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# **Horizon Scanning Technology Horizon Scanning Report**

## **Genetic testing for congenital long QT syndrome**

**May 2007**



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Enquiries about the content of the report should be directed to:

HealthPACT Secretariat  
Department of Health and Ageing  
MDP 106  
GPO Box 9848  
Canberra ACT 2606  
AUSTRALIA

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This *Horizon scanning report* was prepared by Ms Linda Mundy, Mr Adrian Purins, A/Prof Annette Braunack-Mayer and Professor Janet Hiller from the National Horizon Scanning Unit, Adelaide Health Technology Assessment, Discipline of Public Health, Mail Drop 511, University of Adelaide, Adelaide, South Australia, 5005.

## Table of Contents

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|  |    |
|--|----|
| Executive Summary .....                          | 1  |
| HealthPACT Advisory .....                        | 3  |
| Introduction .....                               | 4  |
| Background .....                                 | 4  |
| <i>Description of the technology</i> .....       | 4  |
| <i>The procedure</i> .....                       | 7  |
| <i>Intended purpose</i> .....                    | 9  |
| <i>Clinical need and burden of disease</i> ..... | 10 |
| <i>Stage of development</i> .....                | 12 |
| Treatment Alternatives .....                     | 12 |
| Clinical Outcomes .....                          | 15 |
| <i>Safety</i> .....                              | 15 |
| <i>Effectiveness</i> .....                       | 16 |
| Potential Cost Impact .....                      | 23 |
| Ethical Considerations .....                     | 26 |
| Training and Accreditation .....                 | 28 |
| Limitations of the Assessment .....              | 30 |
| <i>Search Strategy used for the Report</i> ..... | 30 |
| <i>Availability and Level of Evidence</i> .....  | 31 |
| Sources of Further Information .....             | 32 |
| Conclusions .....                                | 35 |
| Appendix A: Levels of Evidence .....             | 38 |
| Appendix B: Diagnostic techniques .....          | 40 |
| Appendix C: Profiles of studies .....            | 43 |
| Appendix D: HTA Internet Sites .....             | 46 |
| Appendix E: Glossary of terms .....              | 49 |
| References .....                                 | 51 |

## Tables

---

|         |  |    |
|---------|--|----|
| Table 1 | LQTS gene mutations .....                          | 6  |
| Table 2 | Criteria for diagnosis of LQTS .....               | 13 |
| Table 3 | Molecular analysis in family members .....         | 18 |
| Table 4 | Molecular analysis in unrelated patients .....     | 21 |
| Table 5 | Implantation costs of ICD device in Australia..... | 24 |
| Table 6 | Search terms utilised .....                        | 30 |
| Table 7 | Literature sources utilised in assessment .....    | 31 |
| Table 8 | Ongoing studies into long QT syndrome .....        | 32 |

## Figures

---

|          |   |    |
|----------|---|----|
| Figure 1 | A normal ECG .....                                    | 5  |
| Figure 2 | Methods used to detect LQTS mutations.....            | 8  |
| Figure 3 | Single-strand conformational polymorphism (SSCP)..... | 40 |
| Figure 4 | DHPLC for mutant DNA .....                            | 41 |
| Figure 5 | DHPLC for normal DNA.....                             | 42 |

## Executive Summary

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Congenital long QT syndrome (LQTS) is caused by mutations in a set of genes (LQT 1-8) which code for protein subunits of cardiac ion channels. The main clinical feature of LQTS is the elongation of the QT interval on electrocardiograms (ECGs). The majority of LQTS patients are asymptomatic and are diagnosed either by family history or by virtue of having survived an episode of syncope or severe ventricular arrhythmia. Unfortunately, for many LQTS patients, the first presentation of symptoms is sudden cardiac death, usually occurring in healthy children or young adults.

It is estimated that LQTS affects approximately 1: 5,000 individuals in the United States and that it would be reasonable to expect that this prevalence would hold true in Australia and New Zealand. Research has suggested that a number of deaths in young people that have previously been ascribed to causes such as drowning, motor vehicle accidents and sudden infant death syndrome may be attributable to LQTS.

LQTS is usually diagnosed using clinical indicators; including past episodes of syncope with or without stress; congenital deafness, length of QTc obtained via ECG; torsade de pointes; T-wave alternans; notched T wave in 3 leads; low heart rate for age; and family history all combine to give a patient an overall clinical score. Based on this score patients are rated on their probability of having LQTS.

Mutational analysis of LQTS involves the use of polymerase chain reaction (PCR) with products then analysed using either single-strand conformational polymorphism or denaturing high performance liquid chromatography and, if required, subsequent DNA sequencing. DNA is usually obtained and isolated from a blood sample but also may be obtained via a mouth swab.

Molecular diagnosis is intended to identify asymptomatic family members of identified LQTS probands who have not been identified via clinical screening and may be at risk of sudden death. In addition, individuals deemed to have an intermediate or high risk of LQTS according to the diagnostic criteria of unexplained syncope, a prolonged QTc on an ECG or a family history of sudden cardiac death would be candidates for screening by mutational analysis. Approximately 30-35 per cent of questionable cases of LQTS will *not* be picked up by molecular diagnosis due to the large number of mutations implicated in LQTS and the possibility of a large number of as yet unidentified mutations.

The only safety outcome reported by papers included for assessment in this report was the high number of genotype negative individuals, as the lack of identification of a genetic defect does not rule out the presence of LQTS.

Clinical assessment alone may not be sufficient to detect silent gene carriers who are asymptomatic for LQTS but are at risk of experiencing life-threatening cardiac events. One study reported a high number of asymptomatic carriers (21.4%) in family members of probands, underlining the importance of DNA analysis for individuals with an ambiguous clinical diagnosis (Jongbloed 1999). A high quality study directly compared the results of

molecular analysis to the results of ECG (QTc) and the Schwartz Clinical Score in family members of probands (Priori et al 1999). There was a low penetrance (25%), indicating that although a high number of family members were genotype positive (72%), the majority of these were clinically asymptomatic. When compared to mutational analysis, ECG and clinical score both had a low sensitivity (38%) but 100 per cent specificity. This study therefore demonstrates that molecular analysis is superior to clinical diagnostic techniques for the identification of “silent” LQTS carriers when a known mutation is screened for in family members.

In studies of *unrelated* patients, conflicting results were reported with one study finding a good correlation with clinical criteria and the corrected QT interval and LQTS diagnosis, as both of these factors were statistically significant in the genotype positive group ( $p < 0.0001$ ) (Tester et al 2005 & 2006b). However, this was contradicted in another smaller study, which reported that 32 per cent of the tested population were negative for a LQTS mutation despite the fact that 81 per cent of these individuals had clinical symptoms of LQTS and the mean QTc of this group was elevated at  $484 \pm 46$  ms (Splawski et al 2000).

The follow-up study by Sherman et al (2005) reported on targeted mutational analysis for the rare LQT4 on patients previously found to be genotype negative for the more common mutations (LQT1-3 and LQT5-6). This study demonstrated the value in a *fully* comprehensive initial mutational analysis, as a further nine patients (3.3%) previously thought to be LQTS negative were found to be positive for this rare mutation.

A cost-effectiveness analysis of genetic testing for familial LQTS in *symptomatic index cases* was conducted. The expected cost-effectiveness of genetic testing of *first-degree relatives* or more distant relatives was not included in this analysis. The three most common mutations were examined in the KCNQ1, KCNH2 and the SCN5A genes. Genetic testing of probands was found to be cost-effective compared to no genetic testing, at a cost per year life saved of US\$2,500, well below the standard threshold of US\$50,000 per life-year saved often used to define a cost-effective intervention. A further cost-effectiveness analysis is required to consider the benefits of genetic testing of family members of the proband.

In conclusion, clinical symptoms and an elongated QTc may not necessarily predict LQT and a negative mutation analysis may not rule out a diagnosis of LQTS. Studies included in this assessment reported that mutational analysis appears to be superior in the diagnosis of LQTS carriers when compared to clinical assessment alone. In addition, once a proband has been identified, targeted screening of family members for a specific mutation may be a cost-effective measure for extended family screening. Mutational analysis of suspected LQTS patients appears to be effective in identifying individuals previously thought to be clinically asymptomatic, and this may have long term consequences for their future. As the first symptom of LQTS for many patients is a life-threatening cardiac event, early diagnosis is of utmost importance.

For an individual identified as being at high-risk for long QT syndrome due to clinical assessment including ECG analysis, the direct benefits of genetic testing may be limited as:

- a negative genetic test does not exclude the possibility of long QT syndrome as approximately 30% of deletions or mutations are undetectable with existing PCR tests.
- patients suspected of having long QT are likely to undergo electrocardiological studies to demonstrate propensity for arrhythmias before the implantation of a defibrillator or other medical therapies, regardless of the outcome of genetic testing.

However, a definitive genetic diagnosis may guide treatment and management of long QT patients.

Family members of probands (either genetically or clinically diagnosed) may benefit from genetic testing to assess their risk of such clinical consequences. However, there is limited information on the risk of sudden death or other complications in individuals detected only by genetic screening, or on the effectiveness of medical intervention in these individuals. Therefore the modelling of cost-effectiveness is based on low quality evidence and should be interpreted with caution.

## Introduction

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The National Horizon Scanning Unit, AHTA, Discipline of Public Health, University of Adelaide, on behalf of the Medical Services Advisory Committee (MSAC), has undertaken an Horizon Scanning Report to provide advice to the Health Policy Advisory Committee on Technology (Health PACT) on the state of play of the introduction and use of genetic testing for congenital long QT syndrome.

Genetic testing for long QT syndrome is not routinely conducted in Australia but is conducted by the Cardiac Inherited Diseases Group (CIDG) in New Zealand. The Royal Children's Hospital in Melbourne has recently set up a genetic laboratory to study arrhythmia conditions such as long QT. Although the capacity for long QT screening in this laboratory is limited, it is feasible with further funding that this and other diagnostic laboratories would be able to provide mutational analysis of the gene sequences identified as implicated in inherited long QT syndrome. Mutational analysis for a number of genetic disorders, using similar techniques, is already well established in a number of pathology laboratories in Australia.

This Horizon Scanning Report is intended for the use of health planners and policy makers. It provides an assessment of the current state of development of genetic testing for long QT syndrome, its present use, the potential future application of the technology, and its likely impact on the Australian health care system.

This Horizon Scanning Report is a preliminary statement of the safety, effectiveness, cost-effectiveness and ethical considerations associated with genetic testing for congenital long QT syndrome.

## Background

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### Description of the technology

For all definitions of genetic terms please refer to Appendix E.

#### *Long QT syndrome*

Sudden cardiac death (SCD) is defined as a natural and unexpected death due to cardiac events, within one hour of onset of symptoms. The majority of SCDs (75-80%) occur in patients with coronary heart disease who may, or may not, experience a myocardial infarction. The remainder of SCDs occur in patients with cardiomyopathies (dilated or hypertrophic) or primary electrical abnormalities, including long and short QT syndromes and Brugada syndrome. SCDs in individuals affected by these syndromes usually occur in healthy children or young adults in the absence of coronary artery disease or congestive heart failure (Ching & Tan 2006; Sukhija et al 2007). The majority of long QT syndrome (LQTS) patients are asymptomatic and are diagnosed either by family history or by virtue of having survived an episode of syncope

or severe ventricular arrhythmia (Roberts 2006). Rates of mortality in untreated individuals are high and patients who become symptomatic in the first year of life are at high risk of sudden cardiac death (Schwartz 2005).

The main clinical feature of LQTS is the elongation of the QT interval on electrocardiograms (ECG) (Schwartz 2006a). The basic ECG waveforms are labelled alphabetically beginning with the P wave which represents arterial depolarisation (Figure 1). The QRS complex represents ventricular depolarisation and the ST segment represents ventricular repolarisation. Arterial repolarisation is usually too low in amplitude to be detected (Goldberger 2001). The QT interval represents the time for both ventricular depolarisation and repolarisation to occur, estimating the duration of an average ventricular action potential. A normal QT interval can range from 200 to 400 milliseconds (ms) depending upon heart rate. After correction for heart rate, the normal corrected QT interval (QTc) is less than 440 ms (Figure 1) (Klabunde 2005). QT prolongation results in a fast, abnormal heart rhythm known as ventricular tachyarrhythmia *torsade de pointes* (TdP). Patients with LQTS will typically have a QTc interval of >480 ms (Schwartz 2006a). Episodes of TdP may terminate spontaneously with the patient often being unaware of the episode and recovering from syncope without intervention. However, episodes of TdP may recur with increased frequency and degenerate into ventricular fibrillation, resulting in death if resuscitation is not performed (Vincent 2005).

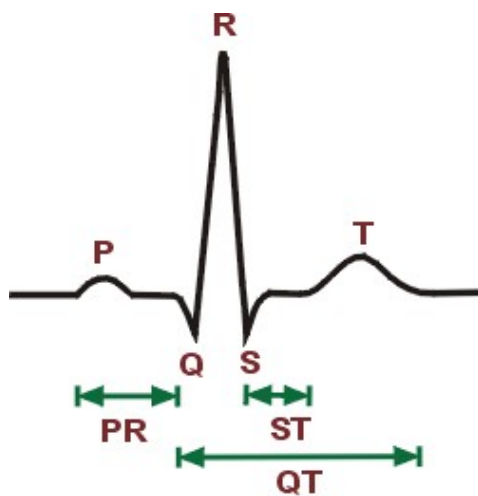


Figure 1 A normal ECG (Klabunde 2005)

Congenital or idiopathic LQTS is caused by mutations in a set of genes which code for protein subunits of cardiac ion channels, and as such may also be referred to as “channelopathies”. As of May 2005, eight major genotypes, LQT1-8, have been identified; however these genotypes are a result of a potential 471 different mutations and 124 polymorphisms. Table 1 provides a brief summary of these genes and mutations (Modell & Lehmann 2006). The three subtypes LQT 1-3 occur with the greatest frequency in LQTS patients (LQT1 = 45-50%, LQT2 = 45%, LQT3 = 7-8%). All of these affected genes encode for cardiac ion channels involved in the control of ventricular repolarisation. The remaining genotypes (LQT 4-8) are associated with rarer forms of the disease. LQT1 results from a mutation in the gene (KCNQ1) coding for a cardiac potassium channel ( $I_{Ks}$ ) and is the most common form of

LQTS. Untreated carriers of the KCNQ1 mutation have an annual risk of sudden death of approximately 0.3%. People with LQT2 have a defect in the HERG, or KCNH2 gene which encodes for a different cardiac potassium channel ( $I_{Kr}$ ) and is the second most common form of LQTS. LQT3 is a rare form of LQTS, and is a mutation in the SCN5A gene which encodes for a cardiac sodium channel ( $I_{Na}$ ) (CIDG 2006). LQT4 is associated with mutations in the ANK2 and ANKB genes. Ankyrin-B is a member of a large family of proteins that link integral membrane proteins to the spectrin-based cytoskeleton. Ankyrin-B is a membrane adaptor protein and was the first non-cardiac channel form of LQTS described, broadening the LQTS field from channelopathies to electropathies (Sherman et al 2005).

**Table 1 LQTS gene mutations (Modell & Lehmann 2006)**

| LQT phenotype | Gene name        | Ion current affected                        | Effect of mutation | Common triggers   |
|---------------|------------------|---|--------------------|---|
| LQT1          | KvLQT1, KCNQ1    | $I_{Ks}$                                    | Loss-of-function   | Exercise, emotional stress                                  |
| LQT2          | HERG, KCNH2      | $I_{Kr}$                                    | Loss-of-function   | Rest/sleep, auditory stimuli, emotional stress, post-partum |
| LQT3          | SCN5A            | $I_{Na}$                                    | Gain-of-function   | Rest/sleep  |
| LQT4          | ANKB, ANK2       | Affects $Na^+$ , $K^+$ , $Ca^{++}$ exchange | Loss-of-function   | Exercise, emotional stress                                  |
| LQT5          | mink, IsK, KCNE1 | $I_{Ks}$                                    | Loss-of-function   | Insufficient data   |
| LQT6          | MiRP1, KCNE2     | $I_{Kr}$                                    | Loss-of-function   | Insufficient data   |
| LQT7          | Kir2.1, KCNJ2    | $I_{K1}$                                    | Loss-of-function   | Insufficient data   |
| LQT8          | CACNA1C          | $I_{Ca,L}$                                  | Gain-of-function   | Hypoglycaemia, sepsis                                       |

There are two forms of the disease; the most common form is autosomal-dominant, also known as Romano Ward syndrome. With autosomal-dominant transmission, males and females are equally affected and each child of an affected parent has a 50 per cent chance of inheriting the abnormal allele, and therefore having LQTS. Although inheritance of the mutation is equal in males and females, females appear to have more frequent symptoms than males. The other rarer form of LQTS is called the Jervell, Lange-Nielsen (JLN) variant, which is inherited in an autosomal-recessive manner (Ching & Tan 2006; Roberts 2006; Vincent 2005). JLN results from homozygous or compound heterozygous mutations on either one or two genes, KCNQ1 (LQT1) and KCNE1 (LQT5). Both of these genes encode proteins which co-assemble to form the potassium cardiac channel (Schwartz et al 2006). With autosomal recessive transmission, if both parents carry the mutation, each child has a 25 per cent chance of inheriting the defective allele from each parent and being homozygous for LQTS. In addition, this variant also affects the potassium channels in the ear, resulting in profound congenital deafness. There is a 50 per cent chance that children may inherit a defective gene from one parent. These heterozygous individuals essentially have the Romano-Ward form of LQTS and have normal hearing. In addition, each child has a 25 per cent chance of having a normal phenotype, in terms of LQTS and deafness, by inheriting a normal allele from each parent (Ching & Tan 2006; Roberts 2006; Vincent 2005).

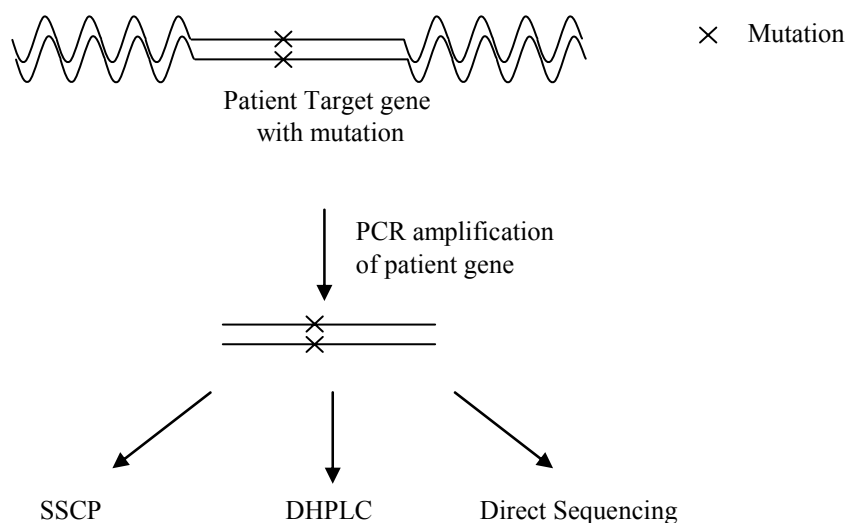
Treatment options for symptomatic patients include administration of  $\beta$ -blockers, left cardiac sympathetic denervation and implantable cardiac defibrillators. Management of asymptomatic patients is more difficult, however they should also receive treatment, usually with  $\beta$ -blockers (Schwartz 2006a). Patients diagnosed with LQTS are advised to avoid competitive sports and vigorous activity, such as swimming, is contraindicated (CIDG 2006). Female probands with congenital LQTS are at significant risk of a cardiac event during the postpartum period and should be treated with  $\beta$ -blockers. This increased risk is thought to be related to an increase in the level of oestrogen and progesterone in addition to an increase in sympathetic nerve activity, which may increase the number of mutated ion channel proteins, precipitating a cardiac event (Wehrens et al 2002).

