Membrane channel interactions underlying rat subthalamic projection neuron rhythmic and bursting activity

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Abstract

A computational model of the rat subthalamic nucleus projection neuron is constructed using electrophysiological and morphological data and a restricted set of channel specifications. The model cell exhibits a wide range of electrophysiological behaviours characteristic of rat subthalamic neurons. It reveals that a key set of three channels play a primary role in distinguishing behaviours: a high voltage activated calcium channel (Ca\textsubscript{1.2.-1.3}), a low voltage activated calcium channel (Ca\textsubscript{3.-}), and a small current calcium activated potassium channel (K\textsubscript{Ca}2.1-2.3). Short and long post-hyperpolarisation rebound responses, low frequency rhythmic bursting (<1 Hz), higher frequency rhythmic bursting (4-7 Hz) and slow action and depolarising potentials are behaviours all mediated by the interaction of these channels. This interaction can generate a robust calcium-dependent extended depolarisation in the dendrites (a depolarising plateau). The diversity observed in the rat subthalamic physiology (such as short or long rebounds, or the presence of low frequency rhythmic bursting) can arise from alterations in both the density and distributions of these channel types and, consequently, their ability to generate this depolarising plateau.

A number of important predictions arise from the model. For example, blocking or disrupting the low voltage activated Ca\textsubscript{3.-} calcium current should mute the emergence of rebound responses and rhythmic bursting. Conversely, increasing this channel current via large hyperpolarising potentials in combination with partial blockade of the high voltage activated calcium channels should lead to the more experimentally elusive in vitro high frequency bursting.

Keywords: subthalamic nucleus, basal ganglia, multi-compartmental model, depolarising plateau, high voltage activated calcium channels, parameter optimisation
1 Introduction

The subthalamic nucleus (STN) plays a pivotal role in the dynamics and function of the basal ganglia, a group of subcortical nuclei implicated in a variety of motor, association and limbic functions (Alexander \textit{et al.}, 1990). It is the only glutamatergic nucleus in the basal ganglia, with projections to the substantia nigra reticulata, entopenduncular nucleus (rat analogue to the primate internal segment of the globus pallidus) and globus pallidus (Kita \textit{et al.}, 1983a; Kita and Kitai, 1987; Parent and Smith, 1987; Bevan \textit{et al.}, 1994; Smith \textit{et al.}, 1998). It receives a direct glutamatergic cortical input (Nakanishi \textit{et al.}, 1987; Fujimoto and Kita, 1993; Maurice \textit{et al.}, 1998) and GABAergic input from the globus pallidus (Shink \textit{et al.}, 1996; Bevan \textit{et al.}, 1997; Ryan and Clark, 1991). The STN also receives an input from the substantia nigra containing tyrosine hydroxylase-positive terminals (Hassani \textit{et al.}, 1997; Prensa \textit{et al.}, 2000).

STN projection neurons are considered to have an important role in both the manifestation of key symptoms of Parkinson’s disease (Albin \textit{et al.}, 1989; Crossman, 2000) and in their treatment (Ashkan \textit{et al.}, 2004; Filho \textit{et al.}, 2001). Periodicity in local field potential recordings from the STN is observed to be dominated by two frequencies (6 and 20Hz) in human Parkinson’s patients (Brown \textit{et al.}, 2001). On leva dopa treatment the low frequency components are reduced. Activity in the primate STN is increased in the MPTP (1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine) model of Parkinson’s disease (Delong, 1990; Bergman \textit{et al.}, 1994). This co-occurs with an increase in the STN population of neurons exhibiting slow periodic bursting activity (4-8Hz) (Bergman \textit{et al.}, 1994). In rat, increases in STN neuron activity are also observed in the 6-OHDA (6-hydroxydopamine) model of Parkinson’s disease (Hassani \textit{et al.}, 1996; Kreiss \textit{et al.}, 1997). A move from regular to bursting activity in the STN is also seen in this model (Ni \textit{et al.}, 2001).

