Chapter 13
Mutations Affecting the Mean Open Time

In the simplest case of Markov models of the form

$$C \underset{k_{co}}{\overset{k_{oc}}{\leftrightarrows}} O, \tag{13.1}$$

we have studied mutations leading to an increased open probability by increasing the rate from closed (C) to open (O), given by k_{co}. We refer to these as CO-mutations and for such mutations we have successfully derived closed state blockers represented as

$$B \underset{k_{bc}}{\overset{k_{cb}}{\leftrightarrows}} C \underset{\mu k_{co}}{\overset{k_{oc}}{\leftrightarrows}} O, \tag{13.2}$$

where $\mu \geqslant 1$ is the mutation severity index and $\mu = 1$ represents the wild type. These blockers can completely repair the equilibrium open probability of the mutant by adjusting the "on rate" divided by the "off rate" of the drug given by

$$\delta_c = \frac{k_{cb}}{k_{bc}}$$

(see, e.g., page 58). The remaining degree of freedom can be found using probability density systems and the resulting drugs have been proven to work exceptionally well in theoretical computations.

A. Tveito, G.T. Lines, *Computing Characterizations of Drugs for Ion Channels and Receptors Using Markov Models*, Lecture Notes in Computational Science and Engineering 111, DOI 10.1007/978-3-319-30030-6_13

There is, however, another way of modeling increased equilibrium open probability. Rather than increasing the rate from C to O, we can reduce the rate from O to C:

$$C \underset{k_{co}}{\overset{k_{oc}/\mu}{\leftrightarrows}} O, \tag{13.3}$$

where again $\mu \geqslant 1$ is referred to as the mutation severity index. This type of mutation is referred to as an OC-mutation and the equilibrium open probability for this Markov model is given by

$$o = \frac{1}{1 + \frac{k_{oc}/\mu}{k_{co}}},$$

which clearly increases for increasing values of μ. Formally, we can carry out the same math to devise a closed state drug that completely repairs the equilibrium open probability of the mutant; however, when this drug is put into the probability density system to determine the remaining degree of freedom of the drug, we quickly observe that the task is impossible and the theoretical drug does not provide significant improvement.

The core difficulty here is that a CO-mutation does not change the mean open time of the channel. A closed state blocker is therefore well suited because such a blocker does not affect the mean open time. However, for an OC-mutation, an increased mean open time is part of the problem and a closed state blocker is not the solution, simply because it cannot affect the mean open time. Rather, an open state blocker must be used.

In this chapter, we will explain the notion of mean open time and study mutations that lead to an increased open probability *and* an increased mean open time. We will show that open state blockers are optimal for such mutations.

13.1 The Mean Open Time

Let us briefly recall the interpretation of the Markov model

$$C \underset{k_{co}}{\overset{k_{oc}}{\leftrightarrows}} O.$$

This scheme means that if the channel is closed (C), the probability of changing the state from closed to open (O) in a small time interval Δt is given by $k_{co}\Delta t$. Clearly, this interpretation only holds for short time intervals, since the probability cannot exceed one. Note also that if the rate k_{co} increases, this leads to an increased probability of moving from C to O during the time step Δt. Similarly, $k_{oc}\Delta t$ denotes the probability of moving from the open state to the closed state in the time step Δt.

Suppose that the channel is open at time $t = 0$. The probability that the channel remains open after a short time step Δt is given by

$$p_1 = 1 - k_{oc}\Delta t.$$

If we take another time step, the probability that the channel is still open at time $t = 2\Delta t$ is given by

$$p_2 = p_1 (1 - k_{oc}\Delta t) = (1 - k_{oc}\Delta t)^2$$

and so on. At time $t = n\Delta t$, the probability of the channel still being open is given by

$$p_n = (1 - k_{oc}\Delta t)^n .$$

If we now introduce time given by

$$t = n\Delta t,$$

we have

$$(1 - k_{oc}\Delta t)^n = (1 - k_{oc}\Delta t)^{\frac{t}{\Delta t}} .$$

The probability of closing a channel that is in the open state during a time step is given by $\Delta t k_{oc}$ and therefore the probability of closing a channel that has remained open for n time steps is given by

$$\Delta t k_{oc} (1 - k_{oc}\Delta t)^{\frac{t}{\Delta t}} .$$

The expected open time is therefore given by

$$\sum_{n=1}^{\infty} n\Delta t (1 - k_{oc}\Delta t)^{\frac{t}{\Delta t}} \Delta t k_{oc}.$$

If we go to the limit of $\Delta t \to 0$ in this expression, we find that

$$\sum_{n=1}^{\infty} n\Delta t (1 - k_{oc}\Delta t)^{\frac{t}{\Delta t}} \Delta t k_{oc} \overset{\Delta t \to 0}{\longrightarrow} \int_0^{\infty} t k_{oc} e^{-k_{oc} t} dt = \frac{1}{k_{oc}}$$

and therefore we have found that the mean open time is given by

$$\tau_o = \frac{1}{k_{oc}}. \tag{13.4}$$

13.1.1 Mean Open Time for More Than One Open State

We have seen that the mean open time for a Markov model of the form

$$C \underset{k_{co}}{\overset{k_{oc}}{\leftrightarrows}} O$$

is given by

$$\tau_o = \frac{1}{k_{oc}}. \tag{13.5}$$

It is straightforward to extend the argument above to see that, for a Markov model of the form

$$C \underset{k_{co}}{\overset{k_{oc}}{\leftrightarrows}} O \underset{k_{ob}}{\overset{k_{bo}}{\leftrightarrows}} B,$$

the mean open time is given by

$$\tau_o = \frac{1}{k_{oc} + k_{ob}}. \tag{13.6}$$

But what happens if there is more than one open state? This situation will become relevant below, where we consider models including a burst mode. The models contain at least two open states. To understand the mean open time in the presence of more than one open state, we consider the generic extension illustrated in Fig. 13.1.

Assuming that the rates are set according to the principle of detailed balance, we have

$$k_{ul} o_u = k_{lu} o_l,$$

where o_u and o_l are the probabilities of being in the states O^u or O^l, respectively, and u and l represent the upper and lower states, respectively.

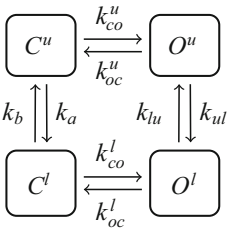

Fig. 13.1 Markov model with two open states (O^u, O^l) and two closed states (C^u, C^l)

As for the derivation above, we assume that the channel is open and our task is to figure out how long we can expect the channel to remain open. We know that, initially, the channel is either in the state O^u or O^l. Let us define q_u and q_l to be the conditional probabilities of being in the upper and lower open states, given that the channel is open. For the upper state we write

$$q_u = P(S = O_u | (S = O_u \text{ or } S = O_l)),$$

where $S = X$ means that the channel is in state X. Since

$$P(A|B) = P(A \text{ and } B)/P(B)$$

and, in our case, since (A and B) = A, we obtain

$$q_u = P(S = O_u)/P(S = O_u \text{ or } S = O_l) = \frac{o_u}{o_u + o_l}$$

and similarly for the lower state; with

$$q_l = P(S = O_l | (S = O_u \text{ or } S = O_l)),$$

we obtain

$$q_l = \frac{o_l}{o_u + o_l}.$$

It follows that $q_u + q_l = 1$ and that

$$q_u = \frac{k_{lu}}{k_{ul} + k_{lu}}$$

and

$$q_l = \frac{k_{ul}}{k_{ul} + k_{lu}}.$$

The probability of remaining in the open states in the first time step is now given by

$$\begin{aligned} p_1 &= \left(1 - \Delta t k^u_{oc}\right) q_u + \left(1 - \Delta t k^l_{oc}\right) q_l \\ &= 1 - \Delta t \left(\frac{k^u_{oc} k_{lu} + k^l_{oc} k_{ul}}{k_{ul} + k_{lu}} \right) \end{aligned}$$

and thus, by following the steps above, we find that

$$p_n = (1 - \Delta t K)^n,$$

where

$$K = \frac{k_{oc}^u k_{lu} + k_{oc}^l k_{ul}}{k_{ul} + k_{lu}}.$$

The probability of closing a channel that is in one of the open states during a time step is given by

$$\Delta t k_{oc}^u q_u + \Delta t k_{oc}^l q_l = \Delta t K$$

and, therefore, the probability of closing a channel in a time step that has remained open for n time steps is given by

$$\Delta t K \left(1 - \Delta t K\right)^n.$$

We find that the expected mean open time is given by

$$\tau_o = \frac{1}{K} = \frac{k_{ul} + k_{lu}}{k_{oc}^u k_{lu} + k_{oc}^l k_{ul}}. \tag{13.7}$$

