

Guidance and practice in the diagnosis and management of two rare inherited metabolic diseases

BY

Alessia Kazakova

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School of Epidemiology and Public Health (SEPH)
Faculty of Medicine
University of Ottawa

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Contribution of Authors

Alessia Kazakova (AK) was primarily responsible for planning and conducting all components of this thesis, including literature reviews and data analysis, with the guidance of her supervisor Dr. Beth Potter (BP), and members of her thesis advisory committee (TAC). Specifically, in addition to providing overall mentorship and guidance with the project, TAC members Drs Pranesh Chakraborty (PC) and Michael Geraghty (MG) provided expertise in clinical metabolic medicine; Ian Graham (IG) provided expertise related to clinical practice guidelines and knowledge synthesis; and Franco Momoli (FM) provided expertise related to data analysis and interpretation. AK is the first author of the two articles included in this thesis and her supervisor and TAC members are co-authors.

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Abstract

By facilitating timely diagnosis and treatment initiation, population-wide newborn screening programs have led to important reductions in morbidity and mortality for many rare diseases, including medium-chain acyl-CoA dehydrogenase (MCAD) deficiency and very-long-chain acyl-CoA dehydrogenase (VLCAD) deficiency. Newborn screening has also expanded the spectrum of disease severity for MCAD and VLCAD deficiencies to include a higher proportion of milder cases, raising questions about appropriate disease management. To date there has been no systematic attempt to characterize best management practices in terms of the guidance that is available to those who provide care for MCAD and VLCAD deficiencies; nor has there been an attempt to understand the extent to which current practices align with such currently available guidance. The two projects that are part of this thesis sought to address these research gaps with a particular focus on two key disease-specific management practices we identified in advance as priorities: the use of carnitine supplementation and the recommended duration of fasting.

The objective of the first project was to systematically review the quality and content of clinical practice guidelines and/or recommendations for the diagnosis and management of both MCAD and VLCAD deficiencies. Two independent reviewers assessed the eligibility of citations retrieved from a comprehensive search of the peer-reviewed and grey literature. We appraised the quality of the reviewed guidance and extracted information on the content of recommendations. From the 25 guidance documents that met our inclusion criteria, only 7 incorporated evidence reviews, indicating that guidance in this field does not generally meet established methodological standards for the rigorous development of clinical practice guidelines. With respect to content, we identified unclear and inconsistent recommendations

regarding fasting times and the use of carnitine supplementation. Further empirical evidence in these areas is necessary to inform the development of future rigorous guidelines.

The objective of the second project was to identify actual practices in the management of MCAD deficiency. We conducted a scoping review of published literature on treatment practices around the world and a secondary analysis of data documenting treatments received by participants in a Canadian pediatric cohort study. For the scoping review, citations retrieved from our comprehensive search strategy were screened by two independent reviewers. We extracted information on study characteristics and disease management. Our secondary analysis included longitudinal data for Canadian children with MCAD deficiency, born between 2006 and 2015 and enrolled in a cohort study at one of 13 centres. For both project components, we described carnitine supplementation and fasting times, overall and according to potential indicators of disease severity (genotype, biochemical phenotype). We identified 5 relevant publications in the scoping review and analyzed data for 107 children participating in the Canadian cohort. Management practices related to carnitine supplementation and fasting times for MCAD deficiency were highly variable based on both data sources. There was some evidence of an association between genotype and carnitine use, which, based on the scoping review, may be due to a relationship between genotype and carnitine deficiency. While actual practice was in some ways aligned with the guidance we reviewed in the first project, these results underscore the need for further evidence to address areas of uncertainty that have been prioritized by patients and families, clinicians, and health researchers, including questions regarding the potential to tailor treatment to predicted disease severity and an emphasis on controversial therapies such as carnitine supplementation.

Chapter 1 – Background

Preface

The overall aim of this thesis was to investigate the extent to which there is an evidence-practice gap in relation to the management of two rare inherited metabolic diseases (IMD), medium-chain acyl-CoA dehydrogenase (MCAD) deficiency and very-long-chain acyl-CoA dehydrogenase (VLCAD) deficiency. This was accomplished by first determining the current state of evidence as reflected in available treatment guidelines or recommendations; and secondly, for MCAD deficiency, by examining actual practice and interpreting that practice in the context of the guidance. This introductory chapter will provide background information which will introduce the rationale for this thesis, focusing on variation in care but also describing the clinical and biochemical presentation of the rare disorders themselves, particularly emphasizing questions that have arisen following the inclusion of both conditions as targets of population-wide newborn screening (NBS) programs.

Rare Diseases

Rare diseases are often defined as those that affect fewer than 1 in 2000 people and although they are individually rare, collectively they are common, with over 7000 diagnosable rare conditions identified to date.¹ According to the Canadian Organization for Rare Disorders (CORD), together, approximately 3 million Canadians (1 in 12) are affected by a rare disease.²

Newborn Screening (NBS)

Population-wide NBS programs are in place in over 64 countries worldwide.³ Blood samples are normally taken within the first 3 days of life, typically via a heel prick blood sample. NBS aims to identify children with rare diseases who would benefit from early detection and

initiation of treatment.⁴ Ideally, the test should be processed quickly and results made available before the baby presents with clinical symptoms⁴ and it is important for clinicians to remain vigilant for these disorders, given that infants have become symptomatic before results were obtained.⁵ NBS programs have expanded in many jurisdictions over the past 10-15 years in terms of the number of diseases targeted, partly in response to the adoption of electrospray ionization tandem mass spectrometry (MS/MS) in NBS, which allows for the detection of a large number of metabolic conditions using a single assay, and also due to the development of new disease therapies. The US and Canada have no national NBS programs, with each state and province having jurisdiction over diseases to screen.^{6,7} In Canada, NBS programs target from 5 to 38 disorders, with phenylketonuria (PKU), cystic fibrosis, congenital hypothyroidism, and medium-chain acyl-CoA dehydrogenase (MCAD) deficiency being the four disorders most commonly included in NBS programs in Canada.³

Inherited Metabolic Diseases (IMD)

The largest category of diseases that are targeted by NBS programs are inherited metabolic diseases (IMD). IMD are a collection of more than 400 single-gene disorders that disrupt normal metabolic functions (only a small subset of IMD have been targets of NBS programs). Depending on both individual patient and disease characteristics, these disorders may lead to a range of clinical manifestations including risk for acute episodic metabolic decompensation, neurodevelopmental consequences⁸ and/or chronic multi-organ sequelae that may be stable or progressive.⁹ Prompting their inclusion in NBS programs, many IMD are treatable, and for several IMD, early diagnosis and treatment initiation have been associated with reduced mortality and morbidity among children diagnosed who have these conditions.^{10,11} Multiple categories of IMD have been targets of NBS, including urea cycle disorders, organic

acidemias, amino acid disorders and fatty acid oxidation disorders (FAOD). NBS has led to an increase in the observed prevalence of disease for many IMD, uncovering some cases of disease that may have resulted in early death without a diagnosis in the absence of screening, and also identifying mild disease cases which may have remained asymptomatic without screening.¹⁰

Newborn Screening for Fatty Acid Oxidation Disorders: Medium Chain Acyl-CoA

Dehydrogenase Deficiency and Very-Long- Chain Acyl-CoA Dehydrogenase Deficiency

Among the FAOD that are targets of many NBS programs are medium-chain acyl-CoA dehydrogenase (MCAD) deficiency and very-long-chain acyl-CoA dehydrogenase (VLCAD) deficiency, both single-gene disorders with an autosomal recessive inheritance pattern, characterized by impairments in metabolic pathways needed to break down body fat.¹²

NBS has facilitated the diagnosis of these FAOD because their presentation was previously difficult to differentiate from other disorders such as sudden infant death syndrome (SIDS) and cyclic vomiting syndrome (CVS).^{13,14} As will be described further, during times of metabolic stress, children with all types of FAOD (including MCAD and VLCAD deficiency) are at risk of acute crises (metabolic decompensation involving hypoketotic hypoglycemia) that can be life-threatening, while long-chain FAOD such as VLCAD deficiency also present with a spectrum of phenotypes that can include chronic manifestations affecting the heart, liver and neuromuscular systems.¹⁵ Knowledge of the risk of acute metabolic decompensation, and prompt preventive management during times of catabolic stress (e.g., prolonged physical activity, illness, fever, and fasting) is highly successful at preventing acute crises for FAOD, which has justified their inclusion as targets of NBS.¹¹ In Canada, all provinces now offer screening for MCAD deficiency. Table 1 summarizes by province the year that NBS started for both disorders.

Table 1. MCAD deficiency and VLCAD deficiency screening by year in Canadian Provinces

	MCAD Deficiency	VLCAD Deficiency
Alberta	2007 ¹⁶	2007 ¹⁶
British Columbia	2004 ¹⁷	2009 ¹⁸
Manitoba	2011 ¹⁹	2011
Newfoundland and Labrador	2005 ⁽¹⁾	2013 ⁽¹⁾
New Brunswick	2007 ⁽²⁾	2007 ⁽²⁾
Nova Scotia	2000 ¹⁷	2004 ⁽³⁾
Ontario	2006 ²⁰	2006 ²¹
PEI	2005 ²²	2007 ⁽⁶⁾
Quebec	2012 ²³	NA
Saskatchewan	2000 ⁽⁴⁾	2000 ⁽⁴⁾
Canadian Territories: Northwest Territories, Nunavut, Yukon	2004-2015 ⁽⁵⁾	2009-2015 ⁽⁵⁾

(1) Panel of markers for mass spectrometry was expanded in 2010 at which time VLCADD marker was added and abnormal results were being reported. Officially, NFL started screening for VLCADD in 2013. Email communication: Sergei Likhodi (Clinical Biochemist, Eastern Health Authority), 'MCADD and VLCADD screening in Newfoundland and Labrador', June 24 2016.

(2) Email communication: Coral MacRae (Healthcare Consultant, New Brunswick Department of Health), 'MCADD and VLCADD screening in NB', June 28 2016.

(3) Email communication: Patti Burrell (IWK Lab Manager), "VLCADD screening in NS", June 27 2016.

(4) Expanded NBS began in 2000 in SK including the entire ACMG recommendations. Email communication: Denis C Lehotay (University of Saskatchewan), 'MCADD and VLCADD screening in Saskatchewan', June 25 2016.

(5) Based on published CORD status reports from 2008-2015, Yukon was screening for VLCADD by 2009. Yukon Labs are managed by BC therefore MCADD screening likely started in 2004, at the same time as BC. The Northwest territories started screening for MCADD and VLCADD in 2012 while all regions of Nunavut joined by 2015. *Canadian Organization for Rare Disorders*. "Newborn Screening in Canada Status Report" 1-3. Print. 2008-2015.

(6) Date of screening for VLCADD in PEI was not found but they are known to have been screening for VLCAD in 2007.

NA Quebec was not screening for VLCADD in 2015, could not identify information whether they are currently screening, but there has been discussion to add VLCADD to the screening panel.

Medium Chain Acyl-CoA Dehydrogenase (MCAD) deficiency

MCAD deficiency is the most common FAOD¹⁰ and has a birth prevalence of about 1 in 14,000 in North American and northern European populations.²⁰ MCAD deficiency is an autosomal recessive inherited disease caused by mutations on the ACADM gene that result in deficiency or insufficient activity of the MCAD enzyme. The MCAD enzyme is required in the process of mitochondrial fatty acid beta oxidation of medium chain (C6-C10) acyl-CoA esters.²⁴

When hepatic glycogen stores are depleted during catabolic states, for example, when there are increased energy demands due to illness, exercise or prolonged fasting, the MCAD enzyme is normally involved in hepatic ketogenesis to provide the major source of fuel. Individuals with MCAD deficiency are not able to respond to catabolic stress in this manner and thus risk experiencing an acute metabolic decompensation. Signs and symptoms of an acute metabolic crisis include hypoketotic hypoglycemia, lethargy, seizures and vomiting, possibly leading to coma and death if untreated.²⁰

Avoidance of fasting is the main long-term treatment strategy for MCAD deficiency and has been demonstrated to effectively prevent metabolic decompensation.²⁵⁻²⁷ During high-risk periods (e.g., fever and fasting when ill), individuals with MCAD deficiency require rapid and readily available carbohydrates to maintain blood glucose levels.^{25,28} Children with MCAD deficiency frequently visit the emergency department during high-risk periods for medical monitoring and preventive interventions such as intravenous glucose. Acute crises often require inpatient admission to prevent coma and death. In addition to avoidance of fasting, a number of other preventive interventions have been used for the long-term management of MCAD deficiency, for example, supplementation with corn starch and/or with carnitine. Carnitine supplementation in particular has been controversial,^{11,29,30} with some recent concern about potential adverse effects.³¹ While avoidance of fasting is critical to prevent crises for MCAD deficiency, there is also concern about overfeeding and the risk of overweight and obesity among children and adults with MCAD deficiency.⁸

Biomarkers used in NBS to identify children who may have MCAD deficiency and require diagnostic follow-up include elevated medium chain acylcarnitines, particularly octanoylcarnitine (C8),⁴ as well as the secondary analytes C6, C10, C10:1, C8/C6, C8/C10 and

C8/C12, with screening thresholds established by specific NBS laboratories.³² A combination of tests are used for the diagnosis of MCAD deficiency among children with positive NBS results for this disorder or with clinical symptoms. These tests can include plasma acylcarnitines, urine organic acids and acylglycines, as well as genetic testing and enzyme assays,^{28,33–35} where residual MCAD activity below 25% is typically indicative of MCAD deficiency.²⁵

NBS for MCAD deficiency has been associated with improved outcomes and decreased mortality in children with the disease.^{11,36,37} Prior to the introduction of NBS for MCAD deficiency, some cases would be detected upon presentation of infants with Reye's syndrome or SIDS.^{38,39} Indeed, in the absence of NBS, the mortality rate associated with an initial metabolic crisis among children with MCAD deficiency was approximately 25%,^{40–42} and approximately 20% of surviving children who experienced a crisis had neurological sequelae.⁴² Due to substantially reduced morbidity and mortality, NBS for MCAD deficiency is considered cost-effective.⁴³

NBS has resulted in a large increase in the observed prevalence of MCAD deficiency, which is believed to be partly due to severe fatal cases that went undiagnosed in the absence of NBS but also due to a potentially large pool of cases of MCAD deficiency that may be relatively mild and would remain asymptomatic in the absence of NBS. Some evidence supporting that some NBS-detected cases are mild is the identification of many novel mutations in the ACADM gene in screened populations.²⁰ The K329E (c.985A→G) mutation is the most common mutation associated with impaired functioning of the MCAD enzyme. Individuals who are homozygous for this mutation are often referred to as having 'classical MCAD deficiency'. While previously about 80% of clinically presenting cases were homozygous for the c.985A>G mutation,^{44,45} with NBS, some studies have found a lower proportion of cases with this genotype (although

estimates vary considerably between studies, from 30 to 71%).^{20,40,44,46} This has led to the suggestion that the common mutation may be associated with a more severe phenotype compared to some novel screen-detected mutations. For example, the c.199T>C mutation is relatively common in screen-detected cases of MCAD deficiency and may be a milder variant.^{32,45} However, associations among genotype, biochemical phenotype (e.g., levels of acylcarnitines), enzyme activity levels, and clinical phenotype (i.e., actual risk of metabolic decompensation with environmental stress) in individuals with MCAD deficiency are not well-established and characterization of disease severity in populations of patients detected asymptotically via NBS is challenging.^{20,36,47–49} Treatment is thus typically recommended as a precautionary measure for all children diagnosed with MCAD deficiency, regardless of predicted severity, given this uncertainty.⁴⁸

Very-Long-Chain Acyl-CoA Dehydrogenase (VLCAD) deficiency

VLCAD deficiency is rarer than MCAD deficiency and is estimated to have a birth prevalence of approximately 1 in 30,000, according to the NIH.⁵⁰ VLCAD deficiency is an autosomal recessive inherited disease caused by mutations on the ACADVL gene that result in deficiency or insufficient activity of the VLCAD enzyme. VLCAD deficiency results in the inability to effectively metabolize very long chain fatty acids (22+ Carbon tail), as the VLCAD enzyme catalyzes the first step in mitochondrial beta fatty acid oxidation. This disorder presents with a wide spectrum of clinical phenotypes. VLCAD deficiency is similar to MCAD deficiency in that individuals with the disorder are at risk of an acute metabolic decompensation characterized by hypoketotic hypoglycemia as a result of an inability to respond to metabolic stress, for example during times of fasting and fever. However, children with VLCAD deficiency

may also experience rhabdomyolysis and kidney failure during times of stress,^{36,51} and many individuals with VLCAD deficiency have chronic manifestations affecting the heart, liver and neuromuscular systems.^{15,52} Thus, VLCAD deficiency has been associated with a risk of more severe consequences relative to MCAD deficiency.⁴⁴ Some authors have classified VLCAD deficiency into three distinct phenotypes: infancy-onset, early childhood onset, and later-onset, with earlier-onset disease being more severe.^{51,53–56} Similar to MCAD deficiency, early initiation of long-term management for VLCAD deficiency aims to prevent cases of metabolic decompensation. Frequent feeding and avoidance of long chain fatty acids are often used to prevent crises⁵⁷ while enzyme treatments are used in some cases to increase enzyme activity levels.¹⁵ Other long-term management strategies that may be used for some patients with VLCAD deficiency include discontinuation of breastfeeding for some symptomatic infants, changes to dietary fat composition (e.g., limiting long chain fat intake and supplementing the diet with some essential fatty acids), and the use medium-chain triglyceride (MCT) supplementation.^{35,58–60} As with MCAD deficiency, carnitine supplementation is controversial.⁵⁸ Triheptanoin, a medium chain fat, has been suggested as a novel treatment for VLCAD deficiency with cardiomyopathy.^{55,61} Regular cardiac monitoring and creatine kinase tests are also recommended for patients diagnosed with VLCAD deficiency, given the risk of chronic multi-system disease.^{58,60} While screening has increased the observed prevalence of the disorder, it has also improved outcomes of infants identified early and treated promptly.^{36,59}

Elevated C14:1 from acylcarnitine profiles on the NBS blood sample are used as the screening marker for VLCAD deficiency,⁶² and secondary analytes include C14:2, C14 and C12:1.⁴ Positive NBS results are followed up with diagnostic testing that may include molecular genetic testing and the measurement of palmitoyl CoA oxidation in fibroblasts and lymphocytes.

Additional confirmatory testing of acylcarnitine markers and organic acids may also be conducted but these may be in the normal range even in the presence of disease among individuals with VLCAD deficiency who are presently well.^{10,58,62}

Also similar to MCAD deficiency, NBS for VLCAD deficiency has led to questions about the potential presence of mild cases detected asymptotically through screening and the challenge in predicting severity. While a number of disease-causing mutation variants have been identified on the ACADVL gene on chromosome 17,⁵⁰ genotype-phenotype associations are not definitive.¹⁰ Indeed there is much more heterogeneity in genetic mutations for VLCAD deficiency compared to MCAD deficiency,⁶² with no dominant genotype in either screened or non-screened patient populations. The extent to which the build-up of long chain fats affects phenotype is unclear, and residual enzyme activity, acylcarnitine blood results and genetic testing have not yet been able to clearly discriminate among phenotypes.⁵⁴ An exception to the lack of genotype-phenotype association is that the most severe phenotype for VLCAD has been associated with the presence of homozygosity for null mutations in the enzyme,^{11,44} while milder forms have been associated more often with missense mutations,¹⁵ with one relatively commonly reported V243A mutation associated with a milder clinical phenotype.⁶³ Again, as with MCAD deficiency, it is unclear whether cases of VLCAD deficiency that are predicted to be milder should be clinically managed in the same manner as classical cases or whether different treatments should be implemented.¹⁰

Thesis rationale: Variation in care for MCAD deficiency and VLCAD deficiency and the need to understand both guidance and current practice

NBS has expanded the clinical spectrum of disease for MCAD and VLCAD deficiencies and thus previously unrecognized variability has resulted in questions about appropriate management of screen-detected cases that are expected to be milder.⁶⁴ Recent surveys of Canadian metabolic physicians and IMD treatment centres have also highlighted variation in views regarding management of MCAD and VLCAD deficiencies⁶⁵ as well as variation in the organization of care.⁶⁶ Specifically, as described earlier, the foundation of long-term preventive management for both diseases is avoidance of fasting; yet there is variation in the way that fasting avoidance is operationalized in terms of specific fasting times,^{26,60,67,68} and potential concern about the risk of development of overweight if there is overfeeding.^{8,26,27,67} Another important specific area of variation in care for both disorders is carnitine supplementation, which has a physiological rationale but little evidence supporting it,⁶⁹ and, as mentioned, recent concern about potential adverse effects.^{60,70}

The collective recent experience with NBS for MCAD and VLCAD deficiencies (over a decade now in many jurisdictions around the world), as well the development of new therapies for both disorders, has likely resulted in changing diagnostic and disease management practices and has contributed to the variation in care that has been observed. However, these are rare diseases with limited empirical data available to understand which treatments are associated with improved outcomes. To date there has been no systematic attempt to characterize best practice in terms of the guidance that is available to those who provide care for MCAD and VLCAD deficiencies; nor has there been an attempt to understand the extent to which current practices align with such currently available guidance. The two projects that are part of this thesis thus

investigated current treatment guidelines or recommendations and actual management practices for MCAD deficiency and for VLCAD deficiency, in Canada and around the world.

The format of this thesis is manuscript-based, with the first article (Chapter 2) presenting the results of our investigation of treatment guidance and the second article (Chapter 3) presenting actual management practices, reported in the literature and from a Canadian cohort of children. The final chapter integrates the two manuscripts, comparing guidance with practice to determine the evidence-practice gap, and outlining future directions for research related to the management of MCAD and VLCAD deficiencies.

Objectives

More specifically, the first objective of this thesis, presented in chapter 2, was to systematically review the quality and content of clinical practice guidelines and/or recommendations for the diagnosis and management of both MCAD and VLCAD deficiencies. We evaluated the quality of the guidance we identified and then reviewed the content of the recommendations made within the guidance, focusing on identifying consistent vs conflicting recommendations. Although we were interested in guidance related to all aspects of diagnosis and management of both MCAD and VLCAD deficiencies, we particularly emphasized guidance related to avoidance of fasting and carnitine supplementation, the two areas of care that we had identified a priori as controversial.

The second objective, presented in chapter 3, was to identify actual practices in the management of MCAD deficiency, with a specific focus on avoidance of fasting and carnitine supplementation. To pursue this second objective, we conducted a scoping review of published literature on treatment practices around the world, and a secondary analysis of data documenting

treatments received by participants in a Canadian cohort study of children with MCAD deficiency. Although we had initially intended to conduct the scoping review and analysis of cohort data for both MCAD and VLCAD deficiencies, we revised this objective to be specific to MCAD deficiency due to the limited literature and very limited data that were available for VLCAD deficiency, and for practical manageability of the scope of the project.

Finally, in chapter 4, an integrative discussion brings together the findings from chapters 2 and 3 to determine the evidence-practice gap, mainly emphasizing MCAD deficiency and focusing on avoidance of fasting and carnitine supplementation. The integrative chapter summarizes the limitations of the thesis and concludes with the main implications of this research and as well suggested directions for future research.

INTERFACE

Given the spectrum of disease severity uncovered by NBS, particularly milder cases and those which may have remained asymptomatic and thus undiagnosed in the absence of screening, it is unclear whether management should be modified. In the project that is reported in Chapter 2, we undertook a systematic review to identify published guidance documents available for health care providers to guide their decision-making in managing MCAD deficiency and VLCAD deficiency, two rare inherited metabolic diseases. We extracted information on the content of the guidance with respect to diagnosis, acute management, and long-term management. We also conducted a quality appraisal of identified guidance documents using the AGREEII tool.

The student (AK) was responsible for conducting the literature search, screening retrieved citations for eligibility, extracting information from eligible included studies, appraising the quality and summarizing the content of the guidance documents, and writing the manuscript. BKP and IG contributed to the conception, design and supervision of the study, the analysis of data from the reviewed articles, and the interpretation of findings and critical revisions to the manuscript. MG, PC, and FM contributed to the conception, design and supervision of the study and the interpretation of findings and critical revisions to the manuscript. JM led the construction of the search strategy. AF contributed to the screening of retrieved citations for eligibility and extraction of information from included studies. JJM, SS, and HV contributed to the conception and design of the study and interpretation of the findings. We plan to submit this manuscript to the journal, *Genetics in Medicine*; it has been formatted accordingly.

Chapter 2 – Systematic Review of the Quality and Content of Practice Guidelines for the Diagnosis and Management of Medium-Chain Acyl-CoA Dehydrogenase (MCAD) deficiency and Very Long-Chain Acyl-CoA Dehydrogenase (VLCAD) deficiency

A Kazakova^a, ID Graham^{a,c}, P Chakraborty^b, MT Geraghty^b, F Momoli^{a,b,c}, J McGowan^a, A Fairfax^a, JJ Mitchell^d, S Stockler^e, H Vallance^e, BK Potter^a, in collaboration with the Canadian Inherited Metabolic Diseases Research Network

Affiliations

^aUniversity of Ottawa, Ottawa, Ontario, Canada

^bChildren's Hospital of Eastern Ontario, Ottawa, Ontario, Canada

^cOttawa Hospital Research Institute, Ottawa, Ontario, Canada

^dMontreal Children's Hospital, Montreal, Quebec, Canada

^eBC Children's Hospital, Vancouver, British Columbia, Canada

Abstract

Purpose To review the quality and the content of existing guidance for the diagnosis and management of medium-chain acyl-CoA dehydrogenase (MCAD) deficiency and very-long-chain acyl-CoA dehydrogenase (VLCAD) deficiency.

Methods We developed a comprehensive search strategy across multiple electronic databases and grey literature sources to identify relevant guidance documents. Two independent reviewers assessed the eligibility of retrieved citations. From the included documents, we extracted information on guidance development methods, appraised guidance quality using the AGREEII tool, and summarized the content of recommendations.

Results The 25 guidance documents that met final inclusion criteria made recommendations about MCAD deficiency only (n=10), VLCAD deficiency only (n=2) or both disorders (n=13). The quality appraisal yielded median scores of >80% on the AGREEII domains of 'Scope and Purpose' and 'Clarity of Presentation' but median scores of <20% on the domains of 'Rigor of Development' and 'Applicability'. Few guidance documents (n=7) incorporated evidence reviews. Recommendations were more consistent for diagnosis than for disease management. Recommendations were conflicting or unclear regarding fasting times and carnitine supplementation, two a priori areas of interest.

Conclusion We identified conflicting guideline recommendations and limited evidence-based guidance for care providers involved in managing these metabolic disorders.

Introduction

Population-wide newborn screening programs have resulted in early diagnosis and treatment initiation for children with many rare treatable conditions that are asymptomatic at birth and may benefit from early initiation of care.¹ The majority of targets of current newborn screening programs are single-gene autosomal recessive inherited metabolic diseases (IMD), including fatty acid oxidation disorders (FAOD) such as medium chain acyl-CoA dehydrogenase (MCAD) deficiency and very long chain acyl-CoA dehydrogenase (VLCAD) deficiency. Newborn screening has been associated with reduced morbidity and mortality for children with these disorders, particularly MCAD deficiency, largely due to the early initiation of treatment to prevent or reduce the severity of episodes of metabolic decompensation that characterize an important risk for both conditions.²⁻⁴

Individuals with MCAD deficiency experience a risk of metabolic decompensation in situations characterized by increased metabolic demand, such as fever and fasting. If not treated promptly and appropriately, a metabolic decompensation can result in neurodevelopmental damage, coma and death.⁵ VLCAD deficiency is characterized by a risk of acute metabolic decompensation, as in MCAD deficiency, but is also frequently characterized by additional manifestations that may be acute or chronic, such as myalgias, rhabdomyolysis and cardiomyopathy.⁶ VLCAD deficiency has been described with 3 distinct phenotypes: a severe and often fatal phenotype marked by cardiomyopathy and hepatomegaly, an MCAD deficiency-like phenotype with recurrent hypoglycemia, and a later-onset phenotype characterized by rhabdomyolysis during times of metabolic stress.⁷⁻⁹ The main treatments for both MCAD deficiency and VLCAD deficiency are prevention of fasting and close medical monitoring during

periods of metabolic stress.^{3,10} Several other treatments are used variably for both conditions, for example, fat restriction/modified fat diets for VLCAD deficiency.^{3,4,11–15}

Newborn screening has led to a substantially increased observed birth prevalence of both MCAD deficiency and VLCAD deficiency.^{2,16} This may partly reflect identification of cases of these diseases that would normally have led to early death without a diagnosis in the absence of newborn screening. However, the large increase in observed prevalence (e.g., at least two-fold for MCAD deficiency)² combined with existing evidence about the clinical course of screen-detected cases⁶ strongly suggests that there may be a relatively large group of milder cases that would have remained asymptomatic and thus would not otherwise have been identified in the absence of diagnosis through screening.^{6,16,17} Thus, newborn screening has led to a broadening of the clinical spectrum of MCAD and VLCAD deficiencies and this in turn has raised questions about both diagnostic definitions of disease and the appropriate clinical management of those NBS-detected cases that may be predicted to be less severe.¹⁸ For example, while some genotype-phenotype correlations have been identified for these disorders it has not been possible to rule out risk for children who are asymptomatic and have a ‘non-classic’ or potentially milder genotype. A common mutation on the ACADM gene causing MCAD deficiency, c.985A>G, was previously responsible for 80% of clinically identified Caucasian cases of the disorder.¹⁹ However, in populations with newborn screening for MCAD deficiency, compound heterozygote status for c.985A>G and another mutation, as well as other genotypes, have been seen more frequently and may result in milder phenotypes.^{20,21} There is no single dominant genotype among patients with VLCAD deficiency, and the genotype-phenotype association is not well-established for that disorder.¹⁶

Due to the rarity of MCAD and VLCAD deficiencies, there is limited evidence to support treatment strategies, which are variable across centres and health care providers, particularly in the current newborn screening era, when most cases are identified asymptotically.²² This study sought to identify and review existing practice guidelines and recommendations (hereafter, “guidance”) for the diagnosis and management of MCAD and VLCAD deficiencies. Specifically, our objectives were to (i) evaluate the quality of the guidance we identified; and (ii) synthesize the content of that guidance, identifying areas where guidance is consistent and areas where it is conflicting. Although we reviewed all aspects of diagnosis and treatment covered in the content of the available guidance, we had a priori interest in two specific interventions: avoidance of fasting and carnitine supplementation. Avoidance of fasting is the foundation of long-term preventive management for MCAD and VLCAD deficiencies but there is some evidence of variation in specific fasting schedules²³ and potential concern about risk of development of overweight with overfeeding.¹³ Carnitine supplementation has been used in the management of both disorders on a physiologic basis but is controversial given a lack of evidence of its effectiveness and recent concern over potential adverse effects.²⁴ Thus, in our analysis and interpretation of the results of this review we placed particular emphasis on these two interventions.

Methods

We conducted a systematic search of published, peer-reviewed scientific literature to identify guidance for the diagnosis and management of MCAD and VLCAD deficiencies. With an experienced health science librarian (JM), we developed a search strategy for multiple electronic bibliographic databases. The search was conducted in MEDLINE, EMBASE (from 1946-November 3rd, 2016). We used both MESH terms and text word searching to capture key

constructs. Box 1 shows the search strategy that was used for Medline(R) and was adapted for the other databases. We also developed a grey literature search strategy using similar search terms and applied it across a list of 54 established guideline databases, websites, unpublished literature and reports, and directives from large research groups and organizations that the research team deemed potentially relevant, including but not limited to the National Guideline Clearinghouse,²⁵ the KT PLUS McMaster database and Turning Research Into Practice (TRIP) database.²⁶ These potential sources of grey literature were searched in December 2016 and January 2017 (one additional source, the International Guidelines Network, was added to the search in May 2018 and searched at that time). A full listing of grey literature sources that we searched is included in Appendix A, Supplement 1. Our search included both English and French literature. Duplicate citations were removed both manually and by autodetection in Covidence.²⁷

Box 1. Search strategy for systematic review of MCAD/VLCAD deficiency diagnosis and management guidance

- 1 exp Acyl-CoA Dehydrogenases/ (2546)
- 2 [MCAD.tw.](#) (568)
- 3 [MCADD.tw.](#) (89)
- 4 (medium-chain acyl-CoA dehydrogenase or Medium-chain acyl-coenzyme A dehydrogenase).tw. (910)
- 5 (medium chain acyl CoA dehydrogenase or Medium chain acyl coenzyme A dehydrogenase).tw. (910)
- 6 [VLCADD.tw.](#) (39)
- 7 [VLCAD.tw.](#) (217)
- 8 (very-long-chain acyl-CoA dehydrogenase or Very long chain acyl CoA dehydrogenase).tw. (270)

- 9 (acyl coa dehydrogenases or acyl-coa dehydrogenases or dehydrogenases acyl-coa).tw. (266)
- 10 Lipid Metabolism, Inborn Errors/ (2521)
- 11 Fatty Acid Desaturases/ (3111)
- 12 fatty acid oxidation disorder\$.tw. (249)
- 13 (FAOD or LC-FAOD).tw. (43)
- 14 Mitochondrial fatty acid [oxidation.tw.](#) (553)
- 15 FAO defect\$.tw. (26)
- 16 Metabolism, Inborn Errors/ (9989)
- 17 or/1-16 (18006)
- 18 exp clinical pathway/ (5474)
- 19 exp clinical protocol/ (143867)
- 20 exp consensus/ (7255)
- 21 exp consensus development conference/ (10502)
- 22 critical pathways/ (5474)
- 23 exp guideline/ (28654)
- 24 exp practice guideline/ (22056)
- 25 (guideline or practice guideline or consensus development conference or consensus development conference, NIH).pt. (36998)
- 26 (position statement* or policy statement* or practice parameter* or best practice*).tw. (20943)
- 27 (standards or guideline or guidelines).tw. (345928)
- 28 ((practice or treatment* or clinical) adj guideline*).tw. (34245)
- 29 (CPG or CPGs).tw. (24357)
- 30 consensus*.tw. (122684)

- 31 consensus*.ab. /freq=2 (17891)
- 32 ((critical or clinical or practice) adj2 (path or paths or pathway or pathways or protocol*)).tw.
(14687)
- 33 recommendat*.tw. (184279)
- 34 (care adj2 (standard or path or paths or pathway or pathways or map or maps or plan or plans)).tw.
(38483)
- 35 (algorithm* adj2 (screening or examination or test or tested or testing or assessment* or diagnosis
or diagnoses or diagnosed or diagnosing)).tw. (5230)
- 36 (algorithm* adj2 (therap* or treatment* or intervention*)).tw. (6639)
- 37 or/18-36 (829312)
- 38 17 and 37 (341)

All citations were screened in two stages by two reviewers (AK and AF) working independently. At the first stage, we screened titles and abstracts against the inclusion and exclusion criteria (Table 1). Those citations deemed definitely or possibly relevant at the first stage of screening passed on to the second stage of screening, where the full text of the paper was screened and specific reasons for exclusion documented. At both stages of screening, conflicts were resolved by discussion; if consensus could not be reached, we consulted a third reviewer (BP or IG). The grey literature sources were searched by one researcher (AK) and potentially relevant documents were retrieved and incorporated into the second stage of screening and reviewed by both reviewers (AK and AF). The number of citations from each source and the results of the two stages of screening were recorded (Figure 1).

