Effect of Life-Long Dietary n-6/n-3 Fatty Acid Ratio on Life Span, Serum Lipids and Serum Glucose in Wistar Rats

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Summary Types of dietary lipid affect the life span of rats. In this study, we investigated the influence of the life-long dietary n-6/n-3 ratio on life span and serum lipid and glucose levels. A semi-purified diet adjusted to a constant saturated : monounsaturated : polyunsaturated fatty acid ratio and an n-6/n-3 ratio of 1 (R1), 4 (R4) or 16 (R16) was fed to rats (n=33) from 4 wk of age until death. There were no significant differences in the food intake or body weight, nor were there survival curve or mean life span variations among the 3 groups. The serum cholesterol levels after feeding the test diet for 6 and 12 mo were significantly lower in the R1 group than in the other groups, and the serum triacylglycerol levels were significantly lower than those in the R16 group. However, no significant differences were noted in the serum cholesterol or triacylglycerol level after feeding for 18 mo among the 3 groups. A significantly higher serum glucose level was noted in the R1 group only at 18 mo of test diet ingestion, compared to that in the R4 group. The results suggest that the influence of the dietary n-6/n-3 ratio on the serum lipid and glucose levels varies, depending on the duration and life stage of feeding. Our findings further suggest that the life span of Wistar rats is not affected even if the ratio of dietary n-6/n-3 changes from 1 to 16.

Key Words fatty acid balance, n-6/n-3 ratio, life span, rats

N-6 and n-3 polyunsaturated fatty acids (PUFA) are essential for normal development and the maintenance of biological functions (1). N-6 and n-3 PUFA have individual physiological actions and are independent essential fatty acids (2, 3). Since they share metabolic enzymes and mutually interfere with metabolism, a balanced ingestion of n-6 and n-3 PUFA is important to maintain health and prevent diseases (4). Although many studies have been performed using various parameters to identify the desirable n-6/n-3 ratio (4–8), it has not yet been determined. Recently, we investigated the influence of the dietary n-6/n-3 ratio on the ratio in the body using rats, and we found that the influence on the ratio in the liver and serum changed at an n-6/n-3 ratio of 4 (9). Since the body’s internal environment is regulated to maintain homeostasis (10), an n-6/n-3 ratio of 4 may have important nutritional significance.

Dietary lipid type-associated variations in the rat and mouse life span have been reported (11–15), suggesting that the dietary n-6/n-3 ratio affects the life span of rats. However, the saturated : monounsaturated : polyunsaturated fatty acid ratio (S : M : P ratio) of test fats used in these experiments was not regulated, so it is not clear whether the influence on lifespan of the lipid type was caused by the n-6/n-3 ratio. In this study, we fed a diet adjusted to an n-6/n-3 ratio of 1, 4 or 16 to rats from 4 wk of age until death, while maintaining a constant S : M : P ratio, and investigated the influences of these ratios on the rat life span.

It has been reported that dietary n-6 and n-3 fatty acids influence the lipid (16–18) and glucose metabolism (19, 20) in rats. However, the influence of the ratio of dietary n-6/n-3 over the rat life span has not been sufficiently clarified. Another purpose of this study was to examine the effect of a life-long dietary n-6/n-3 fatty acid ratio on serum lipids and glucose levels in Wistar rats.

MATERIALS AND METHODS

All animals were treated in accordance with the guidelines for the care and use of laboratory animals (Notification of the Prime Minister’s Office in Japan). The experimental plan was approved by the Laboratory Animal Care Committee of the Central Research Laboratory, Nisshin OilliO Group. Specific pathogen-free (SPF) male Wistar rats (Japan SLC, Inc., Hamamatsu, Japan) were housed in a SPF barrier facility with a one-way airflow and HEPA filtration system. Rats were individually housed in stainless steel wire cages and allowed free access to sterilized water. The temperature of the animal room was set at 22±2°C, with a humidity
Rats were killed after consuming the test diet for 4 wk. Other position of liver and serum lipids, five rats in each group groups following 16 h of food deprivation. Blood was collected the measurement of the fatty acid compositions of liver with an Rats were fed an experimental diet containing test fat 200; sucrose, 100; cellulose, 50; AIN-93G mineral mix ingredients (g/kg diet): cornstarch, 499.48; casein, lenic acid. Experimental diets contained the following 19.8% of energy as fat, carbohydrate, and protein, and test oil, 100. The diets provided 21.9, 58.3, and choline bitartrate, 2.5; 19.8% of energy as fat, carbohydrate, and protein, while maintaining a constant S : M : P ratio (1 : 1.5 : 1). Care was taken that the peroxides never exceeded 10 mmol/kg dietary fat. Each group of rats was given free access to the experimental diet and water.