13.1.1.1 Special Cases

It is interesting to consider the formula for the mean open time given by (13.7) in two special cases. First, we assume that $k_{oc}^u = k_{oc}^l$ and we let k_{oc} denote this common value. Then, by (13.7), we have

$$\tau_o = \frac{1}{k_{oc}}$$

which is the same as we found for the two-state scheme above. Next consider the case of $k_{ul} = k_{lu}$ (and $k_{oc}^u \neq k_{oc}^l$). By (13.7), we find

$$\tau_o = \frac{1}{(k_{oc}^u + k_{oc}^u)/2}. \tag{13.8}$$

13.2 Numerical Experiments

It is useful to have a look at the mean open time computed in specific numerical experiments to determine how well it is represented by the theoretical value derived above. Similarly, it is useful to consider how well the theoretical equilibrium open probability represents the data we observe in actual computations. In this section, we will present experiments that hopefully clarify these matters.

13.2.1 Mean Open Time and Equilibrium Open Probability: Theoretical Values Versus Sample Mean Values

Let us illustrate the result above by a few numerical experiments. We start by considering the Markov model

$$C \underset{k_{co}}{\overset{k_{oc}}{\leftrightarrows}} O,$$

where we set $k_{co} = 1\ \text{ms}^{-1}$ and we let

$$k_{oc} = \frac{1}{m}\text{ms}^{-1}$$

for $m = 1, \ldots, 100$. For every value of k_{oc}, we run a simulation using the Markov model for $T = 10^4$ ms. The time instances when the channel changes state are stored in the sequence $\{t_i\}_{i=0}^{N}$ and the mean open time observed in the simulation is given by[1]

$$\tau_{o,s} = \frac{2}{N}\sum_i (t_i - t_{i-1})_o\,,$$

where

$$(t_i - t_{i-1})_o = \begin{cases} t_i - t_{i-1} & \text{if the channel is open in this interval,} \\ 0 & \text{if the channel is closed in this interval.} \end{cases}$$

With this notation we can also define the sample open probability by

$$o_s = \frac{1}{T}\sum_i (t_i - t_{i-1})_o\,.$$

In Fig. 13.2 (left panel), we plot the sample mean open time $\tau_{o,s}$ and the theoretical mean open time given by

$$\tau_o = \frac{1}{k_{oc}} \tag{13.9}$$

[1]The index s here is used to indicate *sample*, since these are values for a specific computation and not the theoretical value computed above.

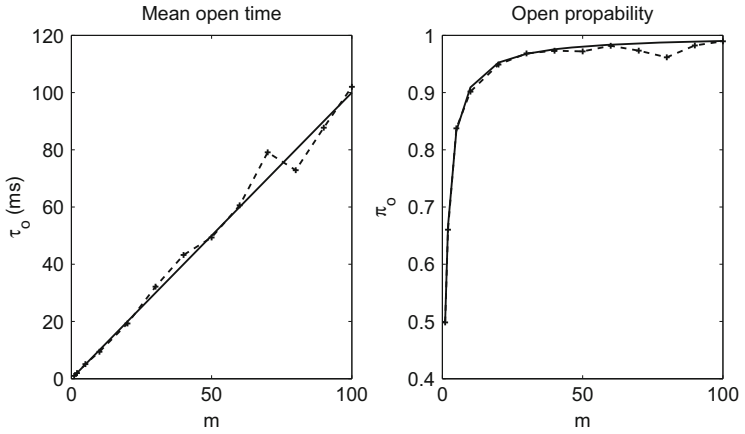

Fig. 13.2 Mean open time (*left*) and open probability (*right*), with $k_{oc} = 1/m$ ms^{-1} and $k_{co} = 1$ ms^{-1}. The sample values (*dashed lines*) correspond well with the theoretical values (*solid line*)

as functions of k_{oc}. We also plot (right panel) the sample open probability o_s and the theoretical equilibrium probability given by

$$o = \frac{1}{1 + \frac{k_{oc}}{k_{co}}}. \tag{13.10}$$

In both plots, we see that the mean values computed in the simulations are quite close to the theoretical values. If we increase the simulation time T, these graphs converge toward the same value.

13.2.2 The Closed to Open Rate k_{co} Does Not Affect the Mean Open Time

We have seen that, theoretically, according to (13.9), the mean open time τ_o is independent of the closed to open rate k_{co}, but the open probability is affected as stated in (13.10). This is illustrated in Fig. 13.3, where we use $k_{oc} = 1$ ms^{-1} and $k_{co} = 1/m$ ms^{-1} and plot the mean open time (left panel) and the open probability (right panel) as functions of m.

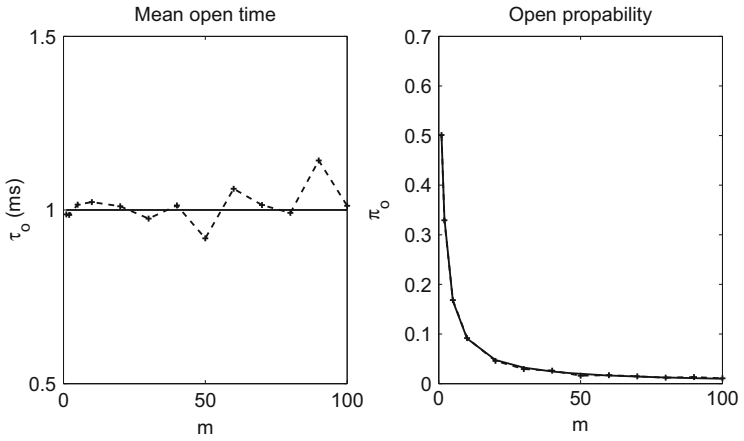

Fig. 13.3 Mean open time (*left*) and open probability (*right*) with $k_{co} = 1/m$ ms^{-1} and $k_{oc} = 1$ ms^{-1}. The mean open time is not affected by changes in k_{co}. The sample values correspond well to the theoretical values

13.2.3 The Mean Open Time in the Presence of Two Open States

In Fig. 13.4, we show the sample mean open time and the theoretical mean open time given by

$$\tau_o = \frac{1}{K} = \frac{k_{ul} + k_{lu}}{k^u_{oc} k_{lu} + k^l_{oc} k_{ul}} \tag{13.11}$$

for the Markov model in Fig. 13.1. In the computations, we have used $k^l_{oc} = 1$ ms^{-1}, $k^u_{oc} = 10$ ms^{-1}, and $k_{lu} = 0.001$ ms^{-1} and k_{ul} varies. The other parameters of the model do not affect the result, as long as detailed balance holds.