Table 1. Inclusion and Exclusion Criteria to Determine Eligibility of Guidance Documents

Inclusion	Exclusion
<ul style="list-style-type: none"> ● published guideline ● formal recommendations for diagnosis and/or management of MCAD/VLCAD deficiency ● based on empirical evidence, expert consensus or a combination ● large scale clinical trials that explicitly provide recommendations for diagnosis/management based on results/findings (e.g. guidance from centres of excellence) ● recommendations provided by national research groups and metabolic care centres (e.g. British Inherited Metabolic Diseases Group) ● guidance described in detail in conference proceedings. 	<ul style="list-style-type: none"> ● guidance that only addresses screening for MCAD/VLCAD deficiency and not diagnosis and/or management ● recommendations related to other IMD where FAOD are not discussed separately ● documents published in languages other than English or French.

For the final set of included papers, one reviewer (AK) abstracted key data into an excel document. A second reviewer (AF) verified all entries. We abstracted data regarding publication characteristics (e.g., title, year, authors), methods of guideline development, and the content of specific guidance provided regarding diagnosis and management of MCAD and/or VLCAD deficiency (Appendix A).

Appraisal of the quality of guidance documents

We used the AGREE II tool, an online reliable and valid tool, to appraise and report on quality (Appendix C).²⁸ This tool consists of 23 items, each scored on a scale of 1-7, within 6 domains: Scope and Purpose, Stakeholder Involvement, Rigour of Development, Clarity of presentation, Applicability and Editorial Independence. Two reviewers (AK and BP) pilot-tested the AGREE II tool with a sample of four included guidance documents. One reviewer (AK) then applied the tool to the remaining documents, with the extracted information and scores verified by a second reviewer (AF). Following the approach published on the AGREEII tool website,²⁸ scaled domain scores were calculated by summing the individual item scores in each domain and applying the following formula,

$$\frac{\text{Obtained score} - \text{Minimum possible score}}{\text{Maximum possible score} - \text{Minimum possible score}} \times 100$$

We calculated the minimum, maximum, and median percentage domain scores for all guidance documents and then separately for peer-reviewed literature and grey literature sources.

Results

Our search strategy yielded 885 references from Embase and 341 from MEDLINE for a total of 1226 citations that were exported from OVID into the Covidence online software for screening. There were no results deemed relevant from the National Guideline Clearinghouse, the KT PLUS McMaster database and Turning Research Into Practice (TRIP) database. The Guidelines International Network was searched after the data extraction was complete, based on the recommendation of an experienced guideline review member (IG), however no relevant results were identified.

A total of 986 unique records from the peer-reviewed literature search were screened in the first phase of screening, of which 918 were excluded as clearly not relevant to the review. The remaining 69 records from the peer-reviewed search were subjected to full-text screening (1 article was excluded as the full-text could not be obtained), along with 9 documents from the grey literature search. The screening process yielded 25 individual guidance documents that met our inclusion criteria, presented in the PRISMA flow chart (Figure 1).²⁹ Upon review of the

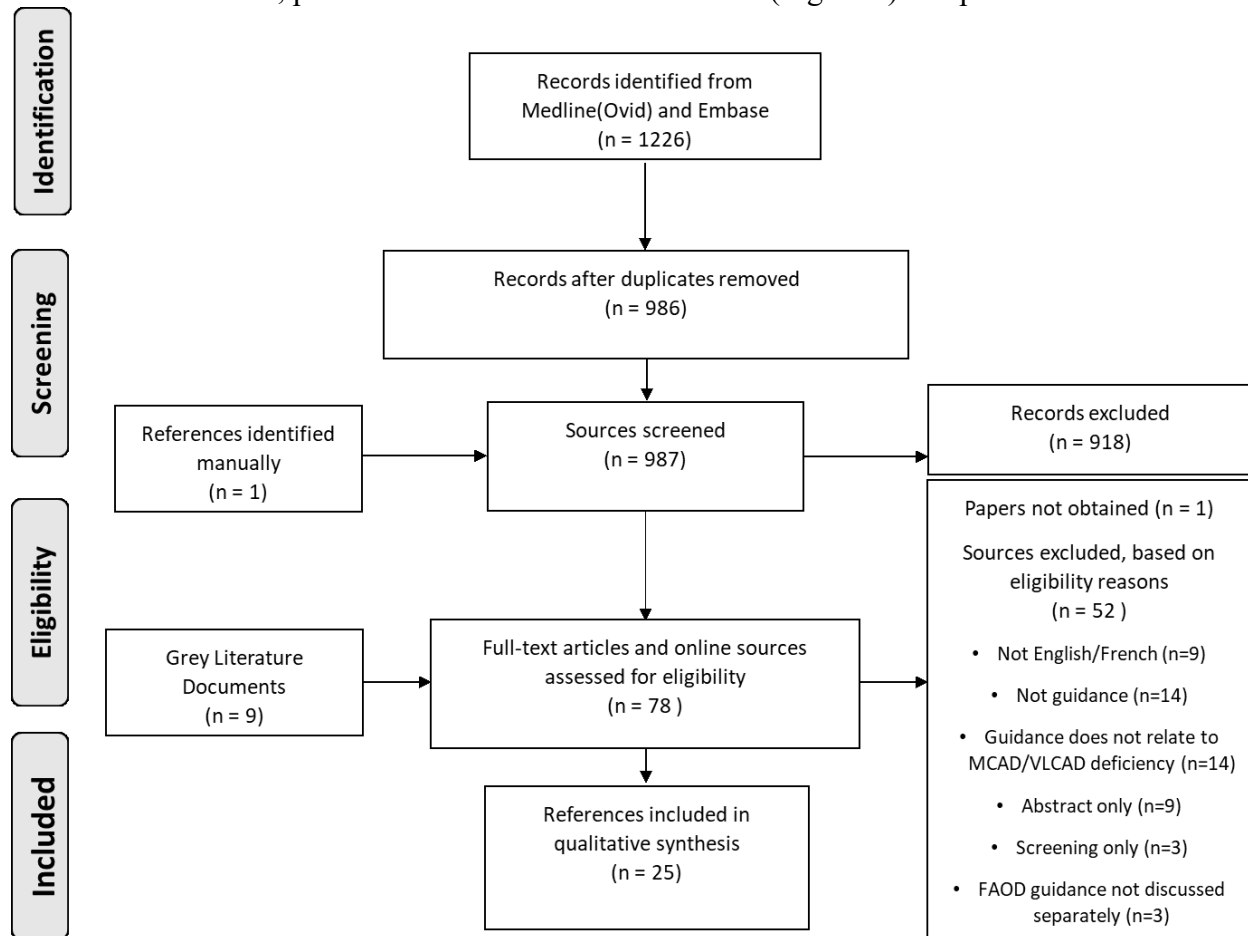


Figure 1. PRISMA flow chart for systematic review.

recommendations provided within these documents, and the structure and organization of the documents themselves, the focus of the guidance and the reporting of their methods seemed to differ according to whether they were published in the peer-reviewed or grey literature. Thus, in some cases, when summarizing the characteristics of the guidance documents in tables, we chose

to separate them according to the source from which they were obtained (peer-reviewed versus grey literature source).

Of the 25 guidance documents, 16 were identified from the peer-reviewed literature, whereas 9 were identified from the grey literature (Tables 2 and 3). Guidance we reviewed from the peer-reviewed literature was published from 1999-2016 while the grey literature guidance was published or last updated (according to the information provided at the source) from 2007-2015. A majority of all guidance documents (peer-reviewed and grey literature) were published in the US (n=9) and UK (n=7), while others were published in Australia (n=1), Central Europe(n=1), France (n=1), Mexico (n=1), Netherlands (n=1) Scotland (n=1) or were the result of an international collaboration (n=3) (Tables 2 and 3).

Ten guidance documents provided recommendations about the diagnosis or management of MCAD deficiency only, 2 provided recommendations about VLCAD deficiency only, and 13 provided recommendations about both diseases (some guidance documents from the grey literature presented recommendations for both MCAD and VLCAD deficiency but did so separately; in such cases we counted the document a single time in Figure 1 and here, but abstracted content separately by disease) (Tables 2 and 3). Peer reviewed guidance (Table 2) covered diagnosis (n=4), management (n=5) or both (n=7), in varying detail. Guidance documents from the grey literature (Table 3) were more likely to focus solely on disease management (n=5), but some covered both diagnosis and management (n=4).

A minority of guidance documents were explicitly developed based on a review of existing evidence (n=3), consensus methods (n=2), or a combination of evidence review and consensus (n=4) (Tables 2 and 3). Details regarding the method of development were not

reported for the remaining documents (n=16). Methods of development were most likely to go unreported for guidance documents from the grey literature (n=7) (Table 3).

Table 2. Characteristics of peer-reviewed papers included in the systematic review of guidance for diagnosis and management of MCAD and VLCAD deficiencies.

Title	First author	Year of publication	Country of release	Journal of publication	Disorder(s) addressed	Diagnosis or management focus	Method of development
A Delphi clinical practice protocol for the management of very long chain acyl coa dehydrogenase deficiency	Arnold GL	2009	International (US and Canada)	Mol Gen Metab	VLCAD deficiency	Both	Combination* (evidence based, consensus)
Investigation of the child with an acute metabolic disorder	Cook P	2011	UK	J Clin Pathol	MCAD deficiency	Diagnosis	Unclear**
Safe and Unsafe duration of fasting for children with MCAD deficiency	Derks TGJ	2007	Netherlands	European J Pediatrics	MCAD deficiency	Management	Evidence based
National Academy of Clinical Biochemistry Laboratory Medicine Practice Guidelines: Follow-up Testing for Metabolic Disease Identified by Expanded Newborn Screening Using Tandem	Dietzen DJ	2009	US	Clin Chem	Both	Diagnosis	Combination (evidence based, consensus***)

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Medium-chain acyl-CoA-dehydrogenase (MCAD) deficiency: French consensus for neonatal screening, diagnosis, and management (FRE)	Feillet F	2012	France	Archives de Pédiatrie	MCAD deficiency	Both	Consensus
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Tandem Mass Spectrometry Newborn Screening for Inborn Errors of Intermediary Metabolism: Abnormal Profile Interpretation	Fernandez-Lainez C	2012	Mexico	Curr Med Chem	Both	Diagnosis	Evidence based
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An overview of Expanded Newborn Screening for Inborn Errors of Metabolism	Goodin B	2006	US	Practical Gastroenterology	MCAD deficiency	Both	Unclear
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The Clinical Aspects of Newborn Screening: Importance of Newborn Screening Follow-up	James PM	2006	US	Ment Retard Dev Disabil Res Rev	Both	Both	Unclear
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Immunizations for patients with metabolic disorders	Kingsley JD	2006	US	American Academy of Pediatrics	MCAD deficiency	Management	Evidence based
Newborn screening for medium chain acyl CoA dehydrogenase deficiency	Leonard JV	2009	UK	Arch Dis Child	MCAD deficiency	Both	Unclear
Fatty Acid Oxidation Defects as a Cause of Neuromyopathic Disease in Infants and Adults	Olpin SE	2005	UK	Clin Lab	Both	Both	Unclear
Inborn errors of metabolism (metabolic disorders)	Rice GM	2016	US	Pediatrics in Review	Both	Both - limited detail	Unclear
Mitochondrial Fatty Acid oxidation disorders and cyclic vomiting syndrome	Rinaldo P	1999	US	Digestive diseases and sciences	Both	Diagnosis - limited detail	Unclear
Treatment recommendations in long-chain fatty acid oxidation defects: consensus from a workshop	Spiekerkoetter U	2009	Central Europe	J Inherit Metab Dis	VLCAD deficiency	Management	Combination (Evidence based, consensus)

Current issues regarding treatment of mitochondrial fatty acid oxidation disorders	Spiekerkoetter U	2010	International	J Inherit Metab Dis	Both	Management	Unclear
Tolerance to fast: rational and practical evaluation in children with hypoketonaemia	Walter JH	2009	UK	J Inherit Metab Dis	MCAD deficiency	Management	Unclear

*Combination refers to using a combination of method to develop the guidelines. ** A separate appraisal in AGREEII was done based on details provided from personal communication with Dr. Valerie Walker. ***A consensus is not explicitly stated; however the document refers to evidence grading by task force committee.

Table 3. Characteristics of grey literature sources included in the systematic review of guidance for diagnosis and management of MCAD and VLCAD deficiencies.

Title	Publishing organization	Year of publication	Country of release	Disorder(s) addressed	Focus of content	Method of development
MCADD Dietary management guidelines for dietitians	British Inherited Metabolic Disease Group (BIMDG®)	2007	UK	MCAD deficiency	Management	Unclear
Additional MCADD/VLCADD diagnosis and management guidelines	British Inherited Metabolic Disease Group (BIMDG)	1 st collection of documents on MCADD and VLCADD diagnosis and management, some with dates, others missing	UK	Both	Both	Unclear
Dietitian Letter	Public Health England, National Health Service (NHS)	Last updated 2015	UK	MCAD deficiency	Management	Unclear

Newborn Screening ACT sheets MCADD, VLCADD	American College of Medical Genetics and Genomics (ACMG)	Last updated 2009	US	Both	Both - limited detail ³	Unclear
GeneReviews® MCADD/VLCADD	National Center for Biotechnology Information (NCBI)	Last update 2014 (VLCADD), 2015 (MCADD)	US	Both	Both	Unclear
MCAD, VLCAD	Genetic Metabolic Dietitians International (GMDI)	Last updated 2008	International (Canada and US)	Both	Both	Combination (evidence based, consensus ⁴)
Acute illness protocol FAODs MCADD, VLCADD	New England Consortium of Metabolic Programs (NECMP)	Last updated 2013	US	Both	Management	Unclear
Paediatric acute care guideline FAOD	Princess Margaret Hospital for Children (PMH), Government of Western Australia, Department of Health	2014	Australia	Both	Management	Consensus ⁵

Dietary management guidelines for MCADD²	IMD Scotland, National Health Service (NHS) Scotland	Not available	Scotland	MCAD deficiency	Management	Unclear
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¹References section provides details on which specific guideline documents from BIMDG were used. ²The guideline document was removed from the website, however a link is provided in the reference to an available online copy. ³Limited detail means that diagnosis and/or management were not covered in great detail. ⁴Stated that members of a task force created guidelines after evidence review. ⁵Guideline development was completed by a Guidelines Team, using NSQHS standards. No other details found.

Quality of Guidance

AGREEII results are presented in Tables 4 and 5. Guidance that met all criteria within a domain for the AGREEII tool received a score of 100%, whereas guidance that contained no information to determine if criteria were met and guidance that did not at all meet criteria received a score of 0%.²⁸

Overall, guidance documents received the highest median scores within the domains covering ‘Scope and Purpose’ (statement of aims, health questions, target population), and ‘Clarity of Presentation’ (language structure and format): medians of 83% (range 39-100%) and 83% (range 33-94%), respectively (Table 4). The lowest median score was in the domain of ‘Applicability’ (median score 17%, range, 4-29%), which reflects the extent to which guidance documents address barriers and facilitators to their implementation and strategies of dissemination and cost, followed by ‘Rigour and Development’ (median score 19%, range 6-52%), which evaluates the authors’ description of methods used for data collection, synthesis and development of recommendations. The other two domains against which the guidance was appraised were ‘Stakeholder Involvement’ (median score 39%, range 11-50%), which describes the extent to which guidelines are developed by the appropriate stakeholders and reflect views of the users or these guidelines; and ‘Editorial Independence’ (median score 25%, range 0-100%), which assesses bias and conflict of interest (Table 4).

We found that median domain scores were generally similar between sources of guidance documents with the exception of ‘Editorial Independence’, where guidance in the grey literature had much lower scores relative to the peer-reviewed literature (median 8% compared to 42%). Overall, there was a wide range of scores across the guidance documents for many domains (Table 5)

Table 4. Domain Scores for AGREEII: Overall and by Source

	Scope and purpose	Stakeholder involvement	Rigour and development	Clarity of presentation	Applicability	Editorial independence
1. SUMMARY OF ALL GUIDANCE (Median and Range of Domain Scores) (n=28)						
Median	83%	39%	19%	83%	17%	25%
Min	39%	11%	6%	33%	4%	0%
Max	100%	50%	52%	94%	29%	100%
2. SUMMARY OF GUIDANCE FROM PEER-REVIEWED LITERATURE (Median and Range of Domain Scores, n=16)						
Median	83%	36%	20%	83 %	15%	42%
Min	39%	11%	6%	33 %	4%	0%
Max	100%	50%	48%	94 %	21%	100%
3. SUMMARY OF GUIDANCE FROM GREY LITERATURE (Median and Range of Domain Scores, n=12*)						
Median	78%	44%	19%	83%	21%	8%
Min	50%	11%	8%	50%	4%	0%
Max	100%	50%	52%	89%	29%	25%

* There were only 9 grey literature documents but three of these contained completely separate information for MCAD deficiency and VLCAD deficiency thus these were appraised separately, yielding 12 items for appraisal.

Table 5. Domain Scores for AGREEII for Individual Guidance Documents from both Peer-Reviewed and Grey Literature

	Scope and purpose	Stakeholder involvement	Rigour and development	Clarity of presentation	Applicability	Editorial independence
1. INDIVIDUAL GUIDANCE DOCUMENTS FROM PEER-REVIEWED LITERATURE (Individual Domain Scores)						
Arnold GL	89%	33%	38%	94%	8%	8%
Cook P*	83%	22%	19%	89%	21%	50%
Derks TGJ	100%	33%	48%	89%	13%	8%
Dietzen DJ	100%	39%	44%	94%	21%	92%
Feillet F	100%	17%	17%	72%	13%	58%
Fernandez-Lainez C	100%	44%	48%	89%	21%	42%
James PM	83%	50%	19%	67%	8%	25%
Kingsley JD	83%	44%	19%	83%	13%	50%
Leonard JV	61%	39%	25%	72%	21%	100%
Olpin SE	61%	11%	19%	72%	13%	0%
Goodin B	89%	50%	13%	33%	17%	0%
Rice GM	94 %	50%	10%	72%	17%	58%
Rinaldo P	61%	17%	6 %	44%	4%	0%
Spiekerkoetter U '09	83%	11%	29%	94%	17%	58%
Spiekerkoetter U '10	78%	44%	48%	89%	21%	42%
Walter JH	39%	11%	21%	83%	4%	42%

*From personal communication with the author, taking into consideration the details provided regarding their search, Rigor of Development would score 29%. As this was not published, this value is not considered in any other calculations.

2. INDIVIDUAL GUIDANCE DOCUMENTS FROM GREY LITERATURE (Individual Domain Scores)

BIMDG - dietitians' guidelines	100%	50%	15%	89%	13%	25%
BIMDG-all other documents	50%	22%	13%	83%	21%	0
NHS Dietitian letter	50%	11%	8%	67%	4%	0
GMDI MCADD	78%	33%	52%	83%	25%	25%
GMDI VLCADD	78%	33%	52%	83%	25%	25%
GeneReviews MCADD	67%	44%	50%	61%	29%	8%
GeneReviews VLCADD	67%	44%	50%	61%	29%	8%
ACT-sheets MCADD	72%	44%	19%	61%	8%	25%
ACT-sheets VLCADD	72%	44%	19%	61%	8%	25%
NECMP MCADD	83%	50%	8 %	83%	21%	0%
NECMP VLCADD	83%	50%	8 %	83%	21%	0%
PMH	78%	33%	19%	50%	8%	0%
Scotland NHS	83%	33%	10%	89%	13%	0%

Content of recommendations contained in guidance regarding MCAD and VLCAD deficiencies

Details of recommendations from each of the guidance documents are presented in data tables separately by peer-reviewed and grey literature, and divided into diagnostic information, acute management, long term management and additional/other guidelines (Appendix A Tables A.S2-A.S17). Here we summarize the similarities and differences across guidance documents within each of these categories. *A consistent recommendation* was defined for our purposes as a recommendation that was discussed in more than one document, and where all documents presented similar recommendations – i.e., all of the guidance on this topic generally agreed, with no conflicting recommendations. *Conflicting recommendations* highlight the areas of controversy or differing recommendations – i.e., these are topics that were discussed in multiple documents, where there were discrepancies in the recommendations across documents. Counts in the summaries below are presented in relation to the denominator for each summary (e.g., when summarizing specific recommendations about diagnostic testing for MCAD deficiency, the denominator reflects all guidance documents that discuss diagnostic testing for MCAD deficiency), as highlighted in Tables 2 and 3.

MCAD Deficiency:

Diagnostic testing for MCAD deficiency was discussed in 14 guidance documents (Appendix Tables A.S2 and A.S3):

Consistent recommendations regarding MCAD deficiency diagnosis

Consistently recommended diagnostic strategies included the following specific tests:

- Plasma acylcarnitines (AC): 13/14 documents^{12,13,15,30–39}
- Urine organic acids (UOA): 11/14 documents^{12,13,15,30,32,34–36,38,40}
- Molecular testing: 11/14 documents^{12,13,30–32,34–39}
- Urine acylglycines (UAG): 6/14 documents^{12,13,34,36,37,39}
- Functional (i.e. enzyme) analysis: 6/14 documents^{12,30,32,34–36}
- Blood glucose: 6/14 documents^{15,30,34,36,37,39}
- Plasma carnitine: 5/14 documents^{13,15,34,36,37}
- Liver function tests (LFT) and assessments of electrolytes and blood gas: 2/14 documents^{37,39}

Two documents^{13,40} also referred to another source which itself endorsed plasma AC, UOA, UAG, blood glucose, LFT, electrolytes and blood gas.³⁹

Conflicting recommendations regarding MCAD deficiency diagnosis

The only area of disagreement noted was that while one guidance document³⁶ stated that fibroblast fatty acid oxidation (FAO), detected using assays or acylcarnitine profiling, is rarely used for confirmatory functional analyses, there were 4/14 documents that discussed fibroblast enzyme analysis for confirmatory testing.^{12,30,32,34}

Acute/Emergency care for MCAD deficiency was discussed in 16 guidance documents, with documents identified from the grey literature tending to provide more detailed descriptions of acute care protocols relative to those published in the peer-reviewed literature (Appendix Tables A.S4 and A.S5):

Consistent recommendations regarding acute/emergency care for MCAD deficiency

Sick day protocols or emergency regimens were explicitly described for parents or for emergency staff in 14/16 guidance documents^{12,13,15,16,32,35,36,38,40-45}, although there was varying detail about the specifics of these protocols and plans. Commonly mentioned sick day or emergency management strategies were: (i) a recommendation to provide a glucose polymer source/high carbohydrate beverage (10/16 of documents)^{12,13,15,35,36,38,41-44}, with some noting that oral rehydration solutions (ORS) alone are usually insufficient to maintain appropriate plasma glucose levels (6/16 documents)^{13,35,38,41-43}; and (ii) a recommendation to use intravenous glucose infusion for emergency treatment when a child is not feeding well, based on symptoms or for surgical procedures (10/16 documents).^{12,13,15,32,33,35,38,42,44,45}

One component of the emergency plan that was mentioned by 4/16 documents^{12,38,44,45}, all of which were from the grey literature, was the recommendation to contact or consult metabolic specialists upon a child's admission to hospital. Similarly, 4/16 documents described specific tests for acute decompensation monitoring upon admission; these included blood glucose, electrolytes, and blood gas, ammonia, LFT.^{38,42,44,45}

Conflicting recommendations regarding acute/emergency care for MCAD deficiency

While 4/16 documents mentioned administration of carnitine for acute care for individuals with MCAD deficiency, recommendations were discrepant.^{13,32,44,45} One document⁴⁵ recommended supplementation at the discretion of the physician given the controversy around carnitine use, two others^{32,44} recommended carnitine at varying doses and one¹³ suggested providing carnitine if a patient's carnitine levels were low or for pregnant women going into labor and until normal feeding is re-established. Nasogastric tube feeding was described by 3/16

guidance document but recommendations also differed for this intervention: in one document¹⁶ it was not recommended; in a second document³⁸ it was recommended when feeding requirements were not met for a max of 23-36 hours; and in a third document⁴⁴, the nasogastric tube was described as an option if a child was not encephalopathic and unwell.

Long-term preventive management for MCAD deficiency was discussed in 15 guidance documents (Appendix Tables A.S6, A.S7, A.S8, and A.S9):

Consistent recommendations for long-term preventive management of MCAD deficiency

Avoidance of fasting was universally described as the foundation of management for MCAD deficiency, to avoid metabolic decompensation, particularly during times of illness (15/15 documents).^{10,12,13,15,16,32–36,38,41–43,46} Of documents stating this, 9/15 provided some information regarding fasting schedules.^{10,13,16,32,33,38,41,43,46} Cornstarch supplementation before bedtime was discussed in 3/15 documents.^{12,13,33} In addition, 7/15 documents^{12,13,36,38,41–43} specifically noted that infants with MCAD deficiency should not receive formula with medium chain triglycerides and/or coconut oil. Also relevant to feeding, 6/15 recommended that both breastfeeding and formula feeding were appropriate for infants with MCAD deficiency.^{13,32,38,41–43} No document definitively recommended carnitine supplementation for all patients as part of long-term management. Furthermore, several documents mentioned the importance of parental education about issues such as avoidance of fasting while guarding against excessively frequent feeds and recognition of symptoms of decompensation (7/15 guidelines).^{13,15,35,39,41–43} Monitoring weight for obesity and avoiding over eating was discussed by 3/15 documents (Appendix Tables A.S8 and A.S9).^{12,13,35} Finally, 3/15 documents recommended using a medical alert bracelet (Table A.S9).^{13,41,44}

Conflicting recommendations for long-term preventive management of MCAD deficiency

Fat content of the diet was described in 6/15 documents, however, this varied by document, with some mentioning ranges and/or limits, following ‘low-fat’ or ‘heart healthy’ diets or general comments about exclusion/limiting of some fat.^{12,13,15,32,33,35} Carnitine supplementation for long-term management was discussed by 9/15 documents.

Recommendations about carnitine use were variable: four documents recommended carnitine supplementation if a deficiency was present,^{13,30,32,36} while five other documents were unclear in their recommendations with respect to the circumstances when carnitine is appropriate.^{15,16,33–35}

VLCAD Deficiency:

Diagnostic testing for VLCAD deficiency was discussed in 11 guidance documents (Appendix Tables A.S10 and A.S11):

Consistent recommendations regarding diagnostic testing for VLCAD deficiency

Consistently recommended diagnostic strategies included the following specific tests:

- Plasma acylcarnitines (AC): 8/11 documents^{12,14,15,31,34,36,47,48}
- Urine organic acids (UOA): 4/11 documents^{34,36,40,48}
- Hypoglycemia: 6/11 documents^{3,12,15,34,36,47}
- Plasma carnitine: 5/11 documents^{3,34,36,37,48}
- Molecular testing: 7/11 documents^{3,14,15,34,40,47,48}

A 9th document referred to another source, which itself recommended plasma AC.³ Measurement of urine acylglycines (UAG), electrolytes, lactate, ammonia, blood gases, liver function tests

(LFT) and creatinine phosphokinase (CPK) were also mentioned in some documents^{3,34,37,47} as laboratory tests that were helpful for diagnosis.

Conflicting recommendations regarding diagnostic testing for VLCAD deficiency

Again, only one area of disagreement was identified: one guidance document³¹ noted that an enzyme activity assay was not generally available, whereas 9/11 documents discussed functional enzyme tests for diagnosis.^{4,14,14,15,34,36,47-49}

Acute/Emergency care for VLCAD deficiency was discussed in 10 guidance documents (Appendix Tables A.S12 and A.S13):

Consistent recommendations regarding acute/emergency care for VLCAD deficiency

Most of the VLCAD guidance documents that addressed acute care mentioned sick day protocols or emergency regimens (8/10 documents)^{4,14,15,36,44,48,50,51} with a 9th document referring to a source which itself provides a protocol.³ Providing a glucose polymer source/high carbohydrate beverage (5/10 documents)^{15,36,44,48,50} and using intravenous glucose for emergency treatment when a child is not feeding, based on symptoms (9/10 documents)^{3,4,14,15,44,47,48,50,51} were also widely recommended.

Acute monitoring for creatine kinase (CK) levels was mentioned by 5/10 guidance documents.^{3,14,14,15,50} Tests for acute decompensation monitoring upon admission, which included measurement of blood glucose, electrolytes, blood gas, LFT and ammonia, were discussed by 5/10 documents,^{3,14,44,50,51} as was cardiac monitoring (5/10 documents).^{3,14,50,51,51} As with MCAD deficiency, generally, documents identified from the grey literature provided more detailed descriptions of acute care compared to those published in the peer-reviewed literature.

Conflicting recommendations regarding acute/emergency care for VLCAD deficiency

Administration of carnitine for acute care was discussed by one guidance document,⁴⁸ whereas another stated that carnitine use during severe metabolic illness should be avoided.⁴

Long-term preventive management for VLCAD deficiency was discussed in 10 guidance documents (Appendix Tables A.S14, A.S15, A.S16 and A.S17):

Consistent recommendations regarding long-term preventive management of VLCAD deficiency

Nearly all guidance documents (9/10 documents)^{3,4,14,15,34,36,48-50} endorsed prevention of fasting as the main treatment for VLCAD deficiency, however of these, only 2 provided specific fasting schedules.^{4,48} 4/10 documents mentioned essential fatty acid (EFA) supplementation,^{4,36,48,49} and all suggested MCT supplementation in variable forms and amounts (10/10 documents).^{3,4,14,34,36,48-51} No guidance document definitively recommended carnitine supplementation for all patients with VLCAD deficiency as part of long-term care. Regular cardiac monitoring was recommended by 3/10 documents.^{4,14,15} Other aspects were discussed in Appendix Tables A.S16 and A.S17, also relevant to long term care. Exercise was discussed in several documents, either in the context of preventing symptoms with additional MCT, or in terms of the need for rest and rehydration post exercise and avoiding high intensity sports (6/11 documents, Appendix Tables A.S16 and A.S17).^{4,14,15,36,48,49}

Conflicting recommendations regarding long-term preventive management of VLCAD deficiency

Fat content of the diet was discussed in all guidance documents (10/10).^{3,4,14,15,34,36,48-51} Of these, some made recommendations based on categorizing VLCAD deficiency according to phenotype severity/family history (5/10 documents),^{3,4,48-50} while others gave more general

recommendations (5/10 documents).^{14,15,34,36,51} Those that specified fat content were variable in their recommendations and there was also variation in recommendations about appropriate percent fat replacement with MCT (6/10 documents).^{3,4,14,14,36,50} Carnitine supplementation was discussed as part of long-term management, with variation in the actual recommendations, by 9/10 guidance documents. Specifically, while one document³ recommended that carnitine supplementation could be required to prevent deficiency, many were unclear about the circumstances under which supplementation would be appropriate.^{4,14,15,34,36,49,51} One document⁵⁰ gave a possible carnitine dose range but stated that carnitine use should generally be avoided while another reported that some centres may supplement with carnitine, and provided a range of appropriate doses.⁴⁸

Discussion

To our knowledge, this is the first systematic review of guidance for the diagnosis and management of MCAD deficiency and VLCAD deficiency. We identified 25 guideline documents (16 published in the peer-reviewed literature and 9 identified from our grey literature search), which varied in both quality and content. With respect to quality, based on their appraisal against the criteria outlined in the AGREEII tool, for all guidance documents together, the highest domain scores were for ‘Scope and Purpose’ and ‘Clarity of Presentation’. This indicates that many guidance documents are clearly identifying the intended purpose of the guidelines/recommendations and presenting them effectively. There was however a wide range across individual documents (Table 4). The domain ‘Rigour of guideline development’ was one of the lowest scoring domains for this review. This suggests that existing guidance for the diagnosis and management of MCAD and VLCAD does not meet generally acceptable standards for clinical practice guideline development. A recent international study of clinical practice

guidelines (CPG) on various topics also used AGREEII to appraise guideline quality; results from our review indicate much lower scores for 5 of the 6 AGREEII domains (the exception was ‘Scope and Purpose’, where our median score was 83% compared to a mean score of 75.8% in the international review).⁵² It is important to state that there is no evidence to specifically link subscale scores for AGREEII to clinical or implementation outcomes, rendering it challenging to set specific thresholds for guideline acceptability (i.e., it is not possible to specifically define the subscale score at which a guideline can be deemed to be high quality).²⁸ Although most of the guidance documents we reviewed did not self-label as “clinical practice guidelines”, they did provide recommendations for care and the low AGREE II scores we observed in some domains, particularly for methodological rigour, speak to the absence of high-quality, evidence-based guidance for disease management in this field.