In addition to the congenital forms, LQTS may be acquired via exposure to particular drugs, including antipsychotics, antihistamines, methadone, antiemetic agents and certain antibiotics. Approximately 50 drugs listed by the Food and Drug Administration may potentially affect the QTc interval. These drugs may induce LQTS in genetically normal individuals but may also precipitate symptoms in congenital LQTS individuals and should therefore be avoided (Ching & Tan 2006; Roden 2004).

#### *The procedure*

The genetic diagnostic assays for LQTS are polymerase chain reaction (PCR) based tests followed by analysis using either single-strand conformational polymorphism (SSCP) or denaturing high performance liquid chromatography (DHPLC) and, if required, subsequent DNA sequencing (Figure 2). Please see Appendix B for more in-depth diagrams of these techniques. DNA is usually obtained and isolated from a blood sample but also may be obtained via a mouth swab. Several regions of the patient's genome are amplified using specifically designed primers. The regions amplified depend on the genes being investigated. Once the PCR products are amplified they are denatured and subjected to either single-strand conformational polymorphism (SSCP) analysis or denaturing high performance liquid chromatography (DHPLC). These techniques allow the initial screening and comparison of the test PCR product against PCR products amplified from normal human reference DNA. Any differences between the test and reference DNA can indicate a possible mutation in the patient's DNA. SSCP relies on the fact that single strands of DNA self-fold into specific conformations. Mutations such as base changes, deletions or insertions affect the conformation of the single stranded DNA and hence the physical properties of the folded DNA. This difference in physical properties can be exploited to differentiate a mutant sequence from a normal sequence. DHPLC relies on the fact that dimers of DNA that have differences in their sequence will have different melting temperatures. Hence DNA that contains a mutation can be distinguished from the normal reference DNA on a DHPLC column due to its melting temperature.

Some mutations are not possible, or are difficult, to detect using these techniques e.g. large deletions will cause the failure to amplify PCR products as the target sequence may not exist (Modell & Lehmann 2006).



**Figure 2      Methods used to detect LQTS mutations**

SSCP is the electrophoretic separation of the denatured single stranded DNA products based on differences in their conformation (Figure 3, Appendix B). This can be achieved in several media such as acrylamide gels or capillary tubes which allow the use of several different coloured fluorophores and hence the up-scaling of the test to accommodate several samples per tube. This allows a greater throughput of samples per time period, reducing costs and decreasing time to diagnosis (Hofman-Bang et al 2006; Modell & Lehmann 2006).

DHPLC involves the heat denaturation of the PCR products, then re-annealing in the presence of similarly denatured PCR products from normal homologous DNA (Figure 4 & 5, Appendix B). The test and normal DNA form duplexes, which may or may not contain a mutation depending on the whether there was a mutation in the patient's DNA sequence being tested. If, when the patient and normal PCR products are re-natured together, if a mutation is present in the patient's DNA sequence, a mix of hetero (mutant patient plus normal DNA) and homo-dimers (either mutant patient plus mutant patient or normal plus normal) will result. If no mutation is present in the patient's DNA sequence then the re-naturation of the patient and normal PCR products will only result in homo-dimers, as the two PCR products are identical despite their differing sources. The different species of dimers can be distinguished due to their differing melting points on a DHPLC column (Modell & Lehmann 2006; Tester et al 2006a).

If mutations are detected by the above methods then sequencing of the patients DNA can be used to determine the exact mutation in the identified gene. It is also feasible to sequence before using either SSCP or DHPLC to identify where the mutations lie, but this is a much slower and expensive method of discovering the exact mutations (Modell & Lehmann 2006).

In the Cardiac Inherited Diseases Group (New Zealand) laboratory, there is currently a six month turnaround for LQT 1-3 analysis and LQT 5 and 6 are tested annually (personal communication CIDG May 2006). It would not be

practical to conduct universal population screening for LQTS, however screening of family members of individuals who have died from sudden cardiac failure should be recommended. PCR and DNA sequencing are robust techniques with high sensitivity and specificity and good reproducibility, however they are expensive and technically complex (Louie et al 2000).

PGxHealth™ (United States) developed a commercial genetic test for LQTS under the name FAMILION™. Genaissance has recently been taken over by Clinical Data Inc (United States), of which, PGxHealth™ is a part. This test currently has 3 forms, comprehensive testing, family specific testing and sodium channel specific testing. Comprehensive testing involves the sequencing of 73 amplicons across 5 genes involved in LQTS, that is LTQ1,2,3,5 and 6. Family specific testing is a subset of this testing regimen with only the gene/s identified previously in an index case being sequenced in the family members. This saves time and money versus repeating comprehensive testing for each family member tested. The sodium channel analysis is targeted at suspected Brugada syndrome patients and involves the sequencing of both intronic and open reading frames of the SCN5A gene.

#### *Intended purpose*

Molecular diagnosis is intended to identify asymptomatic family members of identified LQTS probands who have not been identified via clinical screening and may be at risk of sudden death. In addition, individuals deemed to have an intermediate or high risk of LQTS according to the diagnostic criteria of unexplained syncope, a prolonged QTc on an ECG or a family history of sudden cardiac death would be candidates for screening by mutational analysis (Schwartz 2006a). However, approximately 10 per cent of all mutation carriers have a *normal* QTc interval (<440 ms). Some families have low penetrance of the genetic mutation, displaying a mild phenotype with minimal or no clinical symptoms. These individuals are considered to be “silent gene carriers” as they present as normal on clinical and ECG grounds. Low penetrance mutation carriers may be unaware that they may pass on the LQTS mutation to their offspring, and they are also at risk of sudden death, especially if exposed to drugs that may potentially block potassium ion channels. Therefore molecular screening is recommended for all family members of a positively genotyped individual, regardless of their presenting phenotype (Wehrens et al 2002). Routine mutational analysis is currently limited to the five LQTS genes: KCNQ1, KCNE1, KCNE2, KCNH2 and SCN5A. Approximately 30-35 per cent of questionable cases of LQTS will *not* be picked up by molecular diagnosis due to the large number of mutations implicated in LQTS and the possibility of a large number of as yet unidentified mutations (Schwartz 2006a). The lack of identification of a genetic defect does not rule out the presence of LQTS (Priori & Napolitano 2006).

In addition to being a diagnostic tool, genetic testing has a prognostic value due to correlations between cardiac events and specific genotypes. Individuals with LQT1 and LQT2 are at a higher risk of a cardiac event than those diagnosed with LQT3, with a higher annual risk of sudden cardiac death of about 0.8% and 0.5% in females and males, respectively. Although LQT3 is rare in comparison to LQT1 and LQT2, it is more lethal. Unfortunately the

first clinical presentation of patients carrying the LQT3 mutation is often sudden death. For individuals with this mutation, gender is an additional risk factor with an untreated annual risk of sudden death of 1% in males compared to 0.3% for females (Ching & Tan 2006; CIDG 2006).

Genotype also correlates to an individual's response to pharmacological therapy and other treatment options.  $\beta$ -blockers are effective for LQT1 and LQT2 patients, whereas LQT3 patients may benefit from type Ib sodium channel blockers, although there are some concerns about the long term safety of this therapy. Potassium supplements may be of benefit to LQT7 patients. LQT1 patients may also be treated with propranolol with dosage titrated to achieve a heart rate of 130 beats per minute on treadmill exercise testing. LQT3 patients are more likely to benefit from implantation with a pacemaker to raise their heart rate to >80 beats per minute, as they are prone to SCD at a slower heart rate which is exacerbated during sleep and rest (Ching & Tan 2006).

#### *Clinical need and burden of disease*

It is difficult to estimate the prevalence of LQTS. It is a leading cause of sudden, unexplained cardiac death in children and young adults with a structurally normal heart. It is estimated that LQTS affects approximately 1: 5,000 individuals in the United States and that it would be reasonable to expect that this prevalence would hold true in Australia and New Zealand (Ackerman 2005; SADS 2006). The estimated prevalence in children aged 4-15 years in England, Wales and Ireland is 1.6 – 6 per million (Ching & Tan 2006). Other research has shown that genes known to be involved in LQTS have been identified in 10 per cent of deaths from sudden infant death syndrome (Arnestad et al 2007). Other unsubstantiated estimates in the literature range from 1: 2,500 (Quaglini et al 2006), to between 1: 5,000 and 1: 20,000 (Schwartz et al 2003).

A recent Australian study set out to determine the causes of sudden cardiac death in people aged  $\leq 35$  years. This cross sectional study (level IV aetiology evidence) was conducted on all autopsies performed on people  $\leq 35$  years at a major Sydney forensic unit from January 1994 and December 2002. During the study period there were 10,199 autopsies performed. Of these 2,986 (29.2%) were individuals aged  $\leq 35$  years of whom 193 were classified as sudden cardiac deaths. The cause of sudden death was as follows: not established but probable primary arrhythmia (31%), coronary artery disease (24%), hypertrophic cardiomyopathy / unexplained left ventricular hypertrophy (15%), viral myocarditis (12%), congenital heart disease (7%) and other (11%). Of the 193 sudden cardiac deaths 22 and 38 per cent occurred during exercise and minimal exertion or at rest, respectively. Forty per cent were not witnessed. It may also be likely that a number of deaths in young people that have previously been ascribed to causes such as drowning, motor vehicle accidents and sudden infant death syndrome (SIDS) may be attributable to LQTS (Doolan et al 2004).

A similar study examined all (n=49) sudden unexplained deaths referred by the coroner to the Mayo Clinic in the United States for molecular autopsy (level IV aetiology evidence). The decedents were older than one year (mean

age  $14.2 \pm 10.9$  years) with no family history of LQTS, and death was sudden, unexpected and unexplained after autopsy. The majority of deaths occurred during sleep (34.7%) or during exertion (24.5%) and two women (4.1%) died in the postpartum period. Six of the LQTS susceptible genes were screened for: KCNQ1, KCNE1, KCNE2, KCNH2, SCN5A and KCNJ2. In addition, a targeted analysis of 10 ANK2 exons implicated in LQT4 and TS1 (LQT8) was conducted. Ten LQTS associated mutations were identified in 10/49 (20%) decedents. Five decedents were identified with LQT1, three with LQT2 and two with LQT3. The authors concluded that a large proportion of sudden unexplained cardiac deaths may be directly attributable to LQTS, which may have screening and treatment implications for surviving family members (Tester & Ackerman 2007).

As described above, LQTS has been implicated as a possible cause of death in cases of drowning and SIDS. A Finnish study conducted molecular autopsies on 165 consecutive bodies found in water, screening for only two of the LQTS mutations, KCNQ1 and KCNH2 (level IV aetiology evidence). Only one body was found to be positive for the KCNH2 mutation, giving a prevalence of 0.61 per cent, 95%CI [0.02, 3.33]. Toxicology reports, however, on this decedent revealed the presence of drugs and alcohol, so attributing this death to LQTS may be misleading. Conversely the prevalence of LQTS mutations in this group may have been higher if all LQTS mutations were screened for (Lunetta et al 2003). Defects in the SCN5A gene are thought to be responsible for some cases of SIDS.

Post-mortem molecular analysis on the SCN5A gene in 93 cases of SIDS or undetermined death in infants was conducted (level IV aetiology evidence). Two of the SIDS cases were positive for the SCN5A mutation, a prevalence of 2.2 per cent (Ackerman et al 2001). A more recent Italian study performed molecular analysis for seven genes<sup>1</sup> associated with LQTS on 201 SIDS cases and 182 deceased infant and adult controls (level III-3 aetiology evidence). Mutations were identified in 26/201 (12.9%) of SIDS cases, with the majority in the SCN5A gene (13/26, 50%). Five cases (19%) had a mutation in the KCNQ1 gene and a different five cases had a mutation in the KCNH2 gene. The CAV3 mutation occurred in three cases (11%) and only one case had a mutation in the KCNE2 gene (4%). One case (4%) had a mutation in two genes, SCN5A and CAV3. When the functional effect of these mutations was considered, the authors concluded that eight mutations and seven rare variants were implicated in the death of 19/201 (9.5%) of the SIDS infants (Arnestad et al 2007).

Based on the findings of these SIDS studies many authors are calling for ECG screening of all newborns (Berul & Perry 2007; Schwartz 2006b) whilst others urge caution (van Langen & Wilde 2006). A 1998 cost-effectiveness study on mass neonatal screening with ECG estimated that 100 infants would have to be treated with  $\beta$ -blockers to save two lives. However, the commonest type of LQTS detected in SIDS infants is LQT3, which does not respond to  $\beta$ -blocker therapy.  $\beta$ -blocker therapy is counter intuitive for this potential patient group as SIDS deaths occur at rest and not at times of adrenalin surge (Skinner 2005).

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<sup>1</sup> KCNQ1, KCNH2, SCN5A, KCNE1, KCNE2, KCNJ2 and CAV3

### *Stage of development*

The Cardiac Inherited Diseases Group in Auckland, New Zealand, conducts genetic testing for patients with overt LQTS, allowing identification of subtypes and thus enabling patients to receive appropriate treatment and lifestyle advice. To date, over 100 patients have been screened, with 50 genetic mutations identified. In addition, 160 family members have been screened for the same mutations. Approximately 20 patient samples have been sent from Australia to New Zealand for genetic testing (personal communication CIDG May 2006).

A genetic laboratory has recently been set up at the Royal Children's Hospital in Melbourne for genetic studies to test for arrhythmia conditions such as Long QT Syndrome. The capacity for screening in this laboratory is limited due to lack of funding (personal communication, Royal Children's Hospital, May 2006).

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## **Treatment Alternatives**

### **Existing comparators**

Patients are assessed for LQTS for several reasons: aborted sudden death; unexplained syncope; ECG screening of other family members; or other reasons (Moss & Robinson 1992). The current standard approach to diagnose LQTS was developed in 1993 (Schwartz & Locati 1985; Schwartz et al 1993) and is used to assess several clinical indicators and the patient's family history to give the patient an overall clinical score (Table 2). Based on this score patients are rated on their probability of having LQTS. The clinical indicators are: past episodes of syncope (fainting), either with or without stress; congenital deafness in the patient; length of the patient's QTc in milliseconds; torsade de pointes; T-wave alternans; Notched T wave in 3 leads; and low heart rate for age. The relevant family history indicators are: family member with definite LQTS; and, unexplained sudden cardiac death in an immediate family member aged less than 30 years. Each indicator is weighted to reflect the relevance of that particular indicator to LQTS. Patients receiving a score of:  $\leq 1$  have a low probability of having LQTS; 2-3 points have an intermediate chance of LQTS; or,  $\geq 4$  have a high probability of having LQTS.

**Table 2**      **Criteria for diagnosis of LQTS (Khan 2002)**

| <b>Characteristics</b>   | <b>Points</b> |
|--|---------------|
| <u>Clinical history</u>  |               |
| Syncope  |               |
| With stress  | 2             |
| Without stress   | 1             |
| Congenital deafness  | 0.5           |
| <u>Family history*</u>   |               |
| Family members with definite LQTS  | 1             |
| Unexplained sudden cardiac death at age <30 y among immediate family members | 0.5           |
| <u>Electrocardiographic findings†</u>  |               |
| QTc  |               |
| ≥480 ms  | 3             |
| 460-470 ms   | 2             |
| 450 ms (in males)  | 1             |
| Torsade de pointes   | 2             |
| T-wave alternans   | 1             |
| Notched T wave in 3 leads  | 1             |
| Low heart rate for age (<2nd percentile)                                     | 0.5           |

Scoring: ≤1 point = low probability, 2-3 points = intermediate probability, ≥4 points = high probability.

Torsade de pointes and syncope are mutually exclusive.

\*The same family member cannot be counted twice, †In absence of medications or disorders known to affect these electrocardiographic features.

ECG testing may take several forms, depending on the patient's clinical condition. Tests include: resting ECG, exercise stress tests, medicated stress tests; Holter monitoring; or event ECG recording. A heart-rate corrected QT interval (QTc) of greater than 450ms for men and 460 for women and children is considered extended, although approximately 2.5 per cent of people considered normal will fall within this category. In addition, approximately 10 per cent of individuals considered to have LQTS will fall below these thresholds (Kanters et al 2006; Schwartz et al 1993).

Testing with ECG may give characteristic signs specific for particular gene mutations and hence guide the clinician as to which specific genes should be assessed during genetic testing (Kanters et al 2006). However, the use of ECGs alone to diagnose LQTS is unreliable. A study by Viskin et al (2005) presented four ECG traces, two from patients with congenital LQTS and two from normal healthy individuals, to 902 physicians from 12<sup>2</sup> different countries. Twenty-five QT specialists agreed that the ECGs were correct results. The ECG traces were then presented to arrhythmia specialists (n=106), cardiologists (n=329), and non-cardiac internal medicine physicians (n=442) who were asked to measure the QT, calculate the QTc and determine whether or not the QT interval was prolonged or normal. For the LQTS patients, >80 per cent of the arrhythmia experts, but <50 per cent of cardiologists and <40 per cent of the non-cardiac physicians, correctly calculated the QTc interval. The QTc tended to be underestimated in the LQTS patients and overestimated

<sup>2</sup> Australia, Austria, Brazil, Canada, China, England, France, Israel, Japan, Mexico, Paraguay and the United States

in the healthy individuals. Correct classification of all QT intervals was achieved by 62 per cent of arrhythmia experts, but by only <25 per cent of cardiologists and non-cardiac physicians. Inter-observer agreement was moderate amongst arrhythmia experts and low amongst cardiologists and non-cardiac physicians (kappa coefficient 0.44 and <0.3, respectively) (Viskin et al 2005).

