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Research Article

Marked Continuous-Time Markov Chain Modelling of Burst Behaviour for Single Ion Channels

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Patch clamp recordings from ion channels often show *bursting* behaviour, that is, periods of repetitive activity, which are noticeably separated from each other by periods of inactivity. A number of authors have obtained results for important properties of theoretical and empirical bursts when channel gating is modelled by a continuous-time Markov chain with a finite state space. We show how the use of marked continuous-time Markov chains can simplify the derivation of (i) the distributions of several burst properties, including the total open time, the total charge transfer, and the number of openings in a burst, and (ii) the form of these distributions when the underlying gating process is time reversible and in equilibrium.

1. Introduction

Movement of ions across biological membranes is selectively controlled by specialised protein molecules, called ion channels, which thereby regulate many aspects of cell function. The many kinds of ion channels vary in location, size, chemical structure and function; see, for example, Sakmann and Neher [1]. Usually, ion conduction occurs through a single aqueous pore having a gate that is controlled, for example, by a neurotransmitter, voltage, or membrane tension. Understanding the behaviour of ion channels is important in the study of cell regulation and its pathologies; certain diseases and drugs may affect behaviour of particular channels, and consequently cell functioning. Recordings of the ion flux (tiny current of the order of a few picoamperes) from a single channel are possible through the patch clamp technique (Hamill et al. [2]). At typical recording time resolution, channel gating appears instantaneous, and at any particular time the channel is in one of its stable conductance levels; the simplest channel types exhibit just two, commonly termed *open* (conducting) and *closed* (nonconducting), though some have multiple conductance levels.

Gating behaviour of a single ion channel is usually modelled by a continuous-time homogeneous finite state Markov chain; see Colquhoun and Hawkes [3]. (Other background and references can be gleaned from the work of Sakmann and Neher [1].) Two complications often need to be addressed in such modelling: because each conductance level may arise from several states, there may be *aggregation* of states into conductance classes which partition the state space, into open and closed states in the case of just two conductance levels; also, because of inherent limitations of the recording procedure, very brief sojourns in a class may not be observable (see, e.g., Ball et al. [4] and Hawkes et al. [5]).

Periods of repetitive open channel activity known as *bursts* are often present in a single channel record, and these are noticeably separated from each other by periods of inactivity. Essentially, a burst is a sequence of periods during which the channel is open together with the intervening short closed times, commonly called *gaps*; neighbouring bursts are separated by much longer closed times, termed *interburst sojourns*. Two types of burst have been studied: *theoretical bursts* depend on a partitioning of the closed states into short-lived and long-lived states; *empirical bursts* depend on closed-times being classified as short or long according to whether they do not or do exceed some specified critical time t_c. In practice, from a single channel record only empirical bursts can be determined, and some of their global properties (such as total charge transfer—see Section 4.2) may be less sensitive to problems caused by missed brief sojourns than individual open and closed sojourns. Furthermore, activity *within* a burst is likely to come from only *one* channel even when there are several channels in the patch; consequently data from within empirical bursts are often used for statistical analyses (see, e.g., Colquhoun et al. [6] and Beato et al. [7]). Ball et al. [8, 9] have discussed other reasons for studying bursts.

For a channel with two conductance levels, Colquhoun and Hawkes [3] showed, under diagonalisability assumptions, that the distributions of the duration, total open time, and number of openings in a theoretical burst are each linear combinations of (resp.) exponential or geometric distributions, and that the numbers of these components can be related to the structure of the underlying gating process. Empirical bursts were first considered by Colquhoun and Sakmann [10]; later studies include Ball [11], Li et al. [12], and Yeo et al. [13].

Ball et al. [8, 9] developed a multivariate semi-Markov framework for analysing burst properties of multiconductance channels, that encompassed both theoretical and empirical bursts in a unified fashion, and investigated the form of distributions of burst properties when the underlying channel is in equilibrium and time reversible. (In the absence of a free energy source, any plausible model of channel gating should be time reversible, see Laüger [14].) The aim of the present paper is to show how the results in Ball et al. [8, 9] can be accessed more easily through a marked continuous-time Markov chain (cf. He and Neuts [15]) which is derived from the underlying continuous-time Markov model describing the channel gating behaviour by deleting closed sojourns and concatenating the open sojourns; the marks allow transitions corresponding to the deleted closed sojourns to be labelled according to whether they are gaps or interburst periods. Concatenated processes have been used previously to explain some burst properties; see, for example, Colquhoun and Hawkes [3, pages 20–22] and Ball et al. [8, page 192], [9, page 217]. However, they have not been used previously to provide a systematic approach like that developed in the present paper for derivation of burst properties.

Some background and basic notation for Markov modelling of a single ion channel is given in Section 2, along with definitions of bursts and the key marked continuous-time Markov chain. Section 3 develops some fundamental structural properties of transition-rate matrices and equilibrium distributions relevant for study of bursts, and shows that the key marked process inherits time reversibility from the underlying process. Section 4 then presents derivations for some particular burst properties, the total open time, total charge transfer, and number of openings during a burst. In addition, it summarizes results for other properties, such as the time spent in and the number of visits to a subclass of the open states during a burst. Section 5 makes concluding remarks about some extensions, the advantages and disadvantages of the present approach relative to previous ones, and other applications.

Throughout this paper, vectors and matrices are rendered in bold, all vectors are column vectors, and "$^\top$" denotes transpose, which is used to express row vectors. Furthermore, $\boldsymbol{I}$ denotes an identity matrix, $\mathbf{1}$ a column vector of ones, and $\mathbf{0}$ a matrix (vector) of zeros, dimensions of these being clear from their context.

2. Background and notation

We assume that the gating mechanism of a single ion channel is modelled by an irreducible homogeneous continuous-time Markov chain $\{X(t)\} = \{X(t) : t \geq 0\}$, with finite state space $E = \{1,2,\dots,n\}$, transition-rate matrix $\boldsymbol{Q} = [q_{ij}]$, and equilibrium distribution $\boldsymbol{\pi} = [\pi_1,\pi_2,\dots,\pi_n]^\top$. The state space is partitioned as $E = O \cup C$, where $O = \{1,2,\dots,n_O\}$ and $C = \{n_O+1,n_O+2,\dots,n\}$ correspond to the channel being open and closed, respectively. The closed states are further partitioned as $C = S \cup L$, where $S = \{n_O+1,n_O+2,\dots,n_O+n_S\}$ and $L = \{n_O+n_S+1,n_O+n_S+2,\dots,n\}$ are the short-lived and long-lived closed states, respectively. Let $n_C = n - n_O$ be the number of closed states and $n_L = n - n_O - n_S = n_C - n_S$ be the number of long-lived closed states.

The transition-rate matrix $\boldsymbol{Q}$ may be partitioned in various ways according to the problem under consideration, for example, by the open and closed classes O and C, or by the open, short-lived closed and long-lived closed classes O, S, and L, giving, respectively,

$$\boldsymbol{Q} = \begin{bmatrix} \boldsymbol{Q}_{OO} & \boldsymbol{Q}_{OC} \\ \boldsymbol{Q}_{CO} & \boldsymbol{Q}_{CC} \end{bmatrix} = \begin{bmatrix} \boldsymbol{Q}_{OO} & \boldsymbol{Q}_{OS} & \boldsymbol{Q}_{OL} \\ \boldsymbol{Q}_{SO} & \boldsymbol{Q}_{SS} & \boldsymbol{Q}_{SL} \\ \boldsymbol{Q}_{LO} & \boldsymbol{Q}_{LS} & \boldsymbol{Q}_{LL} \end{bmatrix}. \tag{2.1}$$

Corresponding partitions are used for the equilibrium distribution $\boldsymbol{\pi}$, that is, $\boldsymbol{\pi}^\top = [\boldsymbol{\pi}_O^\top,\boldsymbol{\pi}_C^\top] = [\boldsymbol{\pi}_O^\top,\boldsymbol{\pi}_S^\top,\boldsymbol{\pi}_L^\top]$.

