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Essential oils can cause false-positive results of medium-chain acyl-CoA dehydrogenase deficiency

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ABSTRACT

Newborn screening is a public health care program worldwide to prevent patients from critical illness or conditions. Tandem mass spectrometry allows multiplex, inexpensive, and rapid newborn screening. However, mass spectrometry used for newborn screening to date is not able to separate peaks of compounds with similar *m/z*, which could lead to false-positive results without additional second-tier tests, such as fragmentation. We experienced three neonatal cases with high levels of markers, octanoylcarnitine and octanoylcarnitine/decanoylcarnitine ratio used to pick up possible cases of medium-chain acyl-CoA dehydrogenase (MCAD) deficiency. The babies were born consecutively in a maternity hospital. Their second acylcarnitine profiles were normal, and the genetic tests for *ACADM* were negative. Analysis of samples extracted from their first Guthrie cards where blood was not stained also showed peaks equivalent to octanoylcarnitine and decanoylcarnitine, indicating contamination. Environmental surveillance in the maternity ward suggested that essential oils used there might contain the contaminated compound. LC-HRMS/MS and *in silico* analysis revealed that false-positive results might be due to contamination with the essential oils in Guthrie cards, and causal agents were sphinganine (d17:0) and 2-[2-hydroxyethyl(pentadecyl)amino]ethanol. Thus, health care providers should be cautioned about use of essential oils when collecting blood samples on Guthrie cards. False-positive results can waste costly social resources and cause a physical and psychological burden for children and parents.

1. Introduction

Newborn screening (NBS) is a healthcare program to identify patients with inborn errors who need medical care as soon as possible after birth to prevent irreversible damage or metabolic crisis [1]. The NBS program saves lives and intellectual abilities of children in a cost-effective manner and with high-throughput and accuracy. Liquid chromatography-tandem mass spectrometry (LC-MS/MS) is critical to this process and is used to investigate a wide variety of disorders for NBS. Medium-chain acyl-CoA dehydrogenase (MCAD) deficiency (MIM# 201450) is one of the fatty acid oxidation disorders (FAODs). It occurs in 1/100,000 Japanese newborns; an incidence 10-fold less than in Caucasian [2]. MCAD deficiency can cause a decompensated metabolic state *via* febrile and intercurrent illness combined with energy deficiency or vomiting [3,4]. The metabolic attack may lead to death. Avoiding excessive fasting or catabolic episodes can prevent irreversible damage, and NBS for MCAD deficiency reduces morbidity and mortality [5,6].

Octanoylcarnitine (C8) is a universal NBS marker for MCAD deficiency. In addition to MCAD deficiency, elevated levels of C8 can be caused in other ways. Medium-chain triglyceride supplementation and valproate increase levels of C8 [7]. Elevated C8 may be seen in neonates who show heterozygous pathogenic variant or premature infants [8]. 2-Ethylhexanoic acid in common plasticizers is reported to induce false-

Abbreviations: NBS, newborn screening; LC-MS/MS, liquid chromatography-tandem mass spectrometry,; MCAD, medium-chain acyl-CoA dehydrogenase,; FAOD, fatty acid oxidation disorder,; C8, octanoylcarnitine,; C10, decanoylcarnitine,; LC-HRMS/MS, liquid chromatography-high resolution-tandem mass spectrometry.

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positive results of C8 in two neonates treated with extracorporeal membrane oxygenation [9]. However, other causes have not been previously reported to induce false-positive results for MCAD deficiency.

In this report, we describe three false-positive cases of MCAD deficiency which occurred in births at the same maternity hospital.

2. Materials and methods

2.1. Diagnostic criteria for MCAD deficiency in Japan

C8 and octanoylcarnitine/decanoylcarnitine (C8/C10) ratio is used to screen for MCAD deficiency in Japan. MCAD deficiency is suspected in cases of elevated C8 and C8/C10 ratio on NBS, and further tests are then performed. MCAD deficiency is also considered in cases of non-ketotic hypoglycemia or acute encephalopathy with unknown causes, especially in patients who were born before 2014 in which Japan initiated NBS for MCAD deficiency [10] and show elevated C8 and C8/C10 ratio with blood acylcarnitine analysis. The diagnosis is confirmed if at least one of the enzyme activity assay, *in vitro* probe assay, immunoblot assay, or genetic analysis shows a disease-specific result, such as other FAODs [11].

2.2. Sample preparations

Dried blood spot (DBS) samples were collected from the infants when they were 4–5 days old and transferred to us with the informed consent of their parents. The Guthrie cards used in our cases were manufactured by Toyo Roshi Co., Ltd. (Tokyo, Japan). Essential oils used in the maternity hospital, *Lavandula angustifolia* ssp. *angustifolia* (lavender), *Melaleuca alternifolia* (tea tree), and *Cupressus sempervirens* (cypress) were purchased from Pranarom International (Ghislenghien, Belgium). Guthrie cards were stained with essential oils and left for 24 h at room temperature in a fume hood. Then, 3-mm punch samples of the cards were used to investigate the components of the essential oils. Extraction used 100 μL 100% methanol (FUJIFILM-Wako pure chemicals Co. Ltd., Osaka, Japan) with vortexing and sonication in a water bath for 10 min. MS-FINDER software [12] was used to predict the molecular formula from the pattern of mass spectral peaks and to identify the molecular structure. Reference standard material was purchased from Cayman Chemical (Sphinganine (d17:0), Ann Arbor, MI) and Angene International Limited (2-[2-hydroxyethyl(pentadecyl)amino]ethanol, Nanjing, China).

