Proposal form for the evaluation of a genetic test for NHS Service
Gene Dossier

Test – Disease – Population Triad

Disease – name and description (please provide any alternative names you wish listed) (A)-Testing Criteria	Medium chain acyl-CoA dehydrogenase deficiency (MCADD) is an autosomal recessive disorder and the most common defect of the mitochondrial fatty acid oxidation pathway. Affected individuals are unable to utilise medium-chain fatty acids as an energy source, leading to hypoketotic hypoglycaemia and accumulation of medium-chain metabolites in plasma and urine. Clinical presentation of MCADD usually results from fasting during periods of increased metabolic stress during infancy. Symptoms include drowsiness and lethargy that can progress to coma. Approximately 25% of patients die during their first metabolic crisis. In the children that survive this early crisis, up to 30% are reported to experience some form of developmental delay as a result hypoglycaemic CNS damage The prognosis for MCADD is excellent and adverse effects can be largely prevented by early identification and pre-symptomatic treatment.
OMIM number for disease	201450
Gene – name and description (please provide any alternative names you wish listed)	ACADM
OMIM number for Gene	607008
Mutational spectrum for which you test	Full screen for small-scale mutations within the ACADM gene, including the common c.985A>G mutation.
Technical Method (s)	Bi-directional sequence analysis of coding regions and intron/exon boundaries of the ACADM gene. Subsequent analysis of sequence data using Mutation Surveyor software to identify any variants.
Validation Process Note please explain how this test has been validated for use in your laboratory	Prior to 2007, the laboratory offered screening for 5 exons of the ACADM gene that would pick up the most common published MCADD mutations including the c.985A>G in exon 11. Screening of all 12 exons of ACADM was developed to offer a comprehensive service to patients and their families. The extended mutation screening was validated on a panel of 41 samples, including a number where the genotype had previously been ascertained in another laboratory. In total 7 samples were confirmed with

	an MCADD genotype (4 c.985A>G homozygotes, 2 heterozygous for c.985A>G and another mutation and 1 compound heterozygous for two rare mutations. Five rare mutations were identified, one of which (c.430_432delAAG) had not previously been reported. In addition 10 carriers were identified. Through sequence analysis of the ACADM gene, the coding region was found to be extremely polymorphic. This would preclude mutation scanning as a testing strategy and means that sequencing is the method of choice for diagnostic testing.
Are you providing this test already? If yes, how many reports have you produced? Please give the number of mutation positive/negative samples you have reported	The laboratory in Dundee is currently offering complete screening of the ACADM screen as a diagnostic service. In total 202 MCADD reports have been issued by the laboratory for patients and family members.
For how long have you been providing this service?	Sequence analysis of the entire coding region of ACADM gene has been offered since 2007.
Is there specialised local clinical/research expertise for this disease?	Yes / No / Please provide details The Molecular Genetic laboratory has very close links with the local Biochemistry Dept. In addition, the laboratory has recently been designated as the centre to offer extended mutation screening for ACADM mutations when neonatal blood-spot screening for MCADD is established in Scotland.
Are you testing for other genes/diseases closely allied to this one? Please give details	No
Your Activity How many tests do you (intend to) provide annually in your laboratory?	Within Scotland, expect approximately 10 cases/year. If offered as part of UKGTN, approximately 60-70 per year.
Based on experience how many tests will be required nationally (UK)? Please identify the information on which this is based	Results from a 6 UK labs that piloted neonatal screening for MCAD over 2 years using tandem MS, identified 103 cases from 750,000 babies that were screen positive. Of these, 48 were homozygous for the c.985A>G mutation and 55 required extended mutation screening. The cohort of neonates screened is slightly greater than the annual UK birth rate and thus at most, nationally 55 samples per annum would require extended screening for MCAD mutations.