Rat STN projection neurons have several distinguishing characteristics. \textit{In vitro}, STN cells fire in a slow rhythmic manner in the absence of external input, yet do not exhibit a sub-threshold oscillation in the absence of action potentials. A persistent sodium current is demonstrated to contribute to this regular rest firing pattern (Bevan and Wilson, 1999; Do and Bean, 2003). In early studies depolarising current applied to continuously hyperpolarised cells revealed both “slow action potentials” and “slow depolarising potentials” (Nakanishi \textit{et al.}, 1987). Both are forms of a plateau potential, where the former consists of an all-or-none calcium dependent slow action potential lasting around 30ms, the latter a depolarisation long outlasting the duration of the depolarising stimulus.
Physiological differences within the rat STN projection neuron population have been recently observed. Releasing a cell from hyperpolarisation (induced either via current injection or GABAergic afferent stimulation) leads to a calcium dependent rebound burst of spikes in a majority of cells (Bevan et al., 2002). This rebound can be either short (<100 ms) or long, often having a duration of many hundreds of milliseconds (Bevan et al., 2002). The duration of the short bursts can be extended with the application of apamin (an antagonist of calcium activated potassium channels).

Sustained hyperpolarisation of a subpopulation of rat STN cells can induce a slow rhythmic bursting (Beurrier et al., 1999). It is argued that constant hyperpolarisation together with the application of apamin is required for the generation of slow rhythmic bursts (Hallworth et al., 2003; Wilson et al., 2004). Similar low frequency rhythmic bursting is also observed with the application of N-methyl-D-aspartate (NMDA) (Wilson et al., 2004; Zhu et al., 2004). Multiple bursting modes have been reported, from pure modes (consisting of long lasting bursts of constant duration) to mixed modes (alternating short and long bursts) (Beurrier et al., 1999). The recorded frequencies of in vitro rhythmic bursting range from 0.1-0.5Hz (Beurrier et al., 1999; Hallworth et al., 2003).

Many of the defining characteristics of the STN neuron are observed under hyperpolarised conditions. Bevan et al. (2002) demonstrate a subset of these key characteristics by inducing inhibitory postsynaptic potentials (IPSP) acting on GABA_\text{A} receptors. As the STN has a large GABAergic input, it is possible that many of these characteristics have a functional role in the information processing of these neurons.

The aim of this paper is to present a new model of single rat subthalamic nucleus neurons that enables us to look at the interactions of key channel types. The model reveals a consistent set of mechanisms underlying the wide range of behaviours exhibited by these neurons. We also use the model to explore how changes in channel density or distribution can lead to diversity in the observed behaviours among STN neurons. The model provides a number of testable predictions arising from the proposed mechanisms.

2 Methods

We construct a multi-compartmental model of the rat STN projection neuron with the following aims:

1. We wish the model to exhibit many of the well known and characteristic features of STN neurons employing a reduced or minimised parameter set. In the outline of the model below, we describe explicit assumptions that are made in order to reduce the model complexity.
2. For many neuron models of this type there is an issue of how to deal with unknown parameter values. Although increasingly the STN is becoming of interest to many research labs, there is still a limited amount of data available. We would like to adopt a method to compensate for absent data which does not degrade our first aim by increasing model complexity through adding arbitrary free parameters.

3. We make explicit the procedure used for choosing specific parameter values in the simulations.

2.1 Morphology

The dendritic fields of most rat STN projection neurons are flat and oval, their long axis lying parallel to the long axis of the nucleus (Afsharpour, 1985). Dendrites can extend over 500 µm, and on average 3-4 primary dendrites extend from the soma. Therefore, it is possible for a dendritic field to extend across 1,000 µm (Kita et al., 1983a). The morphology of the STN cell model is based on schematic trees described by Kita et al. (1983a) and Afsharpour (1985), consisting of a soma and three identical trees (Figure 1). Distal dendrites are reported to have sparse numbers of spines, contributing a small amount to the overall surface area (Afsharpour, 1985). This is incorporated into the morphological model via a small surface area extension in the distal dendrites. The STN cell morphology has an important role in the function of these neurons. Their enormity should allow for a wide collection of potentially diverse cortical and pallidal input (Nambu et al., 1996).

The effects on the electrical properties of the cell by possible volume under- or over-estimation (for example, due to shrinkage in the preparations) are considered below in the derivation of the passive properties. The morphology is split into smaller compartments where the length of each compartment ranges from 5% to 22% of the electrotonic length (the number of compartments consequently ranged from 440 to 95). Spatial simulation accuracy was varied to speed the parameter search procedure.

