

Malignant Hyperthermia

Authors: Prof Henry Rosenberg¹, Dr Mark Davis, Ms Danielle James, Dr Neil Pollock and Dr Kathryn Stowell

Creation date: November 2004

Scientific Editor: Prof Loïc Guillevin

¹Department of Medical Education and Clinical Research Saint Barnabas Medical Center Livingston, NJ 07039, USA

[Abstract](#)

[Keywords](#)

[Disease Name](#)

[Definition](#)

[Diagnostic Criteria](#)

[Differential Diagnosis](#)

[Etiology of MH](#)

[Clinical Description](#)

[Laboratory Diagnostic Methods](#)

[Epidemiology](#)

[Genetic Counselling](#)

[Treatment /Management of MH](#)

[Unresolved Issues](#)

[Resources](#)

[References](#)

Abstract

Malignant Hyperthermia is a pharmacogenetic disorder of skeletal muscle affecting humans, certain pig breeds, dogs and horses, and probably other animals. In humans the syndrome is inherited in an autosomal dominant pattern. The incidence of MH reactions ranges from one 10,000 to one 100,000 anesthetics. However the prevalence of the genetic abnormalities may be as great as one in 3,000 individuals.

The syndrome is precipitated in humans upon exposure to potent volatile anesthetic gases such as halothane, sevoflurane, desflurane and the depolarising muscle relaxant succinylcholine while in susceptible swine it may be precipitated by stress alone. The classic signs of MH include hyperthermia to marked degree, tachycardia, tachypnea, increase in carbon dioxide production, increased Oxygen consumption, acidosis, muscle rigidity, rhabdomyolysis, all related to a hypermetabolic response. The syndrome is likely to be fatal if untreated. Early recognition of the signs of MH, specifically elevation of end expired carbon dioxide provide the clinical diagnostic clues to the syndrome. Dantrolene sodium is a specific antagonist of the pathophysiologic changes of MH and should be available wherever general anesthesia is administered.

The pathophysiologic changes of MH are due to an uncontrolled rise of myoplasmic calcium, which activate biochemical processes related to muscle activation. Due to ATP depletion the muscle membrane integrity is compromised leading to hyperkalemia and rhabdomyolysis. The etiology of the syndrome in most cases is a defect in the ryanodine receptor, the calcium channel that controls, calcium release from the sarcoplasmic reticulum. Over forty mutations have been identified in the gene that elaborates the protein making up the channel. Other genes have also been implicated in some cases of MH.

Diagnostic testing relies on assessing the in vitro contracture response of biopsied muscle to halothane, caffeine, and other drugs. However, elucidation of the genetic changes has led to the introduction, on a limited basis so far of genetic testing for susceptibility to MH.

Thanks to the dramatic progress in understanding the clinical manifestation and pathophysiology of the syndrome the mortality from MH has dropped from over 80% thirty years ago to less than 5%.

Keywords

malignant hyperthermia, anesthetics, dantrolene, ryanodine receptor, molecular genetics, caffeine, halothane, central core disease, succinylcholine, muscular dystrophy, hypermetabolism.

Disease Name

Malignant Hyperthermia is also known as Malignant Hyperpyrexia

Definition

MH is a hypermetabolic response to potent inhalation agents, succinylcholine and rarely, in humans, to stresses such as vigorous exercise and heat. A majority of patients with [Central Core Disease](#), an inherited myopathy characterized by muscle weakness, are susceptible to MH.

Because almost all patients who are MH susceptible have no phenotypic changes without anesthesia, it is impossible to diagnostic susceptibility without either the exposure to the "trigger" anesthetics or by specific diagnostic testing.

The key diagnostic features include an unexplained elevation of expired carbon dioxide, muscle rigidity and rhabdomyolysis, hyperthermia, acidosis and hyperkalemia

Diagnostic Criteria

The diagnosis of Malignant Hyperthermia is based on clinical presentation or laboratory testing (see section on diagnostic methods).

The principal diagnostic features of MH are unexplained elevation of end tidal carbon dioxide (ETCO₂) concentration, muscle rigidity, tachycardia, acidosis, hyperthermia, hyperkalemia.

The variability of order and time of onset of signs often make the clinical diagnosis rather difficult.

A clinical grading scale was developed by Larach and colleagues (Larach, et al 1994) in order to assist in clinical diagnosis. The elements of the scale are in **the table 1**. Differential weighting is given to each of the manifestations of the syndrome. However, the scale lacks sensitivity since not all tests may be performed in an individual episode.

The value of the grading scale is mainly in identifying those subjects with the most convincing episodes of MH for subsequent evaluation of the sensitivity and specificity of the diagnostic tests. The clinical grading scale is useful in evaluating clinical episodes in those cases in which the subject is rated a 6 (almost certainly MH), but lower scores should not be considered for actual diagnosis.

Table 1: Criteria used in the Clinical Grading Scale for Malignant Hyperthermia

Clinical Finding	Manifestation
Respiratory acidosis	End-tidal CO ₂ >55 mmHg; PaCO ₂ >60 mm Hg.
Cardiac involvement	Unexplained sinus tachycardia, ventricular tachycardia or ventricular fibrillation
Metabolic acidosis	Base deficit > 8 m/Eq pH < 7.25
Muscle rigidity	Generalized rigidity; severe masseter muscle rigidity.
Muscle breakdown	Serum creatine kinase concentration >20,000/L units; cola colored urine; excess myoglobin in urine or serum; plasma [K+] >6 mEq/L.
Temperature increase	Rapidly increasing temperature; T >38.8° C.
Other	Rapid reversal of MH signs with dantrolene. Elevated resting serum creatine kinase concentration.
Family history	Consistent with autosomal dominant inheritance.

Differential Diagnosis

A variety of unusual conditions may resemble MH during anesthesia. These include sepsis, thyroid storm, [pheochromocytoma](#), iatrogenic overheating. Hence a high index of suspicion for these disorders as well as the ability to measure end tidal carbon dioxide and obtain blood gas analysis is essential in order to differentiate MH from these disorders. Particularly problematic is unexplained hyperthermia following anesthesia. Since anesthetic gases generally inhibit the febrile response, the first sign of sepsis may be marked hyperthermia on emergence from anesthesia. Response to antipyretics as well the clinical setting often is helpful in differentiating this response from MH.

The differential diagnosis of unexplained increased ETCO₂ includes hyperthermia secondary to sepsis, or iatrogenic warming, machine valve malfunction, rebreathing, as well as faulty equipment.

Outside the operation room, MH like syndrome may occur following ionic contrast agents injected into the cerebrospinal fluid, cocaine overdose, and the neuroleptic malignant syndrome.

If a high ionic, water-soluble radiologic contrast agent is injected intrathecally, usually as a result of drug mix-up a characteristic progression of signs occurs. After the injection, the patient appears to recover normally, but within thirty minutes, involuntary jerking movements begin in the lower extremities and ascend to the upper body finally resulting in seizures and hyperthermia. This is the result of the contrast agent entering the cerebral ventricles. Symptomatic treatment is called for. (Ong et al, 1989)

The response of signs of hyperthermia, tachycardia and tachypnea to dantrolene in such syndromes is non-specific. In other words, the response to dantrolene does not per se prove MH susceptibility

In many countries a "hotline" has been established to provide emergency assistance in the management of MH.

Hyperkalemic Cardiac Arrest in patients with Muscular Dystrophy

A syndrome often confused with malignant hyperthermia, is sudden hyperkalemic cardiac arrest during or shortly after anesthesia in young males. Following sporadic reports of such arrests, Larach and colleagues identified that patients with an occult myopathy, especially a [dystrophinopathy](#) such as Duchenne's muscular dystrophy are at risk to dramatic life-threatening hyperkalemia upon administration of succinylcholine. (Larach et al, 1997) Administration of potent volatile agents to such patients may produce a similar syndrome.

