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

Submitting laboratory:
London South East RGC GSTT

1. Disorder/condition – approved name (please provide UK spelling if different from US) and symbol as published on the OMIM database (alternative names will be listed on the UKGTN website).
If NGS panel test, please provide a name.

If this submission is for a panel test please complete appendix 1 listing all of the conditions included using approved OMIM name, symbol and OMIM number.

Carnitine Deficiency, Systemic Primary; CDSP

2. OMIM number for disorder/condition

If a panel test – see 1. Above. If a number of subpanels exist with different clinical entry points e.g. cancer panel test but different subpanels for different types of cancer (breast cancer, colon, phaeochromocytoma) , then please list the sub panels here:

#212140

3a. Disorder/condition – please provide, in laymen's terms, a brief (2-5 sentences/no more than 50 words) description of how the disorder(s) affect individuals and prognosis.

Patients with systemic primary carnitine deficiency (SPCD) have poor uptake of carnitine, an important chemical required by the cells in our bodies for the breakdown of fats to generate energy particularly during times when our food intake is low e.g. during an illness.

The severe form of SPCD typically manifests during early infancy and include encephalopathy (abnormal brain function), cardiomyopathy (an enlarged weakened heart), muscle weakness, vomiting, liver dysfunction and low blood glucose.

Some people with SPCD can be asymptomatic, however, all patients with SPCD are at risk of heart failure, liver problems and sudden death.

Treatment involves oral administration of carnitine, which if started early and ideally from birth, is effective and can prevent the development of cardiomyopathy and other irreversible organ damage.

3b. Disorder/condition – if required please expand on the description of the disorder provided in answer to Q3a.

4. Disorder/condition – mode of inheritance

If this submission is for a panel test, please complete the mode of inheritance for each condition in the table in appendix 1.

Autosomal recessive

5. Gene – approved name(s) and symbol as published on HGNC database (alternative names will be listed on the UKGTN website)

If this submission is for a panel test please complete appendix 1 listing all of the genes included using approved HGNC name, symbol, number and OMIM number. Please provide subpanel split (described in Q2 above) in appendix 1.

solute carrier family 22 (organic cation/carnitine transporter), member 5; SLC22A5

6a. OMIM number(s) for gene(s)
If a panel test – see 5. above

603377

6b. HGNC number(s) for gene(s) If a panel test – see 5. above
10969
7a. Gene – description(s) If this submission is for a panel test, please provide total number of genes and if there are subpanels, please also list the number genes per sub panel.
Located on Chromosome 5, 5q23.3 *SLC22A5* consists of 10 exons, spans ~3kb, encodes for the Solute Carrier Family 22 Member 5 protein. This protein is a high affinity sodium ion dependent Carnitine transporter.
7b. Number of amplicons to provide this test (molecular) or type of test (cytogenetic) (n/a for panel tests)
11
7c. GenU band that this test is assigned to for index case testing.
Band E
8. Mutational spectrum for which you test including details of known common mutations (n/a for panel tests) If this application is for a panel test to be used for different clinical phenotypes and/or various sub panel tests – please contact the team for advice before completing a Gene Dossier
89 variants (mutations) are currently listed on the public version of the HGMD (110 on HGMD public). The majority are missense, but 2 splice site variants are listed, a few ins/del variants and 9 gross deletions.
9a. Technical method(s) – please describe the test.
PCR and Sanger sequencing of all exons and intron/exon boundaries from extracted DNA. Testing for deletions has been considered. We are aware that a commercial MLPA kit is available from MRC-Holland which could be evaluated in future on a case by case basis. At Guys we are currently looking at more widely applicable techniques (e.g. high resolution array CGH based tests) which could be used for many genes rather than just one gene per MLPA kit.
9b. For panel tests, please specify the strategy for dealing with gaps in coverage.
N/A
9c. Does the test include MLPA? **(For panel tests, please provide this information in appendix 1)**
No
9d. If NGS is used, does the lab adhere to the Association of Clinical Genetic Science Best Practice Guidelines for NGS?
N/A
10. Is the assay to be provided by the lab or is it to be outsourced to another provider? **If to be outsourced, please provide the name of the laboratory and a copy of their ISO certificate or their CPA number.**
Provided by this laboratory
11. Validation process Please explain how this test has been validated for use in your laboratory, including calculations of the sensitivity and specificity for the types of mutations reported to cause the clinical phenotype. Note that

the preferred threshold for validation and verification is ≥95% sensitivity (with 95% Confidence Intervals). Your internal validation documentation can be submitted as an appendix (and will be included in the published Gene Dossier available on the website). The validation information should include data on establishing minimum read depth and horizontal coverage for the regions of interest, reproducibility of the pipeline, accuracy of variant calling, filtering of common variants and artefacts.