After consuming the test diet for 4 wk, rats (n = 5) for the measurement of the fatty acid compositions of liver and serum lipids were killed by decapitation at 10:00 h following 16 h of food deprivation. Blood was collected to obtain serum. Livers were quickly removed, weighed, and stored at –80°C until analyses. The samples were freeze-dried, and their total lipids were extracted by the method of Folch et al. (22). The fatty acid composition was determined by gas-liquid chromatography after methylation (23). The fatty acid methyl esters were analyzed using a Hewlett-Packard 6890 chromatograph (Palo Alto, CA, USA) with a BPX-70 capillary column (30 m, i.d. 0.32 mm; SGC Japan, Yokohama, Japan) for flame-ionization detection with helium as the carrier gas. The initial oven temperature was 150°C, and the temperature was increased by 3°C/min until it reached 230°C.

In 10 randomly selected rats per group, blood was collected from the tail vein using a syringe after feeding the test diet for 6, 12, and 18 mo without killing. The blood sample was collected at 14:00 h following 6 h of food deprivation. The serum total cholesterol and triacylglycerol levels were determined by using the commercial Cholesterol E-test Wako and Triglyceride E-test Wako assay kits (Wako Pure Chemical Industries, Ltd., Osaka, Japan). The levels of serum glucose and adiponectin were analyzed by the method reported previously (24).

Table 1. Fatty acid composition of experimental diets.

| Fatty acid  | R1   | R4   | R16  |
|-------------|------|------|------|
| 12:0<sup>1</sup> | 0.1  | 0.1  | 0.1  |
| 14:0        | 0.5  | 0.5  | 0.5  |
| 16:0        | 23.5 | 23.7 | 23.8 |
| 18:0        | 3.6  | 3.5  | 3.4  |
| 18:1        | 42.5 | 42.5 | 42.5 |
| 18:2 (n-6)  | 14.5 | 22.9 | 26.9 |
| 18:3 (n-3)  | 14.2 | 5.7  | 1.7  |
| 20:0        | 0.4  | 0.4  | 0.4  |
| 20:1        | 0.2  | 0.2  | 0.2  |
| 22:0        | 0.2  | 0.2  | 0.2  |
| Others      | 0.3  | 0.3  | 0.3  |

Table 1. Fatty acid composition of experimental diets.

| Fatty acid  | g/100 g total fatty acids | R1   | R4   | R16  |
|-------------|--------------------------|------|------|------|
| S           | 28.5                     | 28.5 | 28.5 |
| M           | 42.9                     | 42.9 | 42.9 |
| P           | 28.6                     | 28.6 | 28.6 |
| S : M : P   | 1.0 : 1.5 : 1.0          | 1.0  | 1.5  | 1.0  |
| n-6         | 14.5                      | 22.9 | 26.9 |
| n-3         | 14.2                      | 5.7  | 1.7  |
| n-6/n-3 ratio | 1.0                      | 4.0  | 16.0 |

<sup>1</sup>Number of carbon atoms:number of double bonds.

S, saturated; M, monounsaturated; P, polyunsaturated.

Four-week-old rats were randomized into three groups (n = 38/group). To determine the fatty acid composition of liver and serum lipids, five rats in each group were killed after consuming the test diet for 4 wk. Other rats (n = 33) were used for the examination of life span. Rats were fed an experimental diet containing test fat with an n-6/n-3 ratio of 1 (R1), 4 (R4), or 16 (R16) while maintaining a constant S : M : P ratio (1 : 1.5 : 1). The test fats used in the experiment were prepared by mixing palm oil, high oleic safflower oil, safflower oil, and linseed oil (Nisshin OilliO Group, Tokyo, Japan). The fatty acid composition of mixed test fats is shown in Table 1. Palm oil consisted of 44.2% palmitic and 39.5% oleic acids, and high oleic safflower oil comprised 75.6% oleic acid. Safflower oil consisted of 73.7% linoleic acid, and linseed oil comprised 50.6% α-linolenic acid. Experimental diets contained the following ingredients (g/kg diet): cornstarch, 499.48; casein, 200; sucrose, 100; cellulose, 50; AIN-93G mineral mix (21). 35; AIN-93 vitamin mix (21), 10; L-cystine, 3.0; choline bitartrate, 2.5; tert-butylhydroquinone, 0.02; and test oil, 100. The diets provided 21.9, 58.3, and 19.8% of energy as fat, carbohydrate, and protein, respectively. Care was taken that the peroxides never exceeded 10 mmol/kg dietary fat. Each group of rats was given free access to the experimental diet and water.

