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Carnitine Profile by Tandem Mass Spectrometry and Dialysis Patients

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Abstract

Background: Carnitine deficiency is a common condition in hemodialysis patients. Therefore, abnormalities in fatty acid metabolism and organic acid metabolism are also common in dialysis patients. Tandem mass spectrometry is a standard technique in pediatric and neonatal medicine. However, it could be a new powerful tool in other fields for estimating the state of intracellular fatty acid metabolism. ***Summary:*** Tandem mass spectrometry has recently revealed the relationships between carnitine profile and dialysis patients' anemia, reduced physical function, and survival rate. Fatty acid and organic acid metabolism, which could previously only be evaluated qualitatively, can now be quantitatively assessed. ***Key Message:*** The applications of tandem mass spectrometry are expected to expand not only in the field of dialysis but also in clinical medicine in general. © 2019 S. Karger AG, Basel

Introduction

Carnitine deficiency occurs due to aberrations in carnitine regulation in disorders such as diabetes, sepsis, cardiomyopathy, malnutrition, cirrhosis, and endocrine disorders and with aging [1]. In hemodialysis patients, carnitine is constantly removed due to dietary restrictions and decreased kidney function, which results in carnitine deficiency and reduced L-carnitine biosynthesis [2].

Carnitine has 2 main functions in vivo. Its first function is the promotion of β-oxidation. Its second function is the detoxification of mitochondria by removing cytotoxins derived from organic acids. L-carnitine transports long-chain fatty acids that are absorbed into the cytoplasm from the blood into the mitochondria, adjusts the acyl-CoA: CoA ratio in mitochondria, and eliminates harmful acyl-CoA. Acyl-CoA accumulates in the mitochondria of patients with abnormal organic acid and fatty acid metabolism; therefore, L-carnitine plays important roles in these metabolic processes [3].

The determination of the carnitine profile can aid in estimating the dynamics of acylcarnitine organic acid and fatty acid metabolism in cells. Acylcarnitine comprises an acyl group, such as an organic acid or fatty acid, bonded to free carnitine. When analysis reveals a profile containing acyl groups, it aids understanding of the state of intracellular organic acid and fatty acid metabolism.

Using tandem mass spectrometry, amino acids and acylcarnitine can be efficiently analyzed in a short time in a small amount of blood with high sensitivity.

Tandem Mass Spectrometry

Mass spectrometry is a method of measuring the mass and abundance of charged molecules (ions) during their flight through a vacuum; it detects behavioral differences to determine mass. A mass spectrometer is composed of a sample introduction device, an ionization device, a mass spectrometer, an ion detector, and a computer for controlling the components.

Among the groups that can be measured by a tandem mass spectrometer are various types of acylcarnitine and amino acids. The technology was developed by Millington et al. [4] in the early 1990s. Dried blood spots or serum can be used as samples. It is a highly valued mass screening tool, and tandem mass spectrometry is used instead of the Guthrie method in newborn mass screening in each country.

Factors Influencing Tandem Mass Spectrometry Results

Researchers must be cautious when analyzing tandem mass spectrometry results, as acylcarnitine and amino acid values fluctuate due to various factors [5]. Depending on total parenteral nutrition and peripheral parenteral nutrition, amino acids and components similar to octanoylcarnitine (C8) and dodecanoylcarnitine (C12) may be detected and affect tandem mass analysis measurements.

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In addition, the ingestion of medium-chain triglyceride oil increases the measured values of hexanoylcarnitine (C6), octanoylcarnitine (C8), decanoylcarnitine (C10), and dodecanoylcarnitine (C12). Furthermore, some antibiotics cause increases in acylcarnitine levels. For example, antibacterial drugs containing pivoxil increase ovalerylcarnitine (C5) values [6].

If patients have a history of blood transfusion within the past 3 months, their measurements must be carefully evaluated. Red blood cells lose nuclei and mitochondria as they mature from erythroblasts but maintain their levels of acylcarnitine and free carnitine. Erythrocyte lifespan is usually approximately 3 months. Host and guest erythrocytes retain the acylcarnitine produced by metabolism as erythroblasts. In particular, 3-hydroxyisovalerylcarnitine (C5-OH) accumulates in red blood cells due to its difficulty exiting the cells, so it correlates with erythrocyte lifespan. Therefore, blood transfusion affects the measured value of 3-hydroxyisovalerylcarnitine (C5-OH).

Dialysis Patients and Carnitine Profiling

There have been numerous reports with differing views on the efficacy of L-carnitine therapy in hemodialysis patients. The findings include improved cardiac function [7–9], alleviation of muscle spasms [10, 11], effects on renal anemia [12, 13], anti-arteriogenic effects [14], and inhibition of dialysis hypotension [15] caused by L-carnitine therapy. The mechanisms of action have only been qualitatively estimated based on the mechanism of L-carnitine action and have not been quantitatively evaluated for abnormal organic and fatty acid metabolism.

However, using tandem mass spectrometry, negative correlations have been reported between acylcarnitine chain length and Short Form-36 physical composite score, sit-to-stand count, and 6-min walk distance [16]. In our study using tandem mass spectrometry, the erythropoiesis-stimulating agent resistance index positively correlated with long-chain C18 acylcarnitine and negatively correlated with short-chain C5-OH acylcarnitine [17]. In addition, 4-year all-cause mortality negatively correlated with the acetylcarnitine/(palmitoylcarnitine+octadecenoylcarnitine; C2/[C16+C18:1]) ratio [18]. We also found that amelioration of the impaired β-oxidation state after L-carnitine administration may improve prognosis. In fact, the C2:(C16+C18:1) ratio of the L-carnitine administration group ($n = 8$) was 124.0 ± 48.2 and only 42.8 ± 15.3 in the control group ($n = 114$; Student t test, $p < 0.001$). The ability to quantitatively evaluate carnitine has confirmed the possibility of improving β-oxidation through carnitine administration.