If this submission is for a panel test, please provide a summary of evidence of instrument and pipeline validation and complete the tables below.

Sequencing is a standard analytical method used in the laboratory for a range of diagnostic services requiring mutation detection. Analysis is conducted in accordance with CPA standards for which we are fully accredited.

12a. Are you providing this test already?

Yes

12b. If yes, how many reports have you produced?

(3 diagnostic)
(2 carrier tests)

Sanger Based Tests	NGS Based Tests
5	

12c. Number of reports with a pathogenic (or likely pathogenic) mutation identified?

(3 diagnostic)
(2 carrier tests)

Sanger Based Tests	NGS Based Tests
5	

12d. Please provide the time period in which these reports have been produced and whether in a research or a full clinical diagnostic setting.

5 months. Diagnostic laboratory.

13a. Is there specialised local clinical/research expertise for this disorder?

Yes

13b. If yes, please provide details

The Biochemical Genetics Unit works closely with the Inherited Metabolic Disease (IMD) service located at the Evelina London Children's Hospital. This service, led by Dr Mike Champion, is one of the largest in the UK with 5 dedicated IMD consultants and a team of dieticians and specialist nursing staff.

14. If using this form as an Additional Provider application, please explain why you wish to provide this test as it is already available from another provider.

N/A

EPIDEMIOLOGY

15a. Estimated prevalence of conditions in the general UK population

Prevalence is total number of persons with the condition(s) in a defined population at a specific time.

e.g. CF prevalence approx. 12 per 100,000 with UK population of approx. 63 million the prevalence of affected individuals in the UK is 7560

Please identify the information on which this is based.

For panel tests, please provide estimates for the conditions grouped by phenotypes being tested.

Prevalence is unknown

15b. Estimated annual incidence of conditions in the general UK population

Incidence is total number of new cases in a year in a defined population. e.g. CF incidence 1/2650 live births in a UK population with 724,000 live births in a year = 273 new cases a year

Please identify the information on which this is based.

For panel tests, please provide for groups of conditions.

Orphanet website quotes an estimated incidence of 1 in 20,000 - 70,000.Using the annual birth rate figure: if I in 70,000 would expect ~ 10; if 1in 20,000 would expect ~36

16. Estimated gene frequency (Carrier frequency or allele frequency)

Please identify the information on which this is based.

n/a for panel tests.

Extrapolating from information above the expected carrier frequency would be 1 in 70 to 1 in 130

17. Estimated penetrance of the condition. Please identify the information on which this is based

n/a for panel tests

All diagnostic referrals will have initial biochemical testing where possible. We would anticipate that pathogenic variants will be detected in >80% of diagnostic referrals.

18. Estimated prevalence of conditions in the population of people that will be tested.

n/a for panel tests.

All diagnostic referrals will have initial biochemical testing where possible which indicates a diagnosis of SPCD. In the 3 patients tested so far pathogenic variants were detected. We would anticipate that pathogenic variants will be detected in >80% of diagnostic referrals. This may be lower for cases suspected during newborn screening if available.

INTENDED USE (Please use the questions in Annex A to inform your answers)

19. Please tick either yes or no for each clinical purpose listed.

Panel Tests: a panel test would not be used for pre symptomatic testing, carrier testing and pre natal testing as the familial mutation would already be known in this case and the full panel would not be required.

Diagnosis	☒ **Yes**	☐ **No**
Treatment	☒ **Yes**	☐ **No**
Prognosis & management	☒ **Yes**	☐ **No**
Presymptomatic testing (n/a for Panel Tests)	☒ **Yes**	☐ **No**
Carrier testing for family members (n/a for Panel Tests)	☒ **Yes**	☐ **No**
Prenatal testing (n/a for Panel Tests)	☒ **Yes**	☐ **No**

TEST CHARACTERISTICS

20. Analytical sensitivity and specificity

The *analytical sensitivity* of a test is the proportion of positive results correctly identified by the test (true positive/true positive + false negative). The *analytical specificity* of a test is the proportion of negative results correctly identified by the test (true negative/true negative + false positive).
This should be based on your own laboratory data for (a) the specific test being applied for or (b) the analytical sensitivity and specificity of the method/technique to be used in the case of a test yet to be set up. Please specify any types of mutations reported to cause the clinical phenotype that cannot be detected by the test.
Note that the preferred threshold is ≥95% sensitivity (with 95% Confidence Intervals).

Sensitivity: Bi-directional Sanger sequencing of all exons and intron-exon boundaries would identify the vast majority of mutations currently listed on the public version of HGMD. An ACGS study indicated Sanger sequencing in combination with Mutation Surveyor software has a sensitivity of >99.59% sensitivity. Other mutation types are possible (e.g. large deletions, promoter region, intronic) however there is insufficient evidence to suggest additional testing is required.

