NHS

UK Genetic Testing Network

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

Test – Disease – Population Triad

Disease – name	MELANOMA, CUTANEOUS MALIGNANT; CMM
OMIM number for disease	155600
Disease – alternative names please provide any alternative names you wish listed	Familial Atypical Multiple Mole Melanoma (FAMMM) Familial melanoma Malignant melanoma
Disease – please provide a brief description of the disease characteristics	Melanoma arises from melanocytes and may arise *de novo* or from pre-existing skin lesions. Families with germline mutations in CDKN2A and CDK4 are at increased risk of melanoma and pancreatic cancer.
Disease - mode of inheritance	Autosomal Dominant
Gene – name(s)	cyclin-dependent kinase inhibitor 2a (cdkn2a) cyclin-dependent kinase 4 (cdk4)
OMIM number for gene(s)	cyclin-dependent kinase inhibitor 2A (melanoma, p16, inhibits CDK4); CDKN2A (600160) cyclin-dependent kinase 4; CDK4 (123829)
Gene – alternative names please provide any alternative names you wish listed	CDKN2A Alternative names: TP16, CDKN2, p14(ARF), MTS1, p16(INK4) and p16 (INK4A).
Gene – description(s) (including number of amplicons).	4 amplicons representing the entire coding region of CDKN2A (exons 1α, 1β, 2, and 3) 1 amplicon CDK4 (exon 2)
Mutational spectrum for which you test including details of known common mutations.	Point mutations, small deletions and insertions, large deletions.
Technical Method (s)	Bidirectional sequencing of 5 amplicons covering the open reading frame followed by analysis of results using Mutation Surveyor software to detect point mutations in CDKN2A and CDK4. MLPA analysis to detect large deletions in CDKN2A.
Validation Process Note: please explain how this test has been validated for use in your laboratory	For all analyses all primer sequences are checked for the presence of single nucleotide polymorphisms and Blasted using appropriate resources at Ensembl and NCBI. Sequence information obtained is checked against the reference sequences available at Ensembl and NCBI.
Are you providing this test already? If yes, how many reports have you produced?	Confirmation of research findings: 6 reports (all positive) Predictive testing: 26 reports (12 positive/14 negative)

Please give the number of mutation positive/negative samples you have reported	Full screen 6 reports (1 positive/5 negative)
For how long have you been providing this service?	Confirmation of mutations found in research laboratory and predictive testing since December 2002. Mutation screening since January 2009.
Is there specialised local clinical/research expertise for this disease?	**Yes** | **Please provide details** Professor Julia Newton Bishop has worked on Familial Melanoma for 25 years. Her research group is involved in various research projects and is part of the GenoMEL consortium. The original DNA laboratory service confirmed mutations found by Professor Newton-Bishop's research group, before taking the screening on as a diagnostic service in January 2009.
Are you testing for other genes/diseases closely allied to this one? Please give details	The Yorkshire Regional DNA laboratory offers a diagnostic service for other familial cancer syndromes including p53, BRCA1&2 and HNPCC.
Your Activity If applicable - How many tests do you currently provide annually in your laboratory?	Index cases: To date: 8 national referrals (over 4 month period) Family members where mutation is known: 4 cases per annum (average figure over past 6 years)
Your Activity How many tests will you be able to provide annually in your laboratory if this gene dossier is approved and recommended for NHS funding?	Index cases: At least 30 per annum with current resources Family members where mutation is known: At least 20 per annum with current resources.
Based on experience how many tests will be required nationally (UK wide)? Please identify the information on which this is based	Index cases: approximately 25 per annum (based on current referral rate to diagnostic laboratory and referral rate to Professor Julia Newton-Bishop's research study) Family members where mutation is known: approximately 5 referrals per annum based on current referral patterns to the diagnostic laboratory.
National Activity **(England, Scotland, Wales & Northern Ireland)** If your laboratory is unable to provide the full national need please could you provide information on 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. It is appreciated that some laboratories may not be able to answer this question. If this is the case please write "unknown".	We should be able to meet the full national need.