### Safety

None of the papers included in this assessment reported on safety outcomes associated with the mutational analysis of LQTS. PCR and sequencing are established diagnostic tools and when offered by NATA<sup>3</sup> accredited laboratories, are performed by technically qualified personnel following specific protocols of quality assurance, sample preparation, amplification, detection and interpretation of results (White et al 1992). The advantages of PCR are that it has high sensitivity, high specificity and good reproducibility. Its limitations are: the potential for false-positive results from contaminating DNA; the potential for false-negatives due to the presence of PCR inhibitors; it is expensive and it is technically complex (Louie et al 2000). However, if PCR samples are run in triplicate or duplicate the number of false-positives would be reduced.

The major issue in the assessment of individuals suspected of carrying a mutation for LQTS is that clinical assessment alone may not be sufficient to detect silent gene carriers who are asymptomatic for disease but at risk of experiencing life-threatening cardiac events, especially if exposed to drugs capable of blocking potassium channels. Studies, such as the one conducted by Jongbloed (1999), reported a high number of asymptomatic carriers (21.4%) in family members of probands. In addition, “silent gene” carriers considered to have a normal phenotype may be capable of passing on gene mutations to offspring (Priori et al 1999). This underlines the importance of DNA analysis for individuals with an ambiguous clinical diagnosis.

In addition, all of the studies included in this assessment reported the numbers of genotype *negative* individuals. All authors stressed that a negative mutational analysis does not rule out the possibility of LQTS as a diagnosis. The study by Splawski et al (2000) (Table 3) reported 32 per cent of the tested population were negative for a LQTS mutation despite the fact that 81 per cent of these individuals had clinical symptoms of LQTS and the mean QTc of this group was elevated at  $484 \pm 46$  ms. Failure to identify mutations may result from phenotypic errors, incomplete sensitivity of the method used to detect mutations (in this case single strand conformational polymorphism) or the presence of mutations in regulatory sequences. However, it is felt that the main reason for the high number of negative genotype individuals is the presence of mutations which have yet to be detected and characterised (Splawski et al 2000).

Tester et al (2004) reported on the prenatal molecular analysis on the amniotic fluid of a foetus at 16 weeks gestation (Table 2). Analysis was successful and the foetus was confirmed as having a mutation in the KCNQ1 gene (LQT1). When bradycardia was detected in the foetus during labour, therapy with  $\beta$ -blockers was initiated immediately. Although prenatal genetic testing allowed appropriate genetic counselling, perinatal monitoring and immediate and potentially life-saving therapy to be initiated, it is unlikely that prenatal testing

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<sup>3</sup> NATA is the National Association of Testing Authorities, Australia

would become routine due to the inherent risks involved in amniocentesis (Tester et al 2004).

## Effectiveness

Five studies, which conducted LQTS molecular analysis of probands and family members, were included for assessment (Table 3). Although all of these studies used clinical assessment of probands as a basis for conducting molecular analysis, only the high quality study by Priori et al (1999) directly compared the results of molecular analysis to the results of ECGs (QTc) and the Schwartz Clinical Score (level III-2 diagnostic evidence). In this study, probands and family members were only screened for mutations in LQT1 and LQT2, however as all probands returned a positive result for a mutation in either of these genes it is reasonable to expect that mutations in other LQTS genes would not be present. Of the 46 family members screened, 33 per cent were found to be positive for a mutation in either gene. This corresponded to a penetrance of only 25 per cent in this group, with penetrance being defined as the ratio of patients with a clinical phenotype to the total number of family members carrying the mutation. The clinical implication of low penetrance has been described previously. When compared to mutational analysis, ECG and clinical score both had a low sensitivity<sup>4</sup> (38%) but 100 per cent specificity. This study therefore demonstrates that molecular analysis is superior to clinical diagnostic techniques for the identification of “silent” LQTS carriers when a known mutation is screened for in family members.

The comprehensive study by Napolitano et al (2005) reported on the genetic screening of 430 LQTS probands with *clinically* defined Romano Ward syndrome and family members consecutively referred for genetic testing (level III-2 diagnostic evidence). Screening was conducted for the five common mutations LQT1-3 and LQT 5-6. Of the 430 probands, 310 (72%) were identified as having a mutation associated with LQTS. Only the family members of these positive genotyped probands were screened (n=1,115) and 521 (46.7%) were found to be genetically affected. Of the probands with a mutation, 296 (95.5%) were heterozygous carriers of a single mutation. Twelve probands were compound heterozygotes with 2 (n=11) or 3 (n=1) mutations, and two patients (0.6%) were homozygous. The most prevalent defect in those probands with a single mutation was on the KCNQ1 gene (49%) followed by defects in the KCNH2 gene (39%). Ten per cent of the probands had a mutation in the SCN5A gene, 1.7% in the KCNE1 gene and only 0.7% in the KCNE2 gene. Parental DNA was available from 80 per cent of probands for genotyping, revealing that in the majority of cases that the mutation was inherited. Only 29 (12%) of cases were considered to be sporadic. Overall there were 235 different mutations detected, with 139 (59%) being considered *novel* mutations not previously reported. When clinical parameters were compared to the results of genetic testing the mean QTc interval for genetically affected individuals was 474 ms ( $\pm$  46 ms) (median =

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<sup>4</sup> Sensitivity is the ability of a test to *correctly* identify those who *have* the disease, specificity of the test is defined as the ability of the test to *correctly* identify those who *do not have* the disease

467 ms, IQR<sup>5</sup> was 444-495 ms). The mean QTc interval was lower amongst non-carrier family members at 406 ms ( $\pm 27$  ms) (median = 409 ms, IQR 390-425 ms), however no statistical analysis was presented on the difference between these two values. Amongst *probands*, the mean corrected QT interval was significantly longer ( $p < 0.001$ ) 496 ms ( $\pm 46$  ms) (median = 490 ms, IQR 462-520 ms) when compared to genetically affected family members, 461 ms ( $\pm 40$  ms) (median = 458 ms, IQR 436-484 ms).

The two studies by Jongbloed et al (1999) and Piippo et al (2001) demonstrate the value of only screening family members for the mutations detected in probands, which may be an important factor when considering the cost-effectiveness of extended family screening. Jongbloed et al screened probands for LQT 1-5, with the majority having a positive LQT1 or LQT2 mutation. Fourteen families, with 134 family members, were deemed to be large enough to screen for these mutations, which revealed 63 per cent to be genotype positive and of these 21 per cent were asymptomatic carriers. Similarly, Piippo et al probed family members only for the specific substitution and truncation mutations found in the two JLNS probands. This study did not report on the number of “silent” carriers, however the QTc of heterozygous carriers ( $460 \pm 40$  ms) was significantly different to that of non-carriers ( $410 \pm 20$  ms),  $p < 0.001$ .

Studies included in this assessment reported that mutational analysis appears to be superior in the diagnosis of LQTS carriers when compared to clinical assessment alone, and that once a proband has been identified, targeted screening of family members for a specific mutation may be a cost-effective measure for extended family screening. However, the majority of these studies did not report on whether or not genetic screening resulted in a change in management for these individuals. The only study to report on outcomes after mutational analysis and subsequent treatment of the patient, was the case report by Tester et al (2004). This case report demonstrates that it is feasible to conduct mutational analysis on foetal cells obtained via amniocentesis. Direct DNA sequencing was conducted on foetal DNA extracted from the amniotic fluid, confirming the presence of a mutation in the KCNQ1 gene, the same as that detected in the mother. When bradycardia was detected in the foetus during labour, therapy with  $\beta$ -blockers was initiated immediately for the baby. The child remained symptom free until a drug-induced (erythromycin) syncope episode occurred.

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<sup>5</sup> IQR = inter-quartile range

**Table 3 Molecular analysis for long QT syndrome in family members**

| Study                   | Level of Diagnostic Evidence | Study Design   | Population   | Outcomes   |                 |                 |                                |      |  |  |                 |             |    |      |  |  |                 |             |    |      |  |  |               |             |    |      |  |  |               |             |    |      |  |  |               |            |   |
|-------------------------|------------------------------|--|--|--|-----------------|-----------------|--------------------------------|------|--|--|-----------------|-------------|----|------|--|--|-----------------|-------------|----|------|--|--|---------------|-------------|----|------|--|--|---------------|-------------|----|------|--|--|---------------|------------|---|
| Napolitano et al (2005) | III-2                        | Case series of probands and family members   | 430 consecutive LQTS probands and 1,115 family members referred to clinic for genetic testing            | <p><b>Mutational analysis</b></p> <p>310/430 (72.1%) probands genotype positive<br/>           296/310 (95.5%) heterozygous mutations<br/>           144/296 (49%) mutations in KCNQ1<br/>           115/296 (39%) mutations in KCNH2<br/>           30/296 (10%) mutations in SCN5A<br/>           5/296 (1.7%) mutations in KCNE1<br/>           2/296 (0.7%) mutations in KCNE2</p> <p>14/310 (4.5%) compound heterozygotes of which 11/14 (78.6%) were 2 mutations and 1/14 (7.1%) were 3 mutations and 2/14 (14.3%) were homozygous</p> <p>1,115 family members of 310 probands genotyped<br/>           521 /1115 (46.7%) genotype positive</p> <table border="1"> <thead> <tr> <th><u>Genotype</u></th> <th><u>mean QTc</u></th> <th><u>QTc Pen (%)<sup>a</sup></u></th> </tr> </thead> <tbody> <tr> <td>LQT1</td> <td></td> <td></td> </tr> <tr> <td>306/521 (58.7%)</td> <td>457 ± 38 ms</td> <td>55</td> </tr> <tr> <td>LQT2</td> <td></td> <td></td> </tr> <tr> <td>164/521 (31.5%)</td> <td>467 ± 36 ms</td> <td>70</td> </tr> <tr> <td>LQT3</td> <td></td> <td></td> </tr> <tr> <td>33/521 (6.3%)</td> <td>478 ± 52 ms</td> <td>79</td> </tr> <tr> <td>LQT5</td> <td></td> <td></td> </tr> <tr> <td>15/521 (2.9%)</td> <td>438 ± 29 ms</td> <td>33</td> </tr> <tr> <td>LQT6</td> <td></td> <td></td> </tr> <tr> <td>3/521 (0.58%)</td> <td>418 ± 8 ms</td> <td>-</td> </tr> </tbody> </table> | <u>Genotype</u> | <u>mean QTc</u> | <u>QTc Pen (%)<sup>a</sup></u> | LQT1 |  |  | 306/521 (58.7%) | 457 ± 38 ms | 55 | LQT2 |  |  | 164/521 (31.5%) | 467 ± 36 ms | 70 | LQT3 |  |  | 33/521 (6.3%) | 478 ± 52 ms | 79 | LQT5 |  |  | 15/521 (2.9%) | 438 ± 29 ms | 33 | LQT6 |  |  | 3/521 (0.58%) | 418 ± 8 ms | - |
| <u>Genotype</u>         | <u>mean QTc</u>              | <u>QTc Pen (%)<sup>a</sup></u>   |  |  |                 |                 |                                |      |  |  |                 |             |    |      |  |  |                 |             |    |      |  |  |               |             |    |      |  |  |               |             |    |      |  |  |               |            |   |
| LQT1                    |                              |  |  |  |                 |                 |                                |      |  |  |                 |             |    |      |  |  |                 |             |    |      |  |  |               |             |    |      |  |  |               |             |    |      |  |  |               |            |   |
| 306/521 (58.7%)         | 457 ± 38 ms                  | 55   |  |  |                 |                 |                                |      |  |  |                 |             |    |      |  |  |                 |             |    |      |  |  |               |             |    |      |  |  |               |             |    |      |  |  |               |            |   |
| LQT2                    |                              |  |  |  |                 |                 |                                |      |  |  |                 |             |    |      |  |  |                 |             |    |      |  |  |               |             |    |      |  |  |               |             |    |      |  |  |               |            |   |
| 164/521 (31.5%)         | 467 ± 36 ms                  | 70   |  |  |                 |                 |                                |      |  |  |                 |             |    |      |  |  |                 |             |    |      |  |  |               |             |    |      |  |  |               |             |    |      |  |  |               |            |   |
| LQT3                    |                              |  |  |  |                 |                 |                                |      |  |  |                 |             |    |      |  |  |                 |             |    |      |  |  |               |             |    |      |  |  |               |             |    |      |  |  |               |            |   |
| 33/521 (6.3%)           | 478 ± 52 ms                  | 79   |  |  |                 |                 |                                |      |  |  |                 |             |    |      |  |  |                 |             |    |      |  |  |               |             |    |      |  |  |               |             |    |      |  |  |               |            |   |
| LQT5                    |                              |  |  |  |                 |                 |                                |      |  |  |                 |             |    |      |  |  |                 |             |    |      |  |  |               |             |    |      |  |  |               |             |    |      |  |  |               |            |   |
| 15/521 (2.9%)           | 438 ± 29 ms                  | 33   |  |  |                 |                 |                                |      |  |  |                 |             |    |      |  |  |                 |             |    |      |  |  |               |             |    |      |  |  |               |             |    |      |  |  |               |            |   |
| LQT6                    |                              |  |  |  |                 |                 |                                |      |  |  |                 |             |    |      |  |  |                 |             |    |      |  |  |               |             |    |      |  |  |               |             |    |      |  |  |               |            |   |
| 3/521 (0.58%)           | 418 ± 8 ms                   | -  |  |  |                 |                 |                                |      |  |  |                 |             |    |      |  |  |                 |             |    |      |  |  |               |             |    |      |  |  |               |             |    |      |  |  |               |            |   |
| Priori et al (1999)     | III-2                        | Cross classification of patients on ECG, clinical assessment and mutational analysis | 9 families with sporadic LQTS <sup>b</sup> .<br>46 clinically unaffected family members plus 9 probands. | <p><b>Mutational analysis</b></p> <p>9/9 (100%) probands mutation carriers<br/>           15/46 (32.6%) family members mutation carriers<br/>           Penetrance<sup>c</sup> = 25%</p> <p><b>ECG criteria: QTc ≥ 470ms</b></p> <p>9/9 (100%) probands correctly identified<br/>           0/15 (0%) family members <i>with</i> mutation correctly identified</p> <p>Sensitivity ECG = 38%<br/>           Specificity ECG = 100%</p> <p><b>Clinical criteria<sup>d</sup></b></p> <p>9/9 (100%) probands correctly identified with clinical score ≥4<br/>           0/15 (0%) family members <i>with</i> mutation had a clinical score ≥ 4<br/>           4/15 (26.7%) family members <i>with</i> mutation had a clinical score ≥ 2</p> <p>Sensitivity clinical score ≥ 4 = 38%<br/>           Specificity clinical score ≥ 4 = 100%<br/>           Sensitivity clinical score ≥ 2 = 54%<br/>           Specificity clinical score ≥ 2 = 90%</p>   |                 |                 |                                |      |  |  |                 |             |    |      |  |  |                 |             |    |      |  |  |               |             |    |      |  |  |               |             |    |      |  |  |               |            |   |

| Study                  | Level of Diagnostic Evidence | Study Design                               | Population   | Outcomes  |
|------------------------|------------------------------|--|--|---|
| Jongbloed et al (1999) | IV                           | Case series of probands and family members | 24 probands with QTc >440ms, family history of sudden death or history of syncope, and family members (n=134)            | <p>Probands screened for LQT 1-5. Majority positive for LQT1 and LQT2. Only 14/24 families screened for LQT 1 and LQT2 as the remaining families were either too small, affected family members were deceased or clinical data was incomplete.</p> <p>84/134 (62.7%) genotype positive<br/>Of these<br/>66/84 (78.6%) were symptomatic carriers and<br/>18/84 (21.4%) were asymptomatic carriers<br/>50/134 (37.3%) genotype negative</p>   |
| Piippo et al (2001)    | IV                           | Case series of probands and family members | <p>2 probands with JNLS and family members (35 &amp; 28).<br/>114 unrelated probands with RWS and 625 family members</p> | <p><b>Mutational analysis of KCNQ1 gene</b></p> <p><u>Proband 1</u><br/>Homozygous for substitution mutation G589D<sup>b</sup><br/>35 family members genotyped<br/>1/35 (2.9%) homozygous G589D<br/>15/35 (42.9%) heterozygous G589D</p> <p><u>Proband 2</u><br/>Compound heterozygote for substitution mutation G589D<sup>e</sup> and the Y171X truncation<sup>f</sup><br/>28 family members genotyped<br/>9/28 (32.1%) heterozygous G589D<br/>4/28 (14.3%) heterozygous Y171X</p> <p><u>Unrelated RWS</u><br/>34/114 (29.8%) probands heterozygous G589D<br/>80/114 (70.2%) probands non-carriers<br/>625 family members screened<br/>282/625 (45.1%) heterozygous G589D<br/>343/625 (54.9) non-carriers</p> <p><u>Total probands and family members</u><br/>316/739 (42.8%) heterozygous G589D<br/>QTc 460 ± 40 ms<br/>423/739 (57.2%) non-carriers<br/>QTc 410 ± 20 ms <span style="float: right;"><i>p</i> &lt; 0.001</span></p> |
| Tester et al (2004)    | IV                           | Case study                                 | Pregnant woman with family history of LQTS undergoing amniocentesis  | Mother identified as positive for mutation in KCNQ1 gene. Amniocentesis revealed foetus positive for same mutation. β-blocker therapy initiated on baby at birth following detection of Bradycardia during labour.  |

LQTS = long QT syndrome, ECG = electrocardiogram, QTc = corrected QT interval, ms = milliseconds, JNLS = Jervell and Lange-Nielsen Syndrome

<sup>a</sup> QTc Pen = QTc penetrance is the percentage of genetically affected individuals with a prolonged QTc, <sup>b</sup> Sporadic LQT where only one family member is clinically affected, <sup>c</sup> penetrance defined as ratio of patients with clinical phenotype and the total number of family member carriers of mutation identified in proband, <sup>d</sup> clinical criteria see Table 2: Schwartz clinical score ≥ 4= high probability of LQTS, ≥ 2 = intermediate probability of LQTS, <sup>e</sup> G589D substitution mutation resulting in a substitution of aspartic acid for glycine at position 589, <sup>f</sup> Y171X truncation resulting in premature stop codon at position 171

Four studies were included for assessment which described mutational analysis in *unrelated* patients (Table 4). Tester et al (2005 & 2006b) and Sherman et al (2005) all reported on the same group of 541 consecutive unrelated patients who underwent comprehensive mutational analysis for LQT 1-3 and LQT 5-6 (level II diagnostic evidence). In contrast to the poor sensitivity reported by Priori et al (1999) (Table 3), it would appear that

clinical diagnosis based on the Diagnostic Criteria points allocation system and a corrected QT interval >480 ms are an accurate basis for diagnosis for LQTS as both of these factors were statistically significant in the genotype positive group ( $p < 0.0001$ ). The association between syncope, previous cardiac arrest or family history tended towards, but did not reach significance in genotype positive patients ( $p = 0.067$ ). A total of 211 putative LQTS mutations were found in 272/541 (50.3%) of patients, and of these more than half (125/211, 59%) were *novel* mutations not previously reported. The majority of mutations were missense mutations (154/210, 73%) and singleton mutations (only observed in a single patient) (165/210, 79%).

