学 位 論 文 の 要 旨

氏名　　　柴田　直昭

学　位　論　文　名　　　Diversity in the Incidence and Spectrum of Organic Acidemias, Fatty Acid Oxidation Disorders, and Amino Acid Disorders in Asian Countries: Selective Screening vs. Expanded Newborn Screening.

著　　　者　　　名　　　Naoaki Shibata, Yuki Hasegawa, Kenji Yamada, Hironori Kobayashi, Jamiyan Purevsuren, Yanling Yang, Vu Chi Dung, Nguyen Ngoc Khanh, Ishwar C. Verma, Sunita Bijarnia-Mahay, Dong Hwan Lee, Dau-Ming Niu, Georg F. Hoffmann, Yosuke Shigematsu, Toshiyuki Fukao, Seiji Fukuda, Takeshi Taketani, Seiji Yamaguchi

論 文 内 容 の 要 旨

INTRODUCTION

In recent years, mass spectrometric techniques, including gas chromatography-mass spectrometry (GC/MS) and tandem mass spectrometry (MS/MS), have been used for the biochemical diagnosis of inherited metabolic diseases (IMDs) such as organic acidemias (OAs), fatty acid oxidation disorders (FAODs), and amino acid disorders (AAs). Newborn screening (NBS) for OAs, FAODs, and AAs utilizing MS/MS is becoming popular worldwide and is referred to as expanded NBS (ENBS). However, ENBS has yet to be introduced in several Asian countries in which epidemiological data pertaining to IMDs remain limited. Accordingly, Shimane University has provided biochemical IMD diagnostic services using GC/MS and MS/MS for symptomatic patients (defined as selective screening) from several Asian countries, including Vietnam, China, and India, between 2000 and 2015.

We investigated the frequencies of OAs, FAODs, and AAs among Asian countries using our selective screening and ENBS records. Furthermore, we compared the detection rates using selective screening and ENBS in Japan with the aim of reevaluating the target diseases of ENBS.

MATERIALS AND METHODS

Selective Screening for IMDs was performed for symptomatic patients upon request by medical institutes in Japan and other Asian countries, including Vietnam, China, India, Indonesia, Thailand, Mongolia, South Korea, Malaysia, Taiwan, and Turkey. Samples from patients with clinical findings suspected to indicate IMDs, such as metabolic acidosis, ketosis, hyperammonemia, hypoglycemia, lethargy, hypotonia, myopathy-like symptoms, acute encephalopathy, and sudden infant death of unknown cause. If the request included the above symptoms, blood and/or urine samples with the patient's data (e.g., clinical course and administered medication) were sent to Shimane University. Acylcarnitines and amino acids were analyzed in blood samples (dried blood filter papers and/or serum) by MS/MS using in butyl-derivatized specimens. Organic acids were analyzed in urine samples (dried urine filter paper or frozen urine) by GC/MS as previously described. Biochemical diagnoses were based on the results of MS/MS and/or GC/MS by several expert physicians who were familiar with IMDs.

Using these records, we investigated the frequencies of IMDs in Asian countries retrospectively. Additionally, to investigate the detection rate of IMDs using ENBS between Asian countries and Germany (the latter is a representative European country), we obtained nationwide ENBS data from the principal ENBS investigators in Japan, Taiwan, South Korea, and Germany. Furthermore, the incidences of selective screening were compared to those of ENBS in Japan. The study protocol was approved by the Ethics Committee of Shimane University (registration No. 20170920-2).

RESULTS AND DISCUSSION

Among 39,270 patients who underwent selective screening, IMDs were detected in 1,170. Selective screening demonstrated unique characteristics regarding the incidence of OAs in each country. Methylmalonic acidemia (MMA) was most frequently identified in several countries, including Japan (81/377 diagnosed IMDs), China (94/216 IMDs), and India (72/293 IMDs). There were many patients detected MMA in China. It was reported that MMA with homocystinuria is prevalent in China, and it might be attributed to a Chinese population- specific mutation in *MMACHC* gene encoding *Cbl* C (p.W203X). Moreover, β-ketothiolase deficiency (BKTD) was particularly frequent (33/250 IMDs) in Vietnam. The high incidence of BKTD in the Vietnamese population can be attributed to a common Vietnamese-specific *ACAT1* mutation (p.R208X).

ENBS yielded differences in overall IMD incidence by country: 1:8,557 in Japan, 1:7,030 in Taiwan, 1:13,205 in South Korea, and 1:2,200 in Germany. Frequently discovered diseases included propionic acidemia (PPA), 1:41,000 and phenylketonuria (PKU), 1:46,000 in Japan;

3-methylcrotonyl-CoA carboxylase deficiency (MCCD), 1:41,000 and PKU, 1:58,000 in Taiwan; MCCD, 1:111,000 and citrullinemia type I, 1:115,000 in South Korea; and PKU, 1:5000 and medium-chain acyl-CoA dehydrogenase deficiency (MCAD), 1:10,000 in Germany. ENBS also demonstrated unique characteristics regarding the incidence of IMDs in each country. The incidence rate of MCAD deficiency detected in Germany was over 10-fold higher than that of Japanese patients, because common mutation (p.K329E) of *ACADM* was detected in approximately 90% Caucasian patients with MCAD deficiency. The incidence of PKU in Germany was also 10-fold higher than that in Japan and Taiwan. These findings suggest that the incidence rates of IMDs differ between European and Asian populations.

Furthermore, in Japan, selective screening and ENBS yielded respective PPA frequencies of 14.7% and 49.4% among all organic acidemias. This can be attributed to a common Japanese-specific mutation (p.Y435C) of *PCCB*. That is generally associated with mild phenotypes and unlikely to be detected during selective screening. However, no previous report has described natural history of patients with mild PPA. Our results suggested that such diseases could potentially be excluded in the future after determining the natural disease history.

This study had several limitations. During selective screening, the diagnoses of most patients were based mainly on the results of biochemical analysis. There were some cases in which gene analysis should be preferable for final diagnosis. We could not conclude whether the incidence of each IMD was influenced by the genetic background. Indeed, it may be difficult to prove that the high prevalence rates of some diseases could be attributed solely to population-specific common mutations. Nevertheless, the differences in incidence rates from before to after the implementation of ENBS will help other Asian countries to determine which diseases should be included in ENBS panels.

CONCLUSION

Our study identified diverse IMD incidence rates and disease spectra among Asian countries. The spectra of IMDs determined by selective screening differed from those detected by ENBS. The target diseases identified by ENBS are generally extremely rare. Therefore, international collaborative activities such as the current study are important to the clarification of the natural histories of these diseases, to develop diagnostic methods and therapies, and to elucidate genetic backgrounds.