We now give formal definitions of the two types of burst. For a *theoretical burst*, a sojourn of $\{X(t)\}$ in the class C is classified as an *interburst sojourn* if it contains a visit to L, and is classified as a *gap* if it is purely within the class S. The interburst sojourns are used to partition the channel record into bursts. Thus, a given burst begins at the start of the first O sojourn following an interburst sojourn, and ends at the start of the subsequent interburst sojourn. An *empirical burst* is defined by specifying a *critical time* $t_c > 0$ and classifying sojourns in C of duration $>t_c$ as interburst sojourns and those of duration $\leq t_c$ as gaps. A given burst is then defined as for a theoretical burst but with this new definition of interburst sojourns and gaps.

Some basic results for aggregated continuous-time Markov chains, required in the sequel, are now summarized. For $t \geq 0$, let $\boldsymbol{P}_C(t) = [p_{ij}^C(t)]$, where

$$p_{ij}^C(t) = P(X(t) = j,\ X(u) \in C \text{ for } 0 \leq u \leq t \mid X(0) = i) \quad (i, j \in C). \tag{2.2}$$

Then, a standard forward argument (see, e.g., Colquhoun and Hawkes [3, pages 9, 10]) shows that

$$\boldsymbol{P}_C(t) = \exp(\mathbf{Q}_{CC}t) \quad (t \geq 0), \tag{2.3}$$

where $\exp(\mathbf{Q}_{CC}t) = \sum_{k=0}^{\infty} t^k \mathbf{Q}_{CC}^k / k!$ denotes the usual matrix exponential.

Suppose that $X(0) \in C$ and let $T_C = \inf\{t > 0 : X(t) \in O\}$ denote the time elapsing until the channel enters an open state. Then T_C has (matrix) probability density function given by

$$\boldsymbol{f}_{T_C}(t) = \exp(\mathbf{Q}_{CC}t)\mathbf{Q}_{CO} \quad (t > 0), \tag{2.4}$$

where $\boldsymbol{f}_{T_C}(t) = [f_{ij}^{T_C}(t)]$ with

$$f_{ij}^{T_C}(t) = \frac{d}{dt} P(T_C \leq t,\ X(T_C) = j \mid X(0) = i) \quad (i \in C,\ j \in O). \tag{2.5}$$

Hence, if $\boldsymbol{P}_{CO} = [p_{ij}^{CO}]$, where $p_{ij}^{CO} = P(X(T_C) = j \mid X(0) = i)$ $(i \in C,\ j \in O)$, then

$$\boldsymbol{P}_{CO} = \int_0^{\infty} \boldsymbol{f}_{T_C}(t)\,dt = \int_0^{\infty} \exp(\mathbf{Q}_{CC}t)\mathbf{Q}_{CO}\,dt = (-\mathbf{Q}_{CC}^{-1})\mathbf{Q}_{CO}. \tag{2.6}$$

Note that $\mathbf{Q}_{CC}$ is nonsingular since C is a transient class (as $\{X(t)\}$ is irreducible), and hence by Asmussen [16, page 77] all the eigenvalues of $\mathbf{Q}_{CC}$ have strictly negative real parts.

Let $\{\widetilde{X}(t)\}$ be the process obtained from $\{X(t)\}$ by deleting all closed sojourns and concatenating the open sojourns; see Figures 2.1(a) and 2.1(b). The process $\{\widetilde{X}(t)\}$ is a continuous-time Markov chain, with state space O. Let $\mathbf{Q}_{OO}^{\text{cat}} = [q_{ij}^{\text{cat}}]$ denote the transition-rate matrix for concatenated open-to-open transitions; that is, for $i, j \in O$, q_{ij}^{cat} is the rate that, given the channel is in state i, it leaves the open states and subsequently reenters the open states via state j. Then it follows from (2.6) that

$$\mathbf{Q}_{OO}^{\text{cat}} = \mathbf{Q}_{OC}(-\mathbf{Q}_{CC}^{-1})\mathbf{Q}_{CO}. \tag{2.7}$$

Thus $\{\widetilde{X}(t)\}$ has $n_O \times n_O$ transition-rate matrix, $\widetilde{\mathbf{Q}}$ say, given by

$$\widetilde{\mathbf{Q}} = \mathbf{Q}_{OO} + \mathbf{Q}_{OC}(-\mathbf{Q}_{CC}^{-1})\mathbf{Q}_{CO}. \tag{2.8}$$

