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 and description (please provide any alternative names you wish listed)	Retinitis Pigmentosa (including X-linked recessive, autosomal dominant and autosomal recessive forms)
OMIM number for disease	Retinitis Pigmentosa (OMIM 268000)
Gene – name and description (please provide any alternative names you wish listed)	RETINITIS PIGMENTOSA GTPase REGULATOR; RPGR RETINITIS PIGMENTOSA 2, X-LINKED; RP2 RHODOPSIN; RHO RETINAL DEGENERATION GENE, SLOW; RDS; RDS/peripherin PRECURSOR mRNA-PROCESSING FACTOR 31; PRPF31 PRECURSOR mRNA-PROCESSING FACTOR 8; PRPF8 NEURAL RETINA LEUCINE ZIPPER; NRL IMP DEHYDROGENASE 1; IMPDH1 PDGFA-ASSOCIATED PROTEIN 1; PDAP1; PAP1 RETINITIS PIGMENTOSA 1 GENE; RP1; ORP1
OMIM number for Gene	RPGR(OMIM 312610), RP2(OMIM 312600), Rhodopsin(OMIM 180380), RDS/peripherin (OMIM 179605), PRPF31 (OMIM 606419), PRPF8 (OMIM 607300), NRL (162080), IMPDH1 (146690), PAP1 (607075), ORP1 (603937).,
Mutational spectrum for which you test	A mutation screening cascade has been developed to maximise detection rate and cost efficiency. The screen detects point mutations and small insertion/deletion variants. Whole exon copy number variants are not included in the screen as they are infrequently described in the clinical spectrum represented by RP.
Technical Method (s)	The mutation screening cascade is described in a separate appendix
Validation Process **Note please explain how this test has been validated for use in your laboratory)**	There are no reference materials available to act as validation controls. A series of patients ascertained through research testing have been repeat tested in developing the diagnostic system. Of 16 patients with previously identified mutations in ORF15 we were able to confirm all 16. For the ADRP arm of the cascade, the new technology of Pyrosequencing was validated using 2 positive samples for each mutation, confirming each by conventional sequencing.
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**	The service has been developed and piloted through a National Genetics Reference Laboratory project (2002-2006). During this period 651 clinical reports have been issued.
For how long have you been providing this service?	Reports have been issued since 2003

<table>
<tr><td>Is there specialised local clinical/research expertise for this disease?</td><td>Yes √</td><td>No</td><td>Please provide details Ophthalmic Genetic expertise is available through a collaboration between the Regional Clinical Genetics Service, Manchester Royal Eye Hospital and Moorfields Eye Hospital.</td></tr>
<tr><td>Are you testing for other genes/diseases closely allied to this one? Please give details</td><td colspan="3">The service includes testing for X-linked RP, Sorsby Fundus Dystrophy, Late Onset Retinal Dystrophy, Autosomal Dominant Drusen and Macular Dystrophy.</td></tr>
<tr><td>Local Activity
How many tests do you intend to provide annually in your laboratory?</td><td colspan="3">
<table>
<tr><td>Year</td><td colspan="2">Disease</td></tr>
<tr><td></td><td>XLRP</td><td>ADRP</td></tr>
<tr><td>2003</td><td>32</td><td>-</td></tr>
<tr><td>2004</td><td>91</td><td>-</td></tr>
<tr><td>2005</td><td>195</td><td>94</td></tr>
<tr><td>2006</td><td>146</td><td>93</td></tr>
</table>
Table showing the increase in the number of reports over the last three and a half years. We would estimate that the number of reports will continue to be around 250 per year on average over the next two years.
</td></tr>
<tr><td>National Activity
How many tests are being provided nationally?</td><td colspan="3">250 (through this pilot project)</td></tr>
<tr><td>Based on experience how many tests will be required nationally?
Please identify the information on which this is based</td><td colspan="3">We consider that the current situation represents the steady state with around 250 referrals per year (comprising ADRP and XLRP).</td></tr>
</table>

