

THE EUROPEAN MALIGNANT HYPERTHERMIA GROUP

- Protocol for a 4-chloro-*m*-cresol bolus IVCT -

4-chloro-*m*-cresol: C₇H₇ClO (4-chlor-3-methylphenol)

(4-CmC)

Aldrich-Chemie; D-89555 Steinheim
Cat. No. C 5,540-2; 100 g; DM 11,90-

FLUKA Chemie; D-89231 Neu-Ulm
Product No. 24940; 250 g; DM 15.90-

E. Merck; D-64271 Darmstadt
Product No. 820314; 250 g; DM 15.00-

Stock solution: 25 mM in distilled water
(solubility = 28 mM in aqueous solution; Arch. Biochim. 1955;54:55-61)

Storage: Stock solution should be freshly prepared before each experiment.

Test concentration: Final bath concentration of 75 µmol/l 4-CmC

Bath concentration: The concentration of 4-CmC in the tissue bath must be periodically checked by HPLC.

Test procedure: Preparation and procedure must be according to our protocol.

1. For each test a fresh muscle specimen must be used.
2. Time from biopsy to completion of the test should not exceed 5h.
3. The test should be performed at optimal length (l_0).
4. The muscle specimen should be electrically stimulated with a 1-2 ms stimulus at a frequency of 0.2 Hz.
5. Baseline must not vary more than 2 mN within a 10 min period before addition of 4-CmC.
6. Viability criteria should be according to the standard tests.
7. A single bolus dose technique is used to reach a final bath concentration of 75 µmol/l 4-CmC. The final bath concentration of 4-CmC must be reached as fast as possible (< 1 min!).

- Test parameters:**
1. weight (mg)
length (mm)
twitch height (mN)
maximum preload (mN)
predrug baseline (mN)
 2. onset time (OT) of contracture (min)
time to reach contracture of 2 mN (TC_{2mN} ; min)
time to reach contracture of 10 mN (TC_{10mN} ; min)
time to reach contracture maximum (TC_{max} ; min)
contracture maximum (C_{max} ; mN)

Controls: IVCT with 4-CmC should, if possible, also performed in control biopsies

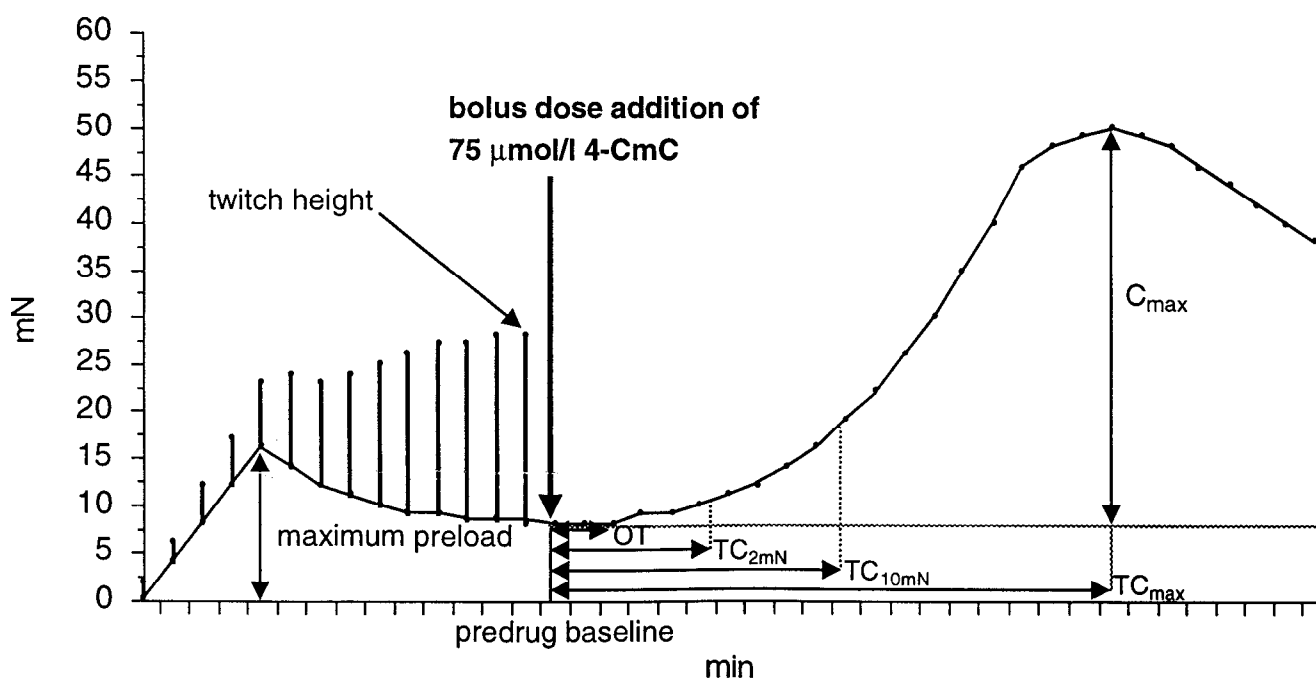


Figure 1: Method of data evaluation.