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Plasma Fatty Acid Composition in Men over 50 in the USA and Japan

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Abstract

Background: Coronary heart disease (CHD) rates differ markedly between the US and Japan. Fatty acid profiles have been linked to risk for CHD. Few studies have compared the plasma fatty acid composition, including trans fatty acids, in Japanese and US subjects. **Methods:** Fasting blood samples were taken from healthy older (>age 50) American (n = 76) and Japanese (n = 44) men, and plasma levels of 23 fatty acids were analyzed by gas chromatography and expressed as a percent of total fatty acids. **Results:** As expected, plasma levels of long-chain $\omega 3$ fatty acids (docosahexaenoic and eicosapentaenoic acids, DHA and EPA) were higher in Japanese men and $\omega 6$ fatty acids (e.g., arachidonic acid, AA) were lower compared with American men. Plasma levels of the major industrially-produced trans fatty acids (IP-TFAs; elaidic and linoelaidic acids) were far higher in American men, and levels of the potentially cardioprotective, primarily ruminant-derived trans fatty acid palmitoelaidic acid (POA) were higher in Japanese. Plasma levels of saturated or monounsaturated fatty acids were also higher in the American men. **Conclusion:** There are multiple differences in plasma fatty acid profiles between American and Japanese older men. The higher levels of DHA and EPA, along with the lower levels of the IP-TFAs, are consistent with the markedly lower risk for coronary heart disease in Japan vs. the US.

Keywords

Plasma Fatty Acid Composition

1. Introduction

Coronary heart disease (CHD) is the leading cause of death worldwide, and cer-

tain dietary fatty acids (FAs) are known to play an important role in CHD risk. As such, some FAs could be modifiable risk factors [1]. Higher intakes of industrially-produced trans fatty acids (IP-TFA) [2] and of saturated fatty acids (SFAs) are associated with increased risk for CHD [3] [4], and higher intakes of both the $\omega6$ (n − 6) polyunsaturated fatty acids (PUFAs) and the omega-3 PUFAs are associated with lower risk of CHD [5] [6]. Since estimation of dietary intakes of FAs using questionnaires is challenging (because of out-of-date databases, reliance on memory, poor estimation of portion sizes, etc.) many researchers have begun to measure plasma/blood levels of FAs as more objective biomarkers of exposure. The two general classes of FAs for which biomarkers are most strongly linked with intakes are the PUFAs (especially the omega-3 class) and IP-TFAs. Because risk for CHD is much lower in Japan than in the US [7], we undertook this study to compare the FA profiles in Japanese and American men over the age of 50.

2. Materials and Methods

2.1. Participants

In Japan, we recruited 44 male volunteers older than 50 who were friends and family members of the research team for this study. Exclusion criteria included the use of medications to treat diabetes, hyperlipidemia, hypertension and/or cardiovascular disease (CVD). Smokers were also excluded. The 76 US men were participants in the Chicago Area Sleep Study, a prospective cohort study to examine risk factors for the development of sleep disorders [8]. This cohort excluded men with known sleep disturbances but did not exclude for the chronic conditions excluded in the Japanese cohort. We collected blood samples after an overnight fast, and plasma was isolated for fatty acid analysis. We obtained an informed consent prior to conducting the protocol which had been approved by the Ethical Committee of Showa Women's University and Saiseikai Shibuya Satellite Clinic. The Chicago Area Sleep Study was approved by the Northwestern University Institutional Review Board.

2.2. Analyses of Plasma Samples

Fatty acids levels were measured in plasma obtained from ethylenediamine tetraacetic acid anticoagulated blood samples. Samples were frozen at -80 degrees until analyzed at Omegaquant, LLC (Sioux Falls, SD, USA). After thawing, an aliquot of plasma was combined (1:40 parts) with the methylating mixture (boron trifluoride in methanol [14%], toluene, and methanol [35/30/35v/v]), shaken at 100˚C for 45 minutes. After cooling, 40 parts of both hexane and distilled water were added. After briefly vortexing, the samples were spun to separate layers, and an aliquot of the hexane layer that contained the fatty acid methyl esters was analyzed by gas chromatography as previously described [9].

As shown in Table 1, there are no statistical differences in plasma levels of total cholesterol, triglyceride, HDL-C, LDL-C, and fasting blood glucose levels

Table 1. Backgrounds of various parameters of healthy old men in Japan and USA.