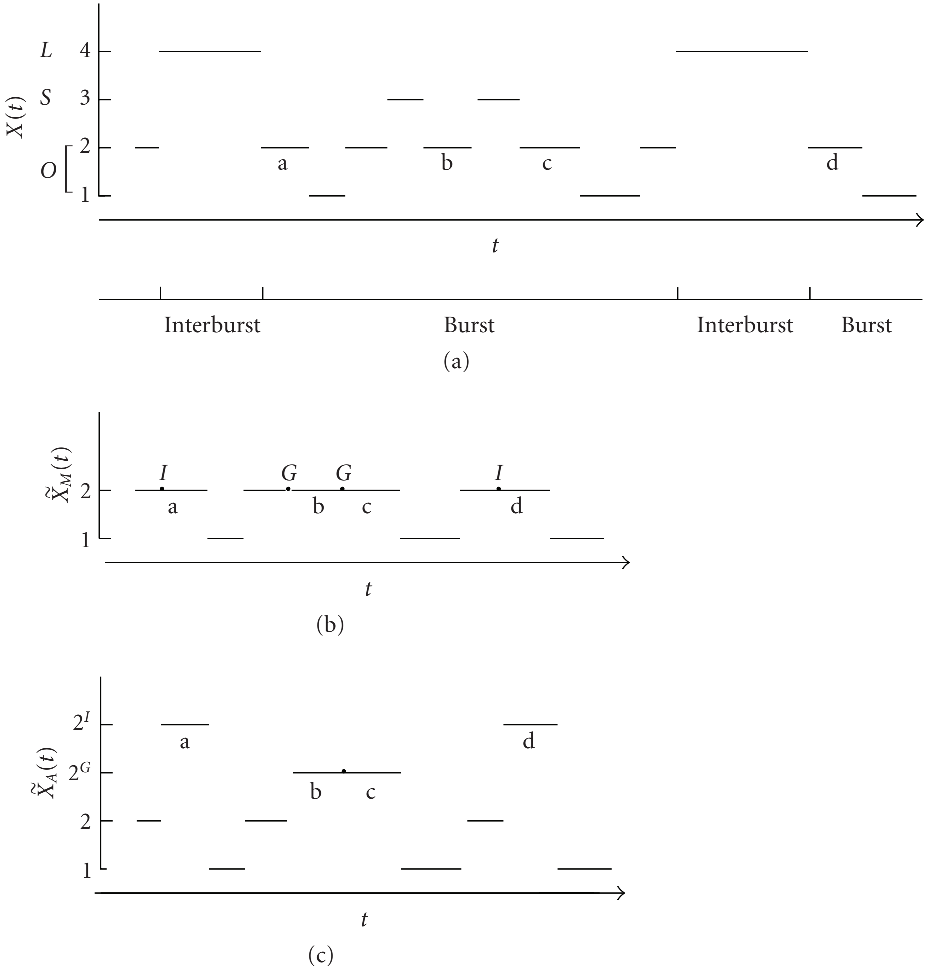

FIGURE 2.1. (a) Partial realization of $\{X(t)\}$ based on $O = \{1,2\}$, $S = \{3\}$, and $L = \{4\}$, with $q_{13} = q_{31} = q_{14} = q_{41} = 0$ and all other entries of $\mathbf{Q}$ nonzero. (For corresponding state space graph see Figure 3.1(a).) Labels a, b, c, d indicate sojourns in an open state immediately preceded by a closed sojourn (allowing these to be tracked in parts (b) and (c)). Also indicated are (theoretical) burst and interburst periods. (b) Corresponding realization of $\{\widetilde{X}_M(t)\}$, that is, realization from (a) after omission of gaps and interburst sojourns, concatenation of neighbouring (open) sojourns, and addition of marks I and G to indicate (preceding omitted) interburst sojourn or gap. Corresponding realization of $\{\widetilde{X}(t)\}$ is obtained by omitting marks. (c) Corresponding realization of augmented process $\{\widetilde{X}_A(t)\}$ (as introduced following proof of Theorem 3.2). This requires two states, denoted 2^G and 2^I, additional to states 1 and 2; these carry information previously indicated by marks I and G in (b).

It is easily verified that $\widetilde{\mathbf{Q}}$ satisfies $\widetilde{\mathbf{Q}}\mathbf{1} = \mathbf{0}$, so it is a proper transition-rate matrix. To see this, start with $\widetilde{\mathbf{Q}}\mathbf{1} = \mathbf{Q}_{OO}\mathbf{1} + \mathbf{Q}_{OC}(-\mathbf{Q}_{CC}^{-1})\mathbf{Q}_{CO}\mathbf{1}$. Expanding $\mathbf{Q}\mathbf{1} = \mathbf{0}$ in partitioned form yields $\mathbf{Q}_{OO}\mathbf{1} + \mathbf{Q}_{OC}\mathbf{1} = \mathbf{0}$ and $\mathbf{Q}_{CO}\mathbf{1} + \mathbf{Q}_{CC}\mathbf{1} = \mathbf{0}$. The latter implies that $(-\mathbf{Q}_{CC}^{-1})\mathbf{Q}_{CO}\mathbf{1} = \mathbf{1}$, whence $\widetilde{\mathbf{Q}}\mathbf{1} = \mathbf{0}$ using the former.

Finally, let $\{\widetilde{X}_M(t)\}$ be defined analogously to $\{\widetilde{X}(t)\}$ except that whenever a closed sojourn of $\{X(t)\}$ is deleted, the corresponding transition of $\{\widetilde{X}(t)\}$ (which may not involve a change of state) is marked G or I, according to whether the closed sojourn of $\{X(t)\}$ is a gap or an interburst sojourn; see Figure 2.1(b).

3. Basic results

The transition-rate matrix, $\widetilde{\mathbf{Q}}$, of $\{\widetilde{X}(t)\}$ can be decomposed as $\widetilde{\mathbf{Q}} = \mathbf{Q}_O + \mathbf{Q}_G + \mathbf{Q}_I$, where $\mathbf{Q}_O$ corresponds to transitions of $\{X(t)\}$ purely within O (i.e., without any deletion and concatenation), and $\mathbf{Q}_G$ and $\mathbf{Q}_I$ to transitions which result from deleted sojourns which were gaps and interburst sojourns, respectively.

Theorem 3.1. *For both types of burst,*

$$\mathbf{Q}_O = \mathbf{Q}_{OO}. \tag{3.1}$$

For a theoretical burst,

$$\mathbf{Q}_G = \mathbf{Q}_{OS}(-\mathbf{Q}_{SS}^{-1})\mathbf{Q}_{SO}, \tag{3.2}$$

$$\mathbf{Q}_I = -[\mathbf{Q}_{OL} + \mathbf{Q}_{OS}(-\mathbf{Q}_{SS}^{-1})\mathbf{Q}_{SL}][\mathbf{Q}_{LL} + \mathbf{Q}_{LS}(-\mathbf{Q}_{SS}^{-1})\mathbf{Q}_{SL}]^{-1}[\mathbf{Q}_{LO} + \mathbf{Q}_{LS}(-\mathbf{Q}_{SS}^{-1})\mathbf{Q}_{SO}]. \tag{3.3}$$

For an empirical burst,

$$\mathbf{Q}_G = \mathbf{Q}_{OC}(-\mathbf{Q}_{CC}^{-1})(\boldsymbol{I} - e^{\mathbf{Q}_{CC}t_c})\mathbf{Q}_{CO}, \tag{3.4}$$

$$\mathbf{Q}_I = \mathbf{Q}_{OC}(-\mathbf{Q}_{CC}^{-1})\, e^{\mathbf{Q}_{CC}t_c}\mathbf{Q}_{CO}. \tag{3.5}$$

Proof. The off-diagonal elements of (3.1) are clear; since $\widetilde{\mathbf{Q}}$ is a proper transition matrix, the diagonal elements of (3.1) follow, respectively, for each type of burst once (3.2) and (3.3), or (3.4) and (3.5), have been established.

For a theoretical burst, (3.2) follows from (2.7) with C replaced by S. To prove (3.3), consider an alternative concatenation of $\{X(t)\}$ in which sojourns in S are deleted unless they are gaps. This yields a continuous-time Markov chain $\{X'(t)\}$ say, with transition-rate matrix $\mathbf{Q}'$ having partitioned form

$$\mathbf{Q}' = \begin{bmatrix} \mathbf{Q}'_{OO} & \mathbf{Q}'_{OS} & \mathbf{Q}'_{OL} \\ \mathbf{Q}'_{SO} & \mathbf{Q}'_{SS} & \mathbf{0} \\ \mathbf{Q}'_{LO} & \mathbf{0} & \mathbf{Q}'_{LL} \end{bmatrix}. \tag{3.6}$$

Now, arguing as for (2.8), $\mathbf{Q}'_{LL} = \mathbf{Q}_{LL} + \mathbf{Q}_{LS}(-\mathbf{Q}_{SS}^{-1})\mathbf{Q}_{SL}$. Also $\mathbf{Q}'_{OL} = \mathbf{Q}_{OL} + \mathbf{Q}_{OS}(-\mathbf{Q}_{SS}^{-1})\mathbf{Q}_{SL}$, where the first term corresponds to transitions directly from O to L and the second to transitions that involve an intervening sojourn in S. Similarly, $\mathbf{Q}'_{LO} = \mathbf{Q}_{LO} + \mathbf{Q}_{LS}(-\mathbf{Q}_{SS}^{-1})\mathbf{Q}_{SO}$. It then follows as in (2.7), with C replaced by L, that $\mathbf{Q}_I = \mathbf{Q}'_{OL}(-\mathbf{Q}'_{LL})^{-1}\mathbf{Q}'_{LO}$, yielding (3.3).