Epidemiology

Estimated prevalence of disease in the general UK population Please identify the information on which this is based	1:3500-4000 in USA and Europe. (OMIM 268000 ; Genetics for Ophthalmologists-G.C.M. Black) RP is highly heterogeneous and this figure includes all inheritance patterns and sporadic cases
Estimated gene frequency (Carrier frequency or allele frequency) Please identify the information on which this is based	It is not possible to calculate gene frequencies in this highly heterogeneous condition.
Estimated penetrance Please identify the information on which this is based	Penetrance is variable between X-linked and autosomal dominant forms as well as between genes. In X-linked RP the penetrance is generally high but can vary in females due to X-inactivation. In autosomal dominant RP the penetrance is high except for those mutations found in PRPF31, PAP1 and PRPF8 which are well documented in the literature to display variable penetrance.
Target Population Please provide details on which population the test is going to be used. For example a description of the particular patient group (see Note 1 below).	Ethnic specific phenotypic and genotypic differences have not been found in X-linked and AD form of RP. Referrals of index cases are accepted according to the following criteria which define the target groups: The families of patients referred for XLRP testing must display a clear X-linked inheritance. We are currently investigating the involvement of mutations in the ORF15 region of RPGR in male cases of sporadic RP. Around 10% of sporadic cases have an ORF15 mutation. The families of patients referred for dominant RP testing must display clear dominant inheritance with affected individuals in more than one generation.
Estimated prevalence of disease in the target population	The target population is defined by the RP phenotype, e.g. only cases with a high index of suspicion (ascertained by clinical geneticists or collaborating ophthalmologists) and a high probability of carrying an RP mutation within the screening cascade are offered mutation scanning.

Note 1- The characteristics of the population for the test should be stated. Features such as ethnicity, age range, and affected families are all appropriate examples of population characteristics. The concept of a “test” includes the population in which it is to be applied, so, for example, a test for the Huntington gene evaluated for use in asymptomatic family members would receive a separate evaluation if it were additionally proposed that it should be used in the general population.

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

Please tick the relevant clinical management criteria that this 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.	On the basis of re-sequencing of positive controls we estimate that the analytical sensitivity and specificity of the techniques used (point mutation screen, mini-sequencing and Sanger sequencing) will be greater than 98%.
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: c*linical sensitivity* since its maximum value can be no greater than the	A pathogenic variant is found in 55% of males with a family history of RP and in 7% of simplex cases. The interpretation of variants in X-linked cases is straightforward as most are frame-shift or nonsense mutations or are mis-sense changes previously described in the literature. The remainder are usually resolved by a family study. Unclassified variants may be counted as potential false positive results. These account for approximately 17% of variants found in the whole cascade. The minimum clinical specificity is therefore 83%.

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 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 and negative predictive value: There is insufficient data to make a reliable estimate of positive and negative predictive values in the target population. Locus heterogeneity dictates that a negative screen in an affected patient does not exclude a genetic mutation as the cause of the condition. However individual mutations in the target group have been described with a high penetrance and within a family where a mutation has been firmly associated with the condition through its inheritance the negative and positive predictive values are >99%.

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 is required to illustrate the clinical utility of the test (a template is provided on page 8). 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 **Please complete the referral pathway diagram on the following page. Include a testing criteria form that may be available to clinicians when assessing merit of testing where possible.**	**Clinical utility of genetic testing for retinitis pigmentosa.** **X-linked retinitis pigmentosa.** X-linked retinitis pigmentosa is one of the more severe forms of RP and accounts for around 20% of RP (Rivolta, 2002). Mutations in 2 genes (RPGR and RP2) account for perhaps 75-90% of all X-linked RP. Mutations in these genes also cause X-linked cone-rod dystrophy (CRD), cone dystrophy (COD) and early onset severe retinal dystrophy (EOSRD). In general males with disease present early and are significantly disabled by the condition during adulthood. By contrast females carriers are more mildly affected and often develop symptoms in middle life or later. Clinical carrier testing (e.g. electrodiagnostic testing) will pick up many carriers but cannot be relied upon in early adult-hood, in particular during reproductive years. The implications of X-linked RP are therefore similar to other X-linked disorders: • Asymptomatic young females, in particular those with experience of their fathers/brothers with disease, are often anxious to know their carrier status. Since this cannot be provided with accuracy from clinical testing, carrier testing is therefore often sought by these females. o As a measure of clinical utility we now have experience of testing in 210 probands with a family history consistent with XLRP. Subsequently carrier tests were performed for 93 females and in 58 (62%) a positive carrier status was confirmed. o Asymptomatic young females, in whom carrier status is excluded are able to be discharged from follow-up. o Females carrying a mutation may be offered PND – indeed we have already had a number of requests for PND while at least one family has been offered PGD for XLRP. o Given the increasing evidence of symptoms in females this test should be regarding as a presymptomatic test as well as a carrier test. • Asymptomatic males – even those who have been reassured that they are unlikely to carry a mutation – are anxious to have a formal genetic confirmation that they do not carry mutations. Males without a mutation can be excluded from follow-up. o We have already undertaken 18 pre-symptomatic tests. o The age at which males should be tested is debatable – however, families are often keen to ensure that boys are diagnosed early since this impacts upon education, training, life choices (e.g. career choices) and management. For this reason examination, or genetic testing, under the age of 5

