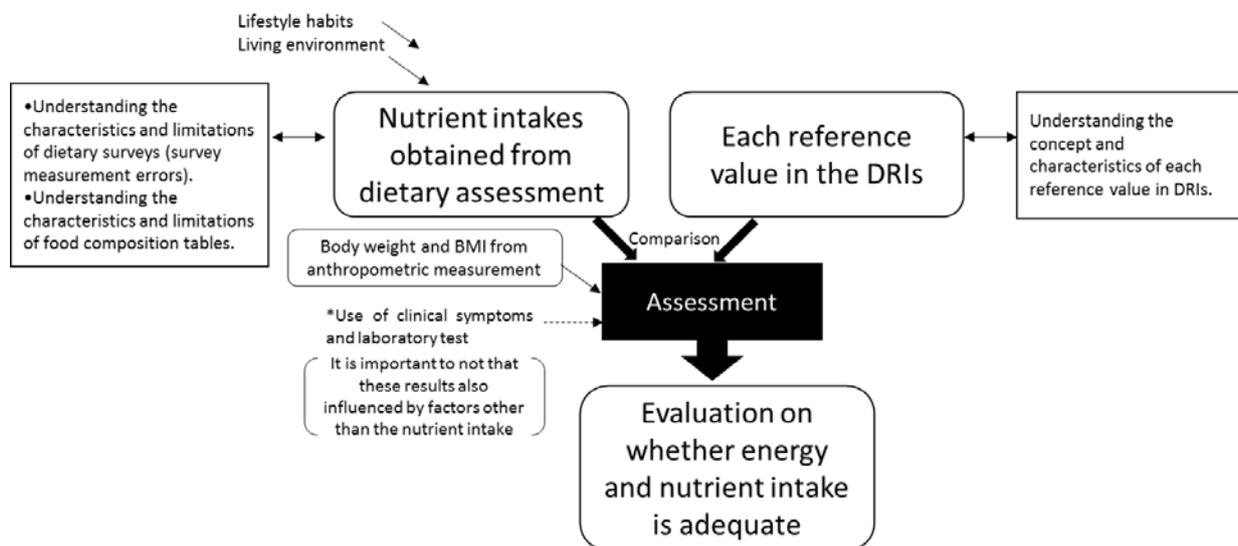
Dietary intake, i.e., intake of energy and nutrients, can be assessed by comparing each of the values in the DRIs with the results obtained from the dietary assessment. To assess excessive or inadequate energy intake, the BMI or amount of change in the body weight should be used.

The intake amounts obtained from dietary assessments are accompanied by measurement errors. To ensure a higher level of accuracy, sufficient consideration should be given to their standardization and accuracy control. The types, characteristics, and degrees of dietary assessment measurement errors need to be considered when dietary intake is assessed. Particularly, attention must be paid to measurements errors such as under- and overreporting, and day-to-day variations.

When energy and nutrient intakes are estimated from dietary assessments, nutritional

values are calculated using food composition tables. However, the nutrient quantities in food composition tables, and the nutrient quantities contained in the actual foods consumed are not necessarily the same. Nutrient calculations, therefore, need to be performed with an understanding of such errors.

Additionally, to assess whether the energy and nutrient intake amount is appropriate, it is necessary to conduct a comprehensive assessment of the target individuals, including clinical symptoms and laboratory test values, and factors such as their living environments and lifestyle habits. It is important to note that clinical symptoms and laboratory test values are also influenced by factors other than the nutrient intake status in question. Figure 4 shows an overview of the application of the DRIs and assessment of dietary intake.



**Figure 4. Applying DRIs and Assessing Dietary Intake**

### • Dietary Assessment

Dietary intake can be assessed in several ways such as the duplicate diet method, meal recording method, dietary recall method, food frequency method, dietary history method, and the use of biomarkers. Each of the methods has its own advantages and disadvantages, and characteristics. It is important to select an appropriate method based on the circumstances and the purpose of the assessment.

The DRIs present reference values for habitual intakes. Therefore, for their application, methods that can estimate habitual intakes must be selected. However, it is quite difficult to obtain accurate data on long-term average nutrient intakes at the individual level. Taking this into account, the food frequency method and diet history method may prove more efficient in the estimation of habitual intakes, for the application of the DRIs to individuals or groups. However, as these methods do not convert consumed foods directly into data, it is necessary to verify their reliability (validity and reproducibility). It is, therefore, best to use a method that

has been internationally recognized in scientific studies published on reliability.

Additionally, for some nutrients, the intake estimation accuracy is low during the dietary assessment. In such cases, estimation methods that use biomarkers such as urinary concentration should also be considered.

#### **4-3. Note for Use of the DRIs**

While the precautions associated with the use of each of the indices are described below, the method used differs depending on the purpose of the indices and the type of nutrients. Therefore, it is important to sufficiently understand the purpose of use, the definitions of the indices, and the characteristics of the nutrients.

##### **● Energy Balance**

BMI is an index of energy intake, and consumption balance maintenance (energy balance). For all practical purposes, excessive or inadequate energy intake is assessed by measuring changes in body weight. Alternatively, a comprehensive assessment, including other factors, is performed to ascertain whether there is a risk of the measured BMI falling below the target BMI range--“deficiency”--or exceeding the target range--“excess.” From the standpoint of preventing LRDs, it is recommended to address energy balance, with an emphasis on individual characteristics, based on the basic concept of body weight management and the desired BMI range (body weight), in each age group. To prevent the progression of LRDs, it is best to adjust the energy balance while assessing the rate of reduction of the body weight and improvement in the health status.

##### **● Estimated Average Requirement**

The EAR indicates a 50% probability of the presence of a deficiency. Since the EAR is assumed to be the nutrient intake level that half of the individuals of a group are estimated to be deficient in, if the intake level falls below this value or there are a number of individuals whose consumption levels fall below it, urgent action must be taken.

##### **● Recommended Dietary Allowance**

The RDA indicates almost no probability of the presence of deficiency in an individual, and at this consumption level, very few individuals in a population are assumed to be deficient. Therefore, when the consumption is close to or greater than this value, it is considered that there is almost no risk of deficiency.

##### **● Adequate Intake**

An AI value is set when sufficient scientific evidence cannot be obtained. Therefore, it is a reference value that is established when the EAR cannot be calculated, and, thus, the risk of deficiency is extremely low if an individual consumes more than the AI. Consequently, there is almost no probability of deficiency when the amount of nutrients consumed is close to the AI, and almost no probability of deficiency in individuals in a group. In addition, the AI should be theoretically higher than the RDA, considering its definition. However, the presence or risk of deficiency cannot be indicated even if an individual's intake is less than the AI.

- **Tolerable Upper Intake Level**

The occurrence risk of adverse health effects, due to excessive intake, is greater than zero in the event that an individual consumes more than the UL. However, it is very unlikely for an individual's consumption to exceed the UL, so long as he/she consumes regular food. Furthermore, it is very difficult to calculate the UL, both theoretically and experimentally, and most of the ULs are calculated on the basis of a small number of incidents. This demonstrates the lack of sufficient scientific evidence on the UL. This is why the UL is understood as “the amount that individuals should avoid approaching as much as possible” rather than “the amount that should not be exceeded.”

The UL is also a value for adverse health effects caused by excessive intake, and is not set for the maintenance and promotion of health or prevention of LRDs. This needs to be fully considered when the UL is used.

- **Tentative Dietary Goal for Preventing Lifestyle-related Diseases**

The DG is a value calculated for the prevention of LRDs. LRDs have a variety of causes, and diet is only one factor. Therefore, from the standpoint of preventing LRDs, it is not sufficient to strictly follow the DG alone.

For example, the excessive intake of sodium (salt) is a risk factor for hypertension, so a DG is calculated for sodium (salt), primarily from this standpoint. However, hypertension has been reported to be associated with the excessive intake of alcohol and inadequate intake of potassium, as well as obesity and lack of exercise<sup>(23)</sup>. The DG for sodium (salt) should be determined with a sufficient understanding of the above issues, target individuals, and populations.

Furthermore, compared to adverse health effects caused by the inadequate or excessive intake of nutrients, LRDs occur as a result of lifestyle habits (including eating habits) continued over a very long period of time. Considering these characteristics of LRDs, long-term (e.g., lifelong) management is more important than short-term, intense management.

#### **4-4. Note for Use According to the Purpose**

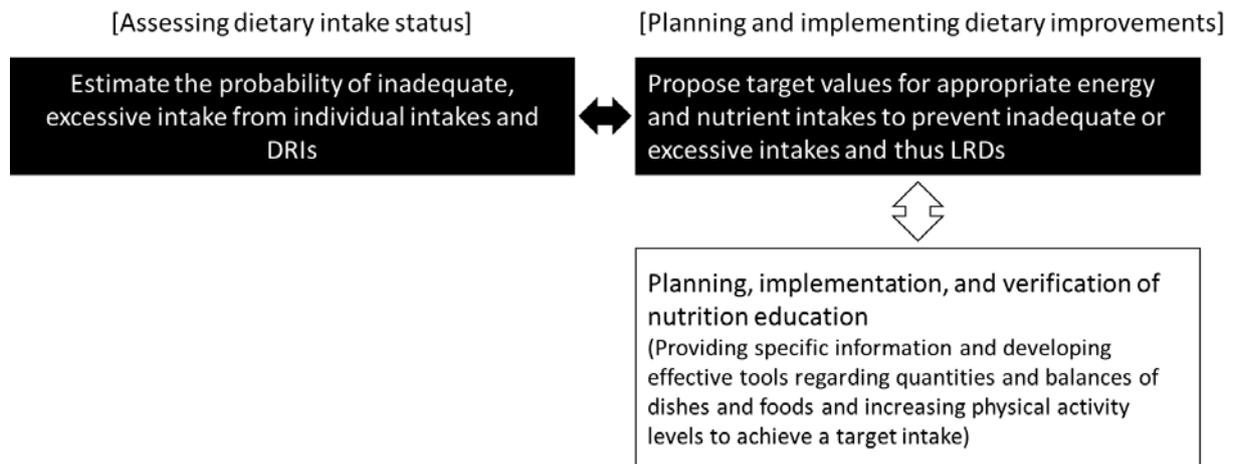
##### **4-4-1. Use in Improving the Diet of Individuals**

The basic concept behind the use of the DRIs in improving the diet of individuals is presented in Figure 5.

The likelihood of inadequate or excessive intake is estimated by assessing the dietary intake status of individuals using the DRIs. On the basis of these results, the DRIs can be used to propose target values for appropriate energy and nutrient intakes in order to prevent inadequate and excessive intakes, as well as LRDs. These assessments should lead to the planning and implementation of dietary improvements.

Furthermore, in order to achieve a target BMI and nutrient intake level, nutrition education should be planned, implemented, and verified for improving the diet of individuals, such as through the development of effective tools and the provision of specific information on

quantities and balance of dishes and food, as well as emphasizing the importance of increasing activity levels.



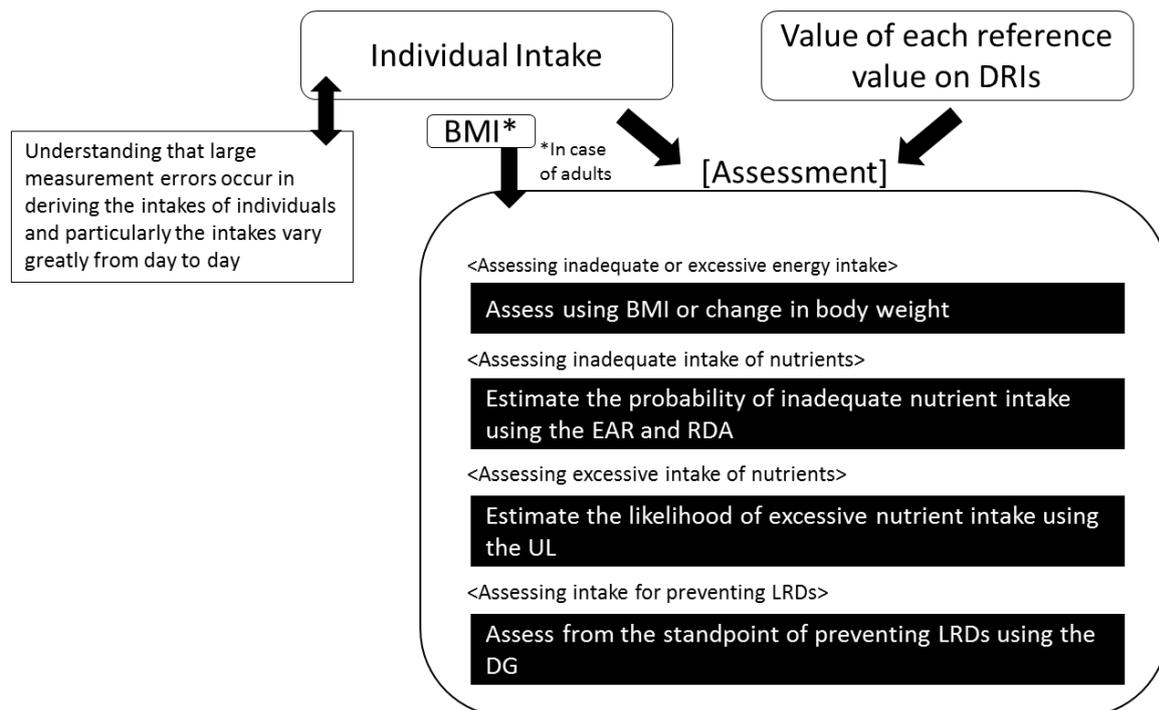
**Figure 5. Basic concept for the use of the DRIs in improving the diet of individuals**

• **Assessing Dietary Intake Status**

An overview of the dietary intake status assessment, using the DRIs, to improve diet in individuals, is presented in Figure 6.

Individual intakes, obtained from dietary assessments, are used in this assessment; however, various factors influence daily intake, such as the different foods selected every day by an individual and differences in appetite, which makes understanding the habitual intakes of individuals challenging. This is why the assessed intakes of individuals include large measurement errors. They vary greatly from day to day; thus, it is important to understand that they do not reflect an individual’s true intake.

Therefore, the assessment of dietary intake status should be performed using the DRIs, considering the above-stated limitations. Moreover, energy intake assessments are performed to evaluate whether the energy balance is positive or negative, and, for this purpose, BMI or change in body weight is used.



**Figure 6. Assessing dietary intake status, using the DRIs, to improve the diet of individuals**

BMI, or change in body weight, is used when assessing inadequate or excessive energy intake in adults. The target BMI range is presented in the present DRIs (See “Energy”). However, even if the BMI is within the target range, in the event that the body weight increases or decreases, careful and appropriate action is required, as this points to either a positive or negative energy balance.

For infants and children, a growth curve (physical growth curve) should be used when assessing inadequate or excessive energy intake. The course of growth should be observed longitudinally to ascertain whether body weight and height measurements follow the growth curve (physical growth curve), whether the body weight deviates greatly from the growth curve, without increases in the observed body weight, and whether there are any increases in the body weight that deviate greatly from the growth curve.

For the evaluation of nutrient intakes, the results of dietary assessment (estimated nutrients intakes) are to be used. This requires a full understanding of the significance and extent of the influence of measurement errors (particularly, under- and overreporting, and day-to-day variations) arising from the dietary assessment method used. It should be considered that day-to-day variations, in individuals, have a large effect on assessments.

The main application of EAR and RDA is in the prevention of the inadequacy of nutrients. When the EAR is not determined, the AI should be used instead. The probability of inadequacy can be estimated from the estimated intake, comparing the EAR and the RDA. When the intake is close to or above the RDA, it can be deemed that there is almost no risk of inadequacy. When the intake is above the EAR, but below the RDA, it is recommended to aim for the RDA. However, the intake is determined comprehensively taking into account factors

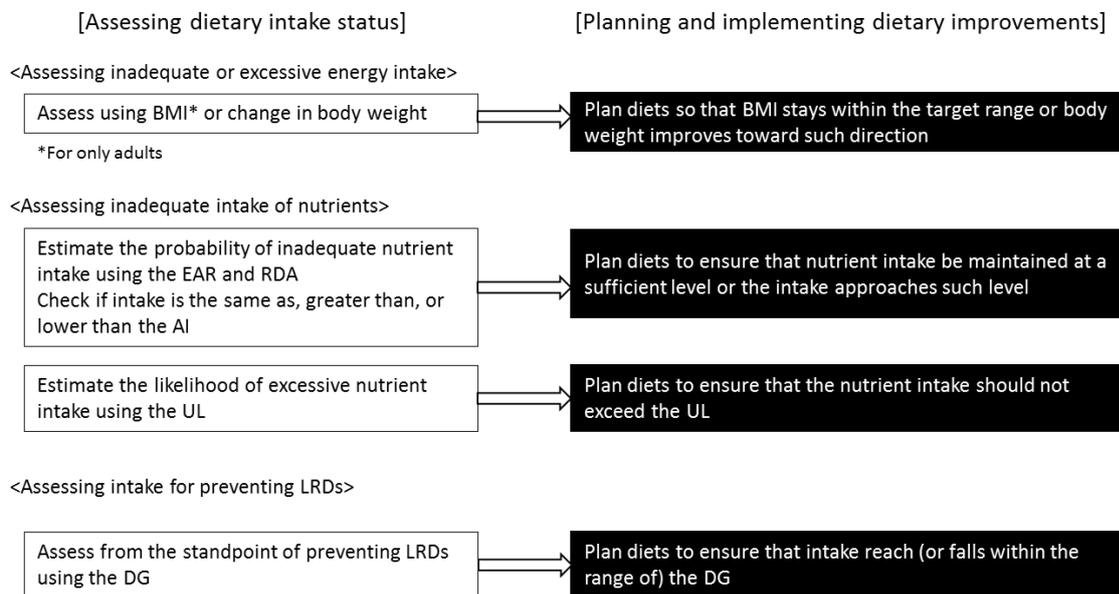
such as the intake status of other nutrients. Since the probability of inadequacy is 50% or more when the intake is less than the EAR, it is necessary to increase the intake. When using the AI, it can be deemed that there is almost no risk of inadequacy, if the intake is higher than the AI. However, the risk of inadequacy cannot be estimated even if the intake is less than the AI, as is evident from the definition of AI.

The UL is used when assessing the risk of the excessive intake of nutrients. When the estimated intake exceeds the UL, it is regarded as excessive intake.

The DG is used for evaluations related to the prevention of LRDs. Since some DGs are presented as a range, they should be compared with the estimated intake, taking into account the characteristics of each DG. Since LRDs develop as a result of a combination of factors, excessive emphasis should not be placed on one nutrient. It is recommended that a comprehensive assessment be performed, with an understanding of the importance of the nutrient in question, in relation to the LRD.

● **Planning and Implementing Dietary Improvements**

An overview of dietary improvement planning and implementation, using the DRIs, on the basis of the results of dietary intake status assessments to improve the diet of individuals, is presented in Figure 7.



**Figure 7. Planning and implementing dietary improvements, using the DRIs, to improve the diet of individuals**

When planning and implementing dietary improvements, it is fundamental to use the results of dietary intake status assessments. Dietary improvements should be planned and implemented based on these evaluations. It is, therefore, important to sufficiently understand the characteristics of individuals. Such characteristics include sex, age, physical activity levels, living environments, and lifestyle habits. Clinical symptoms and clinical test data can be also

used, depending on the purpose.

BMI, or change in body weight, should be used to plan and implement dietary improvements in order to address energy intake inadequacy or excess. Diets should also be planned such that the BMI remains within the target range. When aiming for weight loss or gain, it is recommended to record body weight measurements roughly every 4 weeks, and to perform follow-ups for more than 16 weeks. For example, in a meta-analysis of 493 interventional studies conducted for weight loss, using dietary restrictions and/or exercise, the mean BMI was 33.2 kg/m<sup>2</sup>, the mean intervention period was 16 weeks, and the mean weight loss was 11 kg<sup>(24)</sup>.

The RDA should be used for nutrients for which the corresponding value is determined. When the nutrient intake is close to or exceeds the RDA, the current intake should be maintained; when the nutrient intake falls below the RDA, the intake should be increased so as to bring it closer to the RDA. However, intake should be determined comprehensively, taking into account feasibility, and the intake status of other nutrients. The AI should be used for the nutrients for which the corresponding value is determined. The current nutrient intake should be maintained if it is close to or exceeds the AI. However, the presence and risk of inadequacy cannot be estimated when the nutrient intake falls below the AI. When the nutrient intake is considerably lower than the AI, increasing the intake should be comprehensively considered, together with the intake of energy and other nutrients, anthropometric measurements, and clinical test results.

When the nutrient intake exceeds the UL, plans should be made to ensure that the intake drops below the UL. Nutrient intake that exceeds the UL should be avoided; if the intake exceeds the UL, proper diet should be promptly planned and implemented to resolve the problem.

When the intake of nutrients exceeds the DG range, an appropriate dietary plan must be formulated. However, it is recommended to elucidate the presence and degree of other nutritional factors and non-nutritional factors related to the LRD that is to be prevented and, comprehensively taking these into account, determine the degree of improvement in the intake of the nutrient in question. Moreover, considering the characteristics of the LRD, it is best to devise and implement a feasible, long-term dietary improvement plan.

When creating the above statements, the application examples of the previous Japanese DRIs were considered, based on the approach adopted by the *US–Canada Dietary Reference Intakes*<sup>(25–27)</sup>.

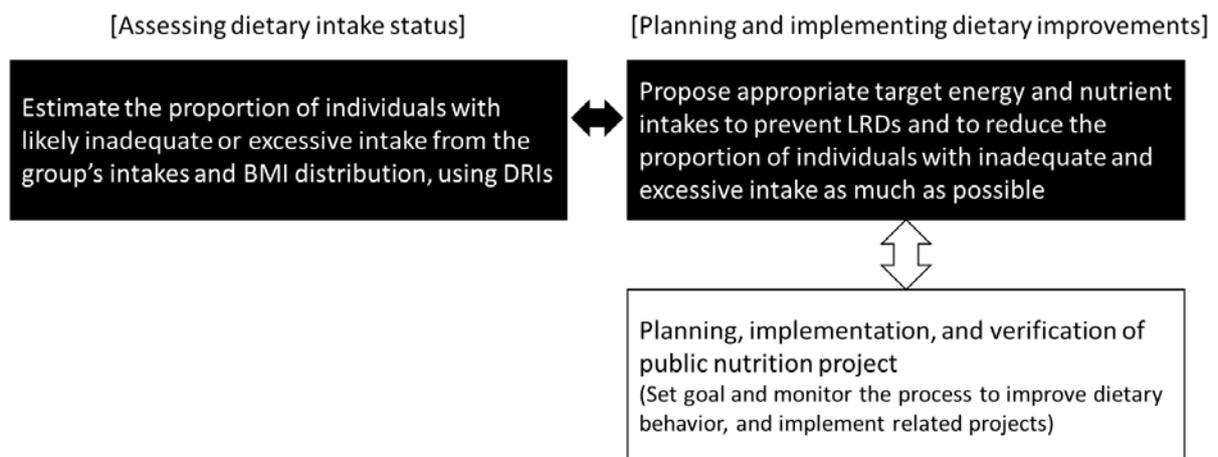
#### **4-4-2. Use of the DRIs to Improve the Diet of Groups**

The basic concept behind the use of the DRIs to improve the diet of groups is presented in Figure 8.

Dietary intake status is assessed by applying the DRIs to estimate the proportion of individuals with possible inadequate or excessive intake, from the intake distribution of a

population. On the basis of the results, the DRIs may be used to propose appropriate energy and nutrient intake targets for the prevention of inadequate or excessive intakes and LRDs, leading to the planning and implementation of dietary improvements.

Furthermore, to achieve the target BMI and nutrient intake, the planning, implementation, and verification of public nutrition projects, such as the establishment of improvement targets for eating behavior/dietary habits and physical activity levels, and their monitoring, as well as the planning and implementation of various effective projects for improvement, can be conducted.



**Figure 8. Basic concept for the use of the DRIs for improving the diet of groups**

- **Assessing Dietary Intake Status**

An overview of the assessment of dietary intake status, using the DRIs to improve the diet of populations, is presented in Figure 9.

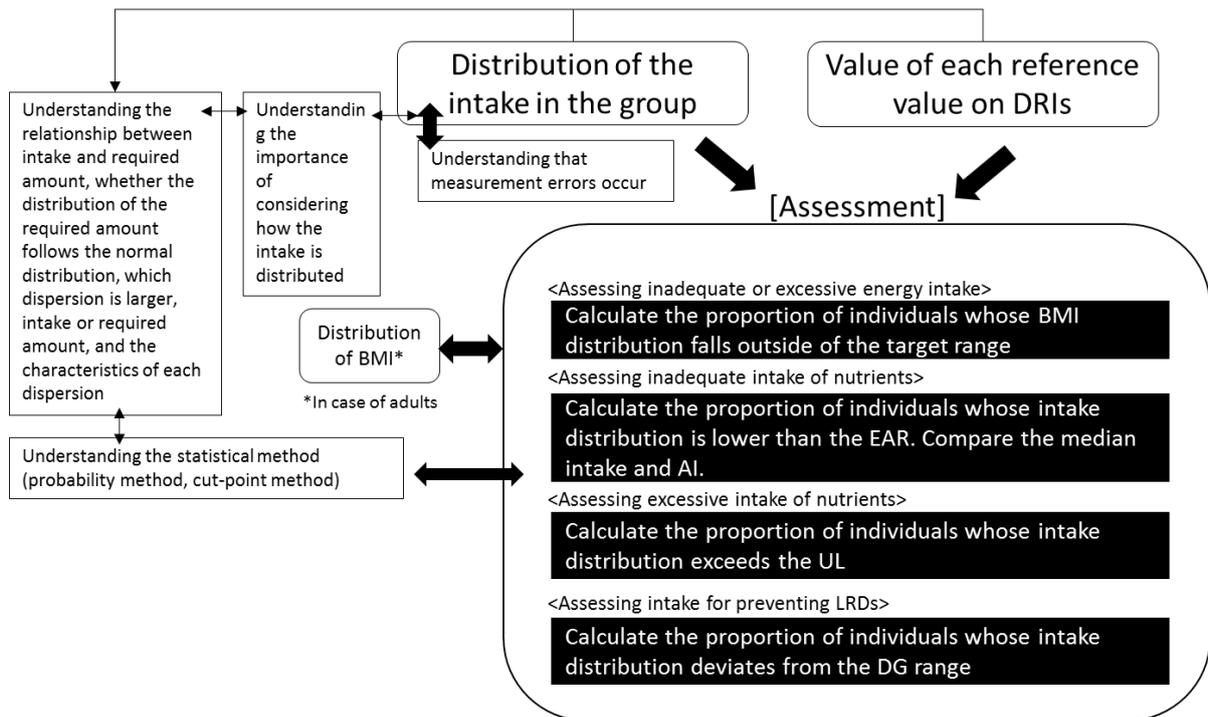


Figure 9. Assessing dietary intake status, using the DRIs, to improve the diet of groups

• Planning and Implementing Dietary Improvements

An overview of the planning and implementation of dietary improvements using the DRIs, on the basis of the results of dietary intake status assessments to improve the diet of populations is presented in Figure 10.

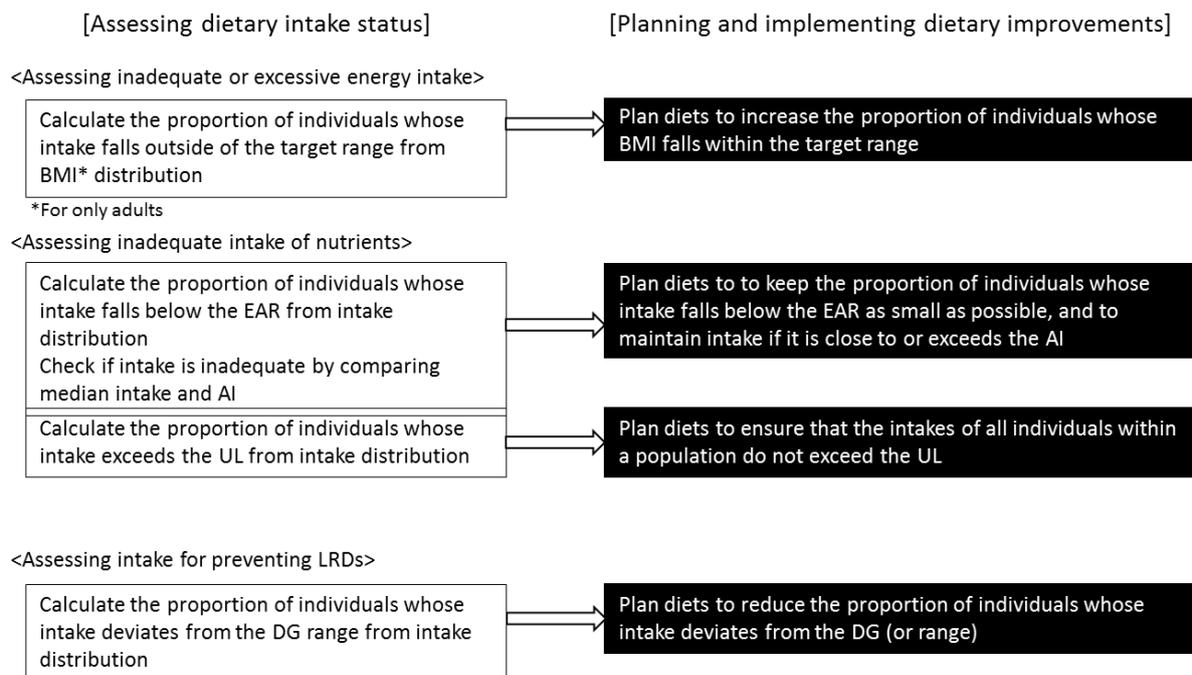


Figure 10. Planning and implementing dietary improvements using the DRIs to improve diet of groups

BMI, or change in body weight, should be used to plan and implement dietary improvements for inadequate or excessive energy intake. Diets should be planned to increase the proportion of individuals whose BMI falls within the target range. Assessments should be performed at least twice, over several months (within at least 1 year), and this evaluation should use changes in body weight.

The EAR or AI should be used to plan and implement dietary improvements for avoiding the inadequate intake of nutrients. Diets should be planned to ensure that the proportion of individuals within a population, whose intake falls below the EAR, is as small as possible. If the median intake is close to or exceeds the AI, plans should be made to maintain this intake. When the median intake falls below the AI, it is impossible to determine if the intake is inadequate. Moreover, when the intake falls considerably below the AI, the need for the improvement of intake should be examined, on the basis of comprehensive judgment, taking into account factors such as the intake of energy and other nutrients, physical measurements, and clinical test results.

The UL should be used to plan and implement dietary improvements to avoid the excessive intake of nutrients. Plans should be formulated to ensure that the nutrient intake of all individuals within a population is lower than the UL. Intakes that exceed the UL should be avoided, and if the intake is found to exceed the UL in some individuals, action should be taken promptly to resolve the problem.

DGs should be used to plan and implement dietary improvements to prevent LRDs. Diets should be planned to increase the proportion of individuals whose intake is within or close to the target range. It is recommended to elucidate the presence and degree of other nutritional factors and non-nutritional factors related to the LRD to be prevented, and taking these comprehensively into consideration, the degree of improvement in the intake of the nutrient in question should be determined. Moreover, considering the characteristics of the LRD, it is best to devise and implement a feasible, long-term dietary improvement plan.

The above statements rely on the practical application of the examples in which the previous Japanese DRIs were considered, based on the approach adopted by the *US–Canada Dietary Reference Intakes*<sup>(25,26,28)</sup>.

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**II**

**Energy and Nutrients**

## Energy

### 1. Background Information

The energy obtained by the body through external sources is used for the maintenance of vital functions, and performance of physical activity. Most of this energy is ultimately released from the body in the form of heat. Energy intake, expenditure, and accumulation in the body, therefore, are expressed in their equivalent calorific values. The unit for energy in the International System of Units is joule (J), although calorie (cal) is often used in nutrition science. Since “1 J” is an extremely small unit, it is more practical to use kJ (or MJ) and kcal. The unit “kcal” is used in the current *Dietary Reference Intakes (DRIs) 2015*. In accordance with the Joint Food and Agriculture Organization of the United Nations (FAO)/World Health Organization (WHO) Special Technical Committee Report 1, 1 kcal = 4.184 kJ.

Energy intake is the sum of each energy conversion factor (amount of energy used per g of each component) calculated for the fats, proteins, and carbohydrates contained in foods. Energy expenditure, meanwhile, is classified into three categories, namely basal metabolism, postprandial thermogenesis, and physical activity. Physical activity is further divided into exercise (intentionally performed to improve physical fitness), activities of daily living, and spontaneous activity (such as postural and muscle tone maintenance).

Energy balance is defined as the balance between energy intake and energy expenditure. In adults, changes in energy balance results in changes in body weight and body mass index (BMI). If individuals’ energy intake continues to exceed their energy expenditure (positive energy balance), their body weight increases; however, if their energy expenditure exceeds their energy intake (negative energy balance), their body weight decreases. Short-term energy imbalance, therefore, can be assessed through changes in body weight. Moreover, energy imbalance can be adjusted on a long-term basis through reciprocal changes in energy intake, energy expenditure, and body weight. For example, if excessive energy intake continues over a long period of time, the energy expenditure increases due to changes in the exercise efficiency associated with weight gain. Weight gain eventually levels off at a certain amount, and the body transitions to a new state in which the energy balance is maintained. In many adults, maintaining a relatively constant body weight and body composition over a long period of time can result in a state in which the energy balance is maintained at almost zero. Energy intake and expenditure are almost equal even in individuals with obesity or malnourishment if no changes in body weight or composition occur. It is, therefore, insufficient, from the perspective of maintaining and promoting health and preventing lifestyle-related diseases (LRDs), to simply satisfy the required intake of energy without excess or deficiency. It is important to consume the amount of energy sufficient to maintain a desirable BMI. This is why BMI has been adopted in the current DRIs as an indicator of the maintenance of energy intake and expenditure balance.

## 2. Energy Intake and Expenditure

### 2-1. Factors Involved in Energy Intake and Expenditure

Various factors, and their interactions, influence energy intake, such as the nutrient composition of meals (energy density<sup>(1,2)</sup>), the energy intake ratio from fats<sup>(3,4)</sup>, amount of proteins<sup>(5)</sup> and dietary fiber<sup>(6)</sup>, and other characteristics of food (taste, color, texture, and palatability),<sup>(7,8)</sup> as well as eating patterns (portion sizes<sup>(9)</sup>, eating speed<sup>(10)</sup>, meal time zone<sup>(11)</sup>, and number of foods<sup>(7,12)</sup>).

Food selection and dietary patterns are influenced by various external and social factors, in modern society: the convenience of obtaining food<sup>(13)</sup>, snack intake<sup>(14)</sup>, communal dining<sup>(12)</sup>, TV viewing<sup>(15)</sup>, food advertising on television<sup>(16)</sup>, food prices<sup>(17)</sup>, and internal and subjective factors such as stress<sup>(18)</sup> are also involved, in addition to the intentional control of intake by individuals.

The mechanisms regulating appetite and satiety *in vivo*<sup>(19,20)</sup> involve the transmission of satiety signals from the liver to the hypothalamus via the vagus nerve, and various appetite-related hormones associated with dietary intake derived from the gastrointestinal tract and pancreas. Various external and internal factors also ultimately control intake by transmitting signals to the hypothalamus via the cerebral cortex. Furthermore, hormones secreted from adipocytes act on the hypothalamus to adjust intake and to maintain a certain amount of body fat (lipostat theory)<sup>(21)</sup>. Factors such as lack of sleep<sup>(22)</sup>, physical activity<sup>(23,24)</sup>, sex<sup>(25)</sup>, menstrual cycle<sup>(26)</sup>, and genetics<sup>(27)</sup> also influence the amount of intake.

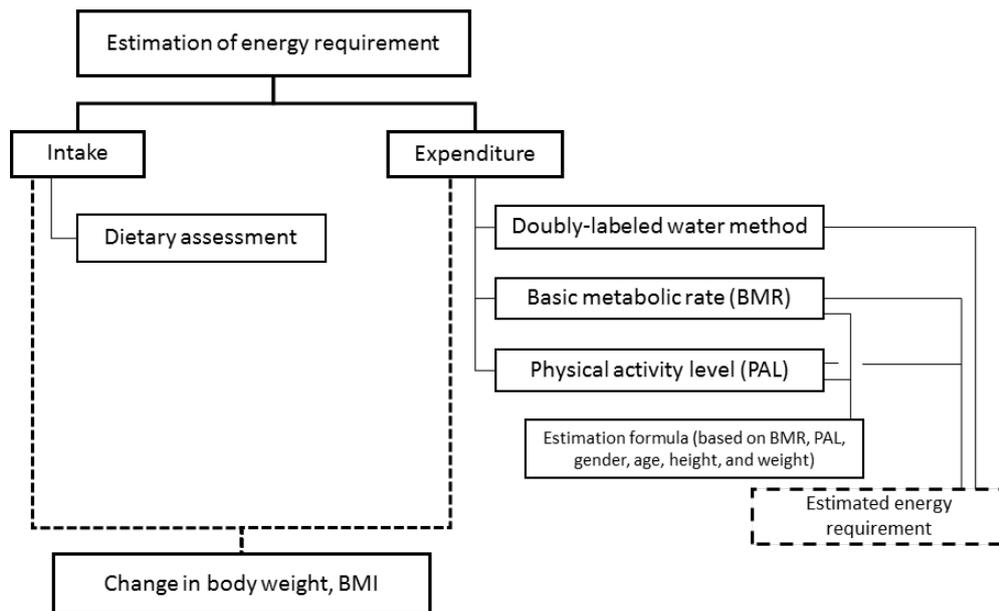
Energy expenditure is composed of parts that vary intentionally (exercise, and activities of daily living), and those that are biologically defined (basal metabolism, postprandial thermogenesis, and spontaneous activity). Energy expenditure during exercise and activities of daily living is determined based on the body weight and degree of obesity. Basal metabolism is determined by factors such as body weight, body composition, age, and sex, and is also affected by energy balance. Postprandial thermogenesis is equivalent to approximately 10% of the calorific value of energy intake, and is also affected by the nutrient composition of foods such as proteins<sup>(28)</sup>. Expenditure for both activities of daily living and spontaneous activity is referred to as non-exercise activity thermogenesis (NEAT). NEAT is affected by energy balance<sup>(29,30)</sup>, and the degree of obesity<sup>(31)</sup>.

Energy intake and energy expenditure, therefore, constitute factors that are influenced by individual biological and external factors, and those that can be controlled intentionally; in addition, these factors are interrelated. When strategically managing energy intake for the maintenance and promotion of health, and prevention of LRDs, it is best to consider facilitation of energy intake control after acquiring a full understanding of the effect of these factors.

## **2-2. Relationship between Energy Intake, Energy Expenditure, and Estimated Energy Requirement**

The methods of estimating energy requirement are broadly divided into those that estimate energy intake under constant weight conditions, and those that measure energy expenditure. Energy intake measurement includes various dietary assessments, while energy expenditure measurement involves the doubly labeled water method, and calculation methods using sex, age, height, body weight, and measured values for basal metabolism and physical activity level (PAL). The doubly labeled water method directly measures energy expenditure. Any types of dietary assessment methods can produce large measurement errors for energy intake. It is, therefore, very difficult to estimate energy requirement from energy intake estimation methods. For this reason, the method in which the energy requirement is estimated closer to the energy expenditure than the energy intake is widely used (Figure 1). The doubly labeled water method, in particular, can directly measure (somewhat habitual) energy expenditure over approximately 2 weeks, and has high measurement accuracy. Therefore, it provides useful basic data for the estimation of energy requirement<sup>(32)</sup>. Energy requirement can be estimated according to PAL, as well as sex and age group. However, individual differences in energy requirements, that cannot be estimated using these methods but cannot be ignored, exist<sup>(33)</sup>. Therefore, it is difficult to estimate energy requirement, at an individual level, even when the energy requirement is estimated taking into account PAL, using the energy expenditure obtained from the doubly labeled water method. This also includes estimation formulas using factors such as basal metabolism and PAL<sup>(34)</sup>. Moreover, different methods are used to measure energy intake and consumption, and each method has its own measurement errors. Thus, there is little sense in comparing measured energy intake, and measured energy expenditure.

The results of energy balance are expressed as BMI, and changes in body weight. Therefore, it is possible to acquire an overview of energy balance, if the BMI and changes in body weight are known. However, it should be noted that, while BMI and changes in body weight merely indicate one of the results of energy balance, they do not indicate energy requirements.



**Figure 1 Measurement methods for estimation of energy requirement and association with change in body weight, BMI, or estimated energy requirement**

### 3. Body Weight Control

#### 3-1. Basic Concept

If the level of physical activity remains unchanged, energy intake control is almost the same as that of body size. It is, therefore, best to assess and change energy intake and expenditure by calculating the results of measurements of body size. In doing so, measurements of estimated energy requirements, using an estimation formula, or measurements of energy intake and supply should not be used. To make these changes, the desired body size must be determined in advance.

As no significant changes in height occur after an individual enters adulthood, body size is primarily controlled by body weight. To control body weight in adults, taking into account differences in height, BMI is primarily used as a body size index. Under normal circumstances, fat—including subcutaneous and visceral fat—and tissues other than fat (primarily muscle) need to be considered. One of the ways to do so is measuring waist circumference. Some reports state that waist circumference or its ratio to height is more strongly correlated with the incidence of diabetes and cardiovascular disease and total mortality than BMI<sup>(35,36)</sup>. However, due to the many accumulated research outcomes, and BMI being the most basic body size index, body weight and BMI are used as the body size indices here. It is recommended to take into account waist circumference as well, when preventing the development and progression of diabetes and cardiovascular disease.

Distribution curves (growth curves) for the height and body weight of Japanese people of a relevant sex and age are used for infants and children.

A high level of physical activity is an effective means of preventing and improving obesity<sup>(37)</sup>, and a PAL of 1.7 or higher is recommended to prevent unhealthy weight gain<sup>(38)</sup>. A

high level of physical activity has also been found to be related to a decrease in total mortality, independent of body weight<sup>(39,40)</sup>. From the standpoint of preventing the development and progression of the LRDs associated with weight gain, having a low PAL (level I) is not advisable; energy needs to be balanced by increasing the level of physical activity.

### **3-2. Prevention**

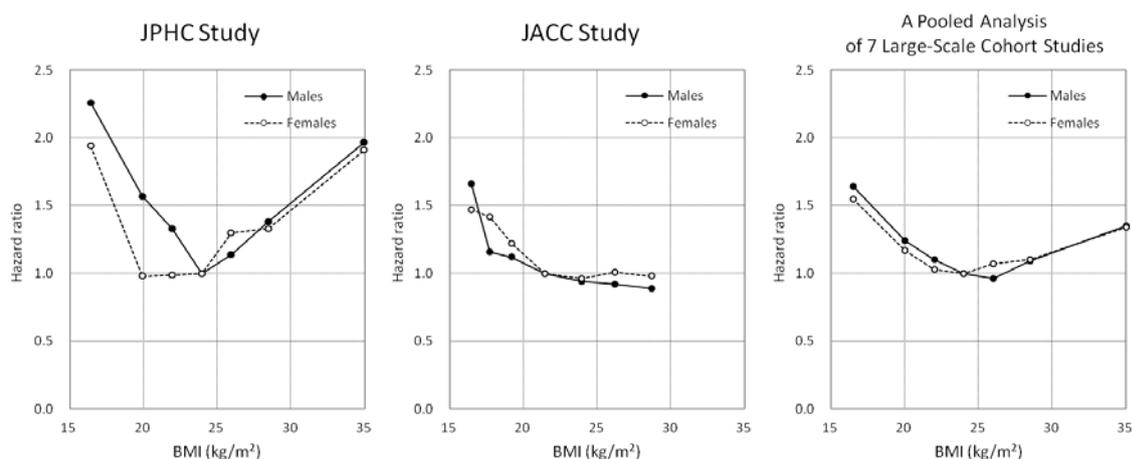
#### **3-2-1. Basic Concept**

To determine a healthy body weight using BMI, in adults, it is necessary to define what is considered “most healthy” in advance, and to examine the effect of BMI on health. A BMI state in which the all-cause mortality (total mortality) was the lowest was considered “healthy” in the current DRIs. Another possible concept is the BMI, at which individuals have the least number of diseases and adverse health effects, at a certain point in time (prevalence), being considered “healthy”. However, the mortality is not necessarily high for diseases or highly prevalent adverse health effects. Therefore, care must be taken, considering that mortality and disease prevalence are not always the same, and that BMI showing lowest rates mortality does not necessarily show corresponding decrease of the latter.

It is also inappropriate to use total mortality in the case of infants and children, and body weight control during pregnancy.

#### **3-2-2. Methods Using Total Mortality as an Indicator**

According to a meta-analysis that summarized the correlation between BMI, at the start of follow-up, and subsequent total mortality, using data from 57 cohort studies in 35–89-year-old individuals (conducted in Europe and the United States of America; total sample size: 894,576 individuals), the lowest mortality was seen in the group with a BMI of 22.5–25.0 kg/m<sup>2</sup> (in both men and women) after adjusting for age<sup>(41)</sup>. However, an analysis of non-smokers alone, conducted with the aim of eliminating the effect of increased mortality and weight loss due to smoking, found that having a slightly lower BMI resulted in the lowest mortality<sup>(42)</sup>. The results of studies in Japan and neighboring East Asian countries need to be referenced in addition to the results of studies in Europe and America. The correlation between BMI (kg/m<sup>2</sup>), at the start of follow-up, and subsequent mortality, in a pooled analysis of two representative cohort studies and seven other cohort studies, in healthy individuals in Japan, is presented in Figure 2<sup>(43–45)</sup>. Representative reports from neighboring East Asian countries are also summarized in Figure 3<sup>(46–48)</sup>.



**Figure 2. The association between BMI at baseline and mortality among normal subjects in JPHC study, JACC study, and a pooled analysis of 7 large-scale cohort studies<sup>(43-45)</sup>**

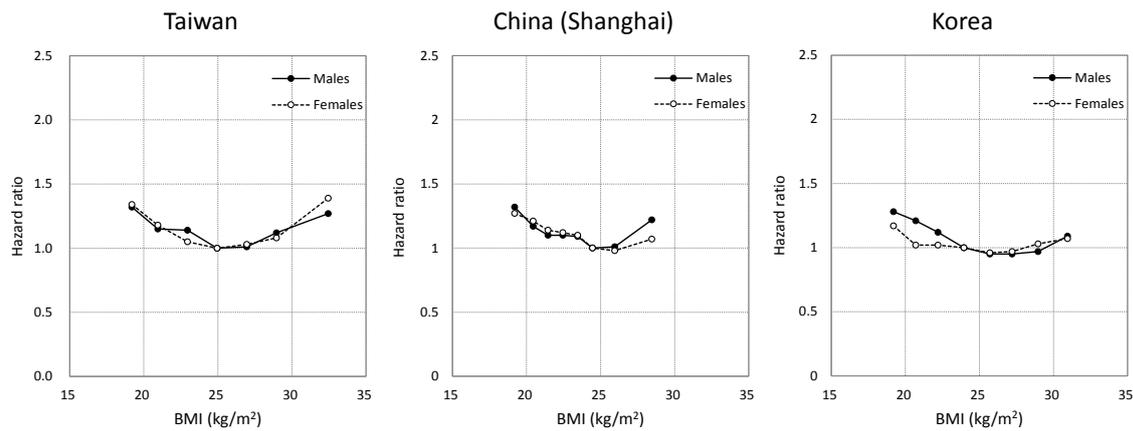
The intermediate values of the BMI ranges studied were shown as plots. The result was not shown if the maximum or minimum value of the highest or lowest BMI categories was not available.

JPHC study: The reference category was BMI=23.0-24.9kg/m<sup>2</sup>. The range of age at baseline was 40-59 years, the mean follow-up duration was 10 years, the analytic sample was 19,500 men and 21,315 women, and the number of death was 943 men and 483 women. The analysis was adjusted for residential area, age, body weight change around 20 years old, alcohol drink, leisure physical activity and educational background.

JACC study: The reference category was BMI=20.0-22.9kg/m<sup>2</sup>. The range of age at baseline was 65-79 years, the mean follow-up duration was 11.2 years, the analytic sample was 11,230 men and 15,517 women, and the number of death was 5,292 men and 3,964 women. The analysis was adjusted for smoking status, alcohol drinking, physical activity, sleep duration, stress, educational background, marital status, green-vegetable intake, history of stroke, history of myocardial infarction and history of cancer.

A pooled analysis of 7 large-scale cohort studies: The reference category was BMI=23.0-24.9kg/m<sup>2</sup>. The range of age at baseline was 40-103 years, the mean follow-up duration was 12.5 years, the analytic sample was 162,092 men and 191,330 women, and the number of death was 25,944 men and 16,036 women. The analysis was adjusted for age, smoking status, alcohol drinking, history of hypertension, leisure or physical activity, and other variables (depending on each cohort). This analysis excluded early follow-up (less than 5 years).

Of the studies presented in Figures 2 and 3, a tendency toward lower mortality with a higher BMI is only seen in the Japan Collaborative Cohort (JACC) study, which limited its analysis to a group of participants aged 65–79 years (at the start of follow-up). As this study demonstrated, the correlation between BMI and total mortality differs depending on age and BMI, with the lowest total mortality showing a tendency to increase as the age at the start of follow-up increases, in both men and women. The South Korean study presented in Figure 3 too did not reveal a clear increase in total mortality, even when the BMI exceeded 30.0 kg/m<sup>2</sup>, in a sub-analysis of individuals aged 65 years and older<sup>(48)</sup>. Moreover, according to a Japanese study that examined BMI with the lowest total mortality, according to age at the start of follow-up, the BMI values in men and women, respectively, were 23.6 kg/m<sup>2</sup> and 21.6 kg/m<sup>2</sup> for those aged 40–49 years, 23.4 kg/m<sup>2</sup> and 21.6 kg/m<sup>2</sup> for those aged 50–59 years, 25.1 kg/m<sup>2</sup> and 22.8 kg/m<sup>2</sup> for those aged 60–69 years, and 25.5 kg/m<sup>2</sup> and 24.1 kg/m<sup>2</sup> for those aged 70–79 years<sup>(49)</sup>. Furthermore, the results of a pooled analysis (results of lifetime non-smokers), summarizing data from 19 cohort studies in American Caucasian individuals (total: 1.46 million individuals),



**Figure 3. The association between BMI at baseline and mortality among normal subjects in East Asian representative 3 cohort studies<sup>(46-48)</sup>**

The intermediate values of the BMI ranges studied were shown as plots. The result was not shown if the maximum or minimum value of the highest or lowest BMI categories was not available.

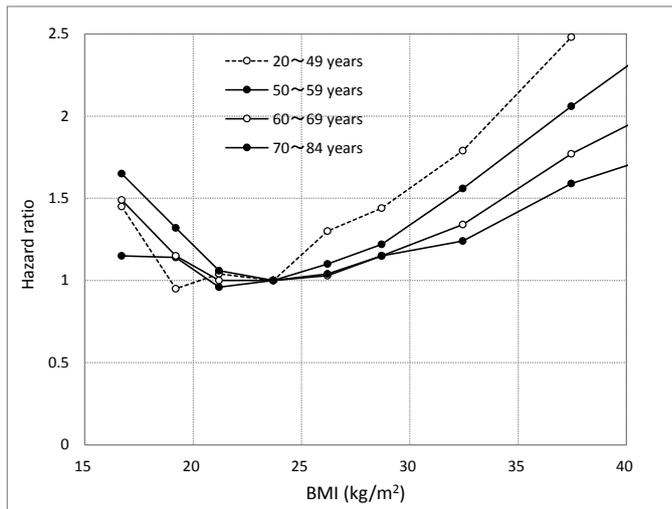
**Taiwan:** The reference category was BMI=24.0-25.9 kg/m<sup>2</sup>. The range of age at baseline was 20 years and higher, the mean follow-up duration was 10 years, the analytic sample was 58,738 men and 65,718 women, and the number of death was 3,947 men and 1,549 women. The analysis was adjusted for age, alcohol drink, educational background, smoking status, income and use of beta nuts.

**China (Shanghai):** The reference category was BMI=24.0-24.9 kg/m<sup>2</sup>. The range of age at baseline was 40 years and higher, mean follow-up duration was 8.3 years, the analytic sample was 158,666 men and women, and the number of death was 10,047 men and 7,640 women. The analysis was adjusted for age, alcohol drink, physical activity, residential area and urbanization of the residential area.

**Korea:** The reference category was BMI=23.0-24.9 kg/m<sup>2</sup>. The range of age at baseline was 30-95 years, mean follow-up duration was 12 years, the analytic sample was 770,556 men and 443,273 women, and the number of death was 58,312 men and 24,060 women. The analysis was adjusted for age, smoking status, alcohol drinking, exercise participation, fasting plasma glucose, systolic blood pressure and serum cholesterol levels.

are presented in Figure 4. These results revealed that the BMI producing a supposed hazard ratio lower than  $\pm 0.1$ , with 22.5–24.9 kg/m<sup>2</sup> as the standard, was 18.5–24.9 kg/m<sup>2</sup> in those aged 20–49 years, 20.0–24.9 kg/m<sup>2</sup> in those aged 50–59 years, and 20.0–27.4 kg/m<sup>2</sup> in those aged 60–69 and 70–84 years<sup>(42)</sup>. Incidentally, the above-mentioned studies inevitably included individuals who had already experienced weight loss due to preexisting latent diseases, or adverse health effects at the time of the baseline survey; this may have led to some kind of “reversal causality”. The possibility of a phenomenon, whereby total mortality is lowest for a slightly higher BMI, which contradicts the true correlation, cannot be ruled out. Some schools of thought question the existence of this phenomenon and its effect on results; however, no consensus has yet been reached<sup>(50,51)</sup>.

Another report states that weight gain or a loss of 5 kg or more, over a 5-year period, is correlated with an increase in mortality, irrespective of BMI<sup>(52)</sup>. However, the effects of weight gain or loss on health are thought to differ, depending on whether the gain or loss is intentional or unintentional. One report found that the mortality in a group of obese individuals, who intentionally lost weight, was significantly lower than that of a group of individuals whose weight did not change<sup>(53)</sup>; however, a meta-analysis found that the effect of intentional weight

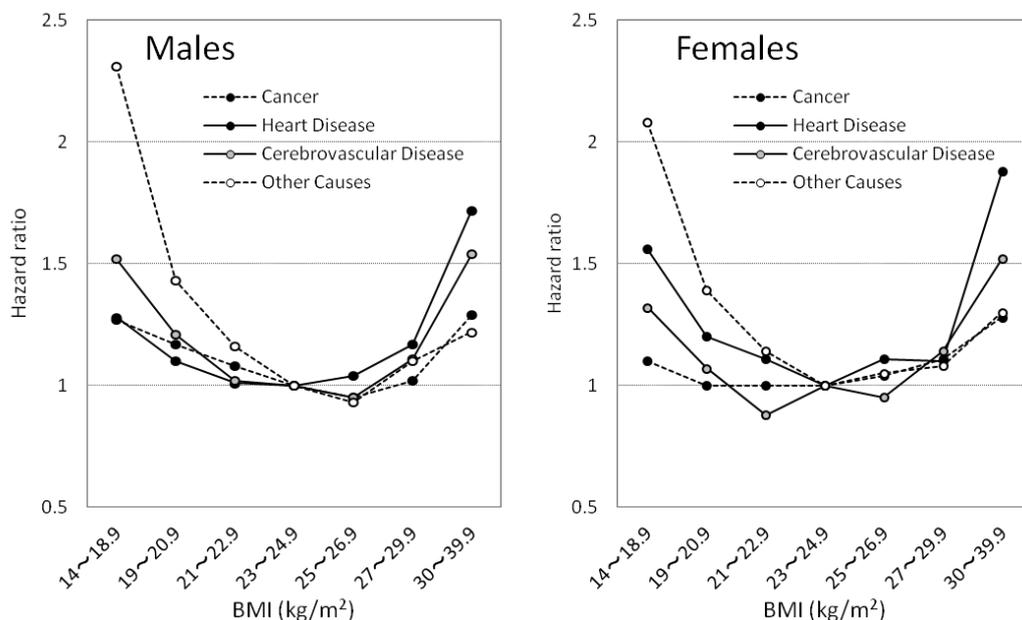


**Figure 4. The hazard ratio for mortality by age categories from the results of a pooled analysis of 19 cohort studies (1,460,000 white Americans): the analysis of never smokers<sup>(42)</sup>**

The intermediate values of the BMI ranges studied were shown as plots. The reference category was BMI=22.5-24.9 kg/m<sup>2</sup>. The range of age at baseline was 19-84 years (median 58), and the mean follow-up duration was 10 years (range: 5-28). The analysis was adjusted for sex, alcohol drink, educational background, marital status and physical activity.

loss on decreased mortality is not always clear<sup>(54)</sup>, and conclusions are yet to be drawn.