For an empirical burst, using (2.4), $\mathbf{Q}_G = \int_0^{t_c}\mathbf{Q}_{OC}e^{\mathbf{Q}_{CC}t}\mathbf{Q}_{CO}\,dt$ and $\mathbf{Q}_I = \int_{t_c}^{\infty}\mathbf{Q}_{OC}e^{\mathbf{Q}_{CC}t}\mathbf{Q}_{CO}\,dt$; hence (3.4) and (3.5) follow. □

The process $\{\widetilde{X}(t)\}$ inherits irreducibility from $\{X(t)\}$, so $\{\widetilde{X}(t)\}$ possesses an equilibrium distribution, $\widetilde{\boldsymbol{\pi}}$ say (of dimension n_O). It is intuitively clear that

$$\widetilde{\boldsymbol{\pi}} = (\boldsymbol{\pi}_O^\top \mathbf{1})^{-1}\boldsymbol{\pi}_O, \tag{3.7}$$

since concatenating closed sojourns does not affect the long-term relative proportions of time that $\{X(t)\}$ spends in the different open states. More formally, it is easily verified that $\widetilde{\boldsymbol{\pi}}^\top \widetilde{\mathbf{Q}} = \mathbf{0}$. For example, for empirical bursts, $\boldsymbol{\pi}_O^\top \widetilde{\mathbf{Q}} = \boldsymbol{\pi}_O^\top \mathbf{Q}_{OO} + \boldsymbol{\pi}_O^\top \mathbf{Q}_{OC}(-\mathbf{Q}_{CC}^{-1})\mathbf{Q}_{CO}$. Now, $\boldsymbol{\pi}^\top \mathbf{Q} = \mathbf{0}$, since $\boldsymbol{\pi}$ is the equilibrium distribution of $\{X(t)\}$, and expanding this in partitioned form yields $\boldsymbol{\pi}_O^\top \mathbf{Q}_{OC} + \boldsymbol{\pi}_C^\top \mathbf{Q}_{CC} = \mathbf{0}$, so $\boldsymbol{\pi}_O^\top \mathbf{Q}_{OC}(-\mathbf{Q}_{CC}^{-1})\mathbf{Q}_{CO} = \boldsymbol{\pi}_C^\top \mathbf{Q}_{CO}$. Hence, $\boldsymbol{\pi}_O^\top \widetilde{\mathbf{Q}} = \boldsymbol{\pi}_O^\top \mathbf{Q}_{OO} + \boldsymbol{\pi}_C^\top \mathbf{Q}_{CO} = \mathbf{0}$, since $\boldsymbol{\pi}^\top \mathbf{Q} = \mathbf{0}$. Thus $\widetilde{\boldsymbol{\pi}}^\top \widetilde{\mathbf{Q}} = \mathbf{0}$, as required. A similar argument holds for theoretical bursts.

Recall that $\{X(t)\}$ is reversible if and only if the detailed balance conditions

$$\pi_i q_{ij} = \pi_j q_{ji} \quad (i, j \in E) \tag{3.8}$$

are satisfied. Let $\boldsymbol{W} = \operatorname{diag}(\boldsymbol{\pi})$ be the diagonal matrix whose entries on the diagonal are those of $\boldsymbol{\pi}$. Then (3.8) can be written as

$$\boldsymbol{W}^{1/2}\mathbf{Q}\boldsymbol{W}^{-1/2} = (\boldsymbol{W}^{1/2}\mathbf{Q}\boldsymbol{W}^{-1/2})^\top. \tag{3.9}$$

Expanding (3.9) in partitioned form yields (cf. Fredkin et al. [17]) that if $A \subseteq E$ and $\boldsymbol{W}_A = \operatorname{diag}(\boldsymbol{\pi}_A)$ then

$$\boldsymbol{W}_A^{1/2}\mathbf{Q}_{AA}\boldsymbol{W}_A^{-1/2} = (\boldsymbol{W}_A^{1/2}\mathbf{Q}_{AA}\boldsymbol{W}_A^{-1/2})^\top, \tag{3.10}$$

while if $A, B \subset E$ are disjoint then

$$\boldsymbol{W}_A^{1/2}\mathbf{Q}_{AB}\boldsymbol{W}_B^{-1/2} = (\boldsymbol{W}_B^{1/2}\mathbf{Q}_{BA}\boldsymbol{W}_A^{-1/2})^\top. \tag{3.11}$$

Theorem 3.2. *For both theoretical and empirical bursts, if* $\{X(t)\}$ *is reversible, then so are* $\{\widetilde{X}(t)\}$ *and* $\{\widetilde{X}_M(t)\}$.

Proof. Again this is clear on intuitive grounds. For a formal proof we show that detailed balance holds for the three types of transition in $\{\widetilde{X}_M(t)\}$, that is, that $\widetilde{\boldsymbol{W}}^{1/2}\mathbf{Q}_O\widetilde{\boldsymbol{W}}^{-1/2}$, $\widetilde{\boldsymbol{W}}^{1/2}\mathbf{Q}_G\widetilde{\boldsymbol{W}}^{-1/2}$, and $\widetilde{\boldsymbol{W}}^{1/2}\mathbf{Q}_I\widetilde{\boldsymbol{W}}^{-1/2}$ are all symmetric, where $\widetilde{\boldsymbol{W}} = \operatorname{diag}(\widetilde{\boldsymbol{\pi}})$. Note that, because of (3.7), it is sufficient to show that $\boldsymbol{W}_O^{1/2}\mathbf{Q}_O\boldsymbol{W}_O^{-1/2}$, $\boldsymbol{W}_O^{1/2}\mathbf{Q}_G\boldsymbol{W}_O^{-1/2}$, and $\boldsymbol{W}_O^{1/2}\mathbf{Q}_I\boldsymbol{W}_O^{-1/2}$ are all symmetric. Setting $A = O$ in (3.10) and recalling (3.1) shows that $\boldsymbol{W}_O^{1/2}\mathbf{Q}_O\boldsymbol{W}_O^{-1/2}$ is symmetric for both types of burst.

For theoretical bursts, using (3.2),

$$\boldsymbol{W}_O^{1/2}\mathbf{Q}_G\boldsymbol{W}_O^{-1/2} = \boldsymbol{W}_O^{1/2}\mathbf{Q}_{OS}\boldsymbol{W}_S^{-1/2}\left[\boldsymbol{W}_S^{1/2}(-\mathbf{Q}_{SS})\boldsymbol{W}_S^{-1/2}\right]^{-1}\boldsymbol{W}_S^{1/2}\mathbf{Q}_{SO}\boldsymbol{W}_O^{-1/2}, \tag{3.12}$$

which is symmetric, because of (3.10) with $A = S$ and (3.11) with $A = O$ and $B = S$. A similar argument shows that $\boldsymbol{W}_O^{1/2}\mathbf{Q}_I\boldsymbol{W}_O^{-1/2}$ is symmetric.