Epidemiology

Estimated prevalence of disease in the general UK population Please identify the information on which this is based	The incidence of melanoma in the UK is approximately 10 cases per 100, 000 per annum. Roberts et al. (1992) UK Guidelines for the management of cutaneous melanoma. British Journal of Dermatology 2002 **146** 7-17. Less than 1-2% of melanoma cases are thought to be attributable to mutations of the melanoma susceptibility genes (CDKN2A and CDK4) Melanoma Genetic Consortium Consensus Statement
Estimated gene frequency (Carrier frequency or allele frequency) Please identify the information on which this is based	Unknown
Estimated penetrance Please identify the information on which this is based	Figures differ between population based studies and those based on multiple-case families. There is also significant geographical variation. In the population study by Begg et al (1) the risk of melanoma in CDKN2A mutation carriers was 14% by the age of 50 years rising to 28% by the age of 80. In a study of high risk families by Bishop et al (2) the risk for European carriers was 13% by the age of 50 years rising to 58% by the age of 80. Penetrance data for codon 24 CDK4 mutations is not well established as it is based on small numbers. Goldstein et al (3) estimated the penetrance of the p.Arg24Cys mutation (2 families) to be 63% and the penetrance of p.Arg24His mutation to be 59% (one family). 1. Begg et al (2005) Life-time Risk of Melanoma in CDKN2A Mutation Carriers in a Population based Sample. J. Natl. Cancer. Inst. **97** p1507-1515 2. Bishop et al (2002) Geographical variation in the penetrance of CDKN2A. J. Natl. Cancer. **94** p894-903. 3. Goldstein et al (2002) Rariety of CDK4 germiline mutations in familial melanoma. Melanoma Research **12** p51-55
Target Population Description of the population to which this test will apply (i.e. description of the population as defined by the minimum criteria listed in the testing criteria)	a) Individuals from families with 3 or more cases of melanoma. b) Individuals from families with 2 cases of melanoma in first degree relatives, with multiple primary melanoma in at least one case. c) Individuals from families with 1 case of melanoma and pancreatic cancer in a first degree relative.

Estimated prevalence of disease in the target population	a) The frequency of mutations in families with 3 or more cases of melanoma is approximately 57% (Goldstein et al [2007]; JNB data). b) The frequency of mutations in pedigrees with 2 cases of melanoma, with multiple primaries in at least one case is 25% (7/28 pedigrees) [~15% in 2 case pedigrees overall] (JNB data). c) The frequency of mutations in families with 1 case of melanoma and pancreatic cancer in a first degree relative has not yet been established. Goldstein et al [2007] Features associated with germline CDKN2A mutations: a GenoMEL study of melanoma-prone families from three continents. J. Med. Genet **44** 99-106 JNB data – data from Julia Newton Bishop's group

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

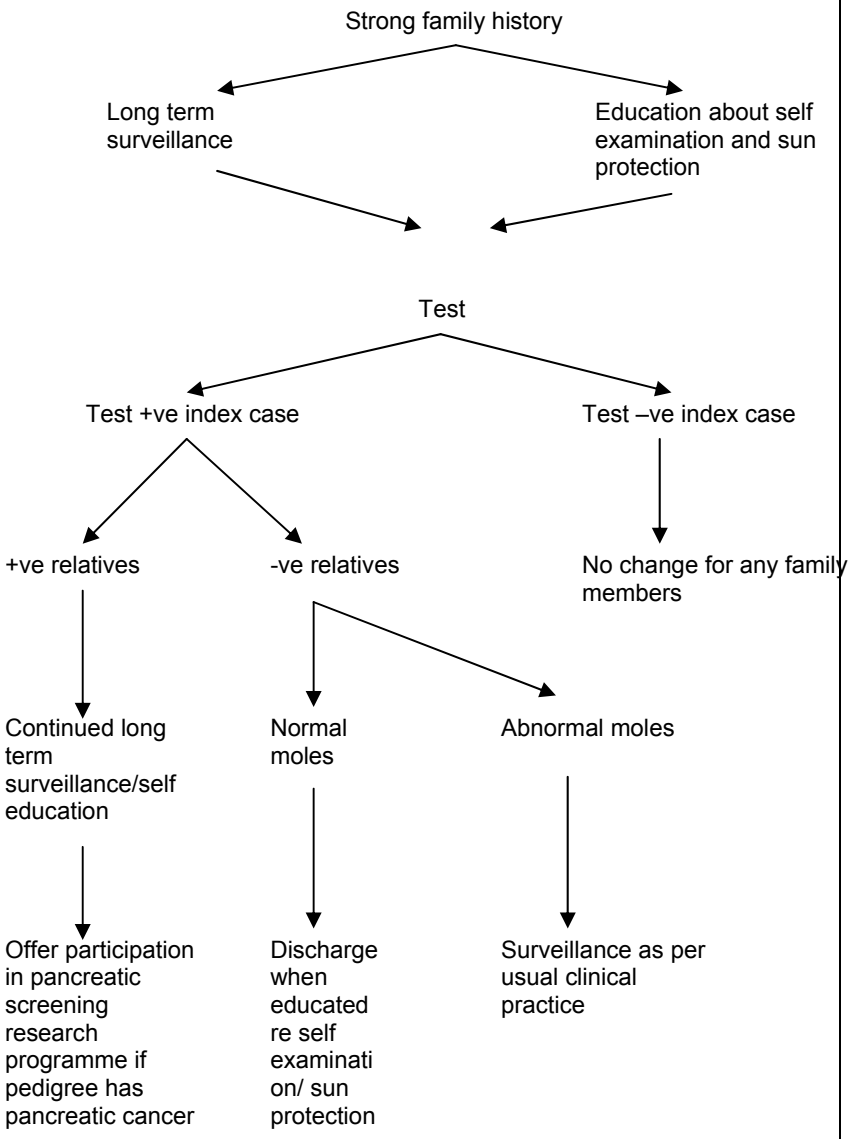
Please tick the relevant clinical purpose of testing	**YES**	**NO**
Diagnosis		√
Treatment		√
Prognosis & Management	√	
Presymptomatic testing	√	
Risk Assessment for family members	√	
Risk Assessment – prenatal testing		√