13.2.4 Changing the Mean Open Time Affects the Dynamics of the Transmembrane Potential

We consider the stochastic model of the transmembrane potential given by

$$v_t = g_K(V_K - v) + \gamma g_{Na}(V_{Na} - v), \tag{13.12}$$

where γ is a stochastic variable governed by the two-state Markov model

$$C \underset{k_{co}}{\overset{k_{oc}}{\leftrightarrows}} O.$$

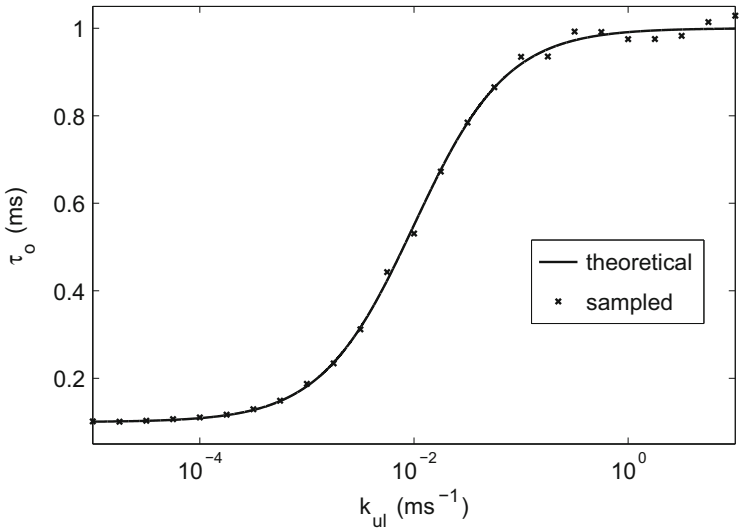

Fig. 13.4 Mean open time for a Markov model with two open states

We use the parameters

$$g_K = \frac{1}{10}\ \text{ms}^{-1},\ g_{Na} = 1\ \text{ms}^{-1}, \tag{13.13}$$
$$V_K = -85\ \text{mV},\ V_{Na} = 45\ \text{mV},$$

and compute solutions using the standard scheme

$$v_{n+1} = v_n - \Delta t\,(g_K\,(v_n - V_K) + \gamma_n g_{Na}(v_n - V_{Na})), \tag{13.14}$$

where the time step is assumed to satisfy the condition

$$\Delta t < \frac{1}{g_K + g_{Na}}. \tag{13.15}$$

Under this condition, we have seen above that, for solutions computed by (13.12), an invariant region is given by

$$\Omega = (V_K, V_+)\,, \tag{13.16}$$

where

$$V_+ = \frac{g_K V_K + g_{Na} V_{Na}}{g_K + g_{Na}}.$$

In Fig. 13.5, we show numerical solutions of (13.12) for

$$k_{oc} = k_{co} = 0.1\ \text{ms}^{-1}, 1\ \text{ms}^{-1}, 10\ \text{ms}^{-1}, 100\ \text{ms}^{-1}.$$

According to the considerations above, the equilibrium open probability is given by

$$o = \frac{1}{1 + \frac{k_{oc}}{k_{co}}},$$

which is constant for the four parameter sets used in Fig. 13.5. The mean open time, however, varies with k_{oc} as

$$\tau_o = \frac{1}{k_{oc}}.$$

For the cases studied in Fig. 13.5, the mean open times are 10, 1, 1/10, and 1/100 ms and we observe that the reduced mean open time greatly reduces the variations of the transmembrane potential.

13.3 Changing the Mean Open Time Affects the Probability Density Functions

The stationary version of the probability density system governing the states of the Markov model

$$C \underset{k_{co}}{\overset{k_{oc}}{\leftrightarrows}} O$$

is given by

$$\begin{aligned} \frac{\partial}{\partial v}(a_o \rho_o) &= k_{co}\rho_c - k_{oc}\rho_o, \\ \frac{\partial}{\partial v}(a_c \rho_c) &= k_{oc}\rho_o - k_{co}\rho_c, \end{aligned} \tag{13.17}$$

where

$$\begin{aligned} a_o &= g_K(V_K - v) + g_{Na}(V_{Na} - v), \\ a_c &= g_K(V_K - v). \end{aligned} \tag{13.18}$$

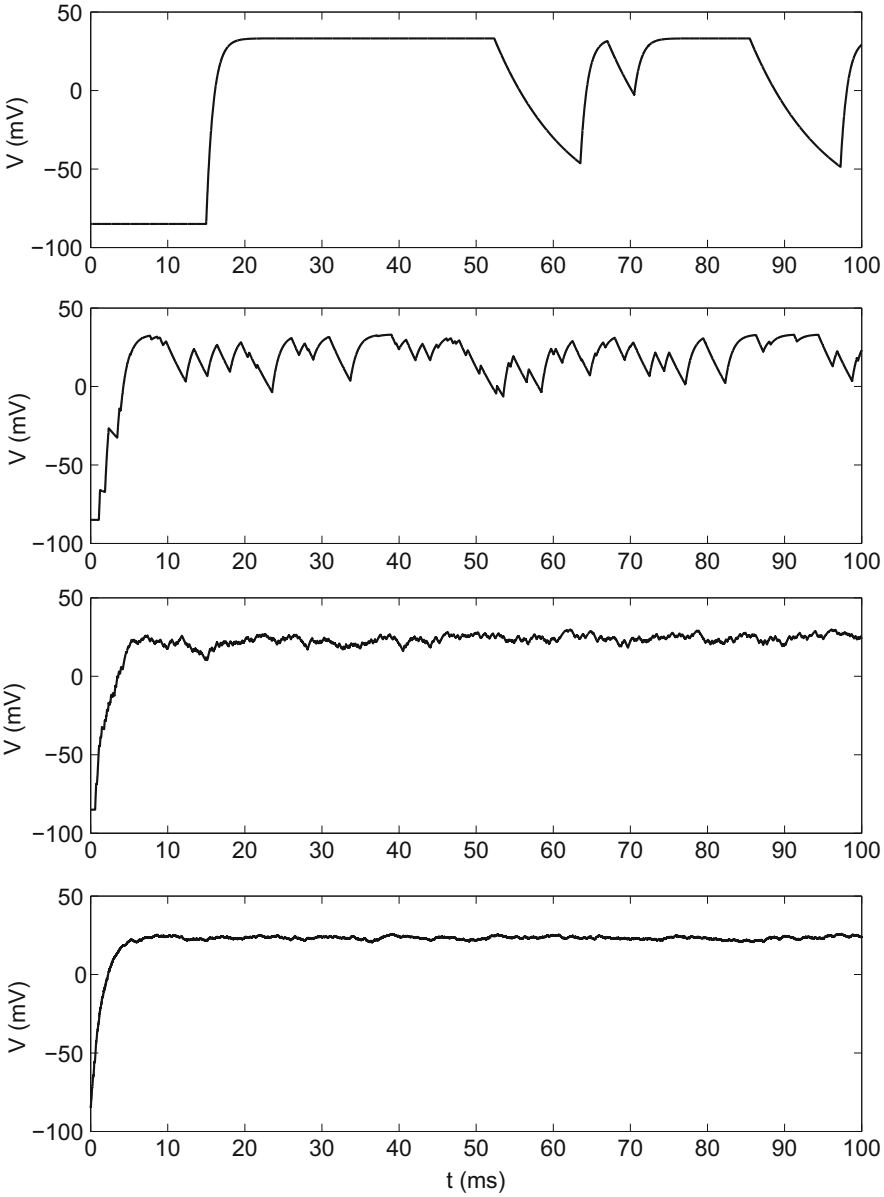

Fig. 13.5 Simulations based on the numerical scheme (13.14) with changing reaction rates for the Markov model. From top to bottom, $k_{oc} = k_{co} = 0.1$, 1, 10, and 100 ms^{-1}. Since $k_{oc} = k_{co}$ for all values, the open probability is kept constant but the mean open time given by $1/k_{oc}$ is decreasing from top to bottom

The analytical solution of this problem is given by

$$\rho_o(v) = Kg_K(V_+ - v)^{\frac{k_{oc}}{g}-1}(v - V_K)^{\frac{k_{co}}{g_K}},$$
$$\rho_c(v) = Kg(V_+ - v)^{\frac{k_{oc}}{g}}(v - V_K)^{\frac{k_{co}}{g_K}-1},$$

where

$$g = g_{Na} + g_K, V_+ = \frac{g_{Na}V_{Na} + g_K V_K}{g_{Na} + g_K}$$

and K is chosen such that

$$\int_{V_K}^{V_+} \rho_o + \rho_c = 1,$$

which is given by

$$1/K = \frac{k_{co} + k_{oc}}{a + b}(V_+ - V_K)^{(a+b)}B(a, b),$$

with $a = k_{co}/g_K, b = k_{oc}/g$, and $B(a, b) = \Gamma(a)\Gamma(b)/\Gamma(a + b)$.