Since the most common muscular dystrophy (Duchenne's) is found with a frequency of 1 in 3500 live male births, and the onset of symptoms of muscle weakness may be as late as 6-8 years of age, some apparently healthy children may really be at risk for succinylcholine induced [hyperkalemia](#). Hence when a young child or young adult experience sudden, apparently unexpected cardiac arrest, think of hyperkalemia, document and treat it in the standard fashion (calcium, bicarbonate, glucose and insulin and hyperventilation). Muscle tissue should be obtained and preserved for testing for a myopathy

In general, the patient with a [dystrophinopathy](#) that develops these anesthetic-related complications does not also exhibit classic signs and symptoms of MH, such as hyperthermia or marked muscle rigidity. Therefore, this reaction

is not malignant hyperthermia per se, since the dystrophinopathies are caused by mutations on the X chromosome. Nevertheless, many anesthesiologists will avoid anesthetic triggering agents in patients with a congenital myopathy.

In response to the presentation of over 30 such cases to the FDA in 1992, a warning was issued to avoid the use of the drug in children and young adolescents for elective cases. Succinylcholine should be reserved for those cases of full stomach and possibly airway related emergencies.

Rhabdomyolysis

Rhabdomyolysis refers to the breakdown of skeletal muscle, which is associated with excretion of myoglobin in the urine. Classically MH presents with hypercarbia, tachycardia, cardiac arrhythmias, pyrexia rigidity, metabolic acidosis and rhabdomyolysis as a late sign. Several reports of isolated rhabdomyolysis apparent immediately following anaesthesia or developing up to 24 hours post anaesthesia have been published (Fierocche et al 1998). Increased creatine kinase (CK) measurement and a positive IVCT (In vitro contracture test) have been obtained in these patients indicating MH susceptibility. However MH like muscle responses can represent false positive diagnoses and an underlying myopathic process may produce a positive IVCT (Lehmann-Horn et al 1990) so there must remain some doubt on the validity of this feature *i.e.* rhabdomyolysis as an expression of MH. Burns et al however state that MH should be considered in all patients presenting with rhabdomyolysis where the degree of muscle necrosis exceeds that expected for the severity of the accompanying disorder.

Etiology of MH

It has become clear that MH is a disorder of skeletal muscle related to uncontrolled release of calcium from the sarcoplasmic reticulum. Other organ systems are involved only secondarily.

The consequence of enhanced release of calcium is muscle contraction/contracture due to release of inhibition of actin-myosin interaction. ATP and creatine kinase levels decline due to activation of processes to re-sequester calcium leading to anaerobic and aerobic metabolism and acidosis.

Presumably, the declining levels of ATP lead to breakdown of membrane integrity release of intracellular enzymes such as creatine kinase, and intracellular constituents such as potassium and hydrogen ions.

Evidence of abnormal calcium control even without exposure to anaesthetic agents is

suggested by nuclear magnetic resonance studies in exercising human muscle *in vivo*. These studies demonstrate greater inorganic phosphate levels (due to ATP breakdown), at rest and with exercise, exercise induced acidosis and slower recovery of ATP levels in MH susceptible patients. Fortunately these changes do not lead to clinical signs of MH.

Further refinement of our understanding of the pathophysiology of MH has focused on the mechanisms responsible for calcium control in muscle. Most attention has been focused on the ryanodine receptor, a calcium channel that mediates excitation contraction coupling in skeletal muscle. Indeed molecular biologic studies have shown that as many as 50% of families susceptible to MH harbor one of about 30 mutations in the gene that elaborates this protein.(Brandt, et al 1999) However, at least 6 other genes have been implicated in MH including another protein that mediates excitation contraction, the dihydropyridine receptor,(Monnier et al., 1997) and the sodium channel as well (Vita et al,1995) as other genes whose function is not yet clarified in relation to the MH syndrome.

Although mutations in the ryanodine receptor may be an important factor in the pathophysiology of MH, it is important to remember that not families show linkage to the ryanodine receptor gene. The presence of a mutation does not explain the inter and intra-individual variability in the human MH syndrome in terms of clinical expression. In several families there is discordance between the MH-ryanodine genotype and the phenotype as determined by the halothane-caffeine contracture test.

A variety of poorly understood modulators may influence the clinical expression of MH. For example, skeletal muscle from MH susceptible patients exhibits elevated fatty acid production. These fatty acids have been demonstrated to increase the sensitivity of halothane induced calcium release *in vitro*.(Fletcher *et al*, 1991,1993) In addition, in MH muscle there is a shift of subtypes of sodium channels leading to longer membrane depolarization and an increased period of calcium release from the terminal cisternae.(Wieland *et al*, 1989). There also is evidence for linkage of MH to chromosome 17 at or near the locus encoding the sodium channel alpha subunit. Hence, these changes in sodium channel function, either through sodium channel mutations or through effects of fatty acids on sodium channel and other proteins, may be essential for the phenotypic expression of certain aspects of the MH syndrome such as muscle rigidity.

Clinical Description

MH may occur at any time during anesthesia and in the early postoperative period. The earliest signs are tachycardia, rise in end expired carbon dioxide concentration despite increased minute ventilation, accompanied by muscle rigidity, especially following succinylcholine. Body temperature elevation is a dramatic, but *often a late sign of MH*. Nevertheless, core temperature should be monitored in all patients undergoing general anesthesia for periods lasting more than 20 minutes, as temperature elevation may be an important confirmatory sign. Other signs include acidosis, tachypnea and hyperkalemia.

The progression of the syndrome may be rapid and dramatic, particularly if precipitated by succinylcholine or more slowly and not become manifest until after several hours of anesthesia.

ALL INHALATION ANESTHETICS EXCEPT NITROUS OXIDE AND SUCCINYLCOLINE ARE TRIGGERS FOR MH. No other anesthetic drugs appear to be triggers, including propofol and ketamine. Neither are catecholamines, non depolarizing muscle relaxants, catechol congeners, digitalis or similar agents MH triggers.(Hopkins et al, 2000)

Although end tidal CO₂ is a sensitive early sign of MH, in recent years, with a decline in the use of succinylcholine, rather than an abrupt rise in CO₂, a more gradual rise is often noted. Indeed by increasing minute ventilation it is possible to mask this rise.(Karan et al, 1994)

Hyperthermia when it occurs, is marked by increase in core temperature at a rate of 1-2° C every five minutes. Severe hyperthermia (core temperature greater than 44°C) may occur, and lead to a marked increase in oxygen consumption, carbon dioxide production, widespread vital organ dysfunction, and disseminated intravascular coagulation (DIC).

Uncontrolled hypermetabolism leads to cellular hypoxia that is manifested by a progressive and worsening metabolic acidosis. If untreated, continuing myocyte death and rhabdomyolysis result in life-threatening hyperkalemia; myoglobinuria may lead to acute renal failure. Additional life-threatening complications include DIC, congestive heart failure, bowel ischemia, and compartment syndrome of the limbs secondary to profound muscle swelling and renal failure from rhabdomyolysis.