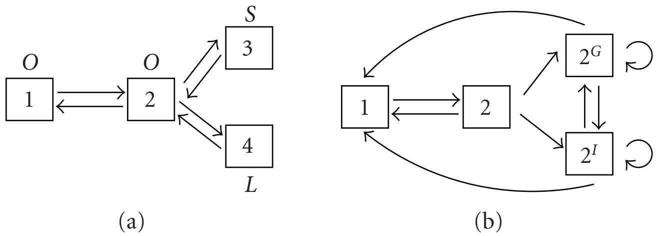

FIGURE 3.1. State space graphs based on $O = \{1,2\}$, $S = \{3\}$, and $L = \{4\}$, with $q_{13} = q_{31} = q_{14} = q_{41} = 0$ and all other entries of $\boldsymbol{Q}$ nonzero. (a) State space graph for underlying process. (b) State space graph for augmented process corresponding to (a); note that only states 1, 2, and additional states 2^G and 2^I are required in this case. The augmented process is clearly nonreversible; for example, state 1 can be reached from, but not followed by, 2^G or 2^I.

For empirical bursts, noting that $-\boldsymbol{Q}_{CC}^{-1}(\boldsymbol{I} - e^{\boldsymbol{Q}_{CC}t_c}) = \sum_{k=1}^{\infty} \boldsymbol{Q}_{CC}^{k-1} t_c^k/k!$,

$$\boldsymbol{W}_O^{1/2}\boldsymbol{Q}_G\boldsymbol{W}_O^{-1/2} = \boldsymbol{W}_O^{1/2}\boldsymbol{Q}_{OC}\boldsymbol{W}_C^{-1/2}\left[\sum_{k=1}^{\infty}(\boldsymbol{W}_C^{1/2}\boldsymbol{Q}_{CC}\boldsymbol{W}_C^{-1/2})^{k-1}t_c^k/k!\right]\boldsymbol{W}_C^{1/2}\boldsymbol{Q}_{CO}\boldsymbol{W}_O^{-1/2}, \tag{3.13}$$

which is symmetric, because of (3.10) with $A = C$ and (3.11) with $A = O$ and $B = C$. Similarly, $\boldsymbol{W}_O^{1/2}\boldsymbol{Q}_I\boldsymbol{W}_O^{-1/2}$ is symmetric. □

The marked process $\{\widetilde{X}_M(t)\}$ could in principle be modelled by augmenting the state space of $\{\widetilde{X}(t)\}$ to indicate whether the current state was immediately preceded by another open state, a deleted gap, or a deleted interburst sojourn. This augmented process, $\{\widetilde{X}_A(t)\}$ say, is a continuous-time Markov chain. Suppose that $\{X(t)\}$ has state space graph as in Figure 3.1(a). In this example, since state 1 cannot be reached directly from either state 3 or state 4, only states 1 and 2, and two additional states, 2^G and 2^I (say), are required for the augmented process. Figure 3.1(b) gives the state space graph and shows the nonreversibility of this augmented process; see Figure 2.1(c) for a typical (partial) realization of $\{\widetilde{X}_A(t)\}$, corresponding to those for $\{X(t)\}$ and $\{\widetilde{X}_M(t)\}$ in Figures 2.1(a) and 2.1(b). In general, the augmented process requires a state space which is up to three times the size of that of the marked process: $\{1,2,\dots,n_O,\ 1^G,2^G,\dots,n_O^G,\ 1^I,2^I,\dots,n_O^I\}$ (say). Hence, this approach would not be so useful because, as well as increasing the size of the state space, such an augmented process need not be reversible.

Let $\{J_k\}$ be the discrete-time Markov chain that records the entry state of successive bursts, that is, the state of $\{\widetilde{X}_M(t)\}$ immediately following successive I-marked transitions. The transition matrix of $\{J_k\}$ is $\boldsymbol{P}_B = -\widetilde{\boldsymbol{Q}}_O^{-1}\boldsymbol{Q}_I$, where $\widetilde{\boldsymbol{Q}}_O = \boldsymbol{Q}_O + \boldsymbol{Q}_G$. (By analogy with (2.3), the (matrix) probability that $\{\widetilde{X}_M(t)\}$ does not have an I-transition in $(0,t]$ is $\exp(\widetilde{\boldsymbol{Q}}_O t)$, so $\boldsymbol{P}_B = \int_0^{\infty}\exp(\widetilde{\boldsymbol{Q}}_O t)\boldsymbol{Q}_I\,dt = -\widetilde{\boldsymbol{Q}}_O^{-1}\boldsymbol{Q}_I$. The matrix $\widetilde{\boldsymbol{Q}}_O$ is nonsingular because its eigenvalues have strictly negative real parts, since $\exp(\widetilde{\boldsymbol{Q}}_O t) \to \boldsymbol{0}$ as $t \to \infty$). Note that $\{J_k\}$ also inherits irreducibility from $\{X(t)\}$, though the state space of $\{J_k\}$ may be a proper subset of O, for example, if there are open states which cannot be entered directly from C. If $\{J_k\}$ is also aperiodic, as is necessarily the case when $\boldsymbol{Q}$ is such that $q_{ij} > 0$ if and only if $q_{ji} > 0$ (a condition that is satisfied by most physically plausible channel gating models and by all time reversible models), then $\{J_k\}$ possesses an equilibrium distribution, $\boldsymbol{\psi} = [\psi_1, \psi_2, \dots, \psi_{n_O}]^\top$ say, where ψ_i is the equilibrium probability that a burst begins in state i. (If the state space of $\{J_k\}$ is a proper subset of O, then some of the elements of $\boldsymbol{\psi}$ are zero.)

Lemma 3.3. *The equilibrium distribution* $\boldsymbol{\psi}$ *of* $\{J_k\}$ *is given by* $\boldsymbol{\psi}^\top = \boldsymbol{\pi}_O^\top \mathbf{Q}_I / (\boldsymbol{\pi}_O^\top \mathbf{Q}_I \mathbf{1})$.

Proof. Recall that $\widetilde{\mathbf{Q}} = \widetilde{\mathbf{Q}}_O + \mathbf{Q}_I$ and, using (3.7), that $\boldsymbol{\pi}_O^\top \widetilde{\mathbf{Q}} = \mathbf{0}$. Thus $\boldsymbol{\pi}_O^\top \mathbf{Q}_I = -\boldsymbol{\pi}_O^\top \widetilde{\mathbf{Q}}_O$, so using $\boldsymbol{P}_B = -\widetilde{\mathbf{Q}}_O^{-1} \mathbf{Q}_I$ gives $\boldsymbol{\pi}_O^\top \mathbf{Q}_I \boldsymbol{P}_B = \boldsymbol{\pi}_O^\top \mathbf{Q}_I$. Hence $\boldsymbol{\psi}^\top \boldsymbol{P}_B = \boldsymbol{\psi}^\top$, as required. $\square$

The equilibrium distribution in Lemma 3.3 is intuitively clear in view of (3.7) and the fact that a burst is immediately preceded by an I-transition of $\{\widetilde{X}_M(t)\}$. Alternative expressions for $\boldsymbol{\psi}$ have been given by, for example, Colquhoun and Hawkes [3, Equation (3.2)] for theoretical bursts, and Ball [11, Equation (3.9)] and Li et al. [12, Equation (2.10)] for empirical bursts.