2.3. Mass spectrometric analysis

MS/MS analysis used a TQD (Waters, Manchester, UK). Precursor ion (*m/z*), fragment ion (m/z), Dwell (s), Cone (V), Collision (V) of C8 and C10 were set to 288.1 or 316.1, 85, 0.050, 40 or 37, 22, respectively. Liquid chromatography-high resolution MS/MS (LC-HRMS/MS) analysis was performed using an Orbitrap Fusion (Thermo Fisher Scientific, San Jose, CA, USA), and a Vanquish UHPLC system (Thermo Fisher Scientific). HRMS and HRMS/MS were analyzed in positive ion mode. Spray voltages were set at 3500 V. Sheath gas (arb), auxiliary gas (arb), sweep gas (arb), ion transfer tube temperature (°C), vaporizer temperature (°C), *m/z* range, resolution at HRMS, resolution at HRMS/MS, and higher collision-induced dissociation energy (%) were set to 50, 10, 0, 275, 350, 200–1500, 500,000, 500,000, and 50%, respectively. Formic acid/water (0.1:100, *v*/v) and formic acid/acetonitrile (0.1100, v/v) were used as mobile phases A and B, respectively. The ratio of mobile phase A/B was set at 1:1. Flow rate was 0.3 mL/min. Injection volume was set at 1 μL.

3. Results

3.1. Case descriptions of three false-positive results

Case 1 was a female with a birth weight of 3152 g who born at 39 weeks' gestation by spontaneous delivery without any complications. She was breastfed and showed no adverse perinatal events or abnormalities. Blood sampling for NBS was performed at the age of 4 days.

Case 2 was a female born by spontaneous delivery at 39 weeks' gestation with a birth weight of 3426 g. There were no complications during gestation and delivery. She was breastfed and no adverse perinatal events or abnormalities were noted. Her blood sample for NBS was collected at the age of 5 days.

Case 3 was a male with a birth weight of 3484 g born at 39 weeks' gestation by spontaneous delivery without any complications. He was mainly breastfed but never fed formula containing medium-chain triglycerides. His blood sampling for NBS was performed at the age of 4 days. The next day, he had a fever, and his weight decreased by 9.4% compared with his birth weight. He was hospitalized and worked up in a general hospital. The finding of metabolic decompensation was not observed because the results did not show hypoglycemia, liver enzyme elevation, or hyperammonemia. The blood analysis showed no elevated white blood cells or C-reactive protein levels, and the culture results for blood and urine were negative. Serum acylcarnitine analysis at the age of 5 days did not reveal any elevation of C8 or C8/C10 (0.08 μM or 0.57, respectively). His body temperature normalized without any treatment when he was 9 days old.

These three cases were born in the same maternity hospital and over a 2-week period. They were referred to our hospital at 9–13 days after birth for second tests because the results of their C8 levels and C8/C10 ratios of their first Guthrie card tests were elevated. However, their second acylcarnitine profiles in using DBS were normal (Table 1). Genetic tests showed no pathogenic variants in *ACADM* genes (NM_000016.5). They appeared to not be cases of MCAD deficiency but false-positive cases because their acylcarnitine profiles were similar and they were born in the same maternity hospital within 2 weeks.

3.2. LC-HRMS/MS analysis revealed the initial Guthrie cards contained agents with almost equivalent m/z but not identical MS/MS patterns to octanoylcarnitine or decanoylcarnitine

Essential oils (lavender, tea tree, and cypress) were mixed, vaporized, and diffused to provide a healing effect to postpartum mothers in

Table 1
Acylcarnitine profiles of the three false-positive cases using dried blood spots.

	Case 1	Case 2	Case 3	Control (N = 2827)
Initial levels	(age 4 days)	(age 5 days)	(age 4 days)	
C2 (μmol/L)	16.40	16.91	20.42	18.146 ± 5.715
C6 (μmol/L)	0.03	0.01	0.02	0.054 ± 0.024
C8 (μmol/L)	0.35	0.34	0.42	0.028 ± 0.009
C10:1 (μmol/L)	0.06	0.05	0.09	0.056 ± 0.021
C10 (μmol/L)	0.17	0.15	0.20	0.088 ± 0.043
C8/C10 ratio	2.06	2.27	2.10	0.629 ± 0.180
Second levels	(age 9 days)	(age 11 days)	(age 13 days)	
C2 (μmol/L)	10.16	13.50	8.07	
C6 (μmol/L)	0.02	0.02	0.03	
C8 (μmol/L)	0.05	0.05	0.06	
C10:1 (μmol/L)	0.04	0.05	0.07	
C10 (μmol/L)	0.05	0.08	0.13	
C8/C10 ratio	1.00	0.63	0.46	

The cutoff value for C8 level and C8/C10 ratio for MCAD deficiency is >0.3 μM and > 1.4, respectively.