After consuming the test diet for 4 wk, rats (n = 5) for the measurement of the fatty acid compositions of liver and serum lipids were killed by decapitation at 10:00 h following 16 h of food deprivation. Blood was collected to obtain serum. Livers were quickly removed, weighed, and stored at –80°C until analyses. The samples were freeze-dried, and their total lipids were extracted by the method of Folch et al. (22). The fatty acid composition was determined by gas-liquid chromatography after methylation (23). The fatty acid methyl esters were analyzed using a Hewlett-Packard 6890 chromatograph (Palo Alto, CA, USA) with a BPX-70 capillary column (30 m, i.d. 0.32 mm; SGC Japan, Yokohama, Japan) for flame-ionization detection with helium as the carrier gas. The initial oven temperature was 150°C, and the temperature was increased by 3°C/min until it reached 230°C.

In 10 randomly selected rats per group, blood was collected from the tail vein using a syringe after feeding the test diet for 6, 12, and 18 mo without killing. The blood sample was collected at 14:00 h following 6 h of food deprivation. The serum total cholesterol and triacylglycerol levels were determined by using the commercial Cholesterol E-test Wako and Triglyceride E-test Wako assay kits (Wako Pure Chemical Industries, Ltd., Osaka, Japan). The levels of serum glucose and adiponectin were analyzed by the method reported previously (24).

Statistical analyses. All statistical analyses were performed with SPSS for Windows, version 10.0J (SPSS Japan, Inc., Tokyo, Japan). Data are expressed as the mean±SE. Survival rates and mean life spans were analyzed using the Kaplan-Meier test. Surviving rats at the completion of the study (115 wk) were treated asensored events. Other data were analyzed by one-way analysis of variance (ANOVA). Significant differences between the diet groups were determined by Scheffe’s test. All statistical tests were considered significant at p<0.05.

RESULTS

Body weight, food intake, and life span

There were no significant differences in the body weight (Fig. 1) or food intake (Fig. 2) among the 3 groups throughout the study period. The body weight
The serum triacylglycerol levels were significantly lower after feeding the test diet for 6 and 12 mo than in the R1 group, but no significant differences were noted among the 3 groups at 18 mo. The serum total cholesterol level at 12 mo was also significantly lower in the R1 group than in the other 2 groups, and no significant differences were noted among the 3 groups at 18 mo. The serum adiponectin levels of the R1 group than in the other 2 groups, but no significant differences in food intake among the 3 groups throughout the study period. 

To analyze the survival rate and mean life span, we used the data of 33 rats per group. There were no significant differences in the survival rate or mean life span among the 3 groups (Fig. 3). The first dead animal was noted between 66 and 75 wk in all groups. The number of surviving rats at the completion of the study (115 wk) was 2 in the R4 group and 4 each in the R1 and R16 groups.

**Serum analysis**

The serum total cholesterol level after feeding the test diet ingestion and tended to decrease thereafter. At 100 wk of test diet ingestion, there were large changes and variations in the food intake.

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**Serum analysis**

The serum total cholesterol level after feeding the test diet for 6 mo was significantly lower in the R1 group than in the other 2 groups (Table 2). The serum total cholesterol level at 12 mo was also significantly lower in the R1 group than in the other 2 groups, but no significant difference was noted among the 3 groups at 18 mo. The serum triacylglycerol levels were significantly lower after feeding the test diet for 6 and 12 mo.

**Fig. 2.** Food intake in rats fed a diet with an n-6/n-3 ratio of 1 (R1), 4 (R4), or 16 (R16). Data are the means±SE, n=33. There were no significant differences in food intake among the 3 groups throughout the study period.

**Fig. 3.** Survival curves and mean life span in rats fed a diet with an n-6/n-3 ratio of 1 (R1), 4 (R4), or 16 (R16). Data are the means±SE, n=33. There were no significant differences in the survival rate or mean life span among the 3 groups.