Contributing to the low scores on the AGREE II items within the domain related to rigour of development, only 7 of the 25 guidance documents we reviewed were explicitly reported to be wholly or partially based on a review of evidence (Tables 2 and 3). This may reflect the limited available evidence supporting diagnostic testing and treatment for these rare diseases. However, with respect to the content covered within the guidance documents, for some of the most widely recommended practices that were areas of agreement across guidance documents, there is some available evidence. For example, one of the documents we reviewed was from the National Academy of Clinical Biochemistry committee and was based on an evaluation of the literature to grade diagnostic markers for MCAD and VLCAD deficiencies, among other disorders, using the criteria from the US Preventive Task Force.³¹ Some of the diagnostic practices, for example, confirmatory genetic testing for the most common mutation associated with classical MCAD deficiency (A985G), were based on “AI”-grade evidence.³¹ A

further example where recommendations within the guidance documents were in agreement and are supported by evidence relates to the use of emergency protocols, including emergency letters for families to present upon arrival to hospital and strategies for managing intercurrent illness so as to avoid and reduce the severity of metabolic crises.⁵³

Areas where guidance documents provided conflicting advice suggest more worrying areas of uncertainty for clinical providers who care for patients diagnosed with, or facing a potential diagnosis of, these fatty acid oxidation disorders. Given the scarcity of empirical evidence to support advice in these areas, they could be prioritized for future evidence generation. For example, and of particular interest to us, in the guidance we reviewed, fasting avoidance was routinely identified as the most important single strategy for managing FAODs in order to prevent acute crises, but there was considerable variation in the specific fasting schedules or fasting times when these were specified. Obesity/excessive weight has been identified as a concern in older children with MCAD deficiency and VLCAD deficiency,^{5,10,13,48} highlighting that fasting schedules need to be developed to manage the risk of decompensation associated with prolonged fasting while also avoiding over-feeding. While a number of different fasting schedules were identified for both disorders among the guidance documents,^{4,13,32,33,38,41,43,48,49} Derks et al. (2007) provided the only schedule for children with MCAD deficiency that was reported to be based on empirical evidence (from fasting studies).¹⁰ One additional guidance document compiled various fasting schedules from the literature and personal communications.⁴⁶ Fasting studies are challenging because they carry some risk, rendering it difficult to generate high-quality evidence to inform this important area of treatment uncertainty.

A second content area that we had identified as being of particular interest was the use of carnitine. Among the guidance documents, carnitine use was discussed in the context of emergency administration as well as long term usage. Some guidance recommended carnitine use but did not give specific criteria for its indication,^{4,14,15,33–36,49,51} while other documents recommended carnitine only when a patient with an FAOD has low plasma carnitine levels or a carnitine deficiency.^{3,13,30,32,36} No guidance recommended carnitine supplementation definitively for all patients as part of either acute or chronic management. We also identified recommendations against the use of carnitine, with guidance authors stating a lack of evidence of clinical benefit and no controlled trials.^{35,49} A Cochrane review on the use of carnitine for inherited metabolic diseases concluded that there was insufficient evidence available to recommend carnitine supplementation.²⁴ Reports of adverse effects from carnitine supplementation range from potential acute effects such as nausea and odor, which are relatively mild, to more severe longer-term effects such as a possible risk of atherosclerosis,^{54–56} further highlighting the importance of developing the evidence base and consequently relying on it to guide disease management.

Finally, although the guidance documents overall had relatively high scores for the AGREEII domain of ‘Scope and Purpose’, they frequently did not provide concrete recommendations in topic areas of greatest interest to health care providers and families of patients with FAOD. For example, there was limited consideration of disease severity, including the potential to differently manage asymptomatic disease in patients predicted to have milder cases based on genotype or other criteria. An exception was that some authors have categorized VLCAD deficiency into 3 phenotypic categories^{7,8,57} and some guidance documents provided recommendations separately for these categories (e.g., regarding low fat diets, and MCT oil or

formula).^{3,4,48-50} This generally limited availability of guidance tailored to predicted disease severity may again reflect the lack of an empirical basis on which such guidance might be formulated, particularly given the imperfect correlation of genotype with clinical risk for both diseases and the lack of other robust markers of predicted disease severity.

One of the main limitations of this systematic review was in developing a search strategy that would identify clinical practice guidelines, especially given that so much of the guidance about the diagnosis and management of MCAD and VLCAD deficiencies was not specifically self-identified as 'clinical practice guidelines'. Our searches and screening process may thus have missed some guidance documents. Another limitation was that only one reviewer extracted and appraised the data, even though a second reviewer verified extractions and AGREEII scores. This could result in some mis-classification or missing information. Finally, we were limited to the amount of detail provided by the authors of the guidance documents and thus our quality appraisal reflects both actual methods and reporting of methods of guidance development.

We found that existing guidance for the diagnosis and management of MCAD deficiency and VLCAD deficiency was not generally reported to be developed using robust evidence reviews and did not meet established criteria for the rigorous development of clinical practice guidelines. With respect to content, there was more agreement with respect to recommendations about diagnostic care relative to disease management among guidance documents for both conditions. We identified important variation in recommendations regarding key areas of preventive care in particular, including fasting times and the use of carnitine supplementation. Further empirical evidence in these areas is necessary to inform the development of future rigorous guidelines. However, such evidence is challenging to produce in part due to the

potential risk of evaluating a less cautionary approach for asymptomatic cases of disease identified by newborn screening that are predicted to have a milder phenotype.^{58,59}

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Reference 38, Additional BIMDG documents, included the following guidance documents, by title/and or description when no title was available:

- **MCADD clinical management guidelines: Presumptive positive MCADD**
- **MCAD deficiency: Management of newborn babies with a family history**
- **Adult emergency management Medium chain fatty acid oxidation disorders (MCADD)**
- **Adult emergency management Long chain fatty acid oxidation defects (VLCADD)**
- **General dietary information for emergency regimens**
- **Medium chain fat oxidation disorders/Long chain fat oxidation disorders – Acute decompensation**
- **Protocol for immediate management of MCADD and VLCADD**
- **Emergency regimen recipes by age**
- **Adult emergency management oral emergency regimen (ER)**
- **Making intravenous fluids for metabolic patients**
- **Child's Glasgow Coma Scale**
- **Management of surgery in children with disorders of fatty acid oxidation (MCADD and VLCADD)**
- **Rhabdomyolysis in young children**
- **Management of newborn babies with a family history of a fatty acid oxidation disorder (even if only suspected)**

Appendix A

Supplement 1. Grey literature search

A systematic review of existing published treatment guidelines for MCADD and VLCADD - Grey Literature search October 2016

Search using these terms from the database search:

- exp Acyl-CoA Dehydrogenases
- MCAD
- MCADD
- medium-chain acyl-CoA dehydrogenase or Medium-chain acyl-coenzyme A dehydrogenase
- medium chain acyl CoA dehydrogenase or Medium chain acyl coenzyme A dehydrogenase
- VLCADD
- VLCAD
- very-long-chain acyl-CoA dehydrogenase or Very long chain acyl CoA dehydrogenase
- acyl coa dehydrogenases or acyl-coa dehydrogenases or dehydrogenases acyl-coa
- Lipid Metabolism, Inborn Errors
- Fatty Acid Desaturases/
- fatty acid oxidation disorders
- FAOD or LC-FAOD
- Mitochondrial fatty acid oxidation
- FAO defects

Databases searched:

National Guidelines Clearinghouse

<https://guidelines.gov>

KT PLUS (McMaster)

<https://plus.mcmaster.ca>

Search engines searched:

TRIP database: <http://www.tripdatabase.com/>

Google: https://www.google.ca/advanced_search

Google Scholar: <https://scholar.google.com/intl/en/scholar/about.html>

Specific sites searched:

Babys first: <http://www.babysfirsttest.org/newborn-screening/conditions/very-long-chain-acyl-coa-dehydrogenase-deficiency> AND <http://www.babysfirsttest.org/newborn-screening/conditions/medium-chain-acyl-coa-dehydrogenase-deficiency>

British inherited metabolic group: <http://www.bimdg.org.uk/site/guidelines.asp>

Genetic metabolic dieticians: <http://www.gmdi.org/Resources/Nutrition-Guidelines/MCAD>

Global genes rare diseases: <https://globalgenes.org/rare-diseases-facts-statistics/>

Kids Health WA: <http://kidshealthwa.com/guidelines/fatty-acid-oxidation-disorders/>

MCADD-UK: <http://www.mcadd-uk.net>

Medical home portal: <https://www.medicalhomeportal.org/diagnoses-and-conditions/mcadd>

National Newborn Screening and Global Resource Centre: <http://genes-r-us.uthscsa.edu>

NIH Genetics: <https://ghr.nlm.nih.gov/condition/very-long-chain-acyl-coa-dehydrogenase-deficiency> AND <https://ghr.nlm.nih.gov/condition/medium-chain-acyl-coa-dehydrogenase-deficiency>

NIH rare diseases: <https://rarediseases.info.nih.gov/diseases/5508/vlcad-deficiency> AND <https://rarediseases.info.nih.gov/diseases/540/medium-chain-acyl-coenzyme-a-dehydrogenase-deficiency>

[GeneReviews](#): provides current, expert-authored, peer-reviewed, full-text articles describing the application of genetic testing to the diagnosis, management, and genetic counseling of patients with specific inherited conditions.

[Orphanet Emergency Guidelines](#): expert-authored and peer-reviewed that is intended to guide health care professionals in emergency situations involving this condition

[Genetic Metabolic Dietitians International \(GMDI\)](#): developed nutrition guidelines for multiple metabolic conditions.

NHS (UK): <http://www.nhs.uk/Conditions/MCADD/Pages/Introduction.aspx>

NHS (UK):

https://www.gov.uk/government/uploads/system/uploads/attachment_data/file/511514/MCADD_confirmed_A5_version.pdf

NHS (UK):

<http://www.imd.scot.nhs.uk/Documents/Guidelines/MCADD%20Dietary%20Guidelines%20-%20Sept%202010%20v1%200.pdf>

Newborn screening:

<http://www.newbornscreening.info/Parents/fattyaciddisorders/MCADD.html> and

<http://www.newbornscreening.info/Parents/fattyaciddisorders/VLCADD.html>

Newborn Screening Ontario: <https://www.newbornscreening.on.ca/en/disease/medium-chain-acyl-coa-dehydrogenase-deficiency-mcadd>

New England Consortium: <http://newenglandconsortium.org/for-professionals/acute-illness-protocols/fatty-acid-oxidation-disorders/medium-chain-acyl-coa-dehydrogenase-deficiency-mcadd/>

ORPHA net: <http://www.orpha.net/consor/cgi-bin/index.php> and

<http://www.orpha.net/national/CA-EN/index/homepage/>

Rare best practice: <http://www.rarebestpractices.eu>

Rare Diseases: <https://rarediseases.org>

Rare disease EU: <http://www.rare-diseases.eu>

Rare diseases FDA:

<http://www.fda.gov/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/ucm423881.htm>

Rare diseases foundation: <https://www.rarediseasefoundation.org>

Rare disease international: <http://www.rarediseasesinternational.org>

Rare disorders: <https://www.raredisorders.ca>

Save babies: <http://www.savebabies.org/resources.html>

Sick Kids: <http://www.sickkids.ca/CGenetics/What-we-do/Newborn-screening-program/index.html>

Canada – provinces - screening

<http://www.health.alberta.ca/professionals/newborn-metabolic-screening.html>

<http://www.perinatalervicesbc.ca/our-services/screening-programs/newborn-screening-program>

<https://www.newbornscreening.on.ca>

<http://www.gov.mb.ca/health/publichealth/cpl/baby.html>

<https://www.saskatchewan.ca/government/health-care-administration-and-provider-resources/saskatchewan-disease-control-laboratory/laboratory-services>

http://www4.gouv.qc.ca/EN/Portail/Citoyens/Evenements/DevenirParent/Pages/progr_depst_neo_nt_sangn.aspx

<http://www.iwk.nshealth.ca/newbornscreening>

<http://www.easternhealth.ca/WebInWeb.aspx?d=2&id=1965&p=1861#PPNL>

General Grey:

Government of Canada: <http://publications.gc.ca/site/eng/search/eCollection.html>

GreyNet International: <http://www.greylit.org>

SIGLE (System for Information on Grey Literature in Europe): <http://www.opengrey.eu>

National Technical Information Service (NTIS): <http://www.ntis.gov>

International databases:

Agency for Healthcare Research and Quality (see for policy makers):

<http://www.ahrq.gov/research/index.html>

LILACS - Latin-American and Caribbean Center on Health Sciences Information:

<http://lilacs.bvsalud.org/en/>

WHO (WHOLIS):

<http://dosei.who.int/uhtbin/cgiirsi/Tue+Apr++5+17:45:43+MEST+2016/0/49>

Thesis:

Center for Research Libraries Foreign Dissertation: <https://www.crl.edu/collections/topics/dissertations>

DART-Europe E-theses Portal: <http://www.dart-europe.eu/basic-search.php>

Electronic Theses Online Service (ETHOS) | British Library:

<http://ethos.bl.uk/Home.do;jsessionid=D96E9CF245B0FE0199DDDB94FF4BD2A7>

Open access dissertations: <https://oatd.org>

Thesis Canada Portal: <http://www.bac-lac.gc.ca/eng/services/theses/Pages/theses-canada.aspx>

Tables A.S2-A.S17 Data extractions for peer-reviewed and grey literature sources for systematic review of diagnosis and/or management guidelines for MCAD and VLCAD deficiencies

Note: Each table presents data from only those guidance documents that covered at least one of the topics described in the table's column headers. Where cells have been left blank, the topic described in the column header was not covered in the guidance document.

Table A.S2. MCAD deficiency peer-reviewed diagnostic guidance

Guidance document (First Author)	Cook	Dietzen	Feillet	Fernandez-Lainez	Goodin	James	Leonard	Olpin	Rice	Rinaldo
Guidance Topic										
Acylcarnitines (AC)	Plasma AC	C6, C8, C10 species and plasma AC analysis	Plasma AC Also used for diagnosis confirmation		✓	Plasma AC for follow-up evaluation. Fibroblast AC profiling for diagnosis only if heterozygous or other mutation.	Repeat AC for diagnosis	PAC profile	✓ PAC as part of confirmatory diagnosis	✓ Plasma AC also part of specialized testing
Organic Acids (OA)	✓		Urine OA Also used for diagnosis confirmation	Urine OA		Urine OA for follow-up evaluation	Repeat urine OA for diagnosis	Urine OA	✓	Specialized testing urine OA
Acylglycines (AG)						Urine AG for follow-up evaluation		Urine AG		Specialized testing urine AG
Blood Glucose	Acute testing for hypoglycemia					✓		Plasma glucose	hypoglycemia	Part of routine plasma
Ammonia	✓									Part of routine plasma
Fatty Acids	Plasma FFA: 3-hydroxybutyrate ratio							Non-essential fatty acids (NEFA)		Specialized testing: Plasma free fatty acids, Plasma 3-OH fatty acid

Carnitine						Total/free carnitine		Total and free carnitine	Total and free carnitine	Specialized testing: Plasma free and total carnitine
Gases										Part of routine plasma
Electrolytes										Part of routine plasma
Other follow-up diagnostic and ancillary confirmatory testing								3-hydroxybutyrate (ketone body)	Ketones	Part of routine plasma: Lactate, Pyruvate, Liver function tests, Creatine phosphokinase Routine urine ketones
Enzyme Analysis	For confirmation using skin fibroblasts		Measure activity in leucocytes or fibroblasts only if homozygosity for c.985A>G mutation not detected or only 1 copy detected.				Confirmation with enzymes studies if appropriate.	Fibroblast FAO rarely used for confirmation.		
Mutation analysis	For confirmatory purposes.	Molecular genetic analysis for common A985G mutation.	Confirmatory diagnosis based on Plasma AC and urine OA coupled with c.985A>G analysis.			MCAD genotype	Confirmation with genotype analysis.	DNA (K304E)		Final confirmation by molecular analysis
Other		Provide algorithm for general diagnostic procedure.	Diagnostic and management algorithm provided.	Abnormal results should be accompanied by ACMG ACT sheets and confirmatory algorithm.						

Table A.S3. MCAD deficiency grey literature diagnostic guidance

Guidance document	BIMDG Additional	GeneReviews	ACMG ACT sheets	GMDI
Guidance Topic				
Acylcarnitines (AC)	Plasma AC	Plasma AC (C6-C10 species, particularly C*)	Plasma AC	Plasma AC profile
Organic Acids (OA)	Urine OA	Urine OA	Urine OA	Urine OA
Acylglycines (AG)		Urine AG	Urine AG	Urine AG
Blood Glucose			Part of routine lab	
Carnitine				Plasma carnitine
Gases			Blood gases part of routine lab.	
Electrolytes			Part of routine lab	
Others follow-up diagnostic and ancillary confirmatory testing			Liver function tests part of routine lab.	
Enzyme Analysis		MCAD enzyme activity in leukocytes, fibroblasts, tissues		
Mutation analysis	c.985A>G analysis	Identification of biallelic pathogenic variants in ACADM	MCAD gene analysis.	DNA mutation analysis
Other		Beta oxidation flux in fibroblasts. Refer to diagnostic algorithm from ACMG ACT sheets.		Repeat NBS not sufficient for diagnosis. Refer to diagnostic algorithm from ACMG ACT sheets.

Table A.S4. MCAD deficiency peer-reviewed emergency/acute care guidance

Guidance document (First Author)	Feillet	Fernandez-Lainez	Goodin	Leonard	Olpin	Rice	Spiekerkoetter
Guidance Topic							
Fluids and Solutions				<p>Low calorie drinks not to be given without additional carbs.</p> <p>As per BIMDG (use high carb drinks).</p>	<p>Emergency regimen should always be available - consisting of readily available high carb drinks.</p>	<p>Dextrose (10% with salt). Use fluids early for preventing hypoglycemia.</p>	<p>Supplementation (ex. maltodextrin) in cases of food refusal/illness.</p>
Intravenous Infusions	<p>Surgery: ≤1 yr - 8mg/kg/min glucose, ≤3 - 7mg/kg/min, ≤6 yr - 6.5 mg/kg/min, ≤14 5.5 mg/kg/min, teens - 4.5mg/kg/min, adult - 3.5 mg/kg/min.</p> <p>Decompensation: 0.5-1 g/kg IV glucose at emerge, as per surgery. Maintain glucose >5mmol/L, infuse with insulin.</p>		<p>IV glucose for vomiting, fever, diarrhea.</p>	<p>As per BIMDG (10% glucose IV).</p>		<p>IV fluids if child doesn't feed well during illness.</p>	
Other Carbohydrates	<p>During infection: frequent snacking and low glycemic index foods.</p>			<p>Small quantities given day and night, until recovery.</p>			
Dietary modifications	<p>Avoid milk with MCT if neonate has symptoms.</p> <p>During infection: 10% increase in caloric intake.</p>						

Carnitine	Carnitine for neonates: 20-50 mg/kg/d (in 2 administrations), for others: 100mg/kg/day Levocarnil.						
Contact specialist							Referral to hospital early when recurrent vomiting.
Other	Complications arising from decompensation should be treated to standard, but without delaying glucose and carnitine administration. Parents require emergency letter.						Tube feeding not recommended.
Emergency regimens, protocols and related procedures	Explain emergency protocol to parents in case of symptoms.	Refers to ACMG ACT sheets.		Refers to BIMDG guidelines.		Emergency department protocol should be provided to all affected families.	Family need sick day protocol.

Table A.S5. MCAD deficiency grey literature emergency/acute care guidance

Guidance document	BIMDG Dietitians'	BIMDG Additional	ACMG ACT sheets	Dietitian Letter	NECMP	GeneReviews	PMH	GMDI	IMD Scotland	
Guidance Topic										
Fluids and Solutions	<p>0-12m: 10% CHO Glc polymer drink 1yr: 15% CHO drink 2yr+: 20% CHO drink 10yrs+: 25% CHO drink</p> <p>ORS not to be given without extra Glc for energy</p>	<p>Glc polymer drink with concentration increasing with age (as per BIMDG Dietitians').</p> <p>May add electrolytes</p> <p>Preoperative drinks same concentrations as per BIMDG ages.</p> <p>Re-introduce oral feeds ASAP post surgery.</p>	<p>Post positive NBS, if signs of illness or unwell baby, emergency treatment includes IV Glc.</p>	<p>Glc polymer ER drink.</p> <p>ORS recipe (as per BIMDG).</p>				<p>High carb drink orally or via nasogastric tube if child not encephalopathic and brought to hospital due to food refusal/single vomit.</p>	<p>Acute management: small and frequent meals/fluids.</p> <p>Pedialyte good ORS, with sweetened fluid of dose 1/2tsp sugar/ounce.</p> <p>If no IV carnitine for increased carnitine requirements due to illness, liquid oral carnitine appropriate, may be at double the dosage.</p> <p>Clear liquids >4 hrs before surgery.</p>	<p>ORS not to be given without additional Glc polymer. ER should be given every 2-3hrs day and night when unwell.</p> <p><1yr ER: 10% CHO. 1-2yrs: ER: 15%CHO or prepared drink with at least 10.5g/100ml CHO. 2-9yrs ER: 20%CHO or prepared drink with at least 17g/100ml CHO. 10+ yrs and adults ER: 25%CHO or prepared drink with at least 17g/100ml CHO. Feed volumes by age provided.</p>
Intravenous Infusions		<p>If there exists doubt or >5% change in weight IV line to be put up.</p>		<p>10% dextrose and electrolyte IV (if ER drink not tolerated).</p>	<p>IV based on symptom indication –</p>	<p>IV if needed/PO intake, initiate with 2ml/kg 25%dextrose, followed with</p>	<p>Insert IV, 10% Glc at 5ml/kg/hr if after initial</p>	<p>10%dextrose IV with electrolytes x1.5maintenance.</p>		

		<p>Potassium can be added to IV.</p> <p>Give Glc 200 mg/kg at once, normal saline, continue with 10% Glc at 5ml/kg/h until next solution ready.</p> <p>Reduce Glc to 5% rather than insulin if hyperglycemia a problem.</p> <p>Surgery: IV 10%Glc/0.45%Saline.</p>			<p>Hypoglycemia: 25% dextrose 2ml/kg, with continuous 10% dextrose 1.5x maintenance, 7-8 mg/kg/min. Lipids not to be administered in any form.</p> <p>Metabolic acidosis: (HCO3<16mEq/L): IV NaHCO3 1mEq/kg.</p> <p>Poor PO intake: 10%Glc continuous infusion 1.5x maintenance.</p>	<p>10%dextrose and electrolytes 10-12g Glc/kg/min, to maintain Glc>5mmol/L</p>	<p>management child vomits/unwell.</p> <p>IV carnitine 30mg/kg 8 hourly</p>	<p>IV Glc before and after surgical procedures requiring fasting, continued until normal intake.</p> <p>IV carnitine 100mg/kg if low oral carnitine intake/increased carnitine requirements due to illness.</p>	
Other Carbohydrates				Glucose polymer feeds.	Avoidance of fasting when stopping IV, in form of cornstarch supplementation.	Simple carbs orally.		Concentrated Glc source to be administered under tongue/cheek at frequent intervals while awaiting care.	
Monitoring tests	C8 and UOA result review at first follow up visit.	pH and gases, Glc, U&E, full blood count, blood culture. Electrolytes to be rechecked every 24hrs if on IV.		BG, electrolytes, CO2, blood gas, NH3, LFTs.	Assessment upon admission: BG, electrolytes, CO2, blood gas, NH3, LFTs		<p>Capillary/venous blood gas, BGL, Lac, U&E, NH3. If venous blood gas not done initially, should be done if child further vomits/unwell.</p> <p>Treatment decisions not to be based on blood Glc</p>	<p>In asymptomatic diagnosed patients, evaluate PACs, plasma free and total carnitine, UAGs, UOAs, MCAD leukocyte activity assay.</p> <p>In symptomatic diagnosed</p>	

							<p>levels.</p> <p>Upon admission to hospital monitor i) stable condition: blood Glc 4 hourly ii) unstable condition: blood Glc hourly, U&E and venous blood Glc 6 hourly.</p> <p>Baseline observations: HR, RR, Temp, SpO2, capillary refill, BP, neurological observations.</p>	<p>patients, additional tests include BG, blood gas, NH3, Lac, CBC with differential, electrolytes, LFTs, blood cultures.</p>	
Other assessments				<p>Assess for dehydration, fever, infection.</p>			<p>Monitor for encephalopathy and record hourly observations. Monitor fluid input/output, blood, urine dipstick for ketones.</p>		
Contact specialist		<p>Contact metabolic centre if necessary.</p>	<p>Consult with pediatric metabolic specialist.</p>	<p>Contact metabolic team for questions.</p>		<p>Metabolic specialist to be consulted during illness (fever, poor calorie intake).</p>	<p>Contact metabolic team if child further vomits/unwell at hospital.</p>		<p><1yr: contact hospital if not taking ER feeds/vomiting.</p>
Other		<p>Antipyretics as clinically indicated.</p> <p>Tube feeding if requirements not met, for max 24-36 hrs.</p> <p>Give usual medicines as soon as it is safe post surgery. Keep</p>			<p>Carnitine controversial, by decision of metabolic physician.</p> <p>Epinephrine if indicated, with dextrose infusion.</p>		<p>Low threshold for hospital admission. All metabolic patients to be triaged as category 2.</p> <p>Discharge if: tolerates carb drinks, observed for >=2hrs, appears well and consulted</p>	<p>Discharge when sufficient oral intake.</p> <p>Pregnant MCADD females: carnitine 100mg/kg, IV Glc as soon as labor starts and until normal oral intake.</p>	<p>Return to normal feeds (approx. 48hrs after starting ER).</p>

		nasogastric tube in place. Monitoring every 4-6hrs and assess using Glasgow Coma Scale. Child must tolerate 2 successive feeds before discharge.			High carb/low fat diet when coming off IV.		with metabolic team.	Awaken during night for feeding and assessment when ill.	
Emergency regimens, protocols and related procedures	Parents are to be provided with ER for each age group describing CHO intake or ORS.	Emergency protocols available for: Acute Decompensation, ER recipe preparation, Making IV fluids for metabolic patients, Child's Glasgow Coma Scale, Management of surgery in children with FAOD, Management of newborn babies with a family history of MCAD, Adult Emergency Management of medium chain FAOD, MCADD clinical management guidelines for presumptive positive MCADD.		Families instructed on ER and safe fasting times.	Emergency protocol provided (see all above recommendations)	Frequently update emergency letter.	Emergency protocol provided for pediatric emergencies (see all above recommendations)	Families need to be provided with emergency protocol/letter to give to EMS.	Give ER if child appears unwell.

Table A.S6. MCAD deficiency peer-reviewed long-term care guidance

Guidance document (First Author)	Derks	Feillet	Goodin	James	Leonard	Olpin	Rice	Spiekerkoetter	Walter
Guidance Topic									
Fasting avoidance	✓	✓	First line of defense.	Avoid catabolic state.	✓	✓	Avoid prolonged fasts, night time feeds when sick.	Regular meals, avoid prolonged fasting, particularly <6 months of age.	✓
Fasting schedule	6 months to 1 yr: <8 hrs <2 yr: 10 hrs 2+ yr: 12 hrs	Avoid fasting more than 3-4 hrs	Infants: 1 or 2 feeds during night, on-demand during day. Older kids can sleep through night.		As per BIMDG.			Same intervals as in VLCADD (Spiekerkoetter et al., 2009).	Makes reference to 4 different fasting schedules from other authors.
Carbohydrates		Introduce low glycemic index carbs when age permits.	Uncooked cornstarch, for enteral support, at night.	Frequent carb feeds.	High carb intake only when at risk.	Frequent high carb feeds when <24 months or unwell, increasing feeding interval with age.		Additional carbs not generally required.	
Fat		Normal fat content; 30-35% calories.	20-30% calories from fat.		Exclude fat only when child ill.	Avoid MCT.			
Carnitine		20-50 mg/kg/day in 2 administrations, if carnitine deficiency presents during follow up or diagnostic testing.	May include carnitine.	Carnitine supplementation - no additional details provided.	Carnitine discussed but no recommendations made.	May benefit from carnitine supplementation if develop low plasma carnitine.	Supplemental carnitine may be prescribed.	Carnitine use unclear, no recommendation made.	
Other dietary recommendations		No specific regimen; infants can be breast fed.	May include low-fat diet.		Ask all patients to follow dietary advice.	Normal diet other than previous recommendations.	Low-fat diet.		

Table A.S7. MCAD deficiency grey literature long term care guidance

Guidance document	BIMDG dietitians'	BIMDG additional	Dietitian MCADD letter	GeneReviews	GMDI	IMD Scotland (NHS)
Guidance Topic						
months	4 to 6 months: feed before bedtime and once at nighttime. 1 year: starchy bedtime snack, not to miss or have late breakfast.	✓	✓	✓	✓	Avoid long periods without food. Teens and adults should avoid prolonged fasting and weight loss should be planned with dietitian.
Fasting schedule	24-48 hrs: 6 hrs 4-6 months: 8 hrs 8-12 months: 10 hrs 12 months: 12 hrs 2 yrs: 12 hrs 3-9 yrs: 12 hrs 10+ yrs: 12 hrs	Bottle fed babies: on demand feeds, <4 hrs. Volumes increase from 20ml/kg on first day to 150 ml/kg by 7 days. Breast fed: Top up bottle feeds or expressed breast milk in first 72 hrs. Breast feed for ≥10 min, 8 times/day. Top up feeds: Day1: 25ml/kg Day2: 40ml/kg Day3: 60ml/kg			Well infant should be fed as non-MCADD infant: Birth-4 months: <4hrs 5-12 months: 4 hrs (+1hr/month thereafter). Childhood: age-appropriate meal and snack schedules.	Infant: 3-4 hrs, <6 hrs (6-8 feeds/day) Infants <1 yr when unwell: breast/formula every 2-3 hrs. 1+yrs and adults: <12 hrs Child 1-2 yrs: offer snacks at regular times between meals if required (every 1.5-2 hrs post meal) to prevent grazing. Wake infants for nighttime feed.
Carbohydrates	1 yr: Missed meal to be replaced with starchy snack or sugary drink.			Toddlers: 2g/kg uncooked cornstarch at bedtime.	Supplementation in the form of cornstarch or other carbs before bedtime (should not be necessary,	Child 1-2 yrs: CHO drink should be at least 10.5g/100ml.

					unless patient not feeding well/ill).	Child 2-9 yrs: CHO drink should be at least 17g/100ml.
Fat	Exclude MCT	MCT feeds and supplements contraindicated.	Infant formula or enteral feeds with MCT should be avoided.	Avoid infant formulas containing MCT.	Infant formulas: $\leq 55\%$ kcal as fat. Child/Adult: 30% kcal from fat. MCT containing formulas not appropriate.	Avoid pure coconut and coconut oil. Avoid formulas with MCT.
Carnitine					Infant dose: 50-100mg/kg. Patients on carnitine should have periodic assessment of free carnitine. Plasma free carnitine monitoring (goal range 25-35uM) to determine need for supplementation.	
Monitoring tests					Frequent blood Glc measures not required. May measure Glc at end of longest sleeping interval to determine max fasting time. For at home monitoring, use a glucometer sensitive to low blood Glc. Post confirmatory testing: routine blood carnitine.	
Other diet/dietary recommendations	0-4 months: breast feeding or normal infant formula. 6+ months: commence weaning, normal diet, rich in	Within 5 days first face-to-face contact: clinical review including dietetic review and emergency regimen.	No special dietary treatment, breast or bottle feed.	Toddlers: Low fat diet (<30% energy from fat).	Infancy: regular infant formula or breast feeding are appropriate. Breast milk	When well, no need to give sugary drinks.

	<p>starchy foods,</p> <p>1yr+: cow milk allowed, normal diet including starchy foods.</p>				<p>enhancers are not appropriate.</p> <p>Children/adults: heart healthy diet. Include fruits and vegetables, complex carbs.</p> <p>No a priori need for supplementation.</p>	<p>Can give small amounts of ER drink to accustom to taste.</p> <p>Infant: breast feed or normal infant formula. Weaning at 6 months (not earlier than 4 months). Start with pureed fruits and vegetables and then introduce starchy carb foods.</p> <p>1+yr: can start full fat cow milk.</p> <p>1-9 yrs: 3 main meals with starchy carb foods, bedtime snack (includes starchy item).</p> <p>If child didn't take breakfast, offer alternative drink (list provided) and then give mid morning snack.</p> <p>Child 2-9yrs: if skipped breakfast, sports drinks are not a suitable alternative drink.</p>
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Table A.S8. MCAD deficiency peer-reviewed additional guidance

Guidance document (First Author)	Feillet	Kingsley	Leonard	Rice
Guidance Topic				
Siblings	Sibs born to MCADD affected family should be put on glucose in first 2-3 hrs until results obtained.		All siblings should be investigated by blood spot AC and genotype. If first proband affected, additional blood spot AC and genotype at 24-48 hrs of age for future siblings. Assume diagnosis and give frequent feeds.	
Parental education			Parents need to be taught to use own judgment in emergency responses.	Parental education about avoiding fasting during intercurrent illness if most important intervention.'
Physical activity and Obesity	Should not limit physical activity.		Risk of obesity if following MCADD dietary advice	
Vaccination		Full normal immunization schedule. ≥24 months: give 23-valent pneumococcal polysaccharide vaccine	Full vaccination, can be done with anti-pyretic and using ER.	
Visit details and frequency	Follow-up visits; ≤1 yr - every 3 months, ≤3 yrs - every 4 months, ≤12 yrs - every 6 months, 12+ yrs - yearly.			
Other	Offer screening and genetic testing to all related family members.			

