

東海大学大学院平成 28 年度博士論文

Comparison of eicosapentaenoic acid
concentrations in plasma between
ischemic stroke patients and control
subjects

(虚血性脳卒中患者の血漿エイコサペ
ンタエン酸濃度に関する検討)

池谷 義守

Comparison of eicosapentaenoic acid concentrations in plasma between ischemic stroke patients and control subjects.

This doctoral thesis was written on the basis of the original article published in Nutrition.

I got permissions for the submission of the doctoral thesis to the institutional repository of Tokai University, in addition to reuse of part of my article in my doctoral thesis.

Summary

Objective: N-3 fatty acids, including eicosapentaenoic acid (EPA), prevent ischemic stroke.

However, the clinical importance of EPA for ischemic stroke and its subtype has not been fully

elucidated. **Methods:** In a cross-sectional study, we determined whether n-3 fatty acids were

predictive factors for ischemic stroke. We compared common clinical parameters between 65

ischemic stroke patients and 65 control subjects. The parameters included blood chemistry data;

concentrations of EPA, docosahexaenoic acid (DHA) and arachidonic acid (AA); EPA/AA ratio;

smoking; alcohol intake; fish consumption > 4 times/week; and incidence of underlying

diseases. The comparisons were performed using the Mann-Whitney U test, and multiple

logistic regression analysis was applied to the significant factors in the non-parametric test. We

also applied the same approach to the ischemic stroke subtypes, cardioembolism and

large-artery atherosclerosis. **Results:** In the multiple logistic regression analysis after the

Man-Whitney U test, a lower EPA concentration was one of the significant risk factors for

ischemic stroke, as were a lower body mass index (BMI), lower high-density lipoprotein (HDL)

cholesterol and smoking (sensitivity: 0.846, specificity: 0.831, positive predictive value: 0.833).

In the analysis of subtypes, a lower EPA/AA ratio and a lower BMI were the significant risk

factors for cardioembolism (sensitivity: 0.800, specificity: 0.733, positive predictive value:

0.750). However, large-artery atherosclerosis was not related to the EPA concentration or the EPA/AA ratio. **Conclusions:** In our study, the plasma EPA concentration and the EPA/AA ratio were potential predictive risk factors for ischemic stroke, especially for cardioembolism. Further prospective studies are necessary.

Introduction

The n-3 fatty acids found in fish oil have preventive effects on coronary artery disease and stroke. Iso et al. reported that dietary intake of marine n-3 fatty acids was inversely correlated with the risk of myocardial infarction. Yokoyama et al. demonstrated that eicosapentaenoic acid (EPA) supplementation for hypercholesterolemic patients prevents major coronary events in Japan. A meta-analysis of randomized trials indicated that supplementation with the marine n-3 fatty acids EPA and docosahexaenoic acid (DHA) reduced the rate of death from coronary heart disease.

Some reports have suggested that n-3 fatty acids reduce the risk of cardiac arrhythmias. Mozaffarian et al. indicated that fish intake reduced the incidence of atrial fibrillation. Calò et al. suggested that the supplementation of n-3 fatty acids reduced the incidence of atrial fibrillation after coronary artery bypass graft surgery.

With regard to stroke, many reports have suggested that the consumption of n-3 fatty acid from fish and rapeseed oil has a preventive effect. Furthermore, recent biochemical research demonstrated that n-3 fatty acids have a neuroprotective effect after brain ischemia. A recent randomized controlled trial by Tanaka et al. also demonstrated the beneficial effects of EPA on the recurrence risk of ischemic events in hypercholesterolemic Japanese patients with a history of stroke. In addition, Park et al. demonstrated that a low level of n-3 fatty acids in erythrocytes is a risk factor for hemorrhagic stroke and ischemic stroke. We determined whether the EPA concentration is a predictive factor for ischemic stroke by comparing clinical data, including the EPA concentration and the EPA/arachidonic acid (AA) ratio, between ischemic stroke patients and control subjects. We employed a non-parametric test and analyzed the factors with significant differences through a multiple logistic regression analysis.