In Fig. 13.6, we show the open probability density function for the data given in (13.13) with

$$k_{oc} = k_{co} = 0.1\ \text{ms}^{-1}, 1\ \text{ms}^{-1}, 10\ \text{ms}^{-1}, 100\ \text{ms}^{-1}.$$

Again, we recall that as k_{oc} increases, the mean open time decreases and we observe in the figure that the probability density function becomes narrower.

13.4 Theoretical Drugs for OC-Mutations

We have seen earlier that when mutations increase the open probability by increasing the reaction rate from C to O (k_{co}), the effect of the mutation can be completely repaired by using an optimal closed state blocker. Now we are interested in a mutation that increases the open probability by reducing the reaction rate from O to C (k_{oc}) . Such a mutation increases both the open probability and the mean open time and we will observe that a closed state blocker is unable to repair the effect of such a mutation.

We consider the two-state Markov model

$$C \underset{k_{co}}{\overset{k_{oc}/\mu}{\leftrightarrows}} O, \tag{13.19}$$

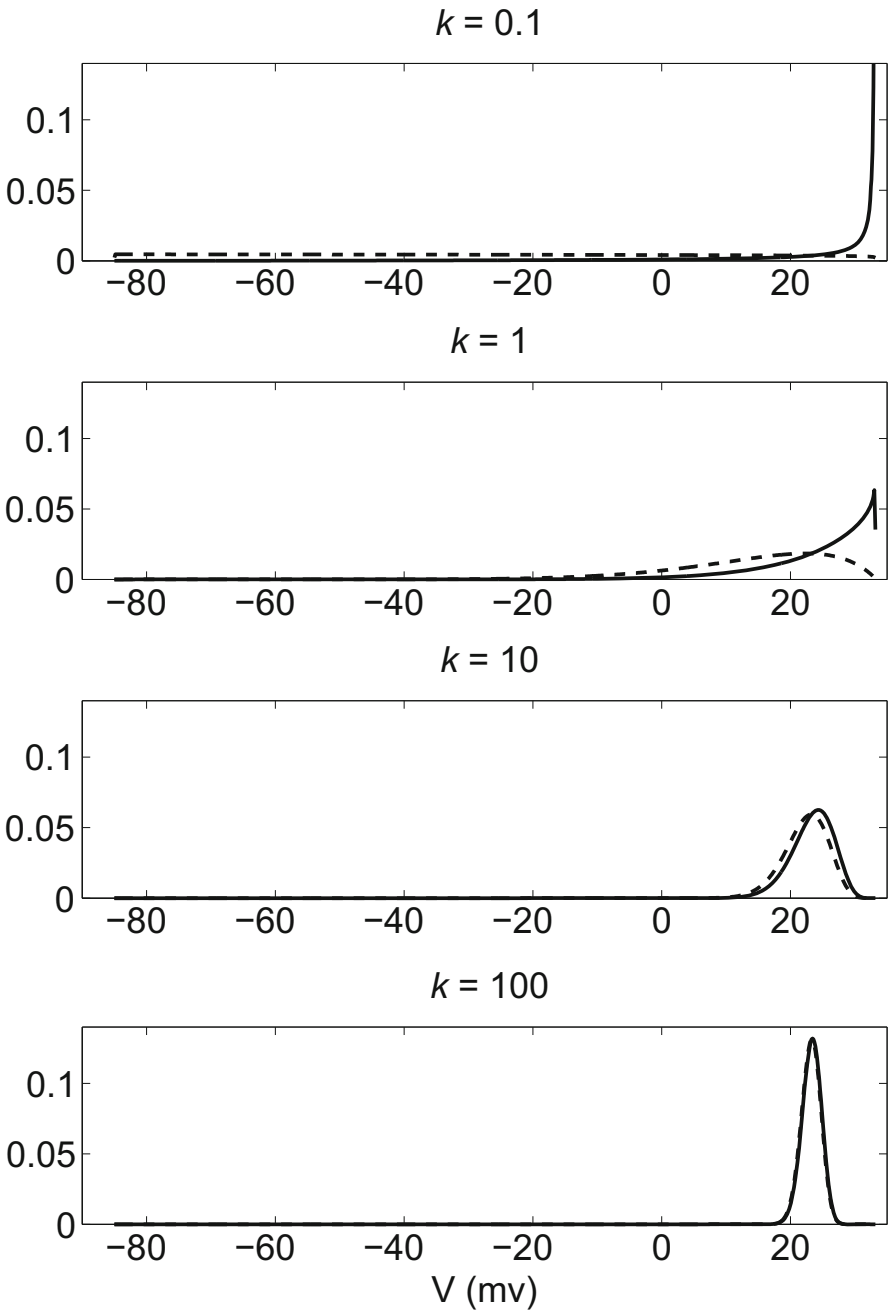

Fig. 13.6 The open probability density function ρ_o (*solid line*) and closed probability density function ρ_c depend on the mean open time given by $1/k_{oc}$. In the figures, we have used $k = k_{oc} = k_{co}$

where $\mu \geqslant 1$ is the mutation severity index; as usual, $\mu = 1$ denotes the wild type. Recall that the equilibrium open probability is given by

$$o = \frac{1}{1 + \frac{k_{oc}}{\mu k_{co}}}$$

and the mean open time is given by

$$\tau_o = \frac{\mu}{k_{oc}},$$

so the mutation clearly increases both the open probability and the mean open time.

13.4.1 The Theoretical Closed State Blocker Does Not Work for the OC-Mutation

Let us start by considering a closed state blocker of the form

$$B \underset{k_{bc}}{\overset{k_{cb}}{\leftrightarrows}} C \underset{k_{co}}{\overset{k_{oc}/\mu}{\leftrightarrows}} O. \tag{13.20}$$

We find that the equilibrium open probability of the mutant in the presence of the closed state blocker is given by

$$o = \frac{1}{1 + \frac{k_{oc}}{k_{co}} \frac{1+\delta_c}{\mu}},$$

where

$$\delta_c = \frac{k_{cb}}{k_{bc}}.$$

Since the wild type equilibrium open probability is given by

$$o = \frac{1}{1 + \frac{k_{oc}}{k_{co}}},$$

the drug will repair the open probability, provided that

$$\frac{1 + \delta_c}{\mu} = 1$$

and therefore the drug must satisfy the usual condition

$$\delta_c = \mu - 1.$$

A drug satisfying this condition will completely repair the equilibrium open probability and that is, of course, good, but it is not enough. Since the mutation represented by (13.19) also affects the mean open time, a drug of the form (13.20) cannot repair that effect of the mutation. To see this, we consider the probability density system defined by

$$\begin{aligned}
\frac{\partial}{\partial v}\left(a_o\rho_o\right) &= k_{co}\rho_c - \frac{1}{\mu}k_{oc}\rho_o,\\
\frac{\partial}{\partial v}\left(a_c\rho_c\right) &= \frac{1}{\mu}k_{oc}\rho_o - \left(k_{co} + (\mu-1)\,k_{bc}\right)\rho_c + k_{bc}\rho_b,\\
\frac{\partial}{\partial v}\left(a_c\rho_b\right) &= (\mu-1)\,k_{bc}\rho_c - k_{bc}\rho_b,
\end{aligned} \tag{13.21}$$

where, as usual, ρ_o, ρ_c, and ρ_b denote the probability density functions of the open (O), closed (C), and blocked (B) states, respectively, and where the fluxes are defined by (13.18). In Fig. 13.7, we compare the open probability density computed by solving the system (13.21) with the open probability density of the wild type. The