Table A.S9. MCAD deficiency grey literature additional guidance

Guidance document	BIMDG dietitians'	BIMDG additional	Dietitian MCADD letter	NECMP	GeneReviews	ACMG ACT sheet	PMH	GMDI	IMD Scotland (NHS)
Guidance Topic									
Siblings		Sibling screening if MCADD diagnosis confirmed and there are unscreened siblings.		All siblings of known MCADD patients should be tested, despite absence of symptoms.	Plasma AC and urine AG in siblings and parents to be evaluated.			If sibling affected with MCADD, and awaiting screening/diagnosis for new neonate, breast feeding can be in conjunction with glucose water/small amounts of formula.	
Parental education	Emphasis on reviewing feeding (breast, bottle), ER and their preparation, feeding frequency and fasting time, emergency contact numbers.		Parents are to be instructed on safe fasting times.			Educate family about fasting avoidance and seeking immediate medical attention if infant ill.		Parents should be educated re: over and under feeding, getting immediate medical attention when child ill or PO intake or blood Glc <60mg/dl.	Parental education on when to contact hospital, preparing ER feeds and managing ER supplies.
Physical activity and Obesity					Follow up to include weight control. Proper nutrition and exercise.			Monitor growth - children should follow normal growth curve. If breast feeding, monitor for appropriate weight gain. Need to avoid over feeding/exceeding kcal intake.	

								Exercise to be accompanied by carb intake and hydration.	
Alert tags	May be useful to wear Medic Alert Bracelet.						Medical tags recommended (provide suggested keywords).	Medical alert tag may be helpful for children.	
Alcohol	10+ yrs: important to give advice re: alcohol.								Alcohol: if excess taken and causes vomiting, go directly to hospital. If meal missed due to drinking, take ER or alternative drink.
Visit items frequency	Weight and length should be measured at each visit. Diet history and feeding problems to be discussed at visits at 1yr+. 3-9 yrs: yearly dietetic review				Monthly visit in first months of life with dietitian involved.			Diet assessment at each clinic visit.	Visit with dietitian 4 times in first year, and then yearly after that.
Other					Biochemical/molecular genetic testing to be offered to both parents.				Take ER supplies when traveling/holidays. Specialist can provide letter about importance of supplies for child to present at customs.

									<p>Schools should keep supply of alternative drinks or older children may carry these around with them.</p> <p>10+yrs: children should know how to make ER.</p>
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Table A.S10. VLCAD deficiency peer-reviewed diagnosis guidance

Guidance document (First Author)	Arnold	Dietzen	Fernandez-Lainez	James	Olpin	Rice	Rinaldo	Spiekerkoetter '09
Guidance Topic								
Acylcarnitines (AC)		For follow-up analysis (particularly C14:0, C14:1, C14:1, C16:0, C16:1, C18:0, C18:1)		Plasma AC for follow-up evaluation. AC profiling in fibroblasts for diagnostic testing.	Infantile and myopathic VLCAD: Detected using AC profiles.	Plasma AC Diagnosis is by Plasma AC profile.	Plasma AC (also part of specialized plasma and urine testing).	
Organic Acids (OA)			Urine OA profile	Urine OA for follow-up evaluation.	✓			
Acylglycines (AG)				Urine AG for follow-up evaluation.			Urine AG (also part of specialized testing).	
Blood Glucose	Additional test if infant unwell.			For follow-up evaluation	✓	Hypoglycemia	Routine plasma	
Ammonia	Additional test if infant unwell.						Routine plasma	
Fatty Acids					Non- essential fatty acids (NEFA).		Specialized plasma: free fatty acids, 3-OH fatty acid.	
Carnitine	Additional test if infant unwell and for VLCAD-C phenotype: plasma carnitine.			Total/free carnitine for follow-up evaluation.	Total/free carnitine.		Specialized plasma: free and total carnitine.	
Gases							Routine plasma	
Electrolytes	Additional test if infant unwell.						Routine plasma	

Other follow-up diagnostic and ancillary confirmatory testing	Additional test if infant unwell: lactate, liver function tests, creatine kinase. For VLCAD-C phenotype: troponin, echocardiogram.				3-hydroxybutyrate	Ketones	Routine plasma: lactate, pyruvate, liver function tests, creatine phosphokinase. Routine urine: ketones.	
Enzyme Analysis	Enzyme assay (or may use FAO probe studies).	Activity assay generally not available.	Enzyme assay		Infantile and myopathic VLCAD: Detected using fibroblast tritiated release assays.	Enzymology often required for diagnosis.		Should be done primarily by enzyme analysis in lymphocytes.
Mutation analysis			Gene defect molecular studies.	VLCAD genotyping		DNA testing often required for diagnosis.		
Other	Endorsed ACMG fact sheets for evaluating screen positive infant. Cardiology evaluation for unwell infant and VLCAD-C phenotype. No specific testing pattern recommended.	Provided algorithm for general diagnostic procedure.	Appropriate confirmatory testing to be done must be included in final report.	VLCAD protein (western blot)				

Table A.S11. VLCAD deficiency grey literature diagnosis guidance

Guidance document	GeneReviews	ACMG ACT sheets	GMDI
Guidance Topic			
Acylcarnitines (AC)	Comprehensive analysis (during time of metabolic stress).	Plasma AC (showing increased C14:1).	Plasma AC profile
Organic Acids (OA)			Urine OA
Blood Glucose		Routine lab	
Carnitine			Plasma carnitine
Ammonia		Routine lab	
Gases		Routine lab	
Electrolytes		Routine lab	
Others follow-up diagnostic and ancillary confirmatory testing		Routine lab: lactate, liver function tests, creatine phosphokinase	
Enzyme Analysis	VLCAD enzyme activity in fibroblasts/lymphocytes.		✓
Mutation analysis	Molecular genetic testing of ACADVL.	Analysis of VLCAD gene.	✓
Other	Functional analyses of FAO in cultured fibroblasts. Immunoreactive VLCAD protein antigen expression.	FAO probe assay, fibroblast culture. Diagnostic algorithm provided.	

Table A.S12. VLCAD deficiency peer-reviewed emergency/acute care guidance

Guidance document (First Author)	Arnold	Olpin	Rice	Spiekerkoetter
Guidance Topic				
Fluids and Solutions		Emergency regimen should always be available, consisting of readily available high carb drinks.	Dextrose (10% with salt)	
Intravenous Infusions	IV Glc fluids should contain appropriate electrolytes and 25-50% normal saline. If no improvement, increase Glc infusion and add insulin by drip.		If not taking sufficient energy orally: dextrose-containing fluid IV.	IV therapy to be initiated for severe illness.
Dietary modifications	Supplement with higher MCT given profoundly abnormal C14:1.			
Carnitine				Supplementation with carnitine during severe metabolic derangement should be avoided.
Other	For ill child monitor Glc, electrolytes, liver function tests, creatine kinase, plasma carnitine, and ammonia for metabolic crisis. For severe VLCADD: troponin and cardiac evaluation		CK levels should be followed during illness.	Nasogastric feeding for metabolic decompensation during illness to avoid IV Glc infusion.
Emergency regimens, protocols and related procedures	Endorsed NECMP for metabolic crisis.		Emergency letter should be provided to all affected families.	Catabolic states need always be managed by emergency regimen.

Table A.S13. VLCAD deficiency grey literature emergency/acute care guidelines

Guidance document	BIMDG Additional	ACMG ACT sheets	NECMP	GeneReviews	PMH	GMDI
Guidance Topic						
Fluids and Solutions	<p>Glucose polymer given as small frequent oral feeds.</p> <p>Vomiting/diarrhea: electrolytes via standard rehydration mixtures, substituting Glc polymer for water.</p>				High carb drink orally or via nasogastric tube if child not encephalopathic and brought to hospital due to food refusal/single vomit.	
Intravenous Infusions	<p>If nauseated, put up IV (or give fluids continuously via nasogastric tube).</p> <p>Treat with IV if can't tolerate fluids, unwell, or if any doubts: 200mg/kg at once over a few minutes, then normal saline 10ml/kg (20ml/kg if poor circulation/patient shocked) as bolus immediately after Glc. Repeat bolus if condition persists. Continue with Glc 10% at 5ml/kg/h until next solution ready (avoid keeping at higher rate). Administer appropriate rate 0.45% saline/10% Glc after calculating deficit and maintenance to correct within 24 hrs.</p>	<p>Post positive NBS, if signs of illness or unwell baby, emergency treatment includes IV Glc (and oxygen).</p>	<p>IV based on symptom indication, never <10% dextrose, 1.5 times maintenance, 7-8 mg/kg/min.</p> <p>Hypoglycemia: 25% dextrose 2ml/kg, with continuous 10% dextrose 1.5x maintenance, 7-8mg/kg/min.</p> <p>Metabolic acidosis (HCO3<16mEq/L): IV NaHCO3 1mEq/kg.</p> <p>Poor PO intake: 10%Glc continuous infusion 1.5x maintenance.</p>	✓	<p>Insert IV, 10% Glc at 5ml/kg/hr if after initial management child vomits/unwell.</p>	<p>Give IV Glc (D10) at 1.5x maintenance.</p>

	<p>Add potassium if required when urine flow normal and plasma concentration known.</p> <p>Unwell baby: transfer to NICU, start IV infusion 10% dextrose at 100ml/kg/day.</p> <p>Unwell adults: triage to high priority. Can give 200ml 25% Glc polymer solution every 2hrs if relatively well, otherwise IV at 50% dextrose 50ml over 30min, then continuous IV 10% dextrose 2mls/kg/hr. Correct volume depletion if required 0.9% NaCl.</p>					
Carnitine					Carnitine controversial during acute illness, should be discussed with metabolic consultant.	During acute illness, carnitine should continue to be administered. IV carnitine may be required at hospital.
Other Carbohydrates	<p>Glucose polymer concentrations by age (yrs):</p> <p>0-1: 10g/100ml 1-2: 15g/100ml 2-10: 20g/100ml >10: 25g/100ml</p>					<p>Parents to provide instant Glc source on way to hospital.</p> <p>Cornstarch may be needed before bedtime to prevent hypoglycemia.</p>
Monitoring tests	<p>For initial admission to hospital complete</p> <p>blood tests: pH and gases, Glc (lab and bedside strip test), urine and electrolytes, full blood count, lactate, blood spot ACs, creatine kinase, liver function tests, blood</p>	<p>Assessment upon admission: blood Glc, electrolytes, CO₂, blood gas, ammonia, liver function tests.</p>		<p>To establish disease extent: baseline plasma creatine kinase concentration and liver transaminases.</p>	<p>Upon admission to hospital monitor i) stable condition: blood Glc 4 hourly ii) unstable condition: blood Glc hourly, urine and electrolytes and venous blood Glc 6 hourly.</p>	

	<p>culture, and urine tests: myoglobin (use urine strip test for blood).</p> <p>Progress monitoring:</p> <p>clinically reassess every 4-6 hrs;</p> <p>Blood: pH, gases, Glc (lab), urine and electrolytes, Glasgow coma scale and BP.</p> <p>Adults should be reassessed frequently including blood tests: pH and gases, urine and electrolytes, creatine kinase, Glc.</p> <p>If on IV, recheck electrolytes daily.</p>			<p>For acute illness: blood Glc and ammonia concentration.</p>	<p>Baseline observations: heart rate, respiratory rate, temperature, oxygen saturation, capillary refill, blood pressure, neurological observations.</p> <p>Monitor fluid input/output, blood, urine dipstick for ketones.</p> <p>Capillary/venous blood gas, blood Glc, lactate, urine and electrolytes, ammonia. If venous blood gas not done initially, should be done if child further vomits/unwell.</p>	
<p>Other assessments</p>	<p>Cardiac monitoring for first 4 days of life or until normal feeds and weight gain established if there is history of sudden sibling death at 2-3 days.</p> <p>For adults upon initial admission, also include c-reactive protein, blood and urine cultures, if appropriate.</p> <p>2D echocardiography if signs of cardio-respiratory problems and for unwell babies with history. Add ECG for adults.</p> <p>Check and treat rhabdomyolysis: put on</p>	<p>Cardiology assessment or at minimum CXR and EKG.</p>		<p>Treat cardiac rhythm disturbance.</p> <p>Monitor rhabdomyolysis, treat by hydration & alkalization.</p> <p>To establish disease extent: echocardiography.</p>	<p>Monitor for encephalopathy and record hourly observations.</p> <p>Creatine kinase level and check for urine myoglobinuria.</p>	

	cardiac monitor; weight and vital signs, blood pressure history and examination.					
Contact specialist		Consult with pediatric metabolic specialist.		To establish disease extent: genetic consultation	Contact metabolic team if child further vomits/unwell at hospital.	After 3 days of care at home, health care professional should be contacted.
Other	<p>Child should be admitted, even if for a short period time, when there are any concerns about condition. Treat any infection.</p> <p>Reintroduce oral feeds as early as possible.</p> <p>Adults: treat infection, give analgesia, anti-pyretic/emetic if required. Dialysis if severe renal impairment.</p>	Epinephrine if indicated, with 10% dextrose infusion.			<p>Treatment decisions not to be based on blood Glc levels.</p> <p>Low threshold for hospital admission. All metabolic patients to be triaged as category 2.</p> <p>Discharge if: tolerates carb drinks, observed for >=2hrs, appears well and consulted with metabolic team.</p>	<p>Surgery/trauma/labor/childbirth/post-partum period - requires extra IV calories and MCT supplementation. MCT orally or by tube as soon as child can tolerate it.</p> <p>MCT oil should continue to be administered during acute illness at home. Intralipid contraindicated.</p>
Emergency regimens, protocols and related procedures	Protocol for blood, urine, ECG, fluids, monitoring, hyperkalemia, oliguria provided.	Emergency protocol described.		Emergency care plan to be kept up to date.	Emergency protocol described.	Emergency protocol should be followed as per NECMP.

Table A.S14. VLCAD deficiency peer-reviewed long-term care guidance

Guidance document (First Author)	Arnold	James	Olpin	Rice	Spiekerkoetter '09	Spiekerkoetter '10
Guidance Topic						
Fasting avoidance	Prevention of fasting key dietary treatment.	Avoid catabolic state.	Avoidance of fasting, frequent high carb feeds.	Avoid prolonged fasts. Nighttime feeds when sick.		Fasting avoidance. More frequent feeds for patients with recurrent hypoglycemia.
Fasting schedule	No one recommended fasting interval or schedule.				<p><i>Neonates</i>: 3 hrs, also at night.</p> <p><i><6 months</i>: 4 hrs, also at night but in well-nourished children can be extended to 6 hrs from 3 months of age.</p> <p><i>>6 months</i>: 4 hrs day time, 6-8 hrs at night but can be extended to 8 hrs from 6 months of age in well-nourished infants.</p> <p><i>>12 months-3 yrs</i>: 4 hrs day time, 10-12 hrs at night.</p> <p><i>4-7 yrs</i>: 4 hrs during day, 10-12 hrs at night.</p>	
Carbohydrates			Slow release CHO (uncooked cornstarch) can be given before bedtime in older infants/children.		No additional carbs/energy required: Routine enrichment with glucose polymers/cornstarch not recommended but part of prophylactic/therapeutic emergency treatment.	Glucose monomer could be added to diet with recurrent hypoglycemia.
Fat		MCT oil supplementation.	Strict reduction of long chain fats.	Dietary fat restriction, MCT supplementation.		Patients with cardiomyopathy: diet comprised primarily of MCT.

			MCT supplementation up to 30% of calories.			
Essential fatty acids			EFA supplementation.		EFA supplementation by age of n-6 Linoleic acid and n-3 Linolenic acid, from walnut/soy/wheatgerm oil.	EFA supplementation for patients with cardiomyopathy.
Carnitine	Only to prevent carnitine deficiency.	✓	✓	Controversial	Controversial Free carnitine monitoring in those not taking carnitine supplements.	No evidence favoring L-carnitine.
Monitoring				Regular cardiac evaluations, echocardiography.	Creatine kinase and transaminases to monitor treatment effects. Echocardiography, documentation of liver size and structure once/year.	
Other dietary recommendations	VLCAD-C: <12months: Symptomatic: Maximally enriched MCT formula. Asymptomatic: bottle-fed (replace standard formula). >12months: LCT restriction and MCT supplementation. VLCAD-M: <12 months: Asymptomatic: breast-fed Symptomatic: supplement MCT >12 months: Asymptomatic: Heart healthy diet with MCT. Symptomatic: Heart healthy diet with MCT.	Low fat and high carb diet.			Low fat diet. LCF intake: <i>Healthy infants:</i> 40-45% total energy. <i>School age children:</i> 30-35% total energy. Asymptomatic: 0-4 months: 0.5 breast milk/infant formula + 0.5 special low fat formula 4+ months: reduction of LCT to 30-40% total energy, 10-15% fat energy from MCT. Symptomatic:	Asymptomatic newborns/normal creatine kinase and transaminase levels: breast-feeding normally suggested. Symptomatic infants/elevated creatine kinase and transaminase levels: diet should contain less/no breast milk and more MCT.

					<p>0-4 months: no breast milk/infant formula, 100% special low-fat formula+EFA+MCT</p> <p>4+months: reduction of LCT to 25-30% total energy, 20% fat energy from MCT, 3-4% EFA</p> <p>Follow-up done at metabolic centres irrespective of severity/choice of treatment.</p>	
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Table A.S15. VLCAD deficiency grey literature long term care guidance

Guidance document	BIMDG additional	NECMP	GeneReviews	GMDI
Guidance Topic				
Fasting avoidance	Frequent feeding.		Avoidance of fasting, including periods of preparation for or recovery from surgery/sedation	Fasting avoidance during, including home care for acute illness. Require snacks during the day and before bedtime.
Fasting schedule				Birth-4 months: <4 hrs 5-12 months: 4 hrs (+1hr/month) up to 8 hrs.
Fat	Formulas where LCT replaced with MCT.	For confirmed VLCADD, may provide MCT oil.	Severe form: low fat formula, supplemental calories through MCT; 13-39% total fat, with 15-18% from MCT.	Total fat energy: Infancy: 40-45% fat intake Severe VLCADD: 10% LCT, 30% MCT Mild/moderate VLCADD: 50% LCT, 50%MCT [Alternatively 2-3g MCT/kg body wt]. Child/adult: 35% intake energy after 1 yr; 10-20% LC, remainder MCT. [Alternatively, 1.0-1.15g MCT/kg/day. MCT as special formula or oil supplement]
Essential fatty acids				EFA intake (with recommended ratio from 5:1 to 10:1): Linoleic acid: 3% intake Linolenic acid: 1% intake DHA (if used) recommended dose for children: <20kg - 65mg/d >20kg - 130mg/d
Carnitine	May give carnitine orally 100-200mg/kg/24h in 4 doses, although should be avoided.	Carnitine controversial, by decision of metabolic physician.	No consensus as to efficacy of carnitine.	Recommended by some centres beginning at dose 50-100 mg/kg.

<p>Monitoring tests</p>			<p>Annual cardiac exam.</p>	<p>Dietary assessments for energy intake, diet composition, vitamins and minerals.</p> <p>AC at each clinic visit.</p> <p>Erythrocyte FFA profile \geqtwice yearly.</p> <p>Monitor triene:tetraene and arachidonic/DHA ratios, plasma free carnitine, urine OA, urine dipstick for myoglobinuria.</p>
<p>Other diet/dietary recommendations</p>	<p>Low-fat diet.</p> <p>Infants with family history: Probably safe to give 50% monogen/50% breast milk.</p> <p>Top ups by nasogastric tube for baby if good milk intake is not met (<15ml/kg/feed).</p>		<p>Avoid high fat diet.</p> <p>Avoid dehydration and myocardial irritation (cardiac catheterization), volatile anaesthetics/with high doses of LCT.</p>	<p>May use special formulas with MCT, and standard infant formula or breast milk as source of LCT.</p> <p>Protein intake ~15%.</p>

Table A.S16. VLCAD deficiency peer-reviewed additional guidance

Guidance document (First Author)	Olpin	Rice	Spiekerkoetter '09	Spiekerkoetter '10
Guidance Topic				
Parental education		Parental education to avoid prolonged fasting.		
Physical activity and Obesity	Avoidance of prolonged exercise to avoid symptoms. For physically active young people: <i>Before:</i> carb-rich meals with MCT <i>During:</i> use of high carb drinks at regular intervals	Competitive and isometric sports must be restricted.	<i>Symptomatic VLCAD:</i> increased MCT or CHO prior to extensive exercise (dose of 0.25-0.5g MCT/kg) for exercise-induced muscle pain/weakness. <i>Asymptomatic VLCAD:</i> MCT prior to exercise for exercise-induced myopathy.	Pre-exercise MCT supplement, with or without carbs may improve exercise ability. Encourage exercise, periods of rest and rehydration.
Other	Multivitamin supplementation.		Supplementation with vitamins/minerals when low intake/documentated deficiency.	

Table S17. VLCAD deficiency grey literature additional guidance

Guidance document	BIMDG additional	NECMP	GeneReviews	ACMG ACT sheet	GMDI
Guidance Topic					
Siblings			Evaluate sibs and relatives who may be at risk.		
Parental education	Ensure parents have clear management plan before returning home post-acute illness.			Educate family about fasting avoidance, regular eating schedule, emergency protocol use, signs of illness, genetic basis and seeking immediate medical attention if infant ill.	Teaching parents about importance of not overfeeding through frequent feeds.
Physical activity and Obesity			Extra MCT beneficial for older individuals with exercise intolerance.		Snack before and during exercise: complex carbs/cornstarch. MCT in snacks may improve exercise tolerance. Post exercise: rest, extra fluid, carb containing snacks. Cornstarch may be needed as additional calories before and during exercise. Avoiding overfeeding and weight reduction.
Other		Recommends physicians to listen to parents who are much more familiar with early signs of decompensation in their children.	Suggest a labor management plan to pregnant women with VLCADD.		New NBS-identified treatment options: i) all or symptomatic only: 10% intake from LCT ii) mild forms: liberalize fat intake iii) asymptomatic: 20-25% LCT intake

INTERFACE

This article describes actual practice in managing MCAD deficiency. We conducted a scoping review to map recent literature from the year 2000. We then conducted a secondary analysis of Canadian pediatric data obtained from an existing multi-centre cohort study that collects chart-abstracted data from participating children. The focus of our analyses was on previously identified areas of controversy, including the use of carnitine supplementation and appropriate fasting schedules for children with MCAD deficiency. We compared management in the cohort data to that reported in the literature.

The student (AK) was responsible for conducting the scoping review literature search, screening retrieved citations for eligibility, extracting information from eligible included studies, and summarizing the content of included studies. AK was also responsible for conducting the secondary descriptive analysis of Canadian cohort data and for interpreting results and writing the manuscript. BKP, MG, IDG, FM, and PC contributed to the conception, design and supervision of the study, the analysis of information from the reviewed articles and of the data from the cohort study, and the interpretation of findings and critical revisions to the manuscript. KT contributed to the analysis and interpretation of the data from the cohort study. JM led the construction of the search strategy. AF contributed to the screening of retrieved citations for eligibility and extraction of information from included studies. We plan to submit this manuscript to the journal, *Orphanet Journal of Rare Diseases*; it has been formatted accordingly.

Chapter 3 – Practice variation in the management of MCAD deficiency: scoping review and descriptive analysis of secondary data from a Canadian pediatric cohort

A Kazakova^a, BK Potter^a, MT Geraghty^b, ID Graham^{a,c}, F Momoli^{a,b,c}, K Tingley^a, J McGowan^a,

A Fairfax^a, Working Group 2 of the Canadian Inherited Metabolic Diseases Research Network, P Chakraborty^b

Affiliations

^aUniversity of Ottawa, Ottawa, Ontario, Canada

^bChildren's Hospital of Eastern Ontario, Ottawa, Ontario, Canada

^cOttawa Hospital Research Institute, Ottawa, Ontario, Canada

Abstract

Purpose We describe management practices for medium-chain acyl-CoA dehydrogenase (MCAD) deficiency with respect to two interventions: (i) the use of carnitine supplementation and (ii) fasting times. We conducted a scoping review of published literature on treatment practices around the world and a secondary analysis of data documenting treatments received by participants in a Canadian pediatric cohort study.

Methods For the scoping review, we included primary studies published since the year 2000 that reported on disease management for MCAD deficiency. We developed a search strategy with a specialist librarian. Citations were screened by two independent reviewers. We extracted information on study characteristics and management related to carnitine supplementation and fasting interventions, describing the use of these interventions overall and by potential indicators of disease severity (genotype, biochemical phenotype). Our secondary analysis included longitudinal data for Canadian children with MCAD deficiency, born between 2006 and 2015 and enrolled in a cohort study at one of 13 centres. We described carnitine supplementation and fasting times by age and according to the same possible indicators of disease severity (genotype, biochemical phenotype).

Results Five publications were eligible for inclusion in the scoping review. There was variation in the use of carnitine across and within studies. Specific fasting times were variable and frequently not defined. We found inconsistent evidence of an association between genotype and carnitine use that may be linked to a relationship between genotype and carnitine deficiency. The Canadian cohort included 107 children with MCAD deficiency. We identified a decreasing trend in carnitine use by calendar time when adjusting for age. Median fasting times increased with age but were variable. A higher proportion of children who were homozygous for the classic

MCAD deficiency genotype were noted to have received carnitine than those with other genotypes. Median fasting times did not appear to differ by genotype.

Conclusion Management practices related to carnitine supplementation and fasting times for MCAD deficiency were highly variable. We identified some evidence of an association between genotype and carnitine use in the literature and we observed a similar pattern in the Canadian cohort. This potential association may be due to a relationship between genotype and carnitine deficiency.

Keywords MCAD deficiency, Current management, Canadian cohort analysis

Introduction

Medium-chain acyl-CoA dehydrogenase (MCAD) deficiency is a recessively inherited single-gene metabolic disorder of impaired fat metabolism. Specifically, mutations in the *ACADM* gene result in insufficient MCAD enzyme activity among those with this disorder, with a corresponding inability to adequately metabolize medium chain fats.¹ The clinical presentation of MCAD deficiency is characterized by a risk of acute metabolic decompensation marked by hypoketotic hypoglycemia. This risk is present particularly in situations of catabolic stress such as long-term fasting, acute illness, and exercise, when medium chain fats are normally an important energy source.² Metabolic decompensation in turn is associated with high morbidity and a risk of mortality.³

Population-wide newborn screening for MCAD deficiency has been implemented in many jurisdictions around the world and has yielded important benefits in terms of reduced morbidity and mortality for this disorder due to the established effectiveness of early, pre-symptomatic preventive management.^{4,5} There has also been a large increase in the observed birth prevalence of MCAD deficiency with its inclusion as a target of newborn screening, likely because of screen-identified cases that would have otherwise died without an MCAD diagnosis, and screen-identified milder cases that are largely asymptomatic and thus typically undiagnosed. Newborn screening has uncovered many new disease-causing mutations for MCAD deficiency, with varying evidence regarding a genotype-phenotype association, particularly when linking genotype to the risk of clinical manifestations.⁶⁻⁸ For example, prior to the inclusion of MCAD deficiency in newborn screening programs, many individuals diagnosed clinically were homozygous for a common mutation in the *ACADM* gene, c.985A>G, which is therefore believed to be associated with a high risk of metabolic decompensation.^{7,9} Among the newly

discovered genetic mutations detected by screening, some have been predicted to lead to milder disease, such as the 199T>C mutation.⁷ Measures of residual enzyme activity may be used as an indicator of disease severity,^{10,11} and some evidence suggests that the milder c.199T>C variant confers a residual enzyme activity level similar to a heterozygous carrier state (one mutation only) where there is virtually no risk of disease symptoms.¹¹ This suggests that patients who are homozygous for this mild variant may be at low risk of metabolic decompensation. However, the link between genotype and risk of metabolic decompensation has not been clearly established in patients followed clinically.¹²

Now that MCAD deficiency is predominantly diagnosed in asymptomatic newborns, questions have been raised regarding appropriate management, particularly the potential to adjust treatment based on predicted disease severity.^{12,13} Given the inconsistent link between clinical phenotype and genotype, a precautionary management approach is typically applied to all diagnosed MCAD deficiency patients, independent of genotype or residual enzyme activity or other estimates of disease severity.^{11,14} Preventive management for MCAD deficiency centres around avoidance of fasting, and, during times of catabolic stress, the use of rapidly available carbohydrates and close medical monitoring.^{1,15,16} Other treatments may be used preventively in some cases, for example carnitine or corn starch supplementation.¹⁷⁻¹⁹ Given generally identical or similar treatment regimens and the success of preventive management for MCAD deficiency, no differences in outcomes, such as hospitalizations or episodes of metabolic decompensation, have been reported based on differing genotype or other potential markers of disease severity in asymptomatic populations identified by screening.¹²

In this study our objective was to identify practices in the management of MCAD deficiency in the newborn screening era. To pursue this objective, we conducted a scoping

review of published literature on treatment practices around the world, and a secondary analysis of data documenting treatments received by participants in a Canadian cohort study of children with MCAD deficiency.²⁰ For both study components, we focused solely on two areas of care that we had identified *a priori* as of interest: (i) the use of carnitine supplementation and (ii) fasting times. In our systematic review of treatment guidance (Chapter 2), none of the guidance we considered recommended routine carnitine supplementation for preventive use in all patients with MCAD deficiency, but we identified conflicting and uncertain recommendations about the circumstances under which it may or may not be appropriate. This mirrors controversy about carnitine supplementation in the literature, with some recent concern about possible long-term adverse effects.^{21–23} While avoidance of fasting is established as the foundation of preventive care for MCAD deficiency, previous literature and our systematic review identified variation in the specific fasting schedules that have been recommended, and there has been some concern about the risk of development of overweight among individuals with MCAD deficiency due to overfeeding.^{24,25} Finally, given the questions raised in the context of newborn screening and the asymptomatic identification of a broader clinical spectrum of disease severity, we were also interested in whether current management practices with respect to carnitine supplementation and fasting times differ according to predicted disease severity, as defined primarily by genotype and also by biochemical phenotype (the latter based on blood analytes used in the screening and diagnostic process).

Methods for literature scoping review

Guided by the Joanna Briggs Institute (JBI) recommendations,²⁶ we conducted a scoping review of literature published since the year 2000. A scoping review is particularly useful in

mapping the literature on a subject to identify the nature and content of a broad range of evidence at an early stage, when it is not possible to establish more precise questions. The JBI method for a scoping review involves the following steps: clearly specifying the research question, including the population, content, and context (PCC). The search strategy includes both peer reviewed and grey literature, initially limited to at least 2 large databases and then using keywords and index words identified from this search to search additional databases.

This scoping review was part of a larger study that initially also included a second fatty acid oxidation disorder (very-long-chain dehydrogenase (VLCAD) deficiency). The citation search and eligibility screening process for the review was integrated across the two disorders so that the methods below include search terms and inclusion criteria that encompass both conditions. However, the rest of the methods and results were separable by disease and in this paper, we focus specifically on MCAD deficiency.

With an experienced health science librarian (JM), we developed a search strategy for multiple electronic databases: MEDLINE, EMBASE, EBM Cochrane Database of Systematic Reviews, EBM Database of Abstracts of Reviews of Effects, PsycInfo, the Joanna Briggs Institute (JBI) EBP Database, and Food Science and Technology Abstracts. This strategy was informed by a related systematic review of the quality and content of disease management guidance (see Chapter 2). Key search terms included Acyl Coa dehydrogenases, MCADD, VLCADD, cohort, case-control studies. Box 1 shows the search strategy that was used for Medline(R) and was adapted for the other databases. The full search strategy can be found in Appendix B. This search was executed on December 13, 2016. We also reviewed a total of 54 grey literature sources, such as website and databases potentially relevant to MCAD deficiency, using similar keywords; the complete list of grey literature sources we searched is included in

Appendix B (Supplement 4). We also did a manual search, reviewing the reference lists of papers that were identified as eligible after full text screening. Our manual search also included reviewing papers and reference lists identified as part of a related systematic review (Chapter 2). Our search included both English and French literature.