In the initial studies by Tester et al, 269/ 547 (49.7%) of clinically positive patients were found to be genotype negative. Targeted analysis for mutations in the ANK2 (LQT4) gene was then conducted by Sherman et al (2005). ANK2 encodes for the membrane adaptor protein ankyrin-B and was the first non-cardiac channel form of LQTS described, extending the LQTS field from channelopathies to electropathies. This study demonstrated the value in a *fully* comprehensive initial mutational analysis, as a further nine patients (3.3%) previously thought to be LQTS negative were found to be positive for the rare LQT4. These patients were initially diagnosed as “atypical” or borderline cases presenting with normal QTc, non-exertional syncope, U waves and/or sinus Bradycardia. In addition, five patients who were already diagnosed as LQTS positive were found to be compound heterozygotes with LQT4, which may have serious health consequences for these patients. Interestingly, a high number (13/200, 6.5%) of control patients (DNA obtained from a DNA bank) were also found to be positive for the ANK2 mutation, which the authors reported warranted further investigation considering that LQTS affects only 1:5,000 (0.02%) individuals.

Although Tester et al (2005 & 2006b) found good correlations between clinical symptoms and elongated QTc times, Splawski et al (2000) contradicted this finding (level IV diagnostic evidence). Splawski et al reported that of 262 unrelated patients tested for mutations in LQT1-3 and LQT 5-6, 32 per cent were genotype negative, however 81 per cent of these individuals had clinical symptoms and the average QTC was considered to be high at  $484 \pm 46$  ms.

Khositseth et al (2004) reported on a cohort of 260 unrelated women (level IV diagnostic evidence). Of these women, 14 (3.6%) reported a personal or family history of post-partum cardiac events, including aborted cardiac arrest, syncope and sudden cardiac death. The majority of mutations were reported in the KCNH2 (LQT2) gene (13/14, 92.9%). This strong association of LQT2 with post – partum cardiac events may facilitate strategic or targeted genotyping in individuals reporting a family history of post-partum cardiac events.

**Table 4 Molecular analysis for long QT syndrome in unrelated patients**

| Study                                       | Level of Diagnostic Evidence | Study Design   | Population  | Outcomes  |                                      |            |  |            |  |             |                               |             |                |         |          |    |      |  |  |  |                |         |          |    |      |  |  |  |               |         |          |    |      |  |  |  |              |         |          |    |      |  |  |  |              |         |         |    |                   |  |  |  |              |         |          |    |
|---|------------------------------|--|---|---|--------------------------------------|------------|--|------------|--|-------------|-------------------------------|-------------|----------------|---------|----------|----|------|--|--|--|----------------|---------|----------|----|------|--|--|--|---------------|---------|----------|----|------|--|--|--|--------------|---------|----------|----|------|--|--|--|--------------|---------|---------|----|-------------------|--|--|--|--------------|---------|----------|----|
| Sherman et al (2005)                        | II                           | Cross classification of patients on ECG, clinical assessment and mutational analysis | 541 consecutive unrelated patients suspected of LQTS                          | <p><b>Mutational analysis</b></p> <p>272/541 (50.3%) mutation carriers<br/>                 269/541 (49.7%) genotype negative for mutations in KCNQ1, KCNH2, SCN5A, KCNE1 and KCNE2 genes<br/>                 Targeted mutational analysis in ANK2 gene (LQT4)<br/>                 9/269 (3.3%) LQTS negative were positive for ANK2<br/>                 5/272 (1.8%) LQTS positive were positive for ANK2<br/>                 13/200 (6.5%) control DNA were positive for ANK2</p>   |                                      |            |  |            |  |             |                               |             |                |         |          |    |      |  |  |  |                |         |          |    |      |  |  |  |               |         |          |    |      |  |  |  |              |         |          |    |      |  |  |  |              |         |         |    |                   |  |  |  |              |         |          |    |
| Splawski et al (2000)                       | III-2                        | Cross classification of patients on ECG, clinical assessment and mutational analysis | 262 unrelated patients with LQTS based on elongated QTc and clinical symptoms | <p><b>Mutational analysis</b></p> <p>177/262 (68%) genotype positive</p> <table border="1"> <thead> <tr> <th></th> <th>Age</th> <th>QTc</th> <th>Symptoms %</th> </tr> </thead> <tbody> <tr> <td>LQT1</td> <td></td> <td></td> <td></td> </tr> <tr> <td>75/177 (42.4%)</td> <td>32 ± 19</td> <td>493 ± 45</td> <td>78</td> </tr> <tr> <td>LQT2</td> <td></td> <td></td> <td></td> </tr> <tr> <td>70/177 (45.2%)</td> <td>31 ± 19</td> <td>498 ± 48</td> <td>71</td> </tr> <tr> <td>LQT3</td> <td></td> <td></td> <td></td> </tr> <tr> <td>14/177 (7.9%)</td> <td>32 ± 24</td> <td>511 ± 42</td> <td>55</td> </tr> <tr> <td>LQT5</td> <td></td> <td></td> <td></td> </tr> <tr> <td>5/177 (2.8%)</td> <td>43 ± 16</td> <td>457 ± 25</td> <td>40</td> </tr> <tr> <td>LQT6</td> <td></td> <td></td> <td></td> </tr> <tr> <td>3/177 (1.7%)</td> <td>54 ± 20</td> <td>457 ± 5</td> <td>67</td> </tr> <tr> <td>Genotype negative</td> <td></td> <td></td> <td></td> </tr> <tr> <td>85/262 (32%)</td> <td>25 ± 16</td> <td>484 ± 46</td> <td>81</td> </tr> </tbody> </table>  |                                      | Age        | QTc                                    | Symptoms % | LQT1                                   |             |                               |             | 75/177 (42.4%) | 32 ± 19 | 493 ± 45 | 78 | LQT2 |  |  |  | 70/177 (45.2%) | 31 ± 19 | 498 ± 48 | 71 | LQT3 |  |  |  | 14/177 (7.9%) | 32 ± 24 | 511 ± 42 | 55 | LQT5 |  |  |  | 5/177 (2.8%) | 43 ± 16 | 457 ± 25 | 40 | LQT6 |  |  |  | 3/177 (1.7%) | 54 ± 20 | 457 ± 5 | 67 | Genotype negative |  |  |  | 85/262 (32%) | 25 ± 16 | 484 ± 46 | 81 |
|   | Age                          | QTc  | Symptoms %  |   |                                      |            |  |            |  |             |                               |             |                |         |          |    |      |  |  |  |                |         |          |    |      |  |  |  |               |         |          |    |      |  |  |  |              |         |          |    |      |  |  |  |              |         |         |    |                   |  |  |  |              |         |          |    |
| LQT1  |                              |  |   |   |                                      |            |  |            |  |             |                               |             |                |         |          |    |      |  |  |  |                |         |          |    |      |  |  |  |               |         |          |    |      |  |  |  |              |         |          |    |      |  |  |  |              |         |         |    |                   |  |  |  |              |         |          |    |
| 75/177 (42.4%)                              | 32 ± 19                      | 493 ± 45   | 78  |   |                                      |            |  |            |  |             |                               |             |                |         |          |    |      |  |  |  |                |         |          |    |      |  |  |  |               |         |          |    |      |  |  |  |              |         |          |    |      |  |  |  |              |         |         |    |                   |  |  |  |              |         |          |    |
| LQT2  |                              |  |   |   |                                      |            |  |            |  |             |                               |             |                |         |          |    |      |  |  |  |                |         |          |    |      |  |  |  |               |         |          |    |      |  |  |  |              |         |          |    |      |  |  |  |              |         |         |    |                   |  |  |  |              |         |          |    |
| 70/177 (45.2%)                              | 31 ± 19                      | 498 ± 48   | 71  |   |                                      |            |  |            |  |             |                               |             |                |         |          |    |      |  |  |  |                |         |          |    |      |  |  |  |               |         |          |    |      |  |  |  |              |         |          |    |      |  |  |  |              |         |         |    |                   |  |  |  |              |         |          |    |
| LQT3  |                              |  |   |   |                                      |            |  |            |  |             |                               |             |                |         |          |    |      |  |  |  |                |         |          |    |      |  |  |  |               |         |          |    |      |  |  |  |              |         |          |    |      |  |  |  |              |         |         |    |                   |  |  |  |              |         |          |    |
| 14/177 (7.9%)                               | 32 ± 24                      | 511 ± 42   | 55  |   |                                      |            |  |            |  |             |                               |             |                |         |          |    |      |  |  |  |                |         |          |    |      |  |  |  |               |         |          |    |      |  |  |  |              |         |          |    |      |  |  |  |              |         |         |    |                   |  |  |  |              |         |          |    |
| LQT5  |                              |  |   |   |                                      |            |  |            |  |             |                               |             |                |         |          |    |      |  |  |  |                |         |          |    |      |  |  |  |               |         |          |    |      |  |  |  |              |         |          |    |      |  |  |  |              |         |         |    |                   |  |  |  |              |         |          |    |
| 5/177 (2.8%)                                | 43 ± 16                      | 457 ± 25   | 40  |   |                                      |            |  |            |  |             |                               |             |                |         |          |    |      |  |  |  |                |         |          |    |      |  |  |  |               |         |          |    |      |  |  |  |              |         |          |    |      |  |  |  |              |         |         |    |                   |  |  |  |              |         |          |    |
| LQT6  |                              |  |   |   |                                      |            |  |            |  |             |                               |             |                |         |          |    |      |  |  |  |                |         |          |    |      |  |  |  |               |         |          |    |      |  |  |  |              |         |          |    |      |  |  |  |              |         |         |    |                   |  |  |  |              |         |          |    |
| 3/177 (1.7%)                                | 54 ± 20                      | 457 ± 5  | 67  |   |                                      |            |  |            |  |             |                               |             |                |         |          |    |      |  |  |  |                |         |          |    |      |  |  |  |               |         |          |    |      |  |  |  |              |         |          |    |      |  |  |  |              |         |         |    |                   |  |  |  |              |         |          |    |
| Genotype negative                           |                              |  |   |   |                                      |            |  |            |  |             |                               |             |                |         |          |    |      |  |  |  |                |         |          |    |      |  |  |  |               |         |          |    |      |  |  |  |              |         |          |    |      |  |  |  |              |         |         |    |                   |  |  |  |              |         |          |    |
| 85/262 (32%)                                | 25 ± 16                      | 484 ± 46   | 81  |   |                                      |            |  |            |  |             |                               |             |                |         |          |    |      |  |  |  |                |         |          |    |      |  |  |  |               |         |          |    |      |  |  |  |              |         |          |    |      |  |  |  |              |         |         |    |                   |  |  |  |              |         |          |    |
| Tester et al (2005) and Tester et al (2006) | II                           | Cross classification of patients on ECG, clinical assessment and mutational analysis | 541 consecutive unrelated patients suspected of LQTS                          | <p><b>Mutational analysis</b></p> <p>272/541 (50.3%) mutation carriers. With a total of 211 different putative mutations detected: 88 in KCNQ1, 89 in KCNH2, 32 in SCN5A, 1 in KCNE1 and 1 in KCNE2</p> <p>120/272 (44.1%) LQT1<br/>                 93/272 (34.2%) LQT2<br/>                 26/272 (9.6%) LQT3<br/>                 3/272 (1.1%) LQT5<br/>                 1/272 (0.4%) LQT6</p> <p>29/272 (10.7%) had 2 LQT mutations</p> <p>269/541 (49.7%) genotype negative</p> <p><b>ECG criteria: QTc ≥ 480ms</b></p> <p>152/268<sup>a</sup> (56.7%) had a QTc ≥ 480ms</p> <table border="1"> <tbody> <tr> <td>Mean QTc of single mutation patients</td> <td>493 ± 50ms</td> </tr> <tr> <td>Mean QTc of multiple mutation patients</td> <td>506 ± 57ms</td> </tr> <tr> <td>Mean QTc of all patients with mutation</td> <td>494 ± 51 ms</td> </tr> <tr> <td>Mean QTc of genotype negative</td> <td>470 ± 60 ms</td> </tr> </tbody> </table> <p>QTc of genotype positive patients significant compared to genotype negative<br/> <math>p &lt; 0.0001</math></p> | Mean QTc of single mutation patients | 493 ± 50ms | Mean QTc of multiple mutation patients | 506 ± 57ms | Mean QTc of all patients with mutation | 494 ± 51 ms | Mean QTc of genotype negative | 470 ± 60 ms |                |         |          |    |      |  |  |  |                |         |          |    |      |  |  |  |               |         |          |    |      |  |  |  |              |         |          |    |      |  |  |  |              |         |         |    |                   |  |  |  |              |         |          |    |
| Mean QTc of single mutation patients        | 493 ± 50ms                   |  |   |   |                                      |            |  |            |  |             |                               |             |                |         |          |    |      |  |  |  |                |         |          |    |      |  |  |  |               |         |          |    |      |  |  |  |              |         |          |    |      |  |  |  |              |         |         |    |                   |  |  |  |              |         |          |    |
| Mean QTc of multiple mutation patients      | 506 ± 57ms                   |  |   |   |                                      |            |  |            |  |             |                               |             |                |         |          |    |      |  |  |  |                |         |          |    |      |  |  |  |               |         |          |    |      |  |  |  |              |         |          |    |      |  |  |  |              |         |         |    |                   |  |  |  |              |         |          |    |
| Mean QTc of all patients with mutation      | 494 ± 51 ms                  |  |   |   |                                      |            |  |            |  |             |                               |             |                |         |          |    |      |  |  |  |                |         |          |    |      |  |  |  |               |         |          |    |      |  |  |  |              |         |          |    |      |  |  |  |              |         |         |    |                   |  |  |  |              |         |          |    |
| Mean QTc of genotype negative               | 470 ± 60 ms                  |  |   |   |                                      |            |  |            |  |             |                               |             |                |         |          |    |      |  |  |  |                |         |          |    |      |  |  |  |               |         |          |    |      |  |  |  |              |         |          |    |      |  |  |  |              |         |         |    |                   |  |  |  |              |         |          |    |

| Study                   | Level of Diagnostic Evidence | Study Design | Population   | Outcomes  |
|-------------------------|------------------------------|--------------|--|---|
|                         |                              |              |  | <p><b>Clinical criteria<sup>b</sup></b><br/> <u>Schwartz score <math>\geq 4</math></u><br/>           108/268<sup>a</sup> (40.3%) genotype positive<br/>           46/269 (17%) of genotype negative<br/>           Schwartz score <math>\geq 4</math> in mutation patients significant compared to genotype negative <math>p &lt; 0.0001</math></p> <p><u>Family history</u><br/>           124/268<sup>a</sup> (46.3%) genotype positive<br/>           102/269 (38%) of genotype negative<br/>           Family history trended towards an increase in mutation positive patients <math>p = 0.067</math></p> |
| Khositseth et al (2004) | IV                           | Case series  | 260 unrelated women suspected of LQTS. Mutational analysis on those women with a personal or family history of post-partum cardiac events. | 14/260 (3.6%) probands with personal or family history of post-partum cardiac events<br>4/14 (28.6%) with personal history of post-partum experienced 7 cardiac events including aborted cardiac arrest (n=2), syncope (n=4) and SCD (n=1)<br>13/14 (92.9%) with mutations in KCNH2 (LQT2) gene<br>1/14 (7.1%) with mutation in KCNQ1 (LQT1) gene<br>Average time from delivery to cardiac event was $10.5 \pm 5.2$ weeks (range 1 hour to 20 weeks, median 8 weeks)  |

LQTS = long QT syndrome, ECG = electrocardiogram, QTc = corrected QT interval, ms = milliseconds, SCD = sudden cardiac death  
<sup>a</sup> data not given for the 4 LQT5 and LQT6 patients, <sup>b</sup> clinical criteria see Table 2 : Schwartz clinical score  $\geq 4$ = high probability of LQTS,  $\geq 2$  = intermediate probability of LQTS

In conclusion, clinical symptoms and an elongated QTc may not necessarily predict LQT and a negative mutation analysis may not rule out a diagnosis of LQTS. In the majority of cases a mutation was found in a single family or an individual. This has implications for the screening of family members as once a proband has been thoroughly screened; family members can be probed for that one mutation with a moderate degree of surety. Successful population screening programmes usually screen for a disease that has a high prevalence, which once diagnosed will result in treatment and a change of management. Genetic screening for LQTS may be considered targeted screening, not population screening. Prevalence of LQTS is low in the general population; however prevalence in the targeted population (ie family members of probands) is high. No papers reported on long term outcomes of individuals who were found to be genotype positive and any subsequent treatment they may have received, or long-term outcomes for those individuals found to be genotype negative. Several papers reported on treatment outcomes for individuals considered positive for LQTS, however they were not included for assessment as they did not report the results of mutational analysis (Goldenberg et al 2006; Hobbs et al 2006).

### Cost Analysis

Phillips et al (2005) conducted a cost-effectiveness analysis of genetic testing for familial LQTS in *symptomatic index cases*. The population in this study was aged 15-40 years, as little is known about LQTS after the age of 40 years. Index cases were assumed to have a compatible family history and clinical presentation consistent with LQTS. The expected cost-effectiveness of genetic testing of *first-degree relatives* or more distant relatives was not included in this analysis. The three most common mutations were examined in the KCNQ1, KCNH2 and the SCN5A genes.

Genetic testing of probands was found to be cost-effective compared to no genetic testing, at a cost per year life saved of US\$2,500, well below the standard threshold of US\$50,000 per life-year saved often used to define a cost-effective intervention (Gold et al 1996). An extensive sensitivity analysis was conducted and the results were generally robust. If the mortality rate for untreated individuals *increases*, the cost-effectiveness of testing *increases* eg if the mortality rate doubles to 30%, cost-effectiveness of testing *decreases* to US\$1,200 per year of life saved. However, testing becomes both more costly and less effective if the mortality rate falls below 1.5%. In addition, as the cost of implantable cardiac defibrillators (ICD) increases, mutational analysis becomes more cost-effective. This is due to the fact that with a definitive diagnosis only a small proportion of patients will receive an ICD, however based on a clinical diagnosis alone, many “borderline” patients would receive an ICD although it is unlikely they will benefit from this preventative therapy. The benefit of mutational analysis for LQTS is that patients are more accurately diagnosed and therefore treated appropriately. A further cost-effectiveness analysis is required to consider the benefits of genetic testing of family members of the proband (Phillips et al 2005).

In New Zealand, the cost of a five gene scan is NZ\$3,000 (screening for LQT 1-3, 5, 6). If a mutation is found in the proband, the cost of familial screening is markedly reduced (NZ\$300) as only the one target mutation is screened for. In New Zealand the cost for screening is covered by the health service. Patients referred from Australia are not directly billed, however the referring clinician or hospital are billed, which may result in the patient bearing the cost (personal communication CIDG May 2006).