	Japanese(n = 44)	American (n = 76)
Age	62.4 ± 9.6	57.5 ± 4.3
Height (m)	1.68 ± 0.07	1.70 ± 0.1
Weight(kg)	68.8 ± 10.9	74.4 ± 12.1
BMI	24.3 ± 3.2	25.3 ± 3.4
Total choesterol (mg/dL)	209.9 ± 32.3	185 ± 33.5
Triglyceride (mg/dL)	126.4 ± 81.3	118.0 ± 62.8
HDL-C (cholesterol) (mg/dL)	60.9 ± 16.6	54.0 ± 15.7
LDL-C (cholesterol)(mg/dL)	123.8 ± 30.2	107.0 ± 29.6
Fasting blood glucose (mg/dL)	91.7 ± 16.3	97 ± 22.3

Mean ± SD.

between Japanese and American old men.

Statistical Analysis: Students' t test was used for the comparison of two groups and $p < 0.05$ was considered as significant difference. Results are expressed as mean ± SD.

3. Results

The ages of the two cohorts was reasonably similar (Japan, 61 ± 10 and US, 57 ± 5 years), as were the body mass indexes (24.9 ± 3.7 vs 25.1 ± 3.4 kg/m^2). Of the fatty acids that constituted at least 1% of the total in either cohort, those that are significantly higher in the Japanese men than the US men were: palmitic, palmitoleic, arachidic, EPA and DHA. Those that were lower in the Japanese men were: LA, DGLA and AA (Table). The IP-TFAs (elaidic and linoelaidic), although of low abundance in both cohorts, were considerably higher in the US than in Japan. The ruminant-derived trans fatty acid palmitoleic was slightly but significantly higher in Japan.

Table 2 shows that plasma levels of myristic, stearic acids, elaidic acid, linoelaidic acid, gamma linoleic acid, eicosadienoic acid, alpha linoleic acid, eicosadienoic acid, behenic acid, DGLA, AA, docosattraenoic acid, and docosapentaenoic acid (n = 6) are higher in Japanese old men. On the other hand, palmitic acid, palmitoelaidic acid, elaidic acid, LA, linoelaidic acid, gamma linolenic acid, eicosadienoic acid, DGLA, AA, lignoceric acid, docosatetraenoic acid, docosapentanoeic acid, and docosapentaenoic acid (n = 6) are higher in American old men.

4. Discussion

We compared plasma levels of fatty acids between Japanese and American men over 50 years of age. We found, not surprisingly, that levels of EPA and DHA are higher in Japanese than American, and that levels of arachidonic acid were lower. This observation, that higher levels of the long chain omega-3 fatty acids are

Table 2. Comparison of fatty acids profiles between Japanese and American men.

Plasma fatty acids (% of total)	Japanese (n = 44)	US (n = 76)	ss
Myristic	0.7 ± 0.2	0.85 ± 0.34	**
Palmitic	22.3 ± 1.3	21.23 ± 2.13	*
Palmitoelaidic (*trans*)	0.2 ± 0.1	0.14 ± 0.07	**
Palmitoleic	1.9 ± 0.6	1.45 ± 0.80	**
Stearic	7.0 ± 0.7	7.24 ± 0.83	
Elaidic (*trans*)	0.1 ± 0.01	0.61 ± 0.34	**
Oleic	20.0 ± 2.6	19.96 ± 3.34	
Linoelaidic (*trans*)	0.2 ± 0.1	0.35 ± 0.12	**
Linoleic (LA)	26.3 ± 4.0	32.83 ± 5,24	**
Arachidic	0.13 ± 0.04	0.13 ± 0.04	
Gamma Linolenic	0.3 ± 0.1	0.53 ± 0.27	**
Eicosenoic	0.2 ± 0.04	0.15 ± 0.05	
Alpha Linolenic	0.7 ± 0.2	0.72 ± 0.32	
Eicosadienoic	0.2 ± 0.03	0.29 ± 0.07	**
Behenic	0.15 ± 0.02	0.20 ± 0.10	**
Dihomo-gamma Linolenic (DGLA)	1.0 ± 0.2	1.56 ± 0.37	**
Arachidonic (AA)	6.0 ± 1.1	7.38 ± 2.18	**
Eicosapentaenoic (EPA)	2.5 ± 1.3	0.77 ± 0.60	**
Lignoceric	0.22 ± 0.10	0.18 ± 0.07	*
Nervonic	0.33 ± 0.18	0.26 ± 0.10	*
Docosatetraenoic	0.12 ± 0.04	0.26 ± 0.11	**
Docosapentaenoic (n6)	0.14 ± 0.04	0.18 ± 0.08	**
Docosapentaenoic (n3)	0.68 ± 0.26	0.59 ± 0.14	*
Docosahexaenoic (DHA)	5.0 ± 1.5	2.14 ± 0.85	**

ss: statistical significance, *p < 0.05, **p < 0.01.