the room at the maternity hospital where blood was sampled for NBS. This situation prompted us to consider the possibility that the essential oils may have volatilized and become absorbed into the Guthrie cards, thereby causing false-positive results. LC-MS/MS analysis showed that C8 and C10 levels were almost the same in extractions of blood-stained and unstained parts of their first Guthrie cards. In contrast, analysis of new Guthrie cards did not show a detectable peak for C8 or C10 (Fig. 1). Thus, their first Guthrie cards were contaminated by some source that led to a false-positive result. LC-HRMS/MS, which has high resolution performance of mass accuracy at sub-ppm levels, unlike triple-quadrupole mass spectrometry used in NBS, revealed two materials with *m/z* of 288.2893 and 316.3206, which were almost equivalent to that of C8 (*m/z* of 288.2) and C10 (*m/z* of 316.3), respectively, although they were not detected in all essential oils (Fig. 2). However, their mass

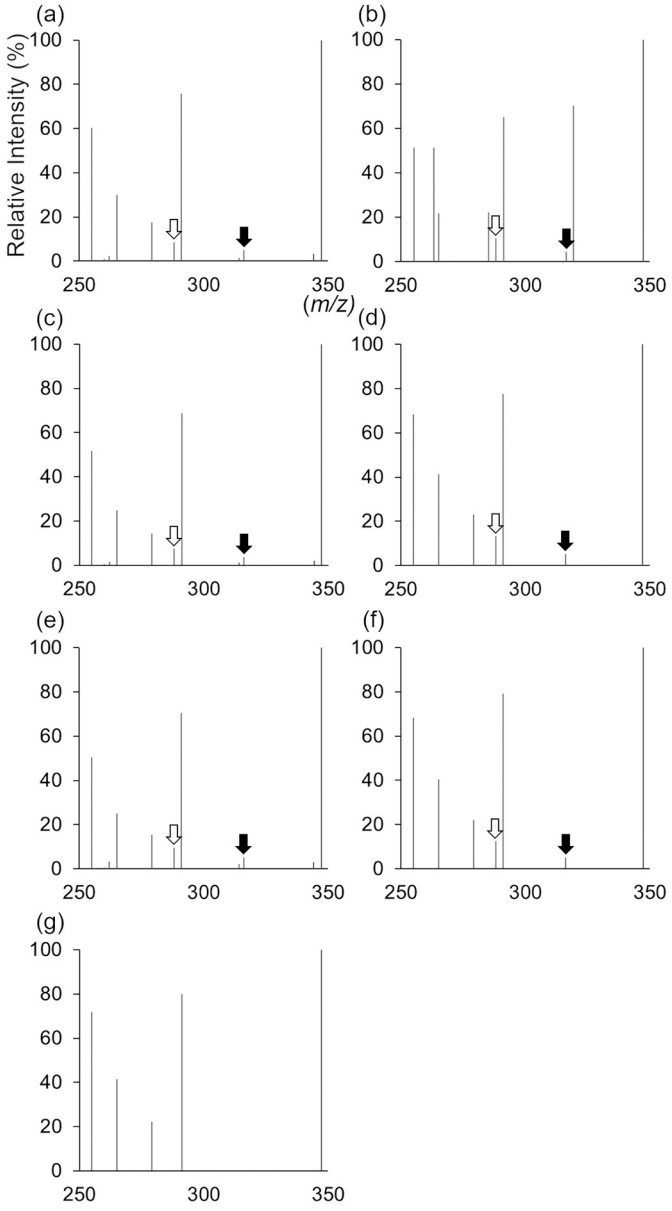

Fig. 1. LC-MS/MS spectra of blood-stained and non-blood-stained portions of the first Guthrie card. (a, c, and e) Blood-stained portions of the first Guthrie card in Cases 1, 2, and 3, respectively. (b, d, and f) Non-blood-stained portions of the first Guthrie card in Cases 1, 2, and 3. respectively (g) Sample taken from a new Guthrie card. The open and closed arrows indicate the peaks of C8 (*m/z* of 288) and C10 (*m/z* of 316), respectively. Abbreviation: LC-MS/MS, liquid chromatography-tandem mass spectrometry.

spectra were not identical to octanoylcarnitine or decanoylcarnitine because the product ion from acylcarnitine species with a monoisotopic mass of 85.02895 [13], which should appear as a peak of *m/z* 84.02 in our experiment, was not detected (Supp. Table 1).

3.3. LC-HRMS/MS and in silico analysis in essential oils shows that sphinganine (d17:0) and 2-[2-hydroxyethyl(pentadecyl)amino]ethanol with monoisotopic mass equivalent to C8 and C10, respectively, are candidate materials for causing false-positive results

Surveilling items placed in the maternity hospital, essential oils were suspected as the source of contamination. Three kinds of essential oils, lavender, tea tree, and cypress, were used to relax family members. First, we prepared the Guthrie cards to which essential oils were added and directly attached. Using their first Guthrie cards and the Guthrie cards stained with the essential oils, LC-HRMS/MS analysis and *in silico* analysis with MS-FINDER [12] identified candidate compounds, sphinganine (d17:0) and 2-[2-hydroxyethyl(pentadecyl)amino]ethanol. LC-HRMS/MS analysis with standard reference materials produced a fragmentation pattern consistent with sphinganine (d17:0) and 2-[2-hydroxyethyl(pentadecyl)amino]ethanol that were almost identical to the peaks to C8 and C10 (Fig. 3). Moreover, when essential oils were volatilized and stained into Guthrie cards, LC-HRMS analysis detected *m/z* 288.2893 in cypress and did not detect *m/z* 316.3206 in all essential oils. However, LC-HRMS/MS analysis of *m/z* 288.2893 and 316.3206, respectively, revealed the same fragment patterns in all essential oils (Fig. 4). MS-FINDER again indicated these MS/MS ion patterns of *m/z* 288.2893 and 316.3206 were consistent with sphinganine (d17:0) and 2-[2-hydroxyethyl(pentadecyl)amino]ethanol, respectively. Guthrie cards at the maternity hospital had been kept in an open file for years. After the occurrence of the false-positive results, the Guthrie cards were replaced with new ones in a closed container, and no additional false-positive cases have been identified in the maternity hospital.