Additionally, according to a study that observed the correlation of cause-specific mortality with BMI, the BMI with the lowest mortality for cardiovascular diseases, particularly cardiac diseases, was lower than the BMI with the lowest total mortality; however, the BMI



**Figure 5 The association between cause-specific mortality and baseline BMI: the hazard ratio compared to BMI=23-24.9kg/m<sup>2</sup> from a pooled analysis of 7 Japanese cohort studies**

The range of age at baseline was 40-103 years, the mean follow-up duration was 12.5 years, the analytic sample was 162,092 men and 191,330 women, and the number of death was 25,944 men and 16,036 women. The analysis was adjusted for age, smoking status, alcohol drinking, history of hypertension, leisure or physical activity, and other variables (depending on each cohort). This analysis excluded early follow-up (less than 5 years).

with the lowest mortality for other diseases, particularly respiratory diseases, tended to be higher<sup>(41,43,45)</sup>. The results of a pooled analysis of seven Japanese cohort studies are presented in Figure 5 as an example. Furthermore, a study found that the incidence of diabetes decreased as the BMI decreased<sup>(55,56)</sup>, and the correlation between the two differed greatly from the correlation observed between BMI and total mortality.

Thus, a summary of the range of BMIs with the lowest total mortality reported in observational epidemiological studies is presented in Table 1.

**Table 1. The range of BMI which showed the lowest all-cause mortality in observational studies (18 years and older)<sup>1</sup>**

Age (years)	BMI (kg/m <sup>2</sup> ) which showed the lowest all-cause mortality
18-49	18.5-24.9
50-69	20.0-24.9
70+	22.5-27.4

<sup>1</sup> For both males and females.

However, as presented in Table 2, on studying the proportion of Japanese individuals whose BMI falls below, within, and above the range with the lowest total mortality, a large discrepancy was observed, in real-world settings, for those aged 18–49 years (10.1% above, 68.4% within, and 21.5% below), 50–69 years (15.8% above, 56.5% within, and 27.7% below), and 70 years or older (45.0% above, 45.5% within, and 9.5% below).

**Table 2. BMI distribution by gender and age groups**

Age (years)		Distribution of BMI (%)						
18-49	Range of BMI	<18.5	18.5-19.9	20.0-22.4	22.5-24.9	25.0-27.4	27.5 ≤	
	Total	10.1	17.3	29.8	21.3	11.6	9.8	
		10.1	68.4			21.5		
	Males	4.7	11.2	16.2	11.4	26.9	15.7	14.0
		4.7	65.7			29.7		
	Females	14.7	22.5	20.7	11.0	16.6	8.1	6.4
14.7		70.8			14.5			
50-69	Range of BMI	<18.5	18.5-19.9	20.0-22.4	22.5-24.9	25.0-27.4	27.5 ≤	
	Total	5.7	10.1	28.0	28.5	17.3	10.3	
		15.8		56.5		27.7		
	Males	2.9	7.2	12.2	12.7	32.3	21.7	11.0
		10.1		57.2		32.7		
	Females	8.1	12.5	18.0	12.6	25.4	13.7	9.8
20.6		56.0		23.5				
70+	Range of BMI	<18.5	18.5-19.9	20.0-22.4	22.5-24.9	25.0-27.4	27.5 ≤	
	Total	8.7	9.9	14.4	12.0	28.6	16.9	9.5
		45.0			45.5		9.5	
		33.0			40.6		26.4	
	Males	7.2	8.9	13.4	11.8	31.9	18.3	8.6
		41.3			50.2		8.6	
29.5			43.7		26.9			
Females	9.9	10.7	15.2	12.2	26.0	15.9	10.2	
	48.0			41.9		10.2		
	35.8			38.2		26.1		

Data source: National Health and Nutrition Survey 2010, 2011

### 3-2-3. Target BMI Range

The current target BMI range, comprehensively determined taking into account mortality, incidence rates of each of the diseases, and their correlation with BMI, the correlation between cause of death and BMI, and the BMI state in Japanese people, as obtained from the results of observational epidemiological studies, is presented in Table 3. In those aged 70 years or older, in particular, a discrepancy was observed between reality and the BMI with the lowest total mortality; therefore, the current target BMI range was set at 21.5–24.9 kg/m<sup>2</sup>, based on the need to consider the prevention of both frailty and LRDs. However, many factors (including genetic factors and environmental factors, such as living habits) contribute to total mortality, and there is little sense in strictly managing only BMI to control body weight. Furthermore, a high level of physical activity is effective in preventing and reducing obesity<sup>(37)</sup>, and reportedly correlates with a decrease in total mortality, independent of body weight<sup>(39,40)</sup>. Therefore, the use of BMI should be limited to the maintenance of health and prevention of LRDs. The correlation of BMI with malnutrition and disease prevention (including stroke prevention) is important from the perspective of preventative care, particularly in those aged 70 years or older, in order to avoid frailty due to advanced old age. However, it is best to manage BMI, taking into consideration the characteristics of each individual.

**Table 3. Target BMI range (18 years and older)<sup>1, 2</sup>**

Age (years)	Target BMI
18-49	18.5-24.9
50-69	20.0-24.9
70+	21.5-24.9 <sup>3</sup>

<sup>1</sup> For both males and females. These values shall be used merely as a reference.

<sup>2</sup> Target range is defined through comprehensive consideration on the association between incidence rate for each disease and BMI, the association between causes of death and BMI, and actual BMI of Japanese people, based on BMI with the lowest all-cause mortality reported in epidemiological observational studies.

<sup>3</sup> For people 70 years and over, the actual BMI deviates from the BMI with the lowest all-cause mortality. The tentative target BMI range is determined to be 21.5-24.9, considering the necessity to take into account both the prevention of frailty and prevention of LRDs.

For example, energy requirements calculated for a normal PAL (level II), using reference values for basal metabolism and reference height, as explained later in this text, in men and women aged 18–29 years, 30–49 years, 50–69 years, and 70 years or older, respectively, are 2,300–3,000 and 1,800–2,400, 2,100–2,800 and 1,800–2,400, 2,100–2,600 and 1,700–2,100, and 2,000–2,400 kcal/day and 1,700–1,900 kcal/day, demonstrating a wide range of requirements. Moreover, it should be noted that there are considerable inter-individual differences in energy requirement, even among those with the same BMI or body weight.

## **(Supplemental Statement) Estimated Energy Requirement Method Used to Calculate the Estimated Energy Requirement**

### **1. Basic Concept behind the Calculation Methods**

If both body weight and body composition remain unchanged, energy intake is equal to energy expenditure, and total energy expenditure can be assessed using the doubly labeled water method. However, as explained earlier, various dietary assessments are typically affected by underreporting in the form of systematic errors, as well as random errors caused by diurnal variations. The estimated energy requirement is, therefore, calculated from the estimated value for total energy expenditure without using the energy intakes obtained from dietary assessments.

The estimated energy requirement for adults (excluding pregnant and lactating women) is calculated as follows:

**Estimated energy requirement = basal metabolism reference value (kcal/kg, body weight/day) × reference body weight (kg) × physical activity level**

In addition, to calculate the estimated energy requirements of infants, children, pregnant women, and lactating women, the amount of energy necessary for growth, the continuation of pregnancy, or lactation is added.

The estimated energy requirement for each sex, age group, and PAL was calculated as shown in Table 5. The factors used in the calculation are described below.

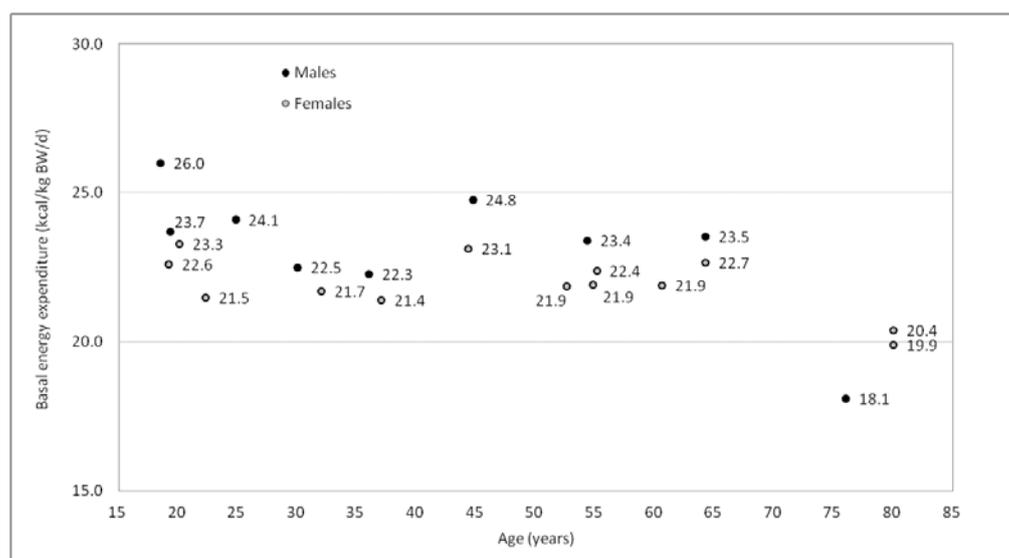
### **2. Basal Metabolism Reference Values**

The basal metabolism reference values are presented in Table 4, based on those for adults, as measured in 13 Japanese studies (Figure 6)<sup>(57-69)</sup>, and a study on individuals aged 6–17 years<sup>(70)</sup>.

The basal metabolism reference value is determined such that the estimated value and the actual measured value in the reference body size match. This is why the estimation errors are greater for body sizes and greatly deviate from the standard. Basal metabolism is overestimated in Japanese people too, when the basal metabolism reference values are used for obese individuals<sup>(71)</sup>. Conversely, basal metabolism is underestimated in slim individuals. The estimated energy requirement, obtained by multiplying this over- or underestimated basal metabolism and the PAL, is likely to be greater than the true energy requirement of obese individuals and lower than that of slim individuals. Therefore, if this estimated energy intake is used to plan the energy intake, the body weight may increase in obese individuals and decrease in slim individuals.

**Table 4. The basal metabolism reference values for the reference bodyweight (BW)**

Gender	Male			Female		
Age (years)	Basal Metabolic Reference value (kcal/kg BW/day)	Reference BW (kg)	Basic Metabolic Rate (kcal/day)	Basal Metabolic Reference value (kcal/kg BW/day)	Reference BW (kg)	Basic Metabolic Rate (kcal/day)
1-2	61	12	700	60	11	660
3-5	55	17	900	52	16	840
6-7	44	22	980	42	22	920
8-9	41	28	1,140	38	27	1,050
10-11	37	36	1,330	35	36	1,260
12-14	31	49	1,520	30	48	1,410
15-17	27	60	1,610	25	52	1,310
18-29	24	63	1,520	22	50	1,110
30-49	22	69	1,530	22	53	1,150
50-69	22	65	1,400	21	53	1,100
70+	22	60	1,290	21	50	1,020



**Figure 6. Reported basal metabolic rates in Japanese adults (13 studies)**

The following estimation formula, for the basal metabolic rate of Japanese people, using age, sex, height, and body weight<sup>(65)</sup>, has been shown not to produce systematic errors due to body weight, up to a BMI of approximately 30 kg/m<sup>2</sup>(34), and can estimate the basal metabolic rate in obese individuals with a BMI of 25–29.9 kg/m<sup>2</sup>.

$$\text{Basal metabolism (kcal/day)} = [0.0481 \times \text{body weight (kg)} + 0.0234 \times \text{height (cm)} - 0.0138 \times \text{age (years)} - \text{constant (men: 0.4235, women: 0.9708)}] \times 1000/4.186$$

Basal metabolic rate was found to be more strongly correlated with fat-free mass than

body weight<sup>(62,65,68,72)</sup>. In the future, it may be possible to estimate basal metabolic rate with a higher degree of accuracy through the appropriate assessment of physical composition.

Incidentally, many reports claim that the basal metabolic rate of diabetes patients is either no different from that of healthy individuals or approximately 5%–7% higher, when corrected for body composition (this is thought to be due to the energy consumed during gluconeogenesis in the liver, among other factors.)<sup>(73–80)</sup>. Few studies have focused on the outcomes of hyperglycemic individuals receiving health guidance; however, a cross-sectional study found that the metabolic rate during sleep exhibited normal glucose tolerance < impaired glucose tolerance (IGT) < diabetes, and changes in the basal metabolism, over time, in the same individual resulted in normal glucose tolerance < IGT (+4%) < diabetes (+3%)<sup>(81)</sup>. Those with hyperglycemia, requiring just health guidance (fasting blood glucose levels: 100–125 mg/dL), therefore, have a basal metabolism that is not very different from that of those with euglycemia. Furthermore, only a few studies have examined the total energy expenditure in diabetes patients using the doubly labeled water method, and no differences in the PAL and total energy expenditure have been observed between diabetes patients and individuals with normal glucose tolerance<sup>(73,75)</sup>.

### 3. Physical Activity Level

#### 3-1. Adults

The PALs of adults, calculated from the measured energy expenditure and estimated basal metabolic rate of healthy Japanese adults (aged 20–59 years; 150 individuals)<sup>(82)</sup>, were used. In other words, the overall PAL derived from the PALs of men and women was  $1.72 \pm 0.26$  (mean  $\pm$  standard deviation), and the overall PAL of 63 individuals with level II physical activity was  $1.74 \pm 0.26$ . Three types of PAL were established based on these data (Table 5).

**Table 5. Daily activities and time of physical activity by PAL**

PAL <sup>1</sup>	Low (I)	Moderate (II)	High (III)
	1.50 (1.40-1.60)	1.75 (1.60-1.90)	2.00 (1.90-2.20)
Daily activities <sup>2</sup>	Corresponds to sedentary lifestyle	Corresponds to sedentary work, however, includes movement and housework such as commuting and shopping, and sports with light intensity.	Individuals who involved in works with high-intensity physical activity or high-intensity leisure-time physical activity such as regular sports habits
Total time per day on physical activity of a moderate intensity (hour/day) <sup>3</sup>	1.65	2.06	2.53
Total length of walking at work (hour/day) <sup>3</sup>	0.25	0.54	1.00

<sup>1</sup> Representative values (approximate range).

<sup>2</sup> Based on reports of Black, et al.<sup>(83)</sup> and Ishikawa-Takata, et al.<sup>(82)</sup> and considered that PAL is largely influenced by physical activity during work

<sup>3</sup> The data was based on Ishikawa-Takata, et al.<sup>(84)</sup>

Metabolic equivalent (MET; indicator of the intensity of each physical activity, expressed as a multiple of metabolic rate at rest, in the sitting position), and activity factor (Af; indicator of the intensity of each physical activity, expressed as a multiple of basal metabolic rate) are the indicators of the intensity of physical activity. As the metabolic rate at rest, in the sitting position, is approximately 10% higher while fasting than the basal metabolic rate measured in the supine position<sup>(85,86)</sup>, the following relational equation was established:  $MET \times 1.1 = Af$ . The METs of various physical activities, in healthy adults, have been summarized by Ainsworth *et al.*<sup>(87)</sup>.

A study of a population of Japanese adults (mean age:  $50.4 \pm 17.1$  years), including a relatively large number of individuals with a high PAL, found a difference between the three PALs, for the total time per day spent on physical activity of a moderate intensity (3–5.9 METs) and walking to work (Table 5)<sup>(84)</sup>. Level II physical activity (normal) corresponds to sedentary work; however, a total of 2 hours/day are spent on movement and housework such as commuting and shopping, and a total of 30 minutes/day are spent on movement within the workplace.

The above study found, however, that the time spent on physical activity during leisure time was almost zero for all three PALs. A more accurate method of estimating PAL will need to be developed in the future, taking into account the time spent on each physical activity and exercise intensity, with a particular focus on work, traveling (commuting, shopping, etc.), and housework.

In addition, when the energy expenditure from physical activity is estimated in the activity logs in the US-Canada DRIs<sup>(33,85)</sup>, excess post-exercise oxygen consumption (EPOC) from hypermetabolism after physical activity is included in the calculations for estimated energy requirement, assuming it accounts for 15% of the energy expenditure during the physical activity in question. However, the EPOC in activities of daily living is, in fact, very low<sup>(86)</sup>.

### 3-2. Elderly Individuals

Elderly individuals are more likely to have PALs that differ from those of other age groups. The typical PAL was set at 1.70, on the basis of reports that measured the PALs in healthy, independent elderly individuals (Table 6)<sup>(88–97)</sup>. Levels I, II, and III were also determined based on a study in which participants were divided into three groups based on their level of physical activity (Table 7)<sup>(98)</sup>. The mean age of the participants in a majority of those reports was 70–75 years; there is a lack of data on those aged 80 years and older. A study that re-assessed 75-year-old participants when they reached age 82 years found that the levels dropped only in men who previously had a high PAL, and that the overall PAL of both men and women was approximately 1.68<sup>(99)</sup>.

**Table 6. Reports that measured the PALs in healthy, independent elderly individuals**

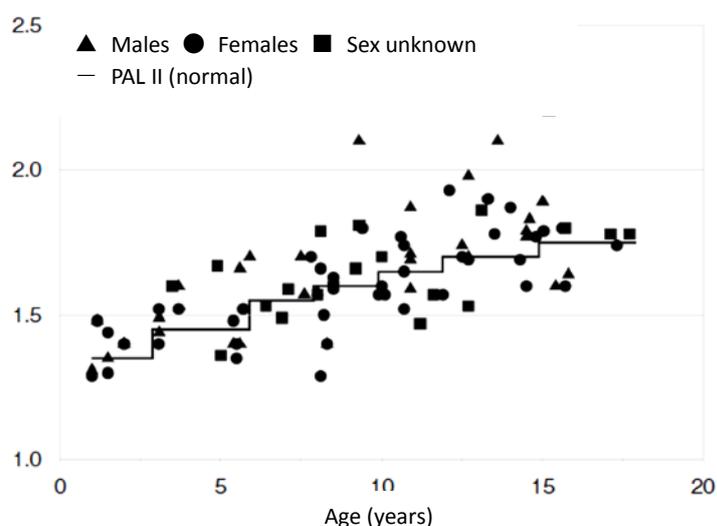
Reference	Subjects	Age (years) mean $\pm$ standard deviation	Sex (number)	BMI (kg/m <sup>2</sup> ) mean $\pm$ standard deviation	PAL mean $\pm$ standard deviation/inter quartile range
87	Healthy subjects	73	males (3), females (9)	25 $\pm$ 3	1.73 $\pm$ 0.25
88	Healthy subjects	74 $\pm$ 6	males (14) females (18)	22.5 $\pm$ 2.5	1.66 $\pm$ 0.24
89	Dependently-living individuals	72.8 $\pm$ 6.1	males (8)	22.4 $\pm$ 2.5	1.4 $\pm$ 0.1
90	Retired subjects	74.0 $\pm$ 4.4	females (10)	24.1 $\pm$ 2.8	1.59 $\pm$ 0.19
91	Healthy subjects	73 $\pm$ 3	females (10)	none	1.80 $\pm$ 0.19
92	Healthy subjects	73.4 $\pm$ 4.1	males (19)	none	1.71 $\pm$ 0.32
93	Black	74.6 $\pm$ 3.2	females (67)	28.6 $\pm$ 5.9	1.69 $\pm$ 0.24
	White	74.6 $\pm$ 3.2	females (77)	26.2 $\pm$ 5.3	1.65 $\pm$ 0.21
	Black	74.8 $\pm$ 2.9	males (72)	27.1 $\pm$ 4.5	1.71 $\pm$ 0.24
	White	75.1 $\pm$ 3.2	males (72)	27.6 $\pm$ 4.2	1.74 $\pm$ 0.22
94	Relatively-healthy subjects	78	males (2) females (9)	24.3 $\pm$ 2.6	1.74 $\pm$ 0.25
95	Home-living subjects	82 $\pm$ 3*	males (17)	24.8 $\pm$ 3.8	1.6 $\pm$ 0.2
96	Disease-free and walkable subjects	74.7 $\pm$ 6.5	males (12) females (44)	25.8 $\pm$ 4.2	1.72 (1.63-1.92)
98	Follow-up study of reference 93	74.7	males (47)	27.0 $\pm$ 4.3	1.77 $\pm$ 0.23
		82.2		27.1 $\pm$ 4.8	1.68 $\pm$ 0.21
		74.5	females (40)	28.4 $\pm$ 4.5	1.68 $\pm$ 0.19
		82.0		28.0 $\pm$ 4.3	1.67 $\pm$ 0.31

\* The value for age and BMI is the mean value of 17  $\pm$  6 individuals (total 23).

### 3-3. Children

Studies that measured the PALs of children, using the doubly labeled water method, were systematically reviewed, and the average, weighted by the number of participants, was set as the PAL. As a general rule, only reports that measured basal metabolism were included in the review<sup>(100–132)</sup>; however, reports that estimated the PAL using the basal metabolic rate in children aged less than 5 years were also included<sup>(133–139)</sup>. The resulting PALs were 1.36 for ages 1–2 years, 1.48 for ages 3–5 years, 1.57 for ages 6–7 years, 1.62 for ages 8–9 years, 1.63 for ages 10–11 years, 1.74 for ages 12–14 years, and 1.81 for ages 15–17 years, demonstrating that the PAL had a tendency to increase with age (Figure 7). A separate meta-analysis, summarizing the results of 17 studies on the relationship between age and PAL in children, also found that PAL increased with age<sup>(140)</sup>. The typical PAL for children was determined based on those reports (Table 7). The typical PALs of those aged 12–14 years and 15–17 years were set

at just 0.05 lower than the weighted average. Some reports found that the PAL exceeded “normal; level II” in these age groups. Moreover, the 2012 Physical Fitness and Athletic Ability Survey found that the ratio of individuals who spent many hours, per day, on exercise and sports was high in this age group, because the typical level II PAL was assumed to be lower than the average level. From the age of 6 years, children were divided into the same three categories as adults, considering individual differences in the PAL. The standard deviations of the averages, weighted by the number of participants in each age group, extracted from the literature, varied, with widths between 0.17 and 0.27, according to the age group, and a mean width of 0.23. The PAL of each category, among children was, therefore, set at just 0.20 higher or lower than the “normal” value for each age group.



**Figure 8. PAL of children by age**

**Table 7. PAL classification according to age categories**

PAL	Low (I)	Moderate (II)	High (III)
1-2 (years)	-	1.35	-
3-5 (years)	-	1.45	-
6-7 (years)	1.35	1.55	1.75
8-9 (years)	1.40	1.60	1.80
10-11 (years)	1.45	1.65	1.85
12-14 (years)	1.50	1.70	1.90
15-17 (years)	1.55	1.75	1.95
18-29 (years)	1.50	1.75	2.00
30-49 (years)	1.50	1.75	2.00
50-69 (years)	1.50	1.75	2.00
70 years and older	1.45	1.70	1.95

### 3-4. For Overweight and Underweight Individuals

In obese individuals, the PAL assessed with a motion sensor, such as an accelerometer, is generally low, and it has been pointed out that obesity can be a cause of decreased activity<sup>(141)</sup>. However, PAL does not correlate with BMI up to a BMI of approximately 30 kg/m<sup>(142,143)</sup>. Moreover, the PAL does not change after weight loss, in obese individuals<sup>(144,145)</sup>. This is likely because obese individuals have poor exercise efficiency, and require more energy to perform certain external tasks<sup>(146,147)</sup>. In conclusion, the use of the same PAL values as those of normal individuals is allowed for obese individuals with a BMI of 25–29.9 kg/m<sup>2</sup>.

## 4. Estimated Energy Requirement

### 4-1. Adults

The estimated energy requirement (kcal/day) in adults (aged 18 years and older) was calculated as follows:

**Estimated energy requirement (kcal/day) = basal metabolic rate (kcal/day) × PAL**

### 4-2. Children

Children in their growth phase (age 1–17 years) must consume extra energy for tissue synthesis and to build tissue (energy deposition), in addition to the energy required for their physical activity. The energy consumed during tissue synthesis is included in the total energy expenditure, so the estimated energy requirement (kcal/day) can be calculated as follows:

**Estimated energy requirement (kcal/day) = basal metabolic rate (kcal/day) × PAL + stored energy storage (kcal/day)**

The energy deposition for the increased tissue mass is calculated as the product of the sum of the daily weight gained in addition to the reference weight, and the energy deposition density<sup>(85)</sup>. See Table 8 for more information on the method of calculation.

**Table 8. Energy deposition accompanied with growth**

Gender	Males				Females			
Age	Reference BW (kg)	BW Increase (kg/year)	For building tissue		Reference BW (kg)	BW Increase (kg/year)	For building tissue	
			Energy Density (kcal/g)	Energy Deposition (kcal/day)			Energy Density (kcal/g)	Energy Deposition (kcal/day)
0-5 (months)	6.3	9.4	4.4	115	5.9	8.4	5.0	115
6-8 (months)	8.4	4.2	1.5	15	7.8	3.7	1.8	20
9-11 (months)	9.1	2.5	2.7	20	8.4	2.4	2.3	15
1-2 (years)	11.5	2.1	3.5	20	11.0	2.2	2.4	15
3-5 (years)	16.5	2.1	1.5	10	16.1	2.2	2.0	10
6-7 (years)	22.2	2.6	2.1	15	21.9	2.5	2.8	20
8-9 (years)	28.0	3.4	2.5	25	27.4	3.6	3.2	30
10-11 (years)	35.6	4.6	3.0	40	36.3	4.5	2.6	30
12-14 (years)	49.0	4.5	1.5	20	47.5	3.0	3.0	25
15-17 (years)	59.7	2.0	1.9	10	51.9	0.6	4.7	10

BW, body weight

BW increase was calculated as following:

Ex) BW increase amount for female children aged 9-11 months,

$$X = [(reference\ BW\ of\ 9-11\ months\ old\ (at\ 10.5\ months) - (reference\ BW\ of\ 6-8\ months\ old\ (at\ 7.5\ months)) / [0.875\ (year\ old) - 0.625\ (year\ old)] + [(reference\ BW\ of\ 1-2\ years\ old) - (reference\ BW\ of\ 9-11\ months\ old)] / [2\ (years\ old) - 0.875\ (years\ old)]]$$

$$BW\ increase = X/2 = [(8.4 - 7.8) / 0.25 + (11.0 - 8.4) / 1.125] / 2 \approx 2.4$$

Energy density for building tissue was calculated according to the US/Canada DRIs<sup>(85)</sup>.

Energy deposition for building tissue was calculated as the product of BW increase and energy density for building tissue.

$$Ex) Energy\ deposition\ for\ female\ children\ aged\ 9-11\ months\ (kcal/day) = [(2.4\ (kg/year) * 1000/365day)] * 2.3\ (kcal/g) = 14.8 \approx 15$$

### 4-3. Infants

Much like children, infants must consume energy for tissue synthesis and energy deposition, in addition to the energy required for physical activity. The energy consumed during tissue synthesis is included in the total energy expenditure, so the estimated energy requirement (kcal/day) can be calculated as follows:

**Estimated energy requirement (kcal/day) = total energy expenditure (kcal/day) + stored energy storage (kcal/day)**

With regards to the total energy expenditure of infants, the FAO/WHO/UNU conducted various studies on the relationship between sex, age (in months), body weight, height, and total energy expenditure, based on the results of previous studies that used the doubly labeled water method. Consequently, it was reported that the total energy expenditure of breastfed infants during infancy can be calculated using the following regression formula, in which body weight is the only independent variable<sup>(148,149)</sup>:

**Total energy expenditure (kcal/day) = 92.8 × reference weight (kg) – 152.0**

No reports have measured total energy expenditure using the doubly labeled water method, in Japanese infants. Therefore, the total energy expenditure per day (kcal/day) was calculated by substituting the reference weight of Japanese people into these formulas.

Much like in the case of children, the energy deposition in infants was calculated as

the product of the sum of the daily weight gained, in addition to the reference weight, and the energy density for the increased tissue (Table 8)<sup>(133)</sup>.

The estimated energy requirement in infants is shown according to age in months (0–5 months, 6–8 months, and 9–11 months). Furthermore, it should be noted that during the ages of 0–5 months, when an infant's body weight varies greatly, large differences arise in the energy requirement, between the first and second half of that period.

Attention must also be paid to the fact that artificially fed infants generally have a larger total energy expenditure than breastfed infants<sup>(148)</sup>. Moreover, the FAO/WHO/UNU state that the total energy expenditure of artificially fed infants can be estimated as follows<sup>(148,149)</sup>:

$$\text{Total energy expenditure (kcal/day)} = 82.6 \times \text{body weight (kg)} - 29.0$$

#### 4-4. Pregnant Women

The estimated energy requirement of pregnant women is calculated as follows:

$$\text{Estimated energy requirement of pregnant women (kcal/day)} = \text{estimated energy requirement before pregnancy (kcal/day)} + \text{additional energy required for pregnancy (kcal/day)}$$

Considering the childbearing age of women covers several age groups, for the estimated energy requirement, the amount of energy that should be additionally consumed compared to the amount consumed before pregnancy needs to be shown as the additional amount of energy required for each trimester to ensure a normal delivery and the maintenance of an appropriate nutritional status during pregnancy.

A longitudinal study using the doubly labeled water method found that the PAL during pregnancy decreased in the first and third trimesters; however, the basal metabolic rate greatly increased in the late stage due to pregnancy-related weight gain<sup>(148–154)</sup>. As a result, the rate of increase in the total energy expenditure was almost the same as the rate of increase in the body weight, across all trimesters, and there was almost no difference in the total energy expenditure per body weight between the trimesters. Therefore, when the amount of change in the total energy expenditure in each pregnancy trimester, compared to the total energy expenditure before pregnancy (estimated energy requirement), was corrected so as to correspond to the final weight gain from pregnancy (11 kg), changes of +19 kcal/day, +77 kcal/day and +285 kcal/day were obtained for the first, second and third trimesters, respectively<sup>(148,149,155)</sup>.

Energy depositions, in the form of protein and fat, corresponding to a final weight gain of 11 kg were calculated from the protein and fat depositions in each pregnancy trimester, and the sum of both was used to obtain the total energy deposition<sup>(148,149)</sup>. As a result, the energy depositions were 44 kcal/day, 167 kcal/day, and 170 kcal/day in the first, second and third trimesters, respectively.

Thus, the additional energy in each pregnancy trimester is ultimately calculated as follows.

$$\text{Additional energy during pregnancy (kcal/day)} = \text{amount of change in total energy}$$

**expenditure due to pregnancy (kcal/day) + energy deposition (kcal/day)**

When rounded to the closest 50 kcal, the additional energy values were calculated to be 50 kcal/day, 250 kcal/day and 450 kcal/day in the first, second and third trimesters, respectively.

#### 4-5. Lactating Women

The estimated energy requirement of lactating women is calculated as follows:

**Estimated energy requirement of lactating women (kcal/day) = estimated energy requirement before pregnancy (kcal/day) + additional energy for lactation (kcal/day)**

A woman's body weight, directly after childbirth, is greater than her weight before pregnancy, and the need for the energy consumed to synthesize breast milk is a factor that increases the basal metabolic rate. However, no clear increase in basal metabolic rate is actually seen<sup>(149)</sup>. In one of four studies that longitudinally examined basal metabolic rate using the doubly labeled water method, the energy used for physical activity was found to decrease significantly<sup>(151)</sup>; however, in the other three studies, the difference was not significant, despite an approximate decrease of 10% in the absolute amount of energy<sup>(152,153,156)</sup>. These studies showed that the total energy expenditure during the lactation period is the same as that before pregnancy<sup>(149,152,153,156)</sup>, and that no specific additional amount of energy needs to be set for lactating women, from the standpoint of changes in total energy expenditure. However, total energy expenditure does not include the amount of energy required to synthesize breast milk, so lactating women need to consume the energy required for this.

The amount of energy required to synthesize breast milk is calculated as follows, assuming the lactated amount is the same as the milk volume (0.78 L/day), and the energy content of breast milk is 663 kcal/L<sup>(157-159)</sup>:

**Amount of energy required to synthesize breast milk (kcal/day) = 0.78 L/day × 663 kcal/L = 517 kcal/day**

In contrast, energy is gained as a result of weight loss (decomposition of body tissue) after delivery (childbirth), but the required energy intake decreases that much more for it. Given an energy decrease of 6,500 kcal per 1 kg of body weight, from weight loss, and a weight loss of 0.8 kg/month<sup>(148,149)</sup>, the amount of energy required can be calculated as follows:

**Energy decrease from weight loss (kcal/day) = 6,500 kcal/kg body weight × 0.8 kg/month ÷ 30 days = 173 kcal/day**

Therefore, if the extra energy that lactating women who experience a normal pregnancy and delivery should consume during the lactation period compared to the energy consumed before pregnancy is assumed to be the amount of additional energy needed for lactation, this amount can be calculated as follows:

**Additional amount of energy for lactation (kcal/day) = amount of energy required to synthesize breast milk (kcal/day) – energy decrease from weight loss (kcal/day)**

Thus, an additional 517–173 = 344 kcal/day of energy was obtained, which was rounded to 350 kcal/day.

**Table appendix Estimated Energy Requirement (kcal/day)**

Gender	Males			Females		
PAL <sup>1</sup>	I	II	III	I	II	III
0-5 months	-	550	-	-	500	-
6-8 months	-	650	-	-	600	-
9-11 months	-	700	-	-	650	-
1-2 years	-	950	-	-	900	-
3-5 years	-	1,300	-	-	1,250	-
6-7 years	1,350	1,550	1,750	1,250	1,450	1,650
8-9 years	1,600	1,850	2,100	1,500	1,700	1,900
10-11 years	1,950	2,250	2,500	1,850	2,100	2,350
12-14 years	2,300	2,600	2,900	2,150	2,400	2,700
15-17 years	2,500	2,850	3,150	2,050	2,300	2,550
18-29 years	2,300	2,650	3,050	1,650	1,950	2,200
30-49 years	2,300	2,650	3,050	1,750	2,000	2,300
50-69 years	2,100	2,450	2,800	1,650	1,900	2,200
70+ years <sup>2</sup>	1,850	2,200	2,500	1,500	1,750	2,000
Pregnant women (additional) <sup>3</sup>	/			+50	+50	+50
Early-stage				+250	+250	+250
Mid-stage				+450	+450	+450
Late-stage						
Lactating women (additional)				+350	+350	+350

<sup>1</sup> PALs (physical activity levels) of I, II, and III indicate low, medium and high activity levels, respectively.

<sup>2</sup> Calculated mainly from reports made on healthy independent subject persons 70-75 years old.

<sup>3</sup> It is important to assess the physique of individual pregnant women, weight increase during pregnancy, and fetal growth.

Note 1: On application of the present table, ensure to conduct assessment of dietary intake, measurement of the body weight and calculation of BMI. Excess energy or inadequate energy shall be evaluated according to change in body weight or BMI.

Note 2: If a subject falls under the category of PAL I, the energy intake may have to be maintained low level to match the low energy consumption level. Such subject needs to increase the level of physical activities from the prospect of health maintenance and promotion.

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## **Protein**

### **1. Background Information**

#### **1-1. Definitions and Classifications**

Proteins are made up of 20 different amino acids linked by peptide bonds. They are important components of organisms, and their types differ depending on the number and types of amino acids, and the sequence of the peptide bonds. Many types of proteins exist, ranging from those with a molecular weight of around 4,000 to viral proteins with molecular weights from several tens of millions to hundreds of millions. Proteins with a small number of amino acids linked by peptide bonds are referred to as peptides. Proteins are composed of 20 different amino acids, which are directly encoded by codons. Of these 20 amino acids, humans can synthesize 11 from other amino acids or intermediate metabolites. The remaining nine amino acids must be consumed from diet, and are referred to as essential amino acids. The essential amino acids are histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan, and valine.

### **2. To Avoid Inadequacy**

#### **2-1. Factors to be Considered in Estimating Requirements**

In the previous versions, the DRIs for proteins were calculated on the basis of the amount required to maintain nitrogen balance. To calculate the DRIs for proteins, using the nitrogen balance method, the following must be considered: (1) technical difficulties, (2) metabolic adaptations associated with changes in protein intakes, (3) the protein-sparing effect of energy, (4) lifestyle, and (5) interindividual variability.

##### **2-1-1. Technical Difficulties Associated with the Nitrogen Balance Method**

The nitrogen balance method requires all nitrogen intakes, and nitrogen excretion to be accurately quantified. However, as it is difficult to collect data on all foods that were not consumed, such as spilled food and food left on the plate, it is likely that the nitrogen intake may be overestimated. Nitrogen is excreted from the body primarily through urine and feces; however, it can also be eliminated via various bodily secretions such as the skin, sweat, desquamation, hair, and nails. This total excretion is more likely to be underestimated than overestimated. This overestimation of protein intake and underestimation of protein excretion may mistakenly produce a positive nitrogen balance. Therefore, this erroneous calculation may lead to the underestimation of protein or amino acid requirements.

##### **2-1-2. Metabolic Changes Associated with Changes in Protein Intake**

When the amount of dietary protein intake is changed, a certain amount of time must normally be given until the change is adapted. This is not only because the human metabolism requires time to adapt to new protein intake, but also because the body's urea pool needs to adjust to the change in protein intake. The urea pool expands or shrinks with increases or

decreases in protein intake with a half-life of approximately 8–12 hours. It takes more than 48 hours for the pool to reach its new size. During this time, urea nitrogen emissions cannot be used as indicators of amino acid oxidation.

In 1985, a joint report by the FAO/WHO/UNU concluded that major adjustments are complete within the first five to seven days, in each sex and age group. On the basis of this conclusion, nitrogen balance studies that allow 1 week or less for adjustments are less likely to produce reliable data; thus, studies that allow a dietary adjustment period of 1–3 weeks must be used.

### **2-1-3. Protein-Sparing Effect of Energy**

Protein utilization efficiency changes with intakes of protein, amino acids, and total nitrogen. Protein metabolism is also affected by intakes of nutrients other than nitrogen compounds. The effect of energy intake on protein metabolism has long been known as the sparing protein effect<sup>(1)</sup>. Energy deficiency reduces protein utilization efficiency, whereas increased energy intake improves nitrogen balance<sup>(2)</sup>. The promotion of protein synthesis, and suppression of decomposition due to the increased secretion of insulin contribute to this. In addition, a report on nitrogen balance in adults (361 individuals) revealed a significant positive correlation between energy intake and nitrogen balance<sup>(3)</sup>. Moreover, a previous experiment regarding protein requirement found that energy balance tended to be positive, and that protein requirement was underestimated. However, it has recently become possible to successfully measure protein requirement in a state of energy equilibrium.

### **2-1-4. Lifestyle Behavior**

#### **2-1-4-1. Physical Activity and Exercise**

In individuals with a large intake, who actively engage in activities, protein requirements can easily be met, and they may not need to worry about the quality of the proteins consumed. However, inactive and elderly individuals are prone to protein or other nutrient deficiencies if they do not pay attention to their diet. Lack of exercise leads to body protein catabolism, while appropriate exercise enhances the use of dietary proteins. Strenuous exercise enhances the breakdown of proteins. Therefore, protein requirement follows a U-shape, depending on the exercise intensity<sup>(4)</sup>. Some studies in children and adults reported that appropriate exercise promotes growth, and enhances the use of dietary proteins<sup>(5,6)</sup>.

Transdermal nitrogen loss generally increases due to perspiration during exercise, resulting in the hypercatabolism of amino acids, and decreased synthesis and increased decomposition of proteins by the body. However, once exercise ends, the synthesis of proteins by the body exceeds their decomposition, and the lost nitrogen is often regained. Protein requirements reportedly do not increase when mild or moderate exercise (200–400 kcal/day) is performed<sup>(7,8)</sup>.

#### **2-1-4-2. Rest and Stress**

Daily stress is only addressed in reports on nitrogen balance tests after 48 hours of sleep deprivation, and at the end of the academic year, in university students. The quantitative impact of mild stress on nitrogen balance is not clear. Moreover, daily stress also has an effect on the participants of nitrogen balance tests, and because this effect is already included in the amount required to maintain nitrogen balance, it was decided not to estimate a safety margin for stress.

### **2-1-4-3. Smoking and Drinking**

Smoking causes cellular free-radical damage, while drinking, both directly and indirectly, affects metabolism. However, the quantitative relationship between protein requirement, and smoking or drinking is unclear.

### **2-1-5. Interindividual Variability**

A wide range (10%–40%) of amounts required to maintain nitrogen balance has been reported to date. This range of variability includes intraindividual variability and researcher-related variability, such as experimental conditions and experimental errors, in addition to interindividual variability. According to the results of the data analysis of 235 participants from 19 studies, inter-researcher variability accounted for 40% of the observed variability, while the remaining 60% was due to intra-researcher variability<sup>(9)</sup>. Furthermore, the outcomes of repeating the measurements in the same participants revealed that two-third of the intra-researcher variability was intraindividual variability, and one-third was true interindividual variability. The coefficient of variation was 12%. However, the coefficient of variation was set at 12.5%, in light of the bias in the variation curve. This was used as the basis for setting an RDA calculation coefficient of 1.25, when calculating the RDA from the EAR.

## **2-2. Methods Used to Set the Estimated Average Requirement and Recommended Dietary Allowances**

### **2-2-1. Adults**

The protein maintenance requirement of high-quality (animal) proteins measured in the nitrogen balance experiment was used as a basis for calculating the reference value for the estimated average requirement (EAR), corrected for the digestive efficiency of mixed proteins in everyday meals. Interindividual variability was then added to this in order to calculate the recommended dietary allowance (RDA). To assess the quality of everyday mixed proteins, amino acid intake was calculated from protein intake and the amino acid compositions of each food group presented in the results of the 2010 and 2011 National Health and Nutrition Survey<sup>(10)</sup>. This amino acid score exceeds 100 even if the 1973 FAO/WHO<sup>(11)</sup> amino acid scoring patterns, the 1985 FAO/WHO/UNU<sup>(12)</sup> amino acid scoring patterns, and the 2007 FAO/WHO/UNU<sup>(13)</sup> scoring patterns are used as reference. Therefore, no quality correction is necessary.

When the values of the 17 studies that examined the nitrogen balance maintenance dose of high-quality (animal) protein were averaged, the protein maintenance requirement was

determined to be 0.65 g/kg body weight (BW)/day (104 mg nitrogen/kg BW/day)<sup>(14–28)</sup>. Consequently, this value was decided to be the protein maintenance requirement (Table 1).

A study that measured the digestive efficiency of everyday mixed proteins in women (12 individuals) reported an average efficiency of 92.2%<sup>(24)</sup>. In addition, the result for men (6 individuals) was 95.4%<sup>(29)</sup>. The digestive efficiency of everyday mixed proteins was, therefore, set at 90%, and the EAR was calculated with the following formula. The RDA was the value for EAR multiplied by an RDA calculation coefficient of 1.25, assuming an interindividual coefficient of variation of 12.5%.

**Reference value for EAR calculation (g/kg BW/day) = protein maintenance requirement ÷ digestive efficiency = 0.65 ÷ 0.90 = 0.72**

**EAR (g/day) = reference value for EAR calculation (g/kg BW/day) × reference BW (kg)**

**RDA (g/day) = EAR (g/day) × RDA calculation coefficient**

### 2-2-2. Elderly Individuals

During adulthood, physiological functions such as maximum voluntary ventilation, renal blood flow, and vital capacity decline, skeletal muscle mass tends to decrease, and body fat mass tends to increase due to aging. While muscle protein metabolism decreases, visceral protein metabolism remains largely unchanged. Decreases in protein metabolic turnover and physiological functions are thought to influence protein utilization efficiency in the elderly. However, some reports found that there were no differences between the EAR of protein among elderly individuals and that among young adults (aged 18–31 years)<sup>(9)</sup>. Elderly individuals generally have inactive daily lives, and, thus, have a low dietary intake, and often experience a loss of appetite. These differences in lifestyle are also thought to influence the EAR of protein.

The mean value of the amount of nitrogen balance maintenance observed in healthy elderly individuals, under normal dietary intake conditions, was considered as the reference value in the determination of the EAR.

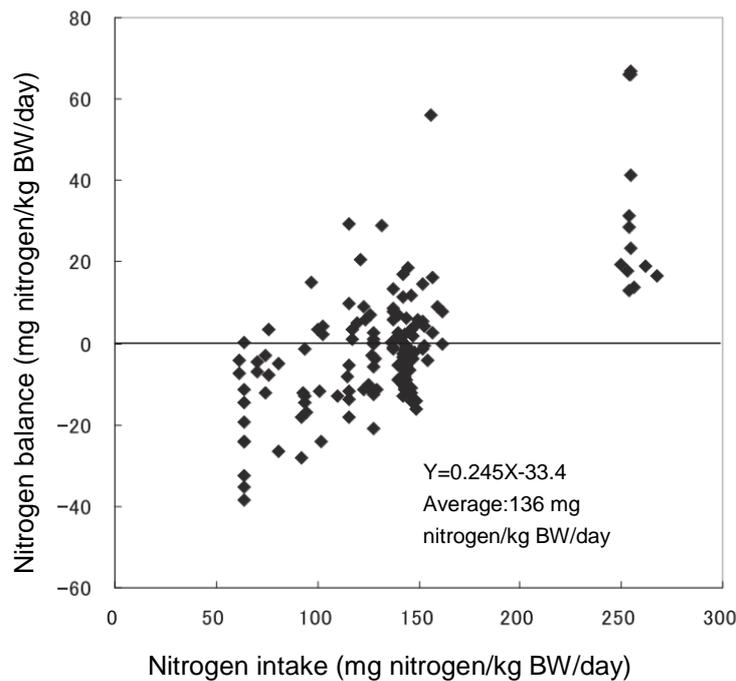
Of the reports that examined protein reference values for EAR calculation in elderly individuals, a pooled analysis was conducted using 144 data on the nitrogen balance of 60 participants, from five studies, that presented the nitrogen balance results of individuals<sup>(28,30–33)</sup>. The average value (0.85 g/kg BW/day [136 mg nitrogen/kg BW/day]) obtained from this pooled analysis was set as the reference value for the EAR calculations (Figure 1). However, the digestion-absorption rate of mixed proteins was 90% with this reference value, which is the rate obtained after correcting the other nitrogen loss values, using the actual values and 5 mg/kg BW/day. Moreover, the RDA was calculated by multiplying the EAR and an RDA calculation coefficient of 1.25, assuming that the interindividual coefficient of variation was the same as that of adults (12.5%).

**EAR (g/day) = reference value for EAR calculation (g/kg BW/day) × reference BW (kg)**

**RDA (g/day) = EAR (g/day) × RDA calculation coefficient**

A significant amount of elderly individuals living in nursing homes or receiving home

care are undernourished and exhibit a negative nitrogen balance<sup>(34)</sup>. Moreover, frailty is frequently observed in elderly individuals with decreased protein intakes<sup>(35)</sup>. When the PAL drops, the protein metabolism of the skeletal muscle decreases, which in turn leads to increases in the EAR of protein. Since the EAR of protein also increases when the energy intake is low, a different amount of protein replenishment than that for healthy individuals needs to be considered for such elderly individuals.



**Figure 1** The nitrogen balance of elderly individuals (from 5 studies' results) <sup>(23,30-33)</sup>

### 2-2-3. Children

The reference value for EAR, in children aged 1–17 years, was calculated using the additive factor method, from the protein maintenance requirement and protein depositions accumulated with growth (Table 2). The utilization efficiency is the protein utilization efficiency during weight maintenance. The EAR was calculated by multiplying the reference value and reference BW. The RDA was calculated by multiplying the EAR and an RDA calculation coefficient of 1.25, assuming the interindividual coefficient of variation was the same as that of adults (12.5%).

**Reference value for EAR calculation (g/kg BW/day) = (protein maintenance requirement ÷ utilization efficiency) + (protein deposition ÷ deposition efficiency)**

**EAR (g/day) = reference value for EAR calculation (g/kg BW/day) × reference BW (kg)**

**RDA (g/day) = EAR (g/day) × RDA calculation coefficient**

In order to obtain the protein maintenance requirement, the mean value (0.67 g/kg BW/day [107 mg nitrogen/kg BW/day]) obtained from the nitrogen balance method results<sup>(36-41)</sup> of growing infants (9–62 months), children (8–9 years), and adolescents (12–14 years) was

used (Table 3). However, the nitrogen loss via routes other than urine and feces was set at  $6.5 \pm 2.3$  mg nitrogen/kg BW/day (5–9 mg nitrogen/kg BW/day) on the basis of currently available reports<sup>(36,42–45)</sup>, and the above-stated maintenance requirement was calculated using this value. Given that no evidence was found stating that the maintenance requirement differs according to the developmental stage (infancy, childhood, and adolescence), this value was used for children of all ages.

Protein deposition was calculated as the amount of accumulated protein, in accordance with growth, using the increase in reference BW in each child age group, and the ratio of body protein to reference BW. The ratio of body protein to a child's BW was calculated on the basis of body composition values from birth to age 10 years<sup>(46)</sup>, from ages 4 months to 2 years<sup>(47)</sup>, and from ages 4 to 18 years<sup>(48)</sup>.

For utilization efficiency, the results of 9–14-month-old infants (with a utilization efficiency of 70% and deposition efficiency of 40% for weight maintenance in 1-year-old infants)<sup>(36)</sup> were used. In addition, the deposition efficiency was considered as 40% throughout childhood, and the utilization efficiency for weight maintenance was considered to be close to the value for adults, in accordance with growth (90%).

Furthermore, values were rounded considering the importance of protein intake in children.

#### 2-2-4. Additional Amount for Pregnant Women

The body protein deposition during pregnancy can be calculated indirectly from an increase in body potassium. The average increase in body potassium in the late-stage of pregnancy is 2.08 mmol/day<sup>(49–52)</sup>. Using a potassium-to-nitrogen ratio of 2.15 mmol potassium/g nitrogen<sup>(49)</sup> and protein conversion factor of 6.25, the body protein deposition was calculated using the following formula.

$$\text{Protein deposition (g/day)} = \text{body potassium increment} \div 2.15 \times 6.25$$

When calculating body protein depositions, changes due to weight gain during pregnancy need to be taken into account. More specifically, the final weight gain was set at 11 kg<sup>(53)</sup>, and the weight gains of participants during pregnancy, in various studies, were corrected to find the increase in body potassium, in each study. Accordingly, the body protein deposition was calculated as shown in Table 4.

Based on a report which stated that the protein deposition ratio of the first, second and third trimesters was 0:1:3.9<sup>(52)</sup>, the total body protein deposition was found for the second and third trimesters if observations had been made during those trimesters in the report (280 days of pregnancy multiplied by 2/3) and the body protein deposition per day was calculated for each trimester after allocating depositions to the second and third trimesters using the simple ratio stated above.

The depositions calculated by the simple averaging of the values obtained from each study were 0 g/day in the early-stage, 1.94 g/day in the mid-stage, and 8.16 g/day in the late-

stage. The protein deposition efficiency was set at 43%<sup>(49)</sup>. Using these values, the additional amounts required during pregnancy (EAR) were set at  $0 \text{ g/day} \div 0.43 = 0 \text{ g/day}$  (rounded to 0 g/day) in the early-stage,  $1.94 \text{ g/day} \div 0.43 = 4.51 \text{ g/day}$  (rounded to 5 g/day) in the mid-stage, and  $8.16 \text{ g/day} \div 0.43 = 18.98 \text{ g/day}$  (rounded to 20 g/day) in the late-stage. The additional amount (RDA) was 0 g/day (rounded to 0 g/day) in the early-stage, 5.64 g/day (rounded to 10 g/day) in the mid-stage, and 23.73 g/day (rounded to 25 g/day) in the late-stage. The RDA values were obtained by multiplying the EAR and a RDA calculation coefficient of 1.25, assuming an interindividual coefficient of variation of 12.5%.

### **2-2-5. Additional Amount for Lactating Women (EAR, RDA)**

A substantial portion of the protein accumulated during pregnancy is lost during delivery. However, some of the protein accumulated in the body remains in the mother. Protein is also lost due to weight loss and lactation during the puerperal period. It was, therefore, decided that the amount of protein added is offset against pregnancy-related residual deposits of protein and residual weight gain. Thus, the amount of protein added during the lactation period is only the amount added for lactation.

The average lactation yield per day, given a child is only fed breast milk for the first 6 months until weaning, was set at  $0.78 \text{ L/day}$ <sup>(54–60)</sup>, and the average protein concentration of breast milk during this period was set at  $12.6 \text{ g/L}$ <sup>(55,56,61–66)</sup>. The conversion efficiency from dietary protein to breast milk protein was set at 70% on the basis of Report 1 by the FAO/WHO/UNU in 1985<sup>(12)</sup>. The additional amount required by lactating women (EAR) was rounded to 15 g/day using these values ( $12.6 \text{ g/L} \times 0.78 \text{ L/day} \div 0.70 = 14.04 \text{ g/day}$ ). The additional amount (RDA) was set at 17.6 g/day (rounded to 20 g/day) by multiplying the EAR and a RDA calculation coefficient of 1.25, assuming an interindividual coefficient of variation of 12.5%.

## **2-3. Methods Used to Set Adequate Intake**

### **2-3-1. Infants**

Since protein requirement cannot be determined using the nitrogen balance method in infants, as is done in adults, it is calculated from the amount of protein contained in the infant formulas and breast milk consumed by healthy infants. This requirement was, therefore, determined based on the concept of adequate intake (AI). Moreover, there is no scientific evidence on the utilization efficiency of protein from infant formulas. The setting of DRIs for protein in artificially fed infants was, therefore, postponed, and a reference value was provided instead.

When infants enter the weaning period, they start to consume protein from sources other than breast milk, and, consequently, a different calculation method of DRIs for protein is used. Therefore, AI was determined for infancy in 3 stages into which the infancy period was divided: 0–5 months, 6–8 months, and 9–11 months.