	is often requested and is considered. • Symptomatic males – where males with symptoms are tested there is unlikely to be an alteration in their care pathway. • Symptomatic females – o Genotype-phenotype correlation. There is emerging evidence that females have a strong likelihood of developing symptoms and that this is under the influence of mutation type. However at present this has no proven predictive value. **Clinical utility of genetic testing for retinitis pigmentosa.** **Autosomal dominant retinitis pigmentosa.** AD retinitis pigmentosa is a progressive, heterogeneous condition accounting for 15—25% of RP (Rivolta, 2002) with mutations in ~10 genes. The implications of ADRP are therefore similar to other progressive autosomal dominant and X-linked disorders. A number of forms including RP9 and RP11 shown reduced expressivity /penetrance • Asymptomatic individuals, in particular those with experience of affected first degree relatives with disease, are often anxious to know their carrier status. Since this cannot be provided with accuracy from clinical testing, carrier testing is often sought by these individuals. • o As with other AD conditions, asymptomatic individuals, in whom carrier status is excluded are able to be discharged from follow-up. o Presymptomatic testing via clinical examination is requested in some families with later onset AD disease – it is likely that this will be requested in some families and will need to be managed as for other presymptomatic tests. • Affected individuals will often be tested in order to allow screening of the wider family. There is unlikely to be an alteration in the care pathway for affected individuals. Rivolta C, Sharon D, DeAngelis MM, Dryja TP (2002) Retinitis pigmentosa and allied diseases: numerous diseases, genes, and inheritance patterns. Hum Mol Genet. **11**:1219-27.

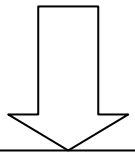
Referral Pathway Template –

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

TARGET

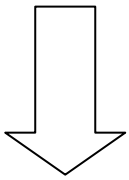
POPULATION

Index cases and relatives at risk of X-linked, autosomal dominant and sporadic Retinitis Pigmentosa



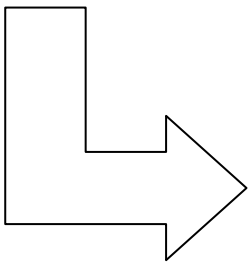
WHAT TYPE AND LEVEL OF PROFESSIONAL OR REFERRER DO YOU ACCEPT SAMPLES FROM?