4. Discussion

We examined sequential positive cases at one maternity hospital that reported MCAD deficiency. These cases were false-positive based on the results of genetic tests and sequential acylcarnitine analysis. LC-HRMS/MS analysis showed that the essential oils had some ingredients that produced peaks equivalent to C8 and C10, indicating that these components of essential oils would vaporize and stain Guthrie cards. Sphinganine (d17:0) and 2-[2-hydroxyethyl(pentadecyl)amino]ethanol in essential oils caused peaks that have *m/z* almost same to C8 and C10. To the best of our knowledge, this is the first report to demonstrate that volatile substances could cause false-positive results in NBS. Health care providers should avoid collecting DBS samples in the room where aromatherapy is used.

False-positive results of NBS should be avoided. Guardians can be exposed to unnecessary stress by such results [14,15],. Perinatal stress of mothers could affect neuronal and behavior development of the baby [16]. Further, medical investigation to rule out the disease consumes resources of time and money [17]. FAODs are sometimes difficult to diagnose due to decreased acylcarnitine levels in anabolic conditions [18–20]. Patients with MCAD deficiency tend to have elevated C8, C10, and C8/C10 ratios throughout their life [21], whereas C8 and C10 levels could decrease in a time-series, especially in equivocal cases [22]. Certainly, aromatherapy may be beneficial for postpartum physiological and psychological states [23]. For example, linalool, one of the main components of lavender, may have anxiolytic effects [24] through modulation of acetylcholine release at the neuromuscular junction [25]. Thus, medical care providers involved in NBS should be careful in handling Guthrie cards to prevent false-positive results due to contamination of essential oils. The methods of storage of Guthrie cards are important because no false-positive cases have been found in the hospital after the storage containers for Guthrie cards were replaced with