Epidemiology

Estimated prevalence of disease in the general UK population Please identify the information on which this is based	Data from the MCADD pilot project (UK Newborn Screening Programme Centre), estimated a disease prevalence in the UK of 1/8333 based on approximately 750,000 neonates screened.
Estimated gene frequency (Carrier frequency or allele frequency) Please identify the information on which this is based	It is estimated that between 1/40-1/80 individuals in the UK are carriers. (UK Newborn Screening Programme Centre)
Estimated penetrance Please identify the information on which this is based	It is difficult to estimate the disease penetrance for MCADD since not all individuals with MCADD based on biochemical ascertainment will present clinically. It has been estimated that MCADD newborn screening will detect 2-3 times more cases of MCADD than will be ascertained clinically (Rhead, 2006: J Inherit Metab Dis. 29:370-377). Data from a study by Schulze et al (2003. Paediatrics, 111: 1399-1406) indicated that 25% of infants detected by newborn screening had a mild phenotype that may not present clinically while the remainder had classic MCADD.
Target Population The essential clinical or family history features defining the target population must be described. (C)-Testing Criteria	Infants who present in the first few years of life, including the neonatal period, with hypoketotic hypoglycaemia, lethargy, encephalopathy and seizures during illness or prolonged fasting. Siblings of confirmed cases of MCADD as well as carrier testing for parents and other family members.
Estimated prevalence of disease in the target population	Data from UK Newborn Screening pilot project confirmed 87 cases of MCADD in 103 cases from tandem mass spectroscopy (84%). Of the remaining 16 cases that were screen positive by tandem MS, 11 were confirmed to be heterozygous for an ACADM mutation, while 5 had no identifiable mutations in the gene. The revised false-positive rate for newborn screening is therefore 4.8%.

Intended Use (Please use the questions in Annex A to inform your answers)

Please tick the relevant clinical purpose of testing	YES	NO
Diagnosis	4	
Treatment	4	
Prognosis & Management	4	
Presymptomatic testing	4	
Risk Assessment	4	

Test Characteristics

Analytical sensitivity and specificity This should be based on your own laboratory data for the specific test being applied for or the analytical sensitivity and specificity of the method/technique to be used in the case of a test yet to be set up. If a number of genes will be tested, please include your testing strategy and data on the expected proportions of positive results for each part of the process. It may be helpful to include a diagram to illustrate the testing strategy.	Bi-directional sequencing coupled with Mutation Surveyor software has a quoted sensitivity of >99%. The quoted specificity is also >99%.
Clinical sensitivity and specificity of test in target population The *clinical sensitivity* of a test is the probability of a positive test result when disease is known to be present; the *clinical specificity* is the probability of a negative test result when disease is known to be absent. The denominator in this case is the number with the disease (for sensitivity) or the number without disease (for specificity)	The clinical specificity is expected to approach 100%, although variants of uncertain clinical significance may be identified during testing of affected or unaffected individuals.

Clinical validity (positive and negative predictive value in the target population) The *clinical validity* of a genetic test is a measure of how well the test predicts the presence or absence of the phenotype, clinical disease or predisposition. It is measured by its *positive predictive value* (the probability of getting the disease given a positive test) and *negative predictive value* (the probability of not getting the disease given a negative test). The denominator in this case is the number of people with a positive or a negative test respectively - not the number with or without the disease. The clinical validity may be calculated knowing the sensitivity and the specificity and the prevalence of the disease in the population being studied. Positive and negative predictive values depend critically on the prevalence of the disease in the test population	For children with hypoglycaemia with raised C8 levels, likely to have a high PPV since if left untreated they have a high probability of developing MCADD. Of those children identified through newborn screening programmes, approximately 50% were homozygous for the c.985A>G mutation. Individuals with this genotype have a greater than 80% risk of developing clinical disease with associated mortality and neurological complications without appropriate treatment. For the non-c.985A>G homozygotes, the risk of developing disease is less clear. Evidence from newborn screening has shown that approximately 2-3 times more cases of MCADD have been identified than will be ascertained clinically.

Clinical utility of test in target population (Please refer to Appendix A) Please provide a full description of the clinical care pathway for those individuals undergoing testing. This should include details of which medical specialties will be able to refer for testing. (B)-Testing Criteria How will the test add to the management of the patient or alter clinical outcome?	There is a significant rate of mortality and morbidity associated with MCADD. Death has been reported in up to 25% of children with MCADD who present with acute symptoms. In those children that survive an early metabolic crisis, up to 30% are reported to experience some form of developmental delay as a result hypoglycaemic CNS damage. Early identification and confirmation of MCAD is therefore vital to prevent these complications. Results from a number of neonatal blood-spot screening programmes have shown a reduction in death rates (from 25% to approx 7%) in infants confirmed with MCADD. Furthermore, to date there have been no reports of neurologic impairment in infants identified early with MCADD. The adverse effects of MCADD can be largely prevented by early identification and pre-symptomatic treatment. In emergency situations, IV dextrose is required. Fasting should be avoided and MCADD dietary management guidelines are available to clinicians and parents. Affected infants should be closely monitored by both clinicians and parents during periods of fever and or vomiting.