Ophthalmologists and Clinical Geneticists



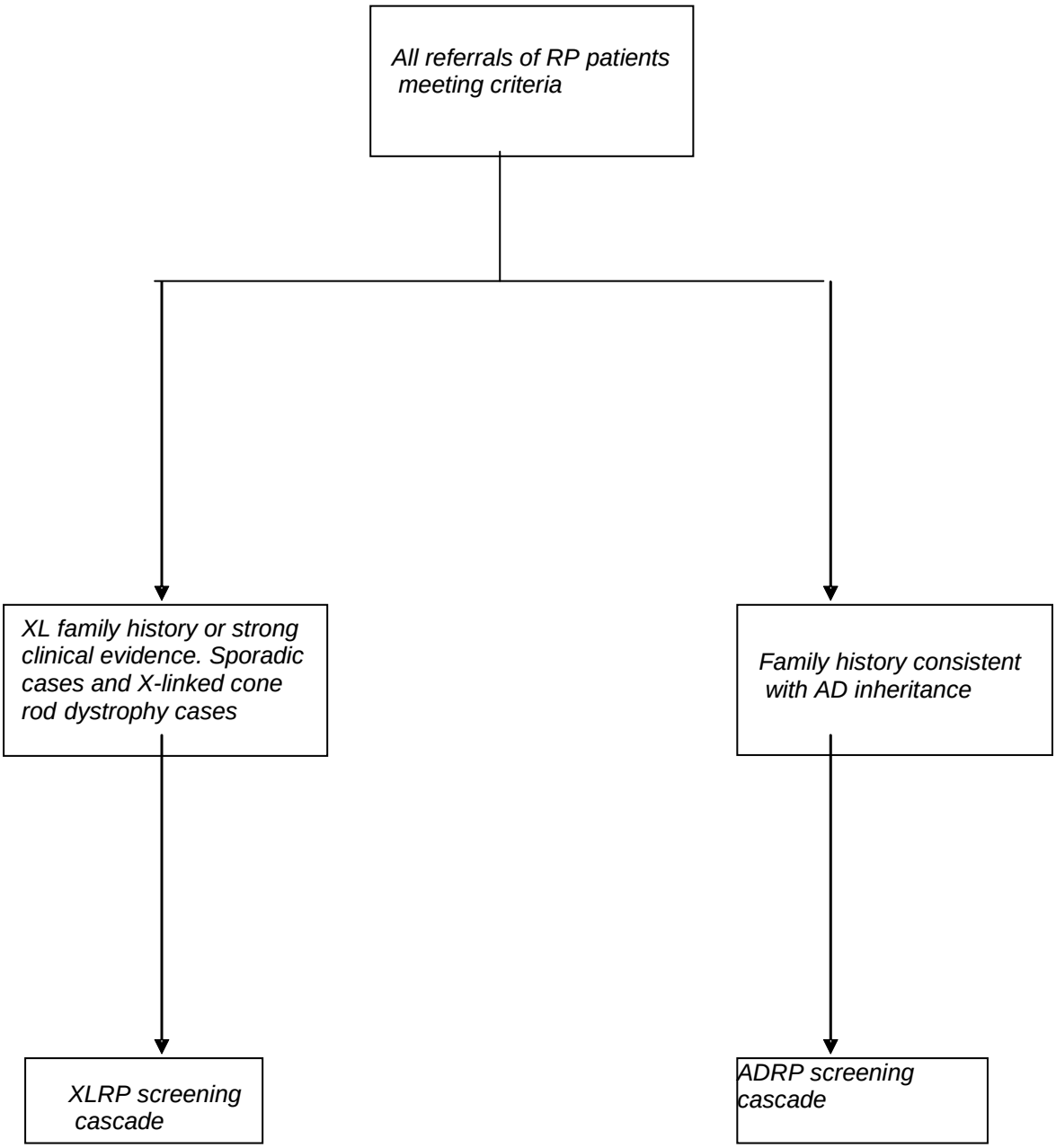
PLEASE PROVIDE DETAILS OF HOW REFERRALS WILL BE ASSESSED FOR APPROPRIATENESS?

All referrals will be assessed by a locally based expert clinical team familiar with the diagnosis of the RP clinical spectrum. The minimum criteria for acceptance will include a statement from the referrer that the index case has been diagnosed as a result of symptoms of initial rod dysfunction, followed by peripheral cone dysfunction,a characteristic retinal appearance and/or characteristic ERG. In addition the referrer should state that finding a pathological mutation in the index case is likely to lead to family testing and/or genetic risk assessment and/or treatment.



HOW MANY TESTS DO YOU EXPECT TO PERFORM ANNUALLY?

Approximately 450



Referral modes for X-linked and autosomal dominant RP.