**Table 2.** Total cholesterol, triacylglycerol, glucose and adiponectin levels of serum in rats fed a diet with an n-6/n-3 ratio of 1, 4, or 16.

| Group | R1 | R4 | R16 |
|-------|----|----|-----|
| Total cholesterol, mg/dl | | | |
| 6 mo | 135±4 | 152±4 | 162±4 |
| 12 mo | 143±8 | 178±5 | 188±9 |
| 18 mo | 154±14 | 157±20 | 181±15 |
| Triacylglycerol, mg/dl | | | |
| 6 mo | 169±12 | 186±10 | 211±9 |
| 12 mo | 162±16 | 196±18 | 239±17 |
| 18 mo | 173±20 | 163±21 | 149±19 |
| Glucose, mg/dl | | | |
| 6 mo | 134±6 | 128±2 | 134±2 |
| 12 mo | 152±5 | 150±4 | 148±4 |
| 18 mo | 131±5 | 113±3 | 119±4 |
| Adiponectin, ng/mL | | | |
| 6 mo | 3.68±0.17 | 3.50±0.15 | 3.28±0.12 |
| 12 mo | 4.07±0.12 | 3.97±0.25 | 3.85±0.20 |
| 18 mo | 3.18±0.22 | 3.24±0.20 | 3.23±0.22 |

1 Values are the means±SE, n=10/group. Within a row, different superscript letters indicate significant differences between dietary groups (p<0.05).

**Table 3.** Fatty acid composition of liver lipids in rats fed a diet with an n-6/n-3 ratio of 1, 4, or 16 for 4 wk (8-wk-old rats).

| Group | R1 | R4 | R16 |
|-------|----|----|-----|
| Fatty acid | g/100 g total fatty acids | | |
| 16:0<sup>2</sup> | 20.3±1.0 | 22.6±0.7 | 23.5±0.3 |
| 16:1 | 1.6±0.1 | 1.9±0.1 | 1.7±0.1 |
| 18:0 | 13.8±0.7 | 13.8±0.2 | 15.8±0.6 |
| 18:1 | 19.7±0.6 | 20.5±0.9 | 20.4±0.7 |
| 18:2 (n-6) | 13.3±0.2<sup>a</sup> | 15.3±0.3<sup>b</sup> | 14.5±0.3<sup>b</sup> |
| 18:3 (n-3) | 12.5±2.8<sup>ab</sup> | 5.9±1.5<sup>b</sup> | 4.1±1.0<sup>b</sup> |
| 20:4 (n-6) | 9.5±0.6<sup>a</sup> | 13.6±0.1<sup>b</sup> | 15.3±0.7<sup>b</sup> |
| 20:5 (n-3) | 2.4±0.4<sup>a</sup> | 0.0±0.0<sup>a</sup> | 0.0±0.0<sup>a</sup> |
| 22:5 (n-3) | 1.2±0.1<sup>b</sup> | 0.8±0.0<sup>a</sup> | 0.7±0.2<sup>a</sup> |
| 22:6 (n-3) | 5.8±0.3<sup>a</sup> | 5.0±0.1<sup>b</sup> | 3.6±0.2<sup>a</sup> |
| Others | 0.5±0.1 | 0.6±0.0 | 0.5±0.0 |
| S | 34.5±1.7<sup>a</sup> | 37.0±0.8<sup>b</sup> | 39.7±0.7<sup>b</sup> |
| M | 21.3±0.7 | 22.4±1.0 | 22.7±0.8 |
| P | 44.2±2.1<sup>b</sup> | 40.7±1.7<sup>a</sup> | 38.2±0.7<sup>a</sup> |
| n-6 | 22.6±0.7<sup>a</sup> | 29.0±0.5<sup>b</sup> | 29.8±0.6<sup>b</sup> |
| n-3 | 21.6±2.3<sup>b</sup> | 11.7±1.6<sup>a</sup> | 8.4±0.9<sup>a</sup> |
| n-6/n-3 ratio | 1.1±0.1<sup>a</sup> | 2.6±0.3<sup>b</sup> | 3.7±0.4<sup>b</sup> |

1 Values are the means±SE, n=5/group.
2 Number of carbon atoms: number of double bonds.
S. saturated; M. monounsaturated; P. polyunsaturated.
Within a row, different superscript letters indicate significant differences between dietary groups (p<0.05).
Table 4. Fatty acid composition of serum lipids in rats fed a diet with an n-6/n-3 ratio of 1.4, or 16 for 4 wk (8-wk-old rats).1