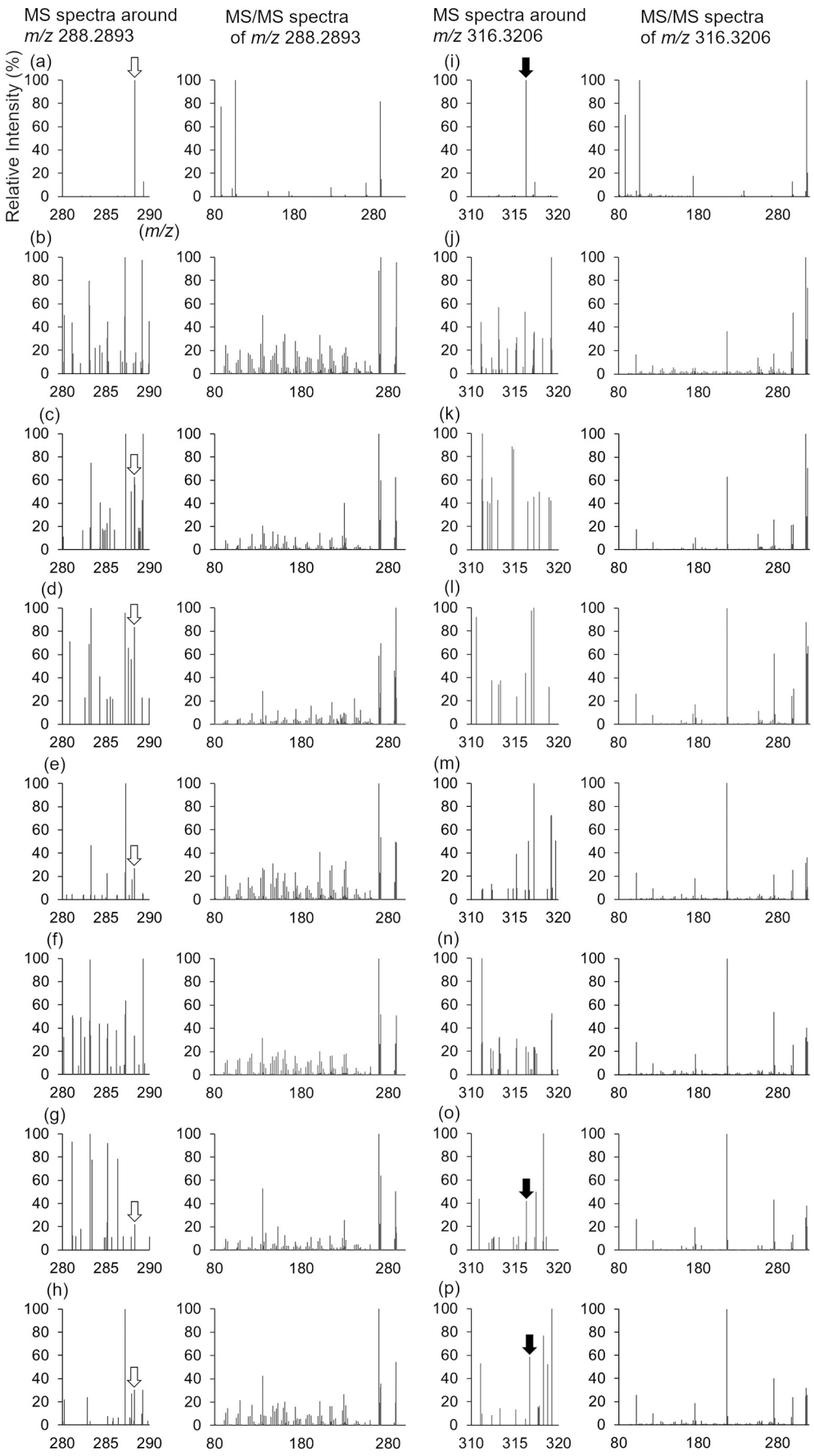

Fig. 2. LC-HRMS/MS spectra of a non-blood-stained portion of the first Guthrie cards compared with Guthrie cards stained with the essential oils from *Lavandula angustifolia* ssp. *angustifolia* (lavender), *Melaleuca alternifolia* (tea tree), and *Cupressus sempervirens* (cypress). (a, i) Non-blood-stained portion of the first Guthrie cards. Guthrie cards were stained with essential oils as follows: (b, j) lavender, (c, k) tea tree, (d, l) cypress, (e, m) lavender and tea tree, (f, n) lavender and cypress, (g, o) tea tree and cypress, and (h, p) lavender, tea tree, and cypress. The columns from left to right are MS spectra around *m/z* 288.2893, MS/MS spectra of *m/z* 288.2893, MS spectra around *m/z* 316.3206, and MS/MS spectra of *m/z* 316.3206. The open and closed arrows indicate the peaks of *m/z* 288 and 316, consistent with C8 and C10, respectively. Abbreviations: LC-HRMS/MS, liquid chromatography-high resolution tandem mass spectrometry; MS, mass spectrometry; MS/MS, tandem mass spectrometry.

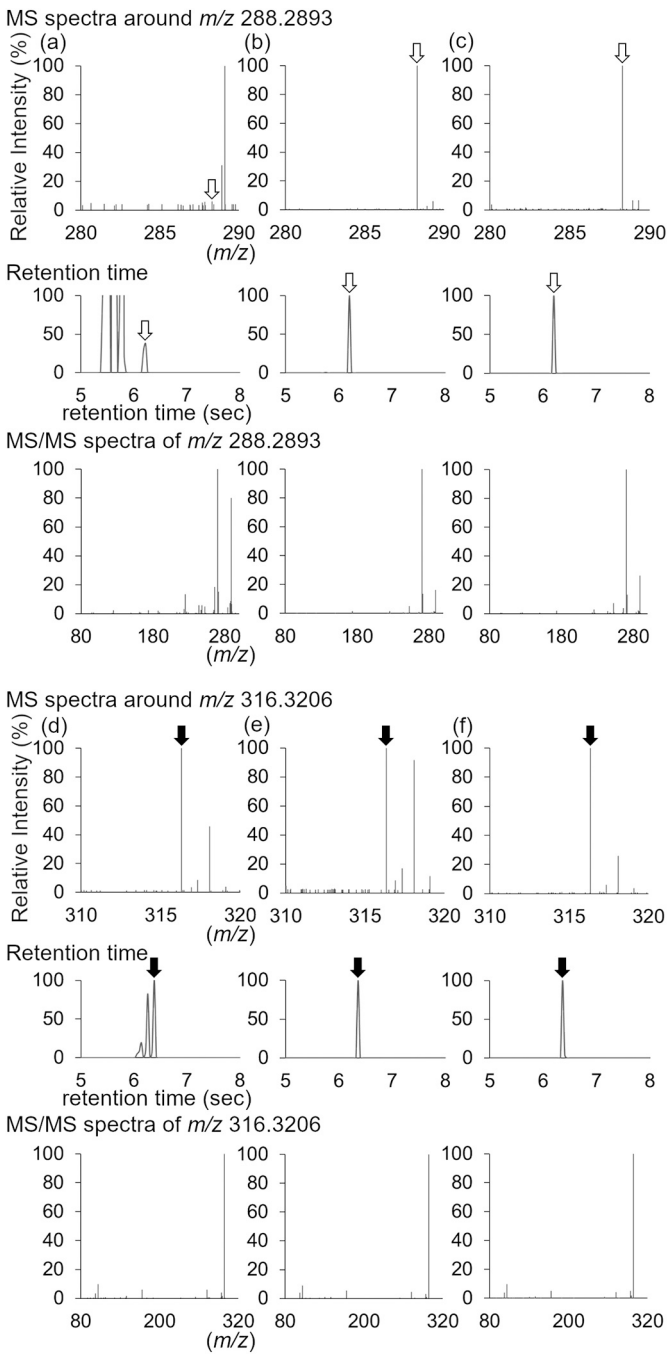

Fig. 3. LC-HRMS/MS analysis of standard reference materials. Upper, middle, and lower panels of each figure show MS, retention time, and MS/MS analysis. Samples are as follows: (a, d) non-blood-stained portion of the first Guthrie cards, (b) sphinganine (d17:0), (e) 2-[2-hydroxyethyl(pentadecyl)amino] ethanol, and (c, f) mixture of sphinganine (d17:0) and 2-[2-hydroxyethyl(pentadecyl)amino]ethanol. (a, b, c) and (d, e, f) were focused on *m/z* 288.2893 and 316.3206, respectively. The open and closed arrows indicate the peaks of *m/z* 288 and 316, which were consistent with C8 and C10, respectively. Retention times of respective peaks matched exactly. Abbreviations: LC-HRMS/MS, liquid chromatography-high resolution tandem mass spectrometry; MS, mass spectrometry; MS/MS, tandem mass spectrometry.

closed containers.

