**General approach**

A muscle fiber was modeled as a cylinder of length *L* and radius *RC*. The intracellular compartment is connected to the extracellular space by a capacitance and 7 or 8 ion channels: voltage-gated K+ (Kv), inward-rectifying K+ (Kir), Chloride (ClC-1), Na-K ATPase pump (NaK), persistent Na+ (NaP), Na+ Leak (NaL), NaKCl cotransporter (NKCCl), and when needed a gating pore passing Na+ (GP). Only 2 compartments were considered, intracellular and extracellular. The t-tubule space was not included as it is expected to behave similarly to the extracellular space when dealing with steady state or slowly changing conditions (Seebohm et al., 2012).

The charge-difference approach (Fraser et al., 2011; Fraser & Huang, 2004) was used to calculate ion fluxes, ion concentration changes, and volume changes. Osmolarity of all compartments was fixed at 300 mOsm. Water flux to maintain osmolarity was instantaneous. Thus the ode solver calculates new ion numbers in each compartment, from which the volume can be calculated based on osmolarity. Then concentrations are calculated from ion numbers and compartment volumes. Impermeant intracellular (*X*) and extracellular (eA) anions were included with valences *zX* and *zeA* respectively to maintain charge neutrality. Additional neutral intracellular osmolyte (*Cosm,i*) was added to balance osmolarity without having to track molecules other than Na+, K+, and Cl-. Even though the volume of each compartment can change, the surface area of each membrane remains the same (Fraser & Huang, 2004). The extracellular space is at a constant, uniform potential at all times.

All simulations were performed in Matlab using a stiff differential equation solver (*ode15s*).

**Main Equations**

where *ion\_chan* is a channel or pump that transports *ion*

where *ion* ϵ {Na+, K+, Cl-, X-}

**Ion channel summary**

The Kv channel uses a standard Hodgkin-Huxley approach. The NaP and GP channels use a modified Hodgkin-Huxley approach where the dependence of steady state gating constants (*m∞, h∞, a∞*) and the time constants (*τm, τh, τa*) on voltage are specified without using α and 𝛽 rate constants. The ClC-1 and Kir channels follow previously defined functions with all parameters unchanged except permeabilities were modified to match time constants from our experiments(DiFranco et al., 2011; Struyk & Cannon, 2008). The NaK ATPase pump follows a previously defined function with all parameters unchanged except the maximum pump activity was reduced to assist with setting a stable resting potential (Wallinga et al., 1999). The NKCCl transporter used a concentration dependent approach with saturation (Fraser and Huang, 2004).

**Ion channels Kv, Kir, ClC-1, NaP, NaL, GP**

Unless V = V’, in which case

**Ion channel Kv**

**Ion channel Kir**

**Ion channel ClC1**

**Ion channels NaP and GP**

where *x* ϵ *{m,h,a}*

for x = m

for x = h,n

**NaK ATPase pump**

**NaKCl cotransporter**

**Ion channel NaL**

**Definitions/Glossary**

|  |  |
| --- | --- |
| Superscripts |  |
| e | Extracellular compartment |
| s | Either the intracellular volume or a membrane quantity across the sarcolemma |
|  |  |
| Subscripts |  |
| sm | Sarcolemma membrane |
|  |  |
|  | Sarcolemmal surface area of a single longitudinal segment (*cm2*) |
| *b* | Activation parameter for Kir |
|  | Max of activation parameter for Kir |
|  | Concentration difference driving force for NKCCl (*mM*) |
|  | Effective driving force including saturation for NKCCl (*mM)* |
| *Cm* | Specific membrane capacitance (*uF/cm2)* |
|  | Sarcolemmal capacitance (*nF*) |
| *Cosm,i* | Concentration of intracellular neutral osmolyte (*mM*) |
|  | Conductance factor for membrane () |
| *eA* | Concentration of extracellular impermeant anion (*mM*) |
|  | Nernst potential for ion (*mV*) |
|  | Voltage-dependent portion of NaK ATPase pump current |
| F | Faraday’s constant (*C/mole*) |
|  | Goldman-Hodgkin-Katz driving force for ion channel transport (*mV mM)* |
| h | Inactivation parameter for NaP channel |
|  | Baseline current for NaK and NKCCl channels (*nA*) |
|  | Current through an ion channel (*nA*) |
|  | Maximum possible current through an ion channel (*nA*) |
|  | Net current from NaK ATPase pump (*nA*) |
|  | Na+ and K+ currents due to NaK ATPase pump (*nA*) |
|  | Net current from NKCCl contransporter (*nA*) |
|  | Na+, K+ , and Cl- currents due to NKCCl cotransporter (*nA*) |
|  | Concentration of ion, where ion ϵ {Na+, K+, Cl-} (*mM*) |
| j | Activation parameter for ClC-1 channel |
|  | Maximum activity for NaK ATPase pump (*µmole/(cm2 s)*) |
|  | Maximum current for NKCCl cotransporter (*nA/cm2*) |
| , | Boltzmann slope factor for ClC-1 rate constants (*mV*) |
|  | Boltzmann slope factor for Kv rate constants (*mV*) |
| *Kb* | Slope factor for Kir voltage dependence (*mV*) |
|  | Affinity constants for [K]o and [Na]s for NaK ATPase pump (*mM*) |
|  | Slope factors for x = m,h,a for NaP and GP (mV) |
| *L* | Length of cell (*cm*) |
| m | Activation parameter for NaP channel |
| n | Activation parameter for Kv channel |
|  | number of molecules of ion in a compartment where ion ϵ {Na+, K+, Cl-} (*nanomoles*) |
|  | Fraction of maximum possible current |
|  | Permeability of ion channel (*cm/s*) |
| Q | Net charge in a compartment (*nC*) |
| *R* | Gas constant (*J/(K mole)*) |
|  | Radius of cell (*cm*) |
| *SSF* | Sigmoid Step Function to describe voltage dependence of gating parameter constants. |
| T | Temperature (*K*) |
| *V* | Voltage across membrane (*mV*) |
| *V’* | Effect of surface charge layers on ion channel current (*mV*) |
|  | Midpoint of V dependence for Kv rate constants (mV) |
|  | Midpoint of m,h,a dependence for NaP and GP (mV) |
|  | Voltage at which ClC-1 rate constants are half maximal (*mV*) |
| *x* | Time varying value of gating parameter, where *x* ϵ {*m,h,a*} |
|  | Voltage dependent steady-state value of gating parameters for Nav and Kv channels where *x* ϵ {*m,h,a*} |
| *X* | Concentration of impermeant intracellular anion (*mM*) |
| *zeA* | Valence of impermeant extracellular anion |
|  | Valence of *ion* |
| *zx* | Valence of impermeant intracellular anion |
|  | Hodgkin Huxley rate constants for Kv ((ms mV)-1 orms-1) |
|  | Rate constants for ClC-1 channel (*ms*) |
|  | Maximum rate constants for ClC-1 channel (*ms*) |
|  | Concentration difference for 50% max driving force for NKCCl (mM4) |
|  | Max driving force for NKCCl cotransporter |
|  | Shift of Kir permeability based on EK and Vm (*mV*) |
|  | [Na]o dependent variable for voltage-dependent part of NaK ATPase pump |
|  | Time constant and factors for Kir channel (*ms* or *mV*) |
| ,,  , | Voltage dependent time constant for gating parameter for NaP and GP channels where *x* ϵ {*m,h,a*} (*ms* or *mV*) |