Succinylcholine induced masseter muscle rigidity (MMR)

Succinylcholine induced masseter muscle rigidity occurs in 1 in 100 children induced with halothane and given succinylcholine.(Schwartz

et al, 1984) The incidence is probably the same following induction with sevoflurane but much less following induction of thiopental. The clinical incidence of MH as defined by arterial blood gas changes is about 15% after MMR. However, muscle biopsy reveals that 50% of patients experiencing MMR are MH susceptible. (O'Flynn, et al 1994)

Patients with generalized rigidity along with MMR are at much greater risk for MH. Kaplan has hypothesized that children with "jaws of steel" after succinylcholine as opposed to mild rigidity are at greater risk for MH. He has hypothesized that the children with the more dramatic masseter rigidity are more often referred for biopsy and hence the high incidence of positive biopsies.

Since MMR may presage MH, it is most advisable to discontinue the anesthetic after MMR. In an emergency the anesthetic may continue with "non-trigger" drugs. Following MMR patients should be admitted to an intensive care unit and monitored for signs of MH. Rhabdomyolysis occurs in virtually all patients experiencing MMR and the CK values should be checked regularly.

Central Core Disease and other myopathies

[Central Core Disease](#) (CCD) is a rare non progressive myopathy with autosomal dominant inheritance presenting in infancy and characterised by hypotonia and proximal muscle weakness. Histological examination of affected muscles shows a predominance of type I fibres containing clearly defined areas (cores) lacking oxidative enzyme activity (Quinlivan et al 2003). An important feature is its close association with MH susceptibility. CCD patients are often susceptible to MH by IVC testing but MH and CCD phenotypes do not always co-segregate within families. Patients with MH may present with cores despite being clinically asymptomatic. And some RYR1 mutations, specifically some of those in the 3'transmembrane domain of the gene, may be specific to CCD. Although RYR1 mutations are the most common identified cause of CCD it does show genetic heterogeneity, with several rare susceptibility loci known (the ACTA1 gene, in association with nemaline myopathy, and the MYH7 gene, in association with hypertrophic cardiomyopathy), with further loci yet to be identified (Shepherd et al 2004).

At least 44 mutations have been reported in the RYR1 gene in association with CCD (Shepherd et al 2004). In general terms single point RYR1 mutations can cause (a) CCD only (b) MH only (c) MH with variable CCD penetrance. In this latter case all individuals with the mutation should be MH susceptible while they may or may

not have CCD. If a mutation is specific to CCD is identified in a family, MH is not automatically excluded as a second mutation may be present and MH susceptibility needs to be assessed by IVCT (Robinson et al 2002). If the mutation has no functional studies performed it is of no use clinically. An MH negative parent eliminates susceptibility in the children.

Other myopathies that have been associated with MH susceptibility include some sodium channel forms of myotonia (myotonia fluctuans), multimimicore disease and Hypokalemic periodic paralysis. (Lambert et al, 1994)

King or King Denborough syndrome is a rare myopathy characterized by dysmorphic facies, ptosis, down-slanting palpebral fissures, hypertelorism, epicanthic folds, low-set ears, malar hypoplasia, micrognathia, high-arched palate, clinodactyly, palmar simian line, pectus excavatum, winging of the scapulae, lumbar lordosis and mild thoracic scoliosis who present congenital hypotonia, slightly delayed motor development, diffuse joint hyperextensibility and mild proximal weakness. Such patients are MH susceptible

Laboratory Diagnostic Methods

The "gold standard" for diagnosis of Malignant Hyperthermia (MH) is currently the *in vitro* contracture test (IVCT) which is based on contracture of muscle fibres in the presence of halothane or caffeine. Two widely used forms of this test have been developed; one by the European Malignant Hyperthermia group (EMHG) and the other by the North American Malignant Hyperthermia Group (NAMHG) (Larach 1989,1992). While similarities exist in performing and interpreting the results of these tests, there are significant differences. Using the EMHG protocol, an individual is considered susceptible to MH (MHS) when both caffeine and halothane test results are positive. A normal (MHN) diagnosis is obtained when both tests are negative. A third diagnosis, MH equivocal (MHE) is obtained when only one of the halothane or caffeine tests is positive. Using the NAMHG protocol, an individual is diagnosed as MHS when either of the halothane or caffeine tests is positive and MNH when both tests are negative. The EMHG protocol may reduce the possibility of false positive and negative results when compared to the NAMHG protocol but overall similar results are obtained (Ording and Bendixen 1992). Sensitivity of 99% and a specificity of 94% are obtained with the EMHG protocol (Ording et al. 1997); while figures of 97% sensitivity and 78 % specificity are reported for the NAMHG. The specificity of either protocol may be affected by neuromuscular disorders,

unrelated to MH, which have an associated increase in myoplasmic calcium concentration. A third variation of the IVCT, the caffeine skinned fibre test, does not appear to be used diagnostically outside of Japan, and has lower specificity and sensitivity than either the EMGH or NAMHG protocols.

IVCT is expensive, confined to specialized testing centres, requires a surgical procedure and can yield equivocal as well as false positive and negative results. Modifications of the EMHG protocol include the use of ryanodine (Bendahan et al. 2004), which binds selectively to the calcium release channel or 4-chloro-m-cresol (Rueffert et al. 2002), but to date these agents have not been included in the standard protocol. Other biochemical, haematological and physical tests have been used in the past but without exception, these lack significant sensitivity and specificity to be used diagnostically.

DNA analysis, however, offers an alternative to the IVCT, requiring only a blood specimen, which can be sent to an accredited diagnostic laboratory. DNA testing for MH was first suggested by in 1990, when a mutation within the ryanodine receptor gene (*RYR1*) encoding the skeletal muscle calcium release channel was identified (McCarthy et al. 1990). Since then about 50% of MH has been linked to *RYR1* with over 60 mutations, associated with MH, identified within this gene. MH is however a heterogeneous genetic disorder with at least five other susceptibility loci being identified. Amongst these loci, mutations with a clear association with MH have been identified in only one gene; *CACNA1S* encoding the alpha subunit of the dihydropyridine receptor (DHPR), the voltage sensor of the skeletal muscle calcium release channel (Monnier 1997; Vainzof et al. 2000; Melzer and Dietze 2001).

Mutations cluster within three regions of *RYR1* with the majority of mutations leading to a single amino acid change in the receptor (Wehner et al. 2004). In addition a deletion has been reported (Sambuughin et al. 2001). While the identification of causative mutations suggests the widespread use of DNA testing for MH, this is confounded by the metabolic complexity and genetic heterogeneity of the disorder. Moreover, discordance between MHS diagnosed by IVCT and the presence of causative mutations has been shown within individual families (Deufel et al. 1995; MacLennan 1995; Fagerlund et al. 1997; Fortunato et al. 1999; Pollock et al. 2002; Robinson et al. 2002; Robinson et al. 2003). At best DNA testing can be used only in selected, genetically characterised families and within the guidelines identified by the EMHG for DNA testing (Urwyler et al. 2001). Because of the

heterogeneity of the disorder, as well as discordance within families, a negative DNA result cannot be used to rule out susceptibility. In addition, only those mutations that have been biochemically characterised to affect sarcoplasmic reticulum calcium release can be used to test for MH susceptibility.