Box 1. Search terms for scoping review of MCAD/VLCAD deficiency reported actual practice

- 1 exp Acyl-CoA Dehydrogenases/
- 2 [MCAD.tw](#).
- 3 [MCADD.tw](#).
- 4 (medium-chain acyl-CoA dehydrogenase or Medium-chain acyl-coenzyme A dehydrogenase).tw.
- 5 (medium chain acyl CoA dehydrogenase or Medium chain acyl coenzyme A dehydrogenase).tw.
- 6 [VLCADD.tw](#).
- 7 [VLCAD.tw](#).
- 8 (very-long-chain acyl-CoA dehydrogenase or Very long chain acyl CoA dehydrogenase).tw.
- 9 (acyl coa dehydrogenases or acyl-coa dehydrogenases or dehydrogenases acyl-coa).tw.
- 10 Lipid Metabolism, Inborn Errors/
- 11 Fatty Acid Desaturases/
- 12 fatty acid oxidation disorder\$.tw.
- 13 (FAOD or LC-FAOD).tw.
- 14 Mitochondrial fatty acid [oxidation.tw](#).
- 15 FAO defect\$.tw.
- 16 Metabolism, Inborn Errors/
- 17 or/1-16

18 [meta-analysis.pt.](#)
19 (MEDLINE or systematic review).tw.
20 intervention\$.ti.
21 or/18-20
22 17 and 21 d
23 limit 22 to (english or french)
24 limit 23 to yr=2000-current
25 animals/ not humans/
26 24 not 25
27 26 not (comment or editorial or letter).pt.
28 exp cohort studies/
29 cohort\$.tw.
30 controlled clinical [trial.pt.](#)
31 exp case-control studies/
32 (case\$ and control\$).tw.
33 (case\$ and series).tw.
34 or/28-33
35 17 and 34
36 limit 35 to (english or french)
37 limit 36 to yr=2000-current
38 animals/ not humans/
39 37 not 38
40 39 not (comment or editorial or letter).pt.
41 27 or 40

We uploaded the citations retrieved from the search to Covidence,²⁷ removing duplicate

citations both manually and by autodetection within Covidence. Two reviewers (AK and AF) worked independently to screen all citations against our inclusion criteria (Table 1) in two stages of screening. Eligible papers were reports of primary studies that used any design, had to include at least 5 participants (i.e., no single case reports), and reported on treatment practices for MCAD deficiency. In the first stage of screening, we considered titles and abstracts only. Citations that were deemed definitely or possibly eligible for inclusion moved on to the second stage of screening, where we considered the full texts of the papers screened. Reasons for exclusion at stage 2 were documented manually. Throughout the screening process, conflicts were resolved by discussion and a third reviewer (BP) was consulted to resolve disagreements.

For the final set of included papers, one reviewer (AK) abstracted key data into an excel document. We abstracted data regarding study characteristics (first author, title and year of publication), study design, target population, sample size, and aspects of acute and chronic disease management, particularly carnitine supplementation practice and fasting time limits. We also extracted any information about differential management according to potential markers of disease severity, notably genotype or biochemical phenotype. Aligned with common practice for scoping reviews,²⁶ we did not do a quality appraisal of the included studies, given that the purpose of this scoping review was to identify and describe published information regarding current practice, regardless of the quality of the research itself.

Table 1. Inclusion and exclusion criteria for scan of literature

Inclusion	Exclusion
<ul style="list-style-type: none"> ● published peer-reviewed literature ● published since 2000 ● published in English or French ● primary studies describing current actual care being provided to patients with FAOD or a survey of health care providers or a published protocol ● description of management available in paper ● any treatments ● any study designs ● study includes ≥ 5 participants or is a provider survey or published protocol 	<ul style="list-style-type: none"> ● primary studies published prior to 2000. ● publications not published in either English or French ● primary studies related to inherited metabolic disorders other than MCADD/VLCADD where FAOD not discussed separately ● studies that do not describe care or management ● case reports or series including < 5 participants (unless the paper is a survey of practitioners or a published protocol)

Methods for secondary data analysis of a Canadian pediatric cohort

The cohort data were analyzed to understand actual practice in managing children with MCAD deficiency who were receiving care in Canada.

Data source and sample

Virtually all Canadian children diagnosed with inherited metabolic diseases (IMD) receive care at one of 16 hereditary metabolic disease treatment centres, based at pediatric academic health sciences centres across Canada.²⁰ With parental consent, the Canadian Inherited Metabolic Diseases Research Network (CIMDRN) cohort study collected clinical data from patient charts for children diagnosed with one of 31 eligible IMD, including MCAD deficiency, who were born between 2006 and 2015 and treated at 1 of 13 of these centres, which participate in the CIMDRN network. Participants were enrolled in CIMDRN starting in 2014. While the cohort study theoretically completely identifies and invites enrollment of all children with MCAD deficiency at the 13 centres (including children who are deceased after receiving some care at a metabolic centre), a small number of neonates with severe disease may be deceased prior to being seen in a metabolic clinic and would not be identifiable to the study; systematic estimates of the relative frequency of very early neonatal death due to MCAD deficiency are not available but this is believed to be quite rare. The CIMDRN cohort study also has a current non-participation rate of 205/942 (22%; across all diseases), mainly due to inability to reach potential participants to invite their participation in the study, which is consent-based. While care for participants is continued at other participating centres if participants have moved between study centres, those who move out of Canada or to a non-participating centre are lost to follow-up; approximately 1% of enrolled participants in CIMDRN (across all disease) have been lost to follow up.

Data collection for CIMDRN was previously approved by research ethics boards (REB) at all participating centres and use of the data for this study was approved by the REB at the Children's Hospital of Eastern Ontario (CHEO REB Protocol# 20140436-01H). The data cover

diagnostic information (clinical, biochemical, molecular) and basic patient characteristics such as date of birth, sex, caregiver information, centre of diagnosis and medical history. In addition, longitudinal data cover disease manifestations and interventions and services received or prescribed, including dietary interventions and pharmacotherapy prescriptions. This information is entered into the database retrospectively from the child's medical chart; the chart itself is typically updated at each clinical encounter with the metabolic treatment centre. Data were originally entered into CIMDRN's central database (held at the Children's Hospital of Eastern Ontario) by research coordinators based at each site, through a secure web-based data entry system (REDCap).²⁹ All data underwent a verification process to ensure that they were complete and to identify potential errors. Enrollment and data entry were still in progress at some centres when this analysis was completed; results reflect analysis of all completed data as of December 22nd, 2017.

Variables

Table 2 identifies the variables from the CIMDRN database that were used in the present analysis, within the categories of diagnosis and classification, demographics, medical history, clinical characteristics, diagnostic care, and clinical management. Diagnostic and classification variables were collected once (baseline data), typically during the diagnostic testing phase in children with MCAD deficiency, and they remain constant over time (for example, ascertainment method and genotype). Symptomatic status was reported in the chart as part of ascertainment. Method of ascertainment was categorized as: newborn screening only; newborn screening in combination with another method (for example, sometimes an infant with MCAD deficiency receives a positive NBS result and that contributes to the diagnosis, but the infant was also symptomatic and was already being investigated diagnostically); or other method only (for

example, an infant with an older sibling who has MCAD deficiency will go directly to diagnostic testing). Age at each chart entry was calculated based on date of birth and recorded date of the clinic visit. Neonatal complications were reported in open text fields and categories were created by grouping equivalent complications.

Because we were particularly interested in how disease management might vary according to potential markers of disease severity (genotype and biochemical phenotype), we also included these variables in our analysis. These were also recorded once, as baseline data collected during the diagnostic process. Genotype categories were created by reviewing allele results from diagnostic genetic testing for children with MCAD deficiency who received such testing. Since the c.985A>G mutation is believed to be associated with a more severe phenotype among individuals with MCAD deficiency⁹, we classified individuals based on the presence of this c.985A>G mutation as homozygous (two copies of this mutation), compound heterozygous (one copy of this mutation and another pathogenic mutation), and other genotypes not including any copies of this mutation. There were some children in the CIMDRN cohort who were siblings with MCAD deficiency, and in some of these families, some but not all siblings had received molecular genetic testing. In such families, we assumed that the siblings who had not been tested but were diagnosed with MCAD deficiency had the same genotype as those who had been tested, and we imputed the genotype accordingly (n=4).

Biochemical phenotype was represented by concentrations of acylcarnitines and free carnitine, which are markers of fatty acid oxidation that are used in NBS and in diagnosis to identify children with MCAD deficiency and may also be a proxy indicator of disease severity. Both NBS and diagnostic analyte levels were entered into open fields either at the participating site or centrally (the latter based on uploaded lab test result forms submitted by the participating

site). Separate measures were included for octanoylcarnitine (C8), a medium chain acylcarnitine that is the primary newborn screening analyte used to identify MCAD deficiency (children with higher C8 levels may have more severe disease), as well as the acylcarnitines C6, C10, and C10:1, which are secondary markers of disease; and C0 (free carnitine).

Regarding disease management practices, we extracted longitudinal data for variables reflecting the two treatments of interest, fasting times (maximum fasting time when well), and carnitine supplementation, which were time-varying (Table 2). Carnitine supplementation could be reported as a medication prescription or as a supplement prescription, therefore multiple fields were searched for this information. Comment fields were also reviewed to determine any additional information regarding carnitine status. For example, a comment may indicate that a patient was prescribed carnitine but was not able to re-fill the prescription, or that therapy was discontinued due to side effects. This additional information sometimes added complexity to classifying treatment status for participants. In all cases, we made the simplifying assumption that the course of treatment described in a patient's chart reflected clinical practice, while recognizing that in reality, prescription, receipt, and adherence to therapy is likely to be the result of an ongoing interaction between the clinician, the patient/family, and external influences. In the remainder of this paper we use the terminology that a patient was "prescribed", "received", or "used" carnitine to refer to the information in the chart. Fasting time duration was reported in an open text field. The exact value (i.e., number of hours fasting time) was used unless a fasting time range was provided, in which case the mean of the range of values was used.

Table 2. Variables used in analysis of CIMDRN data

Category	Variables
Diagnosis and classification	<ul style="list-style-type: none">● Ascertainment method● Symptomatic status at diagnosis
Demographics	<ul style="list-style-type: none">● Age and year of birth● Sex
Medical History	<ul style="list-style-type: none">● Neonatal complications
Screening and Diagnostic Testing Results	<ul style="list-style-type: none">● Newborn screening and diagnostic testing results regarding blood acylcarnitines and free carnitine (biochemical phenotype)● Medical genetic testing (genotype)
Clinical Management	<ul style="list-style-type: none">● Carnitine supplementation● Fasting time duration

Analysis

Using SAS software[®] version 9.4 (SAS Institute Inc., Cary, NC, USA), we performed descriptive data analyses on the baseline and longitudinal data. Due to the privacy policy for the cohort study, we could not report cell sizes of less than 5 cases. We collapsed categories within categorical variables as necessary to adhere to this policy.

We described the study population using proportions and frequencies for method of disease ascertainment, whether the patient was considered symptomatic at ascertainment, year of birth, neonatal complications, and genotype. To explore the association between biochemical phenotype and genotype, median and range of acylcarnitine and free carnitine concentrations

(umol/L) were calculated separately for C8, C10, C6, C10:1 and C0, and stratified by the three genotype groups and screening/diagnosis measures. As described earlier, genotype was classified as homozygous, heterozygous or other based on the presence of the common c.985A>G mutation; the last two genotype categories were combined where required to adhere to the privacy policy. Carnitine was reported as ever or never used, overall or during specified age categories; fasting time was reported in hours, as the median and the range of reported fasting times by age category. Given recent discussion and debate about carnitine supplementation, we also described trends in carnitine use by calendar time within age categories to evaluate potential time trends. To investigate associations between proxy indicators of disease severity and disease management, carnitine was summarized by genotype and by NBS and diagnostic biochemical phenotype (C8). Fasting time was also summarized by genotype.

Carnitine supplementation and fasting times were time-varying and we took a simplifying approach to summarizing their frequency over clinic visits for the purposes of the analysis. For example, carnitine may have been started or stopped multiple times for a single child, as recorded in the database, which was organized by fields completed at each clinic visit, sequenced by calendar time. To report on use of carnitine during a particular age interval, we used a child's birthdate to determine the child's age at each data entry point (the clinic visit). Any documented use of carnitine during an age interval (e.g., 6-12 months), regardless of its frequency or continuity of use during that age category, was considered prevalent use for that interval. For the analysis of carnitine use and fasting periods for different age groups, we allowed the same participants to contribute to the analysis for multiple age categories because of longitudinal data. Because treatment prescriptions were updated only at clinic visits, and clinic visit schedules varied in a way that did not neatly fit within the age intervals in the analysis, we considered

available data prior to, during and after an age interval, to impute values if data were missing but could confidently be imputed based on preceding and following values at each age category. For example, if there was no information for carnitine use recorded at a specific age category for a participant because of no clinic visit during that age interval, but the information about carnitine use was the same for that participant at both the previous and next age category, then we imputed the missing information for the missing age category to be equal to the values for the preceding and following age categories. If the information was different at the preceding versus following time point (for example, if fasting time was missing at a particular age and was recorded as 4 hours at the preceding age but 12 hours at the following age), we did not impute but rather maintained a ‘missing’ entry for that age category and excluded it from the reporting in that age category (casewise deletion). We also carried forward the last observation for fasting time or carnitine supplementation if the child continued to be followed over time (i.e., we had some visit data at later ages) but the data indicated “no update” for these variables. In this case, “no update” means that the chart did not mention the intervention (carnitine supplementation or fasting interval) and thus the assumption was that there was no change to the intervention. While this seemed a reasonable strategy for carnitine supplementation, which is a prescription, we were uncertain about its use for fasting time, out of concern that perhaps fasting times were discussed but not recorded in the chart. We conducted sensitivity analyses where there was missing data for carnitine and maximum fasting time in order to evaluate the robustness of the results against these assumptions about missing data. We also reviewed the frequency with which data were imputed when missing for carnitine and fasting (Appendix B).

This analysis was the first of its kind, as far as we are aware, providing an opportunity to describe treatment practices in a Canadian pediatric cohort diagnosed with MCAD deficiency.

Given the overall exploratory nature of the analysis, we did not perform formal statistical hypothesis tests to evaluate whether the trends and patterns in the data reflected statistically significant associations. Rather, we anticipated that our analyses would identify general patterns that could be compared with related studies from the scoping review and that the findings would generate hypotheses that could be further evaluated in future research. In accordance with CIMDRN's privacy policy, we collapsed variable categories to avoid reporting cell counts of <5.

Results of scoping review:

Our database search identified a total of 2114 citations after removing duplicates (Figure 1). After screening titles and abstracts in the first stage of screening, 33 papers passed on to the second stage of screening (full text). In total, 2 studies from the literature search and 3 studies identified manually were selected for data extraction. Grey literature was searched but no eligible studies were identified. The study characteristics and key findings for each paper included in the scoping review are presented in Table 3.

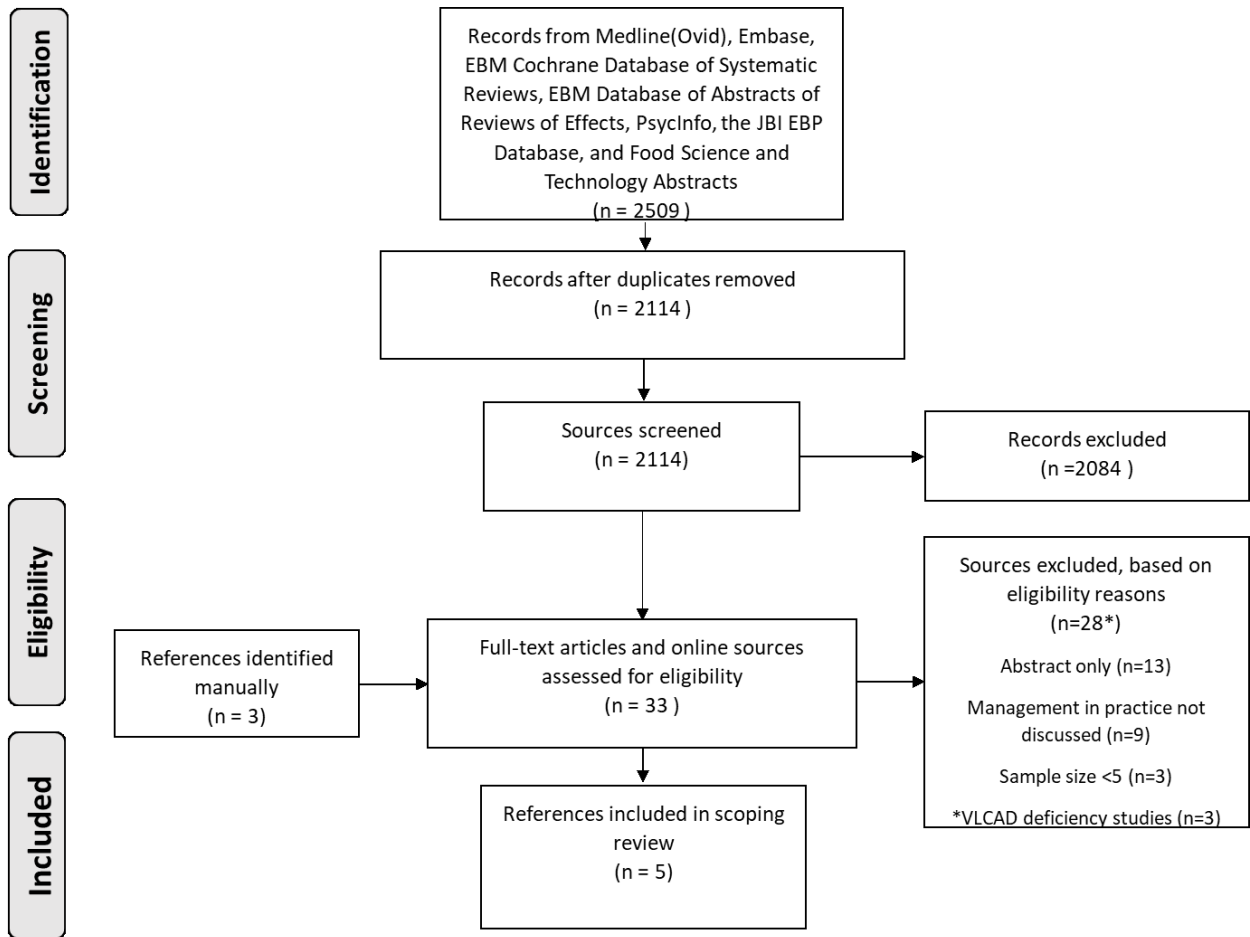


Figure 1. PRISMA flow chart for scoping review.

Table 3. Scoping review literature data extractions on MCAD deficiency management relating to carnitine supplementation and fasting time.

First author, Year, Title	Study design and sample size	Carnitine supplementation	Fasting
Solis et al., 2002 Management of fatty acid oxidation disorders: a survey of current treatment strategies ³⁰	<ul style="list-style-type: none"> A survey sent through a listserv to which many American metabolic dietitians belong N=117 total surveys sent of which n=49 returned (response rate 41.9%) 25/49 (51%) had no FAOD patients, 24/49 (49%) reported having previous/current FAOD patients, n=114 individual patients with MCAD deficiency 	<ul style="list-style-type: none"> Supplementation with carnitine as management strategy reported by 15/19 (79%) metabolic dietitian respondents Reported dosage (mg/kg/day) of carnitine: median value (range): 75 (25-100) 	<ul style="list-style-type: none"> Increased meal frequency as management strategy reported by 18/19 (95%) metabolic dietitian respondents Increased frequency of meals, reported as time (in hours) between meals by age groups; median value (range): <1 year: 4 (2.5-5) Children: 5 (2.5-10) Adults: 5 (4-12)
Walter, 2003 L-carnitine in inborn errors of metabolism: what is the evidence? ³¹	<ul style="list-style-type: none"> A questionnaire posted on electronic mailing list to determine current practice at metabolic clinics 2 questions asked: (i) Use of L-carnitine as a long-term supplement in patients with MCAD deficiency (also asked about propionic/methylmalonic acidemia); (ii) Use of regular oral carnitine (a) in all patients, (b) in those with proven carnitine deficiency in blood or (c) never? Respondents included 31 clinics in Europe, North America, Asia and Australia (28/31 responded to questions regarding carnitine practice) 	<ul style="list-style-type: none"> 11/28 (39%) prescribe l-carnitine to all patients with MCAD deficiency 3/28 (11%) prescribe carnitine for participants <6 yrs. 9/28 (32%) prescribe where proven deficiency. 5/28 (18%) do not prescribe, however 2 of these specified prescribing only when unwell. The reported dosage range of those who prescribe carnitine was 25 to 300 mg/kg/day, although this was not specific to only MCAD deficiency, but also included 3 other IMD. 	<ul style="list-style-type: none"> Not discussed
Lund, Allan et al., 2010 Clinical and Biochemical monitoring of patients with fatty acid oxidation disorders ³²	<ul style="list-style-type: none"> A monitoring approach described specifically for Danish patients with FAOD, specified that it is not an evidence-based standard for monitoring. The monitoring described in the form of a protocol are minimal procedures done for all patients. No additional details provided. Monitoring strategies covered include frequency of visits, clinical monitoring and biochemical monitoring. 	<ul style="list-style-type: none"> Carnitine supplementation is not discussed however plasma carnitine is monitored at every visit to the out-patient clinic, substituted to monitoring every 3 months if plasma levels <12μM. No details provided regarding supplementation if plasma levels are low. 	<ul style="list-style-type: none"> A dietetic review is done at all clinic visits and involves the review of maximum fasting time limits. No values are provided.
Potter, Beth et al., 2012 Variability in the clinical management of fatty acid oxidation disorders: result of a survey of Canadian metabolic physicians ³³	<ul style="list-style-type: none"> A survey of metabolic physicians identified from the Canadian Garrod Association (Canadian Association of Centres for the Management of Hereditary Metabolic Diseases). The questionnaire reported on MCAD, VLCAD and LCHAD/MTP deficiencies: (i) utility of interventions, separated by chronic management or during illness, by assessing whether 	<ul style="list-style-type: none"> Carnitine supplement for chronic management: 33% would use for all or most patients, 33% would use in rare cases or never Carnitine for acute illness: 44% would use for all or most patients, 44% would use in rare cases or never 	<ul style="list-style-type: none"> 94% would recommend avoidance of fasting for all or most patients 44% would recommend increased meal frequency for all or most patients Fasting intervals not reported

	<p>these would be used for all, most or some patients, rarely or never (5-point scale);</p> <p>(ii) value of patient, physiological, social and biochemical characteristics in determining future treatment (4-point scale).</p> <ul style="list-style-type: none"> 18/40 (45%) responded to the survey, of which 17 reported they were physicians involved in care at IMD clinics in the past 12 months, with 77% reporting treatment of MCAD deficiency patients. 		
<p>Couce, Maria Luz et al., 2013</p> <p>Newborn screening for MCADD: regional experience and high incidence of carnitine deficiency³⁴</p>	<ul style="list-style-type: none"> Study population included patients diagnosed through NBS with MCAD deficiency by two Spanish regional NBS programs (Galicia and western Andalusia) and one Portuguese (north and central) program. A total of 45 patients were assessed; 39 had follow-up data and genotypes were also provided, with n=28 homozygous for the c.985A>G mutation and n=11 other genotypes. Follow-up practice was described in the methods section even though the aim of the study was to determine associations between genotype and biochemical findings. 	<ul style="list-style-type: none"> L-carnitine supplement was reported to be prescribed to patients if free carnitine (C0) was <20umol/L in plasma or <12umol/L in blood spots Supplementation was stopped after 2 independent readings showing free carnitine above the respective thresholds. The prescribed dose range was 20-60 mg/kg/day. 32/39 (82%) patients were prescribed a carnitine supplement. 26/28 (93%) patients who were homozygous for c.985A>G required a carnitine supplement, 15 of whom were reported to later stop supplementation (based on biochemical results); 11/15 restarted supplementation. Based on biochemical threshold, carnitine was required continuously in 11 homozygous patients. 6/11 (55%) patients with other genotypes required a carnitine supplement; only 1 remained on carnitine. 	<ul style="list-style-type: none"> It was reported that during follow-up patients were avoiding prolonged periods of fasting and lipolysis, however no limits were described.

The 5 articles included in the scoping review included one Danish protocol³² describing monitoring for the management of all FAOD patients, three practitioner surveys (an international clinic questionnaire survey asking only about carnitine supplementation practice,³¹ a US survey of dietitians,³⁰ and a Canadian survey of physicians³³), and one case series describing the management of patients with MCAD deficiency identified by NBS in Southwest Europe (Spain and Portugal).³⁴ Here we present the scoping review findings regarding carnitine supplementation and avoidance of fasting, as well as information regarding management practices in relation to disease severity.

Carnitine supplementation

Carnitine supplementation was addressed by four of the five papers we reviewed.^{30,31,33,34} The surveys reported high variability in carnitine use. 79% of US dietitians in a 2002 survey reported prescribing carnitine to MCAD patients, however it was not specified whether carnitine was recommended under specific conditions, or for all patients. The median dose prescribed was 75mg/kg/day.³⁰ In the 2012 survey of Canadian physicians, 33% would prescribe carnitine as part of chronic care to all or most patients, while 44% reported that they would prescribe carnitine for acute illness care to all or most patients; however, an equivalent 33% and 44% would prescribe carnitine in rare cases or never as part of chronic care or during acute illness, respectively. Of respondents to a 2003 international clinic survey, 39% reported always using oral carnitine, 11% prescribed carnitine only to those aged <6 years old, 32% only in cases of proven carnitine deficiency and 18% never prescribed carnitine.³¹ The study of patients receiving care in clinics in southwest Europe reported that 82% of patients diagnosed with MCAD deficiency were supplemented with carnitine at some point to keep their carnitine levels 'normal', as defined by these centres.³⁴

Avoidance of fasting

Four of the five reviewed papers reported on practice as it relates to avoidance of fasting and/or increased meal frequency, providing varying detail.^{30,32-34} The 2002 survey of 19 dietitians from the US identified that 95% reported treating MCAD deficiency patients using increased meal frequency, with median fasting times of 4 and 5 hours in children <1 year old and in older children and adults, respectively, but with wide ranges within these age groups.³⁰ In a survey of 18 Canadian metabolic physicians published in 2012, 94% of participants stated that they would recommended avoidance of fasting and 44% would recommend increased meal frequency, for “all or most” individuals with MCAD deficiency.³³ The monitoring protocol for Danish patients did not provide specific details of fasting practice, however it was reported that at each visit, along with a full dietary review, fasting limits are reviewed.³² Similarly, the study from Southwestern Europe reported that fasting avoidance was reported at follow-up visits but no limits were defined.³⁴

Variation in care according to disease severity

We were interested in whether the studies we reviewed identified practice variation according to disease severity, given the questions that have arisen in the context of newborn screening regarding management of cases of MCAD deficiency that may be expected to be mild. The 2012 survey of Canadian physicians reported on characteristics that were considered to be important in determining chronic care for patients. Among varying individual, biochemical and social characteristics, 33% reported that genotype and 22% that biochemical phenotype (plasma acylcarnitines) were “essential or very important” as influences on chronic care decisions.³³ By contrast, neither genotype nor phenotype was described to be taken into consideration in the protocol for monitoring Danish patients.^{32,33} In the study of approximately 45 patients receiving

care in southwest Europe, although the authors did not describe adjusting practice based on genotype, they did report that carnitine supplementation was required much more frequently among patients with the homozygous genotype for the c.985A>G mutation compared to those with other genotypes, given a high incidence of carnitine deficiency in these patients.³⁴ Specifically, 93% of patients with the homozygous genotype and 55% of patients with other genotypes required carnitine supplementation. Also, 53% of patients with the homozygous genotype were reported to later stop supplementation (based on criteria to define carnitine deficiency in the clinics' protocol), but then of these, 73% were required to restart supplementation. Among the participants with other genotypes, only one participant (6%) was required to continue on carnitine.³⁴ None of the other studies discussed either genotype or biochemical phenotype as factors that were considered as influences on management practice with respect to carnitine supplementation or avoidance of fasting.

Results of secondary data analysis of data from the Canadian Metabolic Diseases Research Network (CIMDRN):

Our descriptive analysis of baseline and long-term follow up data included 107 participants with MCAD deficiency enrolled in the Canadian pediatric cohort study (CIMDRN). The counts of non-missing complete data included at each step are presented in Figure 2. Frequencies of missing data by carnitine and fasting time, reflecting the percent of data imputed, are present in Appendix B, Tables B.S3. Baseline sample characteristics are presented in Table 4.

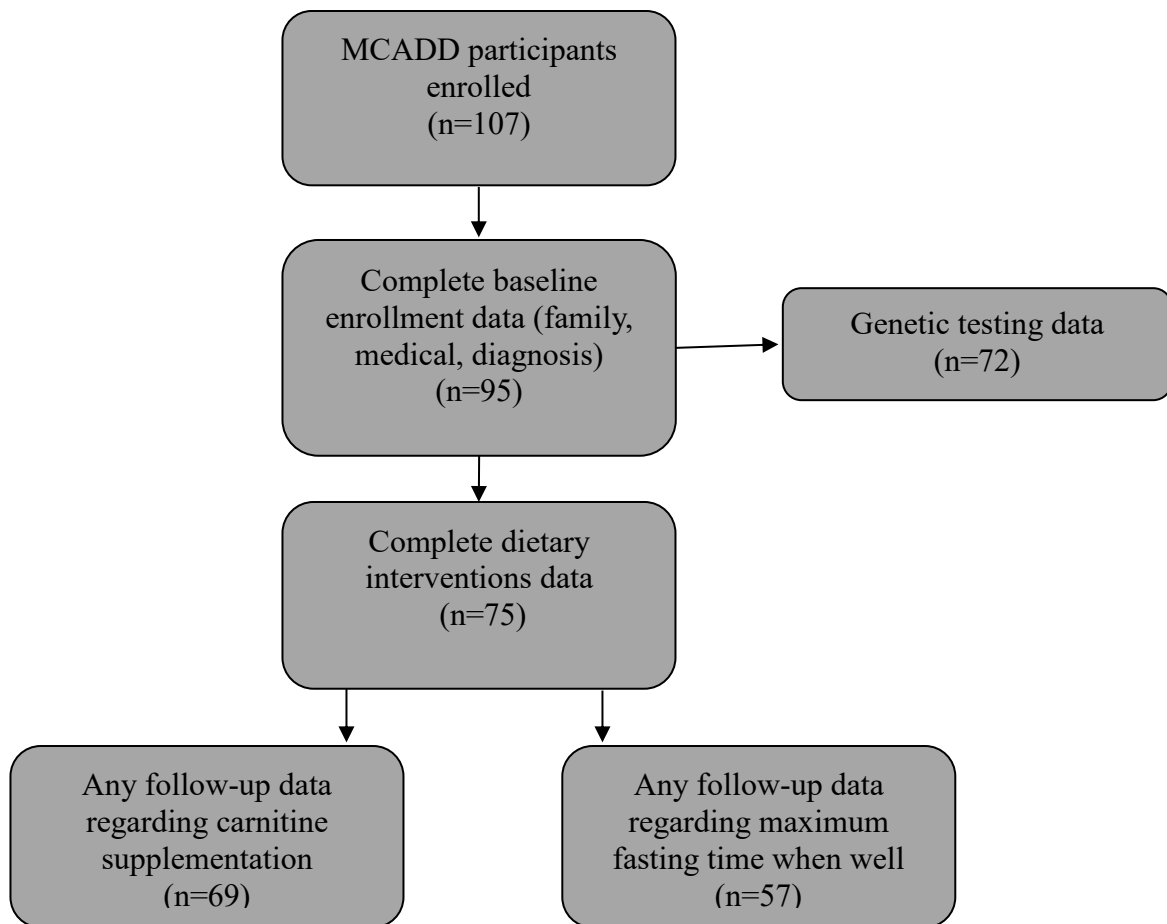


Figure 2. Data flowchart showing the count of complete and non-missing data by the variable of interest in the CIMDRN cohort

Table 4. Baseline characteristics of participants with MCAD deficiency in the CIMDRN cohort.^a

Characteristic (number with valid data for that characteristic)	n (%)
Year of birth (n= 107)	
2006-2007	15 (14)
2008-2009	24 (22)
2010-2011	17 (16)
2012-2013	25 (23)
2014-2015	26 (24)
Sex (n=107)	
Male	58 (54)
Means of ascertainment (n= 96)^b:	
Newborn screening only	81 (84)
Newborn screening and other	>10
Other ascertainment method only	<5
Symptomatic at ascertainment (n=96)	7(7)
Neonatal complications (includes antibiotics, hypoglycemia, IV fluids, respiratory distress and/or other complications) (n=85)	
Yes	27 (32)
Antibiotics	7 (8)
Hypoglycemia	12 (14)
IV fluids	6 (7)
Respiratory distress	5(6)
Other (ex. Meconium aspiration, acidemia, bradycardia)	15(18)
No	58 (68)
Genotype (n=72)	

Homozygous for c.985A>G	29 (40)
Compound Heterozygous for c.985A>G	32(44)
Other mutation	11 (15)
^a Characteristics of all participants enrolled in CIMDRN with MCAD deficiency, with complete data on variable of interest. ^b Exact numbers were suppressed in accordance with CIMDRN's privacy policy for counts <5.	

The vast majority of participants were identified through newborn screening, alone or in combination with another method (e.g., family history, symptomatic); 7% of participants were symptomatic at diagnosis, as reported in the patient chart. There was a relatively even distribution of participants by birth year, with the exception of 2006-2007, which included relatively fewer participants. A third (32%) of participants were reported to have neonatal complications. Among children with known genotype, there were nearly equal proportions who were homozygous (40%) and compound heterozygous (44%) for the common mutation (c.985A>G), while 15% had other mutations in both alleles.

Table 5. Newborn screening and diagnostic testing results of acylcarnitine concentration (units: umol/L) by genotype severity

Severity by genotype	Homozygous for c.985A>G (n=25) median (range*)	Compound Heterozygous for c.985A>G (n=31) median (range*)	Other genotype (n=11) median (range*)
Newborn Screening:			
C8	14.8 (1.13-35.4)	4.43 (0.48-25.5)	2.32 (0.67-14.8)
C10	0.98 (0.26-2.05)	0.88 (0.26-119.0)	0.69 (0.40-1.11)
C6	1.64 (0.30-56.0)	1.01 (0.30-176)	1.04 (0.28-1.80)
C10:1	0.44 (0.27-0.91)	0.36 (0.10-0.89)	0.47 (0.18-0.89)
Diagnosis:			
C8	3.43 (1.38-33.7)	1.83 (0.15-13.5)	0.94 (0.16-38.7)
C10	0.44 (0.13-2.90)	0.40 (0.26-3.07)	0.38 (0.22-2.89)
C6	0.92 (0.37-5.90)	0.47 (0.06-2.85)	0.36 (0.05-3.35)
C0	19.6 (16.0-24.8)	28.7 (13.1-41.2)	32.3**
*Range is reported as minimum to maximum acylcarnitine value. ** Where <5 cases the range was not provided based on CIMDRN's privacy policy.			