| Group | R1 | R4 | R16 |
|-------|----|----|-----|
| Fatty acid | g/100 g total fatty acids |    |     |
| 16:0 | 21.7±0.4 | 22.1±0.4 | 21.8±0.4 |
| 16:1 | 1.9±0.1 b | 1.8±0.0 b | 1.6±0.0 a |
| 18:0 | 7.6±0.2 | 7.1±0.2 | 7.4±0.3 |
| 18:1 | 20.7±0.7 | 20.0±0.5 | 18.6±0.6 |
| 18:2 (n-6) | 15.4±0.3 a | 17.8±0.6 b | 18.1±0.2 b |
| 18:3 (n-3) | 3.9±0.1 c | 1.9±0.1 b | 0.4±0.1 a |
| 20:4 (n-6) | 15.2±0.6 a | 21.7±0.4 b | 27.7±1.0 f |
| 20:5 (n-3) | 5.1±0.2 c | 1.2±0.1 b | 0.1±0.1 a |
| 22:5 (n-3) | 1.7±0.1 c | 1.0±0.0 b | 0.4±0.1 a |
| 22:6 (n-3) | 6.1±0.2 c | 4.8±0.1 b | 3.3±0.1 a |
| Others | 0.5±0.0 | 0.7±0.0 | 0.5±0.0 |

| S | 29.9±0.4 | 29.8±0.6 | 29.7±0.3 |
| M | 22.6±0.7 b | 21.8±0.5 d | 20.2±0.6 a |
| P | 47.5±1.0 | 48.4±0.8 | 50.1±0.9 |
| n-6 | 30.6±0.9 a | 39.5±0.7 b | 45.9±0.9 e |
| n-3 | 16.9±0.2 a | 8.9±0.2 b | 4.2±0.1 a |
| n-6/n-3 ratio | 1.8±0.1 a | 4.5±0.1 b | 10.9±0.4 c |

1 Values are the means±SE. n=5/group.
2 Number of carbon atoms:number of double bonds.
S, saturated; M, monounsaturated; P, polyunsaturated.

Within a row, different superscript letters indicate significant differences between dietary groups (p<0.05).

in the R1 group than in the R16 group, but there was no significant difference among the 3 groups at 18 mo. There was no significant difference in the serum glucose level at 6 or 12 mo among the 3 groups, but the level at 18 mo was significantly higher in the R1 group than in the R4 group. There was no significant difference in the serum adiponectin level at 6, 12, or 18 mo among the 3 groups.

Liver fatty acid composition

The 18:2 and 20:4 fatty acid contents were significantly higher in the R4 and R16 groups than in the R1 group (Table 3). The 18:3 and 22:5 contents were significantly higher in the R1 group than in the R16 group. The 20:5 content was significantly higher in the R1 group than in the other 2 groups. The 22:6 content was significantly higher in the R1 and R4 groups than in the R16 group. The PUFA content was significantly higher in the R1 group than in the R16 group. Compared to the R4 and R16 group, the n-6/n-3 ratio in liver lipids was significantly lower in the R1 group.

Serum fatty acid composition

The 18:2 fatty acid content was significantly lower in the R1 group than in the other 2 groups (Table 4). Compared to the R4 group, the 18:3, 20:5, 22:5, and 22:6 contents were significantly higher in the R1 group and significantly lower in the R16 group, while the 20:4 content was significantly lower in the R3 group and significantly higher in the R16 group. Compared to the R4 group, the n-6 fatty acid content was significantly lower in the R1 group and significantly higher in the R16 group, while the n-3 fatty acid content was significantly higher in the R1 group and significantly lower in the R16 group.

DISCUSSION

This is the first study in which the influence of the n-6/n-3 ratio in a diet adjusted to a constant S:M:P ratio on the life span was investigated over the lifetime of Wistar rats. There was no significant difference in the mean life span among rats fed diets with n-6/n-3 ratios of 1.4, and 16, showing that variation of the dietary n-6/n-3 ratio within a range of 1 to 16 does not affect the rat life span.

No consistent findings have been reported with regard to the influence of dietary fat on life span. Du et al. observed that the life span of mice fed a DHA-rich diet was longer than that of mice fed a lard diet abundant in saturated fatty acids and a safflower oil diet rich in linoleic acid (11), while Jolly et al. reported that the life span of a mouse autoimmune model was prolonged in a group fed n-3 fatty acid-rich fish oil, compared to that in a group fed a corn diet (12). However, it has also been reported that the life spans of BHE/cdb rats (a rat non-obese NIDDM and hyperlipidemia model) and senescence-accelerated mice were shorter in animals fed an n-3 fatty acid-rich diet than in those fed a linoleic acid-rich safflower oil diet (13, 14). Ratnayake et al. showed a positive correlation of life span with dietary saturated fatty acids in stroke-prone spontaneously hypertensive rats, but not with mono, n-6, or n-3 unsaturated fatty acids (15). The influences of dietary fatty acids on life span may vary among animal species and disease models.