A commercial test, FAMILION<sup>®</sup>, is available in the United States produced by PGxHealth<sup>™</sup>. FAMILION<sup>®</sup> is designed to identify mutations in five of the major cardiac ion channel genes. Physicians in the United States take a blood sample from patients, which are then shipped back to PGxHealth<sup>™</sup> for analysis. It is stressed that FAMILION<sup>®</sup> should not be used to exclude the diagnosis of LQTS and that the test is expected to identify 50-75% of mutations that cause LQTS. The test can be conducted in three configurations:

- The comprehensive cardiac ion channel analysis for mutations in the five genes. Analysing 2,331 base pairs (bp) in the KCNQ1 gene, 3,750

bp in the KCNH2 gene, 6,571 bp in the SCN5A gene, 390 bp in the KCNE1 gene and 372 bp in the KCNE2 gene. This analysis is recommended when there is a high index of suspicion of disease such as stress-induced syncope, prolonged QT interval, family history of sudden cardiac death and/or unexplained ventricular tachycardia or fibrillation, or *torsade de pointes*. The cost of full five gene analysis is US\$5,400.

- Sodium channel analysis for mutations in only the SCN5A gene and should be conducted in cases of suspected Brugada Syndrome. The cost of this analysis is US\$2,700.
- Family specific analysis for mutations identified in an index case using either one of the above test configurations and is appropriate for testing blood relatives for this mutation. The cost of this analysis is US\$900 (PGxHealth 2006).

Goldenberg et al (2005) conducted a cost-effectiveness study of implanted cardiac defibrillators (ICDs) in young people with inherited LQTS (Goldenberg et al 2005). A computer-based analytical model was used to compare non-ICD with ICD therapy. Annual probabilities of clinical events including heart failure or sudden cardiac death were used to analyse the course of disease, with outcomes defined as costs per QALY<sup>6</sup>. The analysis included non-health care costs for example the potential for increased productivity attributable to improved survival from the prevention of sudden cardiac death. Costs were estimated in US dollars, with the cost of the defibrillator as US\$17,000, cost of initial ICD implantation of US\$5,000 and the cost of physician visits (three per year) estimated to be US\$125 per visit. According to the recent MSAC report, these costs are comparable to those in Australia when implantation takes place in a public hospital (Table 5) (MSAC 2006).

**Table 5** Implantation costs of ICD device in Australia

| Cost components      | Public hospital (\$) | Private hospital (\$) |
|----------------------|----------------------|-----------------------|
| Hospital cost        | 7,284                | 6,558                 |
| ICD device           | 13,000               | 35,000                |
| Right atrium lead    | 475                  | 1,350                 |
| Right ventricle lead | 1,500                | 8,750                 |
| <b>Total</b>         | <b>22,259</b>        | <b>51,658</b>         |

Results of the Goldenberg et al (2005) study demonstrated that ICDs were cost-effective in high-risk males<sup>7</sup> (QTc >0.5s and/or prior syncope) with an ICER<sup>8</sup> of US\$3,328 per QALY saved and cost saving in high-risk females with an ICER of US\$7,102 gained per QALY saved. Implantation with ICDs was cost-saving in *very* high-risk males and females (cardiac arrest survivor or sustained spontaneous ventricular tachycardia) with an ICER of US\$15,483 and US\$19,393 *gained* per QALY saved, respectively. Implantation with an ICD was not cost-effective for low-risk LQTS patients (QTc ≤0.5s and no

<sup>6</sup> QALY = quality adjusted life year

<sup>7</sup> The risk of cardiac events in LQTS patients is affected by age and gender.

<sup>8</sup> ICER = incremental cost-effectiveness ratio

prior syncope) with an ICER range of US\$400-600,000 *lost* per QALY saved. The authors concluded that in appropriately selected high-risk patients, early intervention with ICD therapy is cost-effective and may result in cost savings due to improved productivity, especially in light of the extended life expectancy of individuals who may otherwise have suffered a sudden cardiac death event (Goldenberg et al 2005).

## Ethical Considerations

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Genetic testing raises a number of ethical issues beyond those usually discussed for new technologies. *Diagnostic* testing is generally not controversial, since it is usually done in the context of providing certainty for establishing an appropriate treatment regime. *Predictive* genetic testing is of ethical concern for a range of reasons that, in this report, will only be touched on briefly. A full discussion of the ethical issues raised by genetic testing and screening can be found in (Chadwick 1999).

### Informed Consent

There are a number of issues of ethical significance that arise when genetic testing is contemplated. Genetic information is complex and seeking the informed consent of patients poses particular challenges for clinicians and counsellors. Clinicians and counsellors need to stress that testing for LQTS is voluntary and optional. Information must be provided in a format that the patient can understand, with particular emphasis on the accuracy of the diagnosis and the fact that test results will not provide definitive information about whether an acute cardiac event will actually occur. The limits of other methods for predictive testing need to be discussed.

### Minimising the risk of harm

The risks that attach to genetic screening are generally psychological and social. Receiving the results of genetic tests often causes considerable anxiety for patients. This may be particularly acute for parents who receive a diagnosis of LQTS in their child. For example, Hendriks et al (2005) found that parents of children who were identified as at risk for LQTS did *not* appear to adjust to the diagnosis over time (Hendriks et al 2005a; Hendriks et al 2005b). They remained distressed and anxious, despite the fact that their child was being treated prophylactically. Such findings are inconsistent with those of studies of parental response to the diagnosis of familial cancer, suggesting that the possibility of sudden death in a child, even when prophylactic treatment is being provided, is acutely distressing. Hendriks et al recommend that anxiety may be alleviated by providing parents with periodic updates as new developments in cardiac genetics occur.

### Genetic privacy

Genetic tests raise specific concerns about privacy. Patients are likely to be concerned about who will have access to test results, how the information will be used and for what purposes. They may be worried that third parties, such as health or life insurers, employers, or financial institutions, may use genetic information to discriminate against them. Confidentiality and privacy in these circumstances is particularly important (Beauchamp & Childress 2001).

Adequate counselling consists not only of helping the person understand and deal with the information, but also judging how much information that person is prepared to divulge, and how much information that person is prepared to acknowledge and handle at a given moment (Liebman 2001).

Family members may also wish to have access to the results of genetic testing for LQTS. In this respect, genetic information is unlike other health information in that, while it is personal and private, it is also familial and non-individual. A number of authors have suggested that clinicians may have a duty to warn at risk family members so that these people can adopt early monitoring and prophylactic treatment if necessary (Liebman 2001). This duty may be justified on two grounds: first, the clinician may regard the entire *family* as his or her patient; in this case informing other members of the family of their genetic risk is not a breach of confidentiality. Second, the clinician may judge that there is a serious risk of significant harm sufficient to warrant breaching confidentiality (Rogers & Braunack-Mayer 2004). Counselling the proband about the need to share information with relatives and then providing genetic information and counselling to those relatives, whether they are affected or not, requires considerable skill and sensitivity.

### **Access Issues**

Suspected LQTS patients from both rural and remote areas, and metropolitan centres would have a blood sample taken by their general practitioner, which would then be sent away for analysis. Currently few centres offer mutational screening for LQTS, with the majority of samples from Australia being sent to New Zealand for analysis. However, patients referred from Australia may bear the cost of this testing, which may be prohibitive for some patients.

### Training

Molecular diagnosis of LQTS would require a dedicated molecular pathology laboratory and a specialist molecular pathologist trained in PCR, molecular sequencing and denatured HPLC, and the interpretation of the results of these techniques.

### Clinical Guidelines

The Cardiovascular Genetics Working Group, comprising members of the Cardiac Society of Australia and New Zealand, recently published “*Guidelines for the Diagnosis and Management of Familial Long QT Syndrome*” (Skinner 2007).

#### *Diagnosis*

LQTS patients often present with syncope or sudden death following an episode of severe emotional stress, strenuous exercise or sudden loud noise. Patients are commonly misdiagnosed as suffering from epilepsy. Seizures experienced following exercise, during sleep or arousal from sleep, should immediately suggest the possibility of LQTS. Diagnosis of LQTS is usually made on clinical grounds satisfying the criteria as outlined in Table 2. QT prolongation due to drugs, biochemical imbalance (low potassium, calcium or magnesium), hypothermia and myocardial disease must be excluded. A detailed family history of close relatives should be taken looking for a history of syncope or sudden unexplained death, including drowning or road traffic accidents. In addition, familial epilepsy (possible misdiagnosis) and sudden infant death syndrome in close family members should be investigated. Sudden death in a family member with a negative post-mortem should be thoroughly investigated. All first-degree relatives should then undergo ECG testing, however one third of asymptomatic gene mutation carriers will have a corrected QT interval (QTc) in the normal range. Exercise testing may also be helpful. For complete certainty, genetic testing of family members should be carried out. Eight genetic forms of LQTS are known, however it should be stressed that *approximately one third of families with LQTS do not yet have a recognised genetic locus*. Once the proband has been identified via a clinical diagnosis, molecular analysis of all known mutations is carried out. Once the mutation is identified in the proband, then familial screening can take place. Counselling is recommended before mutational analysis takes place.

#### *Treatment*

Beta-blockers should be prescribed for symptomatic individuals, those with a definite long QT interval and especially for young or infant patients with LQT1. Administration of beta-blockers results in the up-regulation of beta receptors, therefore, once therapy has commenced, treatment should be continuous as withdrawal of this medication presents a high risk to patients. LQT1 patients, and to a lesser degree LQT2 patients, benefit greatly from

treatment with beta-blockers. LQT3 patients may benefit from implantation with a pacemaker, which would then allow treatment with beta-blockers.

Implantable cardioverter defibrillators (ICD) are indicated for patients who have experienced resuscitated cardiac arrest, persistent syncope whilst on beta-blockers or for patients for whom beta-blockers are contraindicated. ICDs are recommended for asymptomatic patients with a very long QT interval ( $QTc > 0.55s$ ) and in particular males with LQT3. Unless contraindicated, beta-blocker therapy should be continued in these patients to avert the risk of defibrillation shock, which may cause an adrenergic shock, in turn giving rise to a further event or ventricular tachyarrhythmia electrical storm.

For patients with severe LQTS for whom beta-blockers and ICDs are contraindicated, those with LQT3, those implanted with an ICD who experience ventricular tachyarrhythmia electrical storms, or those who experience events whilst asleep, left cervical sympathectomy may be considered. This surgical procedure is rarely performed as it involves cutting the nerves in the neck, which control automatic functions in the body including regulation of heart rhythm.

#### *Asymptomatic family members*

If LQTS cannot be excluded, family members should avoid medications contraindicated for LQTS. Asymptomatic individuals with an elongated QT interval should be treated as if they had experienced syncope, and be prescribed beta-blockers and limit sporting activity. Treatment with beta-blockers of asymptomatic individuals who have a normal QT interval *but* a genetic diagnosis remains controversial.

In 1982, Italy introduced a nationwide systematic screening programme for individuals participating in organised athletic competition, aimed at reducing the incidence of sudden cardiac death in young athletes (Corrado et al 2006). It was hypothesised that medical evaluation of athletic populations prior to competition would identify asymptomatic individuals with cardiovascular abnormalities such as hypertrophic cardiomyopathy and LQTS. Italian law mandates that all adolescents and young adults, aged 12-35 years, involved in an organised competitive sport undergo a clinical evaluation to obtain eligibility. Clinical evaluation consists of the taking of a family and personal history, a physical examination and a 12-lead ECG. Since the introduction of this screening programme, the incidence of sudden cardiac death in young athletes was found to be reduced significantly ( $p < 0.001$ ) from 3.6 to 0.4 per 100,000 person-years, in 1979 and 2004, respectively.

## Limitations of the Assessment

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Methodological issues and the relevance or currency of information provided over time are paramount in any assessment carried out in the early life of a technology.

Horizon Scanning forms an integral component of Health Technology Assessment. However, it is a specialised and quite distinct activity conducted for an entirely different purpose. The rapid evolution of technological advances can in some cases overtake the speed at which trials or other reviews are conducted. In many cases, by the time a study or review has been completed, the technology may have evolved to a higher level leaving the technology under investigation obsolete and replaced.

An Horizon Scanning Report maintains a predictive or speculative focus, often based on low level evidence, and is aimed at informing policy and decision makers. It is not a definitive assessment of the safety, effectiveness, ethical considerations and cost effectiveness of a technology.

In the context of a rapidly evolving technology, an Horizon Scanning Report is a 'state of play' assessment that presents a trade-off between the value of early, uncertain information, versus the value of certain, but late information that may be of limited relevance to policy and decision makers.

This report provides an assessment of the current state of development of genetic testing for congenital long QT syndrome, its present and potential use in the Australian public health system, and future implications for the use of this technology.

### Search Strategy used for the Report

The medical literature (Table 7) was searched utilising the search terms outlined in Table 6 to identify relevant studies and reviews, until January 2007. In addition, major international health assessment databases were searched.

**Table 6 Search terms utilised**

| Search terms   |
|--|
| <b>MeSH</b><br>Long QT syndrome; Death, sudden, cardiac; Genetic screening; Molecular diagnostic techniques; DNA mutational analysis; Polymerase chain reaction; Sequence analysis |
| <b>Text words</b><br>Long QT syndrome; sudden cardiac death; genetic screen*; genetic test; molecular diagnos*; molecular diagnos* technique*; clinical screen*                    |
| <b>Limits</b><br>English, Human  |

**Table 7 Literature sources utilised in assessment**

| Source  | Location  |
|---|---|
| <i>Electronic databases</i>   |   |
| AustHealth  | University library  |
| Australian Medical Index  | University library  |
| Australian Public Affairs Information Service (APAIS) - Health  | University library  |
| Cinahl  | University library  |
| Cochrane Library – including, Cochrane Database of Systematic Reviews, Database of Abstracts of Reviews of Effects, the Cochrane Central Register of Controlled Trials (CENTRAL), the Health Technology Assessment Database, the NHS Economic Evaluation Database | University library  |
| Current Contents  | University library  |
| Embase  | Personal subscription   |
| Pre-Medline and Medline   | University library  |
| ProceedingsFirst  | University library  |
| PsycInfo  | University library  |
| Web of Science – Science Citation Index Expanded  | University library  |
| <i>Internet</i>   |   |
| Australian Clinical Trials Registry   | <a href="http://www.actr.org.au/default.aspx">http://www.actr.org.au/default.aspx</a>                                 |
| Current Controlled Trials metaRegister  | <a href="http://controlled-trials.com/">http://controlled-trials.com/</a>   |
| Health Technology Assessment international  | <a href="http://www.htai.org">http://www.htai.org</a>   |
| International Network for Agencies for Health Technology Assessment   | <a href="http://www.inahta.org/">http://www.inahta.org/</a>   |
| Medicines and Healthcare products Regulatory Agency (UK).   | <a href="http://www.medical-devices.gov.uk/">http://www.medical-devices.gov.uk/</a>                                   |
| National Library of Medicine Health Services/Technology Assessment Text   | <a href="http://www.ncbi.nlm.nih.gov/books/bv.fcgi?rid=hstat">http://www.ncbi.nlm.nih.gov/books/bv.fcgi?rid=hstat</a> |
| National Library of Medicine Locator Plus database  | <a href="http://locatorplus.gov">http://locatorplus.gov</a>   |
| New York Academy of Medicine Grey Literature Report   | <a href="http://www.nyam.org/library/grey.shtml">http://www.nyam.org/library/grey.shtml</a>                           |
| Trip database   | <a href="http://www.tripdatabase.com">http://www.tripdatabase.com</a>   |
| U.K. National Research Register   | <a href="http://www.update-software.com/National/">http://www.update-software.com/National/</a>                       |
| US Food and Drug Administration, Center for Devices and Radiological Health.  | <a href="http://www.fda.gov/cdrh/databases.html">http://www.fda.gov/cdrh/databases.html</a>                           |
| Websites of Specialty Organisations   | Dependent on technology topic area  |

## Availability and Level of Evidence

Nine peer reviewed studies were included for assessment in this Horizon Scanning Report. Five studies reported on LQTS mutational analysis of probands and the prevalence of these mutations in their family members. One study was comparative, reporting cross classification of patients on mutational status compared to QTc interval and clinical symptoms (level III-2 diagnostic evidence) (Priori et al 1999). The remaining studies were either case series (level IV diagnostic evidence) (Jongbloed et al 1999; Napolitano et al 2005; Piippo et al 2001) or a case study (Tester et al 2004). Four studies reported on LQTS mutational analysis in *unrelated* patients. Two studies were comparative, reporting cross classification of patients on mutational status compared to QTc interval and clinical symptoms. In one of these studies, mutational status was blinded to the clinical results (level II diagnostic evidence) (Tester et al 2005). Additional information on this study was supplied in a later paper by the same author (Tester et al 2006b). In a further

study, the same patients underwent mutational analysis for a rare LQTS mutation (Sherman et al 2005). The remaining comparative study was not blinded to clinical results (Splawski et al 2000). One case series was included for assessment as it reported on the prevalence mutations in a cohort of unrelated women who had a personal or family history of post-partum cardiac events (Khositseth et al 2004). See Appendix C for profiles of these studies.

## Sources of Further Information

In 1979 the International Long QT Syndrome Registry was established within the University of Rochester, United States. The Registry maintains an international database on LQTS with the aim of improving understanding of the genetics and natural history of the rare disorder, and to improve treatment options for affected individuals (Moss & Schwartz 2005).

There are currently over 30 studies being conducted, or have just been completed, in the Drug-Induced Arrhythmia Risk Evaluation (DARE) study. Many of these studies are using the same patient group with different outcomes measured. The details of some of these studies, listed on the United Kingdom's Department of Health register of clinical trials web site, are outlined in Table 8 (DoH 2007).