Approximately 22 mutations within *RYR1* have been shown to cause an alteration in calcium release from intracellular stores. A number of functional tests have been successfully used to assess the role of *RYR1* mutations in calcium release. These include the use of lymphoblastoid cell lines generated from MH susceptible individuals (Girard et al. 2001; Tilgen et al. 2001; Sei et al. 2002). COS-7 or HEK293 cells transfected with the cDNA for rabbit *RYR1* carrying point mutations introduced by site-directed mutagenesis (Treves et al. 1994; Querfurth et al. 1998; McCarthy et al. 2000), myotubes generated from muscle biopsy tissue (Wehner et al. 2002; Wehner et al. 2003; Wehner et al. 2003; Wehner et al. 2004) and 1B5 dyspedic myotubes transduced with wild type and mutated *RYR1* cDNA (Yang et al. 2003). Calcium release can be monitored and quantified using calcium-specific indicators like fluo-4 and fura-2 (Gryniewicz et al. 1985), [³H] ryanodine binding assays (Richter et al. 1997; Yang et al. 2003), indirectly by protein release (Klingler et al. 2002). Systems using 1B5 dyspedic myotubes are more physiological as they constitutively express all the components of the skeletal muscle with the exception of *RYR1* (Yang et al. 2003). They also contain larger and more efficiency filled calcium stores than do COS-7 or HEK 293 cells, thus providing a more sensitive measure for calcium release and reloading. To date all mutations functionally characterised have been shown to cause alterations in calcium flux through the ryanodine receptor calcium release channel. It has been argued that lymphoblastoid cells and myotubes derived from MHS patients can not be used to unambiguously demonstrate altered physiological function as the phenotype may be due not only to mutations in *RYR1* but also in other genes encoding protein components of the calcium release channel, sarcoplasmic membrane of calcium signalling. Nevertheless, these systems have proved useful in demonstrating abnormal calcium release associated with *RYR1* mutations and it could be equally argued that these systems are representative of the genetic background of individual patients and therefore provide valuable information *ex vivo*. As lymphoblastoid cell lines do not express the dihydropyridine receptor they could also effectively be used to

functionally test *RYR1* mutations as well as eliminate *RYR1* as a causative factor in MHS individuals who do not show linkage to *RYR1*.

A variety of minimally invasive diagnostic tests are in development at present.

One utilizes nuclear magnetic resonance spectroscopy to evaluate ATP depletion during graded exercise *in vivo*. MH patients have a greater breakdown of ATP and Creatine phosphate as well as an increase in acid content compared to normals. (Bendahan et al, 2004)

Insertion of a microdialysis catheter into muscle, injection of a small amount of caffeine will elicit an enhanced release of carbon dioxide from the muscle tissue, which can be measured by capnography. (Anetseder et al, 2002)

Epidemiology

Reactions develop more frequently in males than females (2:1) and all ethnic groups are affected in all parts of the world. The highest incidence is in young people with a mean age of all reactions 18.3 years. Strazis found that children under 15 years age comprised 52.1% of all reactions. Although described in the newborn the earliest reaction confirmed by testing is 6 months of age (Chamley et al 2000). The oldest 78 years.

Genetically MH exhibits an autosomal mode of inheritance with an estimated prevalence of 1:8,500; but on average, patients require 3 anaesthetics before triggering. Numerous factors could be involved – age, type of anaesthetic, environmental temperature, mitigating drugs administered simultaneously degree of stress (Ording et al 1985). Mauritz found an incidence of 1:37,500 in patients who had been diagnostically tested which is similar to the estimated incidence of Ellis and Robinson (1:30,000), although wide variability is reported. A recent report suggests that the MHS trait may be present in 1:2-3000 of the French population (Monnier et al 2002). Bachand and colleagues examined the incidence of MH, in Quebec, Canada, where many families had been biopsied. They traced the pedigrees of the patients to the original immigrants from France and found an incidence of MH susceptibility of 0.2% in this province. However, that represented only five extended families (Bachand et al, 1997).

MH crises develop not only in humans but in other species particularly pigs which have been a valuable source for research. Reactions have also been described in horses, dogs and other animals (Britt BA 1985).

Genetic Counselling

Genetic testing can be defined as an analysis or test that confirms the presence or the absence of

a genetic condition; this does not necessarily involve the analysis of DNA as there are still many clearly genetic conditions where the gene has not yet been identified. In the context of Malignant Hyperthermia the IVCT test could be considered to be just as much a genetic test as the analysis of the *RYR1* gene.

Genetic testing is different to the traditional medical test in that not only will the result have potential ramifications for the *current* health of that individual but it may also have ramifications for the *future* health of that individual and the future health of their immediate relatives. (Juengst 1999; Halsted 1996). Depending on the test being performed results may leave the individual disadvantaged in terms of their ability to access health insurance or life insurance, employment opportunities and in some cultures may even affect marital opportunities (Burke et al 2001; Fisher 1996).

For this reason it is recommended that each individual accessing any form of genetic testing, and indeed each individual undergoing IVCT or *RYR1* analysis should be fully informed of *all* the implications of each potential result and should be able to provide informed consent (Skene 2002; Grover 2003).

The process of imparting this information and discussing any questions the patient may have, is known as Genetic Counselling. This discussion, with a clinician or genetic counsellor should include the following points of information (Am. Soc. Hum.Gen. 1975):

- Potential implications the result may have on their ability to obtain health/life insurance. (Light 1992; Harper 1993)
- Potential psychological effects of the result. Some parents feel guilty that they may have passed MH sensitivity on to their children, others may feel anxious about the implications of a MH sensitivity (MHS) result and experience an increased fear of surgery, others may even feel guilty if they have not inherited MH when their brother or sister has. (Kessler et al 1984; Marteau et al 1998; Jacobs et al 1999)

Inheritance pattern of the disorder and what implications their test result may have for their children, and the extended family.

If there is also a family history of CCD, it is important that the potential diagnosis of this condition is not lost in the discussions regarding Malignant Hyperthermia. CCD is an extremely variable condition within families and while some individuals may only be very mildly affected other family members may have a more severe phenotype (Quinlivan et al 2003).

Interpreting Risk for Other Family Members.

Malignant Hyperthermia is an autosomal dominant condition. When initiating genetic analysis in a branch of a known family, it is important to test the individual at the highest risk first.

Parents of a proband: the large majority of times an affected proband will have inherited their MH sensitivity from one of their parents. Clarification of which parent may also be MHS is useful for identifying which side of the extended family (ie aunts & uncles) may be at risk.

Siblings of a proband: The risk to the siblings depends of the genetic status of the parents. If a parent is identified as MHS, then each of the proband's siblings have a 1 in 2 or 50% chance of also being MHS. (If both parents receive an MHN result on IVCT & RYR1 analysis – suggesting the mutation is *de novo* in the proband - then the proband's siblings are at no greater risk than the general population.)

Offspring of a proband: The risk for offspring of each individual with proven MHS also has a 50% chance of being MHS. The proband's grandchildren would be considered to be at 25% risk until their parent's genetic status is clarified.

Note: An individual who is MHN cannot pass MH sensitivity on to the next generation, however, if they have an affected parent their siblings may still be at risk.

Interpretation of Risk for Other Family Members in the Context of RYR1 Analysis.

As discussed in earlier sections the identification of a causative RYR1 mutation is sufficient to diagnose MH Sensitivity. However, due to current concerns regarding discordance between IVCT and mutation analysis in some families, current protocols state that a negative mutation result is not sufficient to identify a person as being MHN. In the event of a normal (negative) mutation result IVCT is still recommended to confirm MHN status, and the individual and his/her offspring are still considered to be potentially MHS unless IVCT proves otherwise (Robinson et al 2003).

However, it is important to remember that in the event of a normal (negative) mutation result – the offspring of the individual are no longer at risk of inheriting the characterised family mutation. As the tested individual does not carry the mutation he *cannot* pass it on to his offspring. *Therefore, if an individual is mutation negative but IVCT positive, the only useful test available to the offspring is IVCT.*

Final note on Autonomy in Clinical testing for Malignant Hyperthermia.

Some individuals may wish to delay IVCT or RYR1 analysis while they consider the information they have been given and/or make the necessary preparations. Others may decide that they do not want their risk clarified by clinical testing.