### 2-3-1-1. Infants Aged 0–5 Months

No studies have reported on the protein deficiency caused by lactation in infants aged 0–5 months. Therefore, the AI is calculated from the milk intake and the protein concentration of breast milk. In terms of the milk intake among infants, no clear differences were observed between the values in Japan and those in other countries, at 0.63–0.86 L/day<sup>(54–60)</sup>, so a mean of 0.78 L/day was used. The protein concentration of breast milk is also not considered to differ between races<sup>(55,57,61–66)</sup>. Thus, the average protein concentration of breast milk during this period was set at 12.6 g/L.

$$\text{AI (g/day)} = 12.6 \text{ (g/L)} \times 0.78 \text{ (L/day)} = 9.83$$

### 2-3-1-2. Infants Aged 6–8 Months

Once infants enter the weaning period, their nutrient intake greatly changes. The protein intake from baby foods, in infants aged 6–8 months, was estimated at 6.1 g/day, based on studies in Japanese people<sup>(67)</sup>. Meanwhile, the average milk intake of infants during this period was set at 0.60 L/day<sup>(56,57)</sup>, and the protein concentration of breast milk was set at 10.6 L/day<sup>(56,61,63)</sup>. The AI of protein from breast milk, and sources other than breast milk can, therefore, be calculated as follows:

$$\text{AI (g/day)} = \text{protein concentration of breast milk} \times \text{average milk intake} + \text{amount of protein from baby foods other than breast milk} = 10.6 \text{ (g/L)} \times 0.60 \text{ (L/day)} + 6.1 \text{ (g/day)} = 12.5$$

### 2-3-1-3. Infants Aged 9–11 Months

The protein intake from baby foods, in infants aged 9–11 months, was estimated at 17.9 g/day, based on studies in Japanese people<sup>(67,68)</sup>. Meanwhile, the average milk volume of infants during this period was set at 0.45 L/day<sup>(56,57)</sup>, and the protein concentration of breast milk was set at 9.2 L/day<sup>(56,61–63)</sup>. Therefore, the AI of protein from breast milk, and sources other than breast milk can be calculated as follows.

$$\text{AI (g/day)} = \text{protein concentration of breast milk} \times \text{average milk intake} + \text{amount of protein from baby foods other than breast milk} = 9.2 \text{ (g/L)} \times 0.45 \text{ (L/day)} + 17.9 \text{ (g/day)} = 22.0$$

### 2-3-1-4. Artificially fed Infants

The DRIs for protein, in artificially-fed infants, was provided as a reference value, taking into account the utilization efficiency of protein from infant formulas. The AI reference value for artificially fed infants was calculated as follows, assuming the utilization efficiency of protein from infant formulas to be 70% of that of breast milk<sup>(12)</sup>.

$$\text{0–5 months (g/day): } 12.6 \text{ (g/L)} \times 0.78 \text{ (L/day)} \div 0.70 = 14.0$$

$$\text{6–8 months (g/day): } 10.6 \text{ (g/L)} \times 0.60 \text{ (L/day)} \div 0.70 + 6.1 \text{ (g/day)} = 15.2$$

$$\text{9–11 months (g/day): } 9.2 \text{ (g/L)} \times 0.45 \text{ (L/day)} \div 0.70 + 17.9 \text{ (g/day)} = 23.8$$

### **3. Avoiding Excessive Intake**

#### **3-1. Determining the UL (Tolerable Upper Intake Level)**

The UL of protein must be determined based on the adverse health events caused by the excessive intake of protein. However, at present, there are an insufficient number of reports providing clear evidence on the determination of the UL of protein. Therefore, a UL for protein was not set.

#### **4. Preventing the Development and Progression of Life-style Related Diseases (LRDs)**

The development and progression of LRDs (hypertension, dyslipidemia, diabetes, and chronic kidney disease) are the result of the interaction between environmental factors (lifestyle) and genetic ones. Providing optimal nutrition may be highly significant in preventing the development or progression of these LRDs.

#### **4-2. Methods Used to Set the DG (Tentative Dietary Goal for Preventing LRDs)**

The methods used to set the DG are summarized in “Energy Providing Nutrients’ Balance.”

**DRIs for Proteins**

(EAR, RDA, AI: g/day, DG (median): % energy)

Gender	Males				Females			
Age etc.	EAR	RDA	AI	DG <sup>1</sup> (Median <sup>2</sup> )	EAR	RDA	AI	DG <sup>1</sup> (Median <sup>2</sup> )
0-5 months *	—	—	10	—	—	—	10	—
6-8 months *	—	—	15	—	—	—	15	—
9-11 months *	—	—	25	—	—	—	25	—
1-2 years	15	20	—	13-20(16.5)	15	20	—	13-20(16.5)
3-5 years	20	25	—	13-20(16.5)	20	25	—	13-20(16.5)
6-7 years	25	35	—	13-20(16.5)	25	30	—	13-20(16.5)
8-9 years	35	40	—	13-20(16.5)	30	40	—	13-20(16.5)
10-11 years	40	50	—	13-20(16.5)	40	50	—	13-20(16.5)
12-14 years	50	60	—	13-20(16.5)	45	55	—	13-20(16.5)
15-17 years	50	65	—	13-20(16.5)	45	55	—	13-20(16.5)
18-29 years	50	60	—	13-20(16.5)	40	50	—	13-20(16.5)
30-49 years	50	60	—	13-20(16.5)	40	50	—	13-20(16.5)
50-69 years	50	60	—	13-20(16.5)	40	50	—	13-20(16.5)
70+ years	50	60	—	13-20(16.5)	40	50	—	13-20(16.5)
Pregnant women (additional)	/				+0	+0		
Early-stage					+5	+10	—	—
Mid-stage					+20	+25		
Late-stage								
Lactating women (additional)	/				+15	+20	—	—

\* AIs for infants are values for breast-fed children.

<sup>1</sup> Ranges are expressed as approximate values.

<sup>2</sup> Medians indicate the median values for the given range. They do not indicate most desirable values.

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## Dietary Fat

### 1. Background Information and Definitions

#### 1-1. Definitions and Classifications

Fats are compounds that are insoluble in water but soluble in organic solvents<sup>(1)</sup>. The nutritionally important fats include fatty acids, neutral fat, phospholipids, glycolipids, and sterols. Fatty acids consist of a carboxyl group at the end of a hydrocarbon chain (composed solely of hydrogen and carbon), and have a total number of carbon atoms ranging from 4 to 36. The presence of a carboxyl group allows fatty acids to be metabolized *in vivo* and used as an energy source or cell membrane component. Fatty acids can be saturated - without a double bond between carbon atoms, monounsaturated - with just one double bond in the fatty acid chain, and polyunsaturated - with more than one double bond in the fatty acid chain. Polyunsaturated fatty acids are further differentiated into n-3 fatty acids (third carbon atom from the methyl end) and n-6 fatty acids (sixth carbon atom from the methyl end), based on the position of the first double bond from the methyl end. Unsaturated fatty acids with double bonds are geometric isomers, which are classified into trans and cis isomers. A majority of the unsaturated fatty acids that exist in the natural world are cis isomers; only a few trans isomers exist. Neutral fat can be formed of one, two, or three fatty acids combined with glyceride to form a monoacylglycerol, diacylglycerol, or triacylglycerol (triglyceride, triglycerol, neutral fat). Phospholipids are lipids containing phosphoric acid attached with one or two ester bonds. Glycolipids are lipids in which one or more monosaccharides are attached to a lipid moiety with a glycosidic bond.

Cholesterol is an amphiphilic molecule with a hydrocarbon chain and steroid skeleton composed of a ring of four carbons. We examined dietary cholesterol as a dietary fat.

### 2. DRIs for Dietary Fat

#### 2-1. Characteristics of Reference Setting

The DRIs for total fat, saturated fatty acids, n-6 fatty acids, and n-3 fatty acids were established. The primary role of the macronutrients (fat, carbohydrates, and proteins) is to supply energy to the cells. If the body weight, and physical activity level (PAL) do not change, the energy intake remains largely within a fixed range. Therefore, when the fat intake increases (or decreases), the intake of carbohydrates decreases (or increases). Therefore, the DRIs for fat need to be set taking into account carbohydrate and protein intakes. This is why the DRIs for fat are shown as a percentage of total energy intake, i.e., the energy ratio (%energy: %E), for the tentative dietary goals (DG) for the prevention of lifestyle-related diseases (LRDs) of individuals aged 1 year or older. The DG of infants is shown as %E. Saturated fatty acids are also shown as %E from the standpoint of preventing LRDs. However, the DGs for n-6 and n-3 fatty acids, which are essential fatty acids, are shown as absolute amounts (g/day) not influenced by total energy intake. When body weight correction is necessary, the reference body weight of

each sex and age group was used. The n-3 fatty acids examined were alpha-linoleic acid derived from edible cooking oil, eicosapentaenoic acid (EPA), and docosahexaenoic acid (DHA) derived from fish.

## **2-2. Total Fat (Total Fat Energy Ratio)**

### **2-2-1. Basic Matters and Intakes**

#### **2-2-1-1. Basic Matters**

In the DRIs for fat, the total fat energy ratio was set as the DG for those aged 1 year or older, and adequate intake (AI) for infants.

#### **2-2-1-2. Intakes**

The median dietary fat intake ratio of Japanese people aged 30–49 years, based on the results of the 2010 and 2011 National Health and Nutrition Survey (NHNS)<sup>(2)</sup>, is 25.8%E in men and 28.3%E in women. The fat energy ratio decreases with age. Meanwhile, the median total energy intake of Japanese people aged 30–49 years is 2,078 kcal in men and 1,635 kcal in women. Based on the United States Department of Agriculture's *Continuing Survey of Food Intakes by Individuals* (CSFII, 1994–1996, 1998), the median ratio of fat in those aged 31–50 years is 33.7%E in men and 32.8%E in women. Therefore, the fat energy intake of Japanese people is lower than that of Americans.

### **2-2-2. To Avoid Inadequacy**

#### **2-2-2-1. Infants (Methods Used to Set the AI)**

Breast milk was considered to be an ideal source of nutrients for infants. Therefore, the total fat energy ratio of breast milk<sup>(3,4)</sup> and the average milk intake was calculated (0.78 L/day)<sup>(5,6)</sup> and set as the AI. Infants aged 0–5 months obtain their nutrients from breast milk (or infant formula), but from the age of 6 months, they begin taking baby food, and between ages 6 and 11 months, they obtain their nutrients from both breast milk (or infant formula) and baby food. This period is regarded as the transition period to early childhood, so the average AIs (medians) of 0–5-month-old infants and 1–2-year-old infants were used.

The fat concentration of breast milk consumed by 0–5-month-old infants is 3.5 g/100 g, and, thus, the energy derived from fat in 100 g of breast milk is  $3.5 \text{ g} \times 9 \text{ kcal} = 31.5 \text{ kcal}/100 \text{ g}$ . The total energy in 100 g of breast milk is 65 kcal, which yields a fat energy ratio of 48.46%, as shown below. The AI was rounded to be set at 50%E.

**Fat energy ratio (%E) =  $31.5/65 = 48.46\%E$**

In addition, the fat intake, per day, of infants aged 0–5 months is 27.8 g/day when the fat concentration of the breast milk of Japanese women (35.6 g/L) and the average milk intake (0.78 L/day) are multiplied.

The average AI of infants aged 0–5 months, as obtained from the above formula and

median (male and female average) intake from the 2010 and 2011 NHNS of infants aged 1–2 years, was taken for infants aged 6–11 months. When calculated as follows, a fat energy ratio of 37.9%E was obtained, which was rounded to an AI of 40%E.

**Fat energy ratio (%E) =  $[48.46 + (27.2 + 27.6) / 2] / 2 = 37.9\%E$**

Furthermore, the fat intake per day of infants aged 6–11 months was 29.1 g. This was obtained by taking the average of the fat intake of infants aged 0–5 months (27.8 g/day) and the median (male and female average) intake from the 2010 and 2011 NHNS of infants aged 1–2 years.

### **2-2-3. For the Prevention of the Development and Progression of LRDs**

#### **2-2-3-1. Association with LRDs**

Intake of a low-fat/high-carbohydrate diet increases postprandial blood glucose levels and fasting triacylglyceride levels (neutral fat), and decreases blood high-density lipoprotein (HDL) cholesterol levels<sup>(7,8)</sup>. No studies have shown that this kind of a diet in healthy individuals increases the risk of arteriosclerosis, obesity, or diabetes; however, if these patterns of blood lipid levels persist over a long period of time, the risk of coronary heart disease increases. The data of a large number of interventional studies were reviewed in the US-Canada DRIs<sup>(7)</sup>. These data were used to conduct a regression analysis of the associations between fat or carbohydrate energy ratio, blood HDL cholesterol, total cholesterol/HDL cholesterol, and triglycerides. As a result, it was determined that, to achieve an appropriate blood concentration of these nutrients, a dietary fat energy ratio of at least 20%E is optimal. Additionally, extremely low-fat diet may impair the absorption of fat-soluble vitamins (particularly vitamins A and E)<sup>(9)</sup>, and since there is a positive correlation between the fat and protein contents of food, the intake of sufficient protein may be made difficult. Fat has the highest energy density, and thus it is assumed that the intake of energy is unlikely to be adequate when the intake of fat is low. Therefore, an intake of at least 10–15%E is considered appropriate for adults<sup>(10)</sup>.

Unlike a low-fat/high-carbohydrate diet, a high-fat/low-carbohydrate diet increases HDL cholesterol levels, and decreases fasting triacylglyceride levels. However, low-density lipoprotein (LDL) cholesterol levels, postprandial free fatty acid levels, and postprandial triglyceride levels increase<sup>(11,12)</sup>. Furthermore, it is of concern that the risk of total mortality and type 2 diabetes may increase, as taking a high-fat/low-carbohydrate diet results in an inadequate intake of minerals contained in cereals, and a higher intake of protein<sup>(13)</sup>.

In a meta-analysis of cohort studies published in 2013<sup>(14)</sup>, the consumption of diets with a large intake of fat, compared to carbohydrates, increased the likelihood of total mortality 1.3-fold. Other cohort studies suggested that total mortality may be influenced by the type of lipid. The Nurses' Health Study and Health Professionals' Follow-up Study found that total mortality increased in groups with a high intake of animal-derived foods, whereas the total mortality was decreased in groups with a high intake of plant-derived foods<sup>(15)</sup>. However, a meta-analysis of interventional studies over more than six months (excluding studies on n-3

fatty acids)<sup>(16)</sup>, did not reveal a decrease in total mortality or the prevalence of cardiovascular disease even when total lipid intake was reduced, which was different from the results of the above-mentioned cohort studies.

With regards to the prevention of obesity, a meta-analysis (including 33 interventional studies) conducted in 2012, targeting primarily non-obese individuals, demonstrated that the body weight decreased when the total fat intake reduced<sup>(17)</sup>. Specifically, the body weight decreased by 0.19 kg for every 1%E reduction in the total fat intake. However, it should be noted that a low-carbohydrate diet (fat: 30%E, carbohydrates: 40%E) had a stronger effect on weight loss than a low-fat diet (fat: 20%E, carbohydrates: 55–60%E) in a group of obese individuals with high blood insulin levels and strong insulin resistance<sup>(18)</sup>. In populations with a small number of obese individuals, such as those in Japan, there is concern that the risk of obesity, metabolic syndrome, diabetes, and coronary heart disease may increase when the fat energy ratio rises. A large-scale interventional study in postmenopausal women found that the development of diabetes was significantly reduced when a decrease in the total fat intake and weight loss were observed<sup>(19)</sup>. A high-fat diet increases the intake of saturated fatty acids, and saturated fatty acids raise serum LDL cholesterol levels, which further increase the risk of coronary heart disease. For this reason, the U.S. National Cholesterol Education Program Step I Diet and Step II diets stipulate that a fat energy ratio less than 30% is appropriate<sup>(20)</sup>. According to a report on a meta-analysis of 37 interventional studies that assessed this National Cholesterol Education Program<sup>(21)</sup>, a lipid energy intake of less than 30% resulted in decreased serum total cholesterol, LDL cholesterol, triglyceride and total cholesterol/HDL cholesterol levels, and body weight.

### **2-2-3-2. Children and Adults (Methods Used to Set the DG)**

The DG for saturated fatty acids was set at 7%E or less. The median (AI) intakes of n-6 and n-3 fatty acids in Japanese people are 4–5%E and approximately 1%E, respectively, and the median intake of monounsaturated fatty acids is at least 6%E<sup>(2)</sup>, leading to a fatty acid total of 18–19%E. Triacylglycerides and phospholipids contain glycerol in addition to fatty acids, and account for approximately 10% of all fat. Taking into account glycerol moieties, the lipid energy ratio is 20%E ( $= 18 \div 0.9$ ) to 21%E ( $= 19 \div 0.9$ ). This was rounded to 20%E to be set as the value below the DG range.

Moreover, considering obesity, diabetes prevention, and mortality (reports of cohort studies), an energy ratio less than 30%, which is viewed as a low fat energy ratio in Western countries, is recommended. For this reason, the lipid energy ratio was set at 30%E, which is above the DG. In the US-Canada DRIs<sup>(7)</sup>, because 30%E is considered difficult to achieve in typical individuals due to the current intake status, the value is set at 35%E instead.

## **2-3. Saturated Fatty Acids**

### **2-3-1. Basic Matters and Intake Status**

#### **2-3-1-1. Basic Matters**

Saturated fatty acids include caprylic acid (8:0), capric acid (10:0), lauric acid (12:0), myristic acid (14:0), palmitic acid (16:0), and stearic acid (18:0). Saturated fatty acids are consumed from foods, and can be synthesized from acetyl coenzyme A (CoA) which is an intermediate metabolite of carbohydrate and protein. Therefore, no estimated average requirement (EAR), recommended dietary allowance (RDA), or AI can be set. However, saturated fatty acids are an important source of energy, and are necessary to maintain an appropriate energy ratio and ensure an appropriate fatty acid intake ratio. In addition, interventional studies suggest that lowering the intake of saturated fatty acids can reduce the risk of myocardial infarction. Therefore, a DG was set accordingly.

#### **2-3-1-2. Dietary Intake Status**

The median intakes of Japanese people aged 30–49 years, based on the results of the 2010 and 2011 NHNS<sup>(2)</sup>, are 15.2 g/day and 13.8 g/day, respectively, and the energy ratios are 6.6%E and 7.6%E in men and women, respectively. The median intakes in Americans aged 31–50 years are 31.4 g/day and 20.3 g/day, and the energy ratios are 11.4%E and 11.0%E in men and women, respectively<sup>(7)</sup>. Therefore, the intake of Japanese people is approximately 40% lower than that of Americans, in terms of the energy ratio.

### **2-3-2. For the Prevention of the Development of LRDs**

#### **2-3-2-1. Association with LRDs**

A meta-analysis of cohort studies found that, when saturated fatty acids were substituted with polyunsaturated fatty acids, the hazard ratio for coronary heart disease decreased to 0.87; however, when monounsaturated fatty acids or carbohydrates were substituted, the ratios increased to 1.19 or 1.07, respectively. The lack of a strong correlation between saturated fatty acid intake and myocardial infarction may be attributed to the fact that different effects may be induced depending on the type of saturated fatty acid, and the risk of coronary heart disease varies depending on the intake of foods containing saturated fatty acids<sup>(22)</sup>. The intake of saturated fatty acid derived from dairy products prevents the development of cardiovascular disease, whereas the intake of saturated fatty acid derived from meats is a risk factor for cardiovascular disease<sup>(23)</sup>. A cohort study in Japanese people aged 45–74 years (the Japan Public Health Center-based Prospective [JPHC] study)<sup>(24)</sup>, found a positive correlation between saturated fatty acid intake and myocardial infarction. The hazard ratio for myocardial infarction increased to 1.24 in the intermediate quintile group (saturated fatty acid intake: 16.3 g/day, 7.2%E) and 1.39 in the largest quintile group (saturated fatty acid intake: 24.9 g/day, 10.9%E), compared to the lowest quintile group (saturated fatty acid intake: 9.6 g/day, 4.4%E). Many interventional studies in Western countries have shown that reducing

saturated fatty acid intake lowers the prevalence of coronary heart disease, the degree of arteriosclerosis, and LDL cholesterol levels. For example, a meta-analysis that pooled interventional studies (including primary and secondary prevention) found that, when saturated fatty acids are substituted with polyunsaturated fatty acids, and polyunsaturated fatty acid intake (including both n-6 and n-3 fatty acids) is increased to an average of 14.9%E, the relative risk of myocardial infarction (including death) decreases by 19%, compared to the control group (average polyunsaturated fatty acid intake of 5.0%E).

Pertaining to the relationship between diabetes and obesity, observational studies have demonstrated a positive association between diabetes and saturated fatty acid intake; however, these studies did not find any association between diabetes and saturated fatty acid intake when adjusted for body mass index (BMI)<sup>(25,26)</sup>. However, cross-sectional studies that examined the relationship between saturated fatty acid intake and insulin resistance, which is a cause of diabetes, found a positive correlation between saturated fatty acid intake and insulin resistance even after BMI adjustment<sup>(27-29)</sup>. Interventional studies have also revealed that insulin resistance occurs when a diet high in saturated fatty acids is consumed<sup>(30,31)</sup>. Interventional studies comparing monounsaturated fatty acids demonstrated that insulin sensitivity decreases, and insulin secretion increases when the saturated fatty acid intake is increased<sup>(30,32)</sup>. Therefore, these results suggest that an increased intake of saturated fatty acids causes obesity and insulin resistance (independent of obesity), thereby increasing the risk of diabetes.

Meanwhile, cohort studies in Japanese people revealed that the risk of stroke, particularly cerebral hemorrhage-related mortality or morbidity, increased in those with a low saturated fatty acid intake<sup>(24,33-36)</sup>. The recently published JPHC study found that there is a negative linear correlation between saturated fatty acid intake and cerebral hemorrhage or lacunar infarction, whereby the risks of cerebral hemorrhage and lacunar infarction are reduced as the saturated fatty acid intake increases<sup>(24)</sup>. The hazard ratio for cerebral hemorrhage increased to 0.84 in the intermediate quintile group (saturated fatty acid intake: 16.3 g/day, 7.2%E) and 0.61 in the largest quintile group (saturated fatty acid intake: 24.9 g/day, 10.9%E), compared to the lowest quintile group (saturated fatty acid intake: 9.6 g/day, 4.4%E).

However, animal experiments have not shown that cerebral hemorrhage can be prevented by an increased saturated fatty acid intake<sup>(37)</sup>. Therefore, it is not known if decreases in saturated fatty acid intake may increase the risk of cerebral hemorrhage. Animal protein intake adjustments are insufficient in cohort studies and increased morbidity from diseases such as cerebral hemorrhage may be caused by decreased animal protein intake associated with a reduced saturated fatty acid intake. In fact, a meta-analysis that examined the relationship between the intake of dairy products and stroke found that the relative risk of cerebral hemorrhage decreased to 0.75 in the group with the largest intake of dairy products, compared to the group with the lowest intake<sup>(38)</sup>.

### **2-3-2-2. Adults (Methods Used to Set the DG)**

A positive correlation between saturated fatty acid intake and serum (or plasma) total cholesterol concentration has long been known from Keys's<sup>(39)</sup> and Hedsted's<sup>(40)</sup> formulas. In addition, a meta-analysis of 27 interventional studies revealed results that are very similar to those of another meta-analysis of a large number of studies<sup>(8,41)</sup>. This also applies to LDL cholesterol levels<sup>(8,41)</sup>. However, a meta-analysis that examined serum cholesterol levels according to the number of carbon atoms in saturated fatty acids found significant rises in the levels of lauric acid, myristic acid, and palmitic acid (12–16 carbon atoms), but observed no significant change in the stearic acid levels (18 carbon atoms)<sup>(8)</sup>. It has been pointed out that even among saturated fatty acids, the effect on serum cholesterol levels differs according to the number of carbon atoms in them. Therefore, an excessive intake of saturated fatty acids (all saturated fatty acids, regardless of the number of carbon atoms) is assumed to be a risk factor for arteriosclerosis, and, particularly, myocardial infarction. Nevertheless, a meta-analysis summarizing the results of 21 cohort studies (16 that examined the incidence of myocardial infarction) that examined the relationship between saturated fatty acid intake and the incidence of cardiovascular disease found no significant correlation with myocardial infarction<sup>(42)</sup>. However, serum total cholesterol levels were adjusted in seven of these studies; it has been suggested that over-adjustment may apply to this case at the time of statistical calculation, and, therefore, the relationship between saturated fatty acid intake and cardiovascular disease may not be correctly assessed<sup>(43)</sup>. Two cohort studies were performed in Japanese people: one observed no significant correlation with myocardial infarction mortality<sup>(36)</sup>, while the other observed a significant positive correlation with the incidence of myocardial infarction<sup>(24)</sup>. Incidentally, according to a pooled analysis examining the data of 11 cohort studies on the difference in the risk of myocardial infarction or death when saturated fatty acids, accounting for a fixed total energy intake of 5%E, are switched to the respective amount of fatty acids or carbohydrates, both the incidence of and mortality associated with myocardial infarction are significantly decreased when saturated fatty acids are substituted with polyunsaturated fatty acids<sup>(44)</sup>.

This series of results suggests that, to prevent both the development and progression of arteriosclerosis, particularly myocardial infarction, it is important to not only restrict the intake of saturated fatty acids, but also to simultaneously increase the intake of polyunsaturated fatty acids.

Considering these reports and the feasibility of the amount of and improvement in each citizen's intake, the ideal intake in adults in each country is set at less than 10%E<sup>(45)</sup>. In addition, the American Heart Association (2006 and 2009) and American Diabetes Association (2008) set a ratio of less than 7%E<sup>(45)</sup>. Furthermore, in some cases, guidelines are limited to a qualitative description stating the ratio should be kept "as low as possible" without providing a specific value<sup>(45)</sup>. The saturated fatty acid intake in Japanese people is relatively low compared to that of people in Western countries. The intake was approximately 7.3%E in all participants in the 2011 NHNS (calculated from the average energy intake [1,840 kcal] and average

saturated fatty acid intake [14.85 g]), and 6.9%E when the population was limited to those aged 20 years and older. The health benefits of consuming an amount of saturated fatty acids that is higher than the above-stated value is not clear, with the exception of a possible reduction in the risk of stroke.

With regards to the reduction in the risk of stroke, many cohort studies in Japanese people found that those with a low saturated fatty acid intake are at an increased risk of death or morbidity from stroke, particularly cerebral hemorrhage<sup>(24,33–36)</sup>. However, it is not clear if an increased saturated fatty acid intake can prevent cerebral hemorrhage<sup>(37)</sup>. For this reason, it has not been elucidated whether the correlations in the above-mentioned cohort studies are due to saturated fatty acid intake, or due to the intakes of other nutrients or living habits that show a correlation with saturated fatty acid intake. A negative correlation was demonstrated between saturated fatty acid intake and cerebral hemorrhage in a meta-analysis, but it was not significant<sup>(42)</sup>. There are few reliable data indicating that a low saturated fatty acid intake is a direct risk factor for some LRDs and other diseases. Based on these findings, a reference value, which indicates an enhanced risk of LRDs if the value falls below it, has not been set in the DRIs of other countries or similar guidelines<sup>(45)</sup>. Furthermore, restricting saturated fatty acids intake can lead to the restriction of total fat intake, resulting in an inadequate intake of essential fatty acids. Therefore, caution must be exercised in setting DRIs.

Based on the facts mentioned above, the DG for saturated fatty acids was set at 7%E or less.

### **2-3-2-3. Children (Methods Used to Set the DG)**

Arteriosclerosis has long been known to manifest in childhood, progress throughout early adulthood, and cause coronary heart disease from middle age<sup>(46)</sup>. Several cohort studies in Western countries reported that carotid artery intima-media thickening increases when individuals with high LDL cholesterol levels during childhood (age 4–18 years) enter adulthood (age 18–42 years)<sup>(47–49)</sup>. Low saturated fatty acid intakes during childhood decrease childhood LDL cholesterol levels<sup>(50–52)</sup>. Meanwhile, an excessive intake of saturated fatty acids during childhood can cause coronary heart disease and obesity in middle age. Therefore, a DG of 7%E or less for saturated fatty acids is considered best even during childhood. However, there are only a few descriptive epidemiological studies focusing on the intakes and sources of saturated fatty acids during childhood, studies examining the relationship between saturated fatty acid intake during childhood and arteriosclerosis-related diseases in adulthood, and studies examining the safety of reducing saturated fatty acid intake during childhood (in terms of growth impairment, etc.). Therefore, it was decided not to set a DG for children at this time.

## 2-4. n-6 Fatty Acids

### 2-4-1. Basic Matters and Intake Status

#### 2-4-1-1. Background Information

Linoleic acid (18:2n-6), gamma-linoleic acid (18:3n-6), and arachidonic acid (20:4n-6) are some n-6 fatty acids. Gamma-linoleic acid and arachidonic acid are metabolites of linoleic acid. Since n-6 fatty acids cannot be synthesized from acetyl-CoA *in vivo*, they must be consumed orally. Ninety-eight percent of the n-6 fatty acids consumed by Japanese people is linoleic acid. Few studies have investigated the effect of the consumption of gamma-linoleic acid or arachidonic acid alone on the human body.

#### 2-4-1-2. Dietary Intake Status

Based on the results of the 2010 and 2011 NHNS<sup>(2)</sup>, the median n-6 fatty acid intake of Japanese people aged 30–49 years is 10.0 g/day in men and 8.4 g/day in women, and the energy ratio is 4.3%E in men and 4.6%E in women. In Americans aged 31–50 years, the median linoleic acid intake is 16.1 g/day in men and 11.1 g/day in women, and the energy ratio is 5.9%E in men and 6.0%E in women<sup>(7)</sup>. Therefore, the linoleic acid intake of Japanese people is about 30% lower than that of Americans in terms of energy ratio.

### 2-4-2. To Avoid Inadequacy

#### 2-4-2-1. Factors to be Considered in Estimating Requirements

A deficiency of n-6 fatty acids is observed in those who receive total parenteral nutrition, and the deficiency disappears on administering 7.4–8.0 g/day or 2%E of linoleic acid<sup>(53–56)</sup>. However, no data are available for the setting of the EAR of healthy adults. No studies have reported on conditions such as dermatitis which are thought to be caused by n-6 fatty acid deficiency in healthy adults who lead free daily lives. A DG for n-6 fatty acids was set in light of the possible need for n-6 fatty acids other than linoleic acid.

#### 2-4-2-2. Methods of Determining AI

##### (1) Infants

Breast milk was considered an ideal source of nutrients for infants; therefore, the AI was determined from the fat content of breast milk<sup>(3,4)</sup> and the average milk volume (0.78 L/day)<sup>(5,6)</sup>. Infants aged 0–5 months obtain their nutrients from breast milk (or infant formula). From the age of 6 months, infants begin taking baby foods. Thus, infants aged 6–11 months obtain their nutrients from both breast milk (or infant formula) and baby food. This is regarded as the transition period to early childhood, so the average AIs (medians) of 0–5-month-old infants and 1–2-year-old infants were used.

The AI of infants aged 0–5 months was found by multiplying the n-6 fatty acid concentration of breast milk (5.16 g/L) and the average milk intake (0.78 L/day).

**n-6 fatty acids: AI (g/day) = 5.16 g/L × 0.78 L/day = 4.02 g/day**

For infants aged 6–11 months, the average AI of infants aged 0–5 months and median (male and female average) intake from the 2010 and 2011 NHNS<sup>(2)</sup> of infants aged 1–2 years were taken. The AI was calculated as follows:

**n-6 fatty acids: AI (g/day) = [4.0 + (4.7 + 4.5) /2] /2 = 4.3 g/day**

## **(2) Children and Adults**

The median n-6 fatty acid intake, calculated from the results of the 2010 and 2011 NHNS, was set as the AI for those aged 1 year and older.

## **(3) Pregnant and Lactating Women**

The median n-6 fatty acid intake of pregnant women, calculated from the results of the 2007 to 2011 NHNS<sup>(57)</sup>, is 9 g/day. Thus, the AI was set at 9 g/day because it was considered a value that would not cause fetal developmental problems.

Lactating women are assumed to secrete breast milk containing the average fat components of the breast milk of Japanese women. The median n-6 fatty acid intake of lactating women, calculated from the results of the 2007 to 2011 NHNS<sup>(57)</sup>, is 9 g/day. This value has not been found to cause essential fatty acid deficiencies in a majority of lactating women, and is considered the amount that can be secreted of breast milk containing sufficient n-6 fatty acids. The AI was, therefore, set at 9 g/day.

## **2-4-3. Preventing the Development of LRDs**

### **2-4-3-1. Association with LRDs**

Studies on coronary heart disease include an interventional study that compared blood lipids in Western countries. This interventional study found that LDL cholesterol decreased the most when polyunsaturated fatty acids (primarily n-6 fatty acids), instead of carbohydrates, were consumed compared to when other fats were consumed instead of carbohydrates<sup>(8)</sup>. LDL cholesterol levels also decrease when n-6 fatty acids are consumed instead of saturated fatty acids<sup>(58)</sup>. However, the results of observational studies with coronary heart disease as an endpoint are inconsistent<sup>(59,60)</sup>. In the Nurses' Health Study<sup>64</sup> the largest quintile for linoleic acid intake (7.0%E) had the lowest risk of coronary heart disease; however, recent studies found no correlation with n-6 fatty acid intake<sup>(61–64)</sup>. Many interventional studies have found that the prevalence of coronary heart disease is reduced when saturated fatty acids are substituted with polyunsaturated fatty acids, although no interventional studies have substituted proteins or carbohydrates with polyunsaturated fatty acids. Therefore, it is not clear whether this decrease in the prevalence of coronary heart disease is due to a reduced intake of saturated fatty acids or an increased intake of polyunsaturated fatty acids<sup>(65)</sup>. A meta-analysis of interventional studies published in 2013 (in both healthy individuals and post-myocardial infarction patients) analyzed the effects of n-3 and n-6 fatty acids separately<sup>(66)</sup>. This analysis revealed that a mixed intake of n-3 and n-6 fatty acids reduced the rate of death from myocardial infarction by 19%, while the intake of linoleic acid alone increased mortality by 33%. Meanwhile, a meta-analysis of interventional studies published in 2010 (in both healthy individuals and post-myocardial

infarction patients)<sup>(67)</sup> analyzed nonfatal myocardial infarction, as well as fatal cases and found that a mixed intake of n-3 and n-6 fatty acids reduced the risk of myocardial infarction by 22%, whereas an intake of n-6 fatty acids alone increased this risk by 13%.

A prospective nested case control study of stroke in Japanese people compared a group with a serum lipid linoleic acid ratio of 34% (linoleic acid intake equivalent to approximately 13.3 g/day) with a group with a ratio of 22% (linoleic acid intake equivalent to approximately 9.5 g/day), and found that the odds ratio for stroke decreased to 0.43. However, some cohort studies that examined n-6 fatty acid intake and stroke prevalence did not demonstrate a correlation<sup>(68,69)</sup>.

Although the Nurses' Health Study<sup>(70)</sup> found a weak negative correlation between vegetable oil intake and diabetes, the types of fat contained in vegetable oil were not clarified. A recent study did not show any correlation between n-6 fatty acid intake and diabetes<sup>(71)</sup>.

Pertaining to cancer, a recent cohort study<sup>(72)</sup> and some case control studies<sup>(73,74)</sup> revealed a positive correlation between n-6 fatty acid intake and breast cancer<sup>(75)</sup>.

Linoleic acid is more easily oxidized than oleic acid, which is a monounsaturated acid. The risks of consuming large quantities (10%E or higher) of linoleic acid have not been elucidated<sup>(7)</sup>. As linoleic acid produces prostaglandin and leukotriene<sup>(76)</sup>, which cause inflammation, the safety of consuming it in large quantities is a matter of concern. The increased breast cancer and myocardial infarction morbidities associated with an excessive intake of linoleic acid may be attributed to the ease of oxidation and the inflammatory action of linoleic acid.

However, despite the above-stated risks associated with excessive n-6 fatty acid intake, no DG was set due to the lack of studies in Japanese people.

## **2-5. n-3 Fatty Acids**

### **2-5-1. Background Information and Intake Status**

#### **2-5-1-1. Background Information**

Alpha-linoleic acid (18:3n-3) derived from edible cooking oil, and EPA (20:5n-3), docosapentaenoic acid (DPA; 22:5n-3) and DHA (22:6n-3) derived from fish are some n-3 fatty acids. A small amount of alpha-linoleic acid is converted to EPA and DHA in the body.

These fats cannot be synthesized *in vivo*, and deficiencies result in conditions such as dermatitis<sup>(77,78)</sup>. For this reason, an AI was set.

Apart from the physiological actions of n-3 fatty acids competing with those of n-6 fatty acids, as it is thought that they may also have independent actions, reference intakes were set for n-3 fatty acids themselves as opposed to a ratio of n-3 to n-6 fatty acids. Epidemiological studies also support this approach. The Nurses' Health Study<sup>(79)</sup> of women found that the preventive action of alpha-linoleic acid against coronary heart disease is not influenced by linoleic acid intake. In addition, the Health Professional Study<sup>(80)</sup> of men demonstrated that the preventive action of alpha-linoleic acid or EPA and DHA against coronary heart disease was

not influenced by n-6 fatty acid intake, either.

Moreover, given the environmental contaminants contained in fish, such as mercury and dioxins, and the global shortage of fish resources, the intake of alpha-linoleic acid will become difficult and different sources must be considered in the future. This is why alpha-linoleic acid and fish-derived n-3 fatty acids were both investigated. The AI was set from the standpoint of preventing deficiencies, although it is difficult to distinguish alpha-linoleic acid from fish oil. Reference intakes were, therefore, set for the n-3 fatty acids contained in both alpha-linoleic acid and fish oil. Since many studies have used EPA and DHA intakes in their epidemiological data, EPA and DHA intakes were examined together to find the fish-derived n-3 fatty acid intake.

### **2-5-1-2. Dietary Intake Status**

The median n-3 fatty acid intake of Japanese people aged 30–49 years, based on the results of the 2010 and 2011 NHNS<sup>(2)</sup>, was 2.1 g/day in men and 1.6 g/day in women, and the energy ratio was 0.89%E in men and 0.86%E in women. In Americans aged 31–50 years, the median n-3 fatty acid intake is 1.8 g/day in men and 1.2 g/day in women, and the energy ratio is 0.64%E in men and 0.66%E in women<sup>(7)</sup>. The n-3 fatty acid intake of Japanese people is; therefore, approximately 1.3-fold higher than that of Americans, in terms of energy ratio.

According to calculations based on the results of the 2005 and 2006 NHNS used in the DRIs (2010), the median EPA and DHA intake of Japanese people aged 30–49 years is 0.32 g/day in men and 0.23 g/day in women, and the energy ratio is 0.14%E in men and 0.12%E in women. In Americans aged 31–50 years, the median EPA and DHA intake is 0.086 g/day in men and 0.063 g/day in women, and the energy ratio is 0.031%E in men and 0.034%E in women<sup>(7)</sup>. The EPA and DHA intake of Japanese people is, therefore, increased by approximately 4-fold compared to that of Americans, in terms of energy ratio, which is quite significant. No major difference was observed between the median alpha-linoleic acid intakes of American people and Japanese people.

There is a large bias in the intake distribution of fish oil. Marked differences are observed in the average and median intakes of EPA, DHA, and DPA, and, therefore, it is unclear whether the median is a typical value reflecting the habitual intake of fish oil in the population. This kind of bias is not seen in the intake distribution of alpha-linoleic acid.

## **2-5-2. To Avoid Inadequacy**

### **2-5-2-1. Factors to be Considered in Estimating Requirements**

An AI was set for n-3 fatty acids due to the presence of n-3 fatty acid deficiency<sup>(81)</sup>. Of the patients who could not consume food orally due to enterectomy, encephalopathy, etc., the effects of administered n-3 fatty acid (alpha-linoleic acid + fish oil) were reported in patients who had developed scaly dermatitis, hemorrhagic dermatitis, nodular dermatitis, or growth impairment, and whose n-6 fatty acid intake had been maintained to a certain degree, but n-3

fatty acid intake had been extremely low. With the increase in the blood n-3 fatty acid ratio, skin symptoms were improved through the administration of 0.2–0.3%E n-3 fatty acid<sup>(82,83)</sup>, and increases in weight were observed through the administration of 1.3%E n-3 fatty acid<sup>(77)</sup>. However, because both alpha-linoleic acid and fish oil are administered in many studies, it is unclear which lipid(s) improved symptoms. It may be necessary to consider n-3 fatty acids other than alpha-linoleic acid, EPA and DHA, and, accordingly, an AI was set for n-3 fatty acids.

## **2-5-2-2. Methods of Determining AI**

### **(1) Infants**

Breast milk was considered to be an ideal source of nutrients for infants, and, therefore, the AI was determined from the fat content of breast milk<sup>(3,4)</sup>, and the average milk intake (0.78 L/day)<sup>(5,6)</sup>. Infants aged 0–5 months obtain their nutrients from breast milk (or infant formula), and from the age of 6 months, they begin taking baby foods. Infants aged 6–11 months obtain their nutrients from both breast milk (or infant formula) and baby food. Since this is the transition period to early childhood, the average AIs (medians) of 0–5-month-old infants and 1–2-year-old infants were used.

The AI of infants aged 0–5 months was found by multiplying the n-3 fatty acid concentration of breast milk (1.16 g/L) and the average milk intake (0.78 L/day).

**n-3 fatty acids: AI (g/day) = 1.16 g/L × 0.78 L/day = 0.90 g/day**

For infants aged 6–11 months, the average AI of infants aged 0–5 months, and median (male and female average) intake from the 2010 and 2011 NHNS<sup>(2)</sup> of infants aged 1–2 years were taken. The AI was found as follows.

**n-3 fatty acids: AI (g/day) = [0.9 + (0.7 + 0.8) / 2] / 2 = 0.8 g/day**

### **(2) Children and Adults**

The median total n-3 fatty acid intake, calculated from the results of the 2010 and 2011 NHNS<sup>(2)</sup>, was set as the AI for those aged 1 year and older.

### **(3) Pregnant and Lactating Women**

Arachidonic acid and DHA are important constituent fatty acids of the nervous tissue. DHA is present in large quantities *in vivo*, particularly in the nerve synapses and photoreceptors of the retina. A larger number of n-3 fatty acids are required to create these organs in fetuses during pregnancy<sup>(84)</sup>. The median n-3 fatty acid intake of pregnant women, calculated from the results of the 2007 to 2011 NHNS<sup>(57)</sup>, is 1.8 g/day; thus, this value was considered as that which would not cause fetal developmental problems. Consequently, the AI for n-3 fatty acids was set at 1.8 g/day during pregnancy. Lactating women are assumed to secrete breast milk containing the average fat components of the breast milk of Japanese women. The median n-3 fatty acid intake of lactating women, calculated from the results of the 2007 to 2011 NHNS<sup>(57)</sup>, is 1.8 g/day. This value has not been found to cause essential fatty acid deficiencies in a majority of lactating women, and is considered to be the secretable amount of breast milk with sufficient n-3 fatty acids; therefore, this was set as the AI.

**DRIs for Dietary Fats**

(Percentage of total dietary fat in total energy (fat energy ratio): % energy)

Gender	Males		Females	
Age etc.	AI	DG <sup>1</sup> (median <sup>2</sup> )	AI	DG <sup>1</sup> (median <sup>2</sup> )
0-5 months	50	—	50	—
6-11 months	40	—	40	—
1-2 years	—	20-30(25)	—	20-30(25)
3-5 years	—	20-30(25)	—	20-30(25)
6-7 years	—	20-30(25)	—	20-30(25)
8-9 years	—	20-30(25)	—	20-30(25)
10-11 years	—	20-30(25)	—	20-30(25)
12-14 years	—	20-30(25)	—	20-30(25)
15-17 years	—	20-30(25)	—	20-30(25)
18-29 years	—	20-30(25)	—	20-30(25)
30-49 years	—	20-30(25)	—	20-30(25)
50-69 years	—	20-30(25)	—	20-30(25)
70+ years	—	20-30(25)	—	20-30(25)
Pregnant women	/		—	—
Lactating women			—	—

<sup>1</sup> Ranges are expressed as approximate values.

<sup>2</sup> Medians indicate the median values for the given range. They do not indicate most desirable values.

**DRIs for Saturated Fatty Acid (% energy)**

Gender	Males	Females
Age etc.	DG	DG
0-5 months	—	—
6-11 months	—	—
1-2 years	—	—
3-5 years	—	—
6-7 years	—	—
8-9 years	—	—
10-11 years	—	—
12-14 years	—	—
15-17 years	—	—
18-29 years	$\leq 7$	$\leq 7$
30-49 years	$\leq 7$	$\leq 7$
50-69 years	$\leq 7$	$\leq 7$
70+ years	$\leq 7$	$\leq 7$
Pregnant women	/	—
Lactating women		—

**DRIs for n-6 Fatty Acid (g/day)**

Gender	Males	Females
Age etc.	AI	AI
0-5 months	4	4
6-11 months	4	4
1-2 years	5	5
3-5 years	7	6
6-7 years	7	7
8-9 years	9	7
10-11 years	9	8
12-14 years	12	10
15-17 years	13	10
18-29 years	11	8
30-49 years	10	8
50-69 years	10	8
70+ years	8	7
Pregnant women	/	9
Lactating women		9

**DRIs for n-3 Fatty Acid (g/day)**

Gender	Males	Females
Age etc.	AI	AI
0-5 months	0.9	0.9
6-11 months	0.8	0.8
1-2 years	0.7	0.8
3-5 years	1.3	1.1
6-7 years	1.4	1.3
8-9 years	1.7	1.4
10-11 years	1.7	1.5
12-14 years	2.1	1.8
15-17 years	2.3	1.7
18-29 years	2.0	1.6
30-49 years	2.1	1.6
50-69 years	2.4	2.0
70+ years	2.2	1.9
Pregnant women	/	1.8
Lactating women		1.8

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## Carbohydrates

### 1. Background Information

The nutritional significance of carbohydrates differs depending on the sub-classification (particularly sugars versus polysaccharides, and starch versus non-starch polysaccharides). However, currently, food composition tables (*Standard Tables of Food Composition in Japan 2010*)<sup>(1)</sup> do not list the contents of these carbohydrates in many foods, so it is difficult to measure the dietary intake and supply in Japanese people. Therefore, the DRIs for only (total) carbohydrates and dietary fiber were determined. In addition, alcohol, which is not a carbohydrate but produces energy, and its association with various life-style related diseases (LRDs) is drawing attention; this will be discussed in this chapter.

#### 1-1. Definitions and Classifications

The compositional formula of carbohydrate is  $C_n(H_2O)_n$ . Carbohydrates can be monosaccharides or polymers; monosaccharides are the minimum constituent unit. They are classified according to their chemical, physical, and physiological characteristics. When classified according to the degree of polymerization, which is a chemical characteristic, carbohydrates are divided into saccharides (1 degree or 2 degrees of polymerization), oligosaccharides (3–9 degrees of polymerization), and polysaccharides (10 or more degrees of polymerization)<sup>(2)</sup>. Saccharides are further divided into monosaccharides and disaccharides. Monosaccharides include glucose, fructose, and galactose; and disaccharides include sucrose, lactose, and maltose. Oligosaccharides are divided into maltooligosaccharides (alpha-glucan), and oligosaccharides containing monosaccharides other than glucose. Polysaccharides are divided into starch and non-starch polysaccharides. The former consists of amylose and amylopectin, while the latter includes cellulose, hemicellulose, and pectin<sup>(2)</sup>. In terms of physiological classification, carbohydrates are classified into carbohydrates that can be digested by human digestive enzymes, and carbohydrates that cannot be digested. The term “dietary fiber” is the result of a classification method focused on physiological characteristics; however, the definition of dietary fiber differs slightly between organizations in and outside Japan<sup>(3)</sup>. If only regular foods are consumed, the majority of dietary fiber consumed is in the form of non-starch polysaccharides.

### 2. DRIs for Carbohydrates

#### 2-1. Carbohydrates

The main nutritional role of carbohydrates is to supply glucose to tissues that can usually use only glucose as an energy source, such as the brain, nervous tissue, red blood cells, renal tubules, testes, and oxygen-deficient skeletal muscles. The brain accounts for approximately 2% of the total body weight, but is assumed to consume approximately 20% of an individual's basal metabolic rate<sup>(4)</sup>. If the basal metabolic rate is 1,500 kcal/day, the brain's energy expenditure is 300 kcal/day, which is equal to 75 g/day of glucose. As explained above,

since tissues other than the brain also use glucose as an energy source, the glucose requirement is estimated to be no less than 100 g/day. In other words, the minimum digestible carbohydrate requirement is assumed to be approximately 100 g/day. However, this does not necessarily mean that this is the minimum amount truly required. This is because the liver performs gluconeogenesis using the lactic acid and amino acids released from the muscles, and glycerol released from adipose tissue as needed to supply glucose in the blood. Furthermore, individuals other than infants also usually consume a considerably larger amount (> 100 g/day) of carbohydrate. Therefore, there is little sense or value in calculating the estimated requirement on the basis of this amount. Moreover, as will be explained later on, reports stating that carbohydrates are the direct cause of specific adverse health events are not sufficiently supported theoretically and epidemiologically, with the exception of diabetes--a type of LRD. Consequently, no estimated average requirement (EAR) (and recommended dietary allowance [RDA]), tolerable upper intake limit level (UL), or adequate intake (AI) was set for carbohydrates.

The percentage of energy (%E) derived from carbohydrates as a proportion of total energy intake is more often used to consider the relationship between carbohydrates and diabetes than absolute amounts (g/day). This is because carbohydrates are a major source of energy, and both the intake of carbohydrate as an energy source and the direct effect of carbohydrates on adverse health events must be jointly considered. This is discussed in the chapter "Energy Providing Nutrients' Balance."

The reference intakes for saccharides was not set at this time due to difficulties in measuring their intake in Japanese people.

## **2-2. Dietary Fiber**

### **2-2-1. Basic Concept**

Many reports describe a correlation between the inadequate intake of dietary fiber and the development of LRDs. It was, therefore, deemed appropriate to set a tentative dietary goal (DG) for the prevention of LRDs.

### **2-2-2. Preventing the Development and Progression of LRDs**

#### **2-2-2-1. Association with LRDs**

##### **(1) Association between Dietary Fiber and the Prevention of LRDs Development**

A wide range of LRDs have been examined for their association with dietary fiber intake. Many studies have found that dietary fiber intake is negatively correlated with the development of and death from myocardial infarction<sup>(5,6)</sup>, development of stroke<sup>(7,8)</sup>, development of and death from cardiovascular disease<sup>(6,9)</sup>, development of diabetes<sup>(9,10)</sup>, and development of breast and gastric cancer<sup>(11,12)</sup> (in some of these studies, a significant correlation was not observed). A meta-analysis that examined the association with the development of diabetes showed a significant negative correlation with cereal-derived dietary fiber intake;

however, no association with fruit- or vegetable-derived dietary fiber intake was observed<sup>(10)</sup>. This suggests that sources of dietary fiber need to be considered in terms of whether or not they are effective in preventing the development of diabetes.

A negative correlation between serum (or plasma) LDL cholesterol levels and blood pressure has also been suggested, which is a strong risk factor for cardiovascular disease<sup>(13,14)</sup>. Many epidemiological studies have also shown an association with obesity<sup>(15,16)</sup>. However, study results on the association with cancer, particularly colorectal cancer, are not always consistent<sup>(17)(18)</sup>. A significant negative correlation was observed between dietary fiber intake and colorectal cancer; however, this correlation was no longer significant when the intakes of folic acid, red meat, milk, and alcohol were taken into account, which may be a reason for the inconsistent results<sup>(17)</sup>. Many epidemiological studies have examined the relationship between dietary fiber intake and LRDs. However, few studies have quantitatively (quantity response relationship) demonstrated this relationship.

Dietary fiber intake has been suggested to influence bowel habits (adverse health events such as constipation); one cross-sectional epidemiological study found a negative correlation between dietary fiber intake and the prevalence of constipation<sup>(19)</sup>, whereas no Japanese studies found a correlation between the two<sup>(20)</sup>.

## **(2) Association between Dietary Fiber and Prevention of LRD Progression**

An interventional study found that 20 g/day of dietary fiber increases fecal weight, leading to better bowel movement<sup>(21)</sup>, whereas another study observed an increase in fecal weight but could not conclude that this improved constipation<sup>(22)</sup>. Therefore, it remains to be fully elucidated what amount of dietary fiber consumed from a normal diet affects constipation and what percentage of dietary fiber intake contributes to better bowel habits.

The findings of a meta-analysis of interventional studies also suggested that an increased dietary fiber intake was negatively correlated with blood pressure<sup>(13)</sup>. Another meta-analysis similarly suggested a negative correlation with serum (or plasma) LDL cholesterol levels<sup>(15)</sup>. However, this effect was limited to water-soluble dietary fiber. Meanwhile, a low-glycemic index diet has also been observed to have a LDL cholesterol-lowering effect<sup>(23)</sup>. Diets with a low glycemic index are generally considered rich in dietary fiber, particularly insoluble dietary fiber. Therefore, it is deemed appropriate to recommend dietary fiber to individuals with high LDL cholesterol levels, regardless of whether it is water-soluble or insoluble.

In a meta-analysis report that summarized 15 interventional studies, which observed changes in blood glucose levels after increasing dietary fiber intake, an average decrease of 15.3 mg/dL in fasting blood glucose levels was observed with an average increase of 18.3 g/day in dietary fiber levels<sup>(24)</sup>.

## 2-2-2-2. Methods Used to Set the DG

### (1) Adults

The LRD in which the association with dietary fiber intake is the most evident is myocardial infarction. A pooled analysis that reanalyzed the accumulated data of 10 cohort studies demonstrated a decrease in myocardial infarction-related mortality with a dietary fiber intake of 24 g/day, and an increase in mortality with an intake of less than 12 g/day<sup>(5)</sup>. However, some of these studies included groups of vegetarians, and all of them were conducted in Western countries. Therefore, the dietary fiber intakes were likely larger overall than those of Japanese people. Thus, using these results poses a problem. However, a recently collected meta-analysis showed no clear threshold, and found an almost negative linear correlation with the risk (incidence or mortality) of myocardial infarction<sup>(6)</sup>.

Incidentally, the AI for dietary fiber was set at 14 g/1,000 kcal following a review focusing on each study used in this meta-analysis in the US-Canada DRIs (note: a DG does not exist in the US-Canada DRIs, and, therefore, AI is used instead)<sup>(4)</sup>. This value is based on the typical intake of the group in which dietary fiber was found to have the greatest preventive effect in each of these studies, and is substantially higher than 24 g/day.

Other meta-analyses that examined the development of stroke<sup>(7)</sup> and breast cancer<sup>(25)</sup> found a significant negative correlation, but no clear threshold. For this reason, it is difficult to use these findings as a basis for calculating the DG. Considering the relationship observed between dietary fiber intake and the development of and risk factors for LRDs, the significance of the DG calculated here is nebulous and should be interpreted to mean “it is best to consume as large an amount as possible without excess.” Furthermore, considering the fact that a large dietary fiber intake has not been reported to increase the risk of any LRDs, it is safe to interpret the DG as such.

Despite the above-described limitations, the ideal DG should be at least 24 g/day, and at least 14 g/1,000 kcal if possible, considering the above value. However, the median dietary fiber intake of Japanese people, based on the results of the 2010 and 2011 National Health and Nutrition Surveys<sup>(26)</sup>, is much lower than this in all age groups (Table 1). Thus, the feasibility of setting this value as the DG is low. Therefore, the DG was set using the following method.

The intermediate value (18.9 g/day) between 24 g/day and the median dietary fiber intake (13.7 g/day) of Japanese adults (aged 18 years and older), currently, was set as the reference value for calculating the DG. The mean reference weight (57.8 kg) of adults (aged 18 years and older), and the reference weights of each sex and age group as well as the 0.75th power of body weight were then used to extrapolate intake with the body surface area estimation method, and calculate the DG of each sex and age group. However, the simple average of each sex and age group (all eight groups) was used for the average reference body weight.

This was calculated specifically as follows:

$$18.9 \text{ (g/day)} \times [\text{reference body weight (kg) of each sex and age group} \div 57.8 \text{ (kg)}]^{0.75}$$

After taking the integer of the value obtained with this formula, the value was equalized

between adjacent age groups (Table 1).

No additional amount was set for pregnant or lactating women.

Incidentally, most studies used in the calculation of DGs focused on the dietary fiber derived from normal food, and not dietary fiber derived from supplements, etc. The same health benefits as those described here are, therefore, not guaranteed when the equal amount of dietary fiber is consumed in the form of supplements or sources other than regular food. Furthermore, it should be noted that there is no evidence stating that greater health benefits than those described here can be expected from consuming large amounts of dietary fiber in the form of supplements or other sources in amounts that exceed those that can be consumed from regular food.