associated with lower levels of the long chain n-6 fatty acids, is commonly seen since the former replace the latter in cell membranes [10]. The omega-3 fatty acids are found predominantly in oily fish, whereas arachidonic acid (the major long chain n-6 fatty acid) is contained in meats and eggs, and can be synthesized (albeit very slowly) [11]. The differences between the Japanese and the US men in regards to the consumption of these types of foods can help explain these differences in blood levels [12]. The other major finding of this study was the lower levels of IP-TFA in the Japanese vs the US men.

Currently, CHD death rates in Japan are 3× lower for women and 4× lower for men (ages 35 - 74) compared with the US. Among 30 countries for which the American Heart Association provided CHD death rates in its 2017 Statistical Update [7], Japan had the 2nd and 3rd lowest rate (men and women, respectively)

compared to the US at position 21. Sekikawa *et al.* showed in 2014 that that the calcification of the coronary artery was twice in American compared to Japanese men, but the calcification of Hawaian Japanese was similar to that of people on the US mainland [13]. Since Japanese immigrants to the US have increased CHD mortality [14], although still lower than U.S. Whites, it appears that the Japanese (lifestyle, including diet) must be responsible for this difference. The possibility that differences in dietary fatty acid patterns may contribute to this phenomenon is the subject of this report.

We found that the long chain omega-3 fatty acids EPA and DHA were 2 - 3× higher in Japan vs the US. The relationship of fish and dietary omega-3 fatty acids and cardiovascular disease (CVD) has been investigated in numerous studies and comprehensive reviews and recommendations exist. A recent meta-analysis of randomized trials with omega-3 fatty acids [15] did not find a statistically significant reduction in CVD mortality, but it failed to take into account several factors that may have explained this null outcome [16] [17] [18]. Other systematic reviews have reported mortality benefits for $\omega 3$ fatty acids [19] [20], and omega-3 biomarker levels have been strongly associated with risk for fatal CHD in still other meta-analyses [21] [22]. Hence, higher omega-3 levels could at least partly explain the lower CHD risk in Japan.

We also found that IP-trans fatty acids were lower in Japan vs the US. The reported intake of IP-TFA is 75% lower in Japan than in the US, again supporting the observed differences in biomarker levels. Circulating 18:2 trans fatty acids were shown to be most adversely associated with total mortality, mainly due to the increased risk of CVD [23]. It was also positively associated with total mortality and CHD. In a recent study from Germany, total trans fatty acids in erythrocyte membranes were shown to be inversely associated with mortality, but this was mainly driven by the naturally occurring 16:1 trans (trans-palmitoleic acid) [24]. As to relationship between IP-TFA or SFA intakes and CHD mortality, excessive intakes of both had a greater impact on risk for CHD in the US compared with Japan, whereas insufficient intakes of n6 PUFAs had about the same impact on risk in both countries [25].

Our results also indicate that plasma levels of SFAs are higher in American than in Japanese. Saturated fatty acids are considered to be one of the dietary risk factors of CVD, primarily because these fats raise LDL-cholesterol levels. A reduction in SFAs is a core dietary recommendation that has been issued by many health and government organizations to reduce the risk of CVD. Although this difference in SFA plasma levels may also be one of the reasons that Americans have a higher mortality rate for CVD than Japanese, plasma saturated (and monounsaturated) fatty acids are relatively poor markers of dietary SFA intake [26].

An obvious limitation of the present study is the rather small number of participants, especially from Japan. In addition, inclusion criteria differed between the two cohorts, allowing for the inclusion of some individuals with a history of

cardiometabolic diseases in the US cohort but not the Japanese. Since the differences we observed have been reported in larger cohorts [27], our findings were unlikely to have been affected by these limitations.

5. Conclusion

In conclusion, plasma fatty acid profiles in older men from Japan and the US differed in many ways that are consistent with the lower rate of CHD in the former country. Efforts to lower TFA levels and increase EPA+DHA levels may help lower risk for CHD in the US, and current trends in Japan toward a more western diet [28] should be discouraged.

Acknowledgements

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