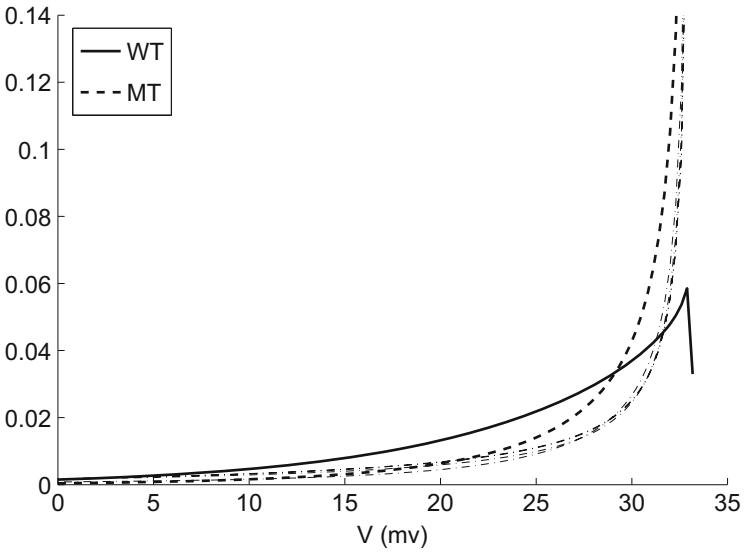

Fig. 13.7 The *solid line* represents the wild type solution and the *dashed line* represents the mutant. Various closed state drugs are applied, but none are able to repair the effect of the mutation

wild type probability density functions are given by

$$\begin{aligned} \frac{\partial}{\partial v}(a_o\rho_o) &= k_{co}\rho_c - k_{oc}\rho_o, \\ \frac{\partial}{\partial v}(a_c\rho_c) &= k_{oc}\rho_o - k_{co}\rho_c, \end{aligned} \tag{13.22}$$

and the probability density functions of the mutant case are given by

$$\begin{aligned} \frac{\partial}{\partial v}(a_o\rho_o) &= k_{co}\rho_c - \frac{1}{\mu}k_{oc}\rho_o, \\ \frac{\partial}{\partial v}(a_c\rho_c) &= \frac{1}{\mu}k_{oc}\rho_o - k_{co}\rho_c. \end{aligned} \tag{13.23}$$

In the computations we have used the parameters given by (13.13) and the rates

$$k_{co} = 1 \text{ ms}^{-1} \text{ and } k_{oc} = 1 \text{ ms}^{-1}.$$

We use three values of the rates k_{bc} and we observe that no parameter is able to repair the open state probability density function of the mutation. In Fig. 13.8, we show the norm of the difference between the open probability density defined by (13.21) and (13.22. The norm is defined by (2.40) on page 46 and we see that no version of the closed state blocker defined by (13.20) is able to repair the effect of the mutations given by (13.19).

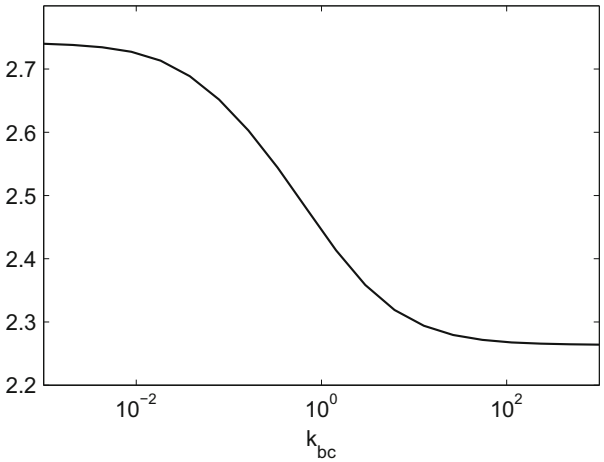

Fig. 13.8 The norm of the difference between the wild type solution and the mutant after the drug is applied. The norm is defined by (2.40) on page 46. We see that no value of the drug parameter k_{bc} for the closed state blocker is able to repair the effect of the mutation

13.4.2 The Theoretical Open State Blocker Repairs the Effect of the OC-Mutation

Next, we consider an open state blocker for the mutation leading to both an increased open probability and an increased mean open time. The theoretical open state blocker can be written in the form

$$C \underset{k_{co}}{\overset{k_{oc}/\mu}{\leftrightarrows}} O \underset{k_{ob}}{\overset{k_{bo}}{\leftrightarrows}} B, \tag{13.24}$$

where the parameters k_{bo} and k_{ob} define the theoretical drug. For this Markov model, the equilibrium open probability is given by

$$o_\mu = \frac{1}{1 + \frac{k_{oc}}{\mu k_{co}} + \frac{k_{ob}}{k_{bo}}}$$

and the mean open time is given by

$$\tau_{o,\mu} = \frac{1}{\frac{1}{\mu}k_{oc} + k_{ob}}.$$

Since the associated wild type values are

$$o = \frac{1}{1 + \frac{k_{oc}}{k_{co}}}$$

and

$$\tau_o = \frac{1}{k_{oc}},$$

we want to define the drug such that

$$1 + \frac{k_{oc}}{\mu k_{co}} + \frac{k_{ob}}{k_{bo}} = 1 + \frac{k_{oc}}{k_{co}}$$

and

$$\frac{1}{\mu}k_{oc} + k_{ob} = k_{oc}.$$

To satisfy these two requirements, we find that the drug must be given by

$$\begin{aligned} k_{ob} &= \frac{\mu - 1}{\mu} k_{oc}, \\ k_{bo} &= k_{co}. \end{aligned} \tag{13.25}$$

13.4.3 The Theoretical Open State Blocker Is Optimal

We will show analytically that the open state blocker defined by (13.24) where the parameters are given by (13.25) is an optimal drug, in the sense that the effect of the mutation is completely repaired. We start by observing that the probability density system associated with the Markov model (13.24) is given by

$$\begin{aligned} \frac{\partial}{\partial v}(a_o \rho_o) &= k_{co}\rho_c - (\mu^{-1}k_{oc} + k_{ob})\rho_o + k_{bo}\rho_b, \\ \frac{\partial}{\partial v}(a_c \rho_c) &= \mu^{-1}k_{oc}\rho_o - k_{co}\rho_c, \\ \frac{\partial}{\partial v}(a_c \rho_b) &= k_{ob}\rho_o - k_{bo}\rho_b. \end{aligned} \tag{13.26}$$

If we insert the drug given by (13.25), we obtain the system

$$\begin{aligned} \frac{\partial}{\partial v}(a_o \rho_o) &= k_{co}\rho_c - k_{oc}\rho_o + k_{co}\rho_b, \\ \frac{\partial}{\partial v}(a_c \rho_c) &= \mu^{-1}k_{oc}\rho_o - k_{co}\rho_c, \\ \frac{\partial}{\partial v}(a_c \rho_b) &= \left(1 - \mu^{-1}\right) k_{oc}\rho_o - k_{co}\rho_b. \end{aligned} \tag{13.27}$$