These decisions should be respected and these individuals considered being MHS until proven otherwise.

Care should then be taken when arranging testing for the offspring of these individuals as a positive result in the next generation will generate a result for the individual who did not want to know (the individual must have carried the gene mutation in order to pass it on).

Treatment /Management of MH

Acute MH crisis

The essential points in the treatment of acute MH crisis are the immediate discontinuation of trigger agents, hyperventilation, administration of dantrolene in doses of 2.5mg/kg repeated prn to limit MH, cooling by all routes available (especially nasogastric lavage), treating hyperkalemia in a standard fashion. Calcium blockers should not be used along with dantrolene since hyperkalemia may occur with such a drug combination. The steps in treatment of acute MH is as follows:

1. Stop potent inhalation agents, Succinylcholine
2. Increase minute ventilation to lower end tidal CO₂
3. Get help
4. Prepare and administer dantrolene-2.5mg/Kg initial dose
Titrate Dantrolene to tachycardia, hypercarbia
10 Mg/Kg suggested upper limit, but more may be given as needed
5. Begin cooling measures, If Hyperthermic, use iced solutions, *i.e.* Ice Packs to groin, axilla, and neck. Nasogastric lavage with iced solution. More aggressive measures as needed.
Stop Cooling Measures At 38.5C
6. Treat arrhythmias as needed. Do not use calcium channel blockers
7. Secure blood gases, electrolytes, CK, blood and urine for myoglobin,
Coagulation profile check values every 6-12 Hours
Treat hyperkalemia with hyperventilation, glucose and insulin as needed

Once crisis is under control, an MH hotline should be contacted for further guidance.

8. Continue Dantrolene at 1mg/Kg every 4-8 hours For 24-48 hours
 9. Insure urine output of 2ml/Kg/Hour with Mannitol, Furosemide, fluids as needed
 10. Evaluate need for invasive monitoring and continued mechanical ventilation
 11. Observe patient in Intensive Care Unit for at least 36 hours
 12. Refer patient and family to MH Testing Center for contracture or DNA testing
- Patients experiencing MH should receive dantrolene and be monitored closely for 48-72 hours, since even despite dantrolene treatment 25% of patients will experience a recrudescence of the syndrome. Tests for disseminated intravascular coagulation (DIC) should be included as well as observation of the urine for myoglobinuric renal failure. DIC is most frequent when body temperature exceeds about 41 degrees C.

Preventive Measures

Preventive measures include:

A thorough anesthetic history to determine the possibility of the patient or a family member having experienced an MH episode. When suspicion of MH exists, family members should not be given trigger anesthetic agents, *i.e.* potent volatile anesthetic agents such as halothane, sevoflurane, desflurane, enflurane, isoflurane and succinylcholine and testing is recommended. Patients with any form of myotonia should not receive succinylcholine.

Patients with hypokalemic periodic paralysis, central core disease, Duchenne or Becker muscular dystrophy, paramyotonia, or myotonia fluctuans should not receive trigger agents.

All patients receiving more than a brief general anesthetic should have their core temperature monitored.

Young patients (below age 12 approximately) should not receive succinylcholine for elective procedures, in order to avoid the possibility of hyperkalemic response in a patient with undiagnosed muscular dystrophy.

Patients who are MH susceptible should be cautioned regarding the remote, but conceivable possibility of heat stroke in environments in which exposure to high heat and humidity are possible.

Management of the MH susceptible for anesthesia.

Patients who are known to be MH susceptible may be anesthetized with regional anesthesia or local anesthesia without problems. If general

anesthesia or sedation is required, the potent volatile agents and succinylcholine should be avoided.

The anesthesia machine should be prepared by flowing 100% Oxygen through the machine at 10L/min for at least 20 minutes. The ventilator should also be included in purging the machine by cycling the ventilator at the time of the Oxygen flow. Vaporizers should be disabled, drained or removed if possible.

All intravenous agents and non-depolarizing relaxants are safe to use.

Once the patient has undergone such an anesthetic without incident he /she may be treated similar to any other patients.

It is no longer felt to be necessary to monitor such patients in the post-anesthesia care unit for four hours routinely.

Pretreatment with dantrolene is also not necessary.

Unresolved Issues

Discordance

Given the confidence provided by functional analysis of *RYR1* mutations, the problem of discordance between *RYR1* mutations and MHS and MH equivocal (MHE) diagnosis still remains the largest problem associated with genetic diagnosis of susceptibility to MH. The MHE diagnosis is the most problematic and exhibits a much higher level of discordance than does MHS. Correlation between *RYR1* mutations and IVCT is greater for the caffeine than the halothane response (Manning et al. 1998) suggesting that the MHE(c) has greater diagnostic potential. The NAMHG protocol does not allow the MHE diagnosis; the potential for discordance between IVCT phenotype and, *RYR1* genotype and, discordance is therefore much greater. In a large UK study investigating the relationship between *RYR1* genotype and IVCT phenotype, discordance was identified in seven families (nine individuals), with five false-positives and four false-negatives (Robinson et al. 2002). Mutation negative/MHS individuals have also been observed (Robinson et al. 2002; Robinson et al. 2003). Clear evidence of the involvement of genes, as well as *RYR1*, has been shown in a New Zealand Maori pedigree where, MHS correlates with a *T4826I* mutation (Brown et al. 2000); but three branches of the family possess unrelated chromosome 19 haplotypes, without the *T4826I* mutation in unambiguous MHS individuals spanning three or four generations. While some discordance may be explained by the existence of other as yet unidentified mutations, false positive IVCT tests (Serfas et al. 1996), and weak contracture

mutations (Robinson et al. 2002) have also been implicated. Clearly, while genetic diagnosis can be used selectively, a greater knowledge of the molecular mechanisms resulting in susceptibility to MH is required before the IVCT can be dispensed with.

Awake MH:

In 1966 the Porcine Stress Syndrome was identified as an “awake” malignant hyperthermia episode. Stresses such as fighting, cause a rapid death in these animals. In 1974 Wingard described an MH susceptible family with exercise and emotional induced pyrexia, in sudden deaths unrelated to surgery. He considered that MH was part of a human stress syndrome. Subsequently, a number of reports of MH reactions in patients given trigger-free anaesthetics appeared. None of these reactions were totally convincing.

However, Gronert *et al* and Denborough both reported patients with “awake” MH episodes, the latter a patient with exercise-induced heat stroke, who responded to Dantrolene. Perhaps the most convincing, though unfortunate, episode of exercise induced MH was reported by Tobin et al. a fatal episode in a 13-year-old boy (Tobin et al, 2001) Brown reported a possible viral trigger.

Further physiological evidence of stress-related MH has been demonstrated by pH changes in MHS muscle recovering from violent exercise (Ellis et al, 1991). The sympathetic nervous system appears to be only secondarily involved (Haggendal et al, 1990), but 5HT agonists may cause an MH-like syndrome in susceptible pigs (Loscher et al, 1990). These agents can also cause MHS contractures in susceptible muscle Wappler et al 1996). Does Serotonin have a role in the stress-induced episodes?

Wappler described a 34-year-old male with recurrent fever, fatigue, muscle cramping, and aching with mild exercise and emotional stress (Wappler et al 2000). IVCT demonstrated an MHS response and a “causative” mutation. Others have reported similar findings (Wappler et al 2001; Davis et al 2002). A possible conclusion that a small subset of MH patients may display muscle damage and, perhaps more ominous signs with the stress of exercise, and may be other stresses. It is recommended that MH is excluded in patients who have had episodes of exertional heat stroke (Grogan et al, 2002). However despite possible links between exertional heat stroke and MH, treatment with dantrolene has had limited results and, this drug should not be used routinely in the management of heat stroke.