**Table 1. Method used to determine the DG for dietary fiber**

Gender	Male				Female			
Age (years)	(A)	(B)	(C)	(D)	(A)	(B)	(C)	(D)
1-2	-	-	-	-	-	-	-	-
3-5	-	-	-	-	-	-	-	-
6-7	10.8	9.2	11	A	10.3	9.1	10	A
8-9	11.8	11.0	12	A	11.7	10.8	12	A
10-11	12.7	13.1	13	B	12.4	13.3	13	B
12-14	14.9	16.7	17	B	13.2	16.3	16	B
15-17	14.0	19.4	19	B	11.8	17.4	17	B
18-29	11.8	20.2	20	B	10.8	17.0	18	B, ↑
30-49	12.6	21.5	20	B, ↓	11.6	17.7	18	B
50-69	14.9	20.7	20	B, ↓	15.1	17.7	18	B
70+	15.5	19.4	19	B	14.8	16.8	17	B, ↑

(A) Median value of the dietary fiber intake in NHNS2010 and NHNS2011 (g/day)

(B) Extrapolated value from the reference value for DG calculation

(C) Value of the DG determined.

(D) Value used as DG (A or B), ↑ and ↓ present the way to smooth the calculated value (up and down)

## (2) Children

Constipation is an adverse health event that is frequently observed in children. Some systematic reviews summarizing the effect of dietary fiber intake on improving constipation describe the existence of reports in which dietary fiber intake improved constipation<sup>(27)</sup>. However, since there are no quantitative discussions, these reports cannot be used to calculate the DG. An interventional study in constipated children (aged 3–14 years) demonstrated a significant improvement in a group of 3–7-year-olds that achieved a dietary fiber intake of at least 10 g/day, and a group of 8–14-year-olds that achieved an intake of at least 14.5 g/day<sup>(28)</sup>.

However, a number of issues remain, such as a control group not having been established (controlled before and after trial), and problems associated with the consideration of the reverse causality among other factors. Moreover, very few supplementary trials used similar study methods.

As explained before, very few reports claim that dietary fiber intake at the time of consumption is directly associated with the prevention of the development and progression of the LRDs featured in the current DRIs. It is, therefore, considered difficult to calculate the DG on the basis of these reports.

However, habitual nutrient intake over a long period of time has an effect on the development of LRDs, and other factors, suggesting that dietary habits during childhood may influence the development of and risk factors for cardiovascular disease after entering adulthood<sup>(29)</sup>. Many reports state that childhood eating habits have a certain impact on eating habits thereafter<sup>(30,31)</sup>. It is, therefore, recommended to also set DRIs for children<sup>(32)</sup>. However, it is difficult to assess intakes in children aged 1–5 years, and the details of the actual intake status in Japan are unknown. Thus, there is little evidence for the calculation of the DG for children. The DG was calculated for those aged 6–17 years using the same method as that in adults. Furthermore, the current median intake was set as the DG when the current median intake was greater than the calculated DG.

**DRIs for Carbohydrates (% energy)**

Gender	Males	Females
Age etc.	DG <sup>1,2</sup> (median <sup>3</sup> )	DG <sup>1,2</sup> (median <sup>3</sup> )
0-5 months	—	—
6-11 months	—	—
1-2 years	50-65 (57.5)	50-65 (57.5)
3-5 years	50-65 (57.5)	50-65 (57.5)
6-7 years	50-65 (57.5)	50-65 (57.5)
8-9 years	50-65 (57.5)	50-65 (57.5)
10-11 years	50-65 (57.5)	50-65 (57.5)
12-14 years	50-65 (57.5)	50-65 (57.5)
15-17 years	50-65 (57.5)	50-65 (57.5)
18-29 years	50-65 (57.5)	50-65 (57.5)
30-49 years	50-65 (57.5)	50-65 (57.5)
50-69 years	50-65 (57.5)	50-65 (57.5)
70+ years	50-65 (57.5)	50-65 (57.5)
Pregnant women	/	—
Lactating women		—

<sup>1</sup> Ranges are expressed as approximate values.

<sup>2</sup> Includes alcohol. However, it does not imply recommendation of alcohol consumption.

<sup>3</sup> Medians indicate the median values for the given range. They do not indicate most desirable values.

**DRIs for Dietary Fiber (g/day)**

Gender	Males	Females
Age etc.	DG	DG
0-5 months	—	—
6-11 months	—	—
1-2 years	—	—
3-5 years	—	—
6-7 years	≥ 11	≥ 10
8-9 years	≥ 12	≥ 12
10-11 years	≥ 13	≥ 13
12-14 years	≥ 17	≥ 16
15-17 years	≥ 19	≥ 17
18-29 years	≥ 20	≥ 18
30-49 years	≥ 20	≥ 18
50-69 years	≥ 20	≥ 18
70+ years	≥ 19	≥ 17
Pregnant women	/	—
Lactating women		—

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## **Energy Providing Nutrients' Balance**

### **1. Background Information**

Energy providing nutrients' balance is the ratio of energy providing nutrients (macronutrients-- namely protein, fat and carbohydrate) including alcohol and the individual components for a particular food item. The current DRIs are determined based on the energy intakes from these nutrients and expressed as the percentage of energy intake (%E). Achieving a balance in the intakes of aforementioned nutrients is aimed at the prevention of not only their inadequate intake, but also the development of lifestyle-related diseases (LRDs). In the prevention of LRD development, nutrient balance should be achieved, based on meeting requirements to avoid inadequacy. Thus, the DRIs for energy-providing nutrients' balance should focus on the tentative dietary goal for preventing LRDs (DGs).

In terms of energy-providing nutrients, protein intake must meet the estimated average requirement (EAR). To avoid insufficiency, it is recommended that the intake be above the recommended dietary allowance (RDA). In terms of fat, adequate intake (AI) is set for n-6 and n-3 fatty acids, while DGs are set for saturated fatty acids. Although carbohydrate is an essential nutrient, its intake is likely to exceed the requirement except under specific conditions.

Therefore, it is appropriate to first determine the amount of protein, followed by that of fat, with the carbohydrate amount set as the residual value for the energy-providing nutrients' balance. While alcohol provides energy, it is not an essential nutrient, and there is no reason to recommend its intake. If alcohol is included in the nutrients' balance, the rest of the protein and fat components are derived from the amount of carbohydrate and alcohol.

For infants aged under 1 year, a desirable nutrients' balance is achieved through the consumption of breast milk. Therefore, the energy-providing nutrients' balance was determined for those aged over 1 year in the DRIs.

### **2. Energy Conversion Factor**

There are slight differences in the energy conversion factors for protein, fat, carbohydrate, and alcohol, depending on the foods from which these nutrients are obtained<sup>(1)</sup>. The Atwater factor is used for the approximation of values (protein, carbohydrate: 4, fat: 9), without consideration of the above-stated differences. It also does not consider the different types of components that comprise these nutrients, namely amino acids, fatty acids, and sugars.

The amount of energy produced by dietary fiber is considered to be 0-2 kcal/g<sup>(2)</sup>, which is lower than that produced by other carbohydrates. Among carbohydrates, dietary fiber should not be included as a source of energy. However, from the standpoint of the applicability and feasibility of the DRIs, the energy-providing nutrients' balance includes dietary fiber as carbohydrates, and 4 kcal/g is used as its energy conversion factor.

In Japan, 7.1 kcal/day is the amount of energy obtained from alcohol<sup>(1)</sup>. However, here, we used the value "7 kcal/g", as the energy conversion factors of the other nutrients are integers.

### **3. For the Prevention of LRD Development and Progression**

It is important for the protein intake to be above the RDA, or at least on par with the EAR. We reviewed studies focusing on protein intakes above the EAR or RDA, from the standpoint of the prevention of LRD development and progression.

In terms of dietary fat, a high intake of saturated fat is a major issue, in this context. Several studies have examined the association between low fat intake and health status.

#### **3–1. Prevention of LRD Development**

##### **3–1–1. Protein**

According to a US cohort study examining the association between protein intake and estimated glomerular filtration rate (eGFR) in women, a significant decrease in the eGFR was observed among high-protein intake (median: 92 g/day) participants with a slightly low renal function level, while there was no significant association among those with normal renal function levels<sup>(3)</sup>.

In another US cohort study examining the association between low-carbohydrate diet and the development of diabetes, the incidence of diabetes was significantly higher in men with a low-carbohydrate and high-animal-protein diet than in those in other groups, with no such results being observed for low-carbohydrate and high-plant-protein diets<sup>(4)</sup>. This finding suggests the importance of considering the influence of the quality of the proteins (amino acids) consumed or their dietary sources on the intakes of other nutrients, rather than the total protein intake. Additionally, the same cohort study reported that the all-cause mortality was significantly higher among men with a low-carbohydrate and high-animal-protein diet, and significantly lower in men with a low-carbohydrate and high-plant-protein diet<sup>(5)</sup>. Similar results were observed in a cohort of American women<sup>(5)</sup>. Although the aforementioned studies did not directly examine the influence of the differences in the amounts of total protein intake, their results suggest that dietary protein sources and the amount consumed may affect health status. A cohort study has shown a significantly higher incidence of cardiovascular diseases in those consumed low-carbohydrate diets (= high-protein diets) in Swedish women<sup>(6)</sup>. It is needed to interpret these results considering the quality of protein (amino acids) and its dietary sources.

A significant negative association was reported between protein intake and the incidence of frailty among elderly American people (aged 65-79 years)<sup>(7)</sup>. In that study, participants were classified into quintiles, according to their protein intake, with those with an intake of 12.4%E in the lowest quintile and those with an intake of 16.0%E in the highest quintile. A Japanese cross-sectional study reported that the prevalence of frailty was significantly lower in those with a protein intake greater than 16.1%E, among 2,108 elderly women (aged 65 years or older)<sup>(8)</sup>.

### **3–1–2. Dietary Fat**

Several studies have focused on the association between fat intake and changes in body weight (BW) later in life. However, the results are inconsistent, with most studies reporting no significant association. A US-based study reported a significant association between fat intake and BW gain only among women younger than 50 years<sup>(9)</sup>, while a European study reported no significant association in any sex or age group<sup>(10)</sup>.

The development of LRDs is affected largely by fatty acid intake (especially saturated fatty acids), rather than total fat intake. This association has been established for myocardial infarction<sup>(11)</sup>, as well as diabetes<sup>(12)</sup>. Thus, it is important to consider the quality of the fats (especially saturated fatty acids) consumed, for the prevention of LRDs.

### **3–1–3. Carbohydrate**

A meta-analysis showed an increased all-cause mortality in those with low-carbohydrate diets, while no association with cardiovascular mortality and incidence was observed<sup>(13)</sup>. However, it is difficult to use these data for the DRIs because this meta-analysis did not report the carbohydrate intake amount.

The development of LRDs is largely influenced by the intake of dietary fiber, the glycemic index of the food consumed, and the types of sugar (monosaccharide, disaccharide or polysaccharide), rather than the total carbohydrate intake amount. For example, some studies have reported on the protective effect of grain dietary fiber<sup>(14)</sup>, dietary glycemic index, and glycemic load<sup>(15,16)</sup> against diabetes development, and the association between the massive intake of sugar-sweetened beverages and obesity<sup>(17,18)</sup>. Therefore, the quality of the carbohydrate consumed is of significance. However, a study reported that there was no significant association between carbohydrate intake and dietary glycemic index, and the development of diabetes<sup>(19)</sup>. At present, these results are inconsistent and further investigation is needed.

## **3–2. Prevention of LRD Progression**

### **3–2–1. Protein**

A meta-analysis summarized the interventional trials that examined the influence of high-protein diets on cardiovascular diseases and metabolic risk factors compared to that of low-protein diets, over a period of more than 12 months<sup>(20)</sup>. The meta-analysis found no significant difference in the BW, waist circumference, serum low-density lipoprotein (LDL) cholesterol level, triglyceride level, blood pressure, fasting blood glucose level, and glycated hemoglobin (HbA1c) level between those with a high-protein diet (30%E in most trials) and those with a low-protein diet (15%E in most trials). In an interventional trial that examined middle-aged obese participants for 2 years, there was no significant difference in the BW change between those with high-protein diets (25%E) and those with low-protein diets (15%E)<sup>(21)</sup>.

Another interventional study of obese patients with diabetes reported a significant BW loss with no significant change in fasting blood glucose and HbA1c levels for 12 months for high protein diet<sup>(22)</sup>, while another study reported there was no significant change in any of the outcomes<sup>(21)</sup>.

A meta-analysis including relatively short-term interventional studies (mean: 12 weeks) reported that significant improvements were observed in BW, body fat, and triglyceride levels in those with high-protein diets (30%E)<sup>(23)</sup>. While several short-term intervention studies have reported positive results for the progression of LRDs through the intake of high-protein diets, few studies included long-term interventions.

### **3–2–2. Dietary Fat**

A meta-analysis summarized interventional studies which examined the effect of low-fat diets (20-30%E in most studies) on blood lipids levels, for more than 12 months<sup>(24)</sup>. The results showed a significant decrease not only in serum LDL cholesterol levels, but also a significant decrease in serum high-density lipoprotein (HDL) cholesterol levels, and a significant increase in triglyceride levels. Another meta-analysis summarizing 6-month interventions showed a similar result, with no significant difference observed in the blood pressure, fasting blood glucose level, and fasting insulin level<sup>(25)</sup>. Another meta-analysis of 19 interventional studies including diabetes patients reported that low fasting insulin and triglyceride levels and high HDL cholesterol levels were observed in those with high-fat diets (low-carbohydrate) compared to those with low-fat diets (high-carbohydrate); no significant differences were noted in the HbA1c, fasting blood glucose, total cholesterol, and LDL cholesterol levels<sup>(26)</sup>.

In an interventional study of middle-aged obese participants, no significant difference was noted in the BW change between individuals with low-fat diets (20%E) and those with high-fat diets (40%E), over a 2-year period<sup>(21)</sup>. However, in another interventional study of middle-aged participants, a greater BW loss was observed in those with high-fat diets (38%E) than those with low-fat diets (30%E), over a 2-year period<sup>(27)</sup>; in that study, the composition of the fatty acids were different between the two diets. A meta-analysis reported a greater BW loss in those with low-fat diets by summarizing 33 interventional studies that examined the effect of decreased dietary fat intake on BW changes<sup>(28)</sup>. In that meta-analysis, the dietary fat intake before the interventions was reported as 28-43%E; therefore, the results cannot be interpreted beyond this range.

Yet another meta-analysis stratified interventional studies examining the effects of low-fat diets on BW and blood lipids, by interventional period; 6 months vs. 12 months<sup>(29)</sup>. In that study, a significant BW loss was observed in the 6-month long trial, but not in the 12-month long trial; no significant effect was observed for blood lipids.

Overall, the influence of total fat intake on health is not as obvious as previously expected, and study results are inconsistent. However, it seems preferable to maintain a lower

fat intake, within the range of 28-43%E, from the standpoint of BW management. However, total fat also includes essential fatty acids. Since the requirements for these fatty acids vary by groups or individuals, the DG for total fat should be set so as to meet the AI of essential fatty acids, considering differences between the life stages. Additionally, several studies have reported that saturated fatty acids have unfavorable effects on several LRDs. Therefore, the DG of total fat should be set such that it does not exceed the DG of saturated fatty acids. Therefore, the DG of total fat should be set within the range observed in the study results, at a value that reduces the health effects of saturated fatty acids as much as possible. Thus, saturated fatty acids must be included in the energy-providing nutrients' balance, considering their health effects.

### **3—2—3. Carbohydrate**

Some meta-analyses have suggested that low-carbohydrate diets may be effective for BW loss and diabetes control<sup>(30,31)</sup>. However, most of those meta-analyses did not provide details on the diets, such as the diet compositions of the control groups.

Another meta-analysis reported that dietary fiber supplementation led to significant decreases in fasting blood glucose and HbA1c levels among patients with diabetes<sup>(32)</sup>. It is possible that carbohydrate quality is also important for the prevention of the development and progression of LRDs. However, further investigation is required.

## **4. Method Used to Set the DG**

### **4—1. Basic Concept**

The achievement of energy-providing nutrients' balance is directly and strongly associated with the prevention of LRD development or progression. However, the type of fatty acid (especially saturated fatty acids), dietary fiber as a carbohydrate, or the source of protein may play a rather more important role. Considering that total fat includes saturated fatty acids and carbohydrate includes dietary fiber, the DG of the energy-providing nutrients' balance was determined in the current DRIs using the methods stated in the following paragraphs. These DG ranges are approximate, however, and the balance needs to be considered in relation with other dietary energy and nutrient intakes.

### **4—2. Protein (DG)**

The lowest DG value was set above the RDA of protein. On expressing the protein/energy ratio of the RDA value (g/day) using estimated energy requirements (EERs: kcal/day) for physical activity level (PAL) I (low), in each age and sex group, the ratio can be calculated as a maximum of 13.3%E in women aged 70 years or older. If pregnant women (in the late stages of pregnancy; aged 18-29 years) and lactating women are included, the maximum ratio can be calculated as 14.3%E. The protein intake should meet the requirement even if the energy intake is low. Considering the importance of protein intake, it may be safe to set values

that are higher than the calculated RDA for the DG.

The highest DG value should be set considering the upper limit (UL). Although there is no UL for protein, some studies recommended an intake level of less than 2.0 g/kg BW/day for adults, especially among elderly individuals, from the standpoint of the prevention of unfavorable metabolic changes as well as azotemia<sup>(33,34)</sup>. Using this value and the EER for PAL II (middle), the protein/energy ratio can be calculated as 19-22%E (in those aged 18-69 years) and 22-23%E (in those aged 70 years and older).

Based on the above-stated estimation, the DG of the protein/energy ratio was determined as 13-20%E. Elderly people (for the prevention of frailty), and pregnant and lactating women (for children's growth) should ensure their protein intake does not reach the lowest range.

#### **4–3. Fat (DG)**

The lowest DG value of the fat/energy ratio was set to secure the AI of essential fatty acids. Therefore, while this value is not aimed at the prevention of LRDs, ensuring the intake meets the AI is of high priority in the prevention of deficiency. For this reason, the lowest DG value for fat was determined considering the AI of essential fatty acids (n-6 and n-3 fatty acids). For more details, see "Dietary Fat."

The highest fat/energy ratio of the DG was determined considering the DG of saturated fatty acids. Most studies examining the health effect of total fat intake used a fat intake value of 30%E for low-fat diets; it is difficult to examine the health effect of fat intakes lower than this value. A meta-analysis (including 33 interventional studies) reported that a greater BW loss was observed in those whose fat intake decreased from 28-43%E<sup>(28)</sup>, suggesting that the results cannot be interpreted beyond this range. Overall, evidence suggesting that a fat/energy ratio lower than 30%E be adopted as the highest DG value of fat is insufficient. Thus, the DG of the fat/energy ratio was determined as 20-30%E.

For saturated fatty acids, the DG was determined as 7%E or less. The DG of saturated fatty acids was not set for children aged 1-17 years due to a lack of data. However, this does not invalidate the need of considering excessive intake of saturated fatty acids in children.

It is necessary to pay attention to the quality of fat consumed, including that of essential fatty acids, as well as the amount of total fat and saturated fatty acids consumed.

#### **4–4. Carbohydrate (DG)**

The carbohydrate/energy ratio, including alcohol, was set as the value of the residual energy ratio of protein and total fat. A high-carbohydrate diet, without quality consideration, may be unfavorable, comprising only highly-refined cereals, sweeteners, sweetened beverages, and alcoholic beverages. Such diets can also lead to an insufficient intake of many vitamins and minerals, as these foods contain lower amounts of vitamins and minerals than other foods. If the lowest energy ratios of protein (13%E) and total fat (20%E) are used, the carbohydrate

## II Energy and Nutrients

### Energy Providing Nutrients' Balance

energy ratio can be calculated as 67%E. Therefore, the highest value of the carbohydrate DG was set at 65%E. It should be noted that the sum of the lowest values of the energy ratio of protein, fat and carbohydrate is not 100%.

The lowest carbohydrate DG value was set according to the highest values of the energy ratios of protein (20%E) and total fat (30%E). It is important to pay attention to carbohydrate quality to ensure the dietary fiber intake does not decrease, in the case of low-carbohydrate diets.

II Energy and Nutrients  
Energy Providing Nutrients' Balance

**DRIs for Energy Providing Nutrient Balance (% energy)**

DG <sup>1</sup> (median <sup>2</sup> ) (For both males and females)				
Age etc.	Proteins	Fats <sup>3</sup>		Carbohydrates <sup>4,5</sup>
		Fats	Saturated fatty acid	
0-11 months	—	—	—	—
1-17 years	13-20 (16.5)	20-30 (25)	—	50-65 (57.5)
18-69 years	13-20 (16.5)	20-30 (25)	≤ 7	50-65 (57.5)
70+ years	13-20 (16.5)	20-30 (25)	≤ 7	50-65 (57.5)

<sup>1</sup> Ranges for each nutrient are expressed as approximate values. The present table shall be applied flexibly if used for the purpose of prevention of LRDs or frailty of elderly persons.

<sup>2</sup> Medians indicate the median values for the given range. They do not indicate most desirable values.

<sup>3</sup> Fats require careful consideration on their qualities, such as their component fatty acids (e.g., saturated fatty acids).

<sup>4</sup> Includes alcohol. However, it does not imply recommendation of alcohol consumption.

<sup>5</sup> Pay extra attention on DGs for dietary fibers.

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## Vitamins

### (1) Fat-soluble Vitamins

## Vitamin A

### 1. Background Information

#### 1-1. Definition and Classification

The vitamin A compounds with potent vitamin A activity *in vivo* after oral intake include retinol, retinal, carotenoids, and 50 types of provitamin A carotenoids, such as  $\beta$ -carotene,  $\alpha$ -carotene, and  $\beta$ -cryptoxanthin. In the current DRIs, we used retinol activity equivalents (RAE) as the vitamin A unit, instead of retinol equivalents (REs) that were used in the previous DRIs.

#### 1-2. Function

Retinol and retinal are necessary for the protection of retinal cells, and photostimulated reaction in the photoreceptor cells. Vitamin A deficiency can cause blindness via corneal xerosis in infants or children, and night blindness due to impaired adaptation to darkness in adults.

#### 1-3. Digestion, absorption, and metabolism

Retinyl ester and provitamin A carotenoids are the main forms of vitamin A contained in animal and plant foods, respectively. Retinyl ester hydrolase catalyzes the hydrolysis of retinyl ester to retinol in the intestinal brush border, and this retinol is then absorbed at a rate of 70% to 90%<sup>(1,2)</sup>. Most  $\beta$ -carotenes produce 2 mol of vitamin A (retinal) in the small intestine, while other provitamin A carotenoids produce 1 mol of retinal. The absorption rate of  $\beta$ -carotene is about one-seventh of a supplement that contains purified  $\beta$ -carotene and oils. In the present DRIs, the absorption rate of  $\beta$ -carotene is one-sixth its total value, which is in accordance with the rate described in the US-Canada DRIs<sup>(3)</sup>.

Assuming that the conversion rate of  $\beta$ -carotene to retinol is 50%, the bioavailability of  $\beta$ -carotene as vitamin A is  $1/12(1/6 \times 1/2)$ , such that 12  $\mu\text{g}$  of food-derived  $\beta$ -carotene would correspond to 1  $\mu\text{g}$  in RAE. Thus, the following formula can be used to convert the values of food-derived vitamin A-related compounds to RAEs:

#### **Retinol activity equivalent ( $\mu\text{gRAE}$ )**

$$= \text{retinol } (\mu\text{g}) + \beta\text{-carotene } (\mu\text{g}) \times 1/12 + \alpha\text{-carotene } (\mu\text{g}) \times 1/24 + \beta\text{-cryptoxanthin } (\mu\text{g}) \times 1/24 + \text{other provitamin A carotenoids } (\mu\text{g}) \times 1/24$$

Caution is to be exercised in calculating the corresponding value for oil-solubilized  $\beta$ -carotene, as its bioavailability as a form of vitamin A is half of its total value; 2  $\mu\text{g}$  of fat-solubilized  $\beta$ -carotene would correspond to 1  $\mu\text{g}$  of retinol.

## 2. To Avoid Inadequacy

### 2–1. Factors to be Considered in Estimating Requirements

Classical vitamin A deficiency leads to corneal xerosis in infants, and possibly blindness and night blindness in adults. Other signs of deficiency include growth retardation, skeletal and neurological developmental defects, disturbed growth and differentiation of epithelial cells, dryness, thickening, and keratinization of the skin, immunodeficiency, and susceptibility to infection<sup>(4)</sup>. Due to the abundant storage of vitamin A in the liver, inadequate intake does not lead to decreased plasma retinol concentrations, unless the hepatic vitamin A storage is lower than 20 µg<sup>(5)</sup>. Thus, plasma retinol concentration cannot be used as an index of vitamin A status. Theoretically, hepatic vitamin A storage is the best index, but its measurement is highly invasive and not applicable to humans.

### 2–2. Method Used to Set the Estimated Average Requirement (EAR) and Recommended Dietary Allowance (RDA)

#### 2–2–1. Calculation of EAR and RDA

Plasma retinol concentrations are maintained at normal levels unless the hepatic vitamin A storage is below 20 µg, even if adults take only meals without vitamin A for up to 4 months. Therefore, as long as the hepatic vitamin A storage is maintained at the minimum level (20 µg/g), relatively minor deficiencies such as immunodeficiency and night blindness will not result<sup>(5,6)</sup>. Thus, the vitamin A intake required to maintain minimal hepatic vitamin A storage can be used to estimate the EAR for vitamin A<sup>(7)</sup>. Compartment analyses, assuming the existence of 3 compartments—serum, liver, and other tissues—have shown that the daily disposal rate of vitamin A is approximately 2%, regardless of the vitamin A storage in the body<sup>(8–10)</sup>. Using this percentage, the daily disposal amount (DDA), daily disposal rate (DDR), body storage (BS) according to body weight (BW), and hepatic storage (HS) of vitamin A can be calculated as follows:

$$\text{DDA } (\mu\text{g/day}) = \text{BS } (\mu\text{g}) \times \text{DDR } (2/100)^{(8)}$$

BS/BW can be calculated as follows:

$$\text{BS/BW } (\mu\text{g}/[\text{kg BW/day}]) = \text{HS } (20 \mu\text{g/g}) \times \text{liver weight/ BW } (21 \text{ g/kg BW})^{(8,11)} \times 10/9^{(8,12)}$$

DDA/BW can be calculated as follows:

$$\text{DDA/BW} = \text{BS } (20 \mu\text{g/g} \times 21 \text{ g/kg} \times 10/9) \times \text{DDR } (2/100) = 9.3 \mu\text{g}/[\text{kg BW/day}]$$

Thus, the amount of vitamin A intake required to compensate for its daily elimination, thereby ensuring that the hepatic storage of vitamin A is maintained and vitamin A deficiency is avoided, is estimated to be 9.3 µgRAE/kg BW/day. This value was set as the reference value for the calculation of the DRIs.

#### 2–2–2. Adult (EAR, RDA)

The EARs of vitamin A for those aged 18 years and above, as calculated by the multiplication of the reference value (9.3 µgRAE/kg BW/day) and the reference BW, are 550

to 650 µgRAE/day for men and 450 to 500 µgRAE/day for women.

Assuming a coefficient of variation of 20%<sup>(3)</sup>, the multiplication of these EAR values by 1.4 yields an RDA of 800 to 900 µgRAE/day ( $\hat{=}$  550 to 600  $\times$  1.4) for men and 650 to 700 µgRAE/day ( $\hat{=}$  450 to 500  $\times$  1.4) for women.

### 2–2–3. Children (EAR, RDA)

There are currently no data that can be used for the estimation of the EAR for children. The extrapolation of the reference value (9.3 µgRAE/kg BW/day) to children aged 1-5 years, based on the BW ratio, may yield a value of 150 to 200 µgRAE/day, at which plasma retinol levels are maintained below 20 µg/100 mL, thus rendering children susceptible to corneal xerosis<sup>(13)</sup>. Therefore, the RDA for those children must be above 200 µgRAE/day, for the prevention of unfavorable outcomes; the EAR for children was determined through the extrapolation of the EAR for adults aged 18 to 29 years by the 0.75<sup>th</sup> power of the BW ratio, which represents the ratio of the body surface area, considering the growth factors<sup>(3)</sup>. For the determination of the RDA for children aged less than 6 years, the DDA was calculated assuming the liver weight/BW ratio to be 42 g/kg BW<sup>(8,11)</sup>:

**DDA/BW (µgRAE/kg BW/day) = BS (20 µg/g  $\times$  42 g/kg  $\times$  10/9)  $\times$  DDR (2/100) = 18.7 µg/kg BW.**

Using the value obtained, the EAR for children aged 1 to 5 years was calculated as follows:

**EAR = 18.7 µg/kg BW/day  $\times$  reference BW  $\times$  (1 + growth factor)**

Assuming the coefficient of variation in the vitamin A requirement to be 20%<sup>(6)</sup>, the RDA was determined as the EAR  $\times$  1.4.

### 2–2–4. Additional Amount for Pregnant and Lactating Women (EAR, RDA)

The amount of vitamin A transported to the fetus through the placenta must be taken into account when estimating the vitamin A requirement for pregnant women. At a gestational age of 37 to 40 weeks, the amount of vitamin A deposited in the fetal liver was found to be 1,800 µg; therefore, the total amount of vitamin A transported to the fetus during pregnancy was estimated at 3,600 µg<sup>(14,15)</sup>. Assuming that the vitamin A absorption rate of the mother is 70%, this amount is accumulated during the last 3 months of pregnancy<sup>(15)</sup>; therefore, the additional amounts required during the early and mid-stages were not determined. The EAR for the additional vitamin A required during the late stage was determined to be 60 µgRAE/day, by rounding 56.3 µgRAE, which, assuming a coefficient of variation of 20%<sup>(3)</sup>, yields an RDA of 80 µgRAE/day (by rounding the 78.9 µgRAE/day observed during the late stage).

Based on the average concentration of vitamin A in breast milk (320 µgRAE/day), the EAR for the additional vitamin A required during lactation was estimated at 300 mg RAE/day, which, assuming a coefficient of variation of 20%<sup>(3)</sup>, yielded an RDA of 450 mg RAE/day by rounding 449 µgRAE/day.

## **2–3. Method Used to Set the Adequate Intake (AI)**

### **2–3–1. Infants (AI)**

The retinol level secreted in breast milk has been reported to be  $352 \pm 18$   $\mu\text{g/L}$  (mean $\pm$ standard deviation [SD]) at  $98 \pm 7$  days after delivery, in Japanese individuals<sup>(16)</sup>. Another study on more than 600 Japanese mothers with healthy babies reported that the vitamin A concentration in breast milk is  $525 \pm 314$   $\mu\text{gRE/L}$  (mean  $\pm$  SD)<sup>(17)</sup>. A more recent study presented precise and detailed data on the concentrations of vitamin A and  $\beta$ -carotene in the breast milk of Japanese mothers, using liquid chromatography-tandem mass spectrometry analysis<sup>(18)</sup>. This study reported that the levels are  $1,026 \pm 398$   $\mu\text{gRE/L}$  0 to 10 days after delivery,  $418 \pm 138$   $\mu\text{gRE/L}$  at 11 to 30 days,  $384 \pm 145$   $\mu\text{gRE/L}$  at 31 to 90 days,  $359 \pm 219$   $\mu\text{gRE/L}$  at 91 to 180 days, and  $267 \pm 117$   $\mu\text{gRE/L}$  at 181 to 270 days (the calculation of the RE in Reference No. 17 is similar to that of the RAE used in the current DRIs; therefore, it is used as the RAE). The concentration of  $\beta$ -carotene is high in colostrum (0.35 to 0.70  $\mu\text{mol/L}$  at 0 to 10 days after delivery), and this decreases to 0.062  $\mu\text{mol/L}$  at 3 months after delivery<sup>(16,18)</sup>. Based on the average vitamin A concentration (411 mg RAE/L)<sup>(18)</sup>, and average milk intake (0.78 L/day)<sup>(19,20)</sup>, the vitamin A intake in breast milk-fed infants aged 0 to 5 months was estimated at 320 mg RAE/day. Thus, the AI for this age group was determined to be 300 mg/day. Based on the extrapolation from the AI for infants aged 0 to 5 months, the AI for infants aged 6 to 11 months was determined to be 400 mg RAE/day by the 0.75<sup>th</sup> power of the BW ratio. The level of provitamin A carotenoids was not taken into account, as its availability is unknown.

## **3. To Avoid Excessive Intake**

### **3–1. Dietary Intake**

The use of a supplement, or having a high intake of liver has been reported to lead to health problems associated with excessive intake<sup>(3)</sup>.

### **3–2. Method Used to Set the Tolerable Upper Intake level (UL)**

An elevated plasma level of retinoic acid is considered responsible for most clinical signs<sup>(21)</sup> and symptoms of vitamin A intoxication, such as headaches. In cases of acute vitamin A intoxication, the cerebrospinal pressure is notably elevated, while increased intracranial pressure, hair loss or muscle pain is observed in cases of chronic intoxication.

Based on a study that focused on the hepatotoxicity caused by excessive vitamin A deposition<sup>(22)</sup>, the no observed adverse effect level (NOAEL) in adults was estimated at 13,500 mg RAE/day, which yielded a UL of 2,700 mg RAE/day, assuming an uncertainty factor of 5. Based on the clinical observation of increased intra-cranial pressure in infants caused by excessive vitamin A intake<sup>(23)</sup>, the NOAEL in infants was estimated at 6,000 mg RAE/day, which yielded a UL of 600 mg RAE/day, assuming an uncertainty factor of 10.

The UL for children aged 1 to 17 years was determined by the extrapolation of the UL for those aged 18 to 29 years based on the BW ratio. For safety reasons, the values for men

were applied to women because of the difference in the reference BW. The extrapolation to infants aged 1 to 2 years old yielded a UL of 500 mg RE/day, which is lower than that for infants aged 6 to 11 months (600 mg RAE/day). Thus, the UL for infants aged 1 to 2 years old was revised to 600 mg RAE/day.

Although a recent study found that ingesting approximately 1,500 mg RAE/day of retinol for 30 years doubled the fracture risk in elderly individuals<sup>(24)</sup>, data from other studies<sup>(25)</sup> contradict this finding. Thus, the determination of a separate UL for vitamin A intake in elderly individuals was not considered in the development of the present DRIs. Moreover, as the excessive intake of  $\beta$ -carotene has not been reported to be associated with the unfavorable consequences of vitamin A intoxication described above<sup>(21,24,26)</sup>, the level of provitamin A carotenoids was also not included in the estimation of the UL.

#### **4. Other Remarks**

##### **4–1. Carotenoid Intake and DRIs for Japanese**

Due to the strict regulations pertaining to their conversion into vitamin A, provitamin A carotenoids, when ingested orally, cannot cause vitamin A intoxication. Unconverted provitamin A carotenoids, as well as carotenoids that are not metabolized to vitamin A are stored *in vivo* as they are. Beneficial actions have been reported with the digestion of these carotenoids, including anti-oxidant activity and immune potentiation.

Although the results of cohort studies suggest that a higher intake of carotenoids is associated with a lower incidence of lung cancer<sup>(27)</sup>, supplementary intervention with  $\beta$ -carotene has been reported to be ineffective or even harmful in the prevention of cancer, especially lung cancer<sup>(28–31)</sup>. Regarding the benefits of specific carotenoids, the prevention of prostate cancer by lycopene<sup>(32,33)</sup>, improvement in age-related macular degeneration by lutein and zeaxanthin<sup>(34,35)</sup>, and maintenance of the retinal pigment by lutein and zeaxanthin have also been reported. The anti-oxidation of carotenoids can aid in the photoprotection of the skin<sup>(36)</sup>. Thus, further research into the efficacy and safety of carotenoids is required. In developing the present DRIs, carotenoids were not considered separately because their deficiency has not been reported.

## Vitamin D

### 1. Background Information

#### 1–1. Definition and Classification

Vitamin D<sub>2</sub> and vitamin D<sub>3</sub> are naturally occurring compounds with potent vitamin D activity. The human body obtains vitamin D from 2 sources--ultraviolet irradiation, which converts the pro-vitamin D<sub>3</sub> (7-dehydrocholesterol) in the skin to pre-vitamin D<sub>3</sub>, which in turn is converted to vitamin D<sub>3</sub> by thermal isomerization; and the dietary intake of vitamin D<sub>2</sub> and vitamin D<sub>3</sub> from sources such as mushrooms and fish, which are good sources of vitamin D<sub>2</sub> and vitamin D<sub>3</sub>, respectively. The current DRIs do not discriminate between vitamin D<sub>2</sub> and D<sub>3</sub> intakes because the compounds have similar characteristics and molecular weights, as well as biological activities that are almost equal. Therefore, the indices for the DRIs of vitamin D are based on the summation of the values of these 2 compounds.

#### 1–2. Function

Vitamin D is first metabolized to 25-hydroxy vitamin D (25OHD) before being metabolized to 1 $\alpha$ ,25-dihydroxy vitamin D (1 $\alpha$ ,25(OH)<sub>2</sub>D)--its active form. In turn, 1 $\alpha$ ,25(OH)<sub>2</sub>D combines with the vitamin D receptor in the core of the target cells and induces the gene expression of vitamin D-dependent proteins. The major actions of vitamin D include the enhancement of the absorption of calcium and phosphate in the intestine and kidneys, and stimulation of bone formation and growth. Vitamin D deficiency impairs calcium absorption from the intestine and kidney, and thus decreases calcium availability, resulting in rickets in children and osteoporosis in adults. Excess vitamin D intake causes hypercalcemia, kidney damage, or calcification disorders in the soft tissues.

#### 1–3. Digestion, Absorption and Metabolism

Although circulating 25OHD levels are dependent on the summation of the vitamin D obtained through the action of ultraviolet radiation and diet<sup>(37)</sup>, 1 $\alpha$ ,25(OH)<sub>2</sub>D is a hormone that regulates calcium metabolism and maintains its blood level in a stable range. Thus, the circulating 25OHD level is the best index of vitamin D status. As vitamin D deficiency and the resultant hypocalcemia cause elevated levels of serum parathyroid hormone (PTH), the serum concentration of PTH can also be a good index of vitamin D deficiency<sup>(38)</sup>.

### 2. To Avoid Inadequacy

#### 2–1. Factors to be Considered in Estimating Requirements

Vitamin D deficiency may result in rickets in children, and osteoporosis in adults. In adults, especially elderly adults, even so-called “vitamin D insufficiency,” which is milder than vitamin D deficiency, can result in decreased bone mineral density.

The current US-Canada DRIs determined the EARs and RDAs for calcium and vitamin

D, while their previous DRIs determined the AIs for both<sup>(39)</sup>. As vitamin D can be induced in the skin through ultraviolet irradiation, there is a lack of scientific evidence on the dose-response relationship between vitamin D intake and the maintenance of bone health. The US-Canada DRIs determined the 25OHD level to be a good index of vital vitamin D, produced through both diet and ultraviolet irradiation. A 25OHD level lower than 30 nmol/L increases the risk of rickets in children and osteoporosis in adults, decreases the absorption of calcium in children and adults, decreases the bone mineral density in children and adults, and increases the prevalence of bone fractures in elderly individuals. For the prevention of bone fractures, based on the maximal effects obtained at 50 nmol/L, the amount of 50% for the requirement was determined to be 40 nmol/L and the amount of 97.5% for the requirement was determined to be 50 nmol/L. These values were determined under circumstances of minimal exposure to sunlight. However, in Japan, few studies have reported data on both serum 25OHD levels and the intake of vitamin D in the same set of individuals, making it difficult to determine an EAR and RDA based on the method stated above.

A cohort study conducted in Nagano, which followed-up 1,470 post-menopausal women ( $63.7 \pm 10.7$ ) for a mean of 7.2 years, showed that 49.6% of the participants had serum 25OHD levels lower than 50 nmol/L<sup>(40)</sup>. The relative risk for long-bone fractures was 2.20 (95% confidence interval [CI] 1.37-3.53) in those who had serum 25OHD levels lower than 62.5 nmol/L compared to the reference (62.5 nmol/L or higher), showing that vitamin D deficiency could increase the risk of osteoporosis-related bone fractures.

Although no intervention studies have been conducted for the prevention of bone fractures in Japan, many such studies have been conducted in other countries. Some of those studies reported that, while a vitamin D intake of 10 µg/day is insufficient, taking more than 17.5 µg/day may inhibit femur fractures<sup>(41,42)</sup>. A recent meta-analysis reported that the effects of vitamin D supplementation on bone mineral density were limited to femur bone, and the effect was insignificant<sup>(43)</sup>. Thus, the results are inconsistent.

## **2-2. Method Used to Set the AI**

### **2-2-1. Adults (AI)**

In the US-Canada DRIs, the EAR (10 µg/day for adults aged over 19 years) and RDA (15 µg/day for adults aged over 19 years and 20 µg/day for adults aged over 70 years) were determined as stated above. However, these values were determined without consideration of the production due to ultraviolet irradiation, and therefore, cannot be applied to the AI in the present DRIs.

One study focused on the duration of sun exposure required to produce 5.5 µg of vitamin D<sub>3</sub>, with the arms and face uncovered, in 3 areas of Japan (Sapporo, Tsukuba and Naha). This study reported that, while vitamin D<sub>3</sub> may be produced even in winter in Naha, this is not the case in Sapporo in December, except around noon<sup>(44)</sup>. Based on the results for Sapporo in December, the production of 5.5 µg took 94.7 minutes at noon, without limitation for sunny

days; this suggests that approximately 7.5 µg of vitamin D<sub>3</sub> can be produced through 2 hours of sun exposure around noon. Thus, the required amount was calculated by subtracting the amount produced by sun exposure (7.5 µg) from the daily requirement (15 µg), yielding 7.5 µg. However, there is no evidence on the daily sun exposure among healthy Japanese adults, and a study reported that 25OHD levels may be affected by walking outdoors in addition to vitamin D intake<sup>(45)</sup>. Many factors should be taken into account. Therefore, the AI values of the previous DRIs could not be updated based on the scientific evidence, and the AI for adults was set at 5.5 µg/day. Due to a lack of data, the AI values for both men and women were the same.

Osteoporosis increases the risk of bone fractures at various sites, and vitamin D deficiency increases the risk of non-vertebral body fractures, especially femur bone fractures, and these bone fractures predominantly occur in elderly individuals<sup>(46)</sup>. In Japan, a high prevalence of fractures was observed in vitamin D-deficient elderly individuals<sup>(40,47)</sup>. Moreover, an intervention study conducted on elderly individuals in a nursing home, who had limited access to sun exposure, showed that 5 µg/day of supplementation was insufficient<sup>(48)</sup>, and in only 40% of the individuals, 50 nmol/L of serum 25OHD was maintained, with 20 µg/day of supplementation<sup>(49)</sup>. Based on these results, the Japanese 2011 guidelines for the prevention and treatment of osteoporosis 2011 (Japan Osteoporosis Society) recommends an intake of 10 to 20 µg/day (Grade B)<sup>(46)</sup>. However, as most of the aforementioned studies targeted elderly individuals in nursing homes<sup>(47-49)</sup>, their results cannot be generalized to the elderly population as a whole. Therefore, the AI was set as 5.5 µg/day for those aged over 70 years, as well as for those aged 18 to 69 years.

A vitamin D intake that is higher than the AI set above may be favorable for those living in areas with shorter daylight hours in winter, those whose performance of outdoor activities is severely limited, or elderly individuals. It is difficult to set these recommendation values at this time.

### **2-2-2. Children (AI)**

One study evaluated the intake of vitamin D and plasma 25OHD levels in 1,380 Japanese children aged 12 to 18 years (672 boys and 718 girls)<sup>(50)</sup>. The study reported that the mean intake of vitamin D was approximately 10 µg/day, and median plasma 25OHD level was 50 nmol/L, regardless of age or sex. As findings on the relationship between vitamin D intake and plasma 25OHD concentration in children are limited, they were considered unsuitable for the setting of the vitamin D AI for children. Thus, the AI for children was determined through the extrapolation of the AI for adults by the 0.75<sup>th</sup> power of the BW ratio and the growth factors.

### **2-2-3. Infants (AI)**

In infants, the development of rickets due to vitamin D deficiency is commonly observed both in Japan and other countries<sup>(51-53)</sup>, suggesting that limited sun exposure or breast feeding are risk factors. In Japan, although the prevalence rate of rickets is unclear, in an

epidemiological study conducted in Kyoto, 22% of neonates were found to have craniotabes, a mineralization defect of the bone likely caused by vitamin D deficiency<sup>(54)</sup>. The incidence of craniotabes exhibited seasonal variation, with a peak and nadir between January and May, and between July and November, respectively. The circulating 25OHD levels were found to be below 25 nmol/L in 37% of all neonates diagnosed with craniotabes, at 1 month after birth.

In breast milk-fed newborn infants, the serum concentration of 25OHD was found to be lower than 25 nmol/L in 57% of the population, and below 12.5 nmol/L in 17%. In contrast, none of the formula-fed infants or those with a combination feed were found to have an inadequate serum 25OHD level. It should be noted that neonates born in a vitamin D-deficient state may not recover to a vitamin D-sufficient state within a short period, and that the serum 25OHD level of breast milk-fed infants decreases further during the winter months<sup>(55)</sup>, indicating that the vitamin D delivered from breast milk may be unsatisfactory. Similar cases were reported in Iowa, US. The rates of breast milk-fed infants without vitamin D supplementation who showed plasma 25OHD levels lower than 27.5 nmol/L were 70%, 57%, 33% and 23% at 112, 168, 224 and 280 days after birth, respectively<sup>(56)</sup>.

The concentration of vitamin D in breast milk, including the active metabolite, is 3.0 µg/L in Japanese individuals<sup>(17)</sup>. Although a recent study using a novel, highly accurate procedure found the average vitamin D concentration in breast milk to be only 0.6 mg/L<sup>(18)</sup>, no consequent data followed. In contrast, a concentration of 0.3 µg/100 g was reported in the Standard Tables of Food Composition in Japan, 2010, through the use of a traditional method<sup>(57)</sup>. The concentrations of vitamin D and its metabolite, which has vitamin D activity, depend on the vitamin D nutrition status of lactating mothers, lactating stage, or season. Therefore, an AI was set for the prevention of rickets.

Breast milk-fed infants with a low degree of sun exposure are at a higher risk of developing rickets<sup>(58)</sup>. Considering that previous research found that infants did not develop rickets after supplementation with 2.5, 5 or 10 µg/day of vitamin D for 6 months, and assuming that infants receive an average of 2.38 µg/day of vitamin D from breast milk, the daily intakes would be 4.88, 7.38 or 12.38 µg/day of vitamin D; therefore, an intake of 4.88 µg/day is satisfactory for the prevention of rickets.

Based on these data, the AI of vitamin D for infants aged 0 to 5 months was determined to be 5 µg/day. The 2003 guideline of the American Pediatric Society set the requirement intake at 5 µg/day for the prevention of rickets<sup>(59)</sup>; in the updated guideline (2008), the value was set at 10 µg/day<sup>(60)</sup>. However, considering that this value requires vitamin D supplementation, and the fact that the actual compliance to this guideline is low<sup>(61)</sup>, the present DRIs did not adopt this value.

The average plasma 25OHD level of 150 18-month-old infants was reported to be higher than 25 nmol/L, and their vitamin D intakes were 8.6 and 3.9 µg/day at 6 months and 12 months, respectively<sup>(62)</sup>. In Norway, infants fed 10 µg/day of vitamin D in winter (the amount of milk intake is unknown) had plasma 25OHD levels at nearly the median of the level

measured in late summer and the level of infants who were fed infant formula. Based on the average milk intake of Japanese infants who were fed infant formula (0.8 L/day)<sup>(63)</sup>, this amount would be equivalent to 8 µg/day. However, achieving this value requires vitamin D supplementation. Moreover, under circumstances of appropriate sun exposure, lower intakes are not considered a severe risk of deficiency.

The AI of vitamin D for 6-11-month-old infants with adequate sun exposure was determined to be 5 µg/day. This value was also applied to infants aged 6 to 11 months with limited sun exposure, due to a lack of evidence on AI determination.

#### **2–2–4. Pregnant and Lactating Women (AI)**

During pregnancy, the requirement of calcium and capacity to produce 1 $\alpha$ ,25(OH)<sub>2</sub>D increases; this decreases after delivery. In a study of pregnant women with limited sun exposure, an inadequate serum 25OHD concentration was observed in those with an average vitamin D intake of 0.75 to 5.3 µg/day<sup>(64)</sup>, but not in those with an average vitamin D intake higher than 7 µg/day<sup>(65)</sup>. As these findings indicate that pregnant women require at least 7 µg/day of vitamin D, the AI of vitamin D required for pregnant women was determined to be 7 µg/day.

For lactating women, as the AI for infants was set based on the prevention of rickets and since rickets and hypocalcemia resulting from vitamin D deficiency have been reported among breast-milk-fed infants<sup>(53)</sup>, the additional amount of vitamin D required for lactating women was determined to be 2.5 µg/day, by multiplying the concentration of vitamin D (including that of the active metabolite) in breast milk (3.0 µg/L)<sup>(17)</sup> and the average milk intake (0.78 L/day)<sup>(19,20)</sup>. The AI for lactating women was set as 8.0 µg/day by adding the additional value to the AI for adults aged over 18 years.

### **3. To Avoid Excessive Intake**

#### **3–1. Dietary Intake**

The production of vitamin D in the skin due to ultraviolet irradiation is controlled, and excess vitamin D is not produced. Thus, an excessive intake of vitamin D is unlikely. Vitamin D is activated in the liver and kidneys in which activation is strictly controlled. Once hypercalcemia occurs, further activation is regulated.

#### **3–2. Method Used to Set the UL**

A prolonged intake of excessive quantities of vitamin D can lead to unfavorable outcomes, such as hypercalcemia, renal dysfunction, soft tissue calcification, and growth retardation. As an increased plasma 25OHD level by itself does not directly cause health problems, the presence of hypercalcemia rather than a high serum 25OHD level, is considered an appropriate indicator for the determination of the UL. For infants, excessive vitamin D intake can cause growth retardation, and this is set as an unfavorable outcome in some studies.

### **3–2–1. Adults (UL)**

In an intervention study that included 10, 20, 30, 60, and 95 µg/day of vitamin D supplementation for 3 months, the serum calcium concentration was found to exceed the reference value in some participants receiving 95 µg/day of vitamin D, but not in those receiving 60 µg/day of vitamin D<sup>(66)</sup>. However, this study had a small sample size, limited to patients with granulomatous diseases that cause hypercalcemia. As no other study has reported the presence of hypercalcemia at vitamin D intake levels lower than 250 µg/day, the NOAEL was determined to be 250 µg/day. This value was divided by an uncertainty factor of 1.2 based on the US-Canada DRIs<sup>(39)</sup>, yielding a UL of 100 µg/day for adults. Since some case reports pointed to the presence of hypercalcemia at a vitamin D intake level of 1,250 µg/day<sup>(67,68)</sup>, using this value as the lowest observed adverse effect level (LOAEL) and an uncertainty factor of 10, a similar value was calculated. Age and sex-related differences were not considered. There is currently no evidence for the setting of other ULs for elderly individuals; therefore, the UL for them was set as the same value as that for adults<sup>(39)</sup>.

An intervention study on pregnant women reported no unfavorable outcomes including hypercalcemia at intake levels lower than 100 µg/day<sup>(69)</sup>. Since there is evidence on pregnant or lactating women having a higher risk of hypercalcemia, the UL for these individuals was set at 100 µg/day<sup>(39,70)</sup>.

### **3–2–2. Infants (UL)**

Based on a study that observed no growth retardation in infants administered an average of 44 (34.5–54.3) µg/day of vitamin D for 6 months<sup>(71)</sup>, the NOAEL for infants was determined to be 44 µg/day, which, assuming an uncertainty factor of 1.8, yielded a UL of 24.4 µg/day in the US-Canada DRIs<sup>(39)</sup>. Accordingly, the UL for infants was set at 25 µg/day.

### **3–2–3. Children (UL)**

As relevant data are unavailable for this age group, the UL for children was determined by extrapolating the UL values for adults aged 18 to 29 years (100 µg/day), and infants (25 µg/day) based on the reference BW. The calculation was conducted by age and sex, and the lowest values in each age group were adopted.

## **4. For the Prevention of the Development and Progression of Lifestyle-Related Diseases**

Vitamin D deficiency is a risk factor for bone fracture, and is indicated to be associated with various lifestyle-related diseases. Since these data remain insufficient, the present DRIs did not consider these associations.

## **5. Future Dietary Reference Intakes for Japanese Individuals**

Reliable data on the sun exposure hours of Japanese individuals, and the association between daily vitamin D intake and plasma 25OHD levels are required.

## Vitamin E

### 1. Background Information

#### 1–1. Definition and Classification

Vitamin E is composed of 8 analogues:  $\alpha$ -,  $\beta$ -,  $\gamma$ - and  $\delta$ -forms, of tocopherol and tocotrienol. Only  $\alpha$ -tocopherol presents in the human blood and various tissues. Therefore,  $\alpha$ -tocopherol was considered in the current DRIs.

#### 1–2. Function

Vitamin E is located in the phospholipid bilayer of the cell membrane, and prevents the propagation of the lipid peroxidation of unsaturated fatty acid or other components of the biological membrane. Animal studies have shown that vitamin E deficiency causes not only infertility, but also cerebromalacia, hepatic necrosis, kidney disorder, hemolytic anemia, and muscular dystrophy. Excess vitamin E can increase an individual's bleeding tendency. An intake of regular food prevents vitamin E deficiency or excess.

#### 1–3. Digestion, Absorption and Metabolism

Digested vitamin E analogues are absorbed via the intestine to the lymphatic system after forming micelles due to bile acid. The absorption rate of this vitamin has been estimated to be 51 to 86 %<sup>(72)</sup>; however, one study reported a rate of only 21 to 29%<sup>(73)</sup>. The actual absorption rate of vitamin E in humans is unknown. After intestinal absorption, vitamin E is packaged into chylomicron, transformed into chylomicron remnants by lipoprotein lipase, and transported to the liver. Of the 8 analogues, only  $\alpha$ -tocopherol is preferentially bound to  $\alpha$ -tocopherol-binding proteins, while the other analogues are metabolized in the liver.  $\alpha$ -tocopherol is then transformed into very low-density lipoprotein (VLDL), and distributed again in the blood flow<sup>(74)</sup>.

### 2. To Avoid Inadequacy

#### 2–1. Factors to be Considered in Estimating Requirements

Erythrocytes are susceptible to hemolysis by hydrogen peroxide when the circulating  $\alpha$ -tocopherol level is between 6 and 12 mmol/L<sup>(75)</sup>, when the plasma  $\alpha$ -tocopherol level is 16.2  $\mu$ mol/L (697  $\mu$ g/dL). However, they are resistant to hemolysis when the serum  $\alpha$ -tocopherol level is higher than 14 mmol/L<sup>(76)</sup>.

An intervention study that administered graded doses (0 to 320 mg/day) of vitamin E to vitamin E-deficient participants reported that 12  $\mu$ mol/L of circulating  $\alpha$ -tocopherol corresponds to 12 mg/day of vitamin E intake<sup>(77)</sup>. These values were not considered appropriate for the estimation of EAR and RDA values, as they were collected many years ago.

## 2–2. Method Used to Set the AI

### 2–2–1. Calculation Method Used for the AI

Several studies that analyzed vitamin E intake and circulating  $\alpha$ -tocopherol levels consistently reported that the average serum  $\alpha$ -tocopherol level exceeded 22  $\mu\text{mol/L}$  in all study populations<sup>(78–80)</sup>. The average vitamin E intake in those studies ranged from 5.6 to 11.1 mg/day, a range that encompasses the values of the 2010 and 2011 National Health and Nutrition Survey (NHNS)<sup>(81)</sup> of median vitamin E intakes (6.2–6.8 mg/day in men and 5.5–6.6 mg/day in women), in each sex and age group. As these findings indicate that the median intake of Japanese individuals likely yields an adequate vitamin E status, the AI values were determined to be the 2010 and 2011 NHNS median values, stratified by sex and age group<sup>(81)</sup>.