UKGTN 'Testing criteria' template

Patient name:
Patient postcode:

Name of referrer:

Title/Position:

Name of Disease/test:
Retinitis Pigmentosa

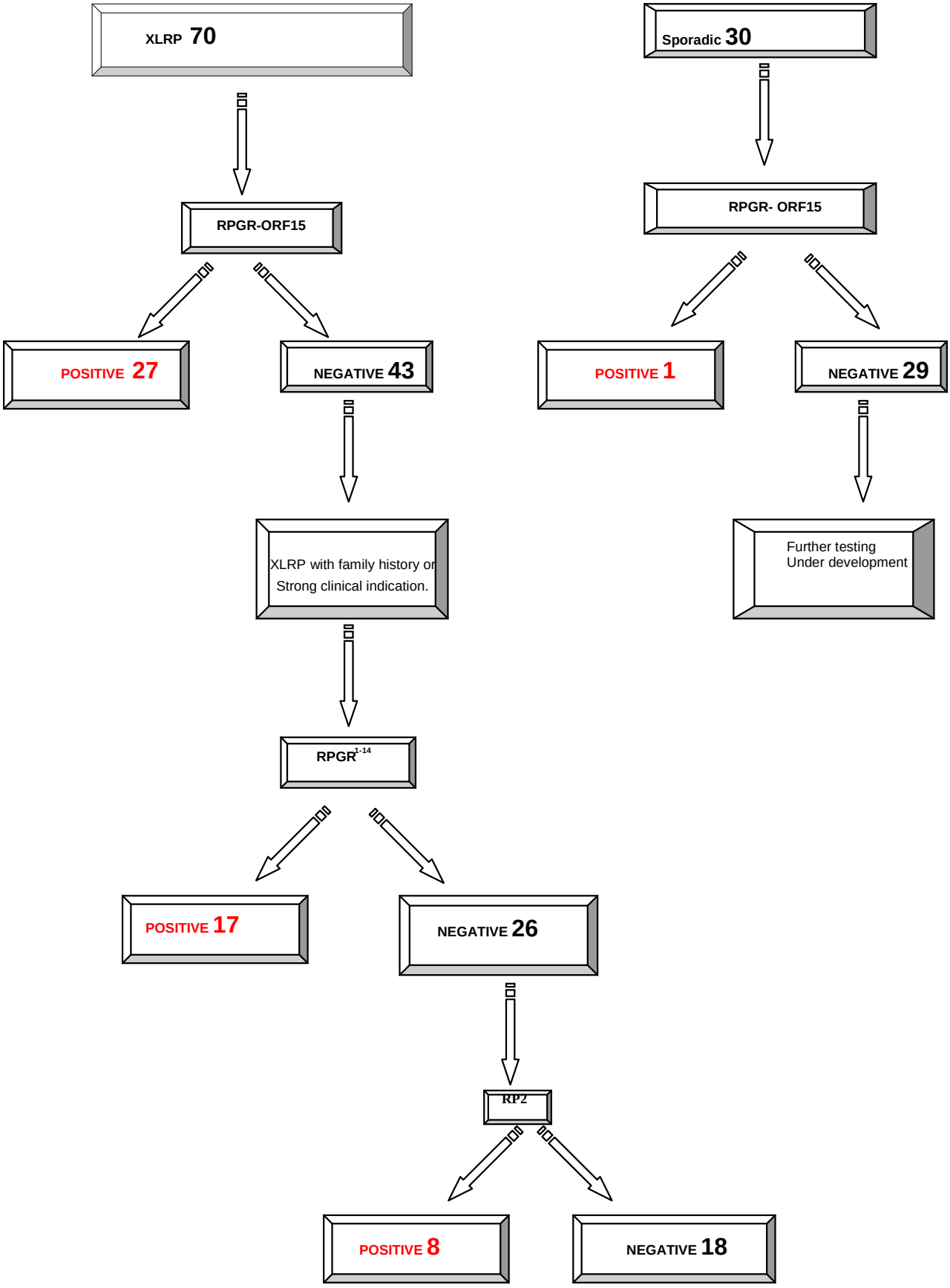
Referrals only will be accepted from one of the following:
(Please indicate with a tick which category refers to the referrer).