Resources

Many anesthesia textbooks, web sites and articles contain very thorough descriptions of MH and related syndromes. However these sources often fail to provide information for patients as well as patient-specific information. Various voluntary organizations throughout the world are dedicated to assisting patients, physicians, anesthesia providers, of all types and any one else in managing the MH susceptible and keeping these individuals up to date with the latest information regarding MH.

In the United States, the Malignant Hyperthermia Association of the United States (MHAUS) provides newsletters, printed information, an informative website <http://www.mhaus.org/> to meet the needs of the various groups interested in MH. In addition a hotline provides direct consultation for providers in real time management of MH episodes or questions related to specific patient as to their likelihood of developing MH and the optimum management of an episode.

MHAUS, similar to other MH patient advocacy organizations is not for profit supported by voluntary contributions.

The North American MH Registry supports a patient-specific database with detailed information as to the phenotypic presentations as well as diagnostic test results. The Registry is a subsidiary of MHAUS and is located at Children’s Hospital of Pittsburgh. The European MH group (<http://www.emhg.org/>) coordinates testing procedures throughout Europe and is made up professionals investigating MH. However, patient supported MH associations exist in France, Germany, Switzerland, Japan, United Kingdom and several other countries. In South Africa, issues related to MH are subsumed under the Muscular Dystrophy Association of that country.

These organizations have been crucial to the education of anesthesia providers in diagnosing and managing MH and helping patients better understand the disorder.

References

American Society of Human Genetics Ad Hoc Committee on Genetic counselling: Genetic Counselling. *Am J Med Genet* 1975; 27: 240 – 242

Anetseder M, Hager M, Muller-Reible C, Roewer N. Diagnosis of susceptibility to malignant hyperthermia by use of a metabolic test. *Lancet* 2002; 359: 1579-80 362(9382) 494-5

Bachand M, Vachond N, Boisvert M *et al*: Clinical reassessment of malignant hyperthermia

in Abitibi-Temiscamingue. *Can J Anaesth* 44:696, 1997

Bendahan D; Kozak-Ribbens G; Rodet L; Confort-Gouny S; Cozzone P. Phosphorus Magnetic Resonance Spectroscopy Characterization of Muscular Metabolic Anomalies in Patients with Malignant Hyperthermia: Application to Diagnosis. *Anesthesiology*. 88(1):96-107, January 1998.

Bendahan D, Guis S, Monnier N, Kozak-Ribbens G, Lunardi J, Ghattas B et al. Comparative analysis of in vitro contracture tests with ryanodine and a combination of ryanodine with either halothane or caffeine: a comparative investigation in malignant hyperthermia. *Acta Anaesthesiol Scand* 2004; 48(8):1019-27.

Brandt A, Schleithoff L, Jurkat-Rott K, et al: Screening of the ryanodine receptor gene in 105 malignant hyperthermia families: novel mutations and concordance with the in vitro contracture test. *Hum Mol Genet* 8: 2055-2062, 1999

Britt BA. Malignant Hyperthermia. *Can Anaesth Soc J* 1985;32(6):666-678

Brown RL, Pollock AN, Couchman KG, Hodges M, Waaka R, Lynch P et al. A novel ryanodine receptor mutation and genotype-phenotype correlation in a large malignant hyperthermia New Zealand Maori pedigree *Human Molecular Genetics* 2000; 9(10): 1515-1524.

Burke W, Pinsky LE, Press NA. Categorizing Genetic Tests to Identify Their Ethical Legal and Social Implications. *Am J Med Genet* 2001; 106: 233 – 240.

Burns AP, Hopkins PM, Hall G, Pusey CD. Rhabdomyolysis and acute renal failure in unsuspected malignant hyperthermia. *Q J Med* 1993; 86:431-34

Central Core Disease are associated with more severe malignant hyperthermia In Vitro Contracture Test Phenotypes. *Human Mutation* 2002;20: 88-97

Chamley D, Pollock NA, Stowell KM, Brown RL. Malignant Hyperthermia in infancy and identification of novel RYR1 mutation. *Br J Anaesth* 2000; 84(4):500-4

Davis M, Brown R, Dickson A, Horton H, James D, Laing N et al. Malignant Hyperthermia associated with exercise-induced rhabdomyolysis or congenital abnormalities and a novel RYR1 mutation in New Zealand and Australian pedigrees. *Br J Anaesth* 2002; 88: 508-515

Denborough MA. Heat stroke and malignant hyperpyrexia. *Med J Aust* 1982; 1(5):204-5

Deufel T, Sudbrak R, Feist Y, Rubsam B, Du Chesne I, Schafer KL et al. Discordance, in a malignant hyperthermia pedigree, between in

vitro contracture-test phenotypes and haplotypes for the MHS1 region on chromosome 19q12-13.2, comprising the C1840T transition in the RYR1 gene. *Am J Hum Genet* 1995; 56(6): 1334-1342.

Ellis FR, Green JH, Campbell IT. Muscle activity, pH and malignant hyperthermia. *Br J Anaesth* 1991; 66(5):535-7

Ellis FR. Malignant Hyperthermia: How important after 30 years. *Acta Anaesthesiologica Belg* 1990; 41: 61-63

Fagerlund TH, Ording H, Bendixen H, Islander G, Ranklev-Twetman E, Berg K. Discordance between malignant hyperthermia susceptibility and RYR1 mutation C1840T in two Scandinavian MH families exhibiting this mutation. *Clin Genet* 1997; 52(6): 416-421.

Fierocche MD, Nivoche Y, Mantz J, Younes E, Veber B, Desmonts J-M. Perioperative severe rhabdomyolysis revealing susceptibility to malignant hyperthermia. *Anesthesiology* 1998; 88:263-5

Fisher NL (ed.) Cultural and Ethnic Diversity - A guide for Genetics Professionals. John Hopkins University Press. 1996.

Fletcher JE, Mayerberger S, Tripolitis L, et al: Fatty acids markedly lower the threshold for halothane-induced calcium release from the terminal cisternae in human and porcine normal and malignant hyperthermia susceptible skeletal muscle. *Life Sci* 49: 1651-7, 1991.

Fletcher JE, Tripolitis L, Rosenberg H, et al: Malignant hyperthermia: halothane- and calcium induced calcium release in skeletal muscle. *Biochem Mol Biol Int* 29: 763-72, 1993.

Fortunato G, Carsana A, Tinto N, Brancadoro V, Canfora G, Salvatore F. A case of discordance between genotype and phenotype in a malignant hyperthermia family. *Eur J Hum Genet* 1999 ; 7(4): 415-20.

Girard T, Cavagna D, Padovan E, Spagnoli G, Urwyler A, Zorzato F, Treves S. B-lymphocytes from malignant hyperthermia-susceptible patients have an increased sensitivity to skeletal muscle ryanodine receptor activators . *J Biol Chem* 2001; 276(51): 48077-82.

Grogan H, Hopkins PM. Heat stroke: implications for critical care and anaesthesia. *Br J Anaesth* 2002; 88(5):700-7

Gronert GA, Thompson RL, Onofrio BM. Human malignant hyperthermia: Awake Episodes and Correction by Dantrolene. *Anesth Analg* 1980; 59(5):377-8

Grover S. The Psychological Dimension of Informed Consent: Dissonance Processes in Genetic Testing. *J Genet Couns* 2003; 12(5): 389 – 403

Gryniewicz G, Poenie M, Tsien RY. A new generation of Ca²⁺ indicators with greatly

improved fluorescence properties. *J Biol Chem* 1985; 260(6): 3440-50.