What impact will this test have on the NHS i.e. by removing the need for alternative management and/or investigations for this clinical population?	Confirmation of MCADD will allow testing of siblings. This is particularly important since although the majority of symptoms occur between 3 months and 3 years, cases of MCADD have been reported in children as old as 7 years and even into adulthood.
Is there an alternative means of diagnosis or prediction that does not involve molecular diagnosis? If so (and in particular if there is a biochemical test) please state the added advantage of the molecular test	Biochemical screening for raised C8 levels in blood samples is the first-line diagnostic test for MCADD both in the context of neonatal screening and for children presenting with unexplained hypoglycaemia. The diagnosis of MCADD can be confirmed by sequence analysis of the ACADM gene following this initial biochemistry result. In families in which MCADD mutations have been identified, molecular genetic testing is the preferred option for identifying carriers or asymptomatic homozygotes who may be at risk from the condition. Molecular genetic testing may be the only diagnostic option following the sudden death of a child, where suitable material for biochemical testing is not available.
Are there specific ethical, legal or social issues with this test?	Furthermore, molecular genetic testing may provide valuable prognostic information particularly regarding genotypes associated with a milder phenotype. This will be particularly important when neonatal MCADD screening rolls out across the whole of the UK. For example the mutation c.199T>C, has not been identified in clinical cases of MCADD but is a relatively common change in babies with elevated C8 levels identified through screening programmes.

Please complete the referral pathway diagram on the following page and the testing criteria form.

Referral Pathway Template –

NHS
UK Genetic Testing Network

NOTE: Please use this page as a template. Please expand the test boxes manually as needed.

TARGET POPULATION

Raised octanoylcarnitine levels from Biochemistry where the proband is greater than 1 year of age (i.e. not been through newborn blood-spot screening programme for MCADD).

Clinical symptoms (hypoglycaemia, lethargy, seizures during illness or prolonged fasting) where no biochemistry is available or possible.

Family history of MCADD with known pathogenic mutation(s)

WHAT TYPE AND LEVEL OF PROFESSIONAL OR REFERRER DO YOU ACCEPT SAMPLES FROM?

Samples will be accepted from Consultant Biochemists, Paediatricians and Clinical Geneticists; samples will also be received from MCADD Newborn Screening centres for confirmatory mutation analysis.

PLEASE PROVIDE DETAILS OF HOW REFERRALS WILL BE ASSESSED FOR APPROPRIATENESS?

See the attached Testing Criteria

Based on figures obtained from the UK Newborn Screening Programme MCADD pilot study, 60-70 cases of MCADD a year would be expected for UK.

UKGTN Testing criteria:

UK Genetic Testing Network

Name of Disease(s):
ACYL-CoA DEHYDROGENASE, MEDIUM-CHAIN, DEFICIENCY OF (201450)

Name of gene(s):
acyl-Coenzyme A dehydrogenase, C-4 to C-12 straight chain; ACADM (607008)

Patient name: **Date of birth:**

Patient postcode: **NHS number:**

Name of referrer:

Title/Position:
Lab ID:

Referrals will only be accepted from one of the following:

Referrer	Tick if this refers to you.
Clinical Geneticists	
Consultant Biochemist	
Consultant Paediatrician	

Minimum criteria required for testing to be appropriate as stated in the Gene Dossier:

Criteria	Tick if this patient meets criteria
Family history of MCADD with known pathogenic mutation(s)	
Clinical symptoms (hypoglycaemia, lethargy, seizures during illness or prolonged fasting) where no biochemistry is available or possible.	
Raised octanoylcarnitine levels from Biochemistry where the proband is greater than 1 year of age (i.e. not been through newborn blood-spot screening programme for MCADD)	

If the sample does not fulfil the clinical criteria or you are not one of the specified types of referrer and you still feel that testing should be performed please contact the laboratory to discuss testing of the sample.