Referrer	**Tick if this refers to you.**
Consultant Clinical Geneticist	
Consultant Ophthalmologists	

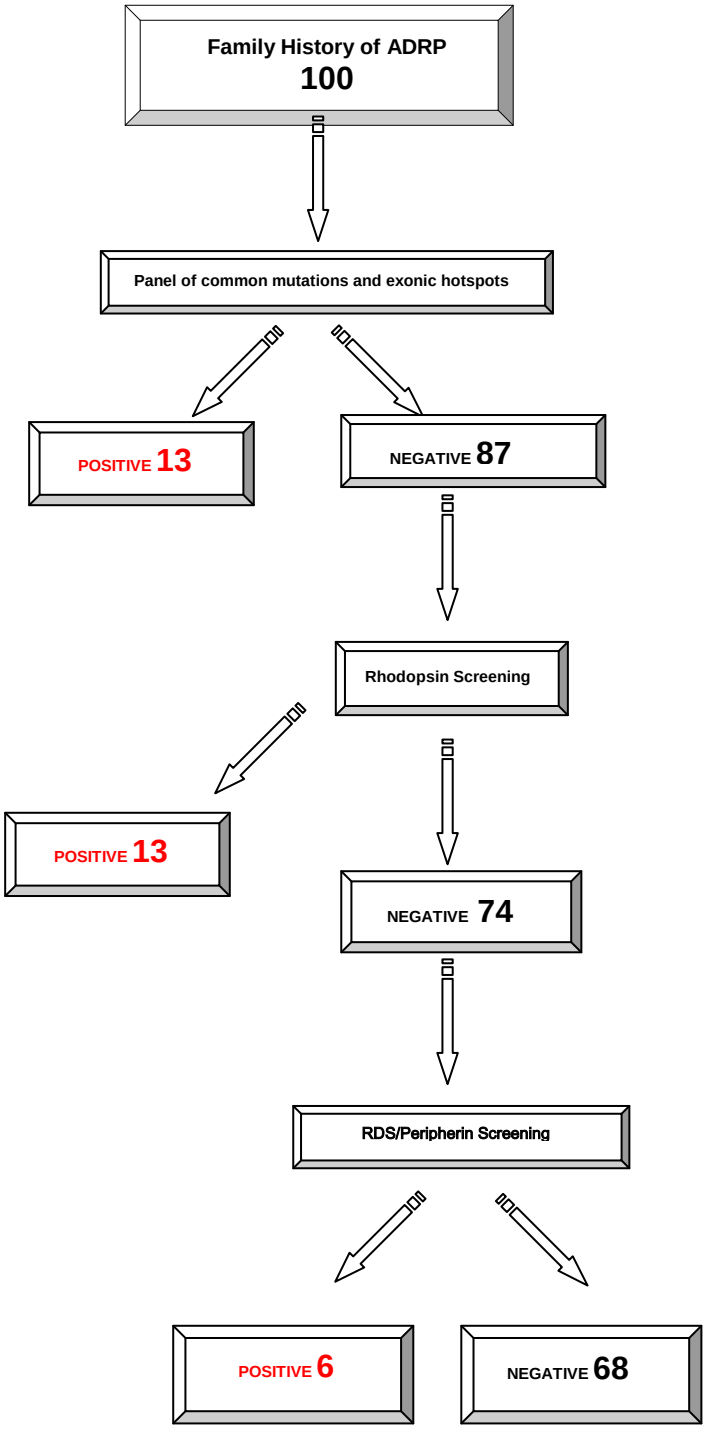
Minimum criteria required for testing to be appropriate as stated in the Gene Dossier:

Criteria	**Tick if this patient meets criteria**
The minimum criteria for acceptance is that the index case has been diagnosed as a result of symptoms of initial rod dysfunction, followed by peripheral cone dysfunction ,a characteristic retinal appearance and/or characteristic ERG.	
Please indicate if the suspected diagnosis is **X –linked** RP **OR**	
If the suspected diagnosis is **Autosomal Dominant** RP	
Please indicate that finding a pathological mutation in the index case is likely to lead to family testing and/or genetic risk assessment and/or treatment.	

If the sample does not fulfil these criteria and you still feel that testing should be performed please contact the Regional Genetics Service on 0161 276 6506 to discuss testing of the sample.



X-linked and sporadic RP testing cascades.



Autosomal dominant RP testing cascade.