Haggendal J, Jonsson L, Carlsten J. The role of sympathetic activity in initiating malignant hyperthermia. *Acta Anaesthesiol Scand* 1990; 34(8): 677-682

Hall LW, Woolf N, Bradley JWP, Jolly DW. Unusual reaction to suxamethonium chloride. *BMJ* 1966;2(525):1305

Halsted CH. Pitfalls of genetic testing. *NEJM* 1996; 334(18): 1192 – 1194

Harper PS. Insurance and Genetic testing. *Lancet*. 1993; 341: 224 – 227

Harwood TN, Nelson TE. Massive Postoperative Rhabdomyolysis after uneventful surgery.: A case report of subclinical malignant hyperthermia. *Anesthesiology* 1998; 88:265-8

Hopkins PM. Malignant hyperthermia: advances in clinical management and diagnosis. *British Journal of Anaesthesia*. 85(1):118-28, 2000

Jacobs LA, Deatrick JA. The Individual, the Family and Genetic Testing. *J Prof Nurs* 1999; 15(5): 313 – 324

Juengst ET. Genetic testing and the moral dynamics of family life. *Public Understanding of Science* 1999; 8(9): 193 – 207

Karan SM, Crowl F, Muldoon SM. Malignant Hyperthermia masked by capnographic monitoring. *Anesthesia and Analgesia* 78:590-2,1994

Kessler S, Kessler H, Ward P. Psychological Aspects of Genetic Counseling. III. Management of Guilt and Shame. *Am J Med Genet* 1984; 17: 673 – 697

Klingler W, Baur C, Georgieff M, Lehmann-Horn F, Melzer W. Detection of proton release from cultured human myotubes to identify malignant hyperthermia susceptibility *Anesthesiology* 2002; 97(5): 1059-66

Lambert C, Blanloeil Y, Krivosic-Horber R, et al: Malignant hyperthermia in a patient with hypokalemic periodic paralysis. *Anesth Analg* 79:1012, 1994

Larach MG, Landis JR, Bunn JS *et al*: Prediction of malignant hyperthermia susceptibility in low-risk subjects. An epidemiologic investigation of caffeine–halothane contracture responses. *Anesthesiology* 76:16, 1992

Larach MG, Localio AR, Allen GC, et al: A clinical grading scale to predict malignant hyperthermia Susceptibility *Anesthesiology* 80: 771-779.1994

Larach MG, Rosenberg H, Gronert GA, Allen GC: Hyperkalemic Cardiac Arrest During Anesthetics in Infants and Children with Occult Myopathies. *Clinical Pediatrics*, January 1997, pp. 9-18

Larach MG: Standardization of the caffeine–halothane muscle contracture test. *Anesth Analg* 69:511, 1989

Lehmann-Horn F, Iazzo P. Are myotonias and periodic paralyses associated with susceptibility to malignant hyperthermia. *Br J Anaesth* 1990; 65: 692-97

Light DW. The practice and ethics of risk-rated health insurance. *JAMA* 1992; 267 (18): 2503 – 2508

Loscher W, Witte U, Fredow G, Ganter M, Bickhardt K. Pharmacodynamic effects of serotonin (5-HT) receptor ligands in pigs: stimulation of 5-HT₂ receptors induces malignant hyperthermia. *Arch Pharmacol* 1990;341(6):483-493

MacLennan DH. Discordance between phenotype and genotype in malignant hyperthermia *Curr Opin Neurol* 1995; 8(5): 397-401.

Manning BM, Quane KA, Ording H, Urwyler A, Tegazzin V, Lehane M et al. Identification of novel mutations in the ryanodine-receptor gene (RYR1) in malignant hyperthermia: genotype-phenotype correlation. *Am J Hum Genet* 1998; 62(3): 599-609.

Marteau TM, Croyle RT. Psychological responses to genetic testing. *Br Med J* 1998; 316: 693 – 696

Mauritz W, Hackl W, Winkler M. Malignant Hyperthermia: state of the art. *Acta Anaesthesiol Scand Suppl*, 1997;111:310-2

Melzer W, Dietze B. Malignant hyperthermia and excitation-contraction coupling. *Acta Physiol Scand* 2001; 171(3): 367-78.

McCarthy TV, Healy JM, Heffron JJ, Lehane M, Deufel T, Lehmann-Horn F et al. Localization of the malignant hyperthermia susceptibility locus to human chromosome 19q12-13.2. *Nature* 1990; 343(6258): 562-564.

McCarthy TV, Quane KA, Lynch PJ. Ryanodine receptor mutations in malignant hyperthermia and central core disease *Hum Mutat* 2000; 15(5): 410-7.

Melzer W, Dietze B. Malignant hyperthermia and excitation-contraction coupling. *Acta Physiol Scand* 2001; 171(3): 367-78.

Monnier N, Krivosic-Horber R, Payen JF, Kozak-Ribbens G, Nivoche Y, Adnet P et al. Presence of two Genetic Traits in Malignant Hyperthermia Families. *Anesthesiology* 2002; 97(5): 1067-74

Monnier N, Procaccio V, Stieglitz P, Lunardi J. Malignant-Hyperthermia Susceptibility is Associated with a Mutation of the α 1-Subunit of the Human Dihydropyridine-Sensitive L-Type Voltage-Dependent Calcium-Channel Receptor in Skeletal Muscle. *Am J Hum Genet* 1997; 60(6): 1316 - 1325.