2.2 Passive Properties

Examples of passive property measurements in rat STN projection neurons are given in Table 1. Extracting passive properties from electrophysiological data is a difficult task (Major et al., 1994; Thurbon et al., 1998). The procedure we follow here is similar to protocols previously reported (Thurbon et al., 1998), and is two pronged. We aim to find a passive model that both is consistent with the properties in Table 1 and exhibits the passive characteristics of transients commonly observed in in vitro intracellular experiments.
(e.g. Nakanishi et al., 1987). It is not possible within the given constraints to determine exact values of the key passive properties of STN cells. However, we can sufficiently represent the passive characteristics of these cells in order to provide a framework to explore the interaction of active properties.

To aid our selection of passive membrane properties we construct an initial model that excludes active channels. The membrane potential of a compartment \( j \) is described by

\[
\frac{d v_j}{dt} = \frac{v_{j-1} - v_j}{r_{j-1,j}} - \frac{v_j - v_{j+1}}{r_{j,j+1}} - I_{\text{ion}} - I_{s_j} \tag{1}
\]

where \( c_{mj} \) is the membrane capacitance for compartment \( j \) (given by the specific membrane capacitance multiplied by surface area, \( C_m A_j \)), \( v_j \) is the membrane potential of compartment \( j \), \( r_{j-1,j} \) and \( r_{j,j+1} \) are the axial resistances between compartments. Axial resistance is given by the specific cytoplasmic resistance (\( R_a \)), length and cross-sectional areas of the joining compartments. This, and Equation 1, take different forms depending on the branching arrangement. \( I_{s_j} \) is the stimulus current applied to compartment \( j \) (usually zero unless the compartment represents the soma). \( I_{\text{ion}} \) is the ionic currents flowing across the membrane of compartment \( j \). In the passive model this is a leak current only (\( I_{L} \)) given by the specific membrane resistance (\( R_m \)), membrane surface area (\( A_j \)) and the reversal potential of the passive channels (\( E_L \))

\[
I_{Lj} = \frac{v_j - E_L}{R_m / A_j} \tag{2}
\]

An additional shunt is added to the soma compartment with the conductance of the shunt given by \( G_{\text{shunt}} \). The shunt represents a change of \( R_m \) at the soma generally influenced by the experimental setup and recording equipment.

We assume a specific membrane capacitance of \( C_m=1 \mu F \ cm^{-2} \). It is likely that the membrane capacitance is slightly different from this value. However, as capacitance and morphology errors trade off with each other we only explore the effects of one of these. Making this assumption, and assuming the morphology given in Figure 1 we have three unknowns: the specific membrane resistance (\( R_m \)), the specific cytoplasmic resistance (\( R_a \)) and a possible shunt at the soma (\( G_{\text{shunt}} \)).

Our procedure is to optimise values of \( R_m \) and \( R_a \) that generate simulation results reproducing passive intracellular data under similar simulated recording conditions (Kita et al., 1983b; Nakanishi et al., 1987; Bevan and Wilson, 1999). Different values of \( G_{\text{shunt}} \) are maintained on a per experiment basis (accounting for potential differences between experimental setups). An error measure can be derived from the comparison of the simula-
tion of the experiments with the published intracellular data. Assuming a particular mor-
phology and for any particular values of $G_{\text{shunt}}$, $R_a$, and, $R_m$ expected values of $R_{\text{in}}$ can also be calculated following the algorithm of Rall (1977). Any difference from the recorded $R_{\text{in}}$ can be added to the accumulated error measure. By minimising the overall error, we select $R_m$ and $R_a$ values that are harmonious both with the passive data and $R_{\text{in}}$ measurements (and providing a estimate of the somatic shunt in each of the experimental setups).

A possible source of error is in the surface area assumed for the morphological model. We allow for this by assuming that such errors are uniform over the morphology. We add a diameter scale factor ($D_s$) to the optimisation procedure above, optimising over four parameters, $R_m$, $R_a$, $G_{\text{shunt}}$ and $D_s$. This generates a passive model that is consistent with recorded passive properties and provides a compensation factor for misestimation of the morphological surface area. The actual minimisation method used in the procedure above is similar to that used in the selection of active property parameters (see Section 2.4 below).