Various efforts to reduce false-positive cases in NBS have been made to date. MS/MS analysis can simultaneously identify many molecules with high accuracy but is not able to discriminate compounds with essentially the very similar *m/z* spectra. For instance, isovalerylcarnitine or 2-methylbutyrylcarnitine, which are markers of isovaleric acidemia, could not be distinguished from pivaloylcarnitine, which is a metabolite of pivalic ester or pivalic acid derivatives [26]. A second-tier test should resolve this problem and allow a correct and prompt decision to be taken [13], although this is time- and effort-consuming because exchanging and setting up the column hampers its use as a practical second-tier test. LC-HRMS/MS also has extremely high resolution capable of differentiating molecules with similar *m/z*; however, its routine use is currently not practical because of the cost. Additionally, it is intriguing that LC-MS/MS in NBS recognized the existence of characteristic ions of acylcarnitine, whereas LC-HRMS/MS confirmed that there were no such ions that had a monoisotopic mass 85.02895. One speculation is that differences in mass accuracy and/or resolution between the mass spectrometers induced the false results. The mass spectrometer used in NBS may have misidentified another compound with an *m/z* similar to that of acylcarnitine-derived ions as acylcarnitine-derived ions. Although exome or genome-wide sequencing could resolve these problems, genetic testing alone cannot identify the pathogenicity of a novel variant. A combination of results reflecting the stages of various biological phenomena can provide clues to judge whether they are truly positive or negative. The C8/C10 ratio is a useful marker to differentiate between mild and severe types of MCAD deficiency [27]. Furthermore, enzyme activity analysis currently plays an important role in assessing metabolic ability [2,28]. Novel techniques could pave the road, such as a report demonstrating the reduction of false-positive cases employing Random Forest machine learning using NBS results [29]. Further studies are necessary to establish a more accurate pipeline in NBS.

Sphinganine (d17:0) and 2-[2-hydroxyethyl(pentadecyl)amino] ethanol would have caused the false-positive results of MCAD deficiency by vaporizing essential oils, although these have not been reported as ingredients in essential oils. 2-[2-hydroxyethyl(pentadecyl) amino]ethanol, candidate for false-positive results of C10, was previously reported to as a non-volatile molecule in baby bottles [30]. Baby bottles would not cause contamination in our cases since little opportunity exists for bottles and Guthrie cards to come into intense contact. Sphinganine (d17:0), would cause a false *m/z* peak for C10, is a sphingolipid and an intermediate in *de novo* synthesis ceramide. C17 bases sphinganine is mainly found in nematodes, not mammalians [31]. No report was found to indicate that essential oils contain sphinganine (d17:0), while plants might possibly produce C17 bases sphinganine [32]. We cannot absolutely exclude the involvement of other usual instruments or environmental compounds. Another limitation is that we could not determine how much essential oils would cause false-positive results because quantification of vaporized and attached sphinganine (d17:0) and 2-[2-hydroxyethyl(pentadecyl)amino]ethanol is difficult. Further, not all essential oils showed peaks at *m/z* 288.2893 and 316.3206 in LC-HRMS analysis, whereas LC-HRMS/MS analysis confirmed the presence of sphinganine (d17:0) and 2-[2-hydroxyethyl (pentadecyl)amino]ethanol in all essential oils. This discrepancy may possibly be due to ion efficacy or a low concentration of material. Fragment patterns of LC-HRMS/MS in essential oils, *in silico* analysis of the fragments, and LC-HRMS/MS analysis using standard materials would confirm our hypothesis. Further study is needed to validate that these compounds were directly extracted from essential oils.

5. Conclusions

Essential oils could cause false-positive results of MCAD deficiency in NBS with LC-MS/MS because the vaporized essential oils contained sphinganine (d17:0) and 2-[2-hydroxyethyl(pentadecyl)amino]ethanol, which have an *m/z* similar to C8 and C10; therefore, Guthrie cards should be carefully handled and stored to avoid contamination, especially when using essential oils.