We define

$$\bar{\rho}_c = \rho_c + \rho_b$$

and add the two latter equations of this system to find that ρ_o and $\bar{\rho}_c$ solve the system

$$\begin{aligned} \frac{\partial}{\partial v}(a_o \rho_o) &= k_{co}\bar{\rho}_c - k_{oc}\rho_o, \\ \frac{\partial}{\partial v}(a_c \bar{\rho}_c) &= k_{oc}\rho_o - k_{co}\bar{\rho}_c, \end{aligned} \tag{13.28}$$

which coincides with the system defining the wild type probability density functions (see (13.22) above). We therefore conclude that the open state blocker defined by

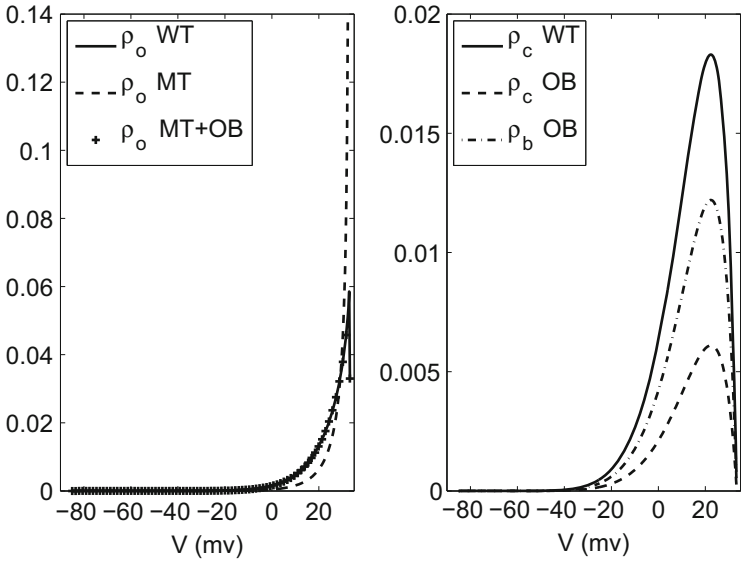

Fig. 13.9 Probability density functions of the wild type, mutant, and mutant in the presence of the open blocker. The open blocker completely repairs the open probability density function of the mutant

the parameters (13.25) completely repairs the probability density functions of the mutant for any value of the mutation severity index.

13.4.3.1 The Probability Density Function of the Blocked State Is Proportional to the Probability Density Function of the Wild Type Closed State

In Fig. 13.9, we show the open probability density functions of the wild type (defined by system (13.22), the mutant (defined by system (13.23) with $\mu = 3$), and the mutant including the optimal drug (defined by system (13.27)). As expected, the open probability is completely repaired by the theoretical drug.

In the right panel of the figure, we show the graph of ρ_c for the wild type (solid line) and for the mutant case in the presence of the open blocker. We show both ρ_c and ρ_b. We note that these graphs seem to have the same shape and we will show that they indeed differ only by a constant.

We start by making the ansatz that for the solution of system (13.27) we have

$$\rho_b = (\mu - 1)\, \rho_c. \tag{13.29}$$

If we insert this into system (13.27), we find that the two latter equations become identical and the system is therefore reduced to the following 2×2 system:

$$\begin{aligned}\frac{\partial}{\partial v}(a_o\rho_o) &= \mu k_{co}\rho_c - k_{oc}\rho_o,\\ \frac{\partial}{\partial v}(a_c\rho_c) &= \mu^{-1}k_{oc}\rho_o - k_{co}\rho_c.\end{aligned} \tag{13.30}$$

Therefore, we can define

$$\rho_c^* = \mu\rho_c$$

and find that ρ_o and ρ_c^* solve system

$$\begin{aligned}\frac{\partial}{\partial v}(a_o\rho_o) &= k_{co}\rho_c^* - k_{oc}\rho_o,\\ \frac{\partial}{\partial v}\left(a_c\rho_c^*\right) &= k_{oc}\rho_o - k_{co}\rho_c^*,\end{aligned} \tag{13.31}$$

which is exactly the wild type system. We therefore conclude that

$$\rho_b = (\mu - 1)\,\rho_c = \frac{\mu - 1}{\mu}\rho_c^*, \tag{13.32}$$

where (ρ_o, ρ_c, ρ_b) solves the system (13.27) and where (ρ_o^*, ρ_c^*) solves the wild type system

$$\begin{aligned}\frac{\partial}{\partial v}\left(a_o\rho_o^*\right) &= k_{co}\rho_c^* - k_{oc}\rho_o^*,\\ \frac{\partial}{\partial v}\left(a_c\rho_c^*\right) &= k_{oc}\rho_o^* - k_{co}\rho_c^*.\end{aligned}$$

13.4.4 Stochastic Simulations Using the Optimal Open State Blocker

In Fig. 13.10, we show the results of numerical simulations using scheme (13.14). We show the result for the wild type model (upper panel), the mutant model (middle panel), and the model of the mutant where the drug defined by (13.25) is used (lower panel).

The graphs show that the effect of the mutation is repaired using the drug (13.25); the solutions are not identical and this is reasonable, since a random number generator is involved in updating the state of the Markov model and therefore two computed solutions will not be identical (not even two wild type solutions).

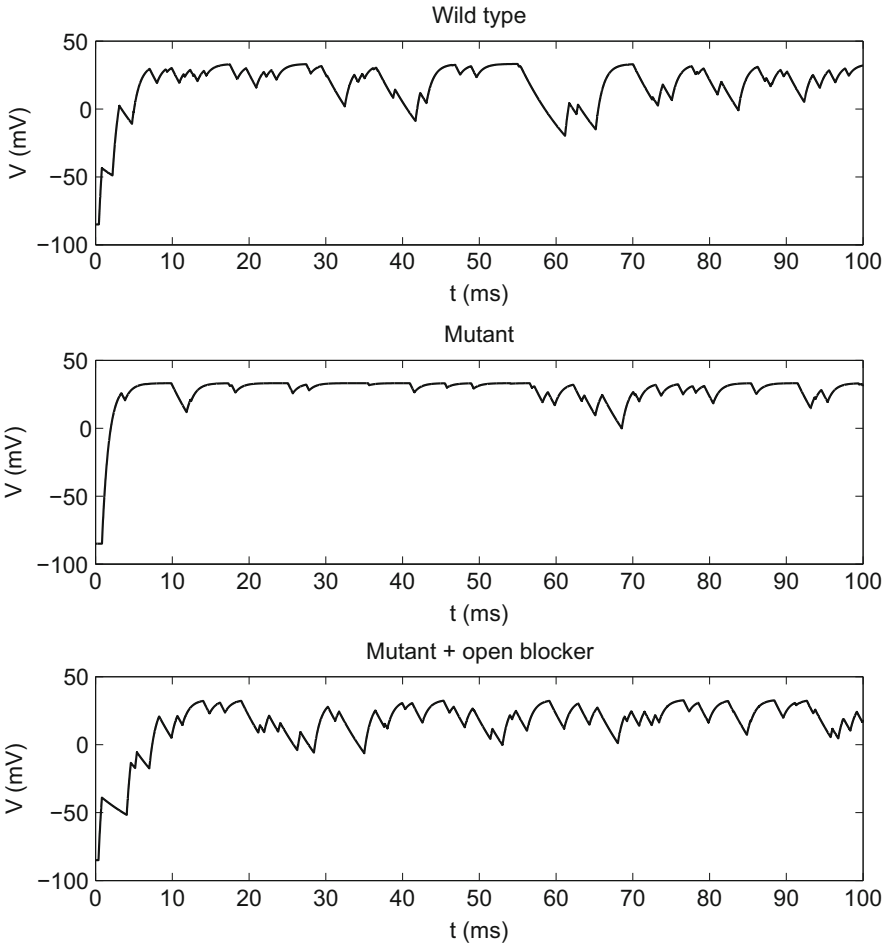

Fig. 13.10 Numerical simulations using scheme (13.14) for wild type data (*upper panel*), mutant data (*center panel*), and mutant data where the drug defined by (13.25) is used (*lower panel*). Observe the long open periods in the middle panel and that these are repaired by the drug (*lower panel*)

However, we note that the qualitative properties of the upper and lower solutions are similar, whereas the mutant case is different due to the increased open probability and prolonged mean open time.