**Parameters**

General Parameters

|  |  |  |
| --- | --- | --- |
| Symbol | Unit | Value |
| *Cm* | µF/cm2 | 0.9a |
| *Cosm,i* | mM | 125.02b |
| eA | mM | 40b |
| *F* | C/Mole | 96485.33 |
| *L* | cm | 1 |
| *R* | J/(mole K) | 8.3144598 |
| *RC* | cm | .003 |
| *T* | K | 305 |
| X | mM | 61.18b |
| zeA |  | -1 |
| zx |  | -1.6477c |

a (Hodgkin & Nakajima, 1972)

b For charge neutrality and total 300 mOsm for WT

c (Fraser & Huang, 2004)

Channel parameters for wild type

|  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Symbol | Unit | Kv | Kir | ClC-1 | NaK | NaP | NKCCl | NaL | GP |
|  | mS/cm2 |  |  |  |  |  |  |  |  |
|  | µmol/(cm2s) |  |  |  | 40x10-6 a |  |  |  |  |
|  | nA/cm2 |  |  |  |  |  | 3.7e-8 |  |  |
|  | mV |  |  | 41b |  |  |  |  |  |
|  | mV |  |  | 25b |  |  |  |  |  |
|  | mV | 4.192 |  |  |  |  |  |  |  |
|  | mV | 17.42 |  |  |  |  |  |  |  |
|  | mM |  |  |  | 1c |  |  |  |  |
|  | mM |  |  |  | 13c |  |  |  |  |
| *P* | cm/s | 2.86e-4 | 1e-5 | 3e-4d |  | 0.9e-8 |  | 2e-9 | 0f |
| *V’* | mV | 0 | 0 | -112.89e |  | 0 |  | 0 | 0 |
|  | mV |  |  | -50b |  |  |  |  |  |
|  | mV |  |  | -90b |  |  |  |  |  |
|  | mV | -47.0 |  |  |  |  |  |  |  |
|  | ms-1 |  |  | 0.03b |  |  |  |  |  |
|  | ms-1 |  |  | 0.16b |  |  |  |  |  |
|  | (ms mV)-1 | 0.0136 |  |  |  |  |  |  |  |
|  | ms-1 | 0.0840 |  |  |  |  |  |  |  |
|  | mM4 |  |  |  |  |  | 1.2e6 |  |  |
|  | mM4 |  |  |  |  |  | 7e6 |  |  |
|  | mV |  | -19.5 |  |  |  |  |  |  |

a Set to improve stability of baseline resting potential.

b (DiFranco et al., 2011a)

c (Wallinga et al., 1999)

d Set to match passive time constant from experiments

e Derived from [Cl]m parameter of (DiFranco et al., 2011a)

f Set to 1.22e-8 for hypoKPP mutation

Gating Parameters for Kir, NaP, and GP

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Symbol | Unit | x = b | x = m | x = h | x = n |
|  | mV | 13.7 | 3 | 3 | 15 |
|  | ms | 5 | 5 | 5 | 5 |
|  | mV | 0 | -50 | -50 | -50 |
|  | mV | 500 | 100 | 100 | 100 |
|  | mV | na | -67.5 | -20 | -60 |

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