With respect to newborn screening and diagnostic biochemical analytes there was an overall pattern of decreasing acylcarnitines by decreasing genotype severity, particularly for the primary MCAD analyte C8 (Table 5), with children who were homozygous for the most common genotype having the most severe biochemical phenotypes (elevated medium chain acylcarnitines). Exceptions were C10:1 (NBS) and C10 (diagnosis), for which there were no clear trends; and C0 (diagnosis), where the trend was in the opposite direction (higher free carnitine with less severe genotype). Across all analytes for both NBS and diagnostic values, ranges were large, indicating high variability within genotypes.

Carnitine supplementation

We identified an increase in the proportion of participants receiving carnitine supplementation with increasing age (age categories were not mutually exclusive as participants could contribute data to more than one category). Within each age category, there was a general decrease in the proportion of participants who were using carnitine with more recent birth years, i.e., a decline in the use of carnitine supplementation with calendar time (Table 6).

A sensitivity analysis was conducted, whereby carnitine data by age category that had been missing but imputed by last observation carried forward, with modification to account for the next value, were re-coded as ‘not on carnitine’, to account for the possibility that an absence of carnitine data in the chart reflected its non-use rather than its continued use (Appendix B, Table B.S1). The overall results were similar with this imputation. There were n=28 values imputed for carnitine and the frequency of missing data by age category is presented in Appendix B, Table B.S3.

Table 6. Participants receiving carnitine supplements at any time during follow-up in each age category, stratified by year of birth.

Age of child ^a	Total	By year of birth		
	n receiving carnitine/ total n (%)	2006 to <2009 n receiving carnitine/ total n (%)	2009 to <2012 n receiving carnitine/ total n (%)	2012 to 2015 n receiving carnitine/ total n (%)
<6 months	27/55 (49)	8/14(57)	10/14(71)	9/27 (33)
6 to <12 months	29/57 (51)	10/14 (71)	8/16 (50)	11/27 (41)
12 to <18 months	32/59 (54)	9/14 (64)	11/17 (65)	12/28 (43)
18 to <24 months	35/56 (63)	11/14 (79)	11/18 (61)	13/24 (54)
24+ months	36/54 (67)	16/19 (84)	11/17 (65)	9/18 (50)

^aData across different age categories are not independent: data were longitudinal, thus participants could contribute data to more than one age category.

Table 7. Newborn screening and diagnostic C8 median and range analyte values (umol/L) for participants according to their receipt of carnitine supplements, by age (years)

	Age (years)					
	<1		1 to <2		2+	
	Yes	No	Yes	No	Yes	No
NBS C8 (umol/L)	14.1	6.83	14.0	8.45	13.6	9.21
Median (range)	(0.96-38.2)	(0.67-25.5)	(0.48-38.2)	(0.75-25.5)	(0.67-38.2)	(0.75-35.4)
(n)	(n=32)	(n=13)	(n=32)	(n=11)	(n=31)	(n=8)
Diagnostic C8 (umol/L)	3.02	1.22	2.70	2.54	3.4	2.54
Median (range)	(0.23-33.7)	(0.16-10.6)	(0.16-33.7)	(0.37-11.3)	(0.16-11.28)	(0.64-10.6)
(n)	(n=22)	(n=10)	(n=24)	(n=9)	(n=22)	(n=7)

We described prescription for carnitine supplementation by median C8 values (C8 is considered the primary analyte that is elevated in children with MCAD deficiency and thus reflects biochemical phenotype) as a proxy indicator of disease severity. Based on both NBS and diagnostic values, participants who received carnitine supplementation had higher C8 values than those who did not receive carnitine across all age categories (Table 7). The range of C8 values, however, is quite large within each group. Similarly, a higher proportion of participants with the homozygous c.985A>G genotype (57-79%) were reported to be receiving carnitine supplementation than participants with other genotypes (29-41%), across all age categories (Table 8).

Table 8. Carnitine supplementation across age categories by genotype.

Age of child ^a (years) n(%)	Genotype	
	Homozygous c.985A>G	Compound heterozygous c.985A>G or Other
<1	13/20 (65)	7/24 (29)
1 to <2	12/21 (57)	9/22 (41)
2+	15/19 (79)	7/17 (41)

^a Data across different age categories are not independent: data were longitudinal, thus participants could contribute data to more than one age category.

Fasting time

Median fasting times by age (defined as maximum fasting time when well) in the full sample increased from 4 hours in infants <6 months of age to 11.4 hours for children 24 months of age or higher (Table 9). Finally, we found no clear pattern when we examined median fasting times by genotype within each age category; median fasting times were very similar and ranges were quite broad (Table 9).

Because we imputed some missing fasting times by carrying forward from a previous observation if a child continued to be followed in our dataset but there was “no update” to the fasting variable, we were concerned that some of the short fasting time outliers were the result of fasting times that had perhaps been lengthened with age but had not been recorded as such in the chart. For this reason, we conducted another sensitivity analysis (Appendix B, Table S.2), where values were not carried forward when no update was provided and were left as missing. The results were not greatly affected by this change (with median fasting times having no change

across age groups). There were n=39 observations in total imputed for fasting time (Appendix B Table B.S3).

Table 9. Maximum fasting time when well by age, overall and by genotype category

Fasting time by age category median(range)	Full Sample	Genotype Category	
	Total	Homozygous c.985A>G	Compound heterozygous c.985A>G or Other
<6 months	4.0 (2.5-6.5) (n=45)	4.0 (3.0-6.0) (n=14)	3.5 (2.5-6.5) (n=21)
6 to <12 months	6.0 (2.5-12.0) (n=42)	6.0 (5.0-12.0) (n=12)	5.5 (2.5-10.0) (n=21)
12 to <18 months	10.0 (3.0 -12.0) (n=39)	10.0 (5.0-12.0) (n=13)	8.0 (3.0-12.0) (n=16)
18 to <24 months	10.0 (4.0-12.0) (n=31)	10.0 (6.0-12.0) (n=14)	10.0 (4.0-11.0) (n=14)
24+ months	11.4 (3.5-12.0) (n=39)	11.7 (3.5-12.0) (n=14)	10.2 (3.5-12.0) (n=13)

Discussion:

We used a scoping review of published literature and a secondary analysis of Canadian data from a pediatric cohort study to identify practices in the management of MCAD deficiency in the newborn screening era. We addressed two important areas of care, namely, the use of carnitine supplementation and fasting times. We also investigated whether there was evidence from either data source (the scoping review or the cohort analysis) that these management practices for MCAD deficiency may differ according to perceived disease severity, particularly as defined by genotype and biochemical phenotype.

Carnitine supplementation

From the scoping review, carnitine supplementation practice was highly variable, and the amount of detail provided in each study differed greatly. The international survey of clinics and the survey of Canadian physicians reported that fewer than 40% of providers/clinics would routinely prescribe carnitine to patients with MCAD deficiency;³³ while among US dietitians, nearly 80% reported prescribing carnitine.³⁰ Among patients with MCAD deficiency in participating clinics in a southwestern European study, 82% were prescribed carnitine based on identified carnitine deficiency. From our secondary analysis of data from a Canadian pediatric cohort, we identified a trend of increasing use of carnitine with increasing age (from 49% of patients <6 months of age to 67% of patients aged 2 years and older), but when stratified by year of birth this trend was less apparent. We also identified a trend of decreasing carnitine use for patients of similar age but born in more recent years (e.g., among patients < 6 months of age, 49% and 33% of those born in 2006-2008 and 2012-2015, respectively, were prescribed carnitine), which may reflect changing practice over time. In the scoping review, carnitine

supplementation was described in two of the studies as frequently prescribed based on evidence of carnitine deficiency^{31,34} and in a third study, carnitine monitoring was recommended.³² We did not review actual plasma carnitine levels in our Canadian cohort for the purposes of this study.

Despite a biological rationale for carnitine supplementation given its involvement in transportation of fatty acids across cellular membranes, it is unclear if supplementation leads to improved outcomes in patients with MCAD deficiency.³⁵ Carnitine supplementation has been investigated in relation to exercise outcomes among individuals with MCAD deficiency but results have been mixed. For example, in one study, impaired fatty acid oxidation was not corrected by l-carnitine supplementation for 4 weeks in MCAD patients²³ while another study reported that 4 weeks of supplementation resulted in increased exercise tolerance.³⁶ There is evidence that patients with MCAD deficiency frequently have low carnitine levels (secondary carnitine deficiency),¹ which may require supplementation, corroborating the scoping review findings that low carnitine levels are frequently reported as a reason for supplementation. In particular, the case series from southwest Europe in our scoping review found that clinics reported a high incidence of carnitine deficiency in their patients.³⁴

A previous summary of 16 NBS programs revealed that six programs recommended carnitine supplementation for children with MCAD deficiency, with two of these (in Oregon and Italy) recommending supplementation only if low carnitine was observed.³⁷ From our previous systematic review of guidance for the management of MCAD deficiency (see Chapter 2), we similarly found variability in recommendations regarding carnitine use; in some cases, carnitine was recommended for patients with low plasma carnitine^{10,17,18,38} but generally, few details were provided about the specific circumstances under which carnitine would be considered appropriate.^{24,39–42} Questions about the appropriate use of carnitine supplementation for MCAD

deficiency are particularly important given that there have been concerns about possible adverse effects.⁴³ Some side effects have been reported in MCAD patients after using doses of 100mg/kg/d for 4 weeks, specifically nausea and heavy fish-like odor,²³ but the potential for more serious adverse effects such as atherosclerosis and spontaneous hypoglycemia has also been discussed as a concern.^{21,30}

Fasting time

With respect to fasting times, four of the five studies included in our scoping review reported on fasting as a management strategy used by practitioners,^{30,32–34} similar to results from our systematic review (Chapter 2). The survey of Canadian physicians reported that 94% and 44% would use fasting avoidance and increased meal frequency, respectively, as long-term management strategies for “all or most patients”.³³ Similarly, 95% of US dietitians reported managing MCAD deficiency with increased meal frequency.³⁰ The other two studies that addressed fasting did not report on specific practices or limits but identified fasting avoidance as a treatment that was reported/discussed with patients. Most papers did not specifically describe the maximum length of fasting time for MCAD deficiency patients, overall or by age. An exception was the survey of 19 US dietitians, which reported the fasting times for infants, children and adults, with much variability in fasting limits prescribed by dietitians.³⁰ With respect to fasting times in the Canadian pediatric cohort study, we found that median fasting times for children with MCAD deficiency, by age, ranged from 4 hours (for infants < 6 months of age) to 11.4 hours (for children 2 years of age and older). It is difficult to compare these results from the cohort study with the scoping review findings, given that fasting time is very inconsistently reported in the literature. In the study of US dietitians that reflected on management of 114 patients with MCAD deficiency, among patients < 1 year of age, reported

fasting duration was a range of 2.5 to 5 hours;³⁰ by comparison, in the Canadian pediatric cohort data, the range of fasting times for infants < 1 year of age was from 2.5 to 10 hours.³³ However, fasting times are heavily influenced by the exact age of the child as well as dietary requirements and growth, therefore data for broad age categories are challenging to interpret.

While it is clear that fasting avoidance is an important component of the management of MCAD deficiency, there is a lack of consensus on appropriate and safe time intervals. The fasting times we identified among Canadian children were similar to those that would be expected in populations of same-age children without MCAD deficiency.^{44,45} Varying fasting schedules have been described for MCAD deficiency in the literature, further emphasizing the lack of consensus regarding appropriate fasting schedules.^{17,46} Table 9 suggests that there may be some patients with abnormally short fasting times, particularly at >12 months of age, at which point a fast of up to 12 hours has been described as appropriate in some literature.⁴⁷ These children may be at risk of overfeeding and behavioral issues.^{17,47,48}

Management of MCAD deficiency by indicators of disease severity, particularly genotype

From our scoping review, genotype was endorsed as “essential or very important” as a determinant of chronic management for MCAD deficiency in only one third of Canadian physicians,³³ while a Danish protocol stated that genetic testing results were not used to determine treatment for participants, given that it may be safer to keep all participants on a preventative diet.³² However, when considering specific interventions, one study from our scoping review reported that 93% of patients who were homozygous for the classic MCAD mutation required carnitine supplementation to achieve normal carnitine levels, compared with only 55% of patients with other genotypes.³⁴ From our cohort analysis, a large proportion

(>50%) of participants with the homozygous genotype for the classic mutation were prescribed carnitine across all ages compared to about a third of participants in the heterozygous or ‘other’ genotype categories (Table 8); actual carnitine levels were not reviewed to determine deficiency in the cohort analysis. Given the potential association between biochemical phenotype and disease severity, we also investigated its association with management. The scoping review identified limited evidence that biochemical phenotype influences clinical management of MCAD deficiency; in the study of Canadian physicians, only 22% reported using plasma acylcarnitines to determine chronic management, while in the Danish monitoring protocol, acylcarnitines were not reported to be monitored at all in their patients.^{32,33} In our cohort study, we observed a pattern of higher median newborn screening and diagnostic C8 values (Table 7) in participants who used carnitine at any time compared to those who never used carnitine. Again, it is not clear whether this finding is related to risk perception or to differences in blood carnitine levels.

In our scoping review we did not identify evidence of different management with respect to fasting times in relation to genotype for patients with MCAD deficiency. Similarly, in our Canadian pediatric cohort, there appeared to be no major differences (Table 9) in fasting times by genotype. Given that avoidance of fasting is the main preventive treatment for MCAD deficiency, and the most severe consequence of extending fasting time too far would be the potential for metabolic decompensation (which in turn carries a risk of coma and mortality), it is possible that a preventative approach is taken with fasting and the same limits are prescribed to all patients regardless of predicted disease severity.

One of the findings of our systematic review (Chapter 2) was that fasting guidelines for MCAD deficiency were not well supported by evidence; to our knowledge, only one study has

defined fasting limits based on patient data from fasting studies in combination with a literature review.¹⁵ All the identified guidance documents considered fasting as the first treatment for MCAD deficiency, and just over half presented fasting schedules with varying detail ranging from recommendations that were to be used as rules of thumb, a recommendation to give an evening meal to prevent hypoglycemia, specific fasting times used by UK dietitians, or others providing no additional information other than ‘fasting avoidance’.^{10,17,31,41,47–49} Previously it has been suggested that fasting guidelines should be dropped, particularly the need for a late evening meal, in patients with mutations which were not usually seen in clinically presenting cases (i.e. potentially milder mutations) where residual enzyme activity was >10%.¹³ One potential explanation for our cohort analysis finding that there was a possible association between genotype and carnitine supplementation but no apparent relationship between genotype and fasting is that the use of carnitine in those with the “classic” MCAD deficiency genotype was in response to low blood carnitine levels and not due to a difference in perception of risk during treatment decision-making.

One challenge in identifying whether treatment of MCAD deficiency differs according to perceived disease severity is that severity is imperfectly predicted by factors such as genotype, given that the associations among genotype, biochemical phenotype (e.g., newborn screening and diagnostic levels of analytes such as C8), and clinical phenotype (risk of metabolic decompensation) are not firmly established. While clinical phenotype is particularly difficult to predict in a population of asymptomatic individuals identified by newborn screening and receiving preventive management, we were able, in our Canadian cohort, to explore the association between genotype and biochemical phenotype (C8 levels). We found that higher newborn screening and diagnostic levels of C8 were associated with the presence of the

c.985A>G mutation, most notably for those who were homozygous relative to those with ‘other’ genotypes; those who were compound heterozygous had intermediate median C8 values (Table 5). Previous studies have also found that homozygosity for the 985A>G mutation is associated with higher biomarker values for C8, particularly when compared to the compound heterozygotes^{50,51} and to other or new genotypes considered to be milder.⁶ By contrast, one study found that biochemical phenotype did not differ between those carrying the 985A>G mutation as either homozygous or compound heterozygous, unless there was the presence of the mild 199T>C mutation in which case a milder biochemical phenotype was present.¹² In our Canadian cohort, despite observing a possible association between genotype and biochemical phenotype, there was a wide range of values of C8 within each genotype category (e.g., newborn screening C8 ranges were 1.1-35 umol/L in the homozygous group and 0.7-14.8 umol/L in the ‘other’ group, Table 5), underscoring the imperfect relationship between these variables.

Limitations

A limitation of our scoping review was difficulty in identifying articles that reported actual practice/current management strategies and protocols, given the challenges in constructing a search strategy that would be sensitive and specific. As a result, we suspect that we did not capture the full range of current practice published in the literature. This concern is supported by the fact that 3/5 papers used in the review were manually identified. The articles that we did include also varied in the amount of detail provided regarding the management of MCAD deficiency and the conclusions we were able to draw were subject to the limitations of those studies. For example, many of the studies were surveys of relatively small numbers of health care providers and they reflected participants’ perceptions about their general management strategies (rather than providing direct evidence of their actual management practices). Thus, we

cannot be certain that the results would accurately reflect the treatments that are currently received by a representative sample of individuals with MCAD deficiency.

Our secondary data analysis of CIMDRN data was subject to a small sample size, although we were able to assemble a relatively large number of cases given the rarity of MCAD deficiency, through a multi-centre approach. There is a potential for bias due to non-participation. CIMDRN does have a non-participation rate of a little over 20%, which we believe is mainly due to the challenge in recruiting families of children who visit the metabolic clinic less frequently due to milder disease and fewer symptoms; thus, our study may over-estimate the use of intensive interventions. There is a small probability that children who died prior to being seen at clinic were excluded, however as NBS has resulted in a decrease in mortality, this is likely to be a very small group. One of the challenges related to the small sample was the need to create broad genotype groups. It would be helpful in future research if a larger sample size is feasible, to further stratify genotype to understand the implications of the presence/absence of potentially milder mutations. As seen in Figure 1, the sample size also decreased further when reviewing specific variables due to missing data. The validity and reliability of our data may be affected by differences across centres in extracting the data from charts, training received by abstractors and differences in reporting. However, our sensitivity analyses suggested that our main findings were robust against the assumptions we made about missing data for carnitine and fasting. Additionally, it's possible that discussions about fasting time between clinicians and families would not be fully reported in patient charts (unlike carnitine supplementation, which would be an actual prescription); therefore the results of the fasting data analysis may not fully reflect actual management practice. Finally, the imputations we were required to make for missing data, in addition to an overall simplifying approach that we used may not accurately

reflect actual treatment practice and/or intensity. No statistical analyses were conducted given the descriptive nature of this work, which limits the strength of the inferences that we can draw; however our description of patterns of care makes a unique contribution to the limited literature in this field.

Conclusions and directions for further research

Our scoping review together with our data for a Canadian sample of pediatric participants provided evidence that there is variation in clinical practice regarding fasting times and carnitine supplementation for MCAD deficiency. While the results of our scoping review confirm essentially universal treatment of MCAD deficiency by fasting avoidance, we identified high variation in fasting times by age category in both the review and cohort analysis. We suggest there is a need for further research regarding necessary fasting length to prevent acute crises while considering risks of overweight with overfeeding in different age groups. Carnitine supplementation is controversial in the management of MCAD deficiency, given that there is limited evidence for its effectiveness and emerging concerns about adverse effects. There does seem to be some evidence of a trend toward decreasing carnitine use over time, based on our secondary data analysis, but both that analysis and the scoping review indicate relatively widespread use of carnitine in the management of MCAD deficiency. With respect to markers of disease severity, we identified some evidence of an association between genotype and carnitine use from the scoping review and in the Canadian cohort, although the literature was inconsistent. This potential association may be due to a relationship between genotype and carnitine deficiency. We identified no evidence supporting an association of markers of disease severity (notably genotype and also biochemical phenotype) with different managing of fasting in MCAD deficiency in the scoping review and cohort analysis. Future research should focus on further

investigating the connection between disease severity, management, and outcomes in individuals with MCAD deficiency identified asymptotically by newborn screening. In particular, our findings emphasize that understanding the relationship between genotype, biochemical phenotype, and clinical phenotype (i.e., risk of metabolic decompensation) is important but challenging.

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Appendix B

Table B.S1. Sensitivity analysis – participants taking carnitine supplements by age category, stratified by year of birth. Imputed values which were missing were re-coded as ‘not receiving carnitine’.

Carnitine supplement by age (months) ^a	Year of birth		
	2006 to <2009	2009 to <2012	2012 to 2015
<6	8(42)	10(53)	9(24)
n (%)	n=19	n=19	n=37
6 to <12	10(53)	8(42)	11(34)
(n%)	n=19	n=19	n=32
12 to <18	9(47)	11(58)	12(38)
(n%)	n=19	n=19	n=32
18 to <24	11(58)	11(58)	13(48)
(n%)	n=19	n=19	n=27
24+	16(84)	11(58)	9(45)
(n%)	n=19	n=19	n=20

^a Data across different age categories are not independent: data were longitudinal so participants could contribute data to more than one age category

Table B.S2. Sensitivity analysis – maximum fasting time when well in each age category. Imputed values from missing data were re-coded and left as missing.

	Age categories (months)				
	<6 (n=45)	6 to <12 (n=36)	12 to <18 (n=30)	18 to <24 (n=18)	24+ (n=30)
Maximum fasting time (hours)^a	4.0 (2.5-6.5)	6.0 (3.0-12.0)	10.0 (3.5-12.0)	10.0 (6.5-12.0)	11.5 (3.5-12.0)
Median (range)					

^a Data across different age categories are not independent: data were longitudinal so participants could contribute data to more than one age category.

Table B.S3. Proportions of missing data that were imputed for carnitine and fasting time by age category.

Age category (months)	n observations of carnitine imputed/total n (n=28)*	n observations of fasting time imputed/total n (n=39)*
<6 n (%)	9/55(16)	0
6 to <12 (n%)	3/57(5)	8/42 (19)
12 to <18 (n%)	4/59(7)	9/39 (23)
18 to <24 (n%)	10/56(18)	13/31(42)
24+ (n%)	2/54(4)	9/39 (23)

*The proportions of imputed data are presented based on the total number of participants who were analyzed after imputation was done in each age category.

Supplement B.4 – Scoping review search strategy

A scoping review of existing published treatment guidelines for MCADD and VLCADD –

Dec 13, 2016

Research question

What is the current management of patients with MCADD and VLCADD?

Search filters used

- Lee E, Dobbins M, DeCorby K, McRae L, Tirilis D, Husson H. An optimal search filter for retrieving systematic reviews and meta-analyses. *BMC Medical Research Methodology* 2012, 12:51 doi:10.1186/1471-2288-12-5
- Observational studies(cohort, case-control, and case series strategy) – BMJ - <http://clinicalevidence.bmj.com/x/set/static/ebm/learn/665076.html>

MEDLINE

Database: Ovid MEDLINE(R) In-Process & Other Non-Indexed Citations and Ovid MEDLINE(R) <1946 to Present>

-
- 1 exp Acyl-CoA Dehydrogenases/ (2795)
 - 2 [MCAD.tw.](#) (631)
 - 3 [MCADD.tw.](#) (96)
 - 4 (medium-chain acyl-CoA dehydrogenase or Medium-chain acyl-coenzyme A

dehydrogenase).tw. (990)

5 (medium chain acyl CoA dehydrogenase or Medium chain acyl coenzyme A dehydrogenase).tw. (990)

6 [VLCADD.tw.](#) (43)

7 [VLCAD.tw.](#) (247)

8 (very-long-chain acyl-CoA dehydrogenase or Very long chain acyl CoA dehydrogenase).tw. (302)

9 (acyl coa dehydrogenases or acyl-coa dehydrogenases or dehydrogenases acyl-coa).tw. (299)

10 Lipid Metabolism, Inborn Errors/ (2654)

11 Fatty Acid Desaturases/ (3372)

12 fatty acid oxidation disorder\$.tw. (270)

13 (FAOD or LC-FAOD).tw. (46)

14 Mitochondrial fatty acid [oxidation.tw.](#) (640)

15 FAO defect\$.tw. (29)

16 Metabolism, Inborn Errors/ (10373)

17 or/1-16 (19078)

18 [meta-analysis.pt.](#) (81217)

19 (MEDLINE or systematic review).tw. (144131)

- 20 intervention\$.ti. (115016)
- 21 or/18-20 (293164)
- 22 17 and 21 (61)
- 23 limit 22 to (english or french) (58)
- 24 limit 23 to yr=2000-current (51)
- 25 animals/ not humans/ (4636428)
- 26 24 not 25 (50)
- 27 26 not (comment or editorial or letter).pt. (50)
- 28 exp cohort studies/ (1714267)
- 29 cohort\$.tw. (434275)
- 30 controlled clinical [trial.pt.](#) (95074)
- 31 exp case-control studies/ (876991)
- 32 (case\$ and control\$).tw. (427549)
- 33 (case\$ and series).tw. (153625)
- 34 or/28-33 (2513289)
- 35 17 and 34 (1096)
- 36 limit 35 to (english or french) (1051)

- 37 limit 36 to yr=2000-current (777)
- 38 animals/ not humans/ (4636428)
- 39 37 not 38 (753)
- 40 39 not (comment or editorial or letter).pt. (742)
- 41 27 or 40 (780)

Joanna Briggs Institute EBP Database - Current to November 30, 2016 – this source was searched but no relevant articles were identified.

EBM Reviews - Cochrane Database of Systematic Reviews <2005 to Dec 01, 2016>

EBM Reviews - Database of Abstracts of Reviews of Effects <1st Quarter 2015>

- 1 [exp Acyl-CoA Dehydrogenases/] (0)
- 2 [MCAD.tw](#). (1)
- 3 [MCADD.tw](#). (2)
- 4 (medium-chain acyl-CoA dehydrogenase or Medium-chain acyl-coenzyme A dehydrogenase).tw. (3)
- 5 (medium chain acyl CoA dehydrogenase or Medium chain acyl coenzyme A dehydrogenase).tw. (3)
- 6 [VLCADD.tw](#). (0)

- 7 [VLCAD.tw](#). (0)
- 8 (very-long-chain acyl-CoA dehydrogenase or Very long chain acyl CoA dehydrogenase).tw.
(0)
- 9 (acyl coa dehydrogenases or acyl-coa dehydrogenases or dehydrogenases acyl-coa).tw. (1)
- 10 [Lipid Metabolism, Inborn Errors/] (0)
- 11 [Fatty Acid Desaturases/] (0)
- 12 fatty acid oxidation disorder\$.tw. (0)
- 13 (FAOD or LC-FAOD).tw. (0)
- 14 Mitochondrial fatty acid [oxidation.tw](#). (0)
- 15 FAO defect\$.tw. (0)
- 16 [Metabolism, Inborn Errors/] (0)
- 17 or/1-16 (4) * note only the first part of the MEDLINE strategy was used

Embase

Database: Embase Classic+Embase <1947 to 2016 December 05>

- 1 medium chain acyl coenzyme a dehydrogenase/ (962)
- 2 long chain acyl coenzyme A dehydrogenase/ (625)
- 3 acyl coenzyme A dehydrogenase/ (1982)

- 4 fatty acid oxidation/ (17394)
- 5 acyl coenzyme A oxidase/ (1423)
- 6 [MCAD.tw](#). (859)
- 7 [MCADD.tw](#). (179)
- 8 (medium-chain acyl-CoA dehydrogenase or Medium-chain acyl-coenzyme A dehydrogenase).tw. (1061)
- 9 (medium chain acyl CoA dehydrogenase or Medium chain acyl coenzyme A dehydrogenase).tw. (1061)
- 10 [VLCADD.tw](#). (103)
- 11 [VLCAD.tw](#). (373)
- 12 (very-long-chain acyl-CoA dehydrogenase or Very long chain acyl CoA dehydrogenase).tw. (373)
- 13 (acyl coa dehydrogenases or acyl-coa dehydrogenases or dehydrogenases acyl-coa).tw. (301)
- 14 fatty acid oxidation disorder\$.tw. (393)
- 15 (FAOD or LC-FAOD).tw. (81)
- 16 FAO defect\$.tw. (47)
- 17 Mitochondrial fatty acid [oxidation.tw](#). (698)

- 18 Fatty Acid [Desaturases.tw](#). (342)
- 19 "disorders of lipid and lipoprotein metabolism"/ (1914)
- 20 inborn error of metabolism/ (12454)
- 21 or/1-20 (34921)
- 22 (MEDLINE or systematic review).tw. (166328)
- 23 intervention\$.ti. (141211)
- 24 meta analysis/ (153770)
- 25 or/22-24 (393791)
- 26 21 and 25 (184)
- 27 limit 26 to (english or french) (175)
- 28 limit 27 to yr=2000-current (167)
- 29 28 not (comment or editorial or letter).pt. (166)
- 30 exp cohort analysis/ (304087)
- 31 exp longitudinal study/ (107215)
- 32 exp prospective study/ (390183)
- 33 exp follow up/ (1257935)
- 34 cohort\$.tw. (624054)

- 35 exp case control study/ (140566)
- 36 (case\$ and control\$).tw. (588302)
- 37 exp case study/ (102730)
- 38 (case\$ and series).tw. (234164)
- 39 or/30-38 (2793119)
- 40 21 and 39 (1789)
- 41 limit 40 to (english or french) (1716)
- 42 limit 41 to yr=2000-current (1513)
- 44 28 or 42 (1650)

Food Science and Technology Abstracts <1969 to 2016 December Week 1>

- 1 exp Acyl-CoA Dehydrogenases/ (0)
- 2 [MCAD.tw.](#) (10)
- 3 [MCADD.tw.](#) (0)
- 4 (medium-chain acyl-CoA dehydrogenase or Medium-chain acyl-coenzyme A dehydrogenase).tw. (13)
- 5 (medium chain acyl CoA dehydrogenase or Medium chain acyl coenzyme A dehydrogenase).tw. (13)

- 6 [VLCADD.tw](#). (1)
- 7 [VLCAD.tw](#). (8)
- 8 (very-long-chain acyl-CoA dehydrogenase or Very long chain acyl CoA dehydrogenase).tw.
(10)
- 9 (acyl coa dehydrogenases or acyl-coa dehydrogenases or dehydrogenases acyl-coa).tw. (3)
- 10 Lipid Metabolism, Inborn Errors/ (0)
- 11 Fatty Acid Desaturases/ (0)
- 12 fatty acid oxidation disorder\$.tw. (2)
- 13 (FAOD or LC-FAOD).tw. (3)
- 14 Mitochondrial fatty acid [oxidation.tw](#). (15)
- 15 FAO defect\$.tw. (0)
- 16 Metabolism, Inborn Errors/ (0)
- 17 or/1-16 (44)
- 18 limit 17 to (english or french) (44)
- 19 limit 18 to yr="2000 -Current" (42) * note only the first part of the MEDLINE strategy was used

Database: PsycINFO <1806 to November Week 4 2016>

- 1 exp Acyl-CoA Dehydrogenases/ (0)
- 2 [MCAD.tw](#). (16)
- 3 [MCADD.tw](#). (6)
- 4 (medium-chain acyl-CoA dehydrogenase or Medium-chain acyl-coenzyme A dehydrogenase).tw. (10)
- 5 (medium chain acyl CoA dehydrogenase or Medium chain acyl coenzyme A dehydrogenase).tw. (10)
- 6 [VLCADD.tw](#). (0)
- 7 [VLCAD.tw](#). (4)
- 8 (very-long-chain acyl-CoA dehydrogenase or Very long chain acyl CoA dehydrogenase).tw. (3)
- 9 (acyl coa dehydrogenases or acyl-coa dehydrogenases or dehydrogenases acyl-coa).tw. (1)
- 10 Lipid Metabolism, Inborn Errors/ (0)
- 11 Fatty Acid Desaturases/ (0)
- 12 fatty acid oxidation disorder\$.tw. (8)
- 13 (FAOD or LC-FAOD).tw. (1)
- 14 Mitochondrial fatty acid [oxidation.tw](#). (11)
- 15 FAO defect\$.tw. (0)

- 16 Metabolism, Inborn Errors/ (0)
- 17 or/1-16 (40)
- 18 limit 17 to (english or french) (40)
- 19 limit 18 to yr="2000 -Current" (39)
- 20 limit 19 to human (31) * note only the first part of the MEDLINE strategy was used

Chapter 4 – Integrated Discussion

The focus of this thesis was on diagnostic and management practices in children with medium-chain acyl-CoA dehydrogenase (MCAD) and very long-chain acyl-CoA dehydrogenase (VLCAD) deficiencies, two fatty acid oxidation disorders (FAOD) that are commonly included in newborn screening programs. The thesis has two major components: (i) a systematic review of the quality and content of published/unpublished practice guidance related to the diagnosis and/or management of both disorders; and (ii) a scoping review of the published literature and secondary analysis of data from a Canadian cohort regarding care practices specific to MCAD deficiency. A large part of the thesis focuses on MCAD deficiency, given that it is the most common FAOD, however the systematic review of guidance also considered VLCAD deficiency, a defect of fatty acid metabolism that is rarer than MCAD deficiency and frequently accompanied by more severe complications. Throughout this thesis, we were particularly interested in guidance and practice regarding two areas of care that we had identified as controversial, carnitine supplementation, and duration of fasting, examining these in context of disease severity where possible.

Best practice: Systematic review of guidance

To our knowledge, ours is the first systematic attempt to characterize best practice in terms of the guidance available to care practitioners managing MCAD and VLCAD deficiencies. Our systematic review identified 25 guidance documents from both peer-reviewed and grey literature. We evaluated the quality of the guidance using the AGREE II tool, finding that although the guidance documents we reviewed generally received high scores for the AGREE II domains related to clarity of presentation and purpose, scores related to the rigor of guideline development were relatively low by comparison. This indicates that guidance in this field does

not generally meet established methodological standards for developing clinical practice guidelines. We classified the methods used to develop recommendations, finding that only 7 of the 25 guidance documents were explicitly reported to be based wholly or partially on an evidence review. Guidelines often identified that there was limited empirical evidence on which to construct recommendations.