- O'Flynn RP**, Shutack JG, Rosenberg H, Fletcher JE, Masseter muscle rigidity and malignant hyperthermia susceptibility in pediatric patients. *Anesthesiology* 80: 1228-33, 1994
- Ong RO**, Rosenberg H: Malignant hyperthermia-like syndrome associated with metrizamide myelography. *Anesthesia and Analgesia* 68:795-797, 1989.
- Ording H**, Bendixen D. Sources of variability in halothane and caffeine contracture tests for susceptibility to malignant hyperthermia *Eur J Anaesthesiol* 1992; 9(5): 367-376.
- Ording H**, Brancadoro V, Cozzolino S, Ellis FR, Glauber V, Gonano EF et al. In vitro contracture test for diagnosis of malignant hyperthermia following the protocol of the European MH Group: results of testing patients surviving fulminant MH and unrelated low-risk subjects. *The European Malignant Hyperthermia Group Acta Anaesthesiol Scand* 1997; 41(8): 955-66
- Ording H**, Glahn K, Gardi T, Fagerlund T, Bendixen D. 4-Chloro-m-cresol test--a possible supplementary test for diagnosis of malignant hyperthermia susceptibility *Acta Anaesthesiol Scand* 1997; 41(8): 967-972.
- Ording H**. Incidence of Malignant Hyperthermia in Denmark. *Anesth Analg* 1985; 64(7):700-4
- Ording H**, Glahn K, Gardi T, Fagerlund T, Bendixen D. 4-Chloro-m-cresol test--a possible supplementary test for diagnosis of malignant hyperthermia susceptibility *Acta Anaesthesiol Scand* 1997; 41(8): 967-972..
- Pollock AN**, Langton EE, Couchman K, Stowell KM, Waddington M. Suspected malignant hyperthermia reactions in New Zealand. *Anaesth Intensive Care* 2002; 30(4): 453-61.
- Querfurth HW**, Haughey NJ, Greenway SC, Yacono PW, Golan DE, Geiger JD. Expression of ryanodine receptors in human embryonic kidney (HEK293) cells. *Biochem J* 1998; 334(Pt 1): 79-86.
- Quinlivan RM**, Muller CR, Davis M, Laing NG, Evans GA, Dwyer J et al. Central core disease: clinical, pathological and genetic features. *Arch Dis Child* 2003;88: 1051-1055
- Richter M**, Schleithoff L, Deufel T, Lehmann-Horn F, Herrmann-Frank A. Functional characterization of a distinct ryanodine receptor mutation in human malignant hyperthermia-susceptible muscle. *J Biol Chem* 1997; 272(8): 5256-5260.
- Robinson RL**, Brooks C, Brown SL, Ellis FR, Halsall PJ, Quinnell RJ et al. Mutations causing central core disease are associated with more severe malignant hyperthermia in vitro contracture test phenotypes. *Hum Mutat* 2002; 20(2): 88-97.
- Robinson RL**, Anetseder MJ, Brancadoro V, van Broekhoven C, Carsana A, Censier K et al. Recent advances in the diagnosis of Malignant Hyperthermia susceptibility: How confident can we be of genetic testing? *Eur J Hum Genet* 2003;11(4):342-48
- Rueffert H**, Koenig F, Meinecke CD, Olthoff D. The Ile2453Thr mutation in the ryanodine receptor gene 1 is associated with facilitated calcium release from sarcoplasmic reticulum by 4-chloro-m-cresol in human myotubes. *Cell Calcium* 2003; 34(2): 163-8.
- Rueffert H**, Olthoff D, Deutrich C, Meinecke CD, Froster UG. Mutation screening in the ryanodine receptor 1 gene (RYR1) in patients susceptible to malignant hyperthermia who show definite IVCT results: identification of three novel mutations. *Acta Anaesthesiol Scand* 2002; 46(6): 692-8.
- Sambuughin N**, McWilliams S, de Bantel A, Sivakumar K, Nelson TE. Single-amino-acid deletion in the RYR1 gene, associated with malignant hyperthermia susceptibility and unusual contraction phenotype *Am J Hum Genet* 2001; 69(1): 204-8 Epub 2001 May 29.
- Sei Y**, Brandom BW, Bina S, Hosoi E, Gallagher KL, Wyre HW et al. Patients with malignant hyperthermia demonstrate an altered calcium control mechanism in B lymphocytes. *Anesthesiology* 2002; 97(5):1052-8.
- Schwartz L**, Rockoff MA, Koka BV: Masseter spasm with anesthesia: incidence and implications *Anesthesiology* 61: 772-5, 1984.
- Serfas KD**, Bose D, Patel L, Wrogemann K, Phillips MS, MacLennan DH, Greenberg CR. Comparison of the segregation of the RYR1 C1840T mutation with segregation of the caffeine/halothane contracture test results for malignant hyperthermia susceptibility in a large Manitoba Mennonite family. *Anesthesiology* 1996; 84(2): 322-329.
- Shepherd S**, Ellis F, Halsall J, Hopkins P, Robinson R. RYR1 mutations in UK central core disease patients: more than just the C-terminal transmembrane region of the RYR1 gene. *J Med Genet*. 2004 Mar;41(3):e33.
- Skene L**, Smallwood R. Informed consent: Lessons from Australia. *Br Med J* 2002; 324: 39 – 41
- Strazis KP**, Fox AW. Malignant Hyperthermia: A Review of Published Cases. *Anesth Analg* 1993;77(2):297-304
- Tilgen N**, Zorzato F, Halliger-Keller B, Muntoni F, Sewry C, Palmucci LM et al. Identification of four novel mutations in the C-terminal membrane spanning domain of the ryanodine receptor 1: association with central core disease and alteration of calcium homeostasis. *Hum Mol Genet* 2001; 10(25): 2879-87.

- Tobin JR**, Jason DR, Challa VR, Nelson TE, Sambuughin N. Malignant Hyperthermia and Apparent Heat Stroke. *JAMA* 286: 168-9,2001
- Treves S**, Larini F, Menegazzi P, Steinberg TH, Koval M, Vilsen B et al. Alteration of intracellular Ca²⁺ transients in COS-7 cells transfected with the cDNA encoding skeletal-muscle ryanodine receptor carrying a mutation associated with malignant hyperthermia. *Biochem J* 1994; 301(Pt 3): 661-665.
- Tobin JR**, Jason DR, Challa VR, Nelson TE, Sambuughin N. Malignant hyperthermia and apparent heat stroke. *JAMA* 2001; 286(2):168-9
- Urwyler A**, Deufel T, McCarthy T, West S. Guidelines for molecular genetic detection of susceptibility to malignant hyperthermia. *Br J Anaesth* 2001; 86(2): 283-7.
- Vainzof M**, Muniz VP, Tsanaclis AM, Silva HC, Rusticci MS. Does the A3333G mutation in the CACNL1A3 gene, detected in malignant hyperthermia, also occur in central core disease? *Genet Test* 2000; 4(4): 383-6
- Vita GM**, Olckers A, Jedlicka AE, et al: Masseter muscle rigidity associated with glycine1306-to-alanine mutation in the adult muscle sodium channel alpha-subunit gene. *Anesthesiology* 82: 1097-103, 1995.
- Wappler F**, Fiege M, Antz M, Schulte am Esch J. Haemodynamic and metabolic alterations in response to graded exercise in a patient susceptible to malignant hyperthermia. *Anesthesiology* 2000; 92(1):268-272
- Wappler F**, Fiege M, Schulte am Esch J. Pathophysiological role of serotonin system in malignant hyperthermia. *Br J Anaesth* 2001; 87(5):794-8
- Wappler F**, Fiege M, Steinfath M, Agarwal K, Scholz J, Singh S et al. Evidence for susceptibility to malignant hyperthermia in patients with exercise-induced rhabdomyolysis. *Anesthesiology* 2001; 94(1):95-100
- Wappler F**, Roewer N, Kochling A, Scholz J, Loscher W, Steinfath M. Effects of the serotonin₂ receptor agonist DOI on skeletal muscle specimens from malignant hyperthermia-susceptible patients. *Anesthesiology* 1996; 84(6):1280-7
- Wehner M**, Rueffert H, Koenig F, Olthoff D. Functional characterization of malignant hyperthermia-associated RyR1 mutations in exon 44, using the human myotube model. *Neuromuscul Disord* 2004; 14(7): 429-37.
- Wehner M**, Rueffert H, Koenig F, Neuhaus J, Olthoff D. Increased sensitivity to 4-chloro-m-cresol and caffeine in primary myotubes from malignant hyperthermia susceptible individuals carrying the ryanodine receptor 1 Thr2206Met (C6617T) mutation *Clin Genet* 2002; 62(2): 135-46.
- Wehner M**, Rueffert H, Koenig F, Olthoff D. Calcium release from sarcoplasmic reticulum is facilitated in human myotubes derived from carriers of the ryanodine receptor type 1 mutations Ile2182Phe and Gly2375Ala. *Genet Test* 2003; 7(3): 203-11.
- Wieland SJ**, Fletcher JE, Rosenberg H, et al: Malignant hyperthermia: slow sodium current in cultured human muscle cells. *Am J Physiol* 257: C759-65, 1989.
- Wingard DW**. Letter: Malignant hyperthermia – a human stress syndrome? *Lancet* 1974; 2:1450-1
- Yang T**, Ta TA, Pessah IN, Allen PD. Functional defects in six ryanodine receptor isoform-1 (RyR1) mutations associated with malignant hyperthermia and their impact on skeletal excitation-contraction coupling. *J Biol Chem* 2003;278(28) : 25722-30