**Table 8** Ongoing or recently completed studies into long QT syndrome

| Investigators  | Study aim  | Completion date |
|--|--|-----------------|
| Department of Medical Genetics, Aberdeen, UK                                 | To discover the frequency with which mutations in the troponin T gene and the <b>QT genes</b> can be detected in sudden adult death syndrome.  | 31 March 2008   |
| Medical Genetics Dept, St George's Hospital, London, UK                      | To determine what percentage of clinically diagnosed individuals with <b>LQTS</b> can have a molecular diagnosis that will show which gene is mutated in their particular instance, and to examine which methods and possible diagnostic flow mechanisms provide the most effective diagnosis in terms of cost and time.   | 1 January 2008  |
| Department of Clinical Genetics, AlderHey Children's Hospital, Liverpool, UK | What is the experience like for children who have undergone pre-symptomatic testing for <b>Long QT Syndrome</b> ?<br>The study is retrospective and requires a qualitative approach to explore in depth the experience of families and in particular adolescents offered predictive testing. Through interview the investigator aims to highlight common themes amongst young people about the way in which they would like to be prepared for genetic testing. The research will involve interviewing parents and with their consent any children between the ages of 12 and 18 years who have been offered pre-symptomatic genetic testing for <b>Long QT Syndrome</b> . | 8 July 2006     |
| Drug Safety Research Unit, Southampton, UK                                   | Drug-induced Arrhythmia Risk Evaluation (DARE STUDY)<br>The principal components and aims are:(1) an epidemiological study to document and follow up cases of drug-induced arrhythmia and compare them with controls; (2) a genetic study to analyse blood samples from cases and controls for mutations and polymorphisms of the cardiac sodium and potassium ion channel genes implicated in the <b>long QT</b> and Brugada syndromes.   | 1 February 2007 |

| Investigators  | Study aim   | Completion date |
|--|---|-----------------|
| Institute Of Medical Genetics, Cardiff University, University Hospital of Wales, Cardiff, UK | <p>Family's Perspective on the inheritance of Long QT syndrome.</p> <p>Would an increase in cardiac genetics knowledge in the cardiac nursing staff have helped during this period of time?</p> <p>To provide an accurate reflection of the emotional outcomes families experience during a period of time when heredity, family illness and possibly a history of sudden death in someone young are high in their thoughts.</p>  | 1 December 2006 |
| Cardiology, Royal United Hospital, Bath, UK  | <p>Drug-induced Arrhythmia Risk Evaluation (DARE STUDY)</p> <p>This project's principle components and aims are: (1) an epidemiological study to systematically document and follow-up incident cases in England and compare them to controls; (2) a genetic study to analyse blood samples from cases and controls for mutations and polymorphisms of the cardiac sodium and potassium ion channel genes implicated in the <b>Long QT</b> and Brugada syndromes. We hypothesise that there is a significant association of genotype with drug-induced arrhythmia. The relative risk of predisposing clinical factors and genetic status will be calculated using conditional logistic regression. Both epidemiological cohorts will be described and outcomes compared. The predictability and awareness of the condition will thus be increased and result in safer prescribing.</p>                                      | 1 July 2008     |
| Cardiological Sciences, St Georges Hospital, London, UK                                      | <p>Drug-induced Arrhythmia Risk Evaluation (DARE STUDY)</p> <p>This project's principle components and aims are: (1) an epidemiological study to systematically document and follow-up incident cases in England and compare them to controls; (2) a genetic study to analyse blood samples from cases and controls for mutations and polymorphisms of the cardiac sodium and potassium ion channel genes implicated in the <b>Long QT</b> and Brugada syndromes. We hypothesise that there is a significant association of genotype with drug-induced arrhythmia. The relative risk of predisposing clinical factors and genetic status will be calculated using conditional logistic regression. Both epidemiological cohorts will be described and outcomes compared. The predictability and awareness of the condition will thus be increased and result in safer prescribing.</p>                                      | 31 July 2008    |
| Cardiology Research Unit, Oldchurch Hospital, Romford, UK                                    | <p>Drug-induced Arrhythmia Risk Evaluation (DARE STUDY)</p> <p>This project's principle components and aims are: (1) an epidemiological study to systematically document and follow-up incident cases in England and compare them to controls; (2) a genetic study to analyse blood samples from cases and controls for mutations and polymorphisms of the cardiac sodium and potassium ion channel genes implicated in the <b>Long QT</b> and Brugada syndromes. We hypothesise that there is a significant association of genotype with drug-induced arrhythmia.</p>  | 1 July 2008     |
| St George's Hospital Medical School, London, UK  | <p>DARE - Drug Induced Arrhythmia Risk Evaluation</p> <p>The objectives are to: establish a unique sample of cases of drug-induced arrhythmic events reported throughout England and representative of the national population, estimate the contribution of individual drugs to the incidence of drug-induced arrhythmic events, look for differences in overall mortality and cardiovascular mortality and morbidity between the drug-induced arrhythmia and control group, determine the relative risk of predictive factors for the development of drug-induced arrhythmias, determine whether mutations and polymorphisms (defined as a variant gene frequency of &gt;1% in the population) of the cardiac ion channel genes are more common in cases of drug-induced ventricular arrhythmias than in control groups, and determine the relative risk for drug-induced arrhythmic events due to mutation carriage.</p> | 1 July 2008     |

| Investigators                                   | Study aim   | Completion date |
|---|---|-----------------|
| St George's Hospital Medical School, London, UK | <p>DARE - Drug Induced Arrhythmia Risk Evaluation</p> <p>This project's principle components &amp; aims are: (1) an epidemiological study to systematically document &amp; follow-up incident cases in England &amp; compare them to controls; (2) a genetic study to analyse blood samples from cases &amp; controls for mutations &amp; polymorphisms of the cardiac sodium &amp; potassium ion channel genes implicated in the <b>Long QT</b> &amp; Brugada syndromes.</p> | 30 June 2008    |
| St George's Hospital Medical School, London, UK | <p>DARE - Drug Induced Arrhythmia Risk Evaluation</p> <p>An epidemiological study to systematically document and follow-up incident cases in England and compare them to controls.</p> <p>A genetic study to analyse blood samples from cases and controls for mutations and polymorphisms of the cardiac sodium and potassium ion channel genes implicated in the <b>Long QT</b> and Brugada syndromes.</p>  | 31 July 2008    |

## Conclusions

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Congenital long QT syndrome (LQTS) is caused by mutations in a set of genes which code for protein subunits of cardiac ion channels. The main clinical feature of LQTS is the elongation of the QT interval on electrocardiograms (ECGs). The majority of LQTS patients are asymptomatic and are diagnosed either by family history or by virtue of having survived an episode of syncope or severe ventricular arrhythmia. Unfortunately, for many LQTS patients, the first presentation of symptoms is sudden cardiac death, usually occurring in healthy children or young adults in the absence of coronary artery disease.

It is estimated that LQTS affects approximately 1: 5,000 individuals in the United States and that it would be reasonable to expect that this prevalence would hold true in Australia and New Zealand. The estimated prevalence in children aged 4-15 years in England, Wales and Ireland is 1.6 – 6 per million. Research has suggested that a number of deaths in young people that have previously been ascribed to causes such as drowning, motor vehicle accidents and sudden infant death syndrome may be attributable to LQTS.

The current standard approach to diagnose LQTS was developed in 1993 by Schwartz et al. Clinical indicators; including past episodes of syncope with or without stress; congenital deafness, length of QTc in milliseconds obtained via ECG; torsade de pointes; T-wave alternans; notched T wave in 3 leads; low heart rate for age; and family history combine to give a patient an overall clinical score. Based on this score patients are rated on their probability of having LQTS.

As of May 2005, eight major genotypes, LQT1-8, have been identified. These genotypes are a result of a potential 471 different mutations and 124 polymorphisms. There are two forms of the disease; the most common form is autosomal-dominant known as Romano Ward syndrome, and the rare autosomal-recessive form called the Jervell, Lange-Nielsen (JLN) variant.

Mutational analysis of LQTS involves the use of polymerase chain reaction (PCR) with products then analysed using either single-strand conformational polymorphism (SSCP) or denaturing high performance liquid chromatography (DHPLC) and, if required, subsequent DNA sequencing. The advantages of PCR are that it has high sensitivity, high specificity and good reproducibility. Its limitations are: the potential for false-positive results from contaminating DNA and the potential for false-negatives due to the presence of PCR inhibitors, however, if PCR samples are run in triplicate or duplicate the number of false-positives may be reduced. DNA is usually obtained and isolated from a blood sample but also may be obtained via a mouth swab.

Molecular diagnosis is intended to identify asymptomatic family members of identified LQTS probands who have not been identified via clinical screening and may be at risk of sudden death. In addition, individuals deemed to have an intermediate or high risk of LQTS according to the diagnostic criteria of unexplained syncope, a prolonged QTc on an ECG or a family history of sudden cardiac death would be candidates for screening by mutational analysis. Approximately 30-35 per cent of questionable cases of LQTS will *not* be picked up by molecular diagnosis due to the large number of mutations

implicated in LQTS and the possibility of a large number of as yet unidentified mutations. The lack of identification of a genetic defect does not rule out the presence of LQTS. In addition to being a diagnostic tool, genetic testing has a prognostic value due to correlations between cardiac events and specific genotypes. A definitive genetic diagnosis may guide treatment management as genotype correlates with an individual's response to pharmacological therapy and other treatment options.

None of the papers included for in this assessment reported on safety outcomes associated with the mutational analysis of LQTS. Clinical assessment alone may not be sufficient to detect silent gene carriers who are asymptomatic for LQTS but are at risk of experiencing life-threatening cardiac events, especially if exposed to drugs capable of blocking potassium channels. Jongbloed (1999) reported a high number of asymptomatic carriers (21.4%) in family members of probands, underlining the importance of DNA analysis for individuals with an ambiguous clinical diagnosis. In addition, a negative mutational analysis does not rule out the possibility of LQTS as a diagnosis.

The high quality study by Priori et al (1999) directly compared the results of molecular analysis to the results of ECG (QTc) and the Schwartz Clinical Score in family members of probands. There was a low penetrance (25%) with 33 individuals genotype positive but the majority being clinically asymptomatic. When compared to mutational analysis, ECG and clinical score both had a low sensitivity (38%) but 100 per cent specificity. This study therefore demonstrates that molecular analysis is superior to clinical diagnostic techniques for the identification of "silent" LQTS carriers when a known mutation is screened for in family members.

Several studies described mutational analysis in *unrelated* patients. Tester et al (2005 & 2006b) reported on 541 consecutive unrelated patients who underwent comprehensive mutational analysis for LQT 1-3 and LQT 5-6. In contrast to the poor sensitivity reported by Priori et al (1999), it would appear that clinical diagnosis based on the Diagnostic Criteria points allocation system and a corrected QT interval >480 ms are an accurate basis for diagnosis for LQTS as both of these factors were statistically significant in the genotype positive group ( $p < 0.0001$ ). The association between syncope, previous cardiac arrest or family history tended towards, but did not reach significance in genotype positive patients ( $p = 0.067$ ). The follow-up study by Sherman et al (2005) reported on targeted mutational analysis for the rare LQT4 on patients previously found to be genotype negative for the more common mutations. This study demonstrated the value in a *fully* comprehensive initial mutational analysis, as a further nine patients (3.3%) previously thought to be LQTS negative were found to be positive for this rare mutation. Although Tester et al (2005 & 2006b) found a good correlation with clinical symptoms, Splawski et al (2000) contradicted this finding reporting that 32 per cent of the tested population were negative for a LQTS mutation despite the fact that 81 per cent of these individuals had clinical symptoms of LQTS and the mean QTc of this group was elevated at  $484 \pm 46$  ms.

Studies included in this assessment reported that mutational analysis appears to be superior in the diagnosis of LQTS carriers when compared to clinical assessment alone, and that once a proband has been identified, targeted

screening of family members for a specific mutation may be a cost-effective measure for extended family screening. However, the majority of these studies did not report on whether or not genetic screening resulted in a change in management for these individuals.

A cost-effectiveness analysis of genetic testing for familial LQTS in *symptomatic index cases* was conducted. The expected cost-effectiveness of genetic testing of *first-degree relatives* or more distant relatives was not included in this analysis. The three most common mutations were examined in the KCNQ1, KCNH2 and the SCN5A genes. Genetic testing of probands was found to be cost-effective compared to no genetic testing, at a cost per year life saved of US\$2,500, well below the standard threshold of US\$50,000 per life-year saved often used to define a cost-effective intervention. A further cost-effectiveness analysis is required to consider the benefits of genetic testing of family members of the proband.

In conclusion, clinical symptoms and an elongated QTc may not necessarily predict LQT and a negative mutation analysis may not rule out a diagnosis of LQTS. Mutational analysis of suspected LQTS patients appears to be effective in identifying individuals previously thought to be clinically asymptomatic, and this may have long term consequences for their future. As the first symptom of LQTS for many patients is a life-threatening cardiac event, early diagnosis is of utmost importance.

## Appendix A: Levels of Evidence

Designation of levels of evidence according to type of research question

| Level | Intervention <sup>§</sup>  | Diagnosis <sup>**</sup>  | Prognosis  | Aetiology <sup>†††</sup>                | Screening   |
|-------|--|--|--|---|---|
| I*    | A systematic review of level II studies  | A systematic review of level II studies  | A systematic review of level II studies  | A systematic review of level II studies | A systematic review of level II studies   |
| II    | A randomised controlled trial  | A study of test accuracy with: an independent, blinded comparison with a valid reference standard, <sup>§§</sup> among consecutive patients with a defined clinical presentation <sup>††</sup>     | A prospective cohort study <sup>***</sup>  | A prospective cohort study              | A randomised controlled trial   |
| III-1 | A pseudorandomised controlled trial (i.e. alternate allocation or some other method)   | A study of test accuracy with: an independent, blinded comparison with a valid reference standard, <sup>§§</sup> among non-consecutive patients with a defined clinical presentation <sup>††</sup> | All or none <sup>§§§</sup>   | All or none <sup>§§§</sup>              | A pseudorandomised controlled trial (i.e. alternate allocation or some other method)                                      |
| III-2 | A comparative study with concurrent controls:<br>Non-randomised, experimental trial <sup>†</sup><br>Cohort study<br>Case-control study<br>Interrupted time series with a control group | A comparison with reference standard that does not meet the criteria required for Level II and III-1 evidence  | Analysis of prognostic factors amongst untreated control patients in a randomised controlled trial | A retrospective cohort study            | A comparative study with concurrent controls:<br>Non-randomised, experimental trial<br>Cohort study<br>Case-control study |
| III-3 | A comparative study without concurrent controls:<br>Historical control study<br>Two or more single arm study <sup>‡</sup><br>Interrupted time series without a parallel control group  | Diagnostic case-control study <sup>††</sup>  | A retrospective cohort study   | A case-control study                    | A comparative study without concurrent controls:<br>Historical control study<br>Two or more single arm study              |
| IV    | Case series with either post-test or pre-test/post-test outcomes   | Study of diagnostic yield (no reference standard) <sup>‡‡</sup>  | Case series, or cohort study of patients at different stages of disease                            | A cross-sectional study                 | Case series   |

## **Tablenotes**

\* A systematic review will only be assigned a level of evidence as high as the studies it contains, excepting where those studies are of level II evidence.

§ Definitions of these study designs are provided on pages 7-8 *How to use the evidence: assessment and application of scientific evidence* (NHMRC 2000b).

† This also includes controlled before-and-after (pre-test/post-test) studies, as well as indirect comparisons (ie. utilise A vs B and B vs C, to determine A vs C).

‡ Comparing single arm studies ie. case series from two studies.

\*\* The dimensions of evidence apply only to studies of diagnostic accuracy. To assess the effectiveness of a diagnostic test there also needs to be a consideration of the impact of the test on patient management and health outcomes. See *MSAC (2004) Guidelines for the assessment of diagnostic technologies*. Available at: [www.msac.gov.au](http://www.msac.gov.au).

§§ The validity of the reference standard should be determined in the context of the disease under review. Criteria for determining the validity of the reference standard should be pre-specified. This can include the choice of the reference standard(s) and its timing in relation to the index test. The validity of the reference standard can be determined through quality appraisal of the study. See Whiting P, Rutjes AWS, Reitsma JB, Bossuyt PMM, Kleijnen J. The development of QADAS: a tool for the quality assessment of studies of diagnostic accuracy included in systematic reviews. *BMC Medical Research Methodology*, 2003, 3: 25.

†† Well-designed population based case-control studies (eg population based screening studies where test accuracy is assessed on all cases, with a random sample of controls) do capture a population with a representative spectrum of disease and thus fulfil the requirements for a valid assembly of patients. These types of studies should be considered as Level II evidence. However, in some cases the population assembled is not representative of the use of the test in practice. In diagnostic case-control studies a selected sample of patients already known to have the disease are compared with a separate group of normal/healthy people known to be free of the disease. In this situation patients with borderline or mild expressions of the disease, and conditions mimicking the disease are excluded, which can lead to exaggeration of both sensitivity and specificity. This is called spectrum bias because the spectrum of study participants will not be representative of patients seen in practice.

‡‡ Studies of diagnostic yield provide the yield of diseased patients, as determined by an index test, without confirmation of accuracy by a reference standard. These may be the only alternative when there is no reliable reference standard.

\*\*\* At study inception the cohort is either non-diseased or all at the same stage of the disease.

§§§ All or none of the people with the risk factor(s) experience the outcome. For example, no smallpox develops in the absence of the specific virus; and clear proof of the causal link has come from the disappearance of small pox after large-scale vaccination.

††† If it is possible and/or ethical to determine a causal relationship using experimental evidence, then the 'Intervention' hierarchy of evidence should be utilised. If it is only possible and/or ethical to determine a causal relationship using observational evidence (ie. cannot allocate groups to a potential harmful exposure, such as nuclear radiation), then the 'Aetiology' hierarchy of evidence should be utilised.

Note 1: Assessment of comparative harms/safety should occur according to the hierarchy presented for each of the research questions, with the proviso that this assessment occurs within the context of the topic being assessed. Some harms are rare and cannot feasibly be captured within randomised controlled trials; physical harms and psychological harms may need to be addressed by different study designs; harms from diagnostic testing include the likelihood of false positive and false negative results; harms from screening include the likelihood of false alarm and false reassurance results.

Note 2: When a level of evidence is attributed in the text of a document, it should also be framed according to its corresponding research question eg. level II intervention evidence; level IV diagnostic evidence; level III-2 prognostic evidence etc.