The final passive parameter values are given in Table 2 and values of the diameters in Figure 1. A large somatic shunt was generated in the minimum fit to the Kita et al. (1983b) transients, and is consistent with their low recording of the membrane time constant ($\tau = 6$ ms). A large shunt may reflect the use of sharp electrodes over patch or whole cell recording. The calculated shunt was small in the other experimental arrangements. As we have highlighted, there are inevitably other possible combinations of passive model parameters that we could have chosen. Due to the unconstrained nature of the parameter selection problem, it is difficult to justify this particular set of passive parameters over others. We can compare this parameter set to a set of directly recorded values as in Table 1. Compared with a recorded $\tau$ of 6 ms and $R_a$ of 123 $\Omega$cm (derived from recorded $R_{\text{in}}$, estimated shunt and given morphology) the passive parameters we derived (Table 2) yield an 80% reduction in error. As the error measure is derived from a comparison with recorded passive transients the selected parameter values generate a significantly improved replication of the observed physiological phenomena. The selected parameter set should be considered simply as being consistent with observed STN passive membrane properties.

### 2.3 Active Properties

Active membrane properties are added to the passive model by modifying the membrane ionic currents ($I_{\text{ioni}}$) in Equation 1. In the active model this is given by

$$I_{\text{ioni}} = I_{\text{Na}_j} + I_{\text{KDR}_j} + I_{\text{Kv31}_j} + I_{\text{skCaj}_j} + I_{\text{hj}} + I_{\text{CaTj}_j} + I_{\text{CaLj}_j} + I_{\text{CaNJ}_j} + I_{\text{Lj}}$$

(3)
where the current components correspond to currents through specific channel types as listed in Table 3. Full equations and parameters for these currents are given in Appendix A.

Active membrane properties involve a significantly increased parameter uncertainty compared to the passive properties above. Not only must we identify what channels are likely to underlie the behaviours of the rat STN projection neuron, but we must also address the myriad of parameters describing individual channel kinetics. Taken together with non-uniform distributions of certain channel types over the dendritic tree and the limited data available from the rat subthalamus, this makes the process of modelling the active properties of these cells enormously under-determined.

We could proceed by declaring the key parameters underlying each of the channels kinetic properties as free, and allow ourselves the luxury of choosing parameters that yield the behaviours in which we are interested. This has the limitation that many combinations of parameter values may yield the same behaviours, providing little information about the key channel interactions and phenomena underlying these characteristics.

We take an alternative approach and exploit the fact that a significant number of active channel properties have been characterised in other areas of the rat brain (for example, thalamus and cortex). Assuming that channel properties vary little across brain areas, we use the kinetic parameter values of these channels in the subthalamic model. This assumption certainly may not be correct. For example, channel subunit constituency can vary across structures. However, if we fix the kinetic parameterisation as experimentally recorded for particular channels (apart from $Q_{10}$ temperature adjustments, see Appendix A) then we dramatically reduce the parameter space within which to explore characteristic STN behaviours. It may now be unrealistic to expect a perfect match to distinguishing STN neuron behaviours. However, if we capture the broad classes of observed STN behaviours in the model, we have a better chance of identifying the key active membrane components that underlie these behaviours.

Table 3 lists the nine channel types included in the model. The choice of channels is based on studies revealing certain channel types or classes in the STN (Wigmore and Lacey, 2000; Bevan and Wilson, 1999; Do and Bean, 2003; Song et al., 2000; Baufreton et al., 2003; Zhu et al., 2004). In a similar manner to passive properties, our goal is not to provide a complete and definitive definition of STN active membrane properties, but rather to provide a realistic set of channels within a modelling framework in order to reveal how key STN behaviours can arise from their interaction. Although the kinetic descriptions and properties of each channel are taken from the associated descriptions (see Table 3) we must align the descriptions to compensate for the differences in experimental recording
temperature. We do this by using a combination of restricted voltage shifts of activation/inactivation curves and rate scale factors (reflecting Q_{10} modifications).