Lee et al. (17) and Jeffery et al. (18) closely investigated the influence of the dietary n-6/n-3 ratio on blood lipids. They fed diets with a constant S:M:P ratio and various n-6/n-3 ratios to rats for 4 wk and found that the serum triacylglycerol level was significantly lower in rats fed a low- than in those fed a high-n-6/n-3 diet, and the blood cholesterol level after feeding for 6 wk was also significantly lower in rats fed a low- than in those fed a high-n-6/n-3 diet. Similarly, the blood cholesterol and triacylglycerol levels after feeding for 6 and 12 mo were significantly lower in rats fed a low-n-6/n-3 diet in our study. Reduced cholesterol release from the liver in hamsters fed an n-3 fatty acid diet, compared to that in animals fed an n-6 diet, has been reported (25), and the results of another study showed that cholesterol and triacylglycerol release from liver cells cultured in the presence of chylomicron remnants containing abundant n-3 fatty acids was reduced, compared to that from cells cultured with n-6 fatty acids (26). The mechanism of the low-n-6/n-3 diet-induced reduction of the blood lipid level might be related to the reduced cholesterol and triacylglycerol release from the liver.

There have been limited studies on the long-term influence of the dietary n-6/n-3 ratio on blood lipids. Du et al. (27) fed linoleic acid-rich safflower oil or α-linolenic acid-rich perilla oil to 17-wk-old mice for 71 wk
and found that the serum total cholesterol level was significantly lower in the perilla oil group than in the safflower oil group. Fukushima et al. (28) fed n-3 fatty acid-rich perilla oil or n-6 fatty acid-rich borage oil to 24-wk-old rats for 15 wk and found that the blood cholesterol level after feeding the test diet for 4 wk was significantly lower in the perilla oil group than in the borage oil group, but differences between the groups were not significant at 8 or 15 wk of test diet ingestion. Similarly, no significant difference was noted in the blood lipid level after 18 mo of test diet ingestion among the 3 groups in our study, suggesting that the influence of the dietary n-6/n-3 ratio on blood lipids varies depending on the duration or life stage of feeding.

Until now, there has been no study in which the influence of the n-6/n-3 ratio on the blood glucose level was investigated over the study subjects’ lifetime. In the present study, the serum glucose level was significantly higher in the low n-6/n-3 diet group at 18 mo of test diet ingestion, suggesting that affects may differ depending on the life stage of feeding. Fickova et al. (29) observed lower blood cholesterol and triacylglycerol levels in rats fed a low-n-6/n-3 diet for 1 wk, but the serum glucose level was not significantly different between those rats and controls. Ghafourunissa et al. (30) also reported that the blood lipid level was significantly reduced by the ingestion of a low-n-6/n-3 diet for 12 wk, but the plasma glucose level was not. The ingestion of a low-n-6/n-3 diet might increase the blood glucose level, but a longer duration of ingestion may be necessary to observe this effect, compared to the effect on the blood lipid level.

Adiponectin (ADN) is a hormone secreted by adipose tissue that improves glucose metabolism. We (31) and others (32, 33) reported that α-linolenic acid, EPA, and DHA increased ADN in rats and mice, whereas n-6 fatty acids have been shown to inhibit ADN release from primary rat adipocytes. In the present study, we measured the serum ADN level after feeding the test diet for 6, 12, and 18 mo, but there was no significant difference in resulting ADN level among the groups. These results suggest that the dietary n-3 fatty acid content may be more important than the dietary n-6/n-3 ratio. The results also suggest that serum ADN was not related to the increase in the serum glucose level by intake of the low-n-6/n-3 diet in this study.

In summary, we investigated the influence of life-long dietary n-6/n-3 ratio on the span and serum lipid and blood glucose levels. Wistar rats were fed a diet with a dietary n-6/n-3 ratio of 1, 4, or 16 from 4 wk of age until death. There were no significant differences in the survival rate or mean life span among the 3 groups. The levels of serum cholesterol and triacylglycerol were lower in the low n-6/n-3 diet group at 6 and 12 mo. The serum glucose concentration was higher in the low n-6/n-3 diet group at 18 mo. The results of this study suggest that the life span of Wistar rats is not affected even if the ratio of dietary n-6/n-3 changes from 1 to 16.

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