4. Properties of bursts

4.1. Total open time during a burst. Suppose that $\{\widetilde{X}_M(t)\}$ is in equilibrium. Then the times of I-transitions of $\{\widetilde{X}_M(t)\}$ form a stationary point process. Let T_O denote the length of a typical interval in this point process (i.e., the time between two successive I-transitions) and let U_O denote a typical excess lifetime (i.e., the time from an arbitrary time point until the next I-transition of $\{\widetilde{X}_M(t)\}$). Note that, because in $\{\widetilde{X}_M(t)\}$ all closed sojourns have been omitted and the open sojourns concatenated, T_O gives the total open time during a typical burst. Since $\{\widetilde{X}_M(t)\}$ is in equilibrium, the survivor function, $\overline{F}_{U_O}(t)$ say, of U_O is given by

$$\overline{F}_{U_O}(t) = \widetilde{\boldsymbol{\pi}}^\top e^{\widetilde{\mathbf{Q}}_O t} \mathbf{1} \quad (t > 0). \tag{4.1}$$

Thus, by the standard relationship between the distributions of a typical lifetime and a typical excess lifetime of a stationary point process, the pdf of T_O, $f_{T_O}(t)$ say, is given by

$$f_{T_O}(t) = \mu_{T_O} \overline{F}''_{U_O}(t) = \mu_{T_O} \widetilde{\boldsymbol{\pi}}^\top \widetilde{\mathbf{Q}}_O^2 e^{\widetilde{\mathbf{Q}}_O t} \mathbf{1} \quad (t > 0), \tag{4.2}$$

where, with D_+ denoting right-hand derivative, $\mu_{T_O} = E[T_O] = [-D_+ \overline{F}_{U_O}(0)]^{-1} = (-\widetilde{\boldsymbol{\pi}}^\top \widetilde{\mathbf{Q}}_O \mathbf{1})^{-1}$; cf. Ball and Milne [18].

Now, suppose that $\{X(t)\}$, and hence $\{\widetilde{X}(t)\}$, is time reversible. Then

$$\overline{F}_{U_O}(t) = \mathbf{1}^\top \widetilde{\boldsymbol{W}} e^{\widetilde{\mathbf{Q}}_O t} \mathbf{1} = \mathbf{1}^\top \widetilde{\boldsymbol{W}}^{1/2} \widetilde{\boldsymbol{W}}^{1/2} e^{\widetilde{\mathbf{Q}}_O t} \widetilde{\boldsymbol{W}}^{-1/2} \widetilde{\boldsymbol{W}}^{1/2} \mathbf{1} \quad (t > 0). \tag{4.3}$$

Now, using the series expression for the matrix exponential, $\widetilde{\boldsymbol{W}}^{1/2} e^{\widetilde{\mathbf{Q}}_O t} \widetilde{\boldsymbol{W}}^{-1/2} = \exp(\widetilde{\boldsymbol{W}}^{1/2} \widetilde{\mathbf{Q}}_O \widetilde{\boldsymbol{W}}^{-1/2} t)$. Further, $\widetilde{\boldsymbol{W}}^{1/2} \widetilde{\mathbf{Q}}_O \widetilde{\boldsymbol{W}}^{-1/2} = \widetilde{\boldsymbol{W}}^{1/2} (\mathbf{Q}_O + \mathbf{Q}_G) \widetilde{\boldsymbol{W}}^{-1/2}$ is symmetric as $\{\widetilde{X}_M(t)\}$ is time reversible. Hence, $\widetilde{\boldsymbol{W}}^{1/2} \widetilde{\mathbf{Q}}_O \widetilde{\boldsymbol{W}}^{-1/2}$ admits the spectral representation

$$\widetilde{\boldsymbol{W}}^{1/2} \widetilde{\mathbf{Q}}_O \widetilde{\boldsymbol{W}}^{-1/2} = \sum_{i=1}^{n_O} \lambda_i \boldsymbol{x}_i \boldsymbol{x}_i^\top, \tag{4.4}$$

where $\lambda_1,\lambda_2,\dots,\lambda_{n_O}$ are the eigenvalues of $\widetilde{\boldsymbol{Q}}_O$, which are all real (as $\widetilde{\boldsymbol{W}}^{1/2}\widetilde{\boldsymbol{Q}}_O\widetilde{\boldsymbol{W}}^{-1/2}$ is symmetric) and strictly negative, and $\boldsymbol{x}_1,\boldsymbol{x}_2,\dots,\boldsymbol{x}_{n_O}$ is a corresponding orthonormal set of right eigenvectors.

Substituting (4.4) into (4.3) yields

$$\overline{F}_{U_O}(t)=\sum_{i=1}^{n_O}\alpha_i\,\mathrm{e}^{\lambda_i t}\quad(t>0),\tag{4.5}$$

where, for $i=1,2,\dots,n_O$, $\alpha_i=\boldsymbol{1}^\top\widetilde{\boldsymbol{W}}^{1/2}\boldsymbol{x}_i\boldsymbol{x}_i^\top\widetilde{\boldsymbol{W}}^{1/2}\boldsymbol{1}=(\boldsymbol{x}_i^\top\widetilde{\boldsymbol{W}}^{1/2}\boldsymbol{1})^2\geq 0$. Thus, if $\{X(t)\}$ is time-reversible and in equilibrium, then T_O is distributed as a mixture of at most n_O negative exponential random variables; this distribution is obtained from Ball et al. [9, Equation (3.17)] (by taking their $\boldsymbol{c}=\boldsymbol{1}$).

4.2. Total charge transfer during a burst. For $i\in O$, let c_i denote the current when $X(t)=i$, that is, when the channel is in open state i. The total charge transfer during a burst is the integral of the current over the burst, which is given by $\int_0^{T_O}c_{\widetilde{X}(t)}\,dt$, assuming that the burst starts at $t=0$ and the current is zero when $X(t)\in C$. Suppose that $c_i>0$ $(i\in O)$. Let $\{\widehat{X}(t)\}$ and $\{\widehat{X}_M(t)\}$ denote the random time-changed versions of $\{\widetilde{X}(t)\}$ and $\{\widetilde{X}_M(t)\}$, respectively, obtained by running the clock at rate c_i^{-1} when $\widetilde{X}(t)=i$ $(i\in O)$. Let $\boldsymbol{C}=\operatorname{diag}(\boldsymbol{c})$, where $\boldsymbol{c}=(c_1,c_2,\dots,c_{n_O})^\top$. The transition-rate matrix, $\widehat{\boldsymbol{Q}}$ say, of $\{\widehat{X}(t)\}$ admits the decomposition $\widehat{\boldsymbol{Q}}=\widehat{\boldsymbol{Q}}_O+\widehat{\boldsymbol{Q}}_G+\widehat{\boldsymbol{Q}}_I$, where $\widehat{\boldsymbol{Q}}_O=\boldsymbol{C}^{-1}\boldsymbol{Q}_O$, $\widehat{\boldsymbol{Q}}_G=\boldsymbol{C}^{-1}\boldsymbol{Q}_{\boldsymbol{G}}$, and $\widehat{\boldsymbol{Q}}_{\boldsymbol{I}}=\boldsymbol{C}^{-1}\boldsymbol{Q}_I$. It is easily verified that $\{\widehat{X}(t)\}$ has equilibrium distribution, $\widehat{\boldsymbol{\pi}}$ say, given by $\widehat{\boldsymbol{\pi}}^\top=(\widetilde{\boldsymbol{\pi}}^\top\boldsymbol{C}\boldsymbol{1})^{-1}\widetilde{\boldsymbol{\pi}}^\top\boldsymbol{C}$, and that $\{\widehat{X}_M(t)\}$ is time reversible if and only if $\{\widetilde{X}_M(t)\}$ is time reversible.