### 2–2–2. Adults (AI)

As described above, the AI was determined to be 6.5 mg/day for men and 6.0 mg/day for women, using the weighted average of the median values of the 2010 and 2011 NHNS<sup>(81)</sup>, stratified by sex and age group, as these values are expected to yield a blood  $\alpha$ -tocopherol level exceeding 12  $\mu\text{mol/L}$ . As aging has not been reported to be associated with the compromised absorption or utilization of vitamin E, the same values were applied to elderly individuals.

**Table 1. Reports of the association between blood  $\alpha$ -tocopherol level and dietary intake among healthy Japanese subjects**

Reference No.	Gender	n	Age (years)	Serum $\alpha$ -tocopherol level ( $\mu\text{mol/L}$ ) <sup>1</sup>	Dietary intake (mg/day) <sup>1</sup>	NHNS <sup>2</sup>	
						Age (years)	Intake (mg/day)
78	male	42	31-58	25.4 $\pm$ 5.6	11.1 $\pm$ 4.9	30-49	6.2
	female	44	24-67	31.8 $\pm$ 10.5	9.5 $\pm$ 3.9	30-49	5.7
79	female	150	21-22	32.0 $\pm$ 10.5	7.0 $\pm$ 2.4 <sup>3</sup>	18-29	5.5
80	female	10	21.6 $\pm$ 0.8	22.2 $\pm$ 2.2	7.1 $\pm$ 2.0 <sup>4</sup>		
		11	21.2 $\pm$ 0.8	26.3 $\pm$ 4.2	6.2 $\pm$ 2.4 <sup>4</sup>		
		10	21.0 $\pm$ 0.7	28.5 $\pm$ 3.6	5.6 $\pm$ 2.0 <sup>4</sup>		

<sup>1</sup> Mean  $\pm$  standard deviation

<sup>2</sup> Dietary intake value among those in relevant age categories reported in NHNS2010, 2011

<sup>3</sup>  $\alpha$ -tocopherol equivalent

<sup>4</sup>  $\alpha$ -tocopherol values calculated from  $\alpha$ -tocopherol intake (mg/kg BW/day) and mean BW (kg).

### 2–2–3. Children (AI)

As data were unavailable for this age group, the median values of the 2010 and 2011 NHNS for children, stratified by sex and age group, were used as the bases for the determination of the AI for children, like in the case of adults. For children aged under 12 years, as there is little difference in body height and weight between boys and girls, the average value of the boys and girls was set as the AI.

#### **2–2–4. Infants (AI)**

The concentration of vitamin E in breast milk gradually decreases, from 6.8 to 23 mg/L in colostrum, and 1.8 to 9 mg/L in mature milk<sup>(82)</sup>. This is not associated with full-term birth or preterm birth, and does not show diurnal variations<sup>(83)</sup>. The AI for infants aged 0 to 5 months was determined to be 3.0 mg/day by multiplying the average  $\alpha$ -tocopherol concentration in the breast milk of Japanese individuals (3.5 to 4.0 mg/L)<sup>(17,18)</sup> and the average milk intake (0.78 L/day)<sup>(19,20)</sup>.

The AI for infants aged 6 to 11 months old was determined to be 4.0 mg/day through the extrapolation of the adult value by the 0.75<sup>th</sup> power of the BW ratio, yielding 3.85 mg/day for boys and 3.80 mg/day for girls.

#### **2–2–5. Pregnant or Lactating Women (AI)**

During pregnancy, the elevated blood lipid level increases the blood  $\alpha$ -tocopherol level<sup>(84)</sup>. The AI for pregnant women was determined to be 6.5 mg/day using the median value of the 2007-2011 NHNS (6.25 mg/day)<sup>(85)</sup> for pregnant women, as there are no reports on vitamin E deficiency during pregnancy. For lactating women, the AI was determined to be 7.0 mg/day using the median value of the 2007-2011 NHNS (6.55 mg/day)<sup>(85)</sup> for lactating women.

### **3. To Avoid Excessive Intake**

#### **3–1. Dietary Intake**

The intake of regular foods does not lead to vitamin E deficiency or excess.

#### **3–2. Method Used to Set the UL**

The basis for determining the UL for vitamin E is its possible effect on an individual's bleeding tendency. Based on the finding that supplementation with 800 mg/day of  $\alpha$ -tocopherol for 28 days did not increase the bleeding tendency in healthy men (average BW, 62.2 kg)<sup>(86)</sup>, the NOAEL was determined to be 800 mg/day. Assuming an uncertainty factor of 1.0, and considering that no data regarding LOAEL are available, the sex- and age-group-stratified UL was calculated by correcting the 800 mg/day value by the BW ratio. As few data are available regarding the UL for infants, and because typical feeding with breast milk or baby food does not cause excessive intake, the UL was not determined for infants.

### **4. For the Prevention of the Development and Progression of LRDs**

Although numerous intervention studies have examined the effect of vitamin E supplementation on the risk of coronary heart diseases, the findings are inconsistent<sup>(87–90)</sup>. A recent study showed an association between excessive vitamin E intake and osteoporosis<sup>(91)</sup>; however, we did not use these data, as the study was conducted on animals, and there are no clinical data supporting this.

## Vitamin K

### 1. Background Information

#### 1–1. Definition and Classification

Naturally occurring vitamin K consists of phyloquinones (PKs; vitamin K<sub>1</sub>) and menaquinones (MKs; vitamin K<sub>2</sub>). Phyloquinones have a phytyl group in the side chain. Menaquinones are further subdivided into 11 analogues depending on the number of isoprene units (4–14) in the prenyl side chain. Among the menaquinones, of nutritional importance are menaquinone-4 (MK-4), which is ubiquitously present in animal foods, and menaquinone-7 (MK-7), which is abundantly present in natto, a traditional Japanese food made from soybeans fermented with *Bacillus subtilis*. At present, data on the determination of the relative biological activity of these analogues are scarce, and no corrections have been made for PKs and MK-4, which have similar molecular weights. MK-7, which has a much larger molecular weight, can be converted into its MK-4 equivalent using the following formula:

$$\text{MK-4 equivalent (mg)} = \text{MK-7 (mg)} \times 444.7/649.0$$

The sum of the quantity of PK, MK-4, and MK-7 as corrected above was employed in determining the DRIs for vitamin K.

#### 1–2. Function

The principal biological action of vitamin K is the activation of prothrombin and other serum coagulation factors, for the enhancement of blood coagulation. Other actions include the modulation of bone formation by the activation of osteocalcin, a bone matrix protein, and inhibition of arterial calcification by the activation of matrix gla protein (MGP), another vitamin-K-dependent matrix protein. Vitamin K deficiency delays blood coagulation. The average dietary pattern of Japanese individuals does not lead to vitamin deficiency.

#### 1–3. Digestion, Absorption and Metabolism

Although long-chain MKs are produced by intestinal bacteria<sup>(92)</sup>, and MK-4 is also produced by enzymatic conversion from PK<sup>(93)</sup>, it is not known to what extent these amounts fulfill requirements. As an intake of 0.8 to 1.0 µg/day kg BW of PK from the regular diet can cause potential vitamin K deficiency<sup>(94)</sup>, the production of MKs by intestinal bacteria in various tissues is not considered to be large enough to contribute to the fulfillment of this requirement.

## 2. To Avoid Inadequacy

### 2–1. Factors to be Considered in Estimating Requirements

Delayed blood coagulation is the only clinically manifested abnormality attributable to vitamin K deficiency. In Japan, coagulation abnormalities due to vitamin K deficiency are rarely observed in healthy individuals. The requirement of vitamin K is considered to be almost fulfilled, except in the case of patients after surgery or those receiving warfarin. The amount of

vitamin K intake required for the activation of blood coagulation is unknown. An intervention study of male volunteers (28.3±3.2 years old) provided vitamin K-deficient meals for 40 days<sup>(94)</sup>; however, the sample size was small, and so the data cannot be used for the determination of the DRIs.

A recent cohort study examined the association between femur bone fractures and vitamin K intake, and reported that individuals taking 100 µg/day or more of vitamin K had a lower risk of fractures than those with a lower intake<sup>(95,96)</sup>. Under-γ-carboxylated osteocalcin (ucOC) has been considered as an indicator of suboptimal vitamin K status, and is reported as an independent risk factor for bone fractures. A much greater amount of vitamin K (more than 500 µg/day) is required to decrease ucOC levels than to activate the coagulation factor in the liver<sup>(97,98)</sup>. The vitamin K intake required for the prevention of bone fractures may be increased, compared to when plasma uncarboxylated prothrombin is used as an index. A meta-analysis reported that vitamin K supplementation decreased the number of bone fracture events, as a result of high doses of MKs (45 mg/day) being administered<sup>(99)</sup>.

Based on these findings, the vitamin K requirement for the prevention of bone fractures is considered to be greater than the amount required for the maintenance of normal blood coagulation function. Here, AIs were set due to a lack of scientific evidence.

## **2–2. Method Used to Set the AI**

### **2–2–1. Adults (AI)**

As described above, the vitamin K intake of healthy Japanese individuals is considered to be almost sufficient. The average values of vitamin K intake from the 2010 and 2011 NHNS data were 185 µg/day and 280 µg/day, respectively<sup>(81)</sup>. The intake of natto has a significant impact on vitamin K intake in Japanese individuals. The vitamin K intakes have been reported to be 336.2±138.2 µg/day and 154.1±87.8 µg/day for those who consumed natto and those did not, respectively<sup>(100)</sup>. As remarkable unfavorable outcomes have not been reported among those who do not consume natto, based on the value above, 150 µg/day was set as the AI.

Elderly individuals may be more susceptible to vitamin K deficiency due to various factors such as impaired intestinal absorption of vitamin K due to the decreased secretion of bile salts and pancreatic juice, or decreased dietary fat intake. Additionally, the presence of chronic diseases or use of antibiotics decreases the MK production in the intestine, and vitamin K activity due to the inhibition of the activity of vitamin K epoxide reductase. Therefore, the AIs for elderly individuals are considered to be elevated, with a study reporting that these individuals require a higher level of vitamin K<sup>(101)</sup>; however, at present, relevant data are scarce, and thus, the AI for elderly individuals--aged above 69 years--was the same as that for those aged 50 to 69 years.

### **2–2–2. Children (AI)**

The AI for children was determined by extrapolating the AI for adults by the 0.75<sup>th</sup>

power of the BW ratio.

### **2–2–3. Infants (AI)**

The average concentration of vitamin K in the breast milk of Japanese mothers has been reported to be 5.17 µg/L<sup>(102)</sup>. Recent studies showed that the concentrations were 3.771 ng/mL for PK, and 1.795 ng/mL for MK-7, using newly-developed measurement methods<sup>(18)</sup>. Newborn infants are susceptible to vitamin K deficiency for various reasons, such as poor transplacental vitamin K transport<sup>(103)</sup>, low vitamin K content in the breast milk<sup>(18,102)</sup>, or low production of vitamin K in the intestinal flora<sup>(102)</sup>. As neonatal vitamin K deficiency is known to cause neonatal melena, a form of gastrointestinal bleeding, and intracranial bleeding, vitamin K is orally administered just after birth, for their prevention<sup>(104)</sup>. An AI of 4.0 mg/day for infants aged 0-5 months was determined by multiplying the average milk intake (0.78 L/day)<sup>(19,20)</sup> and the average vitamin K content of milk (5.17 µg/L), and assuming the presence of the oral administration of vitamin K just after birth in clinical settings. For infants aged 6 to 11 months, the AI was determined to be 7 µg/day, considering the amount of vitamin K received from sources other than breast milk.

### **2–2–4. Pregnant or Lactating Women (AI)**

There is no evidence stating that the vitamin K requirement is increased or that the circulating vitamin K levels are altered in pregnant women. Due to poor transplacental transport, the vitamin K intake in pregnant women is unlikely to affect the vitamin K status of fetuses or neonates. The requirements are technically the same for women who are pregnant and those who are not, and therefore, fulfilling the AI for women who are not pregnant should be sufficient for the prevention of vitamin K deficiency. Thus, the AI for pregnant women was determined to be 150 µg/day.

Since lactating women are not at a higher risk for vitamin K deficiency, the AI for them was determined to be 150 µg/day.

## **3. To Avoid Excessive Intake**

Although menadione--a vitamin K metabolite--can cause toxicity, no toxicity has been reported with regards to PKs and MKs. As 45 mg/d of MK-4 is clinically administered to many patients in Japan with osteoporosis, with no reports of serious adverse events<sup>(46)</sup>, the UL for vitamin K was not determined.

## **4. For the Prevention of the Development and Progression of LRDs**

Vitamin K deficiency has been reported to increase the risk of bone fractures. However, due to a lack of scientific evidence on its preventive effect against bone fracture in intervention studies, DGs were not determined.

II Energy and Nutrients  
 Vitamins (1) Fat-soluble Vitamins

**DRIs for Vitamin A (µg RAE/day) <sup>1</sup>**

Gender	Males				Females			
Age etc.	EAR <sup>2</sup>	RDA <sup>2</sup>	AI <sup>3</sup>	UL <sup>3</sup>	EAR <sup>2</sup>	RDA <sup>2</sup>	AI <sup>3</sup>	UL <sup>3</sup>
0-5 months	—	—	300	600	—	—	300	600
6-11 months	—	—	400	600	—	—	400	600
1-2 years	300	400	—	600	250	350	—	600
3-5 years	350	500	—	700	300	400	—	700
6-7 years	300	450	—	900	300	400	—	900
8-9 years	350	500	—	1,200	350	500	—	1,200
10-11 years	450	600	—	1,500	400	600	—	1,500
12-14 years	550	800	—	2,100	500	700	—	2,100
15-17 years	650	900	—	2,600	500	650	—	2,600
18-29 years	600	850	—	2,700	450	650	—	2,700
30-49 years	650	900	—	2,700	500	700	—	2,700
50-69 years	600	850	—	2,700	500	700	—	2,700
70+ years	550	800	—	2,700	450	650	—	2,700
Pregnant women (additional)	/							
Early-stage					+0	+0	—	—
Mid-stage					+0	+0	—	—
Late-stage					+60	+80	—	—
Lactating women (additional)	/				+300	+450	—	—

<sup>1</sup> Retinol activity equivalent (µgRAE)  
 = retinol (µg) + β-carotene (µg) × 1/12 + α-carotene (µg) × 1/24 + β-cryptoxanthin (µg) × 1/24 + other provitamin A carotenoids (µg) × 1/24

<sup>2</sup> Includes provitamin A carotenoids.

<sup>3</sup> Excludes provitamin A carotenoids.

II Energy and Nutrients  
 Vitamins (1) Fat-soluble Vitamins

**DRIs for Vitamin D (µg/day)**

Gender	Males		Females	
Age etc.	AI	UL	AI	UL
0-5 months	5.0	25	5.0	25
6-11 months	5.0	25	5.0	25
1-2 years	2.0	20	2.0	20
3-5 years	2.5	30	2.5	30
6-7 years	3.0	40	3.0	40
8-9 years	3.5	40	3.5	40
10-11 years	4.5	60	4.5	60
12-14 years	5.5	80	5.5	80
15-17 years	6.0	90	6.0	90
18-29 years	5.5	100	5.5	100
30-49 years	5.5	100	5.5	100
50-69 years	5.5	100	5.5	100
70+ years	5.5	100	5.5	100
Pregnant women			7.0	—
Lactating women			8.0	—

II Energy and Nutrients  
 Vitamins (1) Fat-soluble Vitamins

**DRIs for Vitamin E (mg/day) <sup>1</sup>**

Gender	Males		Females	
Age etc.	AI	UL	AI	UL
0-5 months	3.0	—	3.0	—
6-11 months	4.0	—	4.0	—
1-2 years	3.5	150	3.5	150
3-5 years	4.5	200	4.5	200
6-7 years	5.0	300	5.0	300
8-9 years	5.5	350	5.5	350
10-11 years	5.5	450	5.5	450
12-14 years	7.5	650	6.0	600
15-17 years	7.5	750	6.0	650
18-29 years	6.5	800	6.0	650
30-49 years	6.5	900	6.0	700
50-69 years	6.5	850	6.0	700
70+ years	6.5	750	6.0	650
Pregnant women			6.5	—
Lactating women			7.0	—

<sup>1</sup> Calculated for  $\alpha$ -tocopherol. These do not include vitamin E other than  $\alpha$ -tocopherol.

II Energy and Nutrients  
 Vitamins (1) Fat-soluble Vitamins

**DRIs for Vitamin K (µg/day)**

Gender	Males	Females
Age etc.	AI	AI
0-5 months	4	4
6-11 months	7	7
1-2 years	60	60
3-5 years	70	70
6-7 years	85	85
8-9 years	100	100
10-11 years	120	120
12-14 years	150	150
15-17 years	160	160
18-29 years	150	150
30-49 years	150	150
50-69 years	150	150
70+ years	150	150
Pregnant women	/	150
Lactating women		150

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## (2) Water-Soluble Vitamins

### Vitamin B<sub>1</sub>

#### 1. Background Information

##### 1–1. Definition and Classification

The chemical name of vitamin B<sub>1</sub> is thiamin, and the present DRIs for vitamin B<sub>1</sub> were set as the amount of thiamin hydrochloride; 2-[3-[(4-amino-2-methyl-pyrimidin-5-yl) methyl]-4-methyl]-thiazole-5-yl] ethanol. There are 3 types of thiamin hydrochloride: thiamin monophosphate (TMP), thiamin diphosphate (TDP), and thiamin triphosphate (TTP), and their activity is equimolar to that of vitamin B<sub>1</sub>.

##### 1–2. Function

Vitamin B<sub>1</sub> is involved in the metabolism of glucose and branched-chain amino acids. Insufficient vitamin B<sub>1</sub> intake can cause neuritides or brain damage. Beriberi and Wernicke-Korsakoff syndrome are well-known for being caused by vitamin B<sub>1</sub> deficiency.

##### 1–3. Digestion, Absorption and Metabolism

Vitamin B<sub>1</sub> predominantly exists as TDP in living cells in combination with enzyme proteins. TDP is dissociated from proteins through cooking or digestion, and, thereafter, the released TDP undergoes phosphorylation to become thiamin. Thiamin absorption occurs through an active transportation system in the jejunum and ileum. These processes can be affected by the type of food, and other dietary sources. The relative bioavailability of free vitamin B<sub>1</sub> has been reported to be around 60% in the typical Japanese diet<sup>(1,2)</sup>.

#### 2. To Avoid Inadequacy

##### 2–1. Factors to be Considered in Estimating Requirements

There should be a difference in the requirement estimation between the calculation using the dietary intake required for recovery from deficiency, and that using the association between dietary intake and urinary excretion of vitamin B<sub>1</sub>.

##### 2–1–1. Estimation of the Dietary Intake Required for Recovery from Deficiency

It was reported that recovery from vitamin B<sub>1</sub> deficiency (caused by the intake of meals with lower than 0.03 mg/day of vitamin B<sub>1</sub>) occurred through an intake of 0.7 mg/day of thiamin hydrochloride, in male Japanese student volunteers<sup>(3)</sup>. Considering the relative bioavailability (60%), an intake of 1.17 mg/day of dietary vitamin B<sub>1</sub> is the yield from 0.7 g/day of thiamin hydrochloride. The experimental meals were set at 2,400 kcal/day, and the requirement of dietary vitamin B<sub>1</sub> intake, in the form of thiamin hydrochloride, was considered to be lower than 0.49/1,000 kcal.

### **2–1–2. Estimation of Requirements from Urinary Thiamin Excretion**

Orally administered thiamin is rapidly converted to TDP in the body tissues. Thereafter, excess thiamin is excreted in a free form in the urine. The values obtained through the calculation of excess thiamin may be higher than those required to prevent deficiencies.

The urinary excretion of thiamin sharply increases at an intake of 0.35 mg/1,000 kcal/day of vitamin B<sub>1</sub><sup>(4)</sup>. This value can be considered the body requirement, as the urinary excretion of thiamin increases sharply when the body's requirement is met.

### **2–2. Method Used to Set the Estimated Average Requirement (EAR) and Recommended Dietary Allowance (RDA)**

These values were determined as amounts per energy intake.

#### **2–2–1. Adults and Children (EAR and RDA)**

The DRIs adopted the values obtained from the relation of the inflection points of vitamin B<sub>1</sub> intake and vitamin B<sub>1</sub> excretion. As it is a water-soluble vitamin, excess thiamin is excreted in its free form in the urine. Thus, the EAR of vitamin B<sub>1</sub> was determined as the point at which an increase in urine thiamin excretion is observed.

Since vitamin B<sub>1</sub> plays an important role in energy metabolism, these values were determined as amounts per energy intake. From the data of a meta-analysis of 18 countries<sup>(4)</sup>, the EAR was set at 0.35 mg thiamin (0.45 mg thiamin hydrochloride)/1,000 kcal/day. This value was used as a reference for those aged 1 to 69 years, and the EAR was set using estimated energy requirement values. The RDA was set assuming a coefficient of variation of 20%. No report has stated that the calculation of the values for elderly individuals should be specially considered; thus, the EAR and RDA were determined using the reference value of adults and reference body weight (BW), assuming a coefficient of variation of 10%.

#### **2–2–2. Additional Amount for Pregnant Women (EAR, RDA)**

The additional amounts were calculated based on the assumption that the requirement of vitamin B<sub>1</sub> increases according to the energy requirement. In other words, the additional EAR and RDA for pregnant women were calculated by multiplying the additional estimated energy requirement values (+50 kcal/day for early-term, +250 kcal/day for mid-term, and +450 kcal/day for late-term pregnancies, at a level 2 physical activity) and the vitamin B<sub>1</sub> EAR reference values (0.45 mg/1,000 kcal), to yield values of 0.023 mg/day for early-term, 0.11 mg/day for mid-term, and 0.20 mg/day for late-term pregnancies. These reference values were calculated solely assuming an increase in energy expenditure, and that energy expenditure differs between individuals. Since metabolism is enhanced during pregnancy, the value for late-term pregnancy (0.2 mg/day) was adopted as the additional amount required for pregnant women, yielding 0.2 mg/day (rounding 0.24 mg/day), determined as the EAR × 1.2.

### **2–2–3. Additional Amount for Lactating Women (EAR, RDA)**

The additional amount was calculated based on the assumption that the excreted amount in breast milk is supplemented, using a relative availability of 0.6<sup>(1,2)</sup>, as follows: **0.13 mg/L × 0.78 L/day / 0.6 = 0.169 mg/day.**

The EAR was set at 0.2 mg/day by rounding this value.

The additional RDA was determined as the EAR × 1.2, yielding 0.2 mg/day (rounding 0.24 mg/day).

### **2–3. Method Used to Set Adequate Intake (AI)**

#### **2–3–1. Infants (AI)**

The average concentration of vitamin B<sub>1</sub> in breast milk is 0.13 mg/L<sup>(5–7)</sup>, and the average milk intake is 0.78 L/day<sup>(8,9)</sup>, representing a daily vitamin B<sub>1</sub> intake of about 0.1 mg/day. This value was set as the AI for infants aged 0 to 5 months.

The AI for infants aged 6 to 11 months was calculated using the average of the values from the following 2 expressions:

**Expression 1: the AI for infants aged 0 to 5 months × (reference BW for infants aged 6 to 11 months/reference BW for infants aged 0 to 5 months) 0.75**

**Expression 2: the EAR for adults aged 18 to 29 years × (reference BW for infants aged 6 to 11 months/reference BW for adults aged 18 to 29 years) 0.75 × (1+ growth factor)**

Thus, the AI was determined as 0.2 mg/day for infants aged 6 to 11 months.

### **3. To Avoid Excessive Intake**

#### **3–1. Dietary Intake**

No regular food includes more than 1 mg of vitamin B<sub>1</sub>/100 g. In addition, unfavorable outcomes, as a consequence of the excessive intake of regular food, have not been reported.

#### **3–2. Method Used to Set the Tolerable Upper Intake level (UL)**

A chronic high dose intake of thiamin (50 mg/kg BW/day) has been reported to cause severe toxicity symptoms<sup>(10)</sup>. For example, an intake of 10 g of thiamin hydrochloride every day for 2.5 weeks resulted in headaches, irritability, insomnia, pulsus celer, weakness, contact dermatitis, and itchiness. These symptoms disappeared in 2 days, when the intake was discontinued<sup>(11)</sup>. Nevertheless, there is insufficient evidence for the determination of the UL.

## Vitamin B<sub>2</sub>

### 1. Background Information

#### 1—1. Definition and Classification

The chemical name of vitamin B<sub>2</sub> is riboflavin, and the present DRIs were determined as the amount of riboflavin: 7,8-dimethyl-10-[(2R,3R,4S)-2,3,4,5-tetrahydroxyethyl]benzo[g]pteridine-2,4(3H,10H)-dione. The activities of flavin mononucleotide (FMN) and flavin adenine dinucleotide (FAD) are equimolar to that of vitamin B<sub>2</sub>.

#### 1—2. Function

Vitamin B<sub>2</sub> functions as coenzymes FMN or FAD, and is involved in several metabolic pathways as well as energy production. Vitamin B<sub>2</sub> deficiency causes growth suppression, as the vitamin works in energy metabolism. Its deficiency also causes canker sores, angular cheilitis, tongue inflammation, and seborrheic dermatitis.

#### 1—3. Digestion, Absorption and Metabolism

Riboflavin predominantly exists as FAD or FMN in combination with enzyme proteins. Riboflavin absorption occurs by an active transportation system in the small-intestinal epithelial cells. These processes can be affected by the type of food and other dietary sources. The relative bioavailability of free vitamin B<sub>2</sub> has been reported to be around 64% in the typical Japanese diet<sup>(1)</sup>.

## 2. To Avoid Inadequacy

### 2—1. Factors to be Considered in Estimating Requirements

There should be a difference in the requirement estimation between the calculation using the dietary intake required for the recovery from deficiency, and that from the association between dietary intake and urinary excretion of vitamin B<sub>2</sub>.

#### 2—1—1. Estimation of the Dietary Intake Required for Recovery from Deficiency

An experimental study examined vitamin B<sub>2</sub> deficiency in 4 Japanese individuals (2 men and 2 women)<sup>(12,13)</sup>. Five to 6 weeks after a vitamin B<sub>2</sub>-deficient diet was initiated, the participants complained of sore throat, tongue pain, and pain at the edge of the lips, bleeding from the gums and the oral mucosa, aversion to light, or eye strain<sup>(13)</sup>. The recovery experiment led to acute remission after 0.5 mg/day of vitamin B<sub>2</sub> was administered for 10 days<sup>(12)</sup>. One female participant consuming 1 mg/day of vitamin B<sub>2</sub> reported no complaint. From these results, the vitamin B<sub>2</sub> requirement for the prevention of deficiency can be estimated at about 0.5 mg/day. However, considering the relative bioavailability (64%)<sup>(1)</sup>, this value was set as 0.78 mg/day of dietary vitamin B<sub>2</sub>.

### **2—1—2. Estimation of Requirements from Urinary Riboflavin Excretion**

Usually, only a small amount of riboflavin is excreted in the urine; the level of excretion varies according to the intake of vitamin B<sub>2</sub>. If the body requirement is met, urinary excretion shows a rapid increase. A gradual increase in the intake of free riboflavin to 1.1 mg/day was shown to result in a rapid rise in urinary excretion. This value can be considered as the body requirement, as the urinary excretion of thiamin increases sharply when the body requirement is met.

### **2—2. Method Used to Set the EAR and RDA**

These values were determined as amounts per energy intake.

#### **2—2—1. Adults and Children (EAR and RDA)**

In the determination of the DRIs for vitamin B<sub>2</sub>, the method used was the same as that for vitamin B<sub>1</sub>; the values were obtained from inflection point of the relation between vitamin B<sub>2</sub> intake and excretion. Thus, the EAR of vitamin B<sub>2</sub> was determined as the point at which an increase in urine thiamin excretion was observed. A gradual increase in the intake of free riboflavin to 1.1 mg/day was shown to result in a rapid rise in urinary excretion, in healthy men and women when they received 2,200 kcal/day<sup>(13,14)</sup>. Based on these results, and the involvement of vitamin B<sub>2</sub> in energy metabolism, the EAR was determined as the energy intake/day, i.e., 0.50 mg/1,000 kcal/day for those aged 1-69 years, and the EAR was set using estimated energy requirement values. The RDA was set using a coefficient of variation of 10%.

In terms of elderly individuals, one report stated that the requirement does not differ from that of young adults<sup>(15)</sup>, and another stated that no special consideration is required. Therefore, the EAR and the RDA were determined using the reference value of adults and reference BW, using a coefficient of variation of 10%.

#### **2—2—2. Additional Amount for Pregnant Women (EAR, RDA)**

The additional amounts were calculated based on the assumption that the requirement for vitamin B<sub>2</sub> increases according to the estimated energy requirement. In other words, the additional EAR and RDA for pregnant women were calculated by multiplying the additional values of the estimated energy requirement (+50 kcal/day for early-term, +250 kcal/day for mid-term, and +450 kcal/day for late-term pregnancies at level 2 of physical activity), and the vitamin B<sub>2</sub> EAR reference values (0.50 mg/1,000 kcal), yielding 0.03 mg/day for first-term, 0.13 mg/day for mid-term, and 0.23 mg/day for late-term pregnancies. These reference values were calculated solely assuming an increase in energy expenditure, and that energy expenditure differs between individuals. Since metabolism is enhanced during pregnancy, the value for late-term pregnancy (0.23 mg/day) was adopted as the additional amount required for pregnant women, yielding 0.3 mg/day (rounding 0.27 mg/day) as the additional RDA, determined as the EAR × 1.2.

### **2—2—3. Additional Amount for Lactating Women (EAR, RDA)**

The additional amount was calculated based on the assumption that the excreted amount in breast milk is supplemented, using the relative bioavailability (0.6)<sup>(1,2)</sup>. The mean concentration of riboflavin in breast milk is 0.40 mg/L, and the average milk volume is 0.78 L/day<sup>(5,7-9)</sup>. Thus, the additional EAR was 0.5 (rounding 0.52) mg/day. The additional RDA was determined as the EAR × 1.2, yielding 0.6 (rounding 0.62) mg/day.

### **2—3. Method Used to Set AI**

#### **2—3—1. Infants (AI)**

The daily vitamin B<sub>2</sub> intake of infants is approximately 0.3 (rounding 0.31) mg/day. This value was set as the AI for infants aged 0 to 5 months.

The AI for infants aged 6 to 11 months was calculated using the average of the values from the following 2 expressions:

**Expression 1: the AI for infants aged 0 to 5 months × (reference BW for infants aged 6 to 11 months/reference BW for infants aged 0 to 5 months) 0.75**

**Expression 2: the EAR for adults aged 18 to 29 years × (reference BW for infants aged 6 to 11 months/reference BW for adults aged 18 to 29 years) 0.75 × (1+ growth factor)**

Thus, the AI was determined to be 0.4 mg/day for infants aged 6 to 11 months.

### **3. To Avoid Excessive Intake**

#### **3—1. Dietary Intake**

No regular food contains more than 1 mg of vitamin B<sub>2</sub>/100 g. Additionally, no studies have reported the presence of unfavorable outcomes due to an excessive intake of regular foods.

#### **3—2. Method Used to Set the UL**

A chronic high intake of riboflavin has not been reported to cause severe toxicity. For example, a daily intake of 400 mg of riboflavin for 3 months<sup>(16)</sup>, or a single intravenous injection of 11.6 mg of riboflavin<sup>(17)</sup> caused no deleterious effects. This may be attributed to the rapid excretion of riboflavin in the urine, and also to limited solubility and reduced absorption at higher doses. Thus, there is no evidence for the determination of the UL. Nevertheless, it has been reported that the maximum absorbable amount of riboflavin in a single dose is 27 mg<sup>(17)</sup>; therefore, a single intake of excess vitamin B<sub>2</sub> is rarely effective.

## Niacin

### 1. Background Information

#### 1—1. Definition and Classification

Niacin activity is predominantly exhibited by nicotinic acid, nicotinamide, and tryptophan. The DRIs for niacin are expressed in niacin equivalents (NEs). The Standard Tables of Food Composition in Japan 2010<sup>(18)</sup> lists niacin as the sum of nicotinic acid and nicotinamide, and does not include nicotinamide biosynthesized from tryptophan. Therefore, to calculate the NE in a diet, the amount of nicotinamide biosynthesized from dietary tryptophan should be added to the amount of niacin. The tryptophan to nicotinamide conversion ratio is set at 1/60 on a weight basis. The NE is calculated using the following formula:

**Niacin equivalent (mg NE) = niacin intake (mg) + (1/60) tryptophan intake (mg)**

Most protein contains approximately 1% of tryptophan; therefore, the amount of nicotinamide biosynthesized from tryptophan (mg) is estimated as the amount of protein (g) divided by 6.

#### 1—2. Function

Nicotinic acid and nicotinamide act as coenzymes for enzymes, such as alcohol dehydrogenase, glucose-6-phosphate dehydrogenase, pyruvate dehydrogenase, and 2-oxoglutarate dehydrogenase, in oxidation-reduction reactions, after the conversion of pyridine nucleotide. Niacin is involved in many biological reactions including ATP production, antioxidation via vitamin C or E, as well as fatty acid and steroid synthesis. Nicotinamide adenine dinucleotide (NAD<sup>+</sup>) is used as a substrate of ADP-ribosylation, and is involved in the repair and synthesis of DNA, as well as cell differentiation. Niacin deficiency causes pellagra, in which dermatitis, diarrhea, and neuropsychiatric abnormality are prominent symptoms.

#### 1—3. Digestion, Absorption, and Metabolism

In living cells, niacin exists mainly as the cofactor nicotinamide adenine dinucleotide phosphate (NAD(P)), which binds weakly to enzyme proteins. During the cooking and processing of animal and plant foods, NAD(P) is hydrolyzed to nicotinamide and nicotinic acid, respectively. Any remaining NAD(P) is hydrolyzed to nicotinamide in the gastrointestinal tract. Nicotinamide and nicotinic acid are absorbed in the small intestine. Nicotinic acid predominantly binds to complex carbohydrates in cereal grains, and, therefore, has a lower digestibility<sup>(19)</sup>. The relative availability of dietary niacin to free nicotinamide is approximately 60% in a typical Japanese diet<sup>(1,2)</sup>.

## **2. To Avoid Inadequacy**

### **2—1. Factors to be Considered in Estimating Requirements**

The conversion ratio of tryptophan to nicotinamide is set at 1/60, on a weight basis. In other words, 60 mg of tryptophan is equimolar to 1 mg of niacin.

### **2—2. Method Used to Set the EAR and RDA**

Niacin relates to energy metabolism, and, therefore, the EAR for niacin is expressed as mg NE/1,000 kcal.

#### **2—2—1. Adults and Children (EAR and RDA)**

The requirement was determined from the minimal amount required for the prevention of pellagra. The conversion ratio of tryptophan to nicotinamide is set at 1/60 on a weight basis, according to human studies<sup>(20,21)</sup>. Niacin relates to energy metabolism, and, therefore, the EAR for niacin is expressed as amount per energy intake. Another human study showed that a urinary N<sup>1</sup>-methylnicotinamide level of 1.0 mg/day reflects pellagra-like clinical niacin deficiency<sup>(22)</sup>. An analysis of previous studies showed that the niacin intake equivalent to a urinary N<sup>1</sup>-methylnicotinamide level of 1.0 mg/day is 4.8 mg NE/1,000 kcal<sup>(20-24)</sup>. This value was used as the reference for the setting of the EAR for individuals aged 1 to 69 years, and the EAR was determined using estimated energy requirement values. The RDA was determined as the EAR × 1.2. Based on niacin intake and urinary nicotinamide metabolite data, the niacin activity in older individuals is considered to be the same as that in younger individuals<sup>(25,26)</sup>. Thus, the EAR and RDA were set using the same calculation method as that used in adults.

#### **2—2—2. Additional Amount for Pregnant Women (EAR, RDA)**

There is no evidence for the setting of the EAR using a factorial method, and the additional amounts could be set based on the assumption that the requirement for niacin increases according to the estimated energy requirement; however, the amount of nicotinamide biosynthesized from tryptophan increases during pregnancy, and this compensates for the increase in the niacin requirement<sup>(27)</sup>. Thus, pregnant women do not require additional niacin intake.

#### **2—2—3. Additional Amount for Lactating Women**

The conversion rate of tryptophan to nicotinamide returns to a normal level after delivery<sup>(27)</sup>, and, therefore, lactating women require additional niacin intake to compensate for the loss of niacin through breast milk. Using 2.0 mg/L as the concentration of breast milk, 0.78 L/day as the average milk volume, and 60% as the relative availability<sup>(1,2)</sup>, the additional EAR for lactating women was set at 3 mg NE/day (rounding 2.6 mg NE/day). The additional RDA was set at 3 (rounding 3.0) mg NE/day, determined as the EAR × 1.2.

## **2—3. Method Used to Set the AI**

### **2—3—1. Infants (AI)**

The concentration of niacin in the breast milk of Japanese mothers is 2.0 mg/L<sup>(5-7)</sup>. Considering an average milk intake of 0.78 L/day<sup>(6,7)</sup>, the daily nicotinamide intake is 1.56 mg/day. The AI for infants aged 0 to 5 months was set at 2 mg/day. Nicotinamide is unlikely to be biosynthesized from tryptophan at this stage, and, therefore, the AI is expressed in mg/day<sup>(28)</sup>.

The AI for infants aged 6 to 11 months was calculated using the average of the values from the following 2 expressions:

**Expression 1: the AI for infant boys or girls aged 6 to 11 months (extrapolated from the AI of infants) = AI for infants aged 0 to 5 months × (reference BW for infants aged 6 to 11 months/reference BW for infants aged 0 to 5 months) 0.75**

**Expression 2: the EAR for adults aged 18 to 29 years × (reference BW for infants aged 6 to 11 months/reference BW for adults aged 18 to 29 years) 0.75 × (1 + growth factor)**

The means of these extrapolated values were determined for each sex. The average of the obtained values for each sex is 3.1 mg NE/day. Thus, the AI for infants aged 6 to 11 months was set as 3 mg NE/day.

## **3. To Avoid Excessive Intake**

### **3—1. Dietary Intake**

A high amount of nicotinamide is present in animal food, at a maximum of approximately 10 mg/100 g. Nicotinic acid exists in plant food at less than 10 mg/100 g. No studies have reported the presence of unfavorable outcomes due to an excessive intake of regular food.

### **3—2. Method Used to Set the UL**

Nicotinic acid and nicotinamide are often used in niacin supplements and fortified foods. The UL for niacin, therefore, takes into account the nicotinic acid and nicotinamide obtained from supplements and fortified foods. The large doses of nicotinamide and nicotinic acid used to treat patients with type 1 diabetes and dyslipidemia, respectively, may cause gastrointestinal effects such as dyspepsia, diarrhea, and constipation, and also hepatotoxic symptoms such as dysfunction and fulminant hepatitis. According to previous reports<sup>(29-32)</sup> the no observed adverse effect levels (NOAELs) for nicotinamide and nicotinic acid were set at 25 mg/kg BW and 6.25 mg/kg BW, respectively. The NOAELs were divided by an uncertainty factor of 5, and the obtained values--5 mg/kg BW and 1.25 mg/kg BW--were set as the ULs for nicotinamide and nicotinic acid, respectively. The ULs were determined using these values, according to age and sex group. A pharmacological dose of nicotinic acid has the transient vasodilatory effect of flushing (reddening of the skin), but does not cause adverse health effects. Thus, it is not appropriate to use flushing as symptom for the setting of the UL for nicotinic acid.

## Vitamin B<sub>6</sub>

### 1. Background Information

#### 1—1. Definition and Classification

The chemical substances possessing vitamin B<sub>6</sub> activity are pyridoxine (PN), pyridoxal (PL), and pyridoxamine (PM), and their respective phosphorylated forms. The phosphorylated forms--pyridoxine-5-phosphate (PNP), pyridoxal-5-phosphate (PLP) and pyridoxamine 5-phosphate (PMP)--have an activity that is equimolar to that of vitamin B<sub>6</sub>. The current DRIs were determined as the amount of pyridoxine.

#### 1—2. Function

Vitamin B<sub>6</sub> functions as the coenzyme PLP, and is involved in transamination reaction, decarboxylation, and racemization reaction. Vitamin B<sub>6</sub> is important for the maintenance of immune systems, as vitamin B<sub>6</sub> deficiency decreases the rate of the conversion of linoleic acid to arachidonic acid. Deficiency causes pellagra-like symptoms, seborrheic dermatitis, tongue inflammation, angular cheilitis, or hypolymphemia.

#### 1—3. Digestion, Absorption and Metabolism

Vitamin B<sub>6</sub> predominantly exists as PLP or PMP, in combination with enzyme proteins. Once PLP and PMP dissociate, they are absorbed as PL or PM. Living plant cells contain pyridoxine-5' $\beta$ -glucoside (PNG). PNG is absorbed as PN in humans, and the relative bioavailability of PNG has been estimated to be 50%<sup>(33)</sup>. The digestion processes associated with this vitamin are affected by the type of food and other dietary sources. The relative bioavailability of free vitamin B<sub>6</sub> has been reported to be 75% in the US<sup>(34)</sup>, and 73% in the typical Japanese diet<sup>(1)</sup>.

### 2. To Avoid Inadequacy

#### 2—1. Method Used to Set the EAR and RDA

These values were determined as amounts per protein intake.

##### 2—1—1. Adults and Children (EAR and RDA)

Vitamin B<sub>6</sub> is involved in the catabolism of amino acids and formation of bioactive amines, including some neurotransmitters. The plasma PLP concentration has been reported to reflect the body store of vitamin B<sub>6</sub><sup>(35)</sup>. A low plasma PLP concentration is associated with electroencephalographic changes in young, non-pregnant women<sup>(36)</sup>. Furthermore, a plasma PLP concentration of 30 nmol/L was required to alleviate vitamin B<sub>6</sub> deficiency-induced disorders<sup>(37)</sup>. The EAR for vitamin B<sub>6</sub> was based on the amount of vitamin B<sub>6</sub> required for a maintenance of a plasma PLP level of 30 nmol/L.

The vitamin B<sub>6</sub> requirement increases as the protein intake increases, and the plasma

PLP concentration correlates well with vitamin B<sub>6</sub> intake per protein intake<sup>(38)</sup>. Thus, 0.014 mg pyridoxine/g protein was estimated as the concentration required to maintain a plasma PLP concentration of 30 nmol/L. The EAR reference value was determined (0.014/0.73) using a relative bioavailability of 73%<sup>(1)</sup>. The EARs were calculated by multiplying this value by the RDAs of protein. The RDA was calculated as the EAR × 1.2. To obtain the daily requirement of vitamin B<sub>6</sub>, the EAR of vitamin B<sub>6</sub> was multiplied by the RDA of protein.

In terms of elderly individuals, a previous report stated that plasma PLP levels decrease with increasing age<sup>(39)</sup>; however, due to a lack of data, the EAR and the RDA were determined using the same method as that used in adults.

### **2—1—2. Additional Amount for Pregnant Women (EAR, RDA)**

The plasma PLP concentration reportedly decreases during pregnancy<sup>(40–52)</sup>.

The additional amount required depends on whether plasma PLP should be maintained at levels that are similar to those in non-pregnant women or those in the first-stage of pregnancy, or taking into consideration if the aforementioned decrease in the PLP concentration occurs as a common physiological response to pregnancy.

The previous DRIs adopted the former method, and set the additional amounts of EAR and RDA for pregnant women using the method used in the US-Canada DRIs<sup>(38)</sup>, and the relative bioavailability in Japan<sup>(1)</sup>. However, while no study has reported on vitamin B<sub>6</sub> deficiency in this context, the vitamin B<sub>6</sub> intake of pregnant Japanese pregnant women does not exceed the previous EAR of 1.7 mg/day. Therefore, the additional amount of vitamin B<sub>6</sub> required was reconsidered.

Although vitamin B<sub>6</sub> involves the production of tryptophan metabolites, the proportion of some of these metabolites increases, rather than decreases, under conditions of vitamin B<sub>6</sub> insufficiency. Moreover, their effects on pregnant women and fetuses remain unclear.

The decrease in the plasma PLP level during the late term of pregnancy is considered to be caused by the increased requirement of the fetus<sup>(49)</sup>. This decrease is considered a result of increased placental PL transportation due to the elevated PLP→PL reaction rate that is caused by elevated serum alkaline phosphatase levels in mothers, so as to provide vitamin B<sub>6</sub> to the fetus<sup>(42,47,51,52)</sup>.

During the late stage of pregnancy, to maintain 30 nmol/L of plasma PLP (at a level which is equal to that of women who are not pregnant), an additional 4 to 10 mg/day of pyridoxine is needed<sup>(40)(47)(52)(44)</sup>. However, these amounts are quite different from the potential intakes for the Japanese population, based on the current intake. Lui et al. recommended maintaining a plasma PLP level of 20 nmol/L to prevent vitamin B<sub>6</sub> deficiency<sup>(35)</sup>. Abnormal electroencephalograms have been observed at plasma PLP levels lower than 10 nmol/L in non-pregnant women<sup>(36)</sup>. Another study examined pregnant Japanese women, and reported their mid-term and late-term pregnancy plasma PLP levels (mean±standard deviation) to be 23.3 ± 16.7 nmol/L and 18.3 ± 12.5 nmol/L, respectively<sup>(53)</sup>. Thus, the additional amount of vitamin

B<sub>6</sub> required would be low, considering the enhanced vitamin B<sub>6</sub> metabolism during pregnancy.

However, the requirement for protein increases according to the body protein storage required during pregnancy, enhancing amino acid metabolism.

From these findings, the additional EAR was determined considering the body protein storage for the placenta and fetus. In other words, the value was calculated based on the EAR reference of pyridoxine for non-pregnant women (0.014 mg/protein g) and body protein storage during pregnancy, using the relative bioavailability. During pregnancy, the efficiency of various nutrients increases; however, due to a lack of data, the relative bioavailability was set as 73%<sup>(1)</sup>. The additional EAR was calculated as follows:

**Early-term pregnancy**

$$(0.014 \text{ mg/g protein} \times 0 \text{ g/day} = 0 \text{ mg/day}) / 0.73 = 0 \text{ mg/day}$$

**Mid-term pregnancy**

$$(0.014 \text{ mg/g protein} \times 1.94 \text{ g/day} = 0.027 \text{ mg/day}) / 0.73 = 0.037 \text{ mg/day}$$

**Late-term pregnancy**

$$(0.014 \text{ mg/g protein} \times 8.16 \text{ g/day} = 0.114 \text{ mg/day}) / 0.73 = 0.156 \text{ mg/day}$$

The RDAs were determined as these values  $\times$  1.2, yielding 0 mg, 0.044 mg and 0.187 mg for early-term, mid-term, and late-term pregnancies, respectively.

These values were calculated solely assuming an increase in the amount of protein required, and that requirement differs between individuals. Since metabolism is enhanced during pregnancy, the value for late-term pregnancy (0.156 mg/day) was adopted as the additional amount required by pregnant women, yielding 0.2 mg/day (rounding 0.156 mg/day). The additional RDA was calculated as the additional EAR  $\times$  1.2, yielding 0.2 mg/day (rounding 0.187 mg/day).

**2—1—3. Additional Amount for Lactating Women**

The additional EAR for pregnant women was calculated based on the mean concentration of vitamin B<sub>6</sub> in breast milk (0.25 mg/L)<sup>(54,55)</sup>, the average milk volume (0.78 L/day)<sup>(8,9)</sup>, and the relative bioavailability (73%)<sup>(1)</sup>, i.e., 0.3 mg/day (rounding 0.267 mg/day). The additional RDA was calculated as the additional EAR  $\times$  1.2.

**2—2. Method Used to Set AI**

**2—2—1. Infants (AI)**

For infants aged 0 to 5 months, the vitamin B<sub>6</sub> intake is approximately 0.2 mg/day (rounding 0.195 mg/day) based on the mean concentration of vitamin B<sub>6</sub> in breast milk (0.25 mg/L)<sup>(54,55)</sup> and the average milk intake (0.78 L/day)<sup>(8,9)</sup>. This value was set as the AI.

The AI for infants aged 6 to 11 months was calculated using the average of the values from the following 2 expressions:

**Expression 1: the AI for infants aged 0 to 5 months  $\times$  (reference BW for infants aged 6 to 11 months/reference BW for infants aged 0 to 5 months) 0.75**

**Expression 2: the EAR for adults aged 18 to 29 years × (reference BW for infants aged 6 to 11 months/reference BW for adults aged 18 to 29 years) 0.75 × (1 + growth factor)**

Thus, the AI was determined to be 0.3 mg/day for infants aged 6 to 11 months.

### **3. To Avoid Excessive Intake**

#### **3—1. Dietary Intake**

No regular food contains more than 1 mg of vitamin B<sub>6</sub>/100 g. No reports have suggested the development of unfavorable outcomes due to the excessive intake of regular food.

#### **3—2. Method Used to Set the UL**

A high intake of pyridoxine over a course of several months was shown to result in sensory neuropathy<sup>(56)</sup>. This symptom was used as a criterion for the estimation of the UL for pyridoxine. In contrast, the administration of 100-300 mg pyridoxine/day over a period of 4 months did not cause sensory neuropathy in 24 patients with carpal tunnel syndrome<sup>(57)</sup>. Based on these data, the NOAEL was set at 300 mg/day. Assuming an uncertainty factor of 5, the UL for pyridoxine was set at 60 mg/day-0.86 mg/kg BW. The UL for each age group was obtained by multiplying the UL by the reference BW.

## **Vitamin B<sub>12</sub>**

### **1. Background Information**

#### **1—1. Definition and Classification**

Vitamin B<sub>12</sub> is a cobamide, and there are various B<sub>12</sub> compounds with different upper ligands, such as methylcobalamin, sulfitecobalamin, and cyanocobalamin. The DRIs for vitamin B<sub>12</sub> were set as the amount of cyanocobalamin.

#### **1—2. Function**

Vitamin B<sub>12</sub> is a cofactor for methionine synthetase and L-methylmalonyl-coenzyme A mutase. Vitamin B<sub>12</sub> deficiency causes megaloblastic anemia, white-matter deficit in the spinal cord and brain, and peripheral neuropathy.

#### **1—3. Digestion, Absorption and Metabolism**

Food-bound vitamin B<sub>12</sub> is dissociated from proteins in the presence of acid and pepsin. The released vitamin B<sub>12</sub> then binds to the haptocorrins secreted by the salivary glands. Haptocorrins are partially degraded in the duodenum, releasing vitamin B<sub>12</sub>, which then binds with intrinsic factor. The intrinsic factor-vitamin B<sub>12</sub> complex enters the enterocyte after binding with the receptors in the ileal mucosa. The dietary absorption was reported to be around 50% in healthy participants<sup>(58,59)</sup>.

### **2. To Avoid Inadequacy**

#### **2—1. Factors to be Considered in Estimating Requirements**

The DRIs considered the values required for the treatment of anemia in pernicious anemia patients without intrinsic factors.

#### **2—2. Method Used to Set the EAR and RDA**

##### **2—2—1. Adults and Children (EAR and RDA)**

It is not possible to determine the EAR of vitamin B<sub>12</sub> for healthy adults, because of the intrinsic-factor-mediated B<sub>12</sub> gastrointestinal absorption system and/or the substantial enterohepatic vitamin B<sub>12</sub> circulation. Thus, the EAR for adults was estimated based on clinical data from vitamin B<sub>12</sub>-deficient patients with pernicious anemia, that examined the amount of vitamin B<sub>12</sub> required for the maintenance of an adequate hematological status (mean corpuscular volume < 101 fL) and serum vitamin B<sub>12</sub> level (100 pmol/L or more). Studies reported that an intramuscular injection with varying concentrations (0.1–10 µg/day) of vitamin B<sub>12</sub> showed an increase in the capacity of erythrocyte production at 0.1 µg/day<sup>(44)</sup>, indicating the maximum capacity at 0.5 to 1.0 µg/day<sup>(60)</sup>. Another study reported that an improvement in the mean corpuscular volume was observed at 1.4 µg/day (range 0.5 to 4.0 µg/day) of vitamin B<sub>2</sub> injection in half of the patients with pernicious anemia<sup>(61)</sup>. These data suggest an average

intramuscular requirement of 1.5 mg/day for the maintenance of an adequate hematological status.

Vitamin B<sub>12</sub>-deficient patients with pernicious anemia cannot reabsorb vitamin B<sub>12</sub> (0.5 µg/day) from the bile, due to the lack of an intrinsic-factor-mediated vitamin B<sub>12</sub> absorption system. Thus, under normal physiological conditions, an average intake of 1.0 µg/d is required to compensate for the estimated extra losses of biliary vitamin B<sub>12</sub> (0.5 µg/day) from the average intramuscular requirement (1.5 µg/day). Adjusting for this value with a 50% absorption rate of dietary vitamin B<sub>12</sub>, the EAR was set at 2.0 µg/day for adults. The RDA was calculated as 2.4 mg/day, by multiplying the EAR and 1.2.

Although serum vitamin B<sub>12</sub> levels are known to be higher in women than men<sup>(62–64)</sup>, data on this are insufficient. Therefore, the same values were adopted for both sexes.

The EAR for children was calculated from the EAR for adults aged 18 to 29 years, using the following equation for body surface area, at each age:

**(Reference BW at each age/reference BW of adults aged 18 to 29 years) 0.75 × (1 + growth factor).**

The EARs and DRIs for those aged over 50 years were set at values that were identical to those set for adults aged 18 to 49 years, due to a lack of detailed information on the decrease in vitamin B<sub>12</sub> absorption in elderly individuals<sup>(65,66)</sup>.

## **2—2—2. Additional Amount for Pregnant Women (EAR, RDA)**

The human fetus is estimated to accumulate 0.1 to 0.2 µg/day of vitamin B<sub>12</sub><sup>(67,68)</sup>. Using the median (0.15 µg/day) of the fetal deposition, and the 50% absorption rate of dietary vitamin B<sub>12</sub> in healthy adults, the additional EAR for pregnant women was set at 0.3 µg/day. The additional RDA was estimated as 0.4 µg/day (rounding 0.36 µg/d) by multiplying the additional EAR and 1.2.

## **2—2—3. Additional Amount for Lactating Women**

Using the average vitamin B<sub>12</sub> concentration and secretion of breast milk, and the 50% absorption rate of dietary vitamin B<sub>12</sub> in healthy adults (0.45 µg/L × 0.78 L/day/0.5), the additional EAR for lactating women was set at 0.7 µg/day (rounding 0.702 mg/day). The additional RDA was calculated as 0.8 µg/day (rounding 0.84 µg/d) by multiplying the additional EAR and 1.2.

## **2—3. Method Used to Set AI**

### **2—3—1. Infants (AI)**

For infants aged 0 to 5 months, the mean concentration of vitamin B<sub>12</sub> in breast milk is 0.45 µg/L<sup>(6,7,69)</sup>. The average milk volume is 0.78 L/d<sup>(8,9)</sup>, representing a daily vitamin B<sub>12</sub> intake of about 0.4 µg/day (rounding 0.35 µg/day). This value was set as the AI.

The AI for infants aged 6 to 11 months was calculated using the average of the values

from the following 2 expressions:

**Expression 1: the AI for infants aged 0 to 5 months × (reference BW for infants aged 6 to 11 months/reference BW for infants aged 0 to 5 months) 0.75**

**Expression 2: the EAR for adults aged 18 to 29 years × (reference BW for infants aged 6 to 11 months/reference BW for adults aged 18 to 29 years) 0.75 × (1 + growth factor)**

Thus, the AI was determined to be 0.5 µg/day for infants aged 6 to 11 months.

### **3. To Avoid Excessive Intake**

#### **3—1. Dietary Intake**

Vitamin B<sub>12</sub> absorption is regulated by the intrinsic factor secreted by the stomach in the intestinal absorption system. No reports till date have suggested the presence of unfavorable outcomes due to the excessive intake of regular foods.