From a content perspective, while guidance documents addressed most key aspects of management of MCAD and VLCAD deficiencies, the detail they provided was variable. We identified inconsistencies in some recommendations, particularly regarding long-term management and some aspects of acute care. With respect to our interventions of *a priori* interest, fasting avoidance was consistently identified as the most important intervention to prevent acute crises, but there was important variation in the specific fasting schedules recommended when these were specified in the guidance documents. Most guidance documents discussed carnitine supplementation. No guidance authors recommended routine carnitine supplementation for all individuals with MCAD or VLCAD deficiency, but there was variation in the recommendations about the circumstances under which carnitine supplementation is appropriate. Authors of guidance documents often acknowledged the lack of evidence and many were vague about criteria for supplementation.

Current practice: Scoping review and analysis of Canadian cohort data

The second part of this project was a scoping review of the literature describing current practice for the management of MCAD deficiency and a secondary analysis of clinical data from a Canadian cohort study of children with MCAD deficiency. The scoping review yielded a limited number of published studies of current management practice. We identified five studies: a US⁷¹ survey of metabolic dietitians, an international survey of clinics specific to carnitine

practice,²⁹ a Canadian survey of metabolic physicians,⁶⁵ a monitoring protocol used for Danish patients with MCAD deficiency,⁷² and a study reporting on the regional experience in Southwest Europe clinics, involving a case series of patients with MCAD deficiency.⁷³

Carnitine supplementation was reported in all but one of the studies in the scoping review, with evident variation in carnitine use. While 79% of US dietitians surveyed in one study reported prescribing carnitine,⁷¹ only 33% of Canadian physicians from a second study indicated that they would recommend carnitine for ‘all or most’ patients while a further 33% would recommend carnitine ‘in rare cases’ or ‘never’.⁶⁵ Similarly, of clinics responding to an international survey, 39% reported ‘always’ prescribing carnitine.^{29,65} We identified some evidence of variation in carnitine use based on age, on the basis of an identified carnitine deficiency, or for acute illness.^{29,65,73} With respect to fasting, most studies reported fasting avoidance as a foundation of care, operationalized as increased meal frequency. However, only one broad fasting schedule was reported.⁷¹

There was some evidence from the scoping review of managing patients differently on the basis of disease severity, but this was inconsistent. In one study, 33% of surveyed Canadian physicians reported genotype as ‘essential/very important’ as an influence on chronic care, while biochemical phenotype (the plasma acylcarnitine profile) was used by 22%.⁶⁵ The Danish monitoring protocol specifically reported not taking disease severity into consideration for treatment determination given that the definition was not clear and there was no consensus on this.⁷² Among patients receiving care in clinics in southwest Europe, 93% of those who were homozygous for the ‘classic’ mutation required carnitine to reach normal levels based on carnitine deficiency.⁷³

Our secondary data analysis included 107 children with MCAD deficiency participating in the Canadian cohort. Again, we were particularly interested in receipt (or not) of carnitine supplementation and children's fasting times; and in the potential association of these interventions with markers that may predict disease severity, including genotype and biochemical phenotype. In analyzing carnitine use among cohort study participants, there was an increasing trend of carnitine use by age, such that a higher proportion of older children were receiving carnitine than younger children, but this trend was less apparent after accounting for calendar time trends. We identified a decreasing use of carnitine over calendar time: a smaller proportion of children born in more recent years (2012-2015) were receiving carnitine relative to those born earlier (2006-2009), across all age groups. With respect to fasting time in the Canadian cohort, we saw increasing fasting duration with increasing age, as expected.

With respect to proxy indicators of disease severity, in our cohort analysis, we noted an association between genotype and biochemical phenotype, corresponding to that previously identified in the literature,^{20,32} where both newborn screening and diagnostic C8 values decreased with decreasing disease severity based on the common c.985A>G mutation (homozygous>compound heterozygous>other). In terms of the association between these possible markers of disease severity and management of MCAD deficiency, we found that children who received carnitine had higher NBS and diagnostic C8 levels than those who did not receive carnitine. We also found that a higher proportion of children with the homozygous genotype, often associated with more severe disease, received carnitine compared to those with other genotypes. We did not identify an association between genotype as a proxy indicator of disease severity and fasting time in our cohort.

In comparing reported practice in the scoping review literature with treatments received by Canadian children with respect to carnitine supplementation, it may be particularly relevant to consider the survey of Canadian physicians.⁶⁵ While 33% of participants in that study reported recommending carnitine for all or most patients, in our cohort analysis, 49% of infants <6 months old were on carnitine, with the proportion increasing to 67% at 2 or more years. A limitation of the cohort study was that we were unable to consider the role of carnitine deficiency as a potential influence on supplementation practice, whereas several of the scoping review studies indicated that deficiency was frequently the basis for receipt of carnitine supplements.^{29,73} There were limited specific fasting-time data from the scoping review, with the exception of the US study of dietitians.⁷¹ In comparing the findings of that study with the Canadian cohort, children aged <1 year of age had similar fasting times. However, the fasting times of children >1 year old (and adults) in the US survey of dietitians were half the median fasting time of those of similar-aged children in the Canadian cohort. The Canadian children were fasting a median of over 11 hours by age 2+, while adults in the US were reported to be recommended to fast for only 5 hours on average. The range of fasting time was very large in both the US study and Canadian cohort, reflecting the variation in individual management.

Comparison of best practice and actual practice: The evidence-practice gap and discussion in the context of the broader literature

In this section we integrate the findings from Chapters 2 and 3 to compare best practice in the management of MCAD deficiency with current practice (the evidence-practice gap) and we discuss the integrated findings in the context of the broader related literature.

With respect to the evidence-practice gap in the area of carnitine supplementation for MCAD deficiency, we found that actual practice in carnitine use aligned with published

guidance in a very broad sense: no guidance documents in our systematic review recommended supplementing carnitine for all patients, there was little evidence from the scoping review that all children receive carnitine in the jurisdictions covered by the reviewed studies, and carnitine supplementation in the Canadian cohort was not universal at any specific age or calendar time point. Many guidance documents provided only general recommendations without specific guidance about the appropriate circumstances for supplementation,^{28,58,74–76} while others recommended carnitine when plasma carnitine was low.^{25,27,33,35} From the scoping review of actual practice, carnitine was similarly reported in some papers to be used when there was an identified carnitine deficiency, but additionally there were some reports that noted that carnitine supplementation decisions were based on age, or for acute illness, and some indicated never using carnitine.^{29,65,71,73} In the Canadian cohort, we were unable to identify the specific rationale for carnitine supplementation although there was variation by age and the variation we identified by genotype may have been related to a higher prevalence of carnitine deficiency in children with a more severe genotype.

In terms of the broader literature, low carnitine levels are often described in patients with FAOD, including both MCAD and VLCAD deficiency.^{27,67} Based on evidence of secondary deficiency of carnitine in patients with IMD, treatment with carnitine supplementation was approved in the United States in 1992.⁷⁷ A previous summary of 16 NBS programs from around the world revealed 6 programs (Brussels, Erlangen, Japan, Porto, Switzerland) that reported carnitine supplementation for their patients at follow-up and 2 (in Oregon and Italy) which recommended carnitine supplements only if low carnitine was observed.⁷⁸ Given mixed results regarding the potential for better outcomes^{65,74} and concerns about the side effects for carnitine,^{31,70,74,79} it is important to determine indications for prescription.

With respect to the evidence-practice gap related to fasting, we identified numerous fasting-related recommendations in our systematic review of guidance for management of MCAD deficiency. Specifically, all guidance documents making recommendations for the long-term management of MCAD deficiency (15/15) identified fasting avoidance as a key treatment and 9 of these guidance documents provided some kind of fasting schedules, but with variable detail. These included on demand feeds,⁸⁰ feeding about every 4 hours,^{27,80,81} and then adding 1 hour for every month starting at 5 months,²⁷ schedules resulting in feeding at <10 hours up to 1 year of age, and increasing to a full overnight fast (<12 hours) after 1 year (or near 2 years).^{26,28,68} This was in line with reporting of actual practice from the scoping review, where fasting avoidance was described as an important treatment by most clinics/physicians reporting on their practice, and only 1 schedule was described.^{65,71-73} Some guidance documents also specifically recommended spreading meals during the day to address energy requirements and prevent fasting while avoiding excessive weight gain by means of introducing extra meals. Similarly, from the scoping review, avoidance of fasting and increased meal frequency were recommended for all or most patients by 94 and 44% of Canadian physicians, respectively, and was reported to be used by 95% of US dietitians.^{65,71} Fasting times among children in the Canadian cohort were also generally aligned with recommended practice: median fasting times in the cohort generally fell below the recommended fasting limits in the guidance documents and were closely matched to the only evidence-based fasting limits, from Derks et al., 2007.²⁶ We did identify a small number of children with unusually short fasting durations in our Canadian cohort.

In the context of the broader literature regarding MCAD deficiency and fasting avoidance, there is recognition of adherence to frequent feedings as important for avoiding

metabolic decompensation, particularly in early life. However, recently some authors have expressed concern about the risk of over-feeding and later life obesity.^{8,26-28} Indeed a number of the guidance documents we reviewed addressed monitoring for weight gain and obesity and avoiding over-feeding in recognition of this risk.^{8,26-28,67,82} One study in Portugal indicated a high prevalence (nearly 50%) of overweight/obesity in children with FAOD, which was significantly higher than the prevalence in the general pediatric population.⁸³ Some research suggests that participants with milder diseases, specifically those with higher residual enzyme activity, might not require maximum fasting time regimens at all.⁴⁹ Some authors, however, suggest that because the association between genotype and phenotype is not clearly defined, it may be better to maintain uniform advice for all diagnosed patients, especially given that there is evidence of no difference in outcomes when such advice is given to all patients independent of genotype.⁸⁴

Suggestions for addressing the evidence-practice gap

Future guidance to support the diagnosis and management of MCAD and VLCAD deficiencies can benefit by conforming to established standards for the development of clinical practice guidelines and by using tools such as the AGREEII to ensure the production of rigorous recommendations. In particular, it is challenging to assess the effectiveness of guideline recommendations if their methods of development are unclear or if sufficient details are not provided. With respect to content, it may be worthwhile to address areas of uncertainty that have been prioritized by patients and families, clinicians, and health researchers, including questions regarding the potential to tailor treatment to predicted disease severity and an emphasis on controversial therapies such as carnitine supplementation. Finally, a patient-partnered approach

to guideline development may be beneficial, given that outcomes so highly depend on patients' and parents' understanding and confidence in managing these disorders.

Reports from our scoping review were frequently lacking in detail regarding treatment strategies and generally did not identify the rationale underlying clinical practice. Among other reasons that may partially explain why there is limited available published evidence documenting actual practice, clinics and providers may be relying on their individual accumulated experiences in managing their patients to guide treatment decisions, but they may not have sought to publish such experiences. The evidence base regarding practice needs to be developed so that practice can be more easily presented in context of current guidance and, importantly, so that treatments can be evaluated in order to understand which management strategies may result in better outcomes. In considering carnitine supplementation, understanding the role of carnitine deficiency and determining the influence of genotype and biochemical phenotype on carnitine levels will help to move the evidence forward. Intervention research is also needed to clarify both the benefits and risks of carnitine supplementation in patients with MCAD deficiency with and without carnitine deficiency, with a focus on clinical and patient-reported endpoints. For informing practice on fasting, it could be helpful to describe cases with inappropriate shorter fasting times to determine risk factors, but also to determine under what circumstances children are most vulnerable to acute decompensation, and the association of their clinical phenotype to genotype.

Limitations

Our systematic review of guidance for the diagnosis and management of MCAD and VLCAD deficiencies incorporated many documents that did not self-identify as guidelines. This may in part explain the absence of comprehensive reporting of the methods for development of

guidance in many cases. However, we argue that evaluating the quality of guidance against the AGREE II tool was appropriate given that all of the reviewed documents provided recommendations for disease diagnosis and/or management. A limitation in our quality assessment of guidance was that only one reviewer performed the scoring against the AGREE II tool; a second reviewer verified but did not independently score the guidance and thus we were not able to report on inter-rater reliability.

One of the main limitations of the scoping review was the challenge in comprehensively identifying publications reporting on actual practice in the diagnosis and management of MCAD deficiency, due to the non-specific nature of the indexing of such publications. Given that we identified only five publications for that review, of which three were manually identified and not picked up by the search strategy, we may not have comprehensively captured the full range of current practices.

Regarding our secondary analysis of data from a Canadian cohort, missing data and small counts placed some limits on the analyses we performed. Sensitivity analyses did suggest that the findings were robust against the assumptions we made in imputing some information for carnitine and fasting time where data were missing. The genotype categories that we used to investigate care in association with disease severity for MCAD deficiency were based on the presence of the classic c.985A>G mutation (homozygous, compound heterozygous, other) due to sample size constraints. Ideally, other genotypes, e.g., presence of a common mutation that may be mild (c.199T>C) would be investigated. However, by pooling together across most Canadian centres where children with MCAD deficiency receive care, we were able to add important insights about the relationship between genotype and care received.

Conclusions

This project evaluated best practice in terms of current guidance for the management of MCAD and VLCAD deficiencies, identifying the need for improved methodological rigour in developing such guidance and summarizing their content, in particular with respect to areas of controversy (fasting times, carnitine supplementation, management based on disease severity). We described actual practice in these same areas of care, internationally (the scoping review) and in Canada (secondary analysis of cohort data) and compared this practice with the guidance to identify evidence-practice gaps. Given a growing body of evidence of variation in disease severity according to genotype and the absence of guidance for managing MCAD deficiency based on severity, it is crucial that future research focuses on evaluating care in association with key outcomes, with consideration of whether and how management may be adjusted in the context of predictors of disease severity.

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Appendix C

Table C1. Assessment of learning outcomes

Program Learning Outcomes for Graduate Programs in Epidemiology, April 10, 2017

Self-assessment

Attribute	Learning Outcomes	
	Master's Program	
	Graduating students will be able to:	Self-assessment
Application of knowledge	<ol style="list-style-type: none"> 1. Evaluate and critically appraise the strengths and limitations of an epidemiologic study from its design to the conclusions drawn. 2. Critically and systematically synthesize existing knowledge about a health topic. 3. Proficiently use statistical software to analyze epidemiologic data. 	<p>In conducting a systematic review for my thesis, I reviewed and identified literature and appraised the quality of the guidelines using the AGREEii tool, which allowed me to identify strengths and weaknesses of the guidelines, the evidence on which they were based and consequently gaps in the literature which limited the development of high quality guidelines. I synthesized the information to produce summaries of conflicting and consistent recommendations.</p> <p>I cleaned and analyzed the data, describing the current practice in a pediatric cohort, writing and researching code that would allow me to organize my data and provide descriptive results on management.</p>
Engaged scholarship, knowledge translation, and communication	<ol style="list-style-type: none"> 4. Define the core components of engaged scholarship 5. Describe best practices for facilitating uptake of evidence into policy, programs, patient care, and/or 	<p>I frequently attended lectures and rounds to learn from our researchers and see how epidemiologic methods are applied across different fields, while also reflecting on how these methods could be used in my own research.</p> <p>Appraising guidelines allowed me to</p>

	<p>the design or analysis of future research.</p> <p>6. Prepare a clearly written and logically argued report from epidemiologic analyses.</p> <p>7. Effectively disseminate and communicate epidemiologic evidence to academic and non-academic audiences in written and oral formats.</p> <p>8. Describe the implications of epidemiologic evidence for policy, programs, patient care, and/or future research.</p>	<p>identify gaps in the literature and reflect on the barriers to adopting a ‘universal’ guideline for the management of FAOD. It also helped me identify opportunities for involving patients in future research, while addressing their interests and concerns.</p> <p>This thesis allows an intuitive comparison of guidelines to reported practice to actual practice and results of this research were disseminated at various stages of the project (early and mid-phase of completion), presenting to various audiences including researchers, clinicians, genetic counselors, dietitians, primary health care practitioners and students.</p>
<p>Autonomy, professional capacity, leadership and mentorship</p>	<p>9. Identify and adhere to ethical principles relevant to epidemiologic research that apply locally, nationally, and internationally, including the Tri-Council Policy Statement: Ethical Conduct for Research Involving Humans (TCPS).</p> <p>10. Adhere to principles of academic integrity according to the policies of the University of Ottawa.</p> <p>11. Appreciate the value of and participate in an interdisciplinary research community in their chosen field of health research.</p>	<p>In analyzing such a small cohort, it was important to always be aware that it was highly sensitive data, despite wanting to present so much more information that would be clinically interesting.</p> <p>It was important to consult with my TAC and other team members and colleagues to clearly understand the meaning of the results and to explore options for conducting my research.</p> <p>I shared my results and learned from other researchers at both international and national conferences.</p>

	12. Commit to proactively seeking new knowledge and being aware of current issues of debate in their chosen field of health research.	
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C2. AGREEII tool

**APPRAISAL OF GUIDELINES
FOR RESEARCH & EVALUATION II**



AGREE II

INSTRUMENT

The AGREE Next Steps Consortium
May 2009

UPDATE: December 2017

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DISCLAIMER

The AGREE II Instrument is a generic tool designed primarily to help guideline developers and users assess the methodological quality of guidelines. The authors do not take responsibility for the improper use of the AGREE II Instrument.

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FOR FURTHER INFORMATION ABOUT THE AGREE II, PLEASE CONTACT
AGREE Project Office, agree@mcmaster.ca
AGREE Website, www.agreetrust.org

AGREE NEXT STEPS CONSORTIUM MEMBERSHIP

Dr. Melissa C. Brouwers
Principal Investigator, AGREE Next Steps Consortium
McMaster University, Hamilton, Ontario, Canada

Consortium Members:

Dr. G.P. Browman, Victoria, British Columbia, Canada
Dr. J.S. Burgers, Dutch College of General Practitioners, Utrecht, The Netherlands
Dr. F. Cluzeau, Global Health and Development Group, Imperial College London, UK
Dr. D. Davis, Association of American Medical Colleges, Washington DC, USA
Dr. G. Feder, University of Bristol, UK
Dr. B. Fervers, Cancer et Environnement, Centre Léon Bérard, France
Dr. I. Graham, Ottawa Hospital Research Institute, Ottawa, Ontario, Canada
Dr. J. Grimshaw, Ottawa Hospital Research Institute, Ottawa, Ontario, Canada
Dr. S.E. Hanna, McMaster University, Hamilton, Ontario, Canada
Ms. M.E. Kho, McMaster University, Hamilton, Ontario, Canada
Dr. P. Littlejohns, Kings College London, UK
Ms. J. Makarski, Hamilton, Ontario, Canada
Dr. L. Zitzelsberger, Quebec, Canada

AGREE II VERSIONS & UPDATES

AGREE II Original Public Release and Publication Date: 2009/2010

AGREE II Update: September 2013

AGREE II Update: December 2017

What's new in the December 2017 update?

The August 2017 update includes revisions to the following sections of the Introduction: 'AGREE Website: Resources and References', '10 Years of AGREE', and 'Scoring the AGREE II'. Guidance has been added regarding the use of AGREE II score thresholds to distinguish between higher and lower quality guidelines. In addition, minor editorial changes have been made throughout the Introduction and User's Manual. The content of the AGREE II Instrument itself has not been modified since 2009 and all versions of the AGREE II remain valid for use.

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I. INTRODUCTION

I. OVERVIEW

i) Purpose of the AGREE II Instrument

Clinical practice guidelines ('guidelines') are systematically developed statements to assist practitioner and patient decisions about appropriate health care for specific clinical circumstances (1). In addition, guidelines can play an important role in health policy formation (2,3) and have evolved to cover topics across the health care continuum (e.g., health promotion, screening, diagnosis).

The potential benefits of guidelines are only as good as the quality of the guidelines themselves. Appropriate methodologies and rigorous strategies in the guideline development process are important for the successful implementation of the resulting recommendations (4-6). The quality of guidelines can be extremely variable and some often fall short of basic standards (7-9).

The *Appraisal of Guidelines for REsearch & Evaluation (AGREE) Instrument* (10) was developed to address the issue of variability in guideline quality. To that end, the AGREE instrument is a tool that assesses the methodological rigour and transparency in which a guideline is developed. The original AGREE instrument was refined, which resulted in the AGREE II, and a User's Manual was developed (11-13).

The purpose of the AGREE II, is to provide a framework to:

1. Assess the quality of guidelines;
2. Provide a methodological strategy for the development of guidelines; and
3. Inform what information and how information ought to be reported in guidelines.

The AGREE II replaces the original instrument as the preferred tool and can be used as part of an overall quality mandate aimed to improve health care.

ii) History of the AGREE Project

The original AGREE Instrument was published in 2003 by a group of international guideline developers and researchers, the AGREE Collaboration (10). The objective of the Collaboration was to develop a tool to assess the quality of guidelines. The AGREE Collaboration defined quality of guidelines as *the confidence that the potential biases of guideline development have been addressed adequately and that the recommendations are both internally and externally valid, and are feasible for practice* (10). The assessment includes judgments about the methods used for developing the guidelines, the components of the final recommendations, and the factors that are linked to their uptake. The result of the Collaboration's effort was the original AGREE Instrument, a 23-item tool comprising 6 quality domains. The AGREE Instrument has been translated into many languages, has been cited in over 600 publications, and is endorsed by several health care organizations. More details about the original instrument and related publications are available on the AGREE Website (<http://www.agreetrust.org>).

As with any new assessment tool, it was recognized that ongoing development was required to strengthen the measurement properties of the instrument and to ensure its usability and feasibility among intended users. This led several members of the original team to form the

AGREE Next Steps Consortium (Consortium). The objectives of the Consortium were to further improve the measurement properties of the instrument, including its reliability and validity; to refine the instrument's items to better meet the needs of the intended users; and to improve the supporting documentation (i.e., original training manual and user's guide) to facilitate the ability of users to implement the instrument with confidence.

The result of these efforts is the AGREE II, which is comprised of the new User's Manual and 23 item tool organized into the same six domains, described here. The User's Manual is a significant modification of the original training manual and user's guide and provides explicit information for each of the 23 items. Table 1 compares the items of the original AGREE to the items in the AGREE II.

Table 1. Comparison of the original AGREE and AGREE II items.

Original AGREE Item	AGREE II Item
Domain 1. Scope and Purpose	
1. The overall objective(s) of the guideline is (are) specifically described.	No change
2. The clinical question(s) covered by the guideline is (are) specifically described.	The health question(s) covered by the guideline is (are) specifically described.
3. The patients to whom the guideline is meant to apply are specifically described.	The population (patients, public, etc.) to whom the guideline is meant to apply is specifically described.
Domain 2. Stakeholder Involvement	
4. The guideline development group includes individuals from all the relevant professional groups.	No change
5. The patients' views and preferences have been sought.	The views and preferences of the target population (patients, public, etc.) have been sought.
6. The target users of the guideline are clearly defined.	No change
7. The guideline has been piloted among end users.	Delete item. Incorporated into user guide description of item 19.
Domain 3. Rigour of Development	
8. Systematic methods were used to search for evidence.	No change in item. Renumbered to 7.
9. The criteria for selecting the evidence are clearly described.	No change in item. Renumbered to 8.
	NEW Item 9. The strengths and limitations of the body of evidence are clearly described.
10. The methods for formulating the recommendations are clearly described.	No change
11. The health benefits, side effects, and risks have been considered in formulating the recommendations.	No change
12. There is an explicit link between the recommendations and the supporting evidence.	No change
13. The guideline has been externally reviewed by experts	No change

Original AGREE Item	AGREE II Item
prior to its publication.	
14. A procedure for updating the guideline is provided.	No change
Domain 4. Clarity of Presentation	
15. The recommendations are specific and unambiguous.	No change
16. The different options for management of the condition are clearly presented.	The different options for management of the condition or health issue are clearly presented.
17. Key recommendations are easily identifiable.	No change
Domain 5. Applicability	
18. The guideline is supported with tools for application.	The guideline provides advice and/or tools on how the recommendations can be put into practice. AND Change in domain (from Clarity of Presentation) AND renumbered to 19
19. The potential organizational barriers in applying the recommendations have been discussed.	The guideline describes facilitators and barriers to its application. AND change in order – renumbered to 18
20. The potential cost implications of applying the recommendations have been considered.	The potential resource implications of applying the recommendations have been considered.
21. The guideline presents key review criteria for monitoring and/ or audit purposes.	The guideline presents monitoring and/ or auditing criteria.
Domain 6. Editorial Independence	
22. The guideline is editorially independent from the funding body.	The views of the funding body have not influenced the content of the guideline.
23. Conflicts of interest of guideline development members have been recorded.	Competing interests of guideline development group members have been recorded and addressed.

II. APPLYING THE AGREE II

i) Which guidelines can be appraised with the AGREE II?

As with the original instrument, the AGREE II is designed to assess guidelines developed by local, regional, national or international groups or affiliated governmental organizations. These include original versions of and updates of existing guidelines.

The AGREE II is generic and can be applied to guidelines in any health or disease area targeting any step in the health care continuum, including those for health promotion, public health, screening, diagnosis, treatment or interventions. It is suitable for guidelines presented in paper or electronic format. The AGREE II has not been designed to assess the quality of guidance documents that address health care organizational issues. Its role in the assessment of health technology assessments has not been formally evaluated.

ii) Who can use the AGREE II?

The AGREE II is intended to be used by the following stakeholder groups:

- by **health care providers** who wish to undertake their own assessment of a guideline before adopting its recommendations into their practice;
- by **guideline developers** to follow a structured and rigorous development methodology, to conduct an internal assessment to ensure that their guidelines are sound, or to evaluate guidelines from other groups for potential adaptation to their own context;
- by **policy makers** to help them decide which guidelines could be recommended for use in practice or to inform policy decisions; and
- by **educators** to help enhance critical appraisal skills amongst health professionals and to teach core competencies in guideline development and reporting.

III. AGREE WEBSITE: RESOURCES AND REFERENCES

The AGREE Enterprise website, www.agreetrust.org, contains a variety of tools to assist users in applying the AGREE II.

i) Publications of AGREE Research

- Access publications related to the AGREE II and other AGREE tools.
- The **Key publications: AGREE II** page provides access to publications related to the development and testing of the AGREE II.

ii) AGREE II Training Tools

- Two online tools are available to train new users of the AGREE II:
 - AGREE II Overview Tutorial,
 - AGREE II Practice Exercise.

iii) AGREE II Language Translations

- The AGREE II has been translated into various languages, thanks to members of the international practice guideline community.
- Copies of these translations are available to the public on this webpage.
- If you would like to undertake a new translation, please contact the AGREE Project Office by emailing agree@mcmaster.ca.

iv) My AGREE PLUS

- An online platform called My AGREE PLUS is freely available to the public to complete and track AGREE II appraisals.
- The platform can be used to:
 - Complete individual AGREE II appraisals,
 - Contribute to a group AGREE II appraisal, and
 - Coordinate a group AGREE II appraisal.
- Click the **My AGREE PLUS** tab at www.agreetrust.org to register and use the platform.

v) Other AGREE tools

- Access other AGREE tools to support the development, reporting and appraisal of clinical practice guidelines and health systems guidance:
 - **AGREE Reporting Checklist:** A checklist based on the AGREE II to guide the reporting of clinical practice guidelines (14).
 - **AGREE GRS:** A 4-item tool to assess the quality of clinical practice guidelines when scarce time or resources make it unfeasible to use the more comprehensive AGREE II.
 - **AGREE Recommendations Excellence (AGREE-REX):** A tool to assess the quality and direct the development and reporting of clinical practice guideline recommendations.
 - **AGREE Health Systems (AGREE-HS):** A tool to assess the quality and direct the development and reporting of health systems guidance documents.
 - **CheckUp:** A checklist to guide the reporting of updated clinical practice guidelines (15).

IV. 10 Years of AGREE

In 2013, the AGREE Enterprise marked its 10th anniversary since the original AGREE Instrument was first published and made available for use. To mark this anniversary, an article was published summarizing the academic journey the AGREE instrument has taken, noting the many accomplishments along the way (16).

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**AGREE II:
USER'S MANUAL**

II. USER'S MANUAL: INSTRUCTIONS FOR USING THE AGREE II

This User's Manual has been designed specifically to guide appraisers in the use of the instrument. We suggest reading the following instructions before using the instrument.

I. Preparing to Use the AGREE II

i) Accompanying Guideline Documents

Before applying the AGREE II, users should first carefully read the guideline document in full. In addition to the guideline document, users should attempt to identify all information about the guideline development process prior to the appraisal. This information may be contained in the same document as the guideline recommendations or it may be summarized in a separate technical report, methodological manual or guideline developer policy statement. These supporting documents may be published or may be available publicly on web sites. While it is the responsibility of the guideline authors to advise readers on the existence and location of relevant additional technical and supporting documents, every effort should be made by the AGREE II users to locate and include them as part of the materials appropriate for assessment.

ii) Number of Appraisers

We recommend that each guideline be assessed by at least 2 appraisers, and preferably 4, as this will increase the reliability of the assessment.

II. Structure and Content of the AGREE II

The AGREE II consists of 23 key items organized within 6 domains followed by 2 global rating items ("Overall Assessment"). Each domain captures a unique dimension of guideline quality.

Domain 1. Scope and Purpose is concerned with the overall aim of the guideline, the specific health questions, and the target population (items 1-3).

Domain 2. Stakeholder Involvement focuses on the extent to which the guideline was developed by the appropriate stakeholders and represents the views of its intended users (items 4-6).

Domain 3. Rigour of Development relates to the process used to gather and synthesize the evidence, the methods to formulate the recommendations, and to update them (items 7-14).

Domain 4. Clarity of Presentation deals with the language, structure, and format of the guideline (items 15-17).

Domain 5. Applicability pertains to the likely barriers and facilitators to implementation, strategies to improve uptake, and resource implications of applying the guideline (items 18-21).

Domain 6. Editorial Independence is concerned with the formulation of recommendations not being unduly biased with competing interests (items 22-23).

Overall assessment includes the rating of the overall quality of the guideline and whether the guideline would be recommended for use in practice.

III. Rating Scale and User's Manual Sections

Each of the AGREE II items and the two global rating items are rated on a 7-point scale (1–strongly disagree to 7–strongly agree). The User's Manual provides guidance on how to rate each item using the rating scale and also includes 3 additional sections to further facilitate the user's assessment. The sections include User's Manual Description, Where to Look, and How to Rate.

i) Rating Scale

All AGREE II items are rated on the following 7-point scale:

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Score of 1 (*Strongly Disagree*). A score of 1 should be given when there is no information that is relevant to the AGREE II item, if the concept is very poorly reported, or if the authors state explicitly that criteria were not met.

Score of 7 (*Strongly Agree*). A score of 7 should be given if the quality of reporting is exceptional and where the full criteria and considerations articulated in the User's Manual have been met.

Scores between 2 and 6. A score between 2 and 6 is assigned when the reporting of the AGREE II item does not meet the full criteria or considerations. A score is assigned depending on the completeness and quality of reporting. Scores increase as more criteria are met and considerations addressed. The "How to Rate" section for each item includes details about assessment criteria and considerations specific to the item.

ii) User's Manual Description

This section defines the concept underlying the item in broad terms and provides examples.

iii) Where to Look

This section directs the appraiser to where the information in the guideline can usually be found. Included in this section are common terms used to label guideline sections or chapters. *These are suggestions only*. It is the responsibility of the appraiser to review the entire guideline and accompanying material(s) to ensure a fair evaluation.

iv) How to Rate

This section includes details about assessment criteria and considerations specific to each item.

- The *criteria* identify explicit elements that reflect the operational definition of the item. The more criteria that are met, the higher the score the guideline should receive on that item.
- The *considerations* are aimed to help inform the assessment. As in any evaluation, judgments by the appraisers are required. The more the considerations have been taken into account in the guideline, the higher the score the guideline should receive on that item.

It is important to note that guideline ratings require a level of judgment. The criteria and considerations are there to guide, not to replace, these judgments. Thus, none of the AGREE II items provide explicit expectations for each of the 7 points on the scale.

v) Other Considerations when Applying the AGREE II

On occasion, some AGREE II items may not be applicable to the particular guideline under review. For example, guidelines narrow in scope may not provide the full range of options for the management of the condition (see item 16). The AGREE II does not include a "Not Applicable" response item in its scale. There are different strategies to manage this situation including having appraisers skip that item in the assessment process or rating the item as 1 (absence of information) and providing context about the score. *Regardless of strategy chosen, decisions should be made in advance, described in an explicit manner, and if items are skipped, appropriate modifications to calculating the domain scores should be implemented. As a principle, excluding items in the appraisal process is discouraged.*

IV. Scoring the AGREE II

A quality score is calculated for each of the six AGREE II domains. The six domain scores are independent and should not be aggregated into a single quality score.

i) Calculating Domain Scores

Domain scores are calculated by summing up all the scores of the individual items in a domain and by scaling the total as a percentage of the maximum possible score for that domain.