Discussion

In this study, the plasma EPA concentration and the EPA/AA ratio were significantly lower in the 65 ischemic stroke patients than in the 65 control subjects. In the multiple logistic regression analyses, a lower EPA concentration was one of the significant risk factors for ischemic stroke, along with lower BMI, lower HDL- cholesterol and smoking. Many reports have previously

suggested that lower HDL- cholesterol and smoking are important risk factors for ischemic stroke. In addition to these well-known risk factors, our cross-sectional study suggests that EPA concentrations influence ischemic stroke. Regarding the subtypes of ischemic stroke, we found that for cardioembolism and large-artery atherosclerosis, the EPA concentration and EPA/AA ratio were significantly lower compared with the controls. In the multiple logistic regression analysis, the EPA/AA ratio, but not the EPA concentration, was correlated with cardioembolism. By contrast, neither the EPA concentration nor the EPA/AA ratio was correlated with large-artery atherosclerosis in the multiple logistic regression analysis. Dyerberg et al. suggested that high levels of EPA and low levels of AA could lead to an antithrombotic state in which an active prostaglandin I_3 and a non-active thromboxane A_3 are formed. Our results suggest that the EPA concentration primarily affects the coagulability of blood itself rather than the mechanisms of thrombus formation on the atherosclerotic vascular surface. In other words, different mechanisms are involved in cardioembolism and large-artery atherosclerosis. Cardioembolism is induced mainly by altered blood coagulation associated with arrhythmia and/or valvular disease. In contrast, large-artery atherosclerosis is associated with arteriosclerotic changes of vascular surface in addition to altered blood coagulation. The result of lower EPA/AA ratios in cardioembolism may suggestive high EPA/AA ratio suppress arrhythmia. The high AUC values

in the ROC curve analysis for cardioembolism (0.818 and 0.831) suggested that a plasma EPA concentration of 69.95 $\mu\text{g/mL}$ or an EPA/AA ratio of 0.465 may be a target value for EPA supplement therapy. Recently, Itakura et al. reported in the Japan EPA lipid intervention study (JELIS) sub-analysis that the risk of major coronary events was significantly decreased in the groups with high (150 $\mu\text{g/mL}$ or more) on-treatment plasma EPA concentrations compared with that in the low (less than 87 $\mu\text{g/mL}$) group. This led to the suggestion that cardioembolism might be prevented in patients with atrial fibrillation by increasing EPA concentrations through EPA supplementation therapy or fish consumption. In the ROC curves analyses of the DHA concentration for total ischemic stroke, large-artery atherosclerosis and cardioembolism, all analyses reveal insubstantial AUC values. DHA is one of the n-3 fatty acid. Molecular structure of DHA is similar to that of EPA. However, DHA concentration was not a predictive factor for ischemic stroke in our study. However, our study is only a cross-section of a limited number of cases. In addition, the plasma samples used to determine fatty acid concentrations were obtained in the subacute phase of the disease, when recovery phase processes including inflammatory and anti-inflammatory were active. As a consequence, the fatty acid levels may have been different from the values immediately before the ischemic stroke. It is therefore important that the findings in this study are verified by further prospective studies.

Lower BMI was a significant relative risk factor in our study not only for ischemic stroke but also for cardioembolism and large-artery atherosclerosis. Rajala et al. indicated that mortality and stroke were increased in elderly people with lower BMI. Renzhei et al. indicated that a lower BMI increased the risk of mortality from all types of stroke in both men and women and from ischemic stroke in women. Further prospective studies of these factors are also needed.

Conclusions

In our study, the plasma EPA concentration and the EPA/AA ratio were potential predictive risk factors for ischemic stroke, especially for cardioembolism. Further prospective studies are necessary.