21. Clinical sensitivity and specificity of test in target population

The *clinical sensitivity* of a test is the probability of a positive test result when condition is known to be present; the *clinical specificity* is the probability of a negative test result when disorder is known to be absent. The denominator in this case is the number with the disorder (for sensitivity) or the number without condition (for specificity).
Please provide the best estimate. UKGTN will request actual data after one year service.
For a panel test, the expected percentage diagnostic yield for the test in the target population can be presented as an alternative to clinical sensitivity and specificity?

Assuming the Biochemistry supports the diagnosis we would expect at least 90% pick up (Sanger sequencing will not detect gross deletions). (Data will be collected)

Sensitivity: Insufficient data: however sequence analysis can detect at least one mutation in approximately 70% of affected individuals. Large deletions and duplications of the SLC22A5 gene are not a common mechanism of causing CDSP, but have been detected in at least one individual with this diagnosis**.**

22. 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 condition or predisposition. It is measured by its *positive predictive value* (the probability of getting the condition given a positive test) and *negative predictive value* (the probability of not getting the condition given a negative test).
Not currently requested for panel tests

Most patients will have biochemical testing in the first instance.

23. Testing pathway for tests where more than one gene is to be tested sequentially

Please include your testing strategy if more than one gene will be tested and data on the expected proportions of positive results for each part of the process. Please illustrate this with a flow diagram. This will be added to the published Testing Criteria.

n/a for panel tests

N/A

CLINICAL UTILITY

24. How will the test change the management of the patient and/or alter clinical outcome? Please summarise in 2-3 sentences – no more than 50 words.

Confirmation of biochemical and clinical diagnosis to inform carrier testing and reproductive choices in these disorders which cannot be done on the basis of biochemical testing alone

Biochemical tests may be inconclusive in some cases, therefore molecular testing is essential for confirmation of the diagnosis. Diagnostic confirmation will enable appropriate treatment with carnitine to be given with a reduction in morbidity and mortality.

25. Please provide full description on likely impact on management of patient and describe associated benefits for family members. If there are any cost savings AFTER the diagnosis, please detail them here.

Confirmation of biochemical and clinical diagnosis to inform carrier testing and reproductive choices in these disorders which cannot be done on the basis of biochemistry alone.

26. If this test was not available, what would be the consequences for patients and family members? Please describe in not more than 50 of words.

Carrier testing is not available by biochemical testing. It would therefore not be possible to offer carrier testing to other family members.

Prenatal diagnosis: parents who have had an affected child may request a molecular prenatal test in subsequent pregnancies, so molecular testing is essential in the proband. The option of pre-implantation genetic diagnosis (PGD) can only be carried out if the mutations in the family are known.

27. 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 testing should be carried out as a first line test. However, biochemical testing may be inconclusive. Molecular testing can confirm the diagnosis. Enzyme analysis cannot be used for carrier testing.

28. What unexpected findings could this test show? For example, lung cancer susceptibility when testing for congenital cataract because ERCC6 gene (primarily associated with lung cancer) is included in a panel test for congenital cataract.

N/A

29. Please list any genes where the main phenotype associated with that gene is unrelated to the phenotype being tested by the panel.

N/A

30. If testing highlights a condition that is very different from that being tested for, please outline your strategy for dealing with this situation.

N/A

31. If a panel test, is this replacing an existing panel/multi gene test and/or other tests currently carried out by your lab e.g. Noonan Spectrum Disorders 12 Gene Panel replaced multigene Sanger test for KRAS, RAF1, PTPN11 and SOS1? If so, please provide details below.

N/A

32. Please describe any specific ethical, legal or social issues with this particular test.

N/A

33. REAL LIFE CASE STUDY
Please provide a case study that illustrates the benefits of this test

Molecular diagnosis for primary systemic carnitine deficiency aided the following case;
The index case presented aged 6 months with an acute collapse with cardiomyopathy and unsuccessful resuscitation. The child died and PM revealed features suggestive of an IMD.
A free carnitine level was extremely low and confirmatory genetic testing of the index case confirmed the diagnosis. Parental carrier status was confirmed.
This allowed family counselling for future pregnancies and to allow informed decision regarding prenatal testing. This is considered a treatable condition and the family opted for treatment and testing at birth.
The new baby was born and commenced on treatment.
Initial free carnitine levels measured pre-treatment were inconclusive. The diagnosis and plan for treatment was then dependent on molecular diagnosis. This confirmed the diagnosis within 1 week and the family were informed to continue treatment. The child will require continued follow up with an IMD service.
The ability to confirm the diagnosis in this case was paramount as the biochemical tests were inconclusive.