Example:

If 4 appraisers give the following scores for Domain 1 (Scope & Purpose):

	Item 1	Item 2	Item 3	Total
Appraiser 1	5	6	6	17
Appraiser 2	6	6	7	19
Appraiser 3	2	4	3	9
Appraiser 4	3	3	2	8
Total	16	19	18	53

Maximum possible score = 7 (strongly agree) x 3 (items) x 4 (appraisers) = 84
Minimum possible score = 1 (strongly disagree) x 3 (items) x 4 (appraisers) = 12

The scaled domain score will be:

$$\frac{\text{Obtained score} - \text{Minimum possible score}}{\text{Maximum possible score} - \text{Minimum possible score}}$$
$$\frac{53 - 12}{84 - 12} \times 100 = \frac{41}{72} \times 100 = 0.5694 \times 100 = 57 \%$$

If items are not included, appropriate modifications to the calculations of maximum and minimum possible scores are required.

ii) Interpreting Domain Scores

Domain scores can be used to identify strengths and limitations of guidelines, to compare methodological quality between guidelines, or to select high quality guidelines for adaptation, endorsement, or implementation. At present, there are no empirical data to link specific quality scores with specific implementation outcomes (e.g., speed of adoption, spread of adoption) or specific clinical outcomes; this makes selection of quality thresholds to differentiate between high, moderate, and low quality guidelines a challenge. In the absence of these data, we provide examples of approaches that can be used to set quality thresholds:

- Prioritizing one domain: Through consensus or based on decisions by leadership, one quality domain may be prioritized over the others. Thus, thresholds can be created based on scores for the prioritized domain (e.g., high quality guidelines are those with a Domain 3 score >70%).
- Staged AGREE II appraisal: If users value one domain over the others, they can first appraise the guidelines using that domain only. Only those guidelines that meet a quality threshold for that domain (e.g., >70%) are then appraised using the other five AGREE II domains.
- Considering all domain scores: Users can create a threshold across all six domain scores based on consensus or decisions by leadership (e.g., high quality guidelines are those with domain scores that are all >70%). Alternatively, users might create different thresholds for each of the domains.
- Thresholds for improvement over time: If evaluating changes in scores for guidelines over time, users can create thresholds for improvement (e.g., at least 10% improvement in each domain score for guidelines by a particular developer over a period of five years).

Any decisions about how to define quality thresholds should be made by a panel of all relevant stakeholders before beginning the AGREE II appraisals. Decisions should be guided by the context in which the guideline is to be used and by evaluating the importance of the different domains and items in that context.

V. Overall Assessment

Upon completing the 23 items, AGREE II users will provide 2 overall assessments of the guideline. The overall assessment requires the user to make a judgment as to the quality of the guideline, taking into account the criteria considered in the assessment process. The user is also asked whether he/she would recommend use of the guideline.

The next pages include, by domain, guidance for rating each of the 23 items of the AGREE II when appraising a guideline. Each item includes a description, suggestions for where to find the item information, and guidance for how to rate.

DOMAIN 1. SCOPE AND PURPOSE

1. The overall objective(s) of the guideline is (are) specifically described.
2. The health question(s) covered by the guideline is (are) specifically described.
3. The population (patients, public, etc.) to whom the guideline is meant to apply is specifically described.

SCOPE AND PURPOSE

1. The overall objective(s) of the guideline is (are) specifically described.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

This deals with the potential health impact of a guideline on society and populations of patients or individuals. The overall objective(s) of the guideline should be described in detail and the expected health benefits from the guideline should be specific to the clinical problem or health topic. For example, specific statements would be:

- Preventing (long term) complications of patients with diabetes mellitus
- Lowering the risk of subsequent vascular events in patients with previous myocardial infarction
- Most effective population-based colorectal screening strategies
- Providing guidance on the most effective therapeutic treatment and management of patients with diabetes mellitus.

Where to Look:

Examine the opening paragraphs/chapters for a description of the scope and purpose of the guideline. In some cases, the rationale or need for the guideline is described in a document separate from the guideline, for instance, in the guideline proposal. Examples of commonly labeled sections or chapters in a guideline where this information can be found include: introduction, scope, purpose, rationale, background, and objectives.

How to Rate:

Item content includes the following CRITERIA:

- health intent(s) (i.e., prevention, screening, diagnosis, treatment, etc.)
- expected benefit or outcome
- target(s) (e.g., patient population, society)

Additional CONSIDERATIONS:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?

SCOPE AND PURPOSE

2. The health question(s) covered by the guideline is (are) specifically described.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

A detailed description of the health questions covered by the guideline should be provided, particularly for the key recommendations (see Item 17), although they need not be phrased as questions. Following the examples provided in question 1:

- How many times a year should the HbA1c be measured in patients with diabetes mellitus?
- What should the daily aspirin dosage for patients with proven acute myocardial infarction be?
- Does population-based colorectal screening using the fecal occult blood test reduce mortality of colorectal cancer?
- Is self-monitoring effective for blood glucose control in patients with Type 2 diabetes?

Where to Look:

Examine the opening paragraphs/chapters for a description of the scope and purpose of the guideline. In some cases, the questions are described in a document separate from the guideline, for instance in a search specification. Examples of commonly labeled sections or chapters in a guideline where this information can be found include: questions, scope, purpose, rationale, and background.

How to Rate:

Item content includes the following **CRITERIA**:

- target population
- intervention(s) or exposure(s)
- comparisons (if appropriate)
- outcome(s)
- health care setting or context

Additional **CONSIDERATIONS**:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?
- Is there enough information provided in the question(s) for anyone to initiate the development of a guideline on this topic or to understand the patients/populations and contexts profiled in the guideline?

SCOPE AND PURPOSE

3. The population (patients, public, etc.) to whom the guideline is meant to apply is specifically described.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

A clear description of the population (i.e., patients, public, etc.) covered by a guideline should be provided. The age range, sex, clinical description, and comorbidity may be provided. For example:

- A guideline on the management of diabetes mellitus only includes patients with non-insulin dependent diabetes mellitus and excludes patients with cardiovascular comorbidity.
- A guideline on the management of depression only includes patients with major depression according to the DSM-IV criteria, and excludes patients with psychotic symptoms and children.
- A guideline on screening of breast cancer only includes women, aged between 50 and 70 years, with no history of cancer and with no family history of breast cancer.

Where to Look:

Examine the opening paragraphs/chapters for a description of the target population of the guideline. The explicit exclusion of some populations (for instance children) is also covered by this item. Examples of commonly labeled sections or chapters in a guideline where this information can be found include: patient population, target population, relevant patients, scope, and purpose.

How to Rate:

Item content includes the following **CRITERIA**:

- target population, gender and age
- clinical condition (if relevant)
- severity/stage of disease (if relevant)
- comorbidities (if relevant)
- excluded populations (if relevant)

Additional **CONSIDERATIONS**:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?
- Is the population information specific enough so that the correct and eligible individuals would receive the action recommended in the guideline?

DOMAIN 2. STAKEHOLDER INVOLVEMENT

4. The guideline development group includes individuals from all relevant professional groups.
5. The views and preferences of the target population (patients, public, etc.) have been sought.
6. The target users of the guideline are clearly defined.

STAKEHOLDER INVOLVEMENT

4. The guideline development group includes individuals from all relevant professional groups.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

This item refers to the professionals who were involved at some stage of the development process. This may include members of the steering group, the research team involved in selecting and reviewing/rating the evidence and individuals involved in formulating the final recommendations. *This item excludes individuals who have externally reviewed the guideline (see Item 13). This item excludes target population representation (see Item 5).* Information about the composition, discipline, and relevant expertise of the guideline development group should be provided.

Where to Look:

Examine the opening paragraphs/chapters, acknowledgement section or appendices for the composition of the guideline development group. Examples of commonly labeled sections or chapters in a guideline where this information can be found include: methods, guideline panel member list, acknowledgements, and appendices.

How to Rate:

Item content includes the following **CRITERIA**:

- For each member of the guideline development group, the following information is included:
 - name
 - discipline/content expertise (e.g., neurosurgeon, methodologist)
 - institution (e.g., St. Peter's hospital)
 - geographical location (e.g., Seattle, WA)
 - a description of the member's role in the guideline development group

Additional **CONSIDERATIONS**:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?
- Are the members an appropriate match for the topic and scope? Potential candidates include relevant clinicians, content experts, researchers, policy makers, clinical administrators, and funders.
- Is there at least one methodology expert included in the development group (e.g., systematic review expert, epidemiologist, statistician, library scientist, etc.)?

STAKEHOLDER INVOLVEMENT

5. The views and preferences of the target population (patients, public, etc.) have been sought.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

Information about target population experiences and expectations of health care should inform the development of guidelines. There are various methods for ensuring that these perspectives inform the different stages of guideline development by stakeholders. For example, formal consultations with patients/public to determine priority topics, participation of these stakeholders on the guideline development group, or external review by these stakeholders on draft documents. Alternatively, information could be obtained from interviews of these stakeholders or from literature reviews of patient/public values, preferences or experiences. There should be evidence that some process has taken place and that stakeholders' views have been considered.

Where to Look:

Examine the paragraphs on the guideline development process. Examples of commonly labeled sections or chapters in a guideline where this information can be found include: scope, methods, guideline panel member list, external review, and target population perspectives.

How to Rate:

Item content includes the following **CRITERIA**:

- statement of type of strategy used to capture patients'/public's' views and preferences (e.g., participation in the guideline development group, literature review of values and preferences)
- methods by which preferences and views were sought (e.g., evidence from literature, surveys, focus groups)
- outcomes/information gathered on patient/public information
- description of how the information gathered was used to inform the guideline development process and/or formation of the recommendations

Additional **CONSIDERATIONS**:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?

STAKEHOLDER INVOLVEMENT

6. The target users of the guideline are clearly defined.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

The target users should be clearly defined in the guideline, so the reader can immediately determine if the guideline is relevant to them. For example, the target users for a guideline on low back pain may include general practitioners, neurologists, orthopaedic surgeons, rheumatologists, and physiotherapists.

Where to Look:

Examine the opening paragraphs/chapters for a description of the target users of the guideline. Examples of commonly labeled sections or chapters in a guideline where this information can be found include: target user and intended user.

How to Rate:

Item content includes the following **CRITERIA**:

- clear description of intended guideline audience (e.g. specialists, family physicians, patients, clinical or institutional leaders/administrators)
- description of how the guideline may be used by its target audience (e.g., to inform clinical decisions, to inform policy, to inform standards of care)

Additional **CONSIDERATIONS**:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?
- Are the target users appropriate for the scope of the guideline?

DOMAIN 3. RIGOUR OF DEVELOPMENT

7. Systematic methods were used to search for evidence.
8. The criteria for selecting the evidence are clearly described.
9. The strengths and limitations of the body of evidence are clearly described.
10. The methods for formulating the recommendations are clearly described.
11. The health benefits, side effects, and risks have been considered in formulating the recommendations.
12. There is an explicit link between the recommendations and the supporting evidence.
13. The guideline has been externally reviewed by experts prior to its publication.
14. A procedure for updating the guideline is provided.

RIGOUR OF DEVELOPMENT

7. Systematic methods were used to search for evidence.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

Details of the strategy used to search for evidence should be provided including search terms used, sources consulted, and dates of the literature covered. Sources may include electronic databases (e.g. MEDLINE, EMBASE, CINAHL), databases of systematic reviews (e.g. the Cochrane Library, DARE), handsearching journals, reviewing conference proceedings, and other guidelines (e.g. the US National Guideline Clearinghouse, the German Guidelines Clearinghouse). The search strategy should be as comprehensive as possible and executed in a manner free from potential biases and sufficiently detailed to be replicated.

Where to Look:

Examine the paragraphs/chapters describing the guideline development process. In some cases the search strategies are described in separate documents or in an appendix to the guideline. Examples of commonly labelled sections or chapters in a guideline where this information can be found include: methods, literature search strategy, and appendices.

How to Rate:

Item content includes the following **CRITERIA**:

- named electronic database(s) or evidence source(s) where the search was performed (e.g., MEDLINE, EMBASE, PsychINFO, CINAHL)
- time periods searched (e.g., January 1, 2004 to March 31, 2008)
- search terms used (e.g., text words, indexing terms, subheadings)
- full search strategy included (e.g., possibly located in appendix)

Additional **CONSIDERATIONS**:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?
- Is the search relevant and appropriate to answer the health question? (e.g., all relevant databases and, appropriate search terms used)
- Is there enough information provided for anyone to replicate the search?

RIGOUR OF DEVELOPMENT

8. The criteria for selecting the evidence are clearly described.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

Criteria for including/excluding evidence identified by the search should be provided. These criteria should be explicitly described and reasons for including and excluding evidence should be clearly stated. For example, guideline authors may decide to only include evidence from randomized clinical trials and to exclude articles not written in English.

Where to Look:

Examine the paragraphs/chapters describing the guideline development process. In some cases, the inclusion or exclusion criteria for selecting the evidence are described in separate documents or in an Appendix to the guideline. Examples of commonly labeled sections or chapters in a guideline where this information can be found include: methods, literature search, inclusion/exclusion criteria, and appendices.

How to Rate:

Item content includes the following **CRITERIA**:

- description of the inclusion criteria, including
 - target population (patient, public, etc.) characteristics
 - study design
 - comparisons (if relevant)
 - outcomes
 - language (if relevant)
 - context (if relevant)
- description of the exclusion criteria (if relevant; e.g., *French only* listed in the inclusion criteria statement could logically preclude *non-French* listed in the exclusion criteria statement)

Additional **CONSIDERATIONS**:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?
- Is there a rationale given for the chosen inclusion/exclusion criteria?
- Do inclusion/exclusion criteria align with the health question(s)?
- Are there reasons to believe that relevant literature may not have been considered?

RIGOUR OF DEVELOPMENT

9. The strengths and limitations of the body of evidence are clearly described.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

Statements highlighting the strengths and limitations of the evidence should be provided. This ought to include explicit descriptions - using informal or formal tools/methods - to assess and describe the risk of bias for individual studies and/or for specific outcomes and/or explicit commentary of the body of evidence aggregated across all studies. This may be presented in different ways, for example: using tables commenting on different quality domains; the application of a formal instrument or strategy (e.g., Jadad scale, GRADE method); or descriptions in the text.

Where to Look:

Examine the paragraphs/chapters describing the guideline development process for information on how the methodological quality of the studies (e.g., risk of bias) were described. Evidence tables are often used to summarize quality features. Some guidelines make a clear distinction between description and interpretation of evidence, for instance, in a results section and a discussion section, respectively.

How to Rate:

Item content includes the following **CRITERIA**:

- descriptions of how the body of evidence was evaluated for bias and how it was interpreted by members of the guideline development group
- aspects upon which to frame descriptions include:
 - study design(s) included in body of evidence
 - study methodology limitations (sampling, blinding, allocation concealment, analytical methods)
 - appropriateness/relevance of primary and secondary outcomes considered
 - consistency of results across studies
 - direction of results across studies
 - magnitude of benefit versus magnitude of harm
 - applicability to practice context

Additional **CONSIDERATIONS**:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?
- Are the descriptions appropriate, neutral, and unbiased? Are the descriptions complete?

RIGOUR OF DEVELOPMENT

10. The methods for formulating the recommendations are clearly described.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

A description of the methods used to formulate the recommendations and how final decisions were arrived at should be provided. For example, methods may include a voting system, informal consensus, and formal consensus techniques (e.g., Delphi, Glaser techniques). Areas of disagreement and methods of resolving them should be specified.

Where to Look:

Examine the paragraphs/chapters describing the guideline development process. In some cases, the methods used to formulate the recommendations are described in separate documents or in an appendix to the guideline. Examples of commonly labeled sections or chapters in a guideline where this information can be found include methods and guideline development process.

How to Rate:

Item content includes the following **CRITERIA**:

- description of the recommendation development process (e.g., steps used in modified Delphi technique, voting procedures that were considered)
- outcomes of the recommendation development process (e.g., extent to which consensus was reached using modified Delphi technique, outcome of voting procedures)
- description of how the process influenced the recommendations (e.g., results of Delphi technique influence final recommendation, alignment with recommendations and the final vote)

Additional **CONSIDERATIONS**:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?
- Was a formal process used to arrive at the recommendations?
- Were the methods appropriate?

RIGOUR OF DEVELOPMENT

11. The health benefits, side effects, and risks have been considered in formulating the recommendations.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

The guideline should consider health benefits, side effects, and risks when formulating the recommendations. For example, a guideline on the management of breast cancer may include a discussion on the overall effects on various final outcomes. These may include: survival, quality of life, adverse effects, and symptom management or a discussion comparing one treatment option to another. There should be evidence that these issues have been addressed.

Where to Look:

Examine the paragraphs/chapters describing the guideline development process for a description of the body of evidence, its interpretation, and the translation to practice recommendations. Examples of commonly labeled sections or chapters in a guideline where this information can be found include: methods, interpretation, discussion, and recommendations.

How to Rate:

Item content includes the following **CRITERIA**:

- supporting data and report of benefits
- supporting data and report of harms/side effects/risks
- reporting of the balance/trade-off between benefits and harms/side effects/risks
- recommendations reflect considerations of both benefits and harms/side effects/risks

Additional **CONSIDERATIONS**:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?
- Is the discussion an integral part of the guideline development process? (i.e., taking place during recommendation formulation rather than post-formulation as an afterthought)
- Has the guideline development group considered the benefits and harms equally?

RIGOUR OF DEVELOPMENT

12. There is an explicit link between the recommendations and the supporting evidence.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

An explicit link between the recommendations and the evidence on which they are based should be included in the guideline. The guideline user should be able to identify the components of the body of evidence relevant to each recommendation.

Where to Look:

Define and examine the recommendations in the guideline and the text describing the body of evidence that underpins them. Examples of commonly labeled sections or chapters in a guideline where this information can be found include: recommendations and key evidence.

How to Rate:

Item content includes the following *CRITERIA*:

- the guideline describes how the guideline development group linked and used the evidence to inform recommendations
- each recommendation is linked to a key evidence description/paragraph and/or reference list
- recommendations linked to evidence summaries, evidence tables in the results section of the guideline

Additional *CONSIDERATIONS*:

- Is there congruency between the evidence and recommendations?
- Is the link between the recommendations and supporting evidence easy to find in the guideline?
- When evidence is lacking or a recommendation is informed primarily by consensus of opinion by the guideline group, rather than the evidence, is this clearly stated and described?

RIGOUR OF DEVELOPMENT

13. The guideline has been externally reviewed by experts prior to its publication.

1	2	3	4	5	6	7
Strongly Disagree						Strongly Agree

Comments

User's Manual Description:

A guideline should be reviewed externally before it is published. Reviewers should not have been involved in the guideline development group. Reviewers should include experts in the clinical area as well as some methodological experts. Target population (patients, public) representatives may also be included. A description of the methodology used to conduct the external review should be presented, which may include a list of the reviewers and their affiliation.

Where to Look:

Examine the paragraphs/chapters describing the guideline development process and the acknowledgement section. Examples of commonly labeled sections or chapters in a guideline where this information can be found include: methods, results, interpretation, and acknowledgements.

How to Rate:

Item content includes the following **CRITERIA**:

- purpose and intent of the external review (e.g., to improve quality, gather feedback on draft recommendations, assess applicability and feasibility, disseminate evidence)
- methods taken to undertake the external review (e.g., rating scale, open-ended questions)
- description of the external reviewers (e.g., number, type of reviewers, affiliations)
- outcomes/information gathered from the external review (e.g., summary of key findings)
- description of how the information gathered was used to inform the guideline development process and/or formation of the recommendations (e.g., guideline panel considered results of review in forming final recommendations)

Additional **CONSIDERATIONS**:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?
- Are the external reviewers relevant and appropriate to the scope of the guideline? Was there a rationale given for choosing the included reviewers?
- How was information from the external review used by the guideline development group?

RIGOUR OF DEVELOPMENT

14. A procedure for updating the guideline is provided.

1	2	3	4	5	6	7
Strongly Disagree						Strongly Agree

Comments

User's Manual Description:

Guidelines need to reflect current research. A clear statement about the procedure for updating the guideline should be provided. For example, a timescale has been given or a standing panel is established who receives regularly updated literature searches and makes changes as required.

Where to Look:

Examine the introduction paragraph, the paragraphs describing the guideline development process and the closing paragraphs. Examples of commonly labeled sections or chapters in a guideline where this information can be found include: methods, guideline update, and date of guideline.

How to Rate:

Item content includes the following **CRITERIA**:

- a statement that the guideline will be updated
- explicit time interval or explicit criteria to guide decisions about when an update will occur
- methodology for the updating procedure is reported

Additional **CONSIDERATIONS**:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?
- Is there enough information provided to know when an update will occur or what criteria would trigger an update?

DOMAIN 4. CLARITY OF PRESENTATION

15. The recommendations are specific and unambiguous.
16. The different options for management of the condition or health issue are clearly presented.
17. Key recommendations are easily identifiable.

CLARITY OF PRESENTATION

15. The recommendations are specific and unambiguous.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

A recommendation should provide a concrete and precise description of which option is appropriate in which situation and in what population group, as informed by the body of evidence.

- An example of a specific recommendation is: Antibiotics should be prescribed in children two years or older with a diagnosis of acute otitis media if the pain lasts longer than three days or if the pain increases after the consultation despite adequate treatment with painkillers; in these cases, amoxicillin should be given for 7 days (supplied with a dosage scheme).
- An example of a vague recommendation is: Antibiotics are indicated for cases with an abnormal or complicated course.

It is important to note that in some instances, evidence is not always clear cut and there may be uncertainty about the best care option(s). In this case, the uncertainty should be stated in the guideline.

Where to Look:

Define and examine the recommendations in the guideline. Examples of commonly labeled sections or chapters in a guideline where this information can be found include: recommendations and executive summary.

How to Rate:

Item content includes the following **CRITERIA**:

- statement of the recommended action
- identification of the intent or purpose of the recommended action (e.g., to improve quality of life, to decrease side effects)
- identification of the relevant population (e.g., patients, public)
- caveats or qualifying statements, if relevant (e.g., patients or conditions for whom the recommendations would not apply)

Additional **CONSIDERATIONS**:

- In the event of multiple recommendations (e.g., management guidelines), is there clarity regarding to whom each recommendation applies?
- If there is uncertainty in the interpretation and discussion of the evidence, is the uncertainty reflected in the recommendations and explicitly stated?

CLARITY OF PRESENTATION

16. The different options for management of the condition or health issue are clearly presented.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

A guideline that targets the management of a disease should consider the different possible options for screening, prevention, diagnosis or treatment of the condition it covers. These possible options should be clearly presented in the guideline.

For example, a recommendation on the management of depression may contain the following treatment alternatives:

- a. Treatment with TCA
- b. Treatment with SSRI
- c. Psychotherapy
- d. Combination of pharmacological and psychological therapy

Where to Look:

Examine the recommendations and their supporting evidence. Examples of commonly labeled sections or chapters in a guideline where this information can be found include: executive summary, recommendations, discussion, treatment options, and treatment alternatives.

How to Rate:

Item content includes the following *CRITERIA*:

- description of options
- description of population or clinical situation most appropriate to each option

Additional *CONSIDERATIONS*:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?
- Is this pertaining to a guideline broad or narrow in scope? This item may be more relevant to guidelines that are broad in scope (e.g., covering the management of a condition or issue rather than focusing on a particular set of interventions for a specific condition/issue).

CLARITY OF PRESENTATION

17. Key recommendations are easily identifiable.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

Users should be able to find the most relevant recommendations easily. These recommendations answer the main question(s) that have been covered by the guideline and can be identified in different ways. For example, they can be summarized in a box, typed in bold, underlined or presented as flow charts or algorithms.

Where to Look:

Examples of commonly labeled sections or chapters in a guideline where this information can be found include: executive summary, conclusions, and recommendations. Some guidelines provide separate summaries with key recommendations (e.g., quick reference guide).

How to Rate:

Item content includes the following *CRITERIA*:

- description of recommendations in a summarized box, typed in bold, underlined, or presented as flow charts or algorithms
- specific recommendations are grouped together in one section

Additional *CONSIDERATIONS*:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?
- Are the key recommendations appropriately selected and do they reflect the key messages of the guideline?
- Are specific recommendations grouped in a section placed near the summary of the key evidence?

DOMAIN 5. APPLICABILITY

18. The guideline describes facilitators and barriers to its application.
19. The guideline provides advice and/or tools on how the recommendations can be put into practice.
20. The potential resource implications of applying the recommendations have been considered.
21. The guideline presents monitoring and/or auditing criteria.

APPLICABILITY

18. The guideline describes facilitators and barriers to its application.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

There may be existing facilitators and barriers that will impact the application of guideline recommendations.

For example:

- i. A guideline on stroke may recommend that care should be coordinated through stroke units and stroke services. There may be a special funding mechanism in the region to enable the formation of stroke units.
- ii. A guideline on diabetes in primary care may require that patients are seen and followed up in diabetic clinics. There may be an insufficient number of clinicians available in a region to enable clinics to be established.

Where to Look:

Examine the paragraph/chapter on the dissemination/implementation of the guideline or, if available, additional documents with specific plans or strategies for implementation of the guideline. Examples of commonly labeled sections or chapters in a guideline where this information can be found include: barriers, guideline utilization, and quality indicators.

How to Rate:

Item content includes the following **CRITERIA**:

- identification of the types of facilitators and barriers that were considered
- methods by which information regarding the facilitators and barriers to implementing recommendations were sought (e.g., feedback from key stakeholders, pilot testing of guidelines before widespread implementation)
- information/description of the types of facilitators and barriers that emerged from the inquiry (e.g., practitioners have the skills to deliver the recommended care, sufficient equipment is not available to ensure all eligible members of the population receive mammography)
- description of how the information influenced the guideline development process and/or formation of the recommendations

Additional **CONSIDERATIONS**:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?
- Does the guideline suggest specific strategies to overcoming the barriers?

APPLICABILITY

19. The guideline provides advice and/or tools on how the recommendations can be put into practice.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

For a guideline to be effective it needs to be disseminated and implemented with additional materials. For example, these may include: a summary document, a quick reference guide, educational tools, results from a pilot test, patient leaflets, or computer support. Any additional materials should be provided with the guideline.

Where to Look:

Examine the paragraph on the dissemination/implementation of the guideline and, if available, the specific accompanying materials that have been produced to support the dissemination and implementation of the guideline. Examples of commonly labeled sections or chapters in a guideline where this information can be found include: tools, resources, implementation, and appendices.

How to Rate:

Item content includes the following **CRITERIA**:

- an implementation section in the guideline
- tools and resources to facilitate application:
 - guideline summary documents
 - links to check lists, algorithms
 - links to how-to manuals
 - solutions linked to barrier analysis (see Item 18)
 - tools to capitalize on guideline facilitators (see Item 18)
 - outcome of pilot test and lessons learned
- directions on how users can access tools and resources

Additional **CONSIDERATIONS**:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?
- Is there information about the development of the implementation tools and validation procedures?

APPLICABILITY

20. The potential resource implications of applying the recommendations have been considered.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

The recommendations may require additional resources in order to be applied. For example, there may be a need for more specialized staff, new equipment, and expensive drug treatment. These may have cost implications in the paragraphs that discuss the evidence or decisions behind the recommendations. Examples of commonly labeled sections or chapters in a guideline where this information can be found include: methods, cost utility, cost effectiveness, acquisition costs, and implications for budgets.

Where to Look:

Examine the paragraph(s) on the dissemination/implementation of the guideline or, if available, additional documents with specific plans or strategies for implementation of the guideline. Some guidelines present cost implications in the paragraphs that discuss the evidence or decisions behind the recommendations. Examples of commonly labeled sections or chapters in a guideline where this information can be found include: methods, cost utility, cost effectiveness, acquisition costs, and implications for budgets.

How to Rate:

Item content includes the following **CRITERIA**:

- identification of the types of cost information that were considered (e.g., economic evaluations, drug acquisition costs)
- methods by which the cost information was sought (e.g., a health economist was part of the guideline development panel, use of health technology assessments for specific drugs, etc.)
- information/description of the cost information that emerged from the inquiry (e.g., specific drug acquisition costs per treatment course)
- description of how the information gathered was used to inform the guideline development process and/or formation of the recommendations

Additional **CONSIDERATIONS**:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?
- Were appropriate experts involved in finding and analyzing the cost information?

APPLICABILITY

21. The guideline presents monitoring and/or auditing criteria.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

Measuring the application of guideline recommendations can facilitate their ongoing use. This requires clearly defined criteria that are derived from the key recommendations in the guideline. The criteria may include process measures, behavioural measures, clinical or health outcome measures. Examples of monitoring and audit criteria are:

- The HbA1c should be < 8.0%.
- The level of diastolic blood pressure should be < 95 mmHg.
- 80% of the population aged 50 years should receive colorectal cancer screening rates using fecal occult blood tests.
- If complaints of acute otitis media last longer than three days, amoxicillin should be prescribed.

Where to Look:

Examine the paragraph/chapter on auditing or monitoring the use of the guideline or, if available, additional documents with specific plans or strategies for evaluation of the guideline. Examples of commonly labeled sections or chapters in a guideline where this information can be found include: recommendations, quality indicators, and audit criteria.

How to Rate:

Item content includes the following **CRITERIA**:

- identification of criteria to assess guideline implementation or adherence to recommendations
- criteria for assessing impact of implementing the recommendations
- advice on the frequency and interval of measurement
- descriptions or operational definitions of how the criteria should be measured

Additional **CONSIDERATIONS**:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?
- Are a range of criteria provided including process measures, behavioural measures, and clinical or health outcomes?

DOMAIN 6. EDITORIAL INDEPENDENCE

22. The views of the funding body have not influenced the content of the guideline.
23. Competing interests of guideline development group members have been recorded and addressed.

EDITORIAL INDEPENDENCE

22. The views of the funding body have not influenced the content of the guideline.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

Many guidelines are developed with external funding (e.g., government, professional associations, charity organizations, pharmaceutical companies). Support may be in the form of financial contribution for the complete development, or for parts of it (e.g., printing of the guidelines). There should be an explicit statement that the views or interests of the funding body have not influenced the final recommendations.

Where to Look:

Examine the paragraphs/chapters on the guideline development process or acknowledgements section. Examples of commonly labeled sections or chapters in a guideline where this information can be found include: disclaimer and funding source.

How to Rate:

Item content includes the following *CRITERIA*:

- the name of the funding body or source of funding (or explicit statement of no funding)
- a statement that the funding body did not influence the content of the guideline

Additional *CONSIDERATIONS*:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?
- How did the guideline development group address potential influence from the funding body?

EDITORIAL INDEPENDENCE

23. Competing interests of guideline development group members have been recorded and addressed.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

User's Manual Description:

There are circumstances when members of the development group may have competing interests. For example, this would apply to a member of the development group whose research on the topic covered by the guideline is also funded by a pharmaceutical company. There should be an explicit statement that all group members have declared whether they have any competing interests.

Where to Look:

Examine the paragraphs/chapters describing the guideline development group or acknowledgements section. Examples of commonly labeled sections or chapters in a guideline where this information can be found include: methods, conflicts of interest, guideline panel, and appendix.

How to Rate:

Item content includes the following **CRITERIA**:

- description of the types of competing interests considered
- methods by which potential competing interests were sought
- description of the competing interests
- description of how the competing interests influenced the guideline process and development of recommendations

Additional **CONSIDERATIONS**:

- Is the item well written? Are the descriptions clear and concise?
- Is the item content easy to find in the guideline?
- What measures were taken to minimize the influence of competing interests on guideline development or formulation of the recommendations?

OVERALL GUIDELINE ASSESSMENT

OVERALL GUIDELINE ASSESSMENT

For each question, please choose the response which best characterizes the guideline assessed:

1. Rate the overall quality of this guideline.

1 Lowest possible quality	2	3	4	5	6	7 Highest possible quality
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2. I would recommend this guideline for use.

Yes	
Yes, with modifications	
No	

NOTES

User's Manual Description:

The overall assessment requires the AGREE II user to make a judgment as to the quality of the guideline, taking into account the appraisal items considered in the assessment process.

AGREE II INSTRUMENT

DOMAIN 1. SCOPE AND PURPOSE

1. The overall objective(s) of the guideline is (are) specifically described.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

2. The health question(s) covered by the guideline is (are) specifically described.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

3. The population (patients, public, etc.) to whom the guideline is meant to apply is specifically described.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

DOMAIN 2. STAKEHOLDER INVOLVEMENT

4. The guideline development group includes individuals from all relevant professional groups.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

5. The views and preferences of the target population (patients, public, etc.) have been sought.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

6. The target users of the guideline are clearly defined.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

DOMAIN 3. RIGOUR OF DEVELOPMENT

7. Systematic methods were used to search for evidence.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

8. The criteria for selecting the evidence are clearly described.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

9. The strengths and limitations of the body of evidence are clearly described.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

DOMAIN 3. RIGOUR OF DEVELOPMENT continued

10. The methods for formulating the recommendations are clearly described.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

11. The health benefits, side effects, and risks have been considered in formulating the recommendations.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

12. There is an explicit link between the recommendations and the supporting evidence.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

DOMAIN 3. RIGOUR OF DEVELOPMENT continued

13. The guideline has been externally reviewed by experts prior to its publication.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

14. A procedure for updating the guideline is provided.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

DOMAIN 4. CLARITY OF PRESENTATION

15. The recommendations are specific and unambiguous.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

16. The different options for management of the condition or health issue are clearly presented.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

17. Key recommendations are easily identifiable.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

DOMAIN 5. APPLICABILITY

18. The guideline describes facilitators and barriers to its application.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

19. The guideline provides advice and/or tools on how the recommendations can be put into practice.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

20. The potential resource implications of applying the recommendations have been considered.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

DOMAIN 5. APPLICABILITY continued

21. The guideline presents monitoring and/or auditing criteria.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

DOMAIN 6. EDITORIAL INDEPENDENCE

22. The views of the funding body have not influenced the content of the guideline.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

23. Competing interests of guideline development group members have been recorded and addressed.

1 Strongly Disagree	2	3	4	5	6	7 Strongly Agree
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Comments

OVERALL GUIDELINE ASSESSMENT

For each question, please choose the response which best characterizes the guideline assessed:

1. Rate the overall quality of this guideline.

1 Lowest possible quality	2	3	4	5	6	7 Highest possible quality
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2. I would recommend this guideline for use.

Yes	
Yes, with modifications	
No	

NOTES