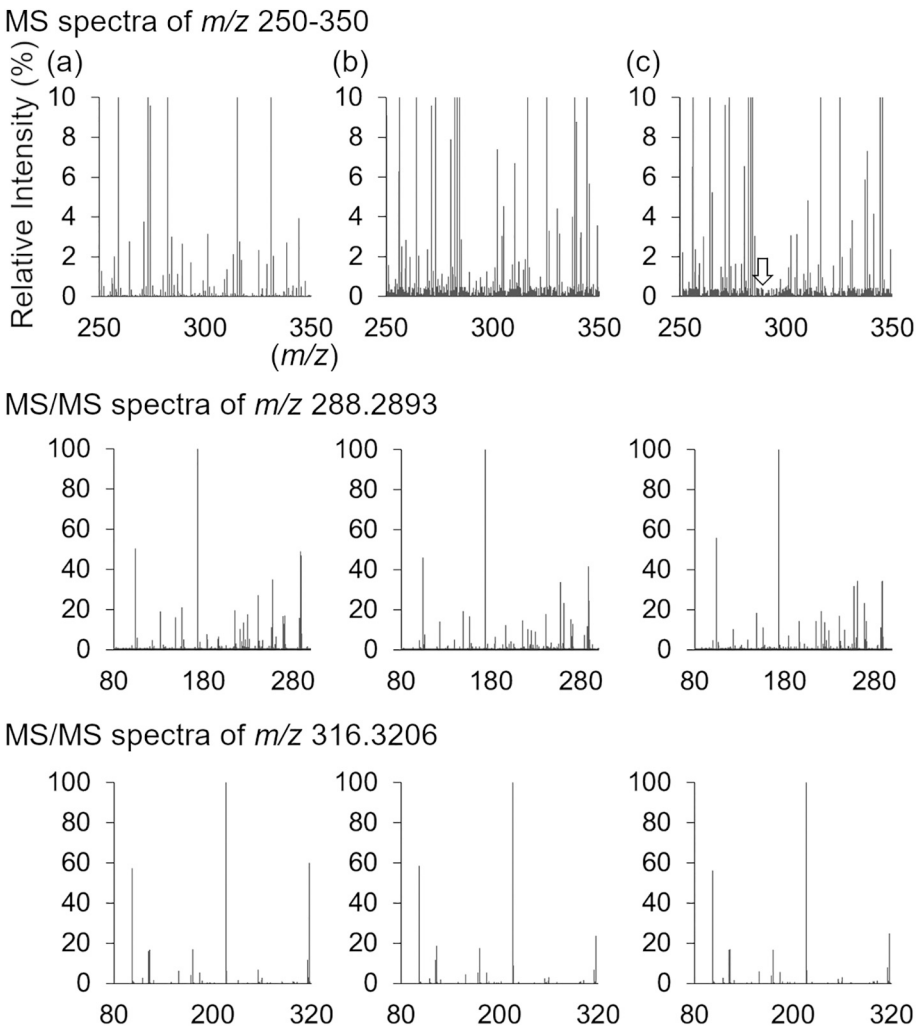

Fig. 4. LC-MS/MS spectra in Guthrie cards stained with vaporized essential oils. (a) Lavender, (b) tea tree, and (c) cypress. MS spectra, MS/MS spectra of *m/z* 288.2893, and MS/MS spectra of *m/z* 316.3206 are shown in order from the top to bottom rows. The open arrow in (c) indicates the peak of *m/z* 288.2893. Abbreviations: MS, mass spectrometry; MS/MS, tandem mass spectrometry.

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Ethics statement

This study was approved by the Ethics Committee of Tohoku University Hospital (approval number: 2020–1-132). All patients' parents provided written informed consent.

Authors' contributions

YM-S and YW conceptualized and designed the study, collected data, drafted the initial manuscript, and reviewed and revised the manuscript. MM designed the study, collected data, and reviewed and revised the manuscript. TK, AK, YS, TY, and NA-I collected data and reviewed and revised the manuscript. SK conceptualized the study and reviewed and revised the manuscript. All authors approved the final manuscript as submitted and agree to be accountable for all aspects of the work.

Declaration of Competing Interest

The authors have no conflicts of interest to disclose.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ymgmr.2020.100674.

References

[1] S.E. McCandless, E.J. Wright, Mandatory newborn screening in the United States: history, current status, and existential challenges, Birth Defects Res. 112 (2020) 350–366.
[2] G. Tajima, et al., Screening of MCAD deficiency in Japan: 16 years' experience of enzymatic and genetic evaluation, Mol. Genet. Metab. 119 (2016) 322–328.
[3] S. Kølvraa, N. Gregersen, E. Christensen, N. Hobolth, In vitro fibroblast studies in a patient with C6-C10-dicarboxylic aciduria: evidence for a defect in general acyl-CoA dehydrogenase, Clin. Chim. Acta 126 (1982) 53–67.
[4] Y. Matsubara, et al., Molecular cloning of cDNAs encoding rat and human medium-chain acyl-CoA dehydrogenase and assignment of the gene to human chromosome 1, Proc National Acad Sci 83 (1986) 6543–6547.
[5] L.N. Venditti, et al., Newborn screening by tandem mass spectrometry for medium-chain acyl-CoA dehydrogenase deficiency: a cost-effectiveness analysis, Pediatrics 112 (2003) 1005–1015.
[6] W.J. Rhead, Newborn screening for medium-chain acyl-CoA dehydrogenase deficiency: a global perspective, J. Inherit. Metab. Dis. 29 (2006) 370–377.
[7] J.L.V. Hove, et al., Medium-chain acyl-CoA dehydrogenase (MCAD) deficiency: diagnosis by acylcarnitine analysis in blood, Am. J. Hum. Genet. 52 (1993) 958–966.
[8] S.E. McCandless, R. Chandrasekar, S. Linard, S. Kikano, L. Rice, Sequencing from dried blood spots in infants with "false positive" newborn screen for MCAD deficiency, Mol. Genet. Metab. 108 (2013) 51–55.