TESTING CRITERIA

34. Are previously approved Testing Criteria available that define the clinical entry point for this

test? No Testing Criteria are available: http://ukgtn.nhs.uk/find-a-test/testing-criteria/

If No, please complete template.

For NGS panel tests, please complete a form for each clinical entry point/subpanel as described in Q2

Please contact the UKGTN office if you are unsure whether testing criteria is available.

35. Please only complete this question if there is previously approved Testing Criteria.

Do you agree with the previously approved Testing Criteria? Yes/No N/A

36. If you do not agree, please provide revised Testing Criteria on the Testing Criteria form and explain below the reasons for the changes.

UKGTN Testing Criteria

Test name: **Carnitine Deficiency, Systemic Primary**		
Approved name and symbol of disorder/condition(s): Carnitine Deficiency, Systemic Primary; CDSP		**OMIM number(s):** **#212140**
Approved name and symbol of gene(s): solute carrier family 22 (organic cation transporter) member 5;	SLC22A5	**OMIM number(s):** ***603377**

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.**
Consultant in Inherited Metabolic Disease	
Consultant Clinical Geneticist	

Minimum criteria required for testing to be appropriate as stated in the Gene Dossier:	
Criteria	**Tick if this patient meets criteria**
Patient with low levels of free carnitine, suspicion of having SCPD	
OR at risk family members where familial mutation is known	

Additional Information:

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.

IS IT A REASONABLE COST TO THE PUBLIC?

37. Based on experience what will be the national (UK wide) activity, per annum, for:

Index cases 10

Family members where mutation is known 20

If a NGS panel test, it is recognised that the full panel will not be used to test family members where the familial mutation is known. Please provide expected number of tests to inform completion of Q40

38. If your laboratory does not have capacity to provide the full national need please suggest how the national requirement may be met.

For example, are you aware of any other labs (UKGTN members or otherwise) offering this test to NHS patients on a local area basis only? This question has been included in order to gauge if there could be any issues in equity of access for NHS patients. If you are unable to answer this question please write "unknown".

This laboratory has sufficient capacity to support the expected number of tests.

39. In order to establish the potential costs/savings that could be realised in the diagnostic care pathway, please list the tests/procedures that are no longer required to make a diagnosis if the index case has a definitive molecular genetic diagnosis from the test proposed in this gene dossier.

	Type of test	Cost (£)
Imaging procedures		
Laboratory pathology tests (other than molecular/cyto genetic test proposed in this Gene Dossier)		
Physiological tests (e.g. ECG)		
Other investigations/procedures (e.g. biopsy)		
Total cost of tests/procedures no longer required (please write n/a if the genetic test does not replace any other tests procedures in the diagnostic care pathway)		

40. Based on the expected annual activity of index cases (Q37), please calculate the estimated annual savings/investments based on information provided in Q39.

Number of index cases expected annually	10
Cost to provide tests for index cases if the genetic test in this Gene Dossier was not available (see Q39)	(b)
Total annual costs pre genetic test submitted for evaluation in this Gene Dossier	
Total annual costs to provide genetic test	10 x £400 (cost of genetic testing for index case) = **£4000**
Additional investment for 100% positive rate for index cases	**£4000**
Percentage of index cases estimated to be negative	(f)
Number of index cases estimated to be negative	(f) x number of index cases = (g)
Costs to provide additional tests for index cases testing negative	(g) x (b) = (h)
Total investment for tests for index patient activity	**£4000**
Total costs for family members	Costs for family member test £160 x 20 (number of family members expected to test in a year) = **£3200** (j)
If there is a genetic test already available and some of the family testing is already being provided, please advise the cost of the family testing already available	Cost for family member testing already available x estimated number of tests for family members already provided (k)
Total costs for family members minus any family member testing costs already provided	(j) – (k) = (l)
Additional costs for all activity expected in a year	£4000 + £3200 **= £7200**

41. Please indicate the healthcare outcomes that apply to this test after diagnosis. It is recognised that all tests recommended by the UKGTN for NHS service improve clinical management and, if a familial mutation is found, allows for prenatal testing and therefore these are not included in the list below.

Healthcare outcomes	Does this apply to this test?
1. Genetic testing alerts significant clinical co-morbidities	No
2. Reduced mortality/saves lives	No
3. Avoids diagnostic invasive procedures/tests and associated in patient episodes	No
4. Confirms targeted therapy	Yes
5. Earlier diagnosis avoiding multi hospital appointments /procedures	No
6. Avoids irreversible harm	Yes
7. Enables access to educational and social support	No
8. At risk family members that test negative for a familial mutation can be discharged from follow up	Yes
9. At risk family members that test positive have appropriate follow up	Yes