Hierarchies adapted and modified from: NHMRC 1999; (Lijmer et al 1999; Phillips et al 2001; Bancher editorial 1999)

## Appendix B: Diagnostic techniques

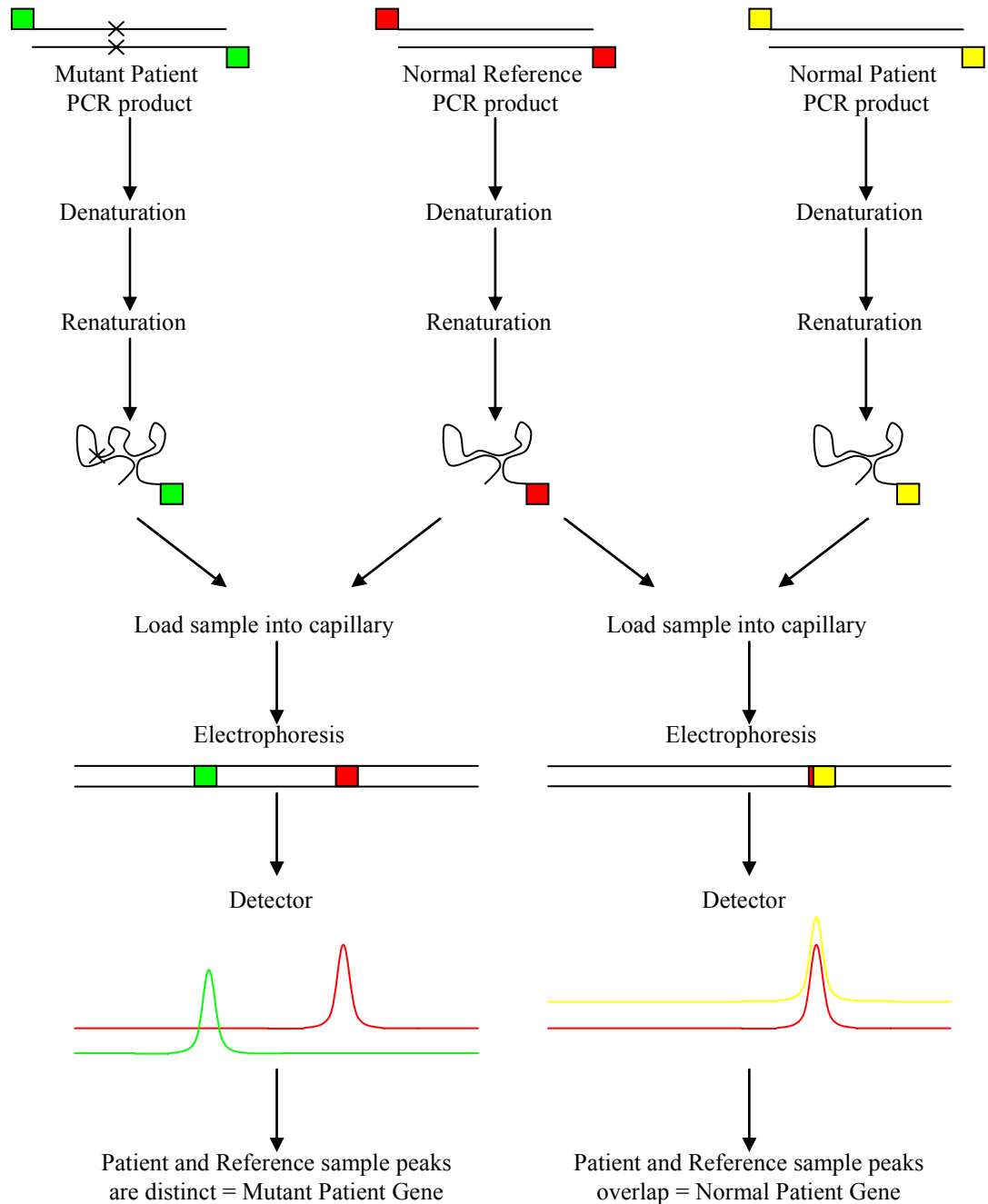


Figure 3 Single-strand conformational polymorphism (SSCP)

If patient gene of unknown status is mutant

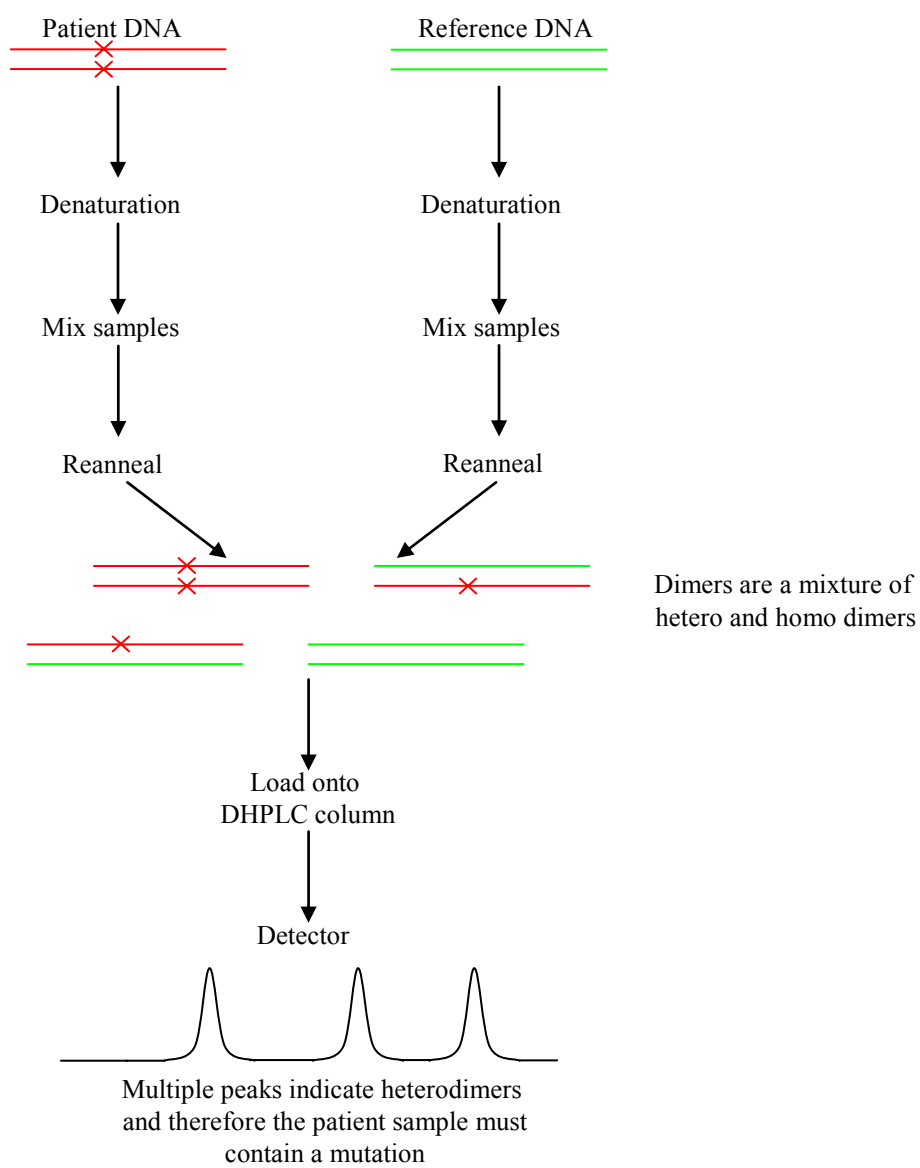


Figure 4 DHPLC for mutant DNA

If patient gene of unknown status is normal

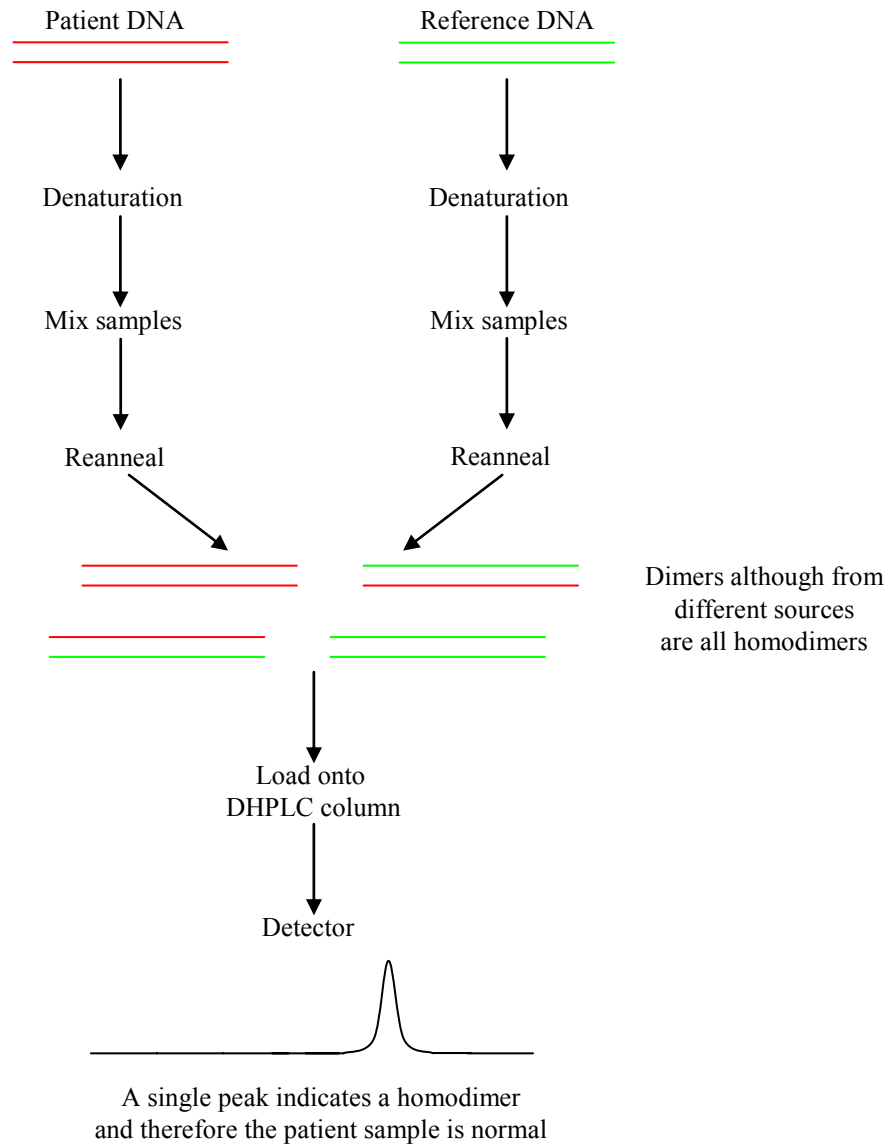


Figure 5 DHPLC for normal DNA

## Appendix C: Profiles of studies

| Study   | Location                    | Study design                    | Study population   | Study details  | Outcomes assessed |
|---|-----------------------------|---------------------------------|--|--|-------------------|
| Jongbloed, R.J.E.<br>Wilde, A.A.M.<br>Geelen, J.L.M.C.<br>Doevendans, P.<br>Schaap, C.<br>van Langen, I.<br>van Tintelen, J.P.<br>Cobben, J.M.<br>Beaufort-Krol, G.C.<br>Geraedts, J.P.M.<br>Smeets, H.J.M.<br>(1999) | Maastricht, The Netherlands | Diagnostic evidence level IV    | 24 probands with QTc >440ms, family history of sudden death or history of syncope, and family members (n= 134)                             | All probands screened for LQT 1-5. Majority positive for LQT1 and LQT2, therefore available family members were screened only for LQT 1 and LQT2. Only 14/24 families underwent molecular analysis as the remaining families were either too small, affected family members were deceased or clinical data was incomplete. | LQTS genotype     |
| Khositseth, A.<br>Tester, D.J.<br>Will, M.L.<br>Bell, C.M.<br>Ackerman, M.J.<br>(2004)  | Minnesota, United States    | Diagnostic evidence level IV    | 260 unrelated women suspected of LQTS. Mutational analysis on those women with a personal or family history of post-partum cardiac events. | All participants underwent molecular analysis of KCNQ1/KVLQT1 (LQT1), KCNH2/HERG (LQT2), SCN5A (LQT3), KCNE1/mink (LQT5) and KCNE2/MiRP1 (LQT6) genes  | LQTS genotype     |
| Napolitano, C.<br>Priori, S.G.<br>Schwartz, P.J.<br>Bloise, R.<br>Ronchetti, E.<br>Nastoli, J.<br>Botteli, G.<br>Cerrone, M.<br>Leonardi, S.<br>(2005)  | Pavia, Italy                | Diagnostic evidence level III-2 | 430 consecutive LQTS probands and 1,115 family members referred to clinic for genetic testing  | All participants underwent molecular analysis of KCNQ1, KCNH2, SCN5A, KCNE1 and KCNE2 genes  | LQTS genotype     |
| Piippo, K.<br>Swan, H.<br>Pasternack, M.<br>Chapman, H.<br>Paavonen, K.<br>Vittasalo, M.<br>Toivonen, L.<br>Kontula, K.<br>(2001)   | Helsinki, Finland           | Diagnostic evidence level IV    | 2 probands with JNLS and family members (35 & 28).<br><br>114 unrelated probands with RWS and 625 family members                           | Initial 2 probands entire coding region of minK and KCNQ1. No mutations in minK gene detected therefore all remaining  | KCNQ1 genotype    |

|  |                          |                                 |  |   |   |
|--|--------------------------|---------------------------------|--|---|---|
|  |                          |                                 |  | participants screened only for all exons in the KCNQ1 gene  |   |
| Priori, S.G.<br>Napolitano, C.<br>Schwartz, P.J.<br>(1998)   | Milan, Italy             | Diagnostic evidence level III-2 | 9 families with sporadic LQTS <sup>a</sup> .<br><br>9 probands with mutations in HERG gene (n=5) and KvLQT1 gene (n=4) (mean age 23 ± 17 years), mean QTc = 504 ± 48ms<br><br>46 clinically unaffected family members (mean age 43 ± 15 years) | All participants underwent ECG and clinical assessment followed by mutational analysis of HERG (LQT2) and KvLQT1 (LQT1) by single-strand conformational polymorphism and sequencing     | LQTS genotype                                 |
| Sherman, J.<br>Tester, D.J.<br>Ackerman, M.J.<br>(2005)<br><br>Same patient group as Tester et al 2005a and 2005b  | Minnesota, United States | Diagnostic evidence level II    | 541 consecutive unrelated patients suspected of LQTS   | All participants underwent ECG and clinical assessment blinded to patient genotype. Genomic DNA was analysed for mutations in ANK2 gene exons 36,37, 39-46.                             | LQTS genotype                                 |
| Splawski, I.<br>Shen, J.<br>Timothy, K.W.<br>Lehmann, M.H.<br>Priori, S.<br>Robinson, J.L.<br>Moss, A.J.<br>Schwartz, P.J.<br>Towbin, J.A.<br>Vincent, M.<br>Keating, M.T.<br>(2000) | United States            | Diagnostic evidence level III-2 | 262 unrelated patients with LQTS based on elongated QTc and clinical symptoms<br><br>Mean age 29 ± 19 years, mean QTc 492 ± 47 ms, 75% with clinical symptoms  | All participants underwent mutational analysis of KVLQT1, HERG, SCN5A, KCNE1 & KCNE2 genes.   | LQTS genotype                                 |
| Tester D.J.<br>Will, M.L.<br>Haglund, C.M.<br>Ackerman, M.J.<br>(2005)<br><br>Same patient group as Sherman et al 2005 and 2005  | Minnesota, United States | Diagnostic evidence level II    | 541 consecutive unrelated patients suspected of LQTS   | All participants underwent ECG and clinical assessment blinded to patient genotype. Genomic DNA was analysed for mutations in 60 protein-coding exons of the KCNQ1, KCNH2, SCN5A, KCNE1 | Correlation of clinical phenotype on genotype |

|   |                          |                              |  |  |               |
|---|--------------------------|------------------------------|--|--|---------------|
|   |                          |                              |  | & KCNE2 genes.   |               |
| Tester D.J.<br>Will, M.L.<br>Haglund, C.M.<br>Ackerman, M.J.<br>(2005)<br><br>Same patient group as Sherman et al 2005 and 2005 | Minnesota, United States | Diagnostic evidence level II | 541 consecutive unrelated patients suspected of LQTS                 | All participants underwent ECG and clinical assessment blinded to patient genotype. Genomic DNA was analysed for mutations in 60 protein-coding exons of the KCNQ1, KCNH2, SCN5A, KCNE1 & KCNE2 genes. | LQTS genotype |
| Tester, D.J.<br>McCormack, J.<br>Ackerman, M.J.<br>(2004)   | Minnesota, United States | Diagnostic evidence level IV | Pregnant woman with family history of LQTS undergoing amniocentesis. | Prenatal molecular analysis of amniotic fluid of an at risk foetus   | LQTS genotype |

LQTS = long QT syndrome, ECG = electrocardiogram, QTc = corrected QT interval, ms = milliseconds, JNLS = Jervell and Lange-Nielsen Syndrome, RWS = Romano-Ward syndrome

<sup>a</sup> Sporadic LQT where only one family member is clinically affected

## Appendix D: HTA Internet Sites

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### AUSTRALIA

- Centre for Clinical Effectiveness, Monash University  
<http://www.med.monash.edu.au/healthservices/cce/evidence/>
- Health Economics Unit, Monash University  
<http://chpe.buseco.monash.edu.au>

### AUSTRIA

- Institute of Technology Assessment / HTA unit  
<http://www.oecaw.ac.at/ita/welcome.htm>

### CANADA

- Agence d'Évaluation des Technologies et des Modes d'Intervention en Santé (AETMIS) <http://www.aetmis.gouv.qc.ca/site/index.php?home>
- Alberta Heritage Foundation for Medical Research (AHFMR)  
<http://www.ahfmr.ab.ca/publications.html>
- Canadian Coordinating Office for Health Technology Assessment (CCHOTA) <http://www.cadth.ca/index.php/en/>
- Canadian Health Economics Research Association (CHERA/ACRES) – Cabot database <http://www.mycabot.ca>
- Centre for Health Economics and Policy Analysis (CHEPA), McMaster University <http://www.chepa.org>
- Centre for Health Services and Policy Research (CHSPR), University of British Columbia <http://www.chspr.ubc.ca>
- Health Utilities Index (HUI)  
<http://www.fhs.mcmaster.ca/hug/index.htm>
- Institute for Clinical and Evaluative Studies (ICES)  
<http://www.ices.on.ca>

### DENMARK

- Danish Institute for Health Technology Assessment (DIHTA)  
[http://www.sst.dk/Planlaegning\\_og\\_behandling/Medicinsk\\_teknologi\\_vurdering.aspx?lang=en](http://www.sst.dk/Planlaegning_og_behandling/Medicinsk_teknologi_vurdering.aspx?lang=en)
- Danish Institute for Health Services Research (DSI)  
<http://www.dsi.dk/engelsk.html>

## **FINLAND**

- FINOHTA <http://www.stakes.fi/finohta/e/>

## **FRANCE**

- L'Agence Nationale d'Accréditation et d'Evaluation en Santé (ANAES)  
<http://www.anaes.fr/>

## **GERMANY**

- German Institute for Medical Documentation and Information (DIMDI)  
/ HTA <http://www.dimdi.de/dynamic/en/>

## **THE NETHERLANDS**

- Health Council of the Netherlands Gezondheidsraad  
<http://www.gr.nl/adviezen.php>

## **NEW ZEALAND**

- New Zealand Health Technology Assessment (NZHTA)  
<http://nzhta.chmeds.ac.nz/>

## **NORWAY**

- Norwegian Centre for Health Technology Assessment (SMM)  
<http://www.kunnskapssenteret.no/>

## **SPAIN**

- Agencia de Evaluación de Tecnologías Sanitarias, Instituto de Salud  
“Carlos III”/Health Technology Assessment Agency (AETS)  
<http://www.juntadeandalucia.es/salud/orgdep/aetsa/default.asp>
- Catalan Agency for Health Technology Assessment (CAHTA)  
<http://www.aatrm.net/html/en/dir394/index.html>

## **SWEDEN**

- Swedish Council on Technology Assessment in Health Care (SBU)  
<http://www.sbu.se/www/index.asp>
- Center for Medical Health Technology Assessment  
<http://www.cmt.liu.se/>

## **SWITZERLAND**

- Swiss Network on Health Technology Assessment (SNHTA)  
<http://www.snhta.ch/>

## UNITED KINGDOM

- NHS Quality Improvement Scotland  
[http://www.nhshealthquality.org/nhsqis/qis\\_display\\_home.jsp?pContentID=43&p\\_applic=CCC&pElementID=140&pMenuID=140&p\\_service=Content.show&](http://www.nhshealthquality.org/nhsqis/qis_display_home.jsp?pContentID=43&p_applic=CCC&pElementID=140&pMenuID=140&p_service=Content.show&)
- National Health Service Health Technology Assessment (UK) / National Coordinating Centre for Health Technology Assessment (NCCHTA)  
<http://www.hta.nhsweb.nhs.uk/>
- University of York NHS Centre for Reviews and Dissemination (NHS CRD) <http://www.york.ac.uk/inst/crd/>
- National Institute for Clinical Excellence (NICE)  
<http://www.nice.org.uk/>

## UNITED STATES

- Agency for Healthcare Research and Quality (AHRQ)  
<http://www.ahrq.gov/clinic/techix.htm>
- Harvard School of Public Health – Cost-Utility Analysis Registry  
<http://www.tufts-nemc.org/cearegistry/index.html>
- U.S. Blue Cross/ Blue Shield Association Technology Evaluation Center (TEC) <http://www.bcbs.com/tec/index.html>

## Appendix E: Glossary of terms

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### Long QT Terms:

LQTS: long QT syndrome

Syncope: A temporary suspension of consciousness due to generalised cerebral ischaemia, a faint or swoon.