[9] L.E. Kratz, J.S. Albert, 2-Ethylhexanoic acid, found in common plasticizers, leads to an artificial increase in C8 acylcarnitine levels in two neonates treated with extracorporeal membrane oxygenation (ECMO), Clin. Chim. Acta 461 (2016) 59–60.
[10] K. Hara, et al., Significance of ACADM mutations identified through newborn screening of MCAD deficiency in Japan, Mol. Genet. Metab. 118 (2016) 9–14.
[11] K. Yamada, T. Taketani, Management and diagnosis of mitochondrial fatty acid oxidation disorders: focus on very-long-chain acyl-CoA dehydrogenase deficiency, J. Hum. Genet. 64 (2019) 73–85.
[12] Z. Lai, et al., Identifying metabolites by integrating metabolome databases with mass spectrometry cheminformatics, Nat. Methods 15 (2018) 53–56.
[13] Y. Shigematsu, I. Hata, G. Tajima, Useful second-tier tests in expanded newborn screening of isovaleric acidemia and methylmalonic aciduria, J. Inherit. Metab. Dis. 33 (2010) S283–S288.
[14] E.A. Gurian, D.D. Kinnamon, J.J. Henry, S.E. Waisbren, Expanded newborn screening for biochemical disorders: the effect of a false-positive result, Pediatrics 117 (2006) 1915–1921.
[15] K. O'Connor, et al., Psychosocial impact on mothers receiving expanded newborn screening results, Eur. J. Hum. Genet. 26 (2018) 477–484.
[16] L.J. Pierce, et al., Association of perceived maternal stress during the perinatal period with electroencephalography patterns in 2-month-old infants, JAMA Pediatr. 173 (2019) 561.
[17] T. Tim-Aroon, H.M. Harmon, M.L. Nock, S.K. Viswanathan, S.E. McCandless, Stopping parenteral nutrition for 3 hours reduces false positives in Newborn screening, J. Pediatr. 167 (2015) 312–316.
[18] I. Schymik, et al., Pitfalls of neonatal screening for very-long-chain acyl-CoA dehydrogenase deficiency using tandem mass spectrometry, J. Pediatr. 149 (2006) 128–130.
[19] A.C. Edmondson, J. Salant, L.A. Ierardi-Curto, C. Ficicioglu, Missed newborn screening case of carnitine Palmitoyltransferase-II Deficiency, JIMD Rep (2016) 93–97.
[20] L. Dowsett, L. Lulis, C. Ficicioglu, S. Cuddapah, Utility of genetic testing for confirmation of abnormal Newborn screening in disorders of long-chain fatty acids: a missed case of carnitine Palmitoyltransferase 1A (CPT1A) deficiency, Int J Neonatal Screen 3 (2017) 10.
[21] D.R. Anderson, K. Viau, L.D. Botto, M. Pasquali, N. Longo, Clinical and biochemical outcomes of patients with medium-chain acyl-CoA dehydrogenase deficiency, Mol. Genet. Metab. 129 (2019) 13–19.
[22] H.-W. Hsu, et al., Spectrum of medium-chain acyl-CoA dehydrogenase deficiency detected by Newborn screening, Pediatrics 121 (2008) e1108–e1114.
[23] S.-S. Tsai, H.-H. Wang, F.-H. Chou, The effects of aromatherapy on postpartum women: a systematic review, J. Nurs. Res. 1 (2019). Latest Articles.
[24] R. Perry, R. Terry, L.K. Watson, E. Ernst, Is lavender an anxiolytic drug? A systematic review of randomised clinical trials, Phytomedicine 19 (2012) 825–835.
[25] L. Re, et al., Linalool modifies the nicotinic receptor–ion channel kinetics at the mouse neuromuscular junction, Pharmacol. Res. 42 (2000) 177–181.
[26] R.S. Carling, et al., Introduction of a simple second tier screening test for C5 isobars in dried blood spots: reducing the false positive rate for Isovaleric Acidaemia in expanded Newborn screening, JIMD Rep. 38 (2018) 75–80.
[27] E.A. Jager, et al., A nationwide retrospective observational study of population newborn screening for medium-chain acyl-CoA dehydrogenase (MCAD) deficiency in the Netherlands, J. Inherit. Metab. Dis. 42 (2019) 890–897.
[28] C.M.L. Touw, et al., Risk stratification by residual enzyme activity after newborn screening for medium-chain acyl-CoA dehydrogenase deficiency: data from a cohort study, Orphanet J. Rare Dis. 7 (2012) 30.
[29] G. Peng, et al., Reducing false-positive results in Newborn screening using machine learning, Int J Neonatal Screen 6 (2020) 16.
[30] W.S. da Oliveira, S. Ubeda, C. Nerín, M. Padula, H.T. Godoy, Identification of non-volatile migrants from baby bottles by UPLC-Q-TOF-MS, Food Res. Int. 123 (2019) 529–537.
[31] J.T. Hannich, D. Mellal, S. Feng, A. Zumbuehl, H. Riezman, Structure and conserved function of iso-branched sphingoid bases from the nematode Caenorhabditis elegans, Chem. Sci. 8 (2017) 3676–3686.
[32] C.R. Dietrich, et al., Loss-of-function mutations and inducible RNAi suppression of Arabidopsis LCB2 genes reveal the critical role of sphingolipids in gametophytic and sporophytic cell viability, Plant J. 54 (2008) 284–298.