13.5 Inactivated States and Mean Open Time

In Chap. 11, we studied a Markov model including the open state (O), closed state (C), and inactivated state (I). The prototypical Markov model is repeated in Fig. 13.11. As usual, we assumed that the principle of detailed balance holds and therefore the parameters of the Markov model satisfy the equation

$$k_{io}k_{oc}k_{ci} = k_{oi}k_{co}k_{ic}. \tag{13.33}$$

We also introduced a mutation that increased the rates k_{io} and k_{ic} and thus reduced the probability of being in the inactivated state. From what we have just seen, we readily observe that such a mutation does not influence the mean open time; however, if data show that the mean open time is affected, the effect of the mutation must be modeled differently. Another way to model the reduced equilibrium probability of being in the inactivated state is to reduce the rates toward the inactivated state. Such a mutation takes the form

$$\begin{aligned} \bar{k}_{ci} &= k_{ci}/\mu, \\ \bar{k}_{oi} &= k_{oi}/\mu, \end{aligned} \tag{13.34}$$

where $\mu \geqslant 1$ and, as usual, $\mu = 1$ represents the wild type. It follows from (13.33) that the principle of detailed balance also holds for the mutant model:

$$k_{io}k_{oc}\frac{k_{ci}}{\mu} = \frac{k_{oi}}{\mu}k_{co}k_{ic}. \tag{13.35}$$

If we repeat the argument above, we find that the mean open time of the model presented in Fig. 13.11 is given by

$$\tau_o = \frac{1}{k_{oc} + k_{oi}}$$

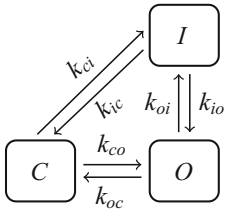

Fig. 13.11 Three-state Markov model. In the mutant case, we replace the rates k_{ci} and k_{oi} by k_{ci}/μ and k_{oi}/μ, respectively, where μ denotes the mutation severity index

for wild type data and

$$\tau_{o,\mu} = \frac{1}{k_{oc} + k_{oi}/\mu}$$

for the mutant case. We note that the mean open time increases as the mutation severity index μ increases. Following the usual steps, we find that the equilibrium probabilities are given by

$$o = \frac{1}{1 + \frac{k_{oc}}{k_{co}} + \frac{k_{oi}}{\mu k_{io}}},$$

$$c = \frac{\frac{k_{oc}}{k_{co}}}{1 + \frac{k_{oc}}{k_{co}} + \frac{k_{oi}}{\mu k_{io}}},$$

$$i = \frac{\frac{k_{oi}}{k_{io}}}{\mu\left(1 + \frac{k_{oc}}{k_{co}}\right) + \frac{k_{oi}}{k_{io}}}.$$

We observe that the equilibrium probability of being in the open and closed states increases as a consequence of the mutation and the equilibrium probability of being in the inactivated state is reduced under the mutation.

13.5.1 A Theoretical Open State Blocker

We observed above that to repair the effect of changes in the mean open time, it is necessary to use an open state blocker. The reason for this is that neither a closed blocker nor an inactivated blocker has any effect on the mean open time and, therefore, it is inconceivable that such blockers can repair the effect of a mutation on the mean open time. An open state blocker directly affects the mean open time and the drug must be tuned to repair the effect of the mutation.

A Markov model that includes an open state blocker is shown in Fig. 13.12. We have already computed formulas for the equilibrium probabilities of a Markov model of this form (see page 170). The inverse ($p = 1/o$) open probability in equilibrium is given by

$$p_\mu = 1 + \frac{k_{oc}}{k_{co}} + \frac{1}{\mu}\frac{k_{oi}}{k_{io}}$$

and thus the wild type inverse open probability is given by

$$p = 1 + \frac{k_{oc}}{k_{co}} + \frac{k_{oi}}{k_{io}}.$$

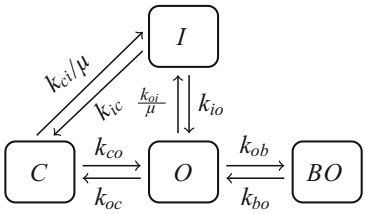

Fig. 13.12 The model represented in Fig. 13.11 is extended to account for the blocker (BO) associated with the open state

Similarly, the inverse open probability in the presence of the open state blocker is given by

$$p_{b,\mu} = 1 + \frac{k_{oc}}{k_{co}} + \frac{1}{\mu}\frac{k_{oi}}{k_{io}} + \frac{k_{ob}}{k_{bo}}.$$

Furthermore, the mean open time of wild type is given by

$$\tau_o = \frac{1}{k_{oi} + k_{oc}}$$

and, when the theoretical drug is included in the mutant case, the mean open time is given by

$$\tau_{o,b,\mu} = \frac{1}{\frac{1}{\mu}k_{oi} + k_{oc} + k_{ob}}.$$

We are now looking for a drug that will repair the equilibrium probability and the mean open time. More precisely, we want to find the parameters k_{bo} and k_{ob} such that $p_{b,\mu} = p$ and $\tau_{o,b,\mu} = \tau_o$. More explicitly, we require that

$$1 + \frac{k_{oc}}{k_{co}} + \frac{1}{\mu}\frac{k_{oi}}{k_{io}} + \frac{k_{ob}}{k_{bo}} = 1 + \frac{k_{oc}}{k_{co}} + \frac{k_{oi}}{k_{io}}$$

and

$$\frac{1}{\mu}k_{oi} + k_{oc} + k_{ob} = k_{oi} + k_{oc}.$$

This is a 2×2 system of equations in the unknowns k_{ob} and k_{bo} and the solution is given by

$$k_{ob} = \left(1 - \mu^{-1}\right) k_{oi} \text{ and } k_{bo} = k_{io}. \tag{13.36}$$

We will see in numerical experiments below that the open state blocker illustrated in Fig. 13.12 where the parameters of the drug are given by (13.36) repairs the effect of the mutation.

13.5.2 Probability Density Functions Using the Open State Blocker

We have found a theoretical drug (see (13.36)) for the mutation affecting the rates from O to I and from C to I and we want to assess the drug's usefulness by considering the open probability density functions. For the wild type case, the probability density functions of the states present in the Markov model of Fig. 13.11 are governed by the system

$$\begin{aligned}
\frac{\partial}{\partial v}\left(a_o\rho_o\right) &= k_{co}\rho_c - \left(k_{oc}+k_{oi}\right)\rho_o + k_{io}\rho_i,\\
\frac{\partial}{\partial v}\left(a_c\rho_c\right) &= k_{oc}\rho_o - \left(k_{co}+k_{ci}\right)\rho_c + k_{ic}\rho_i,\\
\frac{\partial}{\partial v}\left(a_c\rho_i\right) &= k_{oi}\rho_o - \left(k_{io}+k_{ic}\right)\rho_i + k_{ci}\rho_c.
\end{aligned} \tag{13.37}$$

In the mutant case, when the open state blocker is added as indicated in Fig. 13.12, the probability density system is

$$\begin{aligned}
\frac{\partial}{\partial v}\left(a_o\rho_o\right) &= k_{co}\rho_c - \left(k_{oc}+\frac{1}{\mu}k_{oi}+k_{ob}\right)\rho_o + k_{io}\rho_i + k_{bo}\rho_b,\\
\frac{\partial}{\partial v}\left(a_c\rho_c\right) &= k_{oc}\rho_o - \left(k_{co}+\frac{1}{\mu}k_{ci}\right)\rho_c + k_{ic}\rho_i,\\
\frac{\partial}{\partial v}\left(a_c\rho_i\right) &= \frac{1}{\mu}k_{oi}\rho_o - \left(k_{io}+k_{ic}\right)\rho_i + \frac{1}{\mu}k_{ci}\rho_c,\\
\frac{\partial}{\partial v}\left(a_c\rho_b\right) &= k_{ob}\rho_o - k_{bo}\rho_b.
\end{aligned} \tag{13.38}$$