Let $\widehat{T}_O$ be the time elapsing between two successive I-transitions of $\{\widehat{X}_M(t)\}$, that is, the total charge transfer over a typical burst (since all closed sojourns have been omitted and the open sojourns concatenated). Then, in equilibrium, the distribution of $\widehat{T}_O$ is given by (4.2), with $\widetilde{\boldsymbol{\pi}}$ replaced by $\widehat{\boldsymbol{\pi}}$, $\widetilde{\boldsymbol{Q}}_O$ replaced by $\widetilde{\widehat{\boldsymbol{Q}}}_O=\widehat{\boldsymbol{Q}}_O+\widehat{\boldsymbol{Q}}_G$, and μ_{T_O} replaced by $\widehat{\mu}_{T_O}=-(\widehat{\boldsymbol{\pi}}_O^\top\widetilde{\widehat{\boldsymbol{Q}}}_O\boldsymbol{1})^{-1}$. Further, it follows as in Section 4.1, that, if $\{X(t)\}$ is time-reversible, then, in equilibrium, $\widehat{T}_O$ is distributed as a mixture of at most n_O negative exponential random variables; see Ball et al. [9, Equation (3.17)].

4.3. Number of openings during a burst. Let N_O be the number of openings in a burst. Note that $N_O=k$ if and only if, in $\{\widetilde{X}_M(t)\}$, the number of G-marks between two successive I-marks is $k-1$. The (substochastic) transition matrix between two successive marks in $\{\widehat{X}_M(t)\}$ is $-\boldsymbol{Q}_O^{-1}\boldsymbol{Q}_G$ if the second mark is a G, and $-\boldsymbol{Q}_O^{-1}\boldsymbol{Q}_I$ if the second mark is an I. Thus, in equilibrium, and using Lemma 3.3,

$$P(N_O=k)=(\boldsymbol{\pi}_O^\top\boldsymbol{Q}_I\boldsymbol{1})^{-1}\boldsymbol{\pi}_O^\top\boldsymbol{Q}_I(-\boldsymbol{Q}_O^{-1}\boldsymbol{Q}_G)^{k-1}(-\boldsymbol{Q}_O^{-1}\boldsymbol{Q}_I)\boldsymbol{1}\quad(k=1,2,\dots).\tag{4.6}$$

Suppose that $\{X(t)\}$, and hence $\{\widetilde{X}_M(t)\}$, is time reversible. The strictly positive definite matrix $-\boldsymbol{W}_O^{1/2}\boldsymbol{Q}_O\boldsymbol{W}_O^{-1/2}$ is then symmetric, so $(-\boldsymbol{Q}_O)^{-1/2}$ exists and $\boldsymbol{A}_O$ defined by

$\boldsymbol{A}_O = \boldsymbol{W}_O^{1/2}(-\boldsymbol{Q}_O)^{-1/2}\boldsymbol{W}_O^{-1/2}$ is symmetric. Thus,

$$\begin{aligned}
&P(N_O = k)\\
&= (\boldsymbol{\pi}_O^\top \boldsymbol{Q}_I \mathbf{1})^{-1}\mathbf{1}^\top \boldsymbol{W}_O \boldsymbol{Q}_I(-\boldsymbol{Q}_O)^{-1/2}[(-\boldsymbol{Q}_O)^{-1/2}\boldsymbol{Q}_G(-\boldsymbol{Q}_O)^{-1/2}]^{k-1}(-\boldsymbol{Q}_O)^{-1/2}\boldsymbol{Q}_I\mathbf{1}\\
&= (\boldsymbol{\pi}_O^\top \boldsymbol{Q}_I \mathbf{1})^{-1}\mathbf{1}^\top \boldsymbol{W}_O^{1/2}\boldsymbol{W}_O^{1/2}\boldsymbol{Q}_I(-\boldsymbol{Q}_O)^{-1/2}\boldsymbol{W}_O^{-1/2}[\boldsymbol{W}_O^{1/2}(-\boldsymbol{Q}_O)^{-1/2}\boldsymbol{Q}_G(-\boldsymbol{Q}_O)^{-1/2}\boldsymbol{W}_O^{-1/2}]^{k-1}\\
&\qquad \times \boldsymbol{W}_O^{1/2}(-\boldsymbol{Q}_O)^{-1/2}\boldsymbol{Q}_I\boldsymbol{W}_O^{-1/2}\boldsymbol{W}_O^{1/2}\mathbf{1}.
\end{aligned} \tag{4.7}$$

The matrix $\boldsymbol{W}_O^{1/2}(-\boldsymbol{Q}_O)^{-1/2}\boldsymbol{Q}_G(-\boldsymbol{Q}_O)^{-1/2}\boldsymbol{W}_O^{-1/2} = \boldsymbol{A}_O(\boldsymbol{W}_O^{1/2}\boldsymbol{Q}_G\boldsymbol{W}_O^{-1/2})\boldsymbol{A}_O$ is symmetric and positive (semi-) definite (noting that the eigenvalues of $\boldsymbol{Q}_G$ are nonnegative for both types of burst) and hence admits the spectral representation

$$\boldsymbol{W}_O^{1/2}(-\boldsymbol{Q}_O)^{-1/2}\boldsymbol{Q}_G(-\boldsymbol{Q}_O)^{-1/2}\boldsymbol{W}_O^{-1/2} = \sum_{i=1}^{n_O}\rho_i \boldsymbol{y}_i\boldsymbol{y}_i^\top. \tag{4.8}$$

Further, the eigenvalues satisfy $0 \le \rho_i < 1$ ($i = 1,2,\ldots,n_O$) as the matrix on the left in (4.8) is similar to the substochastic matrix $-\boldsymbol{Q}_O^{-1}\boldsymbol{Q}_G$. Substituting (4.8) into (4.7) yields

$$P(N_O = k) = \sum_{i=1}^{n_O}\beta_i\rho_i^{k-1} \quad (k = 1,2,\ldots), \tag{4.9}$$

where $\beta_i = (\boldsymbol{\pi}_O^\top \boldsymbol{Q}_I\mathbf{1})^{-1}\mathbf{1}^\top \boldsymbol{W}_O^{1/2}\boldsymbol{W}_O^{1/2}\boldsymbol{Q}_I(-\boldsymbol{Q}_O)^{-1/2}\boldsymbol{W}_O^{-1/2}\boldsymbol{y}_i\boldsymbol{y}_i^\top \boldsymbol{W}_O^{1/2}(-\boldsymbol{Q}_O)^{-1/2}\boldsymbol{Q}_I\boldsymbol{W}_O^{-1/2}\boldsymbol{W}_O^{1/2}\mathbf{1}$. Now, as $\{\widetilde{X}_M(t)\}$ is reversible,

$$\begin{aligned}
&\boldsymbol{W}_O^{1/2}\boldsymbol{Q}_I(-\boldsymbol{Q}_O)^{-1/2}\boldsymbol{W}_O^{-1/2}\\
&\qquad = \boldsymbol{W}_O^{1/2}\boldsymbol{Q}_I\boldsymbol{W}_O^{-1/2}\boldsymbol{A}_O = [\boldsymbol{A}_O\boldsymbol{W}_O^{1/2}\boldsymbol{Q}_I\boldsymbol{W}_O^{-1/2}]^\top = [\boldsymbol{W}_O^{1/2}(-\boldsymbol{Q}_O)^{-1/2}\boldsymbol{Q}_I\boldsymbol{W}_O^{-1/2}]^\top.
\end{aligned} \tag{4.10}$$