#### **3—2. Method Used to Set the UL**

Vitamin B<sub>12</sub> cannot be absorbed when its intake is excessive, and the intrinsic-factor-regulating absorption system is saturated<sup>(61,70)</sup>. The oral administration of substantial amounts (500 µg) of vitamin B<sub>12</sub> was shown to result in only about 1% absorption in the intestine<sup>(61)</sup>. No harmful effect was observed even when a mega dose (2.5 mg) of vitamin B<sub>12</sub> was administered parenterally<sup>(71)</sup>. Thus, the UL was not determined for vitamin B<sub>12</sub>.

## **Folate**

### **1. Background Information**

#### **1—1. Definition and Classification**

The basic skeleton of folate is pteroylmonoglutamate, which comprises p-aminobenzoic acid with pterin rings and glutamate. Folate naturally occurs in combination with 1 or more molecules of glutamate ( $\gamma$ -binding).

In its narrowest sense, folate is referred to as pteroylmonoglutamate. In a broader sense, it includes coenzyme species in their reduced form, as well as single-carbon compounds and their polyglutamate forms. The present DRIs used the broader definition, as equivalents of pteroylmonoglutamate, in accordance with The Standard Tables of Food Composition 2010.

#### **1—2. Function**

Folate functions as a coenzyme in single-carbon transfers, in the metabolism of nucleic and amino acids. Folate deficiency causes megaloblastic anemia. Folate deficiency in mothers can lead to fetal neural tube defects (NTDs) and anencephalia.

#### **1—3. Digestion, Absorption and Metabolism**

Most naturally occurring folates (food folates) are pteroylpolyglutamates, the activities of which are more easily lost during cooking than those of pteroylmonoglutamates--the form used in vitamin supplements. Pteroylpolyglutamates are hydrolyzed to monoglutamate forms in the gut before absorption across the intestinal mucosa. The digestion processes can be affected by the type of food and other dietary sources. The relative bioavailability of food folate is reported to be 25-81% that of pteroylmonoglutamate<sup>(72-74)</sup>. The relative bioavailability of free-pteroylmonoglutamate is reported to be 50% in the typical Japanese diet<sup>(2)</sup>.

### **2. To Avoid Inadequacy**

#### **2—1. Factors to be Considered in Estimating Requirements**

The relative bioavailability of dietary folate depends on its food sources, and is influenced by other dietary intakes. Naturally occurring folates include various reduced forms, which are a combination of polyglutamate chains and 1 carbon fragment. Polyglutamate is hydrolyzed by conjugase in the jejunum, and is converted to monoglutamate. This is then actively absorbed by specific transporters, and is present in the mucosal cell in its monoglutamate form. Conjugase is an enzyme that comprises zinc as a prosthetic group. It is well-known that orange juice and banana contain the conjugase activity inhibitor<sup>(75)</sup>.

## **2—2. Method Used to Set the EAR and RDA**

### **2—2—1. Adults and Children (EAR and RDA)**

The concentrations of red blood cell folate (300 nmol/L) and plasma total homocysteine (14 µmol/L) were applied as biomarkers to reflect the middle- to long-term folate nutritional status<sup>(76–79)</sup>. The EAR for adults aged 18 to 29 years old was estimated as 200 µg/day. The RDA was calculated as 240 µg/day, by multiplying the EAR and 1.2. The lower value was adopted if the values for the men and women, in each group, were different.

The EAR for children was calculated from the EAR for adults (200 µg/day), using the following equation for body surface area, in each age group:

(Reference BW at each age/reference BW of those aged 18 to 29 years)  $0.75 \times (1 + \text{growth factor})$ .

For adults aged over 50 years old, folate bioavailability was estimated to be equivalent to that of younger adults<sup>(79)</sup>; therefore, the same value as that of adults aged 18 to 29 years old was adopted.

### **2—2—2. Additional Amount for Pregnant Women (EAR, RDA)**

Women with macrocytic anemia during pregnancy recover naturally after delivery<sup>(78)</sup>, indicating a considerable increase in the demand for folate during pregnancy. The addition of 100 µg/day of pteroylmonoglutamate to a diet adequate in food folate has been reported to result in adequate levels of red cell folate<sup>(80,81)</sup>. Thus, this value was set as the additional EAR (200 µg/day; 100/bioavailability rate 0.5<sup>(2,72)</sup>). The additional RDA was calculated by multiplying the additional EAR and 1.2, yielding 240 µg/day.

### **2—2—3. Additional Amount for Lactating Women**

The additional EAR for pregnant women was calculated based on the mean concentration of folate in breast milk (54 µg/L<sup>(5–7)</sup>), the average milk volume (0.78 L/day)<sup>(8,9)</sup>, and the relative bioavailability (50%)<sup>(2,72)</sup>, yielding 80 µg/day (rounding 84 µg/day). The additional RDA was calculated by multiplying the additional EAR and 1.2, yielding 100 µg/day.

## **2—3. Method Used to Set AI**

### **2—3—1. Infants (AI)**

For infants aged 0 to 5 months, the mean concentration of vitamin folate in breast milk is 54 µg/L<sup>(5–7)</sup>. The average milk intake is 0.78 L/day<sup>(8,9)</sup>, representing a daily folate intake of 40 µg/day (rounding 42 µg/day). This value was set as the AI.

The AI for infants aged 6 to 11 months was calculated using the average of the values from the following 2 expressions:

**Expression 1: AI for infants aged 6 to 11 months (extrapolated from the AI for infants) = AI for infants aged 0 to 5 months × (reference BW for infants aged 6 to 11 months/reference BW for infants aged 0 to 5 months) 0.75**

**Expression 2: the EAR for adults aged 18 to 29 years × (reference BW for infants aged 6 to 11 months/reference BW for adults aged 18 to 29 years) 0.75 × (1 + growth factor)**

Thus, the AI was determined to be 60 µg/day for infants aged 6 to 11 months.

### **3. To Avoid Excessive Intake**

#### **3—1. Dietary Intake**

No regular food contains more than 300 µg of folate/100 g except for liver. No study till date has reported the presence of unfavorable outcomes due to an excessive intake of regular food.

#### **3—2. Method Used to Set the Tolerable Upper Intake level**

In the United States (US), adverse health effects such as the masking of pernicious anemia and neurological damage resulting from elevated serum folate levels, caused by the intake of folic acid-supplemented foods, have been reported<sup>(82)</sup>. This masking of pernicious anemia<sup>(83–86)</sup> is the biggest factor involved in the setting of the UL for folate intake. When individuals with insufficient vitamin B<sub>12</sub> levels consume large amounts of pteroylmonoglutamate, the development of pernicious anemia is masked; in addition, it also leads to the progression of more severe disease as well as posterior spinal degeneration<sup>(83–86)</sup>.

These adverse effects may be induced by the dihydropteroylmonoglutamate derived from pteroylmonoglutamate, which inhibits the activities of thymidylate synthase<sup>(87)</sup>, phosphoribosylaminoimidazolecarboxamide transformylase<sup>(88)</sup>, and 5,10-methylenetetrahydrogenase<sup>(89)</sup>. Thus, consuming excessive amounts of pteroylmonoglutamate may inhibit the single-carbon transfer pathways of folate metabolism.

Pteroylmonoglutamate intake is now common, as folate supplementation is recommended before or in the early term of pregnancy, for the prevention of NTDs. However, while supplementation may prevent NTDs, unfavorable outcomes (neurological damage) have been reported. Therefore, the UL for pteroylmonoglutamate should be determined.

The UL of folate intake was determined according to the US-Canada DRIs<sup>(90)</sup>. Women of reproductive age who were administered 0.36-5 mg/day of pteroylmonoglutamate from the preconception period till the gestational age of 3 months had no serious side effects<sup>(90)</sup>. Based on this finding, the adverse effect level was estimated to be 5 mg/day, equivalent to 88 mg/kg BW/day using the reference BW of women aged 19 to 30 years<sup>(91)</sup>. The UL reference was estimated as 18 µg/kg BW/day, by dividing the value by an uncertainty factor of 5. The UL was determined using the reference value and reference BW in each age group. Related studies on this topic have been limited to those on women; therefore, the UL for men was the same as that for women.

### **3—3. Additional Concerns regarding Women of Reproductive Age**

Fetal NTDs are disorders pertaining to the closure of the neural tube (which occurs approximately 28 days after conception), and are clinically diagnosed as anencephaly, spina bifida, and myelomeningocele. Abundant evidence suggests that the preconceptual intake of pteroylmonoglutamate decreases the risk of fetal NTDs<sup>(92–102)</sup>. Genetic polymorphisms of the enzymes related to folate metabolism (e.g., methylene tetrahydrofolate reductase) may be associated with NTD risk<sup>(92–102)</sup>.

Other congenital disorders that can be avoided through the administration of pteroylmonoglutamate are cleft lip/palate<sup>(103,104)</sup> and congenital heart disease<sup>(104)</sup>. Thus, maintaining an adequate maternal folate status is essential for the prevention of NTDs. To estimate the minimum effective dose for the risk reduction of NTDs, the lowest reported preconception dose (0.36 mg/day; at 0.36 to 5 mg/day for over 3 months<sup>(92–102)</sup>) was applied. This value was rounded to 0.4 mg/day, i.e., a dietary folate equivalent of 800 mg/day.

## **4. For the Prevention of the Development and Progression of LRDs**

### **4—1. The Association with LRDs**

#### **4—1—1. Prevention of Disease Development**

##### **4—1—1—1. The Association between Plasma Homocysteine Levels and Cardiovascular or Cerebrovascular Diseases**

Higher folate intakes have been reported to be associated with a decreased risk of stroke or heart disease<sup>(105,106)</sup>. Several randomized controlled trials have investigated the preventive effect of folic acid, but the results are inconsistent<sup>(107,108)</sup>. Inconsistencies in the intervention and observation, and the results of each of those studies must be further studied. The amount of vitamin consumed exceeded the possible dietary intake in the intervention studies; in addition, other types of vitamin B or various polyphenols may have influenced the results of the observational studies.

##### **4—1—1—2. Association between Folate Intake and Cancer**

Previous epidemiological studies have shown that the intake of pteroylmonoglutamate during pregnancy protects against the development of NTDs; however, the risk of cancer is considered to increase with intake. A meta-analysis of approximately 50,000 individuals showed that the risk neither increased nor decreased with long-term pteroylmonoglutamate supplementation<sup>(109)</sup>.

#### **4—1—2. Prevention of Disease Progression**

No data were available in this regard.

### **4—2. Tentative Dietary Goal for Preventing LRDs**

The DG was not determined due to a lack of data.

## **Pantothenic acid**

### **1. Background Information**

#### **1—1. Definition and Classification**

Pantothenic acid exists mainly as coenzyme A (CoA) derivatives, acetyl CoA, acyl CoA, acyl-carrier protein (ACP) and 4-phosphopantetheine, the activities of which are equimolar to that of pantothenic acid. The present DRIs were determined as the amount of pantothenic acid.

#### **1—2. Function**

Pantothenic acid functions as a component of CoA and phosphopantetheine, which are involved in carbohydrate and fatty acid metabolism. Pantothenic acid is widely distributed in foods, and cases of deficiency are rare.

#### **1—3. Digestion, Absorption and Metabolism**

CoA in the diet is hydrolyzed in the intestinal lumen to dephospho-CoA and pantetheine, and these are hydrolyzed to pantothenic acid in its absorbable form. The digestion of this vitamin is affected by the type of food, and other dietary sources. The relative bioavailability of pantothenic acid is reported as 70% in the typical Japanese diet<sup>(1,2)</sup>.

### **2. To Avoid Inadequacy**

#### **2—1. Factors to be Considered in Estimating Requirements**

Pantothenic acid is involved in fatty acid metabolism.

#### **2—2. Method Used to Set AI**

##### **2—2—1. Adults and Children (AI)**

There is no evidence for the setting of the EAR for pantothenic acid, as there are no reports on this vitamin's deficiency in humans. Thus, we estimated the AIs based on the Japanese intake. According to the National Health and Nutrition Survey (NHNS) 2010 and 2011<sup>(110)</sup>, the median dietary pantothenic acid intake among adults and adolescents is 3-7 mg/day. In another dietary assessment study, the mean pantothenic acid intake was reported to be 4.6 mg/day in young Japanese women<sup>(111)</sup>. A study on Japanese individuals aged 32-76 years reported that the mean intakes were 7 mg/day and 6 mg/day in the men and women, respectively<sup>(112)</sup>. There is no evidence that these intake levels lead to pantothenic acid deficiency. Thus, the AIs were adopted from the median dietary pantothenic acid intake determined in the NHNS 2010 and 2011, corresponding to participants' sex and age. The AIs for elderly individuals were set as the same median value, as there are no data indicating the need for special consideration in terms of pantothenic acid nutrition in this population.

### **2—2—2. Infants (AI)**

For infants aged 0 to 5 months, the mean concentration of pantothenic acid in breast milk is 5.0 mg/L<sup>(5,7)</sup>. The average milk volume is 0.78 L/day<sup>(8,9)</sup>, representing a daily pantothenic acid intake of 4.0 mg/day (rounding 3.9 mg/day). This value was set as the AI.

The AI for infants aged 6 to 11 months was calculated using the average of the values from the following 2 expressions:

**Expression 1: the AI for infants aged 0 to 5 months × (reference BW for infants aged 6 to 11 months/reference BW for infants aged 0 to 5 months) 0.75**

**Expression 2: the AI for adults aged 18 to 29 years × (reference BW for infants aged 6 to 11 months/reference BW for adults aged 18 to 29 years) 0.75 × (1 + growth factor)**

Thus, the AI was determined to be 3 mg/day for infants aged 6 to 11 months.

### **2—2—3. Pregnant Women (AI)**

There is no evidence for the determination of the additional pantothenic acid amount for pregnant women by the factorial method. Moreover, there is no indication that the pantothenic acid requirement rises with increases in the energy requirement during pregnancy. Thus, the pantothenic acid intake for pregnant women was estimated using the median of the dietary pantothenic acid intake determined in the NHNS 2010 and 2011<sup>(113)</sup>. The AI for pregnant women was set at 5 mg/day.

### **2—2—4. Lactating Women (AI)**

For pantothenic acid, the estimated AIs are in excess of the pantothenic acid requirement. Thus, the pantothenic acid intakes for lactating women are estimated using the median dietary pantothenic acid intake determined in the NHNS 2010 and 2011<sup>(113)</sup>. The AI for lactating women was set at 5 mg/day.

## **3. To Avoid Excessive Intake**

### **3—1. Dietary Intake**

No regular food contains more than 5 mg of pantothenic acid/100 g except for liver. No study till date has reported unfavorable outcomes due to the excessive intake of regular food.

### **3—2. Method Used to Set the UL**

A pharmacological dose of pantothenic acid, administered over 3 months, in combination with nicotinamide, ascorbic acid, and pyridoxine, was reported to cause adverse effects such as nausea, poor appetite, and abdominal pain in children<sup>(114)</sup>. However, there are no reports stating that a pharmacological dose of pantothenic acid causes adverse health effects. Thus, the UL for pantothenic acid was not set at present.

II Energy and Nutrients  
Vitamins (2) Water-soluble Vitamins  
Folate

#### **4. For the Prevention of the Development and Progression of LRDs**

##### **4—1. The Association with LRDs**

No data were available in this context. The DG was not determined due to a lack of data.

## **Biotin**

### **1. Background Information**

#### **1—1. Definition and Classification**

Biotin is a compound formally known as 5-[(3aS, 4S, 6aR)-2-oxohexahydro-1H-cheno[3, 4d]imidazole-4-yl] pentanoic acid, and only its d-isomer shows physiological activity. The present DRIs were determined as the amount of biotin.

#### **1—2. Function**

Biotin functions as a coenzyme in bicarbonate-dependent carboxylation reactions. Biotin deficiency can cause immune deficiency disorders such as rheumatism, Sjogren's syndrome and Crohn's disease. Insufficient biotin intake can also cause various symptoms such as dermatitis, atrophic gingivitis, lack of appetite, nausea, and facial pallor.

#### **1—3. Digestion, Absorption and Metabolism**

Biotin predominantly exists as protein-bound forms in food. Released biotin is absorbed mainly from the jejunum. The digestion of this vitamin can be affected by the type of food, and other dietary sources. The relative bioavailability of free biotin has been reported to be 80% in the typical Japanese diet<sup>(2)</sup>.

### **2. To Avoid Inadequacy**

#### **2—1. Method Used to Set AI**

##### **2—1—1. Adults and Children (AI)**

There are currently no data on which the EAR for adults can be based. The average daily biotin intake among Americans is 35.5 µg/day<sup>(115)</sup>. The average daily biotin intakes among Japanese individuals are 45.1 µg/day<sup>(116)</sup> and 60.7 µg/day<sup>(117)</sup>. According to the Standard Tables of Food Composition in Japan 2010<sup>(18)</sup> that listed biotin for the first time, the biotin intakes are approximately 30 µg/day<sup>(118)</sup> and 50 µg/day<sup>(119)</sup>. However, in many standard tables, the biotin component values of several foods are still not listed. Thus, the AIs were set based on the average dietary biotin intakes for adults from the previous total dietary assessment methods, i.e., 50 µg/day for adults aged 18 to 69 years.

The AI for children was calculated from the AI for adults (50 µg/day), using the following equation:

**The AI for adults aged 18 to 29 years × (reference BW for children/reference BW for adults aged 18 to 29 years) 0.75 × (1 + growth factor).**

Few studies have investigated the biotin requirements of elderly individuals. There are no data indicating that the biotin requirements of healthy individuals, aged over 70 years, differ from those of young adults. Thus, the AI for those aged over 70 years is the same as that for adults aged 18 to 29 years.

There were insufficient data to allow for the differences in requirements to be discerned between men and women, across all age groups. The lower value was adopted if the values of the men and women varied, in each age group.

### **2—1—2. Infants (AI)**

For infants aged 0 to 5 months, the mean concentration of biotin in breast milk is 5 µg/L<sup>(6,7,120,121)</sup>. The average milk intake is 0.78 L/day<sup>(8,9)</sup>, representing a daily biotin intake of 4.0 µg /day (rounding 3.9 µg /day). This value was set as the AI.

The AI for infants aged 6 to 11 months was calculated using the average of the values from following 2 expressions:

**Expression 1: The AI for infants aged 0 to 5 months × (reference BW for infants aged 6 to 11 months/reference BW for infants aged 0 to 5 months) 0.75**

**Expression 2: the AI for adults aged 18 to 29 years × (reference BW for infants aged 6 to 11 months/reference BW for adults aged 18 to 29 years) 0.75 × (1 + growth factor)**

Thus, the AI was determined to be 10 µg/day for infants aged 6 to 11 months.

### **2—1—3. Pregnant Women (AI)**

Pregnant women have reduced serum biotin concentrations, as well as reduced biotin excretion in the urine. In contrast, the urinary excretion of organic acids such as 3-hydroxyisovaleric acid increases during late pregnancy<sup>(122)</sup>. These findings indicate that pregnancy increases biotin requirements. However, there are no data on the additional amount required by pregnant women. Thus, the AI for pregnant women was set at the AI of non-pregnant women.

### **2—1—4. Lactating Women (AI)**

The amount of biotin required during lactation should be calculated from the differences in the biotin requirements of lactating and non-lactating women of a similar age. However, no such data are available. Thus, the AI for lactating women was set at the AI of non-lactating women.

## **3. To Avoid Excessive Intake**

### **3—1. Dietary Intake**

No regular food contains more than several dozen µg of folate/100 g except for liver. No study till date has reported unfavorable outcomes due to an excessive intake of regular food.

### **3—2. Method Used to Set the UL**

There was insufficient evidence for the determination of the UL for healthy individuals. Excessive biotin intake of 200 mg/day is not associated with adverse effects, even in patients with biotin-responsive inborn errors of metabolism<sup>(114)</sup>.

II Energy and Nutrients  
Vitamins (2) Water-soluble Vitamins  
Biotin

#### **4. For the Prevention of the Development and Progression of LRDs**

##### **4—1. The Association with LRDs**

No relevant data were available. The DG was not determined due to a lack of data.

## Vitamin C

### 1. Background Information

#### 1—1. Definition and Classification

The present DRIs were determined as the amount of ascorbic acid. Vitamin C (ascorbic acid) is a compound of (R)-3, 4-Dihydroxy-5-[(S)-1, 2-Dihydroxyethyl] furan-2(5H)-one, commonly known as L-ascorbic acid or ascorbic acid. Vitamin C freely exists as L-ascorbic acid in its reduced form, or L-dehydroascorbic acid in its oxidized form.

#### 1—2. Function

Vitamin C is essential for the biosynthesis of collagen, skin, and cells. Vitamin C deficiency cause scurvy. Vitamin C also has antioxidant functions.

#### 1—3. Digestion, Absorption and Metabolism

Vitamin C is transported to the blood after intestinal absorption. The digestion processes related to this vitamin can be affected by the type of food, and other dietary sources. The relative bioavailability of vitamin C is 90% up to an intake of 200 mg/day, and less than 50% at more than 1 g/day, pointing to differences between dietary intake and intake in the form of supplements<sup>(123)</sup>. The body's vitamin C level is maintained through various mechanisms, and the plasma concentration is saturated at an intake of about 400 mg/day<sup>(124,125)</sup>.

### 2. To Avoid Inadequacy

#### 2—1. Factors to be Considered in Estimating Requirements

The demand is higher for smokers and passive smokers than non-smokers<sup>(121,126–128)</sup>. Compared to others in the same age group, the vitamin C intakes of these individuals must be higher amounts than the RDA.

#### 2—2. Method Used to Set the EAR and RDA

##### 2—2—1. Adults and Children (EAR and RDA)

Severe vitamin C deficiency results in scurvy, which may be preventable by an ascorbic acid intake of 6-12 mg/day<sup>(129,130)</sup>. Optimal antioxidant activity in the plasma, and the prevention of cardiovascular disease are achieved at a plasma ascorbic acid concentration of 50  $\mu\text{mol/L}$ <sup>(131)</sup>.

A meta-analysis of 36 studies (participants' age: 15 to 96 years) that examined the association between vitamin C intake and plasma concentration reported that an vitamin C intake of 83.4 mg/day was necessary for the plasma vitamin C level to be maintained at 50  $\mu\text{mol/L}$ <sup>(125,132)</sup>. From these findings, the EAR for adults aged 18 to 29 years was determined to be 83.4 mg/day; this method was preferred for the setting of a value for the prevention of scurvy.

The RDA was calculated as the EAR  $\times$  1.2. The differences in the requirements between sexes were not considered<sup>(125)</sup>.

The EAR and RDA for children was calculated from the EAR and RDA for adults aged 18 to 29 years, using the following equation:

**EAR (RDA) for adults aged 18 to 29 years  $\times$  (reference BW for children/reference BW for adults aged 18 to 29 years) 0.75  $\times$  (1 + growth factor).**

The lower value was adopted if the values differed between the men and women, in each age group.

The meta-analysis stated above conducted a separate analysis using studies examining individuals aged 15-65 years, and those examining adults aged 60-96 years. The intake required for the achievement of the same plasma vitamin C level was higher in the latter analysis<sup>(132)</sup>. Therefore, elderly individuals may need to consume a higher amount of vitamin C; however, it was difficult to set a value specifically for this age group. Thus, the EAR and RDA values were adopted from those applicable to adults aged 18 to 69 years.

In a vitamin C depletion–repletion study conducted in men and women, the excretion of unmetabolized ascorbic acid into the urine was not detectable at an intake of 50 to 60 mg/day, but was detectable at an intake of 100 mg/day, under conditions in which the leukocyte vitamin C, as an indicator of body store, was saturated<sup>(124,125)</sup>. This finding supports the setting of an RDA value of 100 mg/day.

## **2—2—2. Additional Amount for Pregnant Women (EAR, RDA)**

The additional amounts were calculated based on the intake of vitamin C required to prevent infant scurvy. Thus, the additional EAR was set at 10 mg/day<sup>(133)</sup>. The additional RDA was set by assuming a coefficient of 1.2, yielding 10 mg/day (rounding 12 mg/day).

## **2—2—3. Additional Amount for Lactating Women (EAR, RDA)**

The additional EAR for lactating women was calculated based on the mean concentration of vitamin C in breast milk (50 mg/L<sup>(6,7,69)</sup>), the average milk volume (0.78 L/day)<sup>(8,9)</sup>, and the relative bioavailability (100%)<sup>(1)</sup>, yielding 40 mg/day (rounding 39 mg/day). The additional RDA was calculated as the EAR  $\times$  1.2, yielding 50 mg/day (rounding 46.8 mg/day).

## **2—3. Method Used to Set AI**

### **2—3—1. Infants (AI)**

The mean concentration of vitamin C in breast milk is 50 mg/L. The average milk intake is 0.78 L/day<sup>(8,9)</sup>, representing a daily vitamin C intake of about 40 mg/day (rounding 39 mg/day). This value was set as the AI.

The AI for infants aged 6 to 11 months was calculated using the average of the values from the following 2 expressions:

**Expression 1: the AI for infants aged 0 to 5 months × (reference BW for infants aged 6 to 11 months/reference BW for infants aged 0 to 5 months) 0.75**

**Expression 2: the EAR for adults aged 18 to 29 years × (reference BW for infants aged 6 to 11 months/reference BW for adults aged 18 to 29 years) 0.75 × (1 + growth factor)**

Thus, the AI was determined to be 40 mg/day for infants aged 6 to 11 months.

### **3. To Avoid Excessive Intake**

#### **3—1. Dietary Intake**

Few regular foods contain more than 100 mg of vitamin C/100 g; however, no studies till date have reported unfavorable outcomes due to the excessive intake of regular food.

#### **3—2. Method Used to Set the Tolerable Upper Intake level**

Generally, the intake of vitamin C intake is regarded as safe for healthy individuals, as the excess intake merely results in a lower absorption rate from the intestine, and enhanced excretion in the urine following absorption<sup>(124,125,134)</sup>. Thus, no UL for vitamin C was set at present.

However, for patients with renal dysfunction, the intake of several grams of vitamin C may increase the risk of kidney stones<sup>(135,136)</sup>. Acute gastrointestinal intolerance was observed following excess intake; for example, an intake of 3 to 4 g/day induced diarrhea<sup>(137)</sup>. An intake higher than 1 g/day from supplements is not advised<sup>(124,125,138)</sup>.

### **4. For the Prevention of the Development and Progression of LRDs**

#### **4—1. The Association with LRDs**

##### **4—1—1. Prevention of Disease Development**

Several reports have stated that there is no benefit in consuming more than 1 g/day of vitamin C<sup>(124–126,136)</sup>. The positive effects of vitamin C supplementation have not been clearly studied<sup>(132)</sup>.

##### **4—1—2. Prevention of Disease Progression**

No relevant data were available. The DG was not determined due to a lack of data.

### **5. Future Dietary Reference Intakes for Japanese**

It is important to reconsider if the use of EAR and RDA, or DG is more appropriate for vitamin C in the DRIs. Additionally, the outcomes used in the setting of the DRIs should also be reviewed in the future.

II Energy and Nutrients  
 Vitamins (2) Water-soluble Vitamins

**DRIs for Vitamin B<sub>1</sub> (mg/day) <sup>1</sup>**

Gender	Males			Females		
	EAR	RDA	AI	EAR	RDA	AI
Age etc.						
0-5 months	—	—	0.1	—	—	0.1
6-11 months	—	—	0.2	—	—	0.2
1-2 years	0.4	0.5	—	0.4	0.5	—
3-5 years	0.6	0.7	—	0.6	0.7	—
6-7 years	0.7	0.8	—	0.7	0.8	—
8-9 years	0.8	1.0	—	0.8	0.9	—
10-11 years	1.0	1.2	—	0.9	1.1	—
12-14 years	1.2	1.4	—	1.1	1.3	—
15-17 years	1.3	1.5	—	1.0	1.2	—
18-29 years	1.2	1.4	—	0.9	1.1	—
30-49 years	1.2	1.4	—	0.9	1.1	—
50-69 years	1.1	1.3	—	0.9	1.0	—
70+ years	1.0	1.2	—	0.8	0.9	—
Pregnant women (additional)	/			+0.2	+0.2	—
Lactating women (additional)				+0.2	+0.2	—

<sup>1</sup> Calculated using estimated energy requirement for PAL II.

Notice: EARs are calculated from the intake where urinary excretion of vitamin B<sub>1</sub> starts to increase (i.e. internal saturation intake), not from the minimum intake required to prevent beriberi (one of the major vitamin B<sub>1</sub> deficiency diseases).

II Energy and Nutrients  
 Vitamins (2) Water-soluble Vitamins

**DRIs for Vitamin B<sub>2</sub> (mg/day) <sup>1</sup>**

Gender	Males			Females		
	EAR	RDA	AI	EAR	RDA	AI
Age etc.						
0-5 months	—	—	0.3	—	—	0.3
6-11 months	—	—	0.4	—	—	0.4
1-2 years	0.5	0.6	—	0.5	0.5	—
3-5 years	0.7	0.8	—	0.6	0.8	—
6-7 years	0.8	0.9	—	0.7	0.9	—
8-9 years	0.9	1.1	—	0.9	1.0	—
10-11 years	1.1	1.4	—	1.1	1.3	—
12-14 years	1.3	1.6	—	1.2	1.4	—
15-17 years	1.4	1.7	—	1.2	1.4	—
18-29 years	1.3	1.6	—	1.0	1.2	—
30-49 years	1.3	1.6	—	1.0	1.2	—
50-69 years	1.2	1.5	—	1.0	1.1	—
70+ years	1.1	1.3	—	0.9	1.1	—
Pregnant women (additional)	/			+0.2	+0.3	—
Lactating women (additional)				+0.5	+0.6	—

<sup>1</sup> Calculated using estimated energy requirement for PAL II.

Notice: EARs are calculated from the intake where urinary excretion of vitamin B<sub>2</sub> starts to increase (i.e. internal saturation intake), not from the minimum intake required to prevent dermatitis such as cheilitis, perleche and glossitis (some of the major vitamin B<sub>2</sub> deficiency diseases).

II Energy and Nutrients  
 Vitamins (2) Water-soluble Vitamins

**DRIs for Niacin (mg NE/day) <sup>1</sup>**

Gender Age etc.	Males				Females			
	EAR	RDA	AI	UL <sup>2</sup>	EAR	RDA	AI	UL <sup>2</sup>
0-5 months <sup>3</sup>	—	—	2	—	—	—	2	—
6-11 months	—	—	3	—	—	—	3	—
1-2 years	5	5	—	60(15)	4	5	—	60(15)
3-5 years	6	7	—	80(20)	6	7	—	80(20)
6-7 years	7	9	—	100(30)	7	8	—	100(25)
8-9 years	9	11	—	150(35)	8	10	—	150(35)
10-11 years	11	13	—	200(45)	10	12	—	200(45)
12-14 years	12	15	—	250(60)	12	14	—	250(60)
15-17 years	14	16	—	300(75)	11	13	—	250(65)
18-29 years	13	15	—	300(80)	9	11	—	250(65)
30-49 years	13	15	—	350(85)	10	12	—	250(65)
50-69 years	12	14	—	350(80)	9	11	—	250(65)
70+ years	11	13	—	300(75)	8	10	—	250(60)
Pregnant women (additional)	/				—	—	—	—
Lactating women (additional)					+3	+3	—	—

NE = niacin equivalent = niacin + 1/60 tryptophan.

<sup>1</sup> Calculated using estimated energy requirement for PAL II.

<sup>2</sup> Quantity as nicotinamide (mg). Values in parentheses are quantities as nicotinic acid (mg). Calculated using the reference weight.

<sup>3</sup> The unit is mg/day.

II Energy and Nutrients  
 Vitamins (2) Water-soluble Vitamins

**DRIs for Vitamin B<sub>6</sub> (mg/day) <sup>1</sup>**

Gender Age etc.	Males				Females			
	EAR	RDA	AI	UL <sup>2</sup>	EAR	RDA	AI	UL <sup>2</sup>
0-5 months	—	—	0.2	—	—	—	0.2	—
6-11 months	—	—	0.3	—	—	—	0.3	—
1-2 years	0.4	0.5	—	10	0.4	0.5	—	10
3-5 years	0.5	0.6	—	15	0.5	0.6	—	15
6-7 years	0.7	0.8	—	20	0.6	0.7	—	20
8-9 years	0.8	0.9	—	25	0.8	0.9	—	25
10-11 years	1.0	1.2	—	30	1.0	1.2	—	30
12-14 years	1.2	1.4	—	40	1.1	1.3	—	40
15-17 years	1.2	1.5	—	50	1.1	1.3	—	45
18-29 years	1.2	1.4	—	55	1.0	1.2	—	45
30-49 years	1.2	1.4	—	60	1.0	1.2	—	45
50-69 years	1.2	1.4	—	55	1.0	1.2	—	45
70+ years	1.2	1.4	—	50	1.0	1.2	—	40
Pregnant women (additional)					+0.2	+0.2	—	—
Lactating women (additional)					+0.3	+0.3	—	—

<sup>1</sup> Calculated using RDAs in DRIs for proteins (excludes additional values for pregnant or lactating women).

<sup>2</sup> Quantity as pyridoxine, not as dietary vitamin B<sub>6</sub>.

II Energy and Nutrients  
 Vitamins (2) Water-soluble Vitamins

**DRIs for Vitamin B<sub>12</sub> (µg/day)**

Gender	Males			Females		
Age etc.	EAR	RDA	AI	EAR	RDA	AI
0-5 months	—	—	0.4	—	—	0.4
6-11 months	—	—	0.5	—	—	0.5
1-2 years	0.7	0.9	—	0.7	0.9	—
3-5 years	0.8	1.0	—	0.8	1.0	—
6-7 years	1.0	1.3	—	1.0	1.3	—
8-9 years	1.2	1.5	—	1.2	1.5	—
10-11 years	1.5	1.8	—	1.5	1.8	—
12-14 years	1.9	2.3	—	1.9	2.3	—
15-17 years	2.1	2.5	—	2.1	2.5	—
18-29 years	2.0	2.4	—	2.0	2.4	—
30-49 years	2.0	2.4	—	2.0	2.4	—
50-69 years	2.0	2.4	—	2.0	2.4	—
70+ years	2.0	2.4	—	2.0	2.4	—
Pregnant women (additional)	/			+0.3	+0.4	—
Lactating women (additional)				+0.7	+0.8	—

II Energy and Nutrients  
 Vitamins (2) Water-soluble Vitamins

**DRIs for Folic Acid ( $\mu\text{g}/\text{day}$ )** <sup>1</sup>

Gender	Males				Females			
Age etc.	EAR	RDA	AI	UL <sup>2</sup>	EAR	RDA	AI	UL <sup>2</sup>
0-5 months	—	—	40	—	—	—	40	—
6-11 months	—	—	60	—	—	—	60	—
1-2 years	70	90	—	200	70	90	—	200
3-5 years	80	100	—	300	80	100	—	300
6-7 years	100	130	—	400	100	130	—	400
8-9 years	120	150	—	500	120	150	—	500
10-11 years	150	180	—	700	150	180	—	700
12-14 years	190	230	—	900	190	230	—	900
15-17 years	220	250	—	900	220	250	—	900
18-29 years	200	240	—	900	200	240	—	900
30-49 years	200	240	—	1,000	200	240	—	1,000
50-69 years	200	240	—	1,000	200	240	—	1,000
70+ years	200	240	—	900	200	240	—	900
Pregnant women (additional)					+200	+240	—	—
Lactating women (additional)					80	+100	—	—

<sup>1</sup> In order to reduce the risk of neural tube closure, an additional intake of 400  $\mu\text{g}/\text{day}$  of pteroylmonoglutamic acid is recommended for women who are planning to become pregnant or may be pregnant.

<sup>2</sup> Quantity as pteroylmonoglutamic acid contained in dietary supplement and vitamin-enriched food.

II Energy and Nutrients  
 Vitamins (2) Water-soluble Vitamins

**DRIs for Pantothenic Acid (mg/day)**

Gender	Males	Females
Age etc.	AI	AI
0-5 months	4	4
6-11 months	3	3
1-2 years	3	3
3-5 years	4	4
6-7 years	5	5
8-9 years	5	5
10-11 years	6	6
12-14 years	7	6
15-17 years	7	5
18-29 years	5	4
30-49 years	5	4
50-69 years	5	5
70+ years	5	5
Pregnant women	/	5
Lactating women		5

II Energy and Nutrients  
 Vitamins (2) Water-soluble Vitamins

**DRIs for Biotin ( $\mu\text{g}/\text{day}$ )**

Gender	Males	Females
Age etc.	AI	AI
0-5 months	4	4
6-11 months	10	10
1-2 years	20	20
3-5 years	20	20
6-7 years	25	25
8-9 years	30	30
10-11 years	35	35
12-14 years	50	50
15-17 years	50	50
18-29 years	50	50
30-49 years	50	50
50-69 years	50	50
70+ years	50	50
Pregnant women	/	50
Lactating women		50

II Energy and Nutrients  
 Vitamins (2) Water-soluble Vitamins

**DRIs for Vitamin C (mg/day)**

Gender	Males			Females		
Age etc.	EAR	RDA	AI	EAR	RDA	AI
0-5 months	—	—	40	—	—	40
6-11 months	—	—	40	—	—	40
1-2 years	30	35	—	30	35	—
3-5 years	35	40	—	35	40	—
6-7 years	45	55	—	45	55	—
8-9 years	50	60	—	50	60	—
10-11 years	60	75	—	60	75	—
12-14 years	80	95	—	80	95	—
15-17 years	85	100	—	85	100	—
18-29 years	85	100	—	85	100	—
30-49 years	85	100	—	85	100	—
50-69 years	85	100	—	85	100	—
70+ years	85	100	—	85	100	—
Pregnant women (additional)	/			+10	+10	—
Lactating women (additional)				+40	+45	—

Notice: EARs are calculated from cardiovascular disease prevention effects and antioxidative effects, not from intake sufficient enough to avoid scurvy.

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## **Minerals**

### **(1) Macrominerals**

## **Sodium**

### **1. Background Information**

#### **1–1. Definition and Classification**

Sodium is an alkali metal element (atomic number: 11, Na).

### **2. To Avoid Inadequacy**

#### **2–1. Factors to be Considered in Estimating Requirements**

##### **Determining the Dietary Reference Intakes (DRIs)**

The WHO's guideline states that the intake of sodium should be no more than 200 to 500 mg/day<sup>(1)</sup>. Based on the belief that endogenous sodium loss is equal to sodium requirement, the estimated average requirement (EAR) was established with the goal of compensating for endogenous loss, using the same method as the DRIs for Japanese 2010<sup>(2)</sup>. However, the value is lower than 1% of the intake distribution value, as determined by the 2010 and 2011 National Health and Nutrition Survey (NHNS)<sup>(3)</sup>. Therefore, the sodium requirements are irrelevant with respect to regular dietary intake, and setting an EAR might not be practical. The EAR was calculated for reference. Since the recommended dietary allowance (RDA) has no significance when the amount recommended is utilized, it was not calculated.

The present DRIs for sodium were set with the aim of preventing the increased risk for lifestyle-related diseases (LRDs); therefore, the tentative dietary goal (DG) for preventing LRDs was determined.

#### **2–2. Method Used to Set the EAR and RDA**

##### **2–2–1. Background Information**

Under conditions of normal renal function, sodium deficiency does not occur, as sodium balance is maintained through sodium reabsorption in the kidneys. The unavoidable sodium loss is the sum of the excretion via urine, stool, skin, or others at a dietary sodium intake of 0. The required sodium intake has been regarded as the loss because the sodium consumed is mainly absorbed by the intestine<sup>(4)</sup>.

##### **2–2–2. Adults and Children (EAR, RDA)**

From the results of a review of traditional studies, the endogenous sodium loss of adults was calculated to be 0.023 mg (0.001 mmol)/kg body weight (BW)/day from the feces, 0.23 mg (0.01 mmol)/kg BW/day from urine, and 0.92 mg (0.04 mmol)/kg BW/day from skin (total: 1.173 mg (0.051 mmol)/kg BW/day)<sup>(4)</sup>. On applying these values to men aged 18 to 29 years, the endogenous sodium loss was 74 (1.173 × 63.0) mg/day or 3.2 (0.051 × 63.0)

mmol/day. The endogenous sodium loss was set at 115 mg/day (5 mmol/day) in the American dietary requirement 1989<sup>(5)</sup>, and 69 to 490 mg/day (3 to 20 mmol/day) in the DRIs for the UK 1991<sup>(6)</sup>. Thus, the endogenous sodium loss of adults is lower than 500 mg/day or 600 mg/day (sodium chloride equivalent of 1.5 g/day) even after considering interindividual variations (with a coefficient of variation of 10%). This value was set as the EAR for adults. In practical terms, the regular dietary intake of salt is lower than 1.5 g/day.

Working or exercising in hot environments can lead to the loss of a significant amount of sodium through profuse perspiration; hydration with a small amount of added salt may be needed in such cases<sup>(7)</sup>. In view of the rising summer temperatures in Japan, it is necessary to consume a moderate amount of salt. However, excessive intakes can be unfavorable in the prevention of the onset and progression of LRDs, and improvement in their severity.

For children, since no data were available, the EAR was not determined.

### **2–2–3. Additional Amount for Pregnant and Lactating Women (EAR, RDA)**

The amount required by pregnant women was estimated to be approximately 21.85 g (950 mmol) for the maintenance of the increase of the mothers' tissues, fetus and placenta<sup>(8)</sup>. These increases occur over a period of 9 months, which is equivalent to 0.08 g (3.5 mmol)/day (a sodium chloride equivalent of 0.2 g/day). Since the regular dietary intake of salt is considered to be sufficient, pregnant women do not require an additional intake of sodium.

The average concentration of sodium in the breast milk of Japanese mothers is 135 mg/L<sup>(9,10)</sup>. Considering the daily milk secretion is 0.78 L/day, the amount of sodium consumed is 105 mg/day (a sodium chloride equivalent of 0.27 g/day). Since their regular dietary intake of salt is considered sufficient, lactating women do not require an additional intake of sodium.

### **2–3. Method Used to Set the Adequate Intake (AI)**

#### **2–3–1. Infants (AI)**

For infants aged 0-5 months, the AI was calculated using the average concentration of sodium in breast milk (135 mg/L)<sup>(9,10)</sup> and average milk intake (0.78 L/day)<sup>(11,12)</sup>, yielding 100 mg/day (a sodium chloride equivalent of 0.3 mg/day).

For infants aged 6 to 11 months, the AI was calculated using the average consumption of sodium from breast milk<sup>(13,14)</sup> and complementary food<sup>(15)</sup>.

The daily sodium intake was calculated as 559 mg/day (72 mg/day (135 mg/L × 0.53 L/day) from breast milk and 487 mg/day from complementary food); therefore, the AI was set at 600 mg/day (a sodium chloride equivalent of 1.5 g/day).

### **3. To Avoid Excessive Intake**

#### **3–1. Dietary Intake**

The major sources of sodium intake are dietary salt (sodium chloride), and sodium-containing seasonings. The amount of sodium chloride can be calculated as follows:

**Sodium chloride equivalent (g) = sodium (g) × 58.5/23 = sodium (g) × 2.54**

The Standard Tables of Food Composition in Japan 2010<sup>(16)</sup> used the above-stated formula to measure the sodium content in food. There are various kinds of sodium compounds other than sodium chloride in various foods, especially in processed foods that contain sodium chloride.

Although sodium exists in foods as sodium salt or sodium ion, humans consume most of the sodium as sodium chloride (NaCl). Therefore, sodium intake is often expressed as sodium chloride equivalents.

### **3–2. The Tolerable the Tolerable Upper Intake level (UL)**

The ULs for sodium have not been determined, as DGs have been set for similar purposes.

## **4. For the Prevention of the Development and Progression of LRDs**

### **4–1. The Association between Sodium and Major LRDs**

The development and maintenance of hypertension are based on the interaction of heritable factors and environmental factors (lifestyle habits). Lifestyle modification plays an important role in the prevention and treatment of hypertension. In addition to patients with hypertension, those with heritable factors for hypertension or a high normal blood pressure (130-139/85-89 mmHg) should work on modifying their lifestyle habits, especially in terms of diet.

Results from large Western clinical studies<sup>(17–22)</sup> showed that the blood pressure was significantly reduced when the sodium intake was within 6 g/day. Worldwide, the major guidelines for the treatment of hypertension recommend that the salt intake be reduced to 6 g/day or less, based on the above-stated findings. The Japanese Guideline for the Management of Hypertension by the Japanese Society of Hypertension (JSH2009)<sup>(23)</sup> also set a threshold of 6 g/day for salt intake.

In Western countries, the guidelines for salt reduction are stricter. In 2010, the American Heart Association issued a recommendation stating that the goal for sodium intake should be 2,300 mg (a sodium chloride equivalent of 5.8 g/day) for healthy adults, and 1,500 mg (a sodium chloride equivalent of 5.8 g/day) for those at risk (having hypertension, belonging to a black race, or being middle-aged)<sup>(24)</sup>. The 2013 WHO guideline for the general population strongly recommends a dietary salt intake goal of 5 g/day<sup>(1)</sup>. Although the 2005 US-Canada DRIs set a salt intake lower than 1,500 mg/day as a dietary goal<sup>(25)</sup>, this goal has been deleted in the recent DRIs due to a lack of relevant data<sup>(26)</sup>. Several studies have reported on the association between dietary salt and cancer, especially gastric cancer. According to a review of the World Cancer Research Fund/American Institute for Cancer Research, the intake of salted foods and dietary salt are likely to increase the risk of gastric cancer<sup>(27)</sup>. Japanese cohort studies have reported that dietary salt intake is positively associated with gastric cancer prevalence and

mortality<sup>(28-30)</sup>, while the frequency of the intake of salt-cured foods was associated with the risk of gastric cancer<sup>(28)</sup>. A meta-analysis reported that a high salt intake increased the risk of gastric cancer<sup>(31)</sup>, while another meta-analysis reported the presence of a dose-response relationship between salt intake and gastric cancer risk<sup>(32)</sup>.

#### 4-2. Method Used to Set the DG

In the DRIs for Japanese 2010<sup>(2)</sup>, the target DG to attain over a period of 5 years was calculated to be less than 9 g/day for men, and less than 7.5 g/day for women. Since that point, the average intake of salt has decreased by approximately 0.5 g in men, and 1 g in women (the median intake was 10.5-11.8 g in men, and 8.8-10.0 g in women in the 2010 and 2011 NHNS)<sup>(3)</sup>. Although the current intake does not meet the DGs set in the previous DRI, the new DG should be set as low as possible, as a salt intake less than 6 g/day is favorable for the prevention and management of hypertension.

The 2013 WHO guideline for the general population strongly recommends achieving a dietary salt intake goal of 5 g/day<sup>(1)</sup>. However, this goal was achieved in only about 5% of the participants in the 2010 and 2011 NHNS<sup>(3)</sup>. Considering that the intraindividual variation of dietary salt intake (34-36%) is greater than the interindividual variation (15-20%)<sup>(33)</sup>, some individuals may consume less than 5 g/day habitually. Therefore, for the sake of feasibility, the DG should not be lower than 5 g/day.

Accordingly, the DG for sodium was set at the median of 5 g/day and the current intake (the median intake from the 2010 and 2011 NHNS<sup>(3)</sup>). This value was applied for the DG, except among those aged 50-69 years; in such cases, smoothing was conducted.

The 2013 WHO guideline for the general population strongly recommends a dietary salt intake goal of 5 g/day<sup>(1)</sup>, through the calculation of the values for children by adjusting the adults' values for energy requirement. The current DRIs also extrapolated the values for adults aged 18 to 29 years into the values for children, using estimated energy requirement (level 2 of physical activity). The DG was set at the median of both these values, and the current intake (the median intake from the 2010 and 2011 NHNS<sup>(3)</sup>) as follows:

$$DG_x = (5.0 \times (EER_x/EER_o) + I_x \text{ [g/day]})/2$$

EER<sub>x</sub>: Estimated Energy Requirement for each age and sex group (kcal/day)

EER<sub>o</sub>: Estimated Energy Requirement for adults aged 18-29 years (kcal/day)

I<sub>x</sub>: the median intake (sodium chloride equivalent) in the 2010 and 2011 NHNS<sup>(3)</sup> (g/day)

Smoothing was conducted for girls aged 12-14 years, and those aged 15-17 years. No additional amount was set for pregnant or lactating women.

II Energy and Nutrients  
 Minerals (1) Macrominerals  
 Sodium

**Table 1. Methods to determine the DG for sodium (salt equivalent: g/day)**

Gender	Male				Female			
Age (years)	(A)	(B)	(C)	(D)	(A)	(B)	(C)	(D)
1-2	1.8	4.3	3.0	3.0	2.3	4.2	3.3	3.5
3-5	2.5	5.9	4.2	4.0	3.2	5.4	4.3	4.5
6-7	2.9	7.2	5.1	5.0	3.7	7.0	5.3	5.5
8-9	3.5	7.8	5.7	5.5	4.4	8.1	6.2	6.0
10-11	4.2	9.1	6.7	6.5	5.4	8.4	6.9	7.0
12-14	4.9	10.7	7.8	8.0	6.2	9.0	7.6	7.5 ↓
15-17	5.4	11.0	8.2	8.0	5.9	9.1	7.5	7.5 ↓
18-29	5.0	10.5	7.8	8.0	5.0	8.7	6.9	7.0
30-49	5.0	10.7	7.9	8.0	5.0	8.8	6.9	7.0
50-69	5.0	11.8	8.4	8.5 ↓	5.0	10.0	7.5	7.5 ↓
70+	5.0	10.7	7.8	8.0	5.0	9.4	7.2	7.0

(A) Recommendation in guideline of WHO in 2013. The values for 1-17 years old were extrapolated using estimated energy requirement.

(B) Median value of the sodium intake (salt equivalent) in NHNS2010 and NHNS2011 (g/day)

(C) Intermediate value of (A) and (B)

(D) Value after rounding, ↑ and ↓ present the way to smooth the calculated value (up and down)

## Potassium

### 1. Background Information

#### 1–1. Definition and Classification

Potassium is an alkali metal element (atomic number: 19: K) that is found in high quantities in fruits and vegetables; the amount of potassium decreases as the degree of processing or refinement increases<sup>(34,35)</sup>.

#### 1–2. Function

As the main cation contained in intracellular fluid, potassium has an important role in the determination of the osmotic pressure of the aqueous humors and maintaining acid-base balance. It also participates in nerve transmission, muscle contraction, and regulation of vascular tone<sup>(36)</sup>.

Potassium deficiency is rarely observed in healthy individuals, and typically affects only those with diarrhea or heavy perspiration, or those taking diuretics. The average sodium intake in Japan is higher than that in many other countries<sup>(3)</sup>. As the urinary excretion of sodium is related to potassium intake, it is believed that increasing the ingestion of potassium is important for Japanese individuals. Moreover, recent animal and epidemiological studies indicated that an increased potassium intake may be associated with a reduction in blood pressure and the prevention of stroke<sup>(34)</sup>.

#### 1–3. Digestion, Absorption and Metabolism

Although the absorption of potassium is passive, it is released actively in the ileum and large intestine. It is released in the large intestine at 25 mEq/L. In the case of severe diarrhea, the plasma potassium level sharply decreases, as more than 16 L/day of intestinal juices could be lost (hypokalemia).

### 2. To Avoid Inadequacy

#### 2–1. Factors to be Considered in Estimating Requirements

Since potassium is present in various foods, its deficiency rarely occurs with the consumption of a regular diet. Few scientific data are available for the establishment of the EAR and RDA.

Therefore, the AI was determined to compensate for endogenous potassium loss and the maintenance of potassium balance at the present intake level. Moreover, the DG was determined for the prevention of the onset and progression of LRDs.

#### 2–2. Method Used to Set the AI

##### 2–2–1. Adults (AI)

The endogenous potassium loss has been estimated as follows: stool, 4.84 mg/kg BW/

day; urine, 2.14 mg/kg BW/ day; skin, 2.34 mg/kg BW/day (5.46 mg/kg BW/ day at high temperatures, at rest); and total, 9.32 mg/kg BW/day (12.44 mg/kg BW/day at high temperatures, at rest)<sup>(4)</sup>. Another study reported the total endogenous potassium loss to be 15.64 mg/kg BW /day<sup>(37)</sup>. A study reported that when the loss from stool is 400 mg/day, and that from urine is 200 to 400 mg/day, the loss from sweat or others can be ignored, indicating that an intake of 800 mg/day is sufficient to maintain balance<sup>(4)</sup>. However, in that study, the plasma potassium levels of some participants decreased at this level; thus, 1,600 mg/day (23 mg/kg BW/day) was reported to be an appropriate amount. In research conducted in other countries, an intake of 1,600 mg was found to be adequate in the maintenance of potassium balance<sup>(38)</sup>. From these findings, 1,600 mg/day can be considered as the amount at which balance can be maintained safely.

Based on data from the 2010 and 2011 NHNS, the median intakes of potassium were 2,309 mg/day and 2,138 mg/day in men and women, respectively<sup>(3)</sup>. The current intake of Japanese individuals was found to exceed the amount required for the maintenance of balance. The median intake of potassium in men aged over 50 years was approximately 2,500 mg/day; therefore, AIs of 2,500 mg/day and 2,000 mg/day were set for men and women, respectively; these values are not unrealizable, considering the differences in energy intake.

#### **2–2–2. Children (AI)**

Based on the AI of adults aged 18 to 29 years, the AI was extrapolated by the 0.75<sup>th</sup> power of the BW ratio, in consideration of the growth factors.

#### **2–2–3. Infants (AI)**

The AI for infants aged 0-5 months infants was calculated using the average concentration of potassium in breast milk (470 mg/L)<sup>(9,10)</sup>, and the daily intake of breast milk (0.78 L/day)<sup>(11,12)</sup>, yielding 367 mg/day.

The AI for infants aged 6 to 11 months was calculated using the amount of potassium obtained from breast milk (249 mg/day (470 mg/L × 0.53 L<sup>(13,14)</sup>) and complementary food (492 mg/day)<sup>(15)</sup>.

By rounding, the AIs were set at 400 mg/day and 700 mg/day for infants aged 0-5 months and 6-11 months, respectively.

#### **2–2–4. Pregnant and Lactating Women (AI)**

During pregnancy, the potassium demand increases, for the development of fetal tissues. This demand was reported to be 12.5 g<sup>(37)</sup>. Considering this value as the demand over a period of 9 months, the daily requirement was calculated as 46 mg/day. This amount can be obtained from regular meals; therefore, an increase in the intake of potassium is not required during pregnancy. The median value from the 2007 to 2011 NHNS<sup>(39)</sup> was calculated to be 1,902 mg/day. The AI for women of childbearing age is 2,000 mg/day. From these data, the AI

for pregnant women was determined to be 2,000 mg/day.

For lactating women, the median value of the 2007 to 2011 NHNS<sup>(39)</sup> was calculated to be 2,161 mg/day. This value is considered to be sufficient for the maintenance of potassium balance, and thus, was adopted as the AI, yielding a value of 2,200 mg/day by rounding.

### 3. To Avoid Excessive Intake

If renal function is normal, the potassium intake from regular meals will not lead to excessive potassium levels. Therefore, the UL was not determined. However, caution must be exercised in terms of potassium intake, if renal disorders are present.

## 4. For the Prevention of the Development and Progression of LRDs

### 4-1. The Association between Sodium and Major LRDs

A meta-analysis of cohort studies<sup>(40)</sup> reported that, while increased potassium levels increased the risk of stroke, they did not affect the risk of cardiovascular diseases. An epidemiological study showed that the Na/K intake ratio was significantly associated with the risk of cardiovascular disease or all-cause mortality in the healthy population<sup>(41)</sup>. Thus, potassium intake should be evaluated in relation with salt intake. A recently published WHO guideline<sup>(34)</sup> recommends a potassium intake higher than 90 mmol (3,510 mg)/day. This value was determined from a meta-analysis which showed that a potassium intake of 90 to 120 mmol/day decreased the systolic blood pressure by 7.16 mmHg.