As usual, ρ_o, ρ_c, ρ_i, and ρ_b denote the probability density functions of the open, closed, inactivated, and blocked states, respectively, and the functions of the flux are given by (13.18). By introducing the drug given by (13.36), we obtain the system

$$\begin{aligned}
\frac{\partial}{\partial v}\left(a_o\rho_o\right) &= k_{co}\rho_c - \left(k_{oc}+k_{oi}\right)\rho_o + k_{io}\rho_i + k_{io}\rho_b,\\
\frac{\partial}{\partial v}\left(a_c\rho_c\right) &= k_{oc}\rho_o - \left(k_{co}+\frac{1}{\mu}k_{ci}\right)\rho_c + k_{ic}\rho_i,\\
\frac{\partial}{\partial v}\left(a_c\rho_i\right) &= \frac{1}{\mu}k_{oi}\rho_o - \left(k_{io}+k_{ic}\right)\rho_i + \frac{1}{\mu}k_{ci}\rho_c,\\
\frac{\partial}{\partial v}\left(a_c\rho_b\right) &= \left(1-\mu^{-1}\right)k_{oi}\rho_o - k_{io}\rho_b.
\end{aligned} \tag{13.39}$$

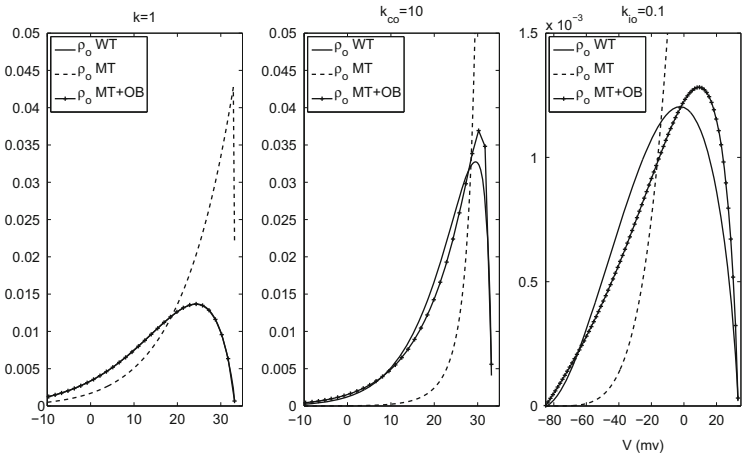

Fig. 13.13 *Left panel*: All rates equal one. The theoretical drug restores ρ_o. *Middle panel*: As in the left panel, except $k_{co} = 10$ ms^{-1}. *Right panel*: As in the left panel, except $k_{io} = 0.1$ ms^{-1}. For all three cases, $\mu = 10$

In Fig. 13.13, we show solutions of the wild type system (13.37), the mutant system, and the mutant system where the drug is added (13.39). Note that the mutant system is equal to the wild type system, except for the change of the rates k_{ci} and k_{oi} given by

$$\bar{k}_{ci} = k_{ci}/\mu, \qquad (13.40)$$
$$\bar{k}_{oi} = k_{oi}/\mu.$$

In Fig. 13.13, we compare the open probability density functions of the three models for three different sets of parameters. In the left panel of Fig. 13.13, we show the open probability of the wild type (solid line), the mutant ($\mu = 10$), and the mutant in the presence of the theoretical open blocker. We see that the effect of the mutation is completely repaired by the drug. Other cases are shown in the center and right panels. The effect of the drug is still good but the effect of the mutation is not completely repaired. These observations are confirmed in Table 13.1. Furthermore, we have tested a large variety of parameters and the results we show here (center and right panels) represent the most difficult cases we could find in experiments. Therefore, we conclude that the theoretical open state blocker illustrated in Fig. 13.12 works very well.

Table 13.1 Statistical properties of ρ_o for the cases shown in Fig. 13.13

	$k = 1$		$k_{co} = 10$		$k_{io} = 0.1$	
	π_o	E_o	π_o	E_o	π_o	E_o
WT	0.333	16.366	0.476	22.995	0.083	−12.867
MT	0.476	23.272	0.833	31.074	0.333	17.702
MT+OB	0.333	16.366	0.476	23.169	0.083	−9.225

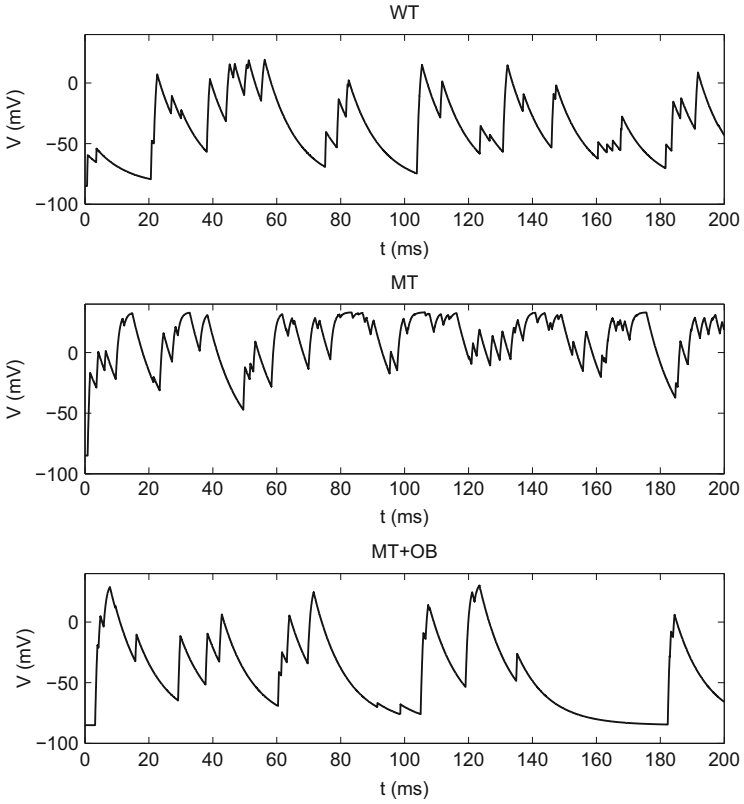

Fig. 13.14 Monte Carlo runs of the case shown in the right panel of Fig. 13.13

13.5.3 Stochastic Simulations Using the Open State Blocker

In Fig. 13.14, we show simulations using the numerical scheme

$$v_{n+1} = v_n - \Delta t \left(g_K \left(v_n - V_K \right) + \gamma_n g_{Na} (v_n - V_{Na}) \right), \tag{13.41}$$

where the value of the variable γ_n is determined by the Markov model given in Fig. 13.11. For the wild type case, the rates k_{ci} and k_{oi} are used and, in the mutant

case, the rates k_{ci}/μ and k_{oi}/μ are used. Furthermore, when the drug is applied in the mutant case, the Markov model is as illustrated in Fig. 13.12, where the rates of the drug are given by (13.36). We observe that, in the mutant case, the channel does not inactivate and therefore more action potentials are generated. When the drug is applied, this effect seems to be removed and the channel again acts more or less as in the wild type case. However, as mentioned above it is not straightforward to compare solutions based on the stochastic model and therefore we emphasis the use of probability density functions.

13.6 Notes

1. The derivation of the formula for the mean open time given by (13.4) can be found in many places (e.g., Keener and Sneyd [42] or Smith [85]).