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	Hemihypertrophy (isolated) Hemihyperplasia (isolated) Hemihypertrophy, or hemihyperplasia, describes an asymmetric overgrowth of a limb or one side of the body. It can occur as part of a wider syndrome, where it is associated with other clinical features, or in isolation. The latter diagnosis is only made after a thorough clinical review has excluded the presence of any associated clinical features.
OMIM number for disease	OMIM 235000
Gene – name and description (please provide any alternative names you wish listed)	H19 (ASM1) IGF2 (somatamedin A) CDKN1C (p57; KIP2) LIT1 (KCNQ10T1)
OMIM number for Gene	H19- 103280 IGF2- 147470 CDKN1C- 600856 LIT1- 604115
Mutational spectrum for which you test	● loss of methylation at LIT1 (KvDMR1 within LIT1) ● hypermethylation of H19 ● duplication of paternally inherited 11p15 region ● paternal uniparental disomy of 11p15 region ● maternal H19 DMR microdeletion ● maternal KvDMR1 microdeletion ● loss of methylation of H19 ● maternal duplication 11p15
Technical Method (s)	1. Methylation-specific multiplex ligation-dependent probe amplification (MS-MLPA) 2. 11p15 microsatellite analysis to confirm uniparental disomy in positive cases identified by MS-MLPA 3. Methylation-specific high resolution melt analysis as back-up confirmation for MS-MLPA
Validation Process **Note please explain how this test has been validated for use in your laboratory)**	We have validated the use of the assays through the blinded analysis of a "test set" of 51 samples (24 normal and 27 abnormal). All samples were identified correctly (Scott et al 2008, attached) Analysis has subsequently been extended to include a total of 200 normal control individuals. We are yet to observe a false positive result. This validation work has been carried out in Prof Rahman's laboratory at ICR, Sutton. Our analyses indicate that MS-MLPA assay is capable, in a single experiment, of detecting all of the 11p15 epigenetic and copy number defects recognised in overgrowth and growth retardation orders. Unlike the existing assays of methylation in the region, it differentiates heritable copy number abnormalities from isolated methylation defects. In addition, the use of MS-MLPA as a first-line assay limits the need for microsatellite analysis to the small number identified with probable UPD. Compared with existing diagnostic testing approaches, this approach therefore both broadens the range of detectable 11p15 abnormalities and reduces the complexity and cost of analysis of the region. In addition we are currently validating the use of a second method (MS-HRM) in the diagnostic lab for confirming positive cases detected by MS-MLPA or for fails.

Are you providing this test already? If yes, how many reports have you produced? **NB please give the number of mutation positive/negative samples you have reported**	No
For how long have you been providing this service?	Not applicable
Is there specialised local clinical/research expertise for this disease?	**Yes** (circled) | **No** | **Please provide details** There is both clinical and research expertise locally as Professor Rahman, whose group (the Childhood Overgrowth (COG) Study) at the nearby Institute of Cancer Research optimised the 11p15 MS-MLPA, is also an Honorary Consultant at St George's. Dr Kate Tatton Brown also has extensive clinical and research expertise in the area of overgrowth.
Are you testing for other genes/diseases closely allied to this one? Please give details	SW Thames Regional Molecular Genetics Diagnostic Laboratory is currently offering NSD1 testing for Sotos syndrome, another overgrowth condition. In addition, the COG study aims to identify novel overgrowth genes and their associated phenotypes. It is envisaged that tests for novel genes, shown to be significant contributors to human overgrowth, will also be offered in the St George's laboratory in the future.
Your Activity How many tests do you (intend to) provide annually in your laboratory?	Approximately 20 tests
Based on experience how many tests will be required nationally? Please identify the information on which this is based	Approximately 40 tests. Information based on number of tests performed in laboratories that are offering testing for these conditions by different techniques.

Epidemiology

Estimated prevalence of disease in the general UK population Please identify the information on which this is based	1 in 13,000 to 80,000 (Higushi et al 1980); 11p15 defects are present in 10-20% of cases (Shuman et al 2006).
Estimated gene frequency (Carrier frequency or allele frequency) Please identify the information on which this is based	No population-based data are available. Based on data from disease-based studies (above), it is estimated that the frequency of 11p15 defects in the general population is 1 in 10,000 – 20,000.
Estimated penetrance Please identify the information on which this is based	Based upon data on large numbers of individuals with overgrowth and 11p15 defects, it is likely that penetrance approaches 100% (Weksberg et al, 2006). However, similar large scale studies have not been undertaken in isolated hemihypertrophy but to date, no unaffected individual has been reported with an 11p15 defect (Netchine et al 2007).
Target Population The essential clinical or family history features defining the target population must be described. (C)-Testing Criteria	Individuals with hemihypertrophy/growth asymmetry
Estimated prevalence of disease in the target population	See prevalence in general population box

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

Please tick the relevant clinical management criteria that his test effects.	**YES**	**NO**
Diagnosis	√	
Treatment		√
Prognosis & Management	√	
Presymptomatic testing	√	
Risk Assessment	√	