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 more than one gene will be tested, please include your testing strategy and data on the expected proportions of positive results for each part of the process. Please illustrate this with a flow diagram.	**Level 1** Bidirectional sequencing – point mutations ➢ 4 cases of melanoma in the family – 63% cases found to have pathogenic mutation* ➢ 3 cases of melanoma in the family – 21% cases have pathogenic mutation* ➢ 2 cases of melanoma in the family – 13% of cases have pathogenic mutation* *Unpublished data provided by Julia Newton Bishop's laboratory ↓ **Level 2** MLPA analysis – deletions Predicted that detection rate would be below 2% (Mistry et al 2005) Bidirectional sequencing of amplicons followed by analysis of results using Mutation Surveyor software has a sensitivity of between 95% and 99% in our laboratory. We will offer a targeted approach to mutation identification in affected patients. In the first instance we will use bidirectional sequencing of *CDKN2A* and *CDK4* (exon 2) to detect point mutations followed by the multiple ligation probe assay (MLPA) to detect deletions at 9p21.
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)	Clinical sensitivity will be dependant on the number of affected relatives within the family and increases as there are more cases of melanoma in the family (see prevalence section). The clinical specificity is not known. Approximately 20% of all mutations identified are unclassified variants, potentially impacting on the sensitivity and specificity of the test.
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	**Positive Predictive value** Figures differ between population based studies and those based on multiple-case families. There is also significant geographical variation. In the population study by Begg et al (2005) the risk of

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).	melanoma in CDKN2A mutation carriers was 14% by the age of 50 years rising to 28% by the age of 80. In a study of high risk families by Bishop et al (2002) the risk for European carriers was 13% by the age of 50 years rising to 58% by the age of 80. **Negative Predictive value** The risk of melanoma for individuals from families with 3 or more cases of melanoma is that they likely have other moderate to high risk genetic mutations which are not yet identified. Melanoma risk to relatives of melanoma cases are based upon registry data: the risk to first degree relatives of 1 cases is twice the population risk and 8 times when 2 first degree relatives have melanoma but there are no data on mutation testing in these individuals.

Clinical utility of test in target population (Please refer to Appendix A) Please provide a description of the clinical care pathway.	Patients with a pathogenic mutation require lifelong surveillance and possible participation in pancreatic cancer screening research programmes if pancreatic cancer present in the pedigree. Patients from positive families with a negative mutation test would normally be discharged after education regarding sun protection. However, those with atypical moles would need long term follow up. For families where no pathogenic mutation was detected there would be no change to clinical followup.
How will the test add to the management of the patient or alter clinical outcome?	A recent paper indicates that reporting CDKN2A/p16 test results to high risk patients significantly improves their compliance with early detection recommendations. Aspinwall et al. (2008) Cancer Epidemiol Biomarkers Prev 17 (6) p1510- 1519.

	 The flow chart shows that a negative test in a family with a known mutation may be discharged from follow up when competent at self examination
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?	As above it will have a small effect by reducing the need to follow up in a small number of patients.
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	If the patient does not undergo genetic testing their management will be determined using family history.

Please describe any specific ethical, legal or social issues with this particular test?	Some individuals have expressed concern that Individuals testing negative will fail to take prevention measures, but there are no data to suggest this will be the case here.

Please complete the testing criteria form.

UKGTN Testing criteria

Name of Disease(s): MELANOMA, CUTANEOUS MALIGNANT; CMM (155600)

Name of gene(s): cyclin-dependent kinase inhibitor 2A (melanoma, p16, inhibits CDK4); CDKN2A (600160)
cyclin-dependent kinase 4; CDK4 (123829)

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	

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

Criteria	Tick if this patient meets criteria
Affected individuals in families with 3 or more cases with primary invasive melanoma.	
OR Affected individual in families with 2 cases of melanoma in first degree relatives with multiple primary melanoma in at least one case.	
OR Affected individual in families with at least one invasive melanoma and two or more other diagnoses of invasive melanoma and/or pancreatic cancer among first- or second-degree relatives on the same side of the family.	

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.