*Torsade de pointes*: rapid ventricular tachycardia

Tachycardia: The excessive rapidity in the action of the heart, the term is usually applied to a heart rate above 100 beats per minute and may be qualified as atrial, junctional (nodal) or ventricular and as paroxysmal.

ECG: electrocardiogram: A recording of the electrical activity of the heart which detects and records the electrical potential of the heart during contraction.

QT interval: measured on an ECG from the beginning of the QRS complex to the end of the T wave, representing the duration of activation and recovery of the ventricular myocardium.

QTc: QT interval corrected for heart rate. QTc longer than 0.44 seconds is considered abnormal, although a normal QTc can be slightly prolonged in female individuals (up to 0.46 sec).

### Genetic Terms:

Allele: Alternative form of a gene. One of the different forms of a gene that can exist at a single locus.

Exon: The region of a gene that contains the code for producing protein. Each exon codes for a specific portion of the complete protein. Exons are separated by introns, long regions of DNA that have no apparent function.

Homozygote: Possessing two identical forms of a particular gene, one inherited from each parent.

Heterozygote: Possessing two different forms of a particular gene, one inherited from each parent.

Compound heterozygote: The presence of *two different* mutant alleles at a particular gene locus, one on each chromosome of a pair.

Locus: The site in a linkage map or on a chromosome where the gene for a particular trait is located. Any one of the alleles of a gene may be present at this site.

Genetic polymorphism: The occurrence together in the same population of more than one allele or genetic marker at the same locus with the least frequent allele or marker occurring more frequently than can be accounted for by mutation alone.

Genotype: The specific allelic composition of a cell, either of the entire cell or more commonly for a certain gene or a set of genes. The genes that an organism possesses.

Missense mutation: A genetic change involving the substitution of one base in the DNA for another which results in the substitution of one amino acid in a polypeptide for another. A missense mutation is a "readable" genetic message although its "sense" (its meaning) is changed.

Open reading frame: A long sequence of DNA that has no stop codon (no signal to stop reading) and therefore may encode part or all of a protein.

PCR: Polymerase chain reaction (PCR): A method for amplifying a DNA base sequence using a heat- stable polymerase and two 20- base primers, one complementary to the (+)- strand at one end of the sequence to be amplified and the other complementary to the (-) - strand at the other end. Because the newly synthesized DNA strands can subsequently serve as additional templates for the same primer sequences, successive rounds of primer annealing, strand elongation, and dissociation produce rapid and highly specific amplification of the desired sequence. PCR also can be used to detect the existence of the defined sequence in a DNA sample.

Penetrance: the proportion of individuals with a specific genotype who manifest that genotype at the phenotype level.

Phenotype:

- (1) The form taken by some character (or group of characters) in a specific individual.
- (2) The detectable outward manifestations of a specific genotype.
- (3) The observable attributes of an organism.

Polymorphism: The occurrence in a population (or among populations) of several phenotypic forms associated with alleles of one gene or homologs of one chromosome. See genetic polymorphism.

Proband: The family member through whom a family's medical history comes to light. The proband may also be called the index case, propositus (if male), or proposita (if female).

Single nucleotide polymorphisms (SNPs): is a DNA sequence variation occurring when a single nucleotide - A, T, C, or G - in the genome differs between members of a species (or between paired chromosomes in an individual). For example, two sequenced DNA fragments from different individuals, AAGCCTA to AAGCTTA, contain a difference in a single nucleotide. In this case we say that there are two alleles: C and T.

<http://helios.bto.ed.ac.uk/bto/glossary/>

<http://linkage.rockefeller.edu/wli/glossary/genetics.html>

<http://www.medterms.com>

## References

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- Ackerman, M. J. (2005). 'Genetic testing for risk stratification in hypertrophic cardiomyopathy and long QT syndrome: fact or fiction?' *Curr Opin Cardiol*, 20 (3), 175-181.
- Ackerman, M. J., Siu, B. L. et al (2001). 'Postmortem molecular analysis of SCN5A defects in sudden infant death syndrome', *Jama*, 286 (18), 2264-2269.
- Arnestad, M., Crotti, L. et al (2007). 'Prevalence of long-QT syndrome gene variants in sudden infant death syndrome', *Circulation*, 115 (3), 361-367.
- Bandolier editorial (1999). *Diagnostic testing emerging from the gloom?* [Internet]. Bandolier. Available from: <http://www.jr2.ox.ac.uk/bandolier/band70/b70-5.html> [Accessed 2004].
- Berul, C. I. & Perry, J. C. (2007). 'Contribution of long-QT syndrome genes to sudden infant death syndrome: is it time to consider newborn electrocardiographic screening?' *Circulation*, 115 (3), 294-296.
- Beauchamp, T. L. & Childress, J. F. (2001). *Principles of Biomedical Ethics*. 5th edition Oxford University Press, New York.
- Chadwick, R. (1999). *The Ethics of Genetic Screening*. Kluwer Academic Publishers, Boston.
- Ching, C. K. & Tan, E. C. (2006). 'Congenital long QT syndromes: clinical features, molecular genetics and genetic testing', *Expert Rev Mol Diagn*, 6 (3), 365-374.
- CIDG (2006). *Cardiac Inherited Diseases Group* [Internet]. Available from: <http://www.cidg.org/webcontent/Default.aspx?tabid=75> [Accessed 24th April 2006].
- Corrado, D., Basso, C. et al (2006). 'Trends in sudden cardiovascular death in young competitive athletes after implementation of a preparticipation screening program', *JAMA*, 296 (13), 1593-1601.
- DoH (2007). *The National Research Register* [Internet]. United Kingdom Department of Health. Available from: <http://www.nrr.nhs.uk/> [Accessed 16th April 2007].
- Doolan, A., Langlois, N. & Semsarian, C. (2004). 'Causes of sudden cardiac death in young Australians', *Med J Aust*, 180 (3), 110-112.
- Gold, M., Siegal, J. et al (1996). *Cost-effectiveness in health and medicine*. Oxford University Press, New York.
- Goldberger, A. L. (2001). *Electrocardiography*, Harrison's principles of internal medicine Vol. Volume 1. 15th edition McGraw-Hill, New York.
- Goldenberg, I., Moss, A. J. et al (2005). 'Cost-effectiveness of implanted defibrillators in young people with inherited cardiac arrhythmias', *Ann Noninvasive Electrocardiol*, 10 (4 Suppl), 67-83.

- Goldenberg, I., Moss, A. J. et al (2006). 'Clinical course and risk stratification of patients affected with the Jervell and Lange-Nielsen syndrome', *Journal of Cardiovascular Electrophysiology*, 17 (11), 1161.
- Hendriks, K. S., Grosfeld, F. J. et al (2005a). 'High distress in parents whose children undergo predictive testing for long QT syndrome', *Community Genet*, 8 (2), 103-113.
- Hendriks, K. S. W. H., Grosfeld, F. J. M. et al (2005b). 'Can parents adjust to the idea that their child is at risk for a sudden death? Psychological impact of risk for Long QT Syndrome', *American Journal of Medical Genetics*, 138 A (2), 107-112.
- Hobbs, J. B., Peterson, D. R. et al (2006). 'Risk of aborted cardiac arrest or sudden cardiac death during adolescence in the long-QT syndrome', *JAMA: Journal of the American Medical Association*, 296 (10), 1249.
- Hofman-Bang, J., Behr, E. R. et al (2006). 'High-efficiency multiplex capillary electrophoresis single strand conformation polymorphism (multi-CE-SSCP) mutation screening of SCN5A: A rapid genetic approach to cardiac arrhythmia', *Clinical Genetics*, 69 (6), 504-511.
- Jongbloed, R. J., Wilde, A. A. et al (1999). 'Novel KCNQ1 and HERG missense mutations in Dutch long-QT families', *Hum Mutat*, 13 (4), 301-310.
- Kanters, J. K., Graff, C. et al (2006). 'Long QT syndrome genotyping by electrocardiography: fact, fiction, or something in between?' *J Electrocardiol*, 39 (4 Suppl), S119-122.
- Khan, I. A. (2002). 'Long QT syndrome: diagnosis and management', *Am Heart J*, 143 (1), 7-14.
- Khositseth, A., Tester, D. J. et al (2004). 'Identification of a common genetic substrate underlying postpartum cardiac events in congenital long QT syndrome', *Heart Rhythm*, 1 (1), 60-64.
- Klabunde, R. E. (2005). *Cardiovascular Physiology Concepts* [Internet]. Available from: <http://www.cvphysiology.com/Arrhythmias/A009.htm> [Accessed 24th April 2006].
- Liebman, J. (2001). 'Some legal, social, and ethical issues related to the genetic testing revolution, as exemplified in the long QT syndrome', *J Electrocardiol*, 34 Suppl, 183-188.
- Lijmer, J. G., Mol, B. W. et al (1999). 'Empirical evidence of design-related bias in studies of diagnostic tests.' *Journal of the American Medical Association*, 282 (11), 1061-1066.
- Louie, M., Louie, L. & Simor, A. E. (2000). 'The role of DNA amplification technology in the diagnosis of infectious diseases', *Cmaj*, 163 (3), 301-309.
- Lunetta, P., Levo, A. et al (2003). 'Molecular screening of selected long QT syndrome (LQTS) mutations in 165 consecutive bodies found in water', *International Journal of Legal Medicine*, 117 (2), 115-117.
- Modell, S. M. & Lehmann, M. H. (2006). 'The long QT syndrome family of cardiac ion channelopathies: a HuGE review', *Genet Med*, 8 (3), 143-155.

- Moss, A. J. & Robinson, J. L. (1992). 'Clinical aspects of the idiopathic long QT syndrome', *Ann N Y Acad Sci*, 644, 103-111.
- Moss, A. J. & Schwartz, P. J. (2005). '25th anniversary of the International Long-QT Syndrome Registry: an ongoing quest to uncover the secrets of long-QT syndrome', *Circulation*, 111 (9), 1199-1201.
- MSAC (2006). *Implantable cardioverter defibrillators for prevention of sudden cardiac death*, The Medical Services Advisory Committee, Department of Health and Ageing, Canberra,  
[http://www.msac.gov.au/internet/msac/publishing.nsf/Content/AD35ED216E990FC7CA2571420004A192/\\$File/MSAC%20Ref%2032%20-%20ICDs.pdf](http://www.msac.gov.au/internet/msac/publishing.nsf/Content/AD35ED216E990FC7CA2571420004A192/$File/MSAC%20Ref%2032%20-%20ICDs.pdf).
- Napolitano, C., Priori, S. G. et al (2005). 'Genetic testing in the long QT syndrome: development and validation of an efficient approach to genotyping in clinical practice', *JAMA*, 294 (23), 2975-2980.
- PGxHealth (2006). *Familion: A genetic test for cardiac ion channel mutations* [Internet]. PGxHealth. Available from:  
<http://www.pgxhealth.com/genetictests/familion/> [Accessed 6th March 2007].
- Phillips, B., Ball, C. et al (2001). *Levels of Evidence and Grades of Recommendations* [Internet]. Centre for Evidence-Based Medicine, Oxford, UK. Available from:  
[http://www.cebm.net/levels\\_of\\_evidence.asp](http://www.cebm.net/levels_of_evidence.asp) [Accessed 28th January 2004].
- Phillips, K. A., Ackerman, M. J. et al (2005). 'Cost-effectiveness analysis of genetic testing for familial long QT syndrome in symptomatic index cases', *Heart Rhythm*, 2 (12), 1294-1300.
- Piippo, K., Swan, H. et al (2001). 'A founder mutation of the potassium channel KCNQ1 in long QT syndrome: implications for estimation of disease prevalence and molecular diagnostics', *J Am Coll Cardiol*, 37 (2), 562-568.
- Priori, S. G. & Napolitano, C. (2006). 'Role of genetic analyses in cardiology - Part I: Mendelian diseases: Cardiac channelopathies', *Circulation*, 113 (8), 1130-1135.
- Priori, S. G., Napolitano, C. & Schwartz, P. J. (1999). 'Low penetrance in the long-QT syndrome: clinical impact', *Circulation*, 99 (4), 529-533.
- Quaglioni, S., Rognoni, C. et al (2006). 'Cost-effectiveness of neonatal ECG screening for the long QT syndrome', *Eur Heart J*, 27 (15), 1824-1832.
- Roberts, R. (2006). 'Genomics and cardiac arrhythmias', *J Am Coll Cardiol*, 47 (1), 9-21.
- Roden, D. M. (2004). 'Drug-induced prolongation of the QT interval', *N Engl J Med*, 350 (10), 1013-1022.
- Rogers, W. & Braunack-Mayer, A. (2004). *Practical ethics for general practice*. Oxford University Press, Oxford.
- SADS (2006). *Australian Sudden Arrhythmia Death Syndromes (SADS) Foundation* [Internet]. Available from: <http://www.sads.org.au/> [Accessed 24th April 2006].
- Schwartz, P. J. (2005). 'Management of long QT syndrome', *Nat Clin Pract Cardiovasc Med*, 2 (7), 346-351

- Schwartz, P. J. (2006a). 'The congenital long QT syndromes from genotype to phenotype: clinical implications', *J Intern Med*, 259 (1), 39-47.
- Schwartz, P. J. (2006b). 'Pro: Newborn ECG screening to prevent sudden cardiac death', *Heart Rhythm*, 3 (11), 1353-1355.
- Schwartz, P. J. & Locati, E. (1985). 'The idiopathic long QT syndrome: pathogenetic mechanisms and therapy', *Eur Heart J*, 6 Suppl D, 103-114.
- Schwartz, P. J., Moss, A. J. et al (1993). 'Diagnostic criteria for the long QT syndrome. An update', *Circulation*, 88 (2), 782-784.
- Schwartz, P. J., Priori, S. G. & Napolitano, C. (2003). 'How really rare are rare diseases? the intriguing case of independent compound mutations in the long QT syndrome', *J Cardiovasc Electrophysiol*, 14 (10), 1120-1121.
- Schwartz, P. J., Spazzolini, C. et al (2006). 'The Jervell and Lange-Nielsen syndrome: natural history, molecular basis, and clinical outcome', *Circulation*, 113 (6), 783.
- Sherman, J., Tester, D. J. & Ackerman, M. J. (2005). 'Targeted mutational analysis of ankyrin-B in 541 consecutive, unrelated patients referred for long QT syndrome genetic testing and 200 healthy subjects', *Heart Rhythm*, 2 (11), 1218-1223.
- Skinner, J. R. (2005). 'Is there a relation between SIDS and long QT syndrome?' *Arch Dis Child*, 90 (5), 445-449.
- Skinner, J. R. (2007). 'Guidelines for the diagnosis and management of familial long QT syndrome', *Heart Lung Circ*, 16 (1), 22-24.
- Splawski, I., Shen, J. et al (2000). 'Spectrum of mutations in long-QT syndrome genes. KVLQT1, HERG, SCN5A, KCNE1, and KCNE2', *Circulation*, 102 (10), 1178-1185.
- Sukhija, R., Mehta, V. et al (2007). 'Implantable cardioverter defibrillators for prevention of sudden cardiac death', *Clin Cardiol*, 30 (1), 3-8.
- Tester, D. J. & Ackerman, M. J. (2007). 'Postmortem long QT syndrome genetic testing for sudden unexplained death in the young', *J Am Coll Cardiol*, 49 (2), 240-246.
- Tester, D. J., McCormack, J. & Ackerman, M. J. (2004). 'Prenatal molecular genetic diagnosis of congenital long QT syndrome by strategic genotyping', *Am J Cardiol*, 93 (6), 788-791.
- Tester, D. J., Will, M. L. & Ackerman, M. J. (2006a). 'Mutation detection in congenital long QT syndrome: cardiac channel gene screen using PCR, dHPLC, and direct DNA sequencing', *Methods Mol Med*, 128, 181-207.
- Tester, D. J., Will, M. L. et al (2005). 'Compendium of cardiac channel mutations in 541 consecutive unrelated patients referred for long QT syndrome genetic testing', *Heart Rhythm*, 2 (5), 507-517.
- Tester, D. J., Will, M. L. et al (2006b). 'Effect of clinical phenotype on yield of long QT syndrome genetic testing', *Journal of the American College of Cardiology*, 47 (4), 764.

van Langen, I. M. & Wilde, A. A. (2006). 'Con: Newborn screening to prevent sudden cardiac death?' *Heart Rhythm*, 3 (11), 1356-1359.

Vincent, G. M. (2005). 'The long QT and Brugada syndromes: causes of unexpected syncope and sudden cardiac death in children and young adults', *Seminars in Pediatric Neurology*, 12 (1), 15.

Viskin, S., Rosovski, U. et al (2005). 'Inaccurate electrocardiographic interpretation of long QT: The majority of physicians cannot recognize a long QT when they see one', *Heart Rhythm*, 2 (6), 569-574.

Wehrens, X. H. T., Vos, M. A. et al (2002). 'Novel insights in the congenital long QT syndrome', *Annals of Internal Medicine*, 137 (12), 981-992.

White, T. J., Madej, R. & Persing, D. H. (1992). 'The polymerase chain reaction: clinical applications', *Adv Clin Chem*, 29, 161-196.