However, the presence of renal disorders requires attention, as these can cause hyperkalemia in their milder forms; therefore, those with renal function disorders should avoid the aggressive intake of potassium.

### 4-2. Method Used to Set the DG

The WHO reported that an intake of 3,510 mg potassium/day is desirable for the prevention of high blood pressure<sup>(34)</sup>. This value is considered an intake goal. However, considering the current intake of Japanese adults, this intake may be difficult to realize. Therefore, the DG was calculated using the following method. The reference was set at the current median intake of Japanese adults--2,384 mg for men and 2,215 mg for women--based on the 2010 and 2011 NHNS<sup>(3)</sup>. Then, the DGs were calculated by extrapolating by the 0.75<sup>th</sup> power of the BW ratio using the average reference BW (57.8 kg for adults) and reference BWs for each age and sex group (the average potassium intake and average reference BW were calculated solely from all the age and sex groups) as follows:

**2,856 mg/day × (reference BW for each age and sex group / 57.8 kg)<sup>0.75</sup>**

The higher of the two values was adopted as the DG. In this method, rounding at each 200 mg/day and smoothing were conducted.

No additional DG was set for pregnant and lactating women.

For children aged 1 to 5 years, few reports present quantitative evidence on potassium

II Energy and Nutrients  
 Minerals (1) Macrominerals  
 Potassium

intake, and its association with the prevention of LRDs. It is difficult to assess potassium intake, and no relevant data were available in Japan. Thus, for children aged 6 to 17 years alone, the DG was determined using the same method as that used for adults. The current average intake was adopted when the calculated amount exceeded it. In the WHO guideline<sup>(34)</sup>, the DG for adults was adjusted for energy requirement; however, the value for girls would be higher if the same DG as that used for boys is employed. Therefore, in the current DRIs, extrapolation was performed using the reference BW.

**Table 2. Method to determine the DG for Potassium (mg/day)**

Gender	Male				Female			
Age (years)	(A)	(B)	(C)	(D)	(A)	(B)	(C)	(D)
6-7	1,393	1,861	(B)	1,800	1,379	1,822	(B)	1,800
8-9	1,658	1,986	(B)	2,000	1,632	1,977	(B)	2,000
10-11	1,986	2,198	(B)	2,200	2,015	2,052	(B)	2,000
12-14	2,523	2,450	(A)	2,600	2,465	2,211	(A)	2,400
15-17	2,926	2,332	(A)	3,000	2,634	1,939	(A)	2,600
18-29	3,054	2,004	(A)	3,000	2,562	1,700	(A)	2,600
30-49	3,244	2,077	(A)	↓ 3,000	2,680	1,843	(A)	2,600
50-69	3,130	2,452	(A)	↓ 3,000	2,676	2,341	(A)	2,600
70+	2,937	2,459	(A)	3,000	2,543	2,293	(A)	2,600

(A) Extrapolated value from the reference value for DG calculation (mg/day).

(B) Median value of the sodium intake (salt equivalent) in NHNS2010 and NHNS2011.

(C) Value of the DG determined.

(D) Value after rounding, ↑ and ↓ present the way to smooth the calculated value (up and down)

## Calcium

### 1. Background Information

#### 1–1. Definition and Classification

Calcium is an alkali earth metal (atomic number: 20: Ca) that accounts for 1%- 2% of the total BW of humans, with more than 99% present in the bones and teeth, and the remaining 1% in the blood, tissue fluid, and cells.

#### 1–2. Function

The calcium concentration in the blood is controlled within a very narrow range (8.5 to 10.4 mg/dL). If the concentration decreases, the parathyroid hormone stimulates the absorption of calcium from bone, which undergoes repeated bone resorption (resorption of calcium from the bones) and bone formation (accumulation of calcium in the bones). Bone mass increases during growth, and begins to decrease in menopause or later, and then continues to decrease during the aging process. Calcium deficiency can cause osteoporosis, high blood pressure, or arteriosclerosis, while excessive calcium intake can cause hypercalcemia, hypercalciuria, calcification of soft tissues, urinary system calculus, prostate cancer, absorption disorders of iron and copper, or constipation.

#### 1–3. Digestion, Absorption and Metabolism

Orally digested calcium is predominantly absorbed in the upper part of the small intestine through active transport. The absorption rate is comparably low, at 25 to 30%, and is affected by various factors such as age, pregnancy/lactation, or other food compositions. Vitamin D promotes calcium absorption.

Absorbed calcium is regulated by bone accumulation, and the urine excretion pathway through the kidney. Therefore, calcium nutrient status should take into account intake, absorption from the intestine, bone metabolism, and urine excretion.

### 2. To Avoid Inadequacy

#### 2–1. Factors to be Considered in Estimating Requirements

As a biomarker for the requirement of calcium, bone health is important. This apart, calcium has been reported to be associated with LRDs such as blood pressure or obesity, although the effect has not been established<sup>(42)</sup>. At present, the requirement cannot be set using biomarkers other than bone health.

A meta-analysis showed a significant association between calcium intake, bone mass and bone mineral density<sup>(43–45)</sup>. A Japanese epidemiological study demonstrated a significant association between increased bone fracture prevalence and low calcium intake<sup>(46)</sup>. A meta-analysis of studies conducted in other countries reported no significant association between calcium intake and bone fracture prevalence<sup>(47)</sup>. While intervention studies reported that

calcium supplementation alone was not associated with the prevention of bone fracture<sup>(48,49)</sup>, calcium supplementation with vitamin D inhibited bone fracture development according to a meta-analysis<sup>(50,51)</sup>. However, another report negated the above-stated finding<sup>(52)</sup>; therefore, the results of epidemiological studies are not necessarily consistent.

In contrast, useful data have been accumulated for the estimation of the calcium intake required for the maintenance of bone mass using factorial methods. The US-Canada DRIs have set the EAR and RDA instead of the AI previously used<sup>(53)</sup>. Although the US-Canada DRIs used data from balance studies, the current DRIs adopted factorial modeling, as no balance study has been conducted in recent times.

## **2–2. Method Used to Set the EAR and RDA**

### **2–2–1. Background**

For those aged over 1 year, the EAR and RDA were calculated using the factorial method, which considers the amount of calcium accumulated in the body, excreted through urine and lost via dermal tissue, as well as the apparent rates. For the RDA, the interindividual difference in the requirement is unclear; however, the coefficient of variation was set as 10%. For infants, the AI was determined.

### **2–2–2. Factors for Factorial Modeling**

#### **2–2–2–1. Calcium Accumulation in the Body**

Few longitudinal studies have examined calcium accumulation in Japanese populations, especially among children. A Chinese study reported that the calcium accumulation in the body was 162.3 mg/day in girls aged 9.5 to 10.5 years, and the accumulation rate was 40.9%<sup>(54)</sup>. In that study, the mean calcium intake was 444 mg/day, which is approximately 200 mg/day lower than that of Japanese girls of a similar age. In a study that examined adolescents, the greatest calcium accumulation was observed at 13.4 years of age in boys with an average calcium intake of 359 mg/day (standard deviation [SD] 82), and at 11.8 years of age in girls with an average calcium intake of 284 mg/day (SD 59)<sup>(55)</sup>. Another study reported that the maximum accumulation was observed at 628.9 mg/day in boys, and the difference between the sexes was 171 mg/day<sup>(56)</sup>. It is known that calcium accumulation varies between ethnicities. A study reported that a calcium intake of 700 mg/day resulted in an accumulation of 367 mg/day in black participants; this value was 183 mg/day in white participants, among adolescent girls<sup>(57)</sup>. Although increased calcium intakes are not associated with race-related differences in the increase in the calcium accumulation, increased calcium intakes are associated with an increase in calcium accumulation, indicating that using the aforementioned results pertaining to calcium intakes higher than those in the normal Japanese diet may be problematic. Taking these into consideration, the accumulation for Japanese individuals was calculated.

The calcium accumulation per year was calculated based on the results of several

studies that examined the total body bone mineral content using dual-energy X-ray absorptiometry<sup>(58-67)</sup>, for each age and sex category. In a cross-sectional study on Japanese children, the accumulation was quite similar to that obtained from the current calculation<sup>(67)</sup>. For children younger than 6 years of age, the calcium accumulation was determined according to increases in the bone mineral content per year<sup>(68)</sup>.

#### **2-2-2-2. Urine Excretion and Percutaneous Loss of Calcium**

Urine calcium excretion can be calculated as  $BW^{0.75} \times 6$  mg/day, when calcium balance is maintained<sup>(69)</sup>. This calculation is similar to the 24-hour urine calcium excretion observed in a balance study in Japanese women<sup>(70,71)</sup>. Another study estimated the percutaneous loss to be approximately one-sixth of the urine excretion<sup>(72)</sup>. Based on these findings, the percutaneous loss was calculated by estimating the urine calcium excretion calculated by the reference BW for each age and sex category.

#### **2-2-2-3. Apparent Absorption**

The apparent absorption rate varies inversely with the calcium intake<sup>(57)</sup>. The intake level is higher in studies conducted in countries other than Japan; therefore, this rate may underestimate the requirement when applied to the Japanese population. The actual absorption rate estimated by the double-isotope method tends to be higher than the apparent rate. Therefore, the apparent rate was estimated based on studies that reported the results of balance tests (through which the apparent rate can be examined), and isotope procedures (through which the actual rate can be examined)<sup>(73-90)</sup>.

**Table 3. Factors for factorial modeling to determine the EAR and RDA**

Age (years)	Reference BW(kg)	(A) Body accumulation (mg/day)	(B) Urinary excretion	(C) Percutaneous loss(mg/day)	(A)+(B)+(C)	Apparent absorption rate (%)	EAR (mg/day)	RDA (mg/day)
<b>Male</b>								
1-2	11.5	99	37	6	143	40	357	428
3-5	16.5	114	49	8	171	35	489	587
6-7	22.2	99	61	10	171	35	487	585
8-9	28.0	103	73	12	188	35	538	645
10-11	35.6	134	87	15	236	40	590	708
12-14	49.0	242	111	19	372	45	826	991
15-17	59.7	151	129	21	301	45	670	804
18-29	63.2	38	135	22	195	30	648	778
30-49	68.5	0	143	24	167	30	557	668
50-69	65.3	0	138	23	161	27	596	716
70+	60.0	0	129	21	150	25	601	722
<b>Female</b>								
1-2	11.0	96	36	6	138	40	346	415
3-5	16.1	99	48	8	155	35	444	532
6-7	21.9	86	61	10	157	35	448	538
8-9	27.4	135	72	12	219	35	625	750
10-11	36.3	171	89	15	275	45	610	732
12-14	47.5	178	109	18	305	45	677	812
15-17	51.9	89	116	19	224	40	561	673
18-29	50.0	33	113	19	165	30	550	660
30-49	53.1	0	118	20	138	25	552	662
50-69	53.0	0	118	20	138	25	552	662
70+	49.5	0	112	19	131	25	524	629

**2–2–3. Adults and Children (EAR, RDA)**

The EAR and RDA were calculated using the factorial method, which considers the amount of calcium accumulated in the body, excreted by urine, and lost via dermal tissue, as well as the apparent rate. For the RDA, the interindividual difference in the requirement is unclear; however, the coefficient of variation was set as 10%.

**2–2–4. Additional Amount for Pregnant and Lactating Women (EAR, RDA)**

A newborn accrues about 28 to 30 g of calcium, most of which is obtained from the

mother and stored<sup>(91)</sup>. The intestinal calcium absorption rate in mothers doubles beginning early in pregnancy<sup>(92)</sup>. A balance study in Japanese women reported that an increased absorption ( $42 \pm 19\%$ ) was observed in the late stages of pregnancy compared to when they were not pregnant ( $23 \pm 8\%$ )<sup>(88)</sup>. As a result, calcium is transferred to the fetus. At the same time, excess calcium absorption increases the urinary excretion in the mother. Therefore, an additional intake of calcium is not required for pregnant women. The US-Canada DRIs adopted this estimation<sup>(53)</sup>. However, Hacker et al. reported that women with an insufficient calcium intake (less than 500 mg/day) may require an additional amount of calcium to meet both their demands and those of the fetus<sup>(93)</sup>.

During lactation, the intestinal calcium absorption slightly increases<sup>(88)</sup>, and the urine calcium excretion decreases<sup>(82,94)</sup>, so as to provide calcium to the breast milk. Thus, there is no requirement for additional calcium intake.

## **2–3. Method Used to Set the AI**

### **2–3–1. Infants (AI)**

The AI for infants was calculated based on the calcium concentration, and the volume of breast milk. For infants aged 0-5 months, the calcium concentration of breast milk was estimated to be 250 mg/L based on Japanese studies<sup>(9,10)</sup>. Using 0.78 L/day as the average milk intake<sup>(11,12)</sup>, the AI was determined to be 200 mg/day by rounding. Infant formula includes nutrient values that are similar to those of breast milk; however, the absorption rates are lower than in the case of breast milk<sup>(73,95)</sup>.

For infants aged over 6 months, the calculation of intake needs to consider breast milk as well as other food. Calcium intake from breast milk was calculated using the average milk intake (0.53 L/day)<sup>(9,11,15)</sup>, and the mean calcium concentration of breast milk (250 mg/L)<sup>(9,11,15)</sup>, and was estimated as 131 mg/day. The calcium intake from foods was estimated to be 128 mg/day for this age group; thus, the total calcium intake was estimated to be 261 mg/day, and the AI was set at 250 mg/day after rounding.

## **3. To Avoid Excessive Intake**

### **3–1. Method Used to Set the UL**

Excess calcium intake can cause hypercalcemia, hypercalciuria, soft tissue calcification, calculus development in the urinary system, prostate cancer, iron and zinc absorption disorders, and constipation<sup>(53)</sup>. The previous DRIs set the lowest observed adverse effect level (LOAEL) using data pertaining to milk-alkali syndrome. Patel and Goldfarb suggested that the name of the syndrome be changed to “calcium-alkali syndrome”<sup>(96)</sup>, and the US-Canada DRIs set the UL according to the results of this case report<sup>(53)</sup>. High serum calcium levels were observed at a calcium intake greater than 3,000 mg/day in a case report on calcium-alkali syndrome<sup>(53)</sup>.

From these data, the LOAEL was set at 3,000 mg/day, and the UL was set at 2,500

II Energy and Nutrients  
Minerals (1) Macrominerals  
Calcium

mg/day using an uncertainty factor of 1.2. The current dietary pattern among Japanese individuals is not considered to exceed 2,500 mg/day. However, calcium supplementation can lead to excessive intake. Bolland et al. reported that the use of calcium supplements elevated the risk of cardiovascular diseases<sup>(97,98)</sup>. Although these reports have attracted debate<sup>(99)</sup>, it should be noted that calcium intake through supplementation or calcium medication may lead to excessive intake. For those aged under 18 years, the UL was not determined due to a lack of data.

## **Magnesium**

### **1. Background Information**

#### **1–1. Definition and Classification**

Magnesium is an alkaline earth metal (atomic number: 12, Mg) that contributes to the maintenance of bone health, and various enzymatic reactions. Approximately 25 g of magnesium exists in the adult body, and is present in bones at levels of 50% to 60%<sup>(100)</sup>.

### **2. To Avoid Inadequacy**

#### **2–1. Factors to be Considered in Estimating Requirements**

The EAR was calculated on the basis of the results obtained by previous studies on magnesium balance. For infants, the AI was determined using the magnesium concentration of breast milk and average milk volume.

#### **2–2. Method Used to Set the EAR and RDA**

##### **2–2–1. Adults (EAR, RDA)**

A magnesium balance study reported 4.7 mg/kg BW/day as the amount required for the maintenance of magnesium balance in 86 Japanese participants aged 18 to 28 years<sup>(101)</sup>. However, another Japanese study reported that the amount was 4.4 mg/kg BW/day among 109 participants aged 18 to 29 years<sup>(102)</sup>. A report examining 31 Japanese participants aged 18 to 26 years from 13 studies pointed to a value of 4.18 mg/kg BW/day after adjusting the magnesium balance<sup>(103)</sup>.

In contrast, an American study suggested that 4.3 mg /kg BW/day is the amount required for the maintenance of magnesium balance<sup>(104)</sup>. A reanalysis of 243 Americans from 27 studies reported that, at an intake of 2.36 mg/kg BW/day of magnesium, the magnesium balance was 0<sup>(105)</sup>.

Compared these findings, as the studies for Japanese individuals were given priority, and 4.5 mg was set as the EAR per an adult's BW. The RDA was set after multiplying it by the reference BW, applying a factor of 1.2, and assuming a coefficient of variation of 10%.

##### **2–2–2. Children (EAR, RDA)**

A magnesium balance study examining Japanese children aged 3 to 6 years estimated the EAR to be 2.6 mg/kg BW/day, based on the observation under conditions of the consumption of a regular diet<sup>(106)</sup>. The results of an American balance test examining 12 boys and 13 girls, aged 9 to 14 years, using a stable magnesium isotope, determined the EAR to be 5 mg<sup>(74)</sup>; this value was adopted since isotope procedures are considered reasonably accurate. This value was subsequently adopted as the RDA after multiplying it by the reference BW, and applying a factor of 1.2, as had been applied to the adult EAR.

### **2–2–3. Additional Amount for Pregnant and Lactating Women (EAR and RDA)**

According to the results obtained in a magnesium balance study of pregnant women<sup>(107)</sup>, an intake of 430 mg/day of magnesium maintained the plus balance in most participants. Considering that the lean BW during pregnancy is 6-9 kg (average 7.5 kg)<sup>(108)</sup>, and the magnesium content per lean BW is 40 mg/kg BW<sup>(109)</sup>, the additional amount of magnesium required can be calculated as 31.5 mg, using 40% as the apparent absorption rate for this period<sup>(73)</sup>. Thus, the additional EAR was determined to be 30 mg. This value was subsequently adopted as the RDA after multiplying it by the reference BW and applying a factor of 1.2.

For lactating women, some studies reported that the urine magnesium concentration does not differ by the presence or absence of lactation<sup>(110,111)</sup>. Therefore, the additional amount required for lactating women was not determined.

### **2–3. Method Used to Set the AI**

#### **2–3–1. Infants (AI)**

The AI for infants aged 0-5 months was calculated using the average concentration of magnesium in breast milk (27 mg/L<sup>(9,10)</sup>), and average milk intake (0.78 L/day<sup>(11,12)</sup>), yielding a value of 20 mg/day by rounding 21.1 mg/day.

The AI for infants aged 6-11 months was calculated using the consumption of magnesium from breast milk, calculated from the concentration of magnesium in breast milk (27 mg/L<sup>(9,10)</sup>) and average milk intake (0.53 L/day<sup>(13,14)</sup>), and complementary food (46 mg/day<sup>(15)</sup>). From these values, the AI was determined to be 60 mg/day.

### **3. To Avoid Excessive Intake**

#### **3–1. Method Used to Set the UL**

The first stage of the unfavorable effects of excessive magnesium intake from sources other than food is diarrhea. Many individuals may experience mild transient diarrhea even without increased magnesium intake. Therefore, it is thought that the development of diarrhea symptoms may be the clearest index for the determination of the UL. The LOAEL was determined to be 360 mg/day based on reports from Western countries pertaining to intake through supplements<sup>(112–115)</sup>.

However, Japan-centric data were not available. As the diarrhea caused by excessive magnesium intake is not severe, and it is a reversible symptom, the uncertainty factor can be set at nearly 1. The US-Canada DRIs adopted these methods<sup>(116)</sup>. Similarly, the UL was determined to be 350 mg/day for adults, and 5 mg/day for children, for intake sources other than food.

In addition, data on unfavorable outcomes due to an excessive intake of magnesium from typical food sources were not found. Therefore, the UL for the intake of typical foods was not determined.

## Phosphorus

### 1. Background Information

#### 1–1. Definition and Classification

Phosphorus is a nitrogen family element (atomic number: 15, P), a maximum of 850 g of which is present in an adult individual, with 85% in the bones, 14% in soft tissues, and the remaining 1% in extracellular fluid.

### 2. To Avoid Inadequacy

#### 2–1. Factors to be Considered in Estimating Requirements

Phosphorus balance studies conducted among Japanese women, aged 18-28 years, reported that the requirement amounts were 18.7 mg/kg BW /day<sup>(103)</sup> and 22.58 mg/kg BW/day<sup>(102)</sup>. Based on these values, the EAR can be calculated as 946 mg/day or 1,143 mg/day, both of which are higher than the RDA value in the US-Canada DRIs (700 mg/day<sup>(116)</sup>). Another study reported that the amount required for even balance was 1,180 mg/day for men, and 970 mg/day for women, among elderly Japanese individuals (the mean age was 74.1 years for men, and 71.9 years for women)<sup>(117)</sup>, which would yield a remarkably higher EAR. In the US-Canada DRIs, the EAR was calculated using the phosphorus intake required for the maintenance of the lowest normal level of plasma phosphorus<sup>(116)</sup>. However, due to a lack of evidence in determining the presumed Japanese EAR and RDA, the AI for phosphorus was determined.

#### 2–2. Method Used to Set the AI

##### 2–2–1. Adults and Children (AI)

According to the 2010 and 2011 NHNS<sup>(3)</sup>, the median phosphorus intake was 944 mg/day. However, the actual intake may be higher, since the phosphorus content of processed foods was not added to this value. A study using a duplicate method reported that the average phosphorus intake was  $1,019 \pm 267$  mg/day<sup>(118)</sup>, which is comparable to the above-stated value.

Therefore, the AI for those aged over 1 year was adopted from the median intake from the 2010 and 2011 NHNS<sup>(3)</sup>, in relation to the US-Canada DRIs<sup>(116)</sup>. The same AI was set for adults aged over 18 years, based on the lowest intake among the age and sex groups.

##### 2–2–2. Infants (AI)

The AI for infants aged 0-5 months was calculated using the average concentration of phosphorus in breast milk (150 mg/L)<sup>(9,10)</sup> and the average milk intake (0.78 L/day)<sup>(11,12)</sup>, yielding 120 mg/day (rounding 117 mg/day).

The AI for infants aged 6 to 11 months was calculated using the average consumption of phosphorus from breast milk (80 mg/day) and complementary food (183 mg/day<sup>(15)</sup>), yielding 260 mg/day.

### **2–2–3. Pregnant and Lactating Women (AI)**

The phosphorus body storage of newborns is reported to be 17.1 g<sup>(119)</sup>. If this value is considered the additional requirement for non-pregnant women, the total dietary requirement can be calculated as 61 mg/day. However, the dietary phosphorus absorption rate is 70% during pregnancy and 60-65% when not pregnant<sup>(116)</sup>. On multiplying the AI for non-pregnant women aged 18-29 years (800 mg/day) and the above-stated values, the phosphorus absorption can be estimated at 560 mg/day for pregnant women, and 480 mg/day for non-pregnant women. As the difference (80 mg/day) is greater than the 61 mg/day above mentioned, pregnant women do not require an additional phosphorus intake.

According to the 2007-2011 NHNS<sup>(39)</sup>, the median phosphorus intake of pregnant Japanese women was 846 mg/day. As mentioned above, sufficient evidence was not available to increase the requirement, in comparison to non-pregnant women. Therefore, the AI was set at 800 mg/day, which is the same as that for non-pregnant women.

The serum phosphorus concentration has been reported to be high in lactating women, although there may be loss due to lactation<sup>(120)</sup>. Furthermore, lactating women exhibit an elevated bone absorption of phosphorus and decreased urinary excretion<sup>(116)</sup>. Therefore, pregnant women do not require an additional amount of phosphorus. The median phosphorus intake of lactating women was 979 mg/day in the 2007-2011 NHNS<sup>(39)</sup>. The AI was set at 800 mg/day, which is the same as that for non-pregnant women.

## **3. To Avoid Excessive Intake**

### **3–1. Dietary Intake**

Phosphorus is present in various foods. Although many processed foods use phosphorus as a food additive, the contribution of such phosphorus to the overall phosphorus intake is unclear since presenting information on the amount present, on food labels, is not mandatory.

### **3–2. Method Used to Set the UL**

If renal function is normal, a high intake of phosphorus may enhance the secretion of parathyroid hormone and fibroblast growth factor 23 (FGF23), which promote phosphorus excretion from the kidney and maintain blood phosphorus concentrations<sup>(121)</sup>. Therefore, the fasting serum phosphorus concentration cannot be used as an indicator of excess phosphorus intake. Postprandial serum phosphorus concentration, urinary phosphorus excretion rate, parathyroid hormone level, and FGF23 level may be indicators for the determination of the UL.

The association between phosphorus intake and parathyroid hormone has been examined<sup>(120,122–130)</sup>. A study reported that hyperparathyroidism occurred when the phosphorus intake from food additives exceeded 2,100 mg/day<sup>(122)</sup>. Additionally, an intake of 1,500-2,500 mg/day of inorganic phosphorus (phosphoric acid)<sup>(123,124)</sup> or 400-800 mg/meal of inorganic phosphorus elevated postprandial parathyroid hormone levels<sup>(125)</sup>. Excess phosphorus intake

reduces the calcium absorption in the intestine and acutely increases the postprandial serum inorganic phosphorus concentration, decreases the serum calcium ion concentration, and elevates the serum parathyroid hormone concentration<sup>(120)</sup>. A report questioned if these reactions may lead to decreased bone mass<sup>(126)</sup>. In contrast, a study reported that phosphorus intake elevated the blood parathyroid hormone concentration, dose-dependently, and bone resorption marker type 1 collagen cross-linked N-telopeptide level, and decreased the bone formation marker level (bone-type alkaline phosphatase) in women, under conditions of low calcium intake<sup>(127)</sup>. The phosphorus and calcium intake ratio may also be considered. However, few human studies have focused on this issue, and it is difficult to determine the UL using the parathyroid hormone level as an indicator.

An increasing number of studies are focusing on FGF23 as an indicator of phosphorus load<sup>(121,125,128–136)</sup>. However, the methods used for the measurement of serum FGF23 levels were different between studies. Moreover, evidence on the effect of serum FGF23 function on human health is scarce, while the association between dietary phosphorus and serum FGF23 in Japanese individuals is unclear.

Several studies have reported an association between dietary phosphorus intake and adverse health effects in body parts other than the bones<sup>(137–141)</sup>. Although it may be possible to use these data for the determination of the UL, the intake levels leading to adverse effects ranged from 1,347–3,600 mg/day across the studies. Due to data insufficiency, it was difficult to set a threshold value.

One study examined the diurnal changes in serum phosphorus concentrations according to phosphorus intake<sup>(142)</sup>. In that study, an intake of 1,500 mg/day phosphorus resulted in normal serum concentrations, while an intake of 3,000 mg/day led to a high serum phosphorus level. A Japanese study reported that an intake of 800 mg/meal (2,400 mg/day) did not lead to serum concentrations that exceeded the normal range, while 1,200 mg/meal (3,600 mg/day) exceeded the normal range<sup>(125)</sup>. There is no standard value for urinary phosphorus excretion, and data on the relationship between urinary phosphorus excretion and adverse health effects are scarce.

The UL was determined based on the relationship between dietary phosphorus intake and the elevation of serum phosphorus concentration.

**Serum inorganic phosphorus = 0.00765 × absorbed phosphorus + 0.8194 × (1-e(-0.2635 × absorbed phosphorus))**

This equation includes both inorganic phosphorus (mmol/L), and absorbed phosphorus (mmol/day)<sup>(143)</sup>. Assuming an absorption rate of 60%<sup>(120)</sup>, serum inorganic phosphorus level of 4.3 mg/dL<sup>(144)</sup> (upper limit of the normal range) and phosphorus molar weight of 30.97, the phosphorus intake can be estimated as 3,686 mg/day, which is the upper limit of the normal range of the serum inorganic phosphorus level. This value was used as the LOAEL. Using an uncertainty factor of 1.2, the UL for adults was set at 3,000 mg/day by rounding 3,072 mg/day.

For children, the UL was not determined due to a lack of data.

**DRIs for Sodium (mg/day. Values in parentheses are salt equivalent [g/day])**

Gender	Males			Females		
Age etc.	EAR	AI	DG	EAR	AI	DG
0-5 months	—	100(0.3)	—	—	100(0.3)	—
6-11 months	—	600(1.5)	—	—	600(1.5)	—
1-2 years	—	—	(<3.0)	—	—	(<3.5)
3-5 years	—	—	(<4.0)	—	—	(<4.5)
6-7 years	—	—	(<5.0)	—	—	(<5.5)
8-9 years	—	—	(<5.5)	—	—	(<6.0)
10-11 years	—	—	(<6.5)	—	—	(<7.0)
12-14 years	—	—	(<8.0)	—	—	(<7.0)
15-17 years	—	—	(<8.0)	—	—	(<7.0)
18-29 years	600(1.5)	—	(<8.0)	600(1.5)	—	(<7.0)
30-49 years	600(1.5)	—	(<8.0)	600(1.5)	—	(<7.0)
50-69 years	600(1.5)	—	(<8.0)	600(1.5)	—	(<7.0)
70+ years	600(1.5)	—	(<8.0)	600(1.5)	—	(<7.0)
Pregnant women	/			—	—	—
Lactating women				—	—	—

**DRIs for Potassium (mg/day)**

Gender	Males		Females	
Age etc.	AI	DG	AI	DG
0-5 months	400	—	400	—
6-11 months	700	—	700	—
1-2 years	900	—	800	—
3-5 years	1,100	—	1,000	—
6-7 years	1,300	≥1,800	1,200	≥1,800
8-9 years	1,600	≥2,000	1,500	≥2,000
10-11 years	1,900	≥2,200	1,800	≥2,000
12-14 years	2,400	≥2,600	2,200	≥2,400
15-17 years	2,800	≥3,000	2,100	≥2,600
18-29 years	2,500	≥3,000	2,000	≥2,600
30-49 years	2,500	≥3,000	2,000	≥2,600
50-69 years	2,500	≥3,000	2,000	≥2,600
70+ years	2,500	≥3,000	2,000	≥2,600
Pregnant women	/		2,000	—
Lactating women			2,200	—

II Energy and Nutrients  
 Minerals (1) Macrominerals

**DRIs for Calcium (mg/day)**

Gender	Males				Females			
Age etc.	EAR	RDA	AI	UL	EAR	RDA	AI	UL
0-5 months	—	—	200	—	—	—	200	—
6-11 months	—	—	250	—	—	—	250	—
1-2 years	350	450	—	—	350	400	—	—
3-5 years	500	600	—	—	450	550	—	—
6-7 years	500	600	—	—	450	550	—	—
8-9 years	550	650	—	—	600	750	—	—
10-11 years	600	700	—	—	600	750	—	—
12-14 years	850	1,000	—	—	700	800	—	—
15-17 years	650	800	—	—	550	650	—	—
18-29 years	650	800	—	2,500	550	650	—	2,500
30-49 years	550	650	—	2,500	550	650	—	2,500
50-69 years	600	700	—	2,500	550	650	—	2,500
70+ years	600	700	—	2,500	550	650	—	2,500
Pregnant women					—	—	—	—
Lactating women					—	—	—	—

**DRIs for Magnesium (mg/day)**

Gender Age etc.	Males				Females			
	EAR	RDA	AI	UL <sup>1</sup>	EAR	RDA	AI	UL <sup>1</sup>
0-5 months	—	—	20	—	—	—	20	—
6-11 months	—	—	60	—	—	—	60	—
1-2 years	60	70	—	—	60	70	—	—
3-5 years	80	100	—	—	80	100	—	—
6-7 years	110	130	—	—	110	130	—	—
8-9 years	140	170	—	—	140	160	—	—
10-11 years	180	210	—	—	180	220	—	—
12-14 years	250	290	—	—	240	290	—	—
15-17 years	300	360	—	—	260	310	—	—
18-29 years	280	340	—	—	230	270	—	—
30-49 years	310	370	—	—	240	290	—	—
50-69 years	290	350	—	—	240	290	—	—
70+ years	270	320	—	—	220	270	—	—
Pregnant women (additional)	/				+30	+40	—	—
Lactating women (additional)					—	—	—	—

<sup>1</sup> No UL is developed for dietary intake from normal food. For dietary intake from sources other than normal food, ULs of 350 mg/day and 5 mg/kg body weight/day are set for adults and children, respectively.

II Energy and Nutrients  
 Minerals (1) Macrominerals

**DRIs for Phosphorus (mg/day)**

Gender	Males		Females	
Age etc.	AI	UL	AI	UL
0-5 months	120	—	120	—
6-11 months	260	—	260	—
1-2 years	500	—	500	—
3-5 years	800	—	600	—
6-7 years	900	—	900	—
8-9 years	1,000	—	900	—
10-11 years	1,100	—	1,000	—
12-14 years	1,200	—	1,100	—
15-17 years	1,200	—	900	—
18-29 years	1,000	3,000	800	3,000
30-49 years	1,000	3,000	800	3,000
50-69 years	1,000	3,000	800	3,000
70+ years	1,000	3,000	800	3,000
Pregnant women			800	—
Lactating women			800	—

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## **(2) Microminerals**

### **Iron**

#### **1. Background Information**

##### **1–1. Definition and Classification**

Iron is a transition metal element (atomic number: 26, Fe). It is predominantly stored as heme iron which is found in combination with protein, as well as non-heme iron: which is inorganic in food.

#### **2. To Avoid Inadequacy**

##### **2–1. Method Used to Set the Estimated Average Requirements (EAR) and Recommended Dietary Allowances (RDA)**

The EAR for iron can be calculated using balance tests and factorial modeling methods, except in the case of infants aged 0-5 months. However, the iron balance in the body can be maintained at low iron intakes, and, as the rate of iron absorption changes depending on dietary intake, the requirement may be underestimated when the results of balance tests are used. Therefore, the EAR was determined using a factorial modeling method. Although a number of studies used factorial modeling methods, few of those studies were conducted in Japanese settings. For those aged over 6 months, the principal calculation methods are based on the US-Canada DRIs<sup>(1)</sup>, using body weight (BW) and menstrual blood loss values from Japanese studies. For infants, the adequate intake (AI) was determined using the iron concentration in breast milk, and the average milk volume (0.78 L/day)<sup>(2,3)</sup>.

##### **2–1–1. Factors Used in the Factorial Modeling Method**

###### **2–1–1–1. Basal Iron Loss**

The basal iron loss measured in 41 people from 4 groups (mean BW 68.6 kg) was 0.9-1.0 mg/ day (mean 0.96 mg/day), and the difference between the groups was relatively small<sup>(4)</sup>; these findings are similar to those of a recent study<sup>(5)</sup>. This value was extrapolated to each sex and age group, using the 0.75<sup>th</sup> power of the BW ratio (Table 1).

**Table 1. Estimation of basal iron loss**

Age	Male				Female			
	Intermediate age (year)	Reference BW (kg)	BW increase (kg/year) <sup>1</sup>	Basal iron loss (mg/day) <sup>2</sup>	Intermediate age (year)	Reference BW (kg)	BW increase (kg/year) <sup>1</sup>	Basal iron loss (mg/day) <sup>2</sup>
6-11 (months)	0.75	8.8	3.6	0.21	0.75	8.1	3.4	0.19
1-2 (years)	2.0	11.5	2.1	0.25	2.0	11.0	2.2	0.24
3-5 (years)	4.5	16.5	2.1	0.33	4.5	16.1	2.2	0.32
6-7 (years)	7.0	22.2	2.6	0.41	7.0	21.9	2.5	0.41
8-9 (years)	9.0	28.0	3.4	0.49	9.0	27.4	3.6	0.48
10-11 (years)	11.0	35.6	4.6	0.59	11.0	36.3	4.5	0.60
12-14 (years)	13.5	49.0	4.5	0.75	13.5	47.5	3.0	0.73
15-17 (years)	16.5	59.7	2.0	0.86	16.5	51.9	0.6	0.78
18-29 (years)	24.0	63.2	0.4	0.90	24.0	50.0	0.0	0.76
30-49 (years)	40.0	68.5	0.1	0.96	40.0	53.1	0.1	0.79
50-69 (years)	60.0	65.3	-	0.93	60.0	53.0	-	0.79
70 years+	-	60.0	-	0.87	-	49.5	-	0.75

<sup>1</sup> Calculated using values of the reference BW

<sup>2</sup> Extrapolated using a value of basal iron loss (0.96mg/day, BW 68.6kg) and the 0.75th power of the BW ratio

## 2-1-1-2. Iron Storage for Growth

Iron is stored according to growth requirements during childhood. The iron storage with growth respect to growth requirements can be stratified into a) iron content in hemoglobin, b) increase in tissue iron (non-storage iron), and c) increase in storage iron.

### (1) Iron storage in hemoglobin

The iron storage with growth for each sex and age group was estimated using the following formulas that were employed in the US-Canada DRIs<sup>(1)</sup>.

**【6-11 months old】** Iron storage in hemoglobin (mg/day) = BW increase (kg/year) × blood volume [70 mL/kg] × hemoglobin concentration [0.12 g/mL] × iron content in hemoglobin [3.39 mg/g] ÷ 365 days

**【1-9 years old】** Iron storage in hemoglobin (mg/day) = (blood hemoglobin level of one level upper age class (g) – blood hemoglobin level at the target age class (g)) × iron content in hemoglobin [3.39 mg/g] ÷ (the middle age of one level upper age class – the middle age of the target age class) ÷ 365 days

**【10-17 years old】** Iron storage in hemoglobin (mg/day) = (reference BW (kg) × increase in hemoglobin concentration (g/L/year) + BW increase (kg/year) × hemoglobin concentration (g/L)) × blood volume [0.075 L/kg] × iron content in hemoglobin [3.39 mg/g] ÷ 365 days

For those aged 1-9 years, the blood volume for each age and sex group was calculated using a regression equation estimated employing the values for those aged 1-11 years<sup>(6)</sup>: (boys:  $0.0753 \times \text{BW (kg)} - 0.05$ , girls:  $0.0753 \times \text{BW (kg)} + 0.01$ ). Blood hemoglobin concentrations were estimated using a regression equation with age and hemoglobin concentration, as

determined in a Canadian study<sup>(7)</sup>. The iron content in hemoglobin was determined as 3.39 mg/g<sup>(8)</sup>.

(2) Increase in tissue iron (non-storage iron) level

The increase in the tissue iron (non-storage iron) level was calculated using the following formula:

$$\text{Tissue iron per BW (0.7 mg/kg)} \times \text{increase in BW (kg/year)} \div 365 \text{ days}$$

(3) Increase in storage iron level

The increase in the storage iron level was reported to be 12% of the total iron storage in children aged 1-2 years<sup>(9)</sup>. Considering this, for children aged 6 months to 2 years, the increase in the storage iron level was estimated to be 12% of the increase in the total iron storage for growth, including the factors mentioned above ((1) and (2)). This percentage was assumed to decrease linearly after age 3 years, and reach 0 at 9 years of age<sup>(9)</sup>. Table 2 shows the increase in the storage iron for each age and sex group.

**Table 2. Estimation of the increase in the storage iron (6 months to 17 years old)**

Gender	Age	Blood volume (L) <sup>1</sup>	Hemoglobin concentration (g/L) <sup>2</sup>	Increase in hemoglobin concentration (g/L/year) <sup>2</sup>	Blood hemoglobin (g) <sup>3</sup>	Iron storage in hemoglobin (mg/day) <sup>4</sup>	Non-storage iron increase (mg/day) <sup>5</sup>	Storage iron increase (mg/day) <sup>6</sup>	Total iron storage (mg/day)
Male	6-11 (months)	-	-	-	-	0.28	0.01	0.04	0.33
	1-2 (years)	0.82	121.8	-	99.4	0.19	0.00	0.02	0.21
	3-5 (years)	1.19	125.3	-	149.4	0.22	0.00	0.02	0.24
	6-7 (years)	1.62	128.8	-	208.9	0.29	0.00	0.01	0.30
	8-9 (years)	2.06	131.6	-	270.9	0.38	0.01	0.00	0.39
	10-11 (years)	2.63	134.4	1.40	353.6	0.46	0.01	-	0.47
	12-14 (years)	-	137.9	1.40	-	0.48	0.01	-	0.49
	15-17 (years)	-	150.4	3.40	-	0.35	0.00	-	0.36
Female	6-11 (months)	-	-	-	-	0.26	0.01	0.04	0.31
	1-2 (years)	0.84	123.2	-	103.3	0.19	0.00	0.03	0.22
	3-5 (years)	1.22	126.0	-	154.0	0.22	0.00	0.02	0.25
	6-7 (years)	1.66	128.7	-	213.5	0.27	0.00	0.01	0.28
	8-9 (years)	2.07	130.9	-	271.4	0.44	0.01	0.00	0.44
	10-11 (years)	2.74	133.1	1.10	365.1	0.44	0.01	-	0.45
	12-14 (years)	-	135.9	1.10	-	0.32	0.01	-	0.32
	15-17 (years)	-	135.6	0.28	-	0.07	0.00	-	0.07

<sup>1</sup> Estimated using regression formulas (male: 0.0753×BW-0.05, female: 0.0753×BW+0.01), according to the report by Hawkins<sup>(6)</sup>

<sup>2</sup> Estimated using a formula of association between age and hemoglobin concentration<sup>(7)</sup>

<sup>3</sup> Blood hemoglobin (g) = blood volume (L) × hemoglobin concentration (g/L)

<sup>4</sup> 6-11 months old: Iron storage in hemoglobin (mg/day) = BW increase (kg/year) × blood volume [70 mL/kg] × hemoglobin concentration [0.12 g/mL] × iron content in hemoglobin [3.39 mg/g<sup>(8)</sup>] / 365 days

1-9 years old: Iron storage in hemoglobin (mg/day) = (blood hemoglobin level of one level upper age class (g) – blood

hemoglobin level at the target age class (g) × iron content in hemoglobin [3.39 mg/g] / (the middle age of one level upper age class – the middle age of the target age class) / 365 days

10-17 years old: Iron storage in hemoglobin (mg/day) = (reference BW (kg) × increase in hemoglobin concentration (g/L/year) + BW increase (kg/year) × hemoglobin concentration (g/L)) × blood volume [0.075 L/kg] × iron content in hemoglobin [3.39 mg/g] / 365 days

<sup>5</sup> Non-storage iron increase (mg/day) = BW increase (kg/year) × storage iron per BW [0.7 mg/kg] / 365 day

<sup>6</sup> Estimated as 12% of total iron storage for 6 months to 2 years old, with decreasing after 3 years old to 0 at 9 years old<sup>(9)</sup>.

### 2–1–1–3. Menstrual Iron Loss

Menstrual iron loss is strongly associated with iron deficiency anemia<sup>(10)</sup>. According to a review of several studies that examined Japanese women aged around 20 years, the geometric mean menstrual blood loss was 37.0 mL/period, and the median duration of the menstrual cycle was 31 days<sup>(11)</sup>; this finding was supported by a recent study<sup>(12)</sup>. However, age-related variations in menstrual blood loss have not been reported in Japanese women aged over 20 years. A Japanese study of high school female students reported that the geometric mean menstrual blood loss was 31.1 mL/period, and the median duration of the menstrual cycle was 31 days<sup>(13)</sup>. From these results, the menstrual blood loss was determined to be 37 mL/period for those aged over 18 years and 31.1 mL/period for those aged 10-17 years, and the duration of the menstrual cycle was determined to be 31 days for all age groups, for the calculation of menstrual iron loss. Using a hemoglobin concentration of 135 g/L<sup>(14)</sup>, and hemoglobin iron content of 3.39 mg/g for all age groups, the requirement values for the compensation of the menstrual loss were estimated to be 3.06 mg/day for those aged 10-17 years, and 3.64 mg/day for those aged 18 years or older.

**Table 3. Estimation of dietary iron requirement for compensation of the menstrual loss (women)**

Subjects	Menstrual blood volume (mL/times)	Menstrual cycle (day)	Iron loss (mg/day) <sup>1</sup>	Required intake to compensate the iron loss(mg/day) <sup>2</sup>
10-17(years)	31.1	31	0.46	3.06
18 years +	37.0	31	0.55	3.64

<sup>1</sup> Iron loss (mg/day) = menstrual blood volume (mL)/Japanese menstrual cycle [31 days]<sup>(13)</sup> × hemoglobin concentration [0.135g/mL]<sup>(14)</sup> × iron concentration in hemoglobin [3.39mg/g]

<sup>2</sup> Iron intake (mg/day) = iron loss (mg/day) / absorption rate [0.15]

Adult menstrual blood loss displays an almost-log-normal distribution. A study reported that the 95<sup>th</sup> percentile value of the loss was 115 mL/period for women without iron deficiency anemia<sup>(15)</sup>, while another study reported that 85% of women lose less than 120 mL/period<sup>(16)</sup>. Although these values are much higher than those used in the definition of hypermenorrhea (>80 mL/period)<sup>(17)</sup>, few relevant studies have been conducted in Japanese populations. Therefore, the EAR and RDA were determined for people without hypermenorrhea, whose menstrual blood loss is less than 80 mL/period. When excluding those with hypermenorrhea, the distribution of adult menstrual blood loss was relatively close to normal, and the mean value of menstrual blood loss could be estimated to be lower than when

hypermenorrhea is included. However, since these data remained unclear, the DRIs used the geometric mean menstrual blood loss including hypermenorrhea (over 20 years old: 37.0 mL/period, 10-17 years old: 31.1 mL/period) for the calculation of the EAR and RDA.

#### **2-1-1-4. Dietary Iron Absorption Rate**

The dietary iron absorption rate is reported to be 16.6% from the normal American diet, 16% from the normal French diet, and 14% from the normal Swedish diet<sup>(14)</sup>. The rate varies based on multiple factors such as the dietary composition ratio of heme iron and non-heme iron, the intake of nutrients and food that promote or inhibit iron absorption, and the need for iron, making it difficult to determine a representative value for the dietary iron absorption rate. In the present DRIs, the dietary iron absorption rate was estimated to be 15% for all age and sex groups, except for infants, in accordance with a value adopted by the WHO and the Food and Agricultural Organization (FAO)<sup>(17)</sup> (15%).

The absorption rate of iron, especially inorganic iron, increases when there is a need for iron. Among Japanese people, inorganic iron intake comprises a majority of the dietary iron intake, due to their high intake of plant foods. Therefore, the absorption rate from the Japanese diet may be higher than 15%; however, this elevated absorption may be derived from low iron intake. Therefore, the iron absorption rate was determined to be 15%, as its use was considered appropriate under conditions of sufficient intake.

#### **2-1-1-5. Interindividual Difference of Requirement**

In the US-Canada DRIs<sup>(1)</sup>, the coefficient of variation for interindividual requirement was determined to be 40% for children aged 8 years or younger, 20% for those aged 11 years, and 10% for those aged 16 years, based on the variance in the increase in body surface area and BW. Few relevant reports have focused on young children; therefore, the coefficient of variation for iron was determined to be 20% for children aged 6 months-14 years, and 10% for those aged over 15 years.

#### **2-1-2. Adults (EAR, RDA)**

##### **2-1-2-1. Men and Non-menstruating Women**

The EAR was calculated as follows:  $EAR = \text{basal iron loss (Table 1)} \div \text{absorption rate (0.15)}$ . The RDA was determined as the  $EAR \times 1.2$ , using 10% as the coefficient of variation.

##### **2-1-2-2. Menstruating Women**

The EAR was calculated as follows:  $EAR = [\text{basal iron loss (Table 1)} + \text{menstrual iron loss (0.55 mg/day (Table 3))}] \div \text{absorption rate (0.15)}$ . The RDA was determined as the  $EAR \times 1.2$ , using 10% as the coefficient of variation. These values were set for those without hypermenorrhea (>80 mL/period). For those with hypermenorrhea, the EAR and RDA were estimated to be more than 13 mg/day and 16 mg/day, respectively. It is difficult to achieve the

dietary iron intakes reported in the National Health and Nutrition Survey through typical foods, and iron supplementation under medical supervision is required.

### **2-1-3. Children (EAR, RDA)**

#### **2-1-3-1. Boys and Non-Menstruating Girls**

The EAR was calculated as follows:  $EAR = [\text{basal iron loss (Table 1)} + \text{iron storage in hemoglobin (Table 2)} + \text{increase in tissue iron (non-storage iron) level (Table 2)} + \text{increase in storage iron level (Table 2)}] \div \text{absorption rate (0.15)}$ . The RDA was determined as the  $EAR \times 1.4$  for those aged 1-14 years, and  $EAR \times 1.2$  for those aged 15 years or older.

#### **2-1-3-2. Menstruating Girls**

For girls aged 10 years or older, the EAR was calculated considering the menstrual iron loss as follows:  $EAR = [\text{basal iron loss (Table 1)} + \text{iron storage in hemoglobin (Table 2)} + \text{increase in tissue iron (non-storage iron) level (Table 2)} + \text{increase in storage iron level (Table 2)} + \text{menstrual iron loss (0.46 mg/day) (Table 3)}] \div \text{absorption rate (0.15)}$ . The RDA was determined as the  $EAR \times 1.4$  for those aged 1-14 years, and  $EAR \times 1.2$  for those aged 15 years or older. These values were set for those without hypermenorrhea (>80 mL/period).

### **2-1-4. Infants**

#### **2-1-4-1. 0-5 months old (AI)**

Fetal hemoglobin is degraded after birth, and iron is released; thereafter, the adult hemoglobin begins its biosynthesis. Accordingly, the blood hemoglobin concentration reaches a minimum level during the 4-6 months after birth, and increases thereafter. As full-term newborns with normal intrauterine growth, weighing more than 3 kg, can maintain normal iron metabolism by utilizing the body iron storage during the first 4 months of life, iron deficiency anemia develops more frequently during the later stages of infancy (such as the weaning period)<sup>(18)</sup>. The AI was estimated by multiplying the iron concentration in the breast milk of Japanese women (0.426 mg/L)<sup>(19)</sup>, and the average milk intake (0.78 L/day)<sup>(2,3)</sup>, as the iron intake from breast milk was considered sufficient for infants aged 0-5 months. The AI was determined as 9.5 mg /day by rounding 0.332 mg/day.

Although there is no apparent difference between the growth of breast milk-fed infants and non-breast milk-fed infants, breast milk-fed infants were reported to have a lower hemoglobin concentration and anemic tendency<sup>(20)</sup>. Iron supplementation using infant formula needs to be considered when needed, among exclusively breast milk-fed infants with iron deficiency anemia, as breast milk may not provide sufficient iron.

#### **2-1-4-2. 6-11 months old (EAR, RDA)**

Lower hemoglobin concentrations have been reported among 6-month-old breast milk-fed Japanese infants. Therefore, the value extrapolated from the AI for children aged 0-5 months

can be lower for the prevention of iron deficiency. Therefore, the EAR for children aged 6-11 months was determined using the same calculation as that used in other children as follows:  $\text{EAR} = [\text{basal iron loss (Table 1)} + \text{iron storage in hemoglobin (Table 2)} + \text{increase in tissue iron (non-storage iron) level (Table 2)} + \text{increase in storage iron level (Table 2)}] \div \text{absorption rate (0.15)}$ . The RDA was determined as the  $\text{EAR} \times 1.4$ , using 20% as the coefficient of variation.

### **2-1-5. Additional Amount for Pregnant Women (EAR, RDA)**

In addition to basal iron loss, fetal iron storage, placental iron storage, and an increase in the hemoglobin mass caused by erythrocyte mass expansion are required during pregnancy. Each of the above-stated factors varies by the pregnancy stage.

For the DRIs, the values shown in Table 4 were used for fetal and placental iron storage<sup>(21)</sup>. The values for the increase in the hemoglobin mass caused by erythrocyte mass expansion were calculated based on the reference weight (age 18-29 years, 50.6 kg), blood volume (0.075 L/kg BW), increase in blood volume during pregnancy (30-50%), hemoglobin concentration standard for pregnant women (110 g/L: criteria for pregnancy anemia), hemoglobin concentration for adult women (135 g/L)<sup>(18)</sup>, and iron content in hemoglobin (3.39 mg/g)<sup>(8)</sup>. Using a BW of 50.6 kg for non-pregnant women, the hemoglobin iron content was estimated to be 1,737 mg ( $50.6 \times 0.075 \times 135 \times 3.39 = 1,737$  mg), and the minimum estimate for the iron content in hemoglobin at the time of delivery, in the absence of pregnancy anemia, was estimated to be 1,840-2,123 mg ( $50.6 \times 0.075 \times 1.3 - 1.5 \times 110 \times 3.39 = 1,840-2,123$  mg). The difference between these values was 103-386 mg; therefore, the total usage of iron throughout pregnancy was estimated to be approximately 300 mg. Furthermore, this demand expands greatly during the second and third trimesters, with an equal division between the terms.

With these estimates, the additional iron requirements for pregnant women were calculated to be 0.32 mg/day, 2.68 mg/day and 3.64 mg/day, in the early, mid, and late stages, respectively. Using a dietary iron absorption rate of 15% in the early stage, and 25% in the second and late stages, the dietary iron requirements were calculated to be 2.1 mg/day, 10.7 mg/day, and 14.6 mg/day for early-, mid- and late-stage pregnancies, respectively. The average value for the mid and late stages was calculated, and this was adopted for the EAR for these two terms. Thus, the additional EAR was determined to be 2.0 mg/day for the early stage, and 12.5 mg/day for the mid and late stages, after rounding. The additional RDA was determined as the  $\text{EAR} \times 1.2$ , using 10% as the coefficient of variation. The RDAs were determined to be 2.5 mg/day and 15.0 mg/day for the mid and late stages of pregnancy. Table 4 translates this calculation, and the values presented are the additional amounts required for the determination of the EAR and RDA of non-pregnant women.

**Table 4. Additional amount during pregnancy: factors used to estimate the EAR and RDA**

	Fetal iron storage (mg/stage) <sup>1</sup>	Placental iron storage (mg/stage) <sup>1</sup>	Required iron for blood increase (mg/stage) <sup>2</sup>	Total (mg/stage)	Total iron requirement throughout pregnancy (mg/day) <sup>3</sup>	Absorption Rate <sup>4</sup>	additional EAR (mg/day) <sup>5</sup>	additional RDA (mg/day) <sup>6</sup>
Early stage	25	5	0	30	0.32	0.15	2.1	2.6
Mid stage	75	25	150	250	0.25	0.25	10.7	12.9
Late stage	145	45	150	340	0.25	0.25	14.6	17.5

<sup>1</sup> According to the report by Bothwell, et al.<sup>(21)</sup>

<sup>2</sup> Calculated based on the reference BW (50.6kg), blood volume per BW (0.075L/kg), blood volume increase during pregnancy (30-50%), hemoglobin concentration standard during pregnancy (11g/dL), hemoglobin concentration in non-pregnant women (135g/L)<sup>(14)</sup>, and iron content in hemoglobin (3.39mg/g)<sup>(8)</sup>.

<sup>3</sup> Total (mg/stage) / (280day)

<sup>4</sup> For early stage, the value for non-pregnant women was used. For mid and late stage, the values were based on the report by Barrett et al.<sup>(22)</sup>

<sup>5</sup> Total iron requirement (mg/day) / absorption rate

<sup>6</sup> Using 10% as the coefficient of variation

Among pregnant Japanese women, the prevalence of iron deficiency anemia (22.9%) was reported to be slightly higher than that among non-pregnant women (15.7%), in spite of their dietary iron intakes being similar<sup>(23)</sup>. This indicates that there is a clear gap between dietary iron intake and iron deficiency anemia among pregnant women. This could be attributed to the fact that iron absorption significantly increases during pregnancy, as the demand increases. A balance study reported that dietary absorption rate was 39% among Japanese women in their 18<sup>th</sup>, 27<sup>th</sup> and 34<sup>th</sup> weeks of pregnancy<sup>(24)</sup>. Using a dietary iron absorption rate of 40% during the mid and late stages, the EAR was estimated to be 6.7 mg/day for the mid stage, and 9.1 mg/day for the late stage. The RDA was estimated at 8.0 mg/day for the mid stage and 10.9 mg/day for the late stage. While these values may be more realistic, they were not established as the EAR and RDA for the DRIs, due to a lack of sufficient evidence.