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.	The validation data presented above indicate that the MS-MLPA test has analytical sensitivity and specificity approaching 100%.
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) *Positive predictive value* and *penetrance* are notionally equivalent for any single genetic allele – the probability of developing disease given a positive test. The relationship is much more complex if more than one gene is responsible for the disease (locus heterogeneity), or if in any one gene there are multiple alleles (allelic heterogeneity), unless all the alleles are tested. In these cases, there are implications for the *clinical sensitivity* of the test and for *its negative predictive value*. For example, for a disease (such as APKD) that may be caused by either of two separate genes, even if each is 100 percent penetrant, the *clinical sensitivity* and the *negative predictive value* (and *clinical validity*) will both be reduced: *clinical sensitivity* since its maximum value can be no greater than the proportion of the disease that is caused by that particular gene, and *negative predictive value* since a negative test on Gene A will be no guarantee that the patient will not develop the phenotype, because the disease may be caused by Gene B. A similar form of analysis may be applied to genes with multiple alleles unless the "test" measures all the alleles	**Clinical Sensitivity** The test is capable of detecting all 11p15 defects reported in association with isolated hemihypertrophy. Clinical sensitivity is therefore estimated at 10-20%, including all cases with reported 11p15 defects. **Clinical Specificity** The validation data presented above indicate that the test has clinical specificity approaching 100%.

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 *Positive predictive value* and *penetrance* are notionally equivalent for any single genetic allele – the probability of developing disease given a positive test. The relationship is much more complex if more than one gene is responsible for the disease (locus heterogeneity), or if in any one gene there are multiple alleles (allelic heterogeneity), unless all the alleles are tested. In these cases, there are implications for the *clinical sensitivity* of the test and for *its negative predictive value*. For example, for a disease (such as APKD) that may be caused by either of two separate genes, even if each is 100 percent penetrant, the *clinical sensitivity* and the *negative predictive value* (and *clinical validity*) will both be reduced: c*linical sensitivity* since its maximum value can be no greater than the proportion of the disease that is caused by that particular gene, and *negative predictive value* since a negative test on Gene A will be no guarantee that the patient will not develop the phenotype, because the disease may be caused by Gene B. A similar form of analysis may be applied to genes with multiple alleles unless the "test" measures all the alleles.	Positive predictive value / penetrance Published data indicate that 11p15 defects detectable with the test may be fully penetrant and have a positive predictive value approaching 100%. Negative predictive value Not applicable. 11p15 analysis is used principally to confirm diagnosis and/or to inform offspring/recurrence risks and estimate Wilms tumour risk rather than to rule out an underlying diagnosis (see clinical utility section). In the case of a negative test result, patients/families are counselled according to empiric risks for individuals in their phenotypic group with normal 11p15 status. In rare families with known heritable 11p15 defects, eg H19 microdeletion, predictive testing can be used in at-risk individuals. In this context, negative predictive value is very high (approaching 100%).

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? 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 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 Are there specific ethical, legal or social issues with this test?	**Testing criteria** *Individuals with hemihypertrophy/growth asymmetry.* **Referring clinicians** Clinical Geneticist (consultant or specialist registrar) Consultant Paediatrician Consultant in fetal medicine / obstetrics **Outcome** 1. Abnormality detected a. Molecular confirmation of diagnosis b. MS-MLPA distinguishes heritable from non-heritable 11p15 defects and enables accurate estimation of recurrence and offspring risks. c. The correct classification of the 11p15 abnormality allows targeting of Wilms tumour surveillance to patients at risk. Certain 11p15 abnormalities are associated with an increased Wilms tumour risk (UPD, 11p15 duplication or hypermethylation at H19) while KvDMR1 loss of methylation is not. Children with KvDMR1 loss of methylation do not require Wilms tumour screening. Avoidance of unnecessary screening in these children, reduces the large financial burden to the NHS and a source of much parental anxiety (Scott et al 2006). 2. No abnormality detected a. Following a negative 11p15 test result, patients/families can be counselled according to empiric risks for individuals in their phenotypic group with normal 11p15 status. For example, the risk of Wilms tumour is not greatly elevated in individuals with isolated hemihypertrophy and a normal 11p15 result. Families of such children can be reassured and unnecessary surveillance avoided (Scott et al 2006). b. Where there is a known heritable 11p15 defect in a family, a negative test result in an 'at-risk' relative allows the reassurance and appropriate counselling of the patient and the avoidance of unnecessary surveillance. There are no ethical, legal or social issues specific to this test.

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

Referral Pathway Template –

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

TARGET POPULATION

Individuals with hemihypertrophy/growth asymmetry.

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

1. Clinical Genetics- Consultant and Specialist registrar
2. Paediatrics- Consultant

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

Testing criteria will be advertised on UKGTN website. After one year the service will be audited with referring clinicians contacted. The types of cases being referred will be analysed and criteria amended if appropriate.

HOW MANY TESTS DO YOU EXPECT TO PERFORM ANNUALLY?

20

UKGTN Testing Criteria:

Name of Disease(s): HEMIHYPERPLASIA, ISOLATED; IH (235000)

Name of gene(s):
H19, imprinted maternally expressed transcript - H19 (103280)
insulin-like growth factor 2 (somatomedin A) - IGF2 (147470)
cyclin-dependent kinase inhibitor 1C (p57, Kip2) - CDKN1C (600856)
KCNQ1 overlapping transcript 1 - KCNQ1OT1 (604115)

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 Geneticist	
Consultant Paediatrician	

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

Criteria	Tick if this patient meets criteria
Isolated hemihypertrophy/growth asymmetry.	

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.