Thus,

$$\beta_i = (\boldsymbol{\pi}_O^\top \boldsymbol{Q}_I\mathbf{1})^{-1}[\mathbf{1}^\top \boldsymbol{W}_O\boldsymbol{Q}_I(-\boldsymbol{Q}_O)^{-1/2}\boldsymbol{W}_O^{-1/2}\boldsymbol{y}_i][\mathbf{1}^\top \boldsymbol{W}_O\boldsymbol{Q}_I(-\boldsymbol{Q}_O)^{-1/2}\boldsymbol{W}_O^{-1/2}\boldsymbol{y}_i]^\top \ge 0, \tag{4.11}$$

so if $\{X(t)\}$ is time-reversible and in equilibrium then N_O is distributed as a mixture of at most rank($\boldsymbol{Q}_G$) geometric random variables; cf. Ball et al. [9, Equation (3.28)].

4.4. Other properties. Various other properties of bursts may be obtained by using appropriate marked processes. For example, suppose that $n_O > 1$ and consider a proper subset $A \subset O$ of the class O of open states. Let T_A denote the time $\{\widetilde{X}(t)\}$ spends in A between two successive I-transitions. For example, if A denotes the open states that have a specified conductance level, then T_A is the total time the channel spends at that conductance level during a typical burst. Let $B = O\backslash A$. Now partition $\widetilde{\boldsymbol{Q}}_O$ and $\boldsymbol{\psi}$, giving, respectively,

$$\widetilde{\boldsymbol{Q}}_O = \begin{bmatrix} \widetilde{\boldsymbol{Q}}_{AA} & \widetilde{\boldsymbol{Q}}_{AB} \\ \widetilde{\boldsymbol{Q}}_{BA} & \widetilde{\boldsymbol{Q}}_{BB} \end{bmatrix}, \qquad \boldsymbol{\psi}^\top = [\boldsymbol{\psi}_A^\top, \boldsymbol{\psi}_B^\top]. \tag{4.12}$$

Then, in equilibrium, the probability that the channel visits A during a burst is given by $P(T_A > 0) = \boldsymbol{\psi}_A^\top \mathbf{1} + \boldsymbol{\psi}_B^\top(-\widetilde{\boldsymbol{Q}}_{BB})^{-1}\widetilde{\boldsymbol{Q}}_{BA}\mathbf{1}$. Note that it is possible for this probability to be strictly less than one, in which case $P(T_A = 0) > 0$.

The distribution of $T_A \mid T_A > 0$ can be obtained by a further concatenation of $\{\widetilde{X}(t)\}$, in which sojourns in B are deleted. Denote the resulting process by $\{X^*(t)\}$ and the corresponding marked process by $\{X_M^*(t)\}$. The transition-rate matrix of $\{X^*(t)\}$, $\boldsymbol{Q}^*$ say, admits the decomposition $\boldsymbol{Q}^* = \boldsymbol{Q}_A^* + \boldsymbol{Q}_G^* + \boldsymbol{Q}_I^*$. Moreover, if $\widetilde{\boldsymbol{Q}}_A^* = \boldsymbol{Q}_A^* + \boldsymbol{Q}_G^*$, then arguing as for (2.8) yields $\widetilde{\boldsymbol{Q}}_A^* = \widetilde{\boldsymbol{Q}}_{AA} + \widetilde{\boldsymbol{Q}}_{AB}(-\widetilde{\boldsymbol{Q}}_{BB}^{-1})\widetilde{\boldsymbol{Q}}_{BA}$.

Suppose that $\{X_M^*(t)\}$ is in equilibrium and let U_A denote a typical excess lifetime from an arbitrary time until the next I-transition of $\{X_M^*(t)\}$. Then U_A has survivor function given by $\overline{F}_{U_A}(t) = (\boldsymbol{\pi}^*)^\top e^{\widetilde{\boldsymbol{Q}}_A^* t}\mathbf{1}$ $(t > 0)$, where $\boldsymbol{\pi}^* = (\boldsymbol{\pi}_A^\top \mathbf{1})^{-1}\boldsymbol{\pi}_A$ is the equilibrium distribution of $\{X^*(t)\}$. Arguing as in Section 4.1 now shows that, if $\{X(t)\}$ is time reversible and in equilibrium, then $T_A \mid T_A > 0$ is distributed as a mixture of at most n_A negative exponential random variables, where n_A is the number of states in A. The distribution of the total charge transfer whilst in A during a burst can be determined using a random time transformation of $\{X_M^*(t)\}$, as in Section 4.2; details concerning this distribution are given in Ball et al. [9, Equations (3.13) and (3.14)].

Let N_A denote the number of visits to A during a burst. The distribution of N_A comprises a point mass at zero, given by $P(N_A = 0) = P(T_A = 0)$, and a (possibly defective) distribution on the positive integers. Moreover, similar arguments to those used in Section 4.3 show that if $\{X(t)\}$ is time-reversible and in equilibrium, then $N_A \mid N_A > 0$ is distributed as a mixture of at most rank$(\boldsymbol{P}_A)$ geometric random variables, where $\boldsymbol{P}_A = (-\widetilde{\boldsymbol{Q}}_{AA}^{-1})\widetilde{\boldsymbol{Q}}_{AB}(-\widetilde{\boldsymbol{Q}}_{BB}^{-1})\widetilde{\boldsymbol{Q}}_{BA}$ is the (substochastic) transition matrix for entry states of two successive visits to A during a burst; for details see Ball et al. [9, Equation (3.32)].

5. Concluding remarks

In previous papers, notably Ball et al. [4, 8, 9], we have derived results about ion channel gating behaviour by exploiting structure arising from relevant Markov renewal processes that are embedded in the underlying Markov or Markov renewal process which describes the channel gating. Especially, in [8, 9] the focus was on derivation of burst properties. The present paper has shown that many of the results of those two papers can be obtained much more simply using a suitably marked continuous-time Markov chain which is derived from the assumed underlying continuous-time Markov chain by deleting closed sojourns and concatenating the open sojourns. Other results in those papers, such as the form of autocorrelation functions of burst properties, can also be obtained using the present framework but details are omitted owing to space restrictions. The clarity of the derivations appears to result from them accessing precisely the details of structure which are relevant in each situation, and from exploiting two other aspects. First, the use of excess lifetimes simplifies the derivation of properties of sojourn time pdfs, since they avoid the use of burst entry process equilibrium distributions and consequently lead more directly to mixtures of exponentials. Second, the expression for the burst entry process equilibrium distribution given in Lemma 3.3 (that arises naturally in the present setting) leads to more efficient derivations of mixture properties than in [9]. The approach of the present paper is not readily applicable when knowledge of the time spent in the deleted closed sojourns is required (e.g., in determining the distribution of burst duration). Also, the method is generally less useful in cases where the channel gating behaviour is modelled by a Markov renewal process that is not a continuous-time Markov chain; the concatenated process is a Markov renewal process but its semi-Markov kernel usually does not take a simple form. Concatenated processes may also prove useful in other areas of application of aggregated processes, such as system reliability (cf. Csenki [19]).

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