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Malignant Hyperthermia: Normal Muscle Calcium Uptake and Abnormal Caffeine and Halothane Contractures

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In 1986, thin-strip muscle calcium uptake was abandoned as a diagnostic test for susceptibility to malignant hyperthermia (MH). Physicians were informed that the test did not discriminate between the normal and the abnormal patient.** It appeared to be biased toward false positive results.[1,2] We now report a patient with a false negative calcium uptake test.

CASE REPORT

This 32-yr-old, 168-cm, 57-kg woman had had a healthy brother who had died at age 20 in 1970 from an apparent MH episode with hyperthermia (41° C) and cardiac arrest. In 1983, during epidural anesthesia, she underwent biopsy of her left vastus lateralis muscle for MH testing. The report was: "Calcium uptake by thin sections (at 37° C): 8.3 μmol · g muscle^{-1} · min^{-1}; actomyosin adenosine triphosphatase activity by thin sections: 0.18 μmol phosphate · g muscle^{-1} · min^{-1}. The results are unquestionably *negative;* therefore, you are not susceptible to malignant hyperthermia."

In 1990, the patient became concerned about the validity of her test results and requested a second testing. During femoral nerve and lateral femoral cutaneous nerve blocks and sedation with morphine 4 mg and midazolam 3 mg, the surgeon excised the old cutaneous scar, exposed the muscle, and removed the specimen from normal-appearing muscle alongside the prior biopsy scar. Histology and histochemistry performed on a portion of the sample was normal except for three type-1 fiber "type groupings" and one area of pyknotic nuclear clumps, presumably representing the remnants of a fascicle. These findings were consistent with previous denervation of a few fibers and subsequent collateral reinnervation.

Contracture testing was performed according to the North American protocol.[3] Bundles were tested in triplicate, and only freshly cut bundles were used for each drug exposure, which consisted of 1) halothane alone at 3%, 2) incremental doses of caffeine in the presence of halothane 1%, and 3) caffeine alone in incremental doses (table 1). Twitch heights of 8–19 g tension confirmed viability. Muscle testing concluded within 2.5 h after excision.

All nine bundles tested showed susceptibility (table 1). 1) Halothane 3% contractures are positive when greater than 0.5–0.8 g (North American protocol).[3] 2) A halothane–caffeine specific concentration less than 0.5 mM is abnormal, and contracture with halothane 1% alone during that test also is abnormal.[3] 3) A caffeine-specific concentration of less than 4 mM is abnormal, as is a contracture increase at 2 mM that is greater than 7% of maximum tension, or an absolute increase of more than 0.2 g at 2 mM.[3] Tension increases with less than 2 mM caffeine are unusual even in susceptible patients (table 1).

DISCUSSION

The currently accepted muscle contracture test requires a fresh, functioning, 8-g specimen; the patient must travel to one of the few laboratories that perform the test. Muscle contracture testing has a solid, 22-yr base of evidence supporting its accuracy,[4–6] but the convenience of mailing a sample to the laboratory helped to popularize muscle calcium uptake as an MH test. Thin-strip muscle calcium uptake required a 0.1–0.3 g specimen, immediately chilled in liquid nitrogen. Ten-micrometer sections were assayed for calcium uptake. This was believed to correlate with calcium uptake measured in specimens of fragmented sarcoplasmic reticulum.[7] One group of investigators, supporting the validity of MH testing by thin-strip calcium uptake, compared it to muscle contracture testing in 11 subjects (5 control and 6 susceptible), and although they did not include specific data, they reported that the test results coincided.[8]

However, there were problems with the test. One case report of a patient experiencing an MH episode and treated with cardiopulmonary bypass included data showing abnormal calcium uptake during the acute episode and normal uptake after recovery.[9] This manifest contradiction of test result and diagnosis was ignored. In a study of masseter spasm, all 12 of the 15 children with masseter spasm who had subsequent muscle biopsies tested positive by calcium uptake; this led to an estimation of MH susceptibility of at least 1% in the general population, if the test was valid.[1] Careful study of the calcium uptake test by another laboratory showed that known normal and MH-susceptible patients are indistinguishable by calcium uptake.[10] The pathology laboratory at the original institution reached a similar conclusion.** Although thin-

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** Hedley-White ET: Massachusetts General Hospital abandons calcium uptake muscle test. The Communicator. Malignant Hyperthermia Association of the United States. 5(3), Spring 1987, p 1.

TABLE 1. Muscle Contracture Responses

Test	Bundle 1	Bundle 2	Bundle 3
Halothane alone 3% (g contracture)	2.81	1.02	6.58
Caffeine with halothane			
Halothane 1% (g contracture)	1.26	0.68	2.75
HCSC (mM)	0.06	0.21	Tore
Caffeine alone			
0.5 mM (g contracture)	0.52	0.14	0.26
1 mM (g contracture)	1.16	0.35	0.68
2 mM (g contracture)	2.35	Tore	2.21
Increase (%) at 2 mM	18	Tore	22
CSC	0.9	Tore	1.2

HCSC = millimolar concentration of caffeine at which tension increases 1 g in the presence of 1% halothane; Tore = muscle bundle tore before test completion; CSC = millimolar concentration of caffeine at which tension increases 1 g.

$$\text{Increase (\%) at 2 mM} = \frac{\text{tension at 2 mM} - \text{baseline tension}}{\text{tension at 32 mM} - \text{baseline tension}} \times 100$$

strip muscle calcium uptake may reflect sarcoplasmic reticulum calcium uptake activity, the basic defect in MH appears to be in sarcoplasmic reticulum calcium release.[11] This explains in part the lack of correlation between the calcium uptake test and MH susceptibility.

For our patient, it is unlikely that the characteristics of the two muscle specimens were discordant. Her physical activity was similar (producing similar ratios of slow-twitch to fast-twitch fibers), and the same muscle was used. Histologic examination confirmed that the sample was functional, with some evidence of denervation and reinnervation, probably due to nerve fibers cut by the biopsy 7 years before or to localized recent trauma. Denervation *per se* does not result in MH characteristics in skeletal muscle.[12] The patient's abnormal test results were unequivocal, in fact, to an extent seldom seen in tests of MH-positive patients.

Our patient had a false negative calcium uptake test—an unexpected finding given the high false-positive rate. False positive results complicate a person's life: there is difficulty in obtaining treatment, and there is the need to have relatives tested. A false negative result, in contrast, jeopardizes the patient with the hazard of future exposure to triggering agents. A proponent of the muscle calcium uptake test stated that his laboratory had tested 1,200 muscle specimens and counseled 600 families.†† We believe that these people should be made aware of contradictions between the tests.

†† Ryan JF: Ask the experts. The Communicator. Malignant Hyperthermia Association of the United States. 5(3), Spring 1987, p 5.

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