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Conclusion

Tandem mass spectrometry is a powerful method of identifying and measuring minor components in living bodies. It is already used as a standard technique in pediatric and neonatal medicine. In other fields, tandem mass spectrometry is a potent new tool that can help to estimate the state of intracellular fatty acid metabolism. It is facilitating the discovery of novel findings in the dialysis field. In the future, applications of tandem mass spectrometry are expected to expand for dialysis and other clinical medicine fields.

Disclosure Statement

The authors declare that they have no conflicts of interest to disclose.

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References

1 Flanagan JL, Simmons PA, Vehige J, Willcox MD, Garrett Q: Role of carnitine in disease. Nutr Metab 2010;7:30.
2 Fornasini G, Upton RN, Evans AM: A pharmacokinetic model for L-carnitine in patients receiving haemodialysis. Br J Clin Pharmacol 2007;64:335–345.
3 Vaz FM, Wanders RJ: Carnitine biosynthesis in mammals. Biochem J 2002;361:417–429.
4 Millington DS, Kodo N, Norwood DL, Roe CR: Tandem mass spectrometry: a new method for acylcarnitine profiling with potential for neonatal screening for inborn errors of metabolism. J Inherit Metab Dis 1990;13:321–324.
5 Clinical and Laboratory Standards Institute: Newborn Screening by Tandem Mass Spectrometry (ed 2). CLSI document NBS04. Wayne, The Clinical and Laboratory Standards Institute, 2017.
6 Abdenur JE, Chamoles NA, Guinle AE, Schenone AB, Fuertes AN: Diagnosis of isovaleric acidaemia by tandem mass spectrometry: false positive result due to pivaloylcarnitine in a newborn screening programme. J Inherit Metab Dis 1998; 21:624–630.
7 van Es A, Henny FC, Kooistra MP, Lobatto S, Scholte HR: Amelioration of cardiac function by L-carnitine administration in patients on haemodialysis. Contrib Nephrol 1992;98:28–35.
8 Sakurabayashi T, Miyazaki S, Yuasa Y, Sakai S, Suzuki M, Takahashi S, Hirasawa Y: L-carnitine supplementation decreases the left ventricular mass in patients undergoing hemodialysis. Circ J 2008;72:926–931.
9 Higuchi T, Abe M, Yamazaki T, Okawa E, Ando H, Hotta S, Oikawa O, Kikuchi F, Okada K, Soma M: Levocarnitine improves cardiac function in hemodialysis patients with left ventricular hypertrophy: a randomized controlled trial. Am J Kidney Dis 2016;67:260–270.
10 Ahmad S, Robertson HT, Golper TA, Wolfson M, Kurtin P, Katz LA, Hirschberg R, Nicora R, Ashbrook DW, Kopple JD: Multicenter trial of L-carnitine in maintenance hemodialysis patients. II. Clinical and biochemical effects. Kidney Int 1990; 38:912–918.
11 Sakurauchi Y, Matsumoto Y, Shinzato T, Takai I, Nakamura Y, Sato M, Nakai S, Miwa M, Morita H, Miwa T, Amano I, Maeda K: Effects of L-carnitine supplementation on muscular symptoms in hemodialyzed patients. Am J Kidney Dis 1998; 32:258–264.

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12 Hurot JM, Cucherat M, Haugh M, Fouque D: Effects of L-carnitine supplementation in maintenance hemodialysis patients: a systematic review. J Am Soc Nephrol 2002;13:708–714.
13 Maruyama T, Higuchi T, Yamazaki T, Okawa E, Ando H, Oikawa O, Inoshita A, Okada K, Abe M: Levocarnitine injections decrease the need for erythropoiesis-stimulating agents in hemodialysis patients with renal anemia. Cardiorenal Med 2017;7:188–197.
14 Higuchi T, Abe M, Yamazaki T, Mizuno M, Okawa E, Ando H, Oikawa O, Okada K, Kikuchi F, Soma M: Effects of levocarnitine on brachial-ankle pulse wave velocity in hemodialysis patients: a randomized controlled trial. Nutrients 2014;6:5992–6004.
15 Kudoh Y, Aoyama S, Torii T, Chen Q, Nagahara D, Sakata H, Nozawa A: Hemodynamic stabilizing effects of L-carnitine in chronic hemodialysis patients. Cardiorenal Med 2013;3:200–207.
16 Murphy WJ, Steiber A, Connery GC, Carder J, Spry L, Hoppel C: Altered carnitine metabolism in dialysis patients with reduced physical function may be due to dysfunctional fatty acid oxidation. Nephrol Dial Transplant 2012;27:304–310.
17 Kamei D, Tsuchiya K, Nitta K, Mineshima M, Akiba T: Association between resistance to erythropoiesis-stimulating agents and carnitine profile in patients on maintenance hemodialysis. Nephrology (Carlton) 2017, Epub ahead of print.
18 Kamei Y, Kamei D, Tsuchiya K, Mineshima M, Nitta K: Association between 4-year all-cause mortality and carnitine profile in maintenance hemodialysis patients. PLoS One 2018;13:e0201591.

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