## **2—1—6. Additional Amounts for Lactating Women (EAR, RDA)**

The average iron requirement for lactation was calculated to be 2.2 mg/day, based on the iron concentration in the breast milk of Japanese mothers (0.426 mg/L)<sup>(25)</sup>, average milk volume (0.78 L/day)<sup>(2,3)</sup>, and absorption rate (15%):  $0.426 \times 0.78 \div 0.15$ . Rounding this value, the additional EAR for lactation was determined to be 2.0 mg/day. The additional RDA was determined as the EAR  $\times$  1.2, yielding a value of 2.5 mg/day by rounding 2.7 mg/day. These values were the additional EAR and RDA of non-pregnant women without menstruation. At the time of delivery, the actual loss of iron through blood (mean  $\pm$  standard deviation) is 328 $\pm$ 236 mL in primiparous women and 279 $\pm$ 235 mL in multiparous women<sup>(26)</sup>. This amount is clearly lower than the increase in the blood circulation during pregnancy, implying that the

iron loss at delivery can be ignored for the establishment of the DRIs for lactating women. Indeed, the prevalence of iron deficiency anemia is lower among lactating women than non-pregnant or non-lactating women<sup>(23)</sup>.

### **3. To Avoid Excessive Intake**

The consumption of a regular diet does not lead to excessive iron intake; however, the inappropriate use of supplemental foods, iron-fortified foods, or medicinal iron for the treatment of anemia can lead to an overdose.

#### **3–1. Method Used to Set the Tolerable Upper Intake level (UL)**

##### **3–1–1. Adult and Children (UL)**

In a double-blind trial involving the administration of 60 mg/day of non-heme iron (fumarate iron), 18 mg/day of a mix of heme and non-heme iron (2 mg/day iron of pig blood-origin heme iron + 16 mg/day of non-heme iron) and a placebo, participants in the non-heme iron group more frequently reported symptoms such as constipation or other gastrointestinal effects<sup>(27)</sup>. Inorganic iron supplement intake can also cause unidentified complaints, including gastric distress even in small doses of 2 mg/day or 10 mg/day as iron<sup>(28,29)</sup>. In contrast, heme iron supplementation at 30 mg/day as iron, for 2 months, was not associated with gastric distress or changes in blood laboratory data<sup>(30)</sup>.

For adults, chronic siderosis is a severe effect of long-term excess iron intake. A regular intake of beer containing a large amount of iron, or iron consumption due to the use of iron pans can cause Bantu siderosis. This has been estimated to occur at iron intakes greater than approximately 100 mg/day<sup>(31)</sup>.

The FAO/WHO set the provisional maximal tolerable intake for iron at 0.8 mg/kg BW, excluding the use of iron oxide colorants, iron supplements for pregnancy and lactation, and medicinal iron<sup>(32)</sup>. Taking this into consideration, the UL for adults aged 15 years or older was calculated using this value and the reference BWs for each age and sex category.

One study reported that a lower BW increase was observed among children aged 12-18 months after supplementation with 3 mg/kg/day of ferrous sulfate<sup>(33)</sup>. According to the Food and Drug Administration (FDA), the most common causes of acute toxicity were accidental overdoses of medicinal iron or iron supplements, in children around 6 years of age; therefore, a limit of 60 mg/kg BW/day was set<sup>(34)</sup>. This value was used to derive the UL for children aged 1-2 years; this was set as the lowest observed adverse effect level (LOAEL). Thus, the UL was calculated to be 2 mg/kg/day, using an uncertainty factor of 30. This uncertainty factor was obtained by multiplying 10 (to account for the extrapolation from the LOAEL to the UL) and 3 (for the protection of susceptible individuals). Among children aged 3-14 years, the UL was determined 1.6 mg/kg/day for those aged 3-5 years 1.4 mg/kg/day for those aged 6-7 years, 1.2 mg/kg/day for those aged 8-9 years, and 1.0 mg/kg/day for those aged 10-14 years.

### **3–1–2. Infants (UL)**

A randomized controlled trial reported poorer growth, in terms of height and head circumference, in infants receiving iron supplementation (1 mg/kg iron) with a normal iron status (hemoglobin concentration >11 g/dL and serum ferritin concentration >50 µg/L)<sup>(35)</sup>. Furthermore, in this study, the odds ratio (OR) for diarrhea was 2.4 in the iron supplementation group, compared to the placebo group, among infants with a hemoglobin concentration > 11 g/dL; the OR was 0.21 among infants with a hemoglobin concentration <11 g/dL. This iron supplementation is equivalent to approximately 7 mg/day for Japanese infants. In contrast, no adverse gastrointestinal effects were reported when 1-month-old infants were supplemented with 5 mg/day of non-heme iron for up to 1 year or 30 mg/day for up to 18 months<sup>(36)</sup>, and when 3-month-old infants were supplemented with 10 mg/day of non-heme iron for up to 21 months<sup>(37)</sup>. Similarly, no significant adverse gastrointestinal effects were reported when children aged 11-14 months were supplemented with 3 mg/kg BW/day (approximately 30 mg/day) of non-heme iron for 3 months<sup>(38)</sup>. From these controversial findings for infants, it was difficult to set an LOAEL or NOAEL (no observed adverse effect level), and, consequently, no UL was established.

### **3–1–3. Pregnant and Lactating Women (UL)**

Supplementation with 60 mg of fumarate iron suppressed zinc absorption in 5 lactating women<sup>(39)</sup>. Similarly, stable zinc absorption was observed among 4 lactating women who received 120 mg/day of iron supplementation during pregnancy, and 76 mg/day of iron during lactation (zinc absorption usually rises during pregnancy)<sup>(40)</sup>. However, a decreased serum zinc concentration was reported when 18 mg/day of iron supplements was administered to young pregnant women aged under 20 years; however, their iron status was improved<sup>(41)</sup>. Although several reports have focused on zinc absorption in relation to iron supplementation, data were limited for the establishment of a UL for pregnant or lactating women.

## Zinc

### 1. Background Information

#### 1–1. Definition and Classification

Zinc is an element belonging to the zinc group (atomic number: 30, Zn). Approximately 2,000 mg of zinc is stored in the body<sup>(42)</sup>, and this is predominantly distributed in the skeletal muscles, bones, skin, liver, brain and kidneys<sup>(43)</sup>.

### 2. To Avoid Inadequacy

#### 2–1. Method Used to Set the EAR and RDA

##### 2–1–1. Adults (EAR, RDA)

No study has focused on zinc metabolism in the Japanese population. Therefore, the EARs for adults were calculated using the values from the US-Canada DRIs<sup>(44)</sup>. The calculation methods used were as follows: 1) Estimate the nonintestinal losses of endogenous zinc (losses via urine, body surface and semen/menstrual blood), 2) Define the relationships between intestinal endogenous excretion (amount that moved from the tissues to feces via the intestine) and absorbed zinc, 3) Calculate the minimum quantity of absorbed zinc necessary to offset endogenous zinc losses, and 4) Calculate the dietary zinc intake corresponding to the average minimum quantity of absorbed zinc.

Considering the results pertaining to endogenous zinc excretion via the intestine, among 18-40-year-old men whose zinc intakes were lower than 20 mg/day, in the United Kingdom and US<sup>(45–51)</sup>, the following equation was estimated:

Endogenous excretion via the intestine =  $0.628 \times \text{quantity absorbed} + 0.2784 \text{ mg/day}$

As total endogenous zinc excretion is the sum of endogenous excretion via the intestine and other routes, the total endogenous excretion can be calculated as follows:

Total endogenous excretion =  $0.628 \times \text{quantity absorbed} + 0.2784 + (\text{urinary loss} + \text{integumental loss} + \text{loss through semen or menstrual blood})$

According to a US balance study of 11 men (mean BW: 75.5 kg), the urinary loss, integumental loss, and loss through semen were 512, 525 and 111  $\mu\text{g/day}$ , respectively<sup>(52)</sup>. Using the 0.75<sup>th</sup> power of the BW ratio and the reference BW for those aged 18-29 years, the total endogenous excretion was extrapolated as follows:

**Men: Total endogenous excretion =  $0.628 \times \text{quantity absorbed} + 0.2784 + (0.448 + 0.460 + 0.097)$  (mg/day)**

**Women: Total endogenous excretion =  $0.628 \times \text{quantity absorbed} + 0.2784 + (0.376 + 0.386 + 0.082)$  (mg/day)**

Therefore, the minimal intake necessary to maintain zinc balance among adults aged 18-29 years, with a reference BW, was calculated to be 3,450 mg/day for men and 3,015 mg/day for women.

However, the relationship between zinc absorption and zinc intake is expressed by the

following equation<sup>(45-51)</sup>: quantity of absorbed zinc =  $1.113 \times \text{zinc intake}^{0.5462}$ . Therefore, the quantities of absorbed zinc were calculated to be 7,936 mg/day and 6,199 mg/day, respectively. Using these values as reference for the EAR, the EAR for each age group was determined through extrapolation, using the 0.75<sup>th</sup> power of the BW ratio.

The RDA was determined as the EAR  $\times$  1.2, using 10% as the coefficient of variation. For women aged 18-29 years, the RDA was determined by smoothing the value calculated.

### **2-1-2. Children (EAR, RDA)**

The EAR for adolescents aged 12-17 years was determined by the extrapolation of the EAR for adults, using the 0.75<sup>th</sup> power of the BW ratio and the growth factors. The RDA was determined as the EAR  $\times$  1.2, using 10% as the coefficient of variation.

In a study of Japanese children (mean BW: 16.34 kg), the minimal intake necessary to maintain zinc balance was estimated to be 3.87 mg/day<sup>(53)</sup>. Using data on the growth factors and integumental zinc loss among US men with a BW of 75.5 kg (0.51 mg/day)<sup>(52)</sup>, the integumental zinc loss of children with a BW of 16.34 kg was estimated to be 0.16 mg/day. The EAR for children with a BW of 16.34 kg was determined by the extrapolation of 4.03 (the sum of the minimal intake required to maintain zinc balance and integumental loss). The EAR for children aged 1-11 years was determined by the extrapolation of 4.03 mg/day to each age group, using the 0.75<sup>th</sup> power of the BW ratio and growth factors. The RDA was determined as the EAR  $\times$  1.2, using 10% as the coefficient of variation.

### **2-1-3. The Additional Amount for Pregnant and Lactating Women**

The decreases in the plasma zinc concentration are reported to be 72.7  $\mu\text{g/dL}$ , 63.8  $\mu\text{g/dL}$  and 62.1  $\mu\text{g/dL}$ , and 63.3  $\mu\text{g/dL}$  in the early term, mid term and late term of pregnancy, and at delivery, respectively<sup>(54)</sup>, and these are the additional intakes required for pregnant women. The EAR for pregnant women (amount to be added to the value of non-pregnant women) was determined to be 1 mg/day by dividing the zinc storage during pregnancy (0.40 mg/day)<sup>(55)</sup> by the zinc absorption of non-pregnant women (27%), and rounding. The RDA (amount to be added to the value of non-pregnant women) was determined as the EAR  $\times$  1.2, using 10% as the coefficient of variation.

The concentration of zinc in breast milk was calculated to be 1.13 mg/day, using the reported average concentration in Japanese women (1.45 mg/L)<sup>(56)</sup>, and the average milk volume (0.78 L/day)<sup>(2,3)</sup>. The EAR (amount to be added to the value of non-pregnant women) was calculated to be 2.13 mg/day, using the above value and an absorption rate of 53%<sup>(57)</sup>. The RDA (amount to be added to the value of non-pregnant women) was calculated as the EAR  $\times$  1.2, using 10% as the coefficient of variation. These values were rounded to yield a value of 3 mg/day.

## **2–2. Method Used to Set AI**

### **2–2–1. Infants (AI)**

Like in the case of the previous DRIs, the AI for Japanese infants aged 0-5 months was set at 2.0 mg/day in accordance with the US-Canada DRIs<sup>(44)</sup>. Several studies have focused on the breast milk of Japanese mothers<sup>(56,58–60)</sup>. Using the reported data on the zinc concentration in the breast milk of Japanese mothers (1.45 mg/L)<sup>(56)</sup>, and average breast milk intake (0.78 L/day)<sup>(2,3)</sup>, the average zinc intake was estimated to be 1.13 mg/day among infants aged 0-5 months. However, there have been no reports on zinc intake and deficiency among Japanese infants since the previous DRIs; therefore, the AI was not changed.

The AI for infants aged 6-11 months was determined as the mean of the following values: 1) the sum of the zinc intake from complementary food and infant formula (3.1 mg/day)<sup>(61)</sup>; and 2) the extrapolation of 2 mg/day using the 0.75<sup>th</sup> power of the BW ratio (2.6 mg/day). Thus, the AI was determined as 3 mg/day by rounding the calculated mean (2.85 mg/day).

### **3. To Avoid Excessive Intake**

Excessive zinc intake can occur when supplemental foods are used inappropriately. There is no evidence on the adverse effects associated with the intake of naturally occurring zinc in food.

Based on the results of a study in which 18 women in the US (age 25-40 years) were administered 50 mg/day of zinc supplements<sup>(62,63)</sup>, the LOAEL of zinc was estimated to be 60 mg/day in women with a BW of 61 kg. Using this value and an uncertainty factor of 1.5<sup>(44)</sup>, the UL for adults was determined to be 0.66 mg/kg/day × the reference BW of each age and sex group. No UL was determined for children, infants, pregnant women and lactating women as no relevant data were available.

## Copper

### 1. Background Information

#### 1–1. Definition and Classification

Copper (atomic number: 29, Cu) is a transition metal element. Approximately 80 mg of copper exists in the human body, 50% of which is distributed in the muscles and bones. As excess intracellular copper is associated with toxicity<sup>(64)</sup>, the homeostasis of copper needs to be regulated by absorption and excretion<sup>(65)</sup>. The liver is a key site in the maintenance of plasma copper concentrations<sup>(66,67)</sup>.

### 2. To Avoid Inadequacy

#### 2–1. Method Used to Set the EAR and RDA

No studies have focused on the dietary copper requirement in Japan. Therefore, the EAR for copper was determined using the plasma copper concentration, serum ceruloplasmin concentration, and erythrocyte superoxide dismutase activity (SOD), in accordance with the US-Canada DRIs<sup>(68)</sup>. Although the use of these indicators for the DRIs has some limitations<sup>(69)</sup>, no better indicator has been suggested till date<sup>(69–74)</sup>.

##### 2–1–1. Adults (EAR, RDA)

Two studies examining the effects of copper intake on the copper status of men in the US reported that the amounts of copper intake that showed no difference in the copper status indicators mentioned above were 0.66 mg/day and 0.79 mg/day<sup>(75,76)</sup>. While a study suggested that an intake of 0.66 mg/day was not sufficient for the maintenance of whole-body copper metabolism<sup>(77)</sup>, in another study, no change in the biomarkers was observed when the dietary copper intake was increased from 0.8 mg/day to 7.5 mg/day<sup>(78)</sup>. Several reviews have suggested that the appropriate intake of copper is between 0.8 and 0.94 mg/day<sup>(69–71)</sup>. From these values, the minimal copper intake required was estimated to be 0.79 mg/day. The EAR for each sex and age group was determined by extrapolation, using the reference BW of the US-Canada DRIs (men aged 18-30 years, 76.0 kg) and the 0.75<sup>th</sup> power of the BW ratio. The RDA was determined as the EAR × 1.3, using 15% as the coefficient of variation.

One report recommended a copper intake of 0.6 mg/1,000 kcal for the prevention of the decrease in the blood copper concentrations among elderly patients with enteral nutrition therapy, who tend to have copper deficiency<sup>(79)</sup>. However, no report has stated that the requirement for healthy elderly individuals is higher than that of adults aged 18-69 years. Therefore, the EAR for those aged over 70 years was determined to be the same as that for younger adults.

##### 2–1–2. Children (EAR, RDA)

The EAR was extrapolated from the values for adults, using the 0.75<sup>th</sup> power of the

BW ratio and growth factors. The RDA was determined as the EAR  $\times$  1.3, using 15% as the coefficient of variation.

### **2–1–3. The Additional Amount for Pregnant and Lactating Women**

A full-term fetus has approximately 13.7 mg of copper<sup>(80)</sup>. A study using a stable isotope reported that the dietary copper absorption is 44-67% among healthy individuals<sup>(77)</sup>. Therefore, using a dietary absorption rate of 60%, the additional EAR for pregnant women was determined as 1.0 mg/day (13.7 mg  $\div$  280 days  $\div$  0.6 = 0.8 mg/day, rounded). The additional RDA was determined as the EAR  $\times$  1.3, using 15% as the coefficient of variation.

For lactating women, based on the average copper concentration in the breast milk of Japanese mothers (0.35 mg/L)<sup>(56)</sup>, the average milk intake (0.78 L/day)<sup>(2,3)</sup>, and a copper absorption rate of 60%, the additional EAR was determined to be 0.5 mg/day (0.35  $\times$  0.78  $\div$  0.6 = 0.455 mg/day, rounded). The additional RDA was determined as the EAR  $\times$  1.3, using 15% as the coefficient of variation.

## **2–2. Method Used to Set Adequate Intake**

### **2–2–1. Infants (AI)**

The average copper concentrations in the breast milk of Japanese women were estimated to be 0.35 mg/L (age 0-5 months) and 0.16 mg/L (age 6-11 months)<sup>(56)</sup>. For infants aged 0-5 months, the AI was determined as 0.3 mg/day, using the average milk volume (0.78 L/day)<sup>(2,3)</sup> (0.35 mg/L  $\times$  0.78 L/day). For infants aged 6-11 months, based on the intake from breast milk (0.16 mg/L<sup>(56)</sup>  $\times$  0.53 L/day<sup>(81,82)</sup>) and the intake from complementary food (median of 0.05-0.34 mg/day)<sup>(54)</sup>, the AI was estimated to be 0.28 mg/day by rounding 0.3 mg/day.

## **3. To Avoid Excessive Intake**

Excessive copper intake can occur when supplemental foods are used inappropriately. No studies have reported on the presence of adverse effects due to the intake of naturally occurring copper in food.

Based on the fact that a study with an administration of 10 mg/day of copper supplements did not observe adverse effects<sup>(83)</sup>, the NOAEL was estimated to be 10 mg and the UL was set at 10 mg/day, using an uncertainty factor of 1.0. No UL was determined for children, infants, pregnant women, and lactating women, as no relevant data were available.

## Manganese

### 1. Background Information

#### 1–1. Definition and Classification

Manganese (atomic number: 25, Mn) is a group 7 element, 12-20 mg of which is present in the adult human body, uniformly distributed across the tissues and organs<sup>(84)</sup>.

### 2. To Avoid Inadequacy

#### 2–1. Method Used to Set the AI

Balance studies, pertaining to manganese, have been conducted in Japan and other countries<sup>(85,86)</sup>. However, only a small percentage of dietary manganese is absorbed, and most of it is excreted in feces<sup>(84)</sup>. Therefore, short-term balance data could not be used to estimate the average requirement for manganese, in accordance with the US-Canada DRIs<sup>(87)</sup>. The AI was set using the Japanese dietary manganese intake, which most likely far exceeds the requirement for manganese balance.

##### 2–1–1. Adults (AI)

Based on a review of the manganese intake of Japanese individuals, the average manganese intake of adults was  $3.8 \pm 0.8$  mg/day in men,  $3.8 \pm 1.4$  mg/day in women, and  $3.6 \pm 1.1$  mg/day in adults, as examined using duplicate methods<sup>(88)</sup>. Another study that examined weighed dietary records reported that the median manganese intake was 4.5 mg/day in men, and 3.9 mg/day in women (aged 30-69 years)<sup>(89)</sup>. To account for the differences in the energy intake between men and women, the AI for adults aged 18 years and older was set at 4.0 mg/day in men, and 3.5 mg/day in women.

##### 2–1–2. Children (AI)

Although several studies have focused on manganese intake among Japanese children<sup>(90,91)</sup>, the estimated intake has a wide range. Therefore, the AI for children and adolescents was determined by the extrapolation of the value for adults, using the 0.75<sup>th</sup> power of the BW ratio and growth factors.

##### 2–1–3. Infants (AI)

Since there are differences in the concentration of manganese in the breast milk of lactating mothers in Japan and the US<sup>(56,92–94)</sup>, the current DRIs used Japanese data. For infants aged 0-5 months, using the average manganese concentration in the breast milk of Japanese women (11 µg/L)<sup>(56)</sup>, and the average milk intake (0.78 L/day)<sup>(2,3)</sup>, the AI was set at 0.01 mg/day by rounding 8.6 µg/day. For infants aged 6-11 months, the average manganese intake was estimated as 0.44 mg/day<sup>(61)</sup>. Therefore, taking the manganese intake from breast milk to be 5.8 µg/day (manganese concentration in breast milk : 11 µg/L<sup>(56)</sup>, and average milk intake to be 0.53

L/day<sup>(81,82)</sup>), the AI was set at 0.5 mg/day by rounding 0.446 mg/day.

#### **2–1–4. Pregnant and Lactating Women (AI)**

The AI for pregnant women was the same as that for non-pregnant women, due to a lack of data on the manganese intake required during pregnancy.

For lactating women, milk production can lead to a loss of 172-286 µg/day of dietary manganese [manganese concentration in breast milk: 11 µg/L × average milk volume: 0.78 L/day ÷ absorption rate: (0.03-0.05) = 172-286 µg/day]. However, this value is much lower than the AI of non-pregnant women; therefore, the AI was set at a value that was equal to that of non-pregnant women.

### **3. To Avoid Excessive Intake**

Excessive manganese intake can occur in the case of strictly vegan diets, and the inappropriate intake of supplemental foods.

The manganese intake from meals comprising grains, beans, and nuts is estimated to be no higher than 10.9 mg/day<sup>(95)</sup>. Similarly, vegetarians may have an intake of 13-20 mg/day of manganese<sup>(96)</sup>. The US-Canada DRIs estimated the NOAEL of manganese to be 11 mg<sup>(88)</sup>.

From these reports, the UL for adults was set at 11 mg/day, using a NOAEL of 11 mg/day and an uncertainty factor of 1.0. No UL was determined for children, infants, pregnant women, and lactating women, as no relevant data were available.

## **Iodine**

### **1. Background Information**

#### **1–1. Definition and Classification**

Iodine (atomic number: 53, I) is a halogen element. A total of 70-80% of the iodine in the body is distributed in the thyroid, as it is an essential component of the thyroid hormone.

### **2. To Avoid Inadequacy**

#### **2–1. Method Used to Set the EAR and RDA**

Japanese people routinely consume marine products, which contain high levels of iodine, and their average iodine intake is estimated to be much higher than in other populations. However, since there are no Japanese studies available for the setting of the requirement of iodine intake, the EAR and RDA were determined based on studies conducted in Western countries.

##### **2–1–1. Adults (EAR, RDA)**

The EAR was determined through the measurement of thyroid iodine accumulation and turnover. Based on the results of 2 US studies, the accumulation of radioiodine by the thyroid gland was estimated to be 95  $\mu\text{g}/\text{day}$ <sup>(97,98)</sup> in adults. This value was used for the EAR of men and women. In accordance with the US-Canada DRIs<sup>(99)</sup>, the RDA was determined to be 130  $\mu\text{g}/\text{day}$ , using the EAR  $\times$  1.4, and a coefficient of variation of 20%.

##### **2–1–2. Children (EAR, RDA)**

For children and adolescents aged 1-17 years, the EAR was determined by the extrapolation of the EAR for adults aged 18-29 years using the 0.75<sup>th</sup> power of the BW ratio and growth factors. The RDA was determined as the EAR  $\times$  1.4, using 20% as the coefficient of variation.

##### **2–1–3. The Additional Amount for Pregnant and Lactating Women**

According to a Western study, the iodine turnover in newborn infants ranged from 50-100  $\mu\text{g}/\text{day}$ <sup>(100)</sup>. Using the median value (75  $\mu\text{g}/\text{day}$ ), the EAR was determined as the amount to be added to the value of non-pregnant women.

A balance study examining 5 pregnant women reported that the iodine intake necessary for the maintenance of iodine balance was approximately 160  $\mu\text{g}/\text{day}$ <sup>(101)</sup>, which is similar to the sum of the EAR for non-pregnant women and the additional EAR for pregnant women (170  $\mu\text{g}/\text{day}$ ). The RDA (to be added to the RDA for non-pregnant women) was determined as the EAR  $\times$  1.4, using 20% as the coefficient of variation.

The iodine loss from breast milk may be large in Japanese women. However, this could be caused by a high intake of iodine; this suggests that there is no requirement for an increase

in the intake, based on the high iodine concentration of breast milk. Therefore, for lactating women, the EAR was determined to be the same as the AI for infants aged 1-5 months. Estimating the iodine absorption to be 100%, the additional EAR was set at 100 µg/day. The additional RDA was determined as the EAR × 1.4, using 20% as the coefficient of variation. The WHO set the recommendation for iodine intake at 250 µg/day for pregnant or lactating women<sup>(102)</sup>.

## **2–2. Method Used to Set the AI**

### **2–2–1. Infants (AI)**

The iodine content of the breast milk of Japanese mothers is reported to be 77-3,971 µg/L (n=39, median 172 µg/L)<sup>(103)</sup>, or 83-6,960 µg/L (n=33, median 207 µg/L)<sup>(104)</sup>. When using the median of these values (189 µg/L), and the average milk intake<sup>(2,3)</sup>, the iodine intake of infants aged 0-5 months can be estimated as 147 µg/day; this is much higher than the AI in the US-Canada DRIs (110 µg/day)<sup>(99)</sup>. Therefore, the AI for infants aged 0-5 months was determined to be 100 g/day, considering the value of the US-Canada DRIs, and the difference in the body size between Japanese individuals and those from the US. The WHO set the recommendation for iodine intake at 90 µg/day for infants<sup>(105)</sup>.

For infants aged 6-11 months, the iodine intake from complementary food ranges widely<sup>(106,107)</sup>, and it is difficult to use these values for the estimation of AI. Therefore, the AI for infants aged 6-11 months was determined by extrapolating the AI for infants aged 0-5 months using the 0.75<sup>th</sup> power of the BW ratio.

## **3. To Avoid Excessive Intake**

### **3–1. Dietary Intake**

The high iodine intake among Japanese individuals has been examined from several angles. Based on a chemical iodine analysis of duplicate diets<sup>(108)</sup>, and the measurement of urinary iodine excretion<sup>(109,110)</sup>, the iodine intakes were regularly lower than 500 µg/day with an intermittent intake of 2-10 mg/day. Based on the annual reports on the consumption of seaweeds, the average iodine intake was estimated as 1.2 mg/day<sup>(111)</sup>. A review of the iodine intake among Japanese individuals showed that the average intake was 1-3 mg/day<sup>(112)</sup>. From these results, the iodine intake in Japanese populations can be estimated as 1-3 mg/day for diets without seaweed (less than 500 µg/day) and those with seaweed. Recent reports on the iodine intake in Japan support this value<sup>(113,114)</sup>.

## **3–2. Method Used to Set the UL**

### **3–2–1. Adults (UL)**

Initially, excessive iodine intake induces hypothyroidism and goiter, a phenomenon referred to as the Wolff-Chaikoff effect. However, this effect does not occur due to a continuous excessive intake of iodine; it is a result of a phenomenon referred to as the “escape phenomenon,”

which maintains the thyroid hormone synthesis within the normal range<sup>(115)</sup>. Excessive iodine intake is assumed to affect Japanese individuals to a lower extent, due to their unique iodine intake pattern and the escape phenomenon. However, despite the presence of the escape phenomenon, excessive iodine intake decreases the synthesis of the thyroid hormone, which can induce hypothyroidism, or goiter at worst<sup>(99)</sup>.

In the US-Canada DRIs, using an iodine intake value of 1.7 mg/day, which results in hypothyroidism, as the LOAEL, the UL was set at 1.1 mg/day among adults<sup>(99)</sup>. Some reports pointed to a higher risk of goiter among those whose iodine intake exceeded 1.5 mg/day (mainly from water) in China and Africa<sup>(116,117)</sup>. In contrast, although the iodine intake in Japanese individuals can be estimated at an average of 1-3 mg/day, the prevalence of hypothyroidism or goiter is exceedingly low. Therefore, for Japanese adults, 3 mg/day was regarded as an upper limit--the NOAEL. Using this value, and an uncertainty factor of 1, the UL was estimated as 3.0 mg/day.

According to several Japanese case studies, unusual iodine intakes, such as intakes of 28 mg/day for a 1-year period (mainly from seaweed soup)<sup>(118)</sup>, or consuming one pack of seaweed chips a day<sup>(119)</sup>, led to the development of hypothyroidism or goiter. Some Japanese experimental studies reported that an iodine intake (seaweed) of 35-70 mg/day for 7-10 days increased the serum thyroid-stimulating hormones (TSH) levels<sup>(120)</sup>, and an iodine preparation of 27 mg/day for 28 days decreased the thyroid activity and increased the thyroid volume<sup>(121)</sup>. Taking these values as the NOAELs, the ULs were estimated as 2.8, 3.5 and 2.7 mg/day, respectively, using an uncertainty factor of 10. An epidemiological study examining people living in the coastal areas of Hokkaido reported that an increased prevalence of hypothyroidism was observed among those with an iodine intake greater than 10 mg/day, based on a urine analysis<sup>(122,123)</sup>. As the urinary iodine concentration was measured only once, this value could not be used for the determination of the UL.

Based on these reports, the UL was estimated to be around 3.0 mg/day; therefore, the UL for adults was set at this value. One study reported that the average iodine intake among those who did not consume seaweed was only 73 µg/day<sup>(124)</sup>. Therefore, as the UL applies to habitual iodine intake, it is not necessary to restrict the habitual intake of seaweed.

### **3-2-2. Children (UL)**

A study examining children aged 6-12 years, worldwide, reported that an iodine intake greater than 500 µg/day could be harmful, as the thyroid volume of Japanese children living in the coastal areas of Hokkaido was significantly larger than that of other populations, and their estimated average iodine intake was 741 µg/day<sup>(125)</sup>. Therefore, the UL was set at 500 µg/day for children aged 6-17 years.

The UL for children aged 1-5 years was extrapolated from that of those aged 6-7 years, using the 0.75<sup>th</sup> power of the BW ratio, and the average of the values for boys and girls was adopted as the UL. Among children aged 12-17 years, the UL was set at 1.2 g/day for the 12-

14 years age group, and 2 mg/day for the 15-17 years group, considering the values for those aged 10-11 years and adults. As the UL applies to habitual iodine intake, it is not necessary to restrict the habitual intake of seaweed.

### **3–2–3. Infants (UL)**

A decrease in serum thyroid hormone levels, and an increase in TSH levels were observed in low-birth weight Korean infants whose iodine intake from breast milk exceeded 100 µg/kg/day<sup>(126)</sup>. Using this value, and an uncertainty factor of 3, 33 µg/kg/day was set as the value for the determination of the UL for each age category. Using this value and the reference BW, the ULs were calculated as 208 µg/day (in boys aged 1-5 months), 195 µg/day (in girls aged 1-5 months), 290 µg/day (in boys aged 6-11 months), and 267 µg/day (in girls aged 6-11 months). However, as the participants of the Korean study were low-birth weight infants, the UL was determined as 250 µg/day by rounding the average value of the four values calculated above. Although this UL applies to habitual iodine intake, breast-feeding mothers need to pay attention to the UL of iodine intake, as infants display a higher sensitivity to iodine<sup>(127)</sup>.

### **3–2–4. Pregnant and Lactating Women (UL)**

In Japanese case reports focusing on hypothyroidism in infants, the mothers' iodine intakes were reported to be 1.9-4.3 mg/day<sup>(128)(129)</sup>. However, it is difficult to use these values for the setting of the UL, due to the inaccuracy of the dietary iodine assessment. The iodine intake of over 500 healthy pregnant or lactating Japanese women was estimated to be 1.4-1.7 mg/day, using a food frequency questionnaire focusing on iodine intake<sup>(130)</sup>, indicating that the iodine intake of pregnant women is not very different from that of normal adults. As infants may display a higher sensitivity to iodine<sup>(127)</sup>, pregnant women should pay attention to excess iodine intake. Therefore, using the UL for adults and an uncertainty factor of 1.5, 2 mg/day was set as the UL for pregnant women. For lactating women, a special UL was not set due to a lack of relevant data. However, a consistent excessive intake of iodine is not recommended for lactating women compared to non-lactating women.

## Selenium

### 1. Background Information

#### 1–1. Definition and Classification

Selenium (atomic number: 34, Se) is a group 16 element. Fish and shellfish are known to contain high levels of selenium. The amount of selenium in plant foods and stock farm products depends on the type of soil and feed, respectively<sup>(131)</sup>.

### 2. To Avoid Inadequacy

#### 2–1. Method Used to Set the EAR and RDA

The EAR and RDA were determined for the prevention of selenium deficiency disorders such as Keshan disease.

##### 2–1–1. Adults (EAR, RDA)

Although the synthesis of selenium-containing protein is strongly associated with selenium intake, stability is maintained at a certain intake level<sup>(132)</sup>. The association between plasma glutathione peroxidase (GPX) and selenium intake has been well-examined. In a study that examined a low-selenium area of China, a saturation in the plasma GPX activity was observed at a selenium intake of 41 µg/day in men with an average weight of 60 kg<sup>(133)</sup>. Together with this result, the US-Canada DRIs used the findings of another study that showed that an intake level of 38 µg/day resulted in saturation<sup>(134)</sup>; accordingly, the EAR was set at 45 µg/day using these average values, at a BW of 76 kg<sup>(135)</sup>. However, the WHO concluded that selenium deficiency may be prevented when two-third of the value of saturated plasma GPX activity is maintained<sup>(136)</sup>.

Several studies have reported the absence of deficiency at low selenium intakes, with unsaturated serum or erythro GPX activity<sup>(137)(138)(139)</sup>, indicating that maintaining two-third of the value of saturated plasma GPX activity is sufficient. Based on the aforementioned Chinese study<sup>(133)</sup>, the selenium intake necessary to maintain two-third of the value of saturated plasma GPX activity was estimated to be 24.2 µg for adults with a BW of 60 kg, using the WHO equation<sup>(136)</sup>. The EAR for selenium in adults aged 18 years and older was calculated by the extrapolation of this value, using the 0.75<sup>th</sup> power of the BW ratio. The RDA was determined as the EAR × 1.2, using 10% as the coefficient of variation.

##### 2–1–2. Children (EAR, RDA)

The EAR for children aged 1-17 years was determined by the extrapolation of the value for adults (24.2 µg/day, BW 60 kg), using the 0.75<sup>th</sup> power of the BW ratio and growth factors. The RDA was determined as the EAR × 1.2, using 10% as the coefficient of variation.

### **2–1–3. The Additional Amount for Pregnant and Lactating Women**

Based on the average body selenium concentration in fetuses ( $250 \mu\text{g}/\text{kg}$ )<sup>(96)</sup>, and the sum of the placental and birth weights (3.5 kg), the fetal and placental selenium storage was estimated to be about  $900 \mu\text{g}$  during pregnancy. The average blood selenium concentration has been reported to be  $170\text{--}198 \mu\text{g}/\text{L}$  (average  $184 \mu\text{g}/\text{L}$ )<sup>(140)</sup>. Therefore, the increased selenium requirement due to an increase in the blood volume (1.5 L) during pregnancy can be estimated as about  $300 \mu\text{g}$ . Using a dietary selenium absorption rate of 90%<sup>(132)</sup>, the EAR added to the value of non-pregnant women, was set at  $5 \mu\text{g}/\text{day}$  ( $(900 + 300) \mu\text{g}/0.9/280$  days, rounded). The additional RDA was determined as the EAR  $\times$  1.2, using 10% as the coefficient of variation.

For lactating women, the additional EAR was determined to be  $15 \mu\text{g}/\text{day}$ , based on the average selenium concentration in the breast milk of Japanese women ( $17 \mu\text{g}/\text{L}$ )<sup>(56)</sup>, average milk volume ( $0.78 \text{ L}/\text{day}$ )<sup>(2,3)</sup>, and a dietary selenium absorption rate of 90%<sup>(132)</sup>. The additional RDA was determined as the EAR  $\times$  1.2, using 10% as the coefficient of variation.

### **2–2. Method Used to Set the AI**

#### **2–2–1. Infants (AI)**

Based on the average selenium concentration in the breast milk of Japanese women ( $17 \mu\text{g}/\text{L}$ )<sup>(56)</sup>, the AI for infants aged 0–5 months was determined as  $15 \mu\text{g}/\text{day}$ , using the average milk intake ( $0.78 \text{ L}/\text{day}$ ).

A study reported that there was no difference in the plasma selenium concentration between exclusively-breastfed infants and infants fed formula and complementary food (aged 12 months)<sup>(141)</sup>. Therefore, the AI for infants aged 6–11 months was calculated by the extrapolation of the AI for infants aged 0–5 months ( $13.3 \mu\text{g}/\text{day}$ ), using the 0.75<sup>th</sup> power of the BW ratio, and was set at  $15 \mu\text{g}/\text{day}$  for both boys and girls.

### **3. To Avoid Excessive Intake**

Excess selenium intake can result from inappropriate supplemental food intake.

#### **3–1. Method Used to Set the UL**

##### **3–1–1. Adults and Children (UL)**

Based on a Chinese report on chronic selenium intoxication, the minimum intake among 5 patients was estimated as  $913 \mu\text{g}/\text{day}$  (average BW 60 kg). After recovery, the average selenium intake was estimated as  $800 \mu\text{g}/\text{day}$ . From these results, the LOAEL was set at  $913 \mu\text{g}/\text{day}$  ( $15.2 \mu\text{g}/\text{kg weight}/\text{day}$ ), and the NOAEL at  $800 \mu\text{g}/\text{day}$  ( $13.3 \mu\text{g}/\text{kg weight}/\text{day}$ )<sup>(142)</sup>. In a study on selenium intoxication among farm animals in the US, no health effect was found among 142 farmers with a maximum selenium intake of  $724 \mu\text{g}/\text{day}$ <sup>(143)</sup>. This supports the setting of an LOAEL of  $800 \mu\text{g}/\text{day}$ . This value was used as the LOAEL, and the UL was determined using an uncertainty factor of 2, as well as the reference BW for each age and sex category.

**3–1–2. Infants (UL)**

The US-Canada DRIs set the UL at 47 µg/L using a breast milk selenium concentration of 60 µg/L, as almost no selenium intoxication was observed at this level<sup>(135,144,145)</sup>. However, a report stated that there were very few cases of selenium intoxication in the hair and nails<sup>(145)</sup>. Therefore, the current DRIs did not set the UL due to insufficient evidence.

**3–1–3. Pregnant and Lactating Women (UL)**

The UL for pregnant or lactating women was not determined due to a lack of adequate information.

## Chromium

### 1. Background Information

#### 1–1. Definition and Classification

Chromium (atomic number: 24, Cr) is group 6 element, and is predominantly consumed as trivalent chromium in the regular diet.

### 2. To Avoid Inadequacy

#### 2–1. Method Used to Set the AI

##### 2–1–1. Adults and Children (AI)

The WHO<sup>(146)</sup> and a UK study<sup>(147)</sup> estimated the requirement of chromium to be 24.5 µg/day, based on a balance study<sup>(148)</sup>. However, that study examined only a small number of elderly people, and did not estimate the intake required for the maintenance of equilibrium; therefore, it was not regarded as evidence for the setting of the EAR. As it was difficult to determine the EAR, the AI was determined based on the chromium intake, in accordance with the US-Canada DRIs<sup>(149)</sup>.

According to a report measuring the chromium content of diets, the chromium intake was estimated as ranging from 20-80 µg/day among adults, including those in Japan<sup>(150)</sup>. In contrast, the chromium intake among Japanese individuals was estimated to be about 10 µg/day, on using the Japanese Standard Food Composition Table 2010<sup>(151)(152)</sup>. These results indicate that there may be differences between the findings of chemical analyses and intake estimations. Therefore, although it is difficult to accurately estimate the chromium intake of Japanese people, it may be more appropriate to use the estimated intake using dietary assessment (10 µg/day)<sup>(151)(152)</sup>. Thus, the AI was set at 10 µg/day. For children, the AI was not determined as there was no information on selenium intake.

##### 2–1–2. Infants (AI)

According to a Japanese report on the chromium concentration of breast milk, the chromium intake was less than 1 µg/L among 48% of participants and 1-2 µg/L among 25%; only 8% of the participants had an intake higher than 5 µg/L (median: 1.00 µg/L)<sup>(153)</sup>. These results were higher than the chromium concentration of breast milk used in the US-Canada DRIs (0.25 µg/L)<sup>(154)</sup>. However, the Japanese intake is within the intake range of the investigation by the WHO/IAEA<sup>(155)</sup>. Based on the median chromium concentration in the breast milk of Japanese mothers (1.00 µg/L), and average milk volume (0.78 L/day)<sup>(2,3)</sup>, the AI for infants aged 0-5 months was determined at 0.8 µg/day. For infants aged 6-11 months, this value was extrapolated to calculate the AI, using the 0.75<sup>th</sup> power of the BW ratio.

##### 2–1–3. Pregnant and Lactating Women (AI)

For pregnant or lactating women, the AI was determined to be the same as that for

non-pregnant/non-lactating adults.

### **3. To Avoid Excessive Intake**

Although excess hexahydric chromium can accumulate in the kidneys, spleen, liver, lung and bones, and cause toxicity<sup>(156)</sup>, hexahydric chromium is artificially produced and it occurs naturally in low amounts. The present DRIs did not consider hexahydric chromium in the determination of the UL.

Some reports focused on the health effects associated with chromium supplementation<sup>(157,158)</sup>. However, those reports did not examine the influence of other medications or supplements. The UL for chromium was not set due to a lack of sufficient data on the quantitative association between trivalent chromium intake and possible adverse effects.

### **4. For the Prevention of the Development and Progression of Lifestyle-related Diseases**

A meta-analysis<sup>(159)</sup> found that while chromium supplementation had a positive effect on blood glucose and HbA1c levels among patients with type 2 diabetes, there was no effect in those without diabetes, including those with impaired glucose tolerance. The studies included in the meta-analysis used chromium chloride and picolinate chromium intake levels (200-1,000 µg/day), and chromium yeast levels (10-400 µg/day).

The findings of other relevant studies on diabetes patients are inconsistent<sup>(160,161)</sup>. However, one study reported the absence of an effect of chromium supplementation (500 or 1,000 µg/day) in those with impaired glucose tolerance, elevated fasting blood glucose levels, or metabolic syndrome<sup>(162)</sup>. Furthermore, decreased insulin sensitivity was observed in normal-weight participants without diabetes who were administered 1,000 µg/day of chromium supplementation<sup>(163)</sup>.

## Molybdenum

### 1. Background Information

#### 1–1. Definition and Classification

Molybdenum (atomic number: 42, Mo) is among the chromium group elements.

### 2. To Avoid Inadequacy

#### 2–1. Method Used to Set the EAR and RDA

##### 2–1–1. Adults (EAR, RDA)

The EAR for molybdenum was determined based on the results of a balance test of 4 men in the US<sup>(164)</sup>. In this study, a positive balance of molybdenum was observed without any molybdenum deficiency in all participants consuming 22 µg/day of molybdenum for 102 days. Estimating the integumental and sweat molybdenum loss to be 3 µg/day, 25 µg/day was set as a reference value for the calculation of the EAR. Using the average BW of the study (76.4 kg) and the 0.75<sup>th</sup> power of the BW ratio, the EAR was determined by the extrapolation for each age and sex group.

The RDA was determined as the EAR × 1.2, using 10% as the coefficient of variation. It is important to note that this EAR and RDA are dependent on the result of one study, and the reliability might need to be considered cautiously; nevertheless, the US-Canada DRIs and WHO have adopted the same method<sup>(154,165)</sup>.

##### 2–1–2. Children (EAR, RDA)

There is little evidence on the EAR of children, and it is difficult to extrapolate the EAR for adults to that of children, since the EAR for adults was determined based on a study of 4 individuals. The EAR and RDA were not determined for children aged under 18 years.

##### 2–1–3. The Additional Amount for Pregnant and Lactating Women

The additional EAR for pregnant women was not determined due to a lack of data.

For lactating women, based on the average molybdenum concentration of the breast milk of Japanese mothers (3.0 µg/L)<sup>(153,166)</sup>, the average milk volume (0.78 L/day)<sup>(2,3)</sup>, and the Japanese dietary molybdenum absorption rate (93%)<sup>(167)</sup>, the EAR was determined as 3 µg/day (3.0 × 0.78 ÷ 0.93, rounded), as the amount to be added to that for non-pregnant women. The RDA was determined as the EAR × 1.2, using 10% as the coefficient of variation.

#### 2–2. Method Used to Set AI

##### 2–2–1. Infants (AI)

Two reports focused on the molybdenum concentration of breast milk in Japanese women. One study reported that the level was 0.8-34.7 µg/L (median 2.9 µg/L)<sup>(166)</sup>, while another reported the range to be 0.1 to 25.91 µg/L (median 3.18 µg/L)<sup>(153)</sup>. Based on the average

of these median values (3.0 µg/L), and the average milk volume (0.78 L/day)<sup>(2,3)</sup>, the EAR for infants aged 0-5 months was determined as 2 µg/day.

For infants aged 6-11 months, the molybdenum intake from complementary foods should be considered. One study estimated the molybdenum intake to be 6.5 µg/day in children aged 6-8 months, and 12.5 µg/day in children aged 9-11 months, based on the content analysis of commercially available complementary foods in Japan<sup>(168)</sup>. Using the average of these values, the AI for infants aged 6-11 months was determined at 10 µg/day.

### **3. To Avoid Excessive Intake**

#### **3—1. Example of Molybdenum Intoxication**

Few studies have focused on molybdenum intoxication. One study reported on the relationship between molybdenum intake and high uric acid levels, and the development of gout symptoms<sup>(169)</sup>. Using the results of that study, the American Environment Preservation Association set an LOAEL of 140 µg/kg weight, and a reference value of 5 µg/kg for chronic oral molybdenum intake, using an uncertainty factor of 30<sup>(170)</sup>. The WHO uses the same reference value<sup>(165)</sup>. However, the National Research Conference of America has concluded that the influence of molybdenum on high uric acid levels and the development of gout has not been substantially established in the aforementioned study<sup>(171)</sup>.

#### **3—2. Dietary Intake**

Since molybdenum is present in high quantities in grains and beans, consuming a strict vegetarian diet may lead to a high intake of molybdenum. The molybdenum intake in Japanese populations has been reported to be 225 µg/day, on average<sup>(172)</sup>. Another study reported that the molybdenum intake was higher than 300 µg/day when a soybean-rich diet was consumed<sup>(167)</sup>. In contrast, 540 µg/day was reported as the mean molybdenum intake in Japanese women (mean BW: 49.1 kg) consuming a strict vegetarian diet; however, no negative effect was observed in that study<sup>(173)</sup>.

#### **3—3. Method Used to Set the UL**

A study reported that no negative effect was observed among 4 Americans who received oral administration of molybdenum stable isotope after the consumption of 1,490 µg/day of molybdenum for 24 days<sup>(174)</sup>. The UL was determined using this result, and was set as the NOAEL. The NOAEL was calculated as 1,500 µg/day (total molybdenum intake)/82 kg (the mean BW) = 18 µg/kg weight/day. Using an uncertainty factor of 2, 9 µg/kg/day was set as the reference value for the calculation of the UL. The ULs were calculated as 550 µg/day for men, and 450 µg/day for women, using the reference BW for those aged over 70 years (the lowest among all adults) after rounding. These values are consistent with the results of the aforementioned Japanese study that reported no negative effects among vegetarian women consuming approximately 500 µg/day of molybdenum<sup>(173)</sup>.

II Energy and Nutrients  
Minerals (2) Microminerals  
Molybdenum

The UL was not determined for infants, children, pregnant women, and lactating women due to a lack of data.

**DRIs for Iron (mg/day) <sup>1</sup>**

Gender	Males				Females					
	EAR	RDA	AI	UL	Not menstruating		Menstruating		AI	UL
					EAR	RDA	EAR	RDA		
0-5 months	—	—	0.5	—	—	—	—	—	0.5	—
6-11 months	3.5	5.0	—	—	3.5	4.5	—	—	—	—
1-2 years	3.0	4.5	—	25	3.0	4.5	—	—	—	20
3-5 years	4.0	5.5	—	25	3.5	5.0	—	—	—	25
6-7 years	4.5	6.5	—	30	4.5	6.5	—	—	—	30
8-9 years	6.0	8.0	—	35	6.0	8.5	—	—	—	35
10-11 years	7.0	10.0	—	35	7.0	10.0	10.0	14.0	—	35
12-14 years	8.5	11.5	—	50	7.0	10.0	10.0	14.0	—	50
15-17 years	8.0	9.5	—	50	5.5	7.0	8.5	10.5	—	40
18-29 years	6.0	7.0	—	50	5.0	6.0	8.5	10.5	—	40
30-49 years	6.5	7.5	—	55	5.5	6.5	9.0	10.5	—	40
50-69 years	6.0	7.5	—	50	5.5	6.5	9.0	10.5	—	40
70+ years	6.0	7.0	—	50	5.0	6.0	—	—	—	40
Pregnant women (additional)	/									
Early stage					+2.0	+2.5	—	—	—	—
Mid to late stage					+12.5	+15.0	—	—	—	—
Lactating women (additional)	/				+2.0	+2.5	—	—	—	—

<sup>1</sup> Developed excluding persons with menorrhagia (menstrual blood loss of 80mL/period or more).

**DRIs for Zinc (mg/day)**

Gender	Males				Females			
Age etc.	EAR	RDA	AI	UL	EAR	RDA	AI	UL
0-5 months	—	—	2	—	—	—	2	—
6-11 months	—	—	3	—	—	—	3	—
1-2 years	3	3	—	—	3	3	—	—
3-5 years	3	4	—	—	3	4	—	—
6-7 years	4	5	—	—	4	5	—	—
8-9 years	5	6	—	—	5	5	—	—
10-11 years	6	7	—	—	6	7	—	—
12-14 years	8	9	—	—	7	8	—	—
15-17 years	9	10	—	—	6	8	—	—
18-29 years	8	10	—	40	6	8	—	35
30-49 years	8	10	—	45	6	8	—	35
50-69 years	8	10	—	45	6	8	—	35
70+ years	8	9	—	40	6	7	—	35
Pregnant women (additional)					+1	+2	—	—
Lactating women (additional)					+3	+3	—	—

**DRIs for Copper (mg/day)**

Gender	Males				Females			
Age etc.	EAR	RDA	AI	UL	EAR	RDA	AI	UL
0-5 months	—	—	0.3	—	—	—	0.3	—
6-11 months	—	—	0.4	—	—	—	0.4	—
1-2 years	0.2	0.3	—	—	0.2	0.3	—	—
3-5 years	0.3	0.4	—	—	0.3	0.4	—	—
6-7 years	0.4	0.5	—	—	0.4	0.5	—	—
8-9 years	0.4	0.6	—	—	0.4	0.5	—	—
10-11 years	0.5	0.7	—	—	0.5	0.7	—	—
12-14 years	0.7	0.8	—	—	0.6	0.8	—	—
15-17 years	0.8	1.0	—	—	0.6	0.8	—	—
18-29 years	0.7	0.9	—	10	0.6	0.8	—	10
30-49 years	0.7	1.0	—	10	0.6	0.8	—	10
50-69 years	0.7	0.9	—	10	0.6	0.8	—	10
70+ years	0.7	0.9	—	10	0.6	0.7	—	10
Pregnant women (additional)					+0.1	+0.1	—	—
Lactating women (additional)					+0.5	+0.5	—	—

**DRIs for Manganese (mg/day)**

Gender	Males		Females	
Age etc.	AI	UL	AI	UL
0-5 months	0.01	—	0.01	—
6-11 months	0.5	—	0.5	—
1-2 years	1.5	—	1.5	—
3-5 years	1.5	—	1.5	—
6-7 years	2.0	—	2.0	—
8-9 years	2.5	—	2.5	—
10-11 years	3.0	—	3.0	—
12-14 years	4.0	—	4.0	—
15-17 years	4.5	—	3.5	—
18-29 years	4.0	11	3.5	11
30-49 years	4.0	11	3.5	11
50-69 years	4.0	11	3.5	11
70+ years	4.0	11	3.5	11
Pregnant women	/		3.5	—
Lactating women			3.5	—

**DRIs for Iodine ( $\mu\text{g}/\text{day}$ )**

Gender	Males				Females			
	EAR	RDA	AI	UL	EAR	RDA	AI	UL
Age etc.	EAR	RDA	AI	UL	EAR	RDA	AI	UL
0-5 months	—	—	100	250	—	—	100	250
6-11 months	—	—	130	250	—	—	130	250
1-2 years	35	50	—	250	35	50	—	250
3-5 years	45	60	—	350	45	60	—	350
6-7 years	55	75	—	500	55	75	—	500
8-9 years	65	90	—	500	65	90	—	500
10-11 years	80	110	—	500	80	110	—	500
12-14 years	100	140	—	1,200	100	140	—	1,200
15-17 years	100	140	—	2,000	100	140	—	2,000
18-29 years	95	130	—	3,000	95	130	—	3,000
30-49 years	95	130	—	3,000	95	130	—	3,000
50-69 years	95	130	—	3,000	95	130	—	3,000
70+ years	95	130	—	3,000	95	130	—	3,000
Pregnant women (additional)					+75	+110	—	— <sup>1</sup>
Lactating women (additional)					+100	+140	—	—

<sup>1</sup> UL of pregnant women is determined to be 2,000  $\mu\text{g}/\text{day}$ .

**DRIs for Selenium (µg/day)**

Gender	Males				Females			
	EAR	RDA	AI	UL	EAR	RDA	AI	UL
Age etc.	EAR	RDA	AI	UL	EAR	RDA	AI	UL
0-5 months	—	—	15	—	—	—	15	—
6-11 months	—	—	15	—	—	—	15	—
1-2 years	10	10	—	80	10	10	—	70
3-5 years	10	15	—	110	10	10	—	110
6-7 years	15	15	—	150	15	15	—	150
8-9 years	15	20	—	190	15	20	—	180
10-11 years	20	25	—	240	20	25	—	240
12-14 years	25	30	—	330	25	30	—	320
15-17 years	30	35	—	400	20	25	—	350
18-29 years	25	30	—	420	20	25	—	330
30-49 years	25	30	—	460	20	25	—	350
50-69 years	25	30	—	440	20	25	—	350
70+ years	25	30	—	400	20	25	—	330
Pregnant women (additional)					+5	+5	—	—
Lactating women (additional)					+15	+20	—	—

**DRIs for Chromium ( $\mu\text{g}/\text{day}$ )**

Gender	Males	Females
Age etc.	AI	AI
0-5 months	0.8	0.8
6-11 months	1.0	1.0
1-2 years	—	—
3-5 years	—	—
6-7 years	—	—
8-9 years	—	—
10-11 years	—	—
12-14 years	—	—
15-17 years	—	—
18-29 years	10	10
30-49 years	10	10
50-69 years	10	10
70+ years	10	10
Pregnant women	/	10
Lactating women		10

**DRIs for Molybdenum (µg/day)**

Gender	Males				Females			
Age etc.	EAR	RDA	AI	UL	EAR	RDA	AI	UL
0-5 months	—	—	2	—	—	—	2	—
6-11 months	—	—	10	—	—	—	10	—
1-2 years	—	—	—	—	—	—	—	—
3-5 years	—	—	—	—	—	—	—	—
6-7 years	—	—	—	—	—	—	—	—
8-9 years	—	—	—	—	—	—	—	—
10-11 years	—	—	—	—	—	—	—	—
12-14 years	—	—	—	—	—	—	—	—
15-17 years	—	—	—	—	—	—	—	—
18-29 years	20	25	—	550	20	20	—	450
30-49 years	25	30	—	550	20	25	—	450
50-69 years	20	25	—	550	20	25	—	450
70+ years	20	25	—	550	20	20	—	450
Pregnant women (additional)					—	—	—	—
Lactating women (additional)					+3	+3	—	—

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