The locations and densities of active channels across the membrane can play an important role in the behaviours exhibited by the cell. However, apart from evidence that the low voltage activated calcium channel (CaT) is not located proximally on the STN projection neuron (Song et al., 2000), there is very little data on the distributions of the channels listed in Table 3. Consequently, we choose channel distributions that best fit observed STN behaviours (see the minimisation method below). Where the distribution is found to be critical to specific behaviours in the model, we are making a prediction of the expected arrangements of channels in the STN neuron. Where there is evidence for channel localisation (such as a non-proximal CaT distribution) it is added as a constraint to the model.

We can reduce the number of parameters specifying the distribution by assuming linear distributions over the dendritic tree. This means that we can only capture linear proximal, distal, and uniform distributions rather than modelling possible non-uniformities. This assumption allows the dependence of observed physiological behaviours on channel co-localisation to be exposed. An important side effect of the linearisation is that it biases against central concentrations (biasing toward either distal or proximal distributions). With this assumption the distribution and density of an individual channel may then be modelled using only four parameters: the channel density at the soma; the overall channel density across all the dendritic trees; the amount of density that is uniform across the trees; a single parameter specifying the linear distribution of the remaining density over the trees (ranging between 1, maximally proximal and -1, maximally distal) (Figure 2). In the case of the CaT channel, the parameters governing the distribution are permitted to vary within the non-proximal constraints (i.e. distributions ranging from near uniform excluding the soma to extremely distal are possible). The use of a multi-compartmental model over a single compartment allowed this constraint to be considered.

The final model has 46 free parameters, which is low for a multi-compartmental model. In the following section we describe the search procedure used in selecting values for these parameters.

2.4 Parameter Selection

We begin by selecting a set of characteristic STN properties and use specific experimentally recorded instances of these to compare with the model neuron. The model is simulated under equivalent conditions for each experiment (including artificial cerebral spinal fluid (aCSF) composition, temperature, current injection protocols, etc.) and corre-
sponding measurements are compared with those recorded in the experimental arrange-
ment. An error value is derived from this comparison and model parameters are then ad-
justed so as to reduce this error.

The STN properties used in the definition of the error space are listed here with ex-
ample simulations of the final model shown in Figure 3.

- Action potential properties characterised and recorded by Beurrier et al. (1999). These comprised six properties delineating action potential and after-
hyperpolarisation (AHP) form. [Figure 3A]

- In vitro resting firing patterns recorded by Bevan and Wilson (1999) and Bevan et
al. (2002) consisting of frequency and coefficient of variation measurements at dif-
ferent temperatures. [Figure 3B,C]

- Hyperpolarisation response characterised by Bevan et al. (2002) focusing on the
shape of the “sag” during the hyperpolarisation, and the presence of a rebound re-
response. The form of the post-stimulus response (short or long) is not included in
the error space, rather the presence of an increased post-hyperpolarisation firing
sequence is simply desired. [Figure 3D]

- Passive properties used in the derivation of the passive parameters are also in-
cluded in the error space (although passive parameters are not modified during the
minimisation of active properties). [Figure 3E]

- Finally, after the initial parameter search, an additional component is added to the
definition of the error space characterising the repetitive burst firing during a hyper-
polarising stimulus and simulated apamin protocol. This is compared to observa-
tions of Hallworth et al. (2003) and Beurrier et al. (1999).

In total there are a maximum of 30 error components. Each is a squared error differ-
ence of a specific measured characteristic in simulation and in experiment. For example,
the squared difference of action potential half width from recorded half width is a single
component. The final model error is calculated as the sum of all components.

These properties were chosen to be characteristic of rat STN projection neuron be-
behaviour, while omitting some of the distinguishing STN features, such as the “slow action
potential” reported by Nakanishi et al. (1987), or the short and long post-hyperpolarising
response classifications of Bevan et al. (2002). This allows us to assess the final model through its ability to replicate these features that were not used in the construction of the error space.

The selection of parameter values that minimise the model’s error proceeds using a form of the simulated annealing method (Press et al., 1988). Each parameter is given an upper and lower bound. The starting point of the parameter search is the selection of random parameters within these set bounds. From the initial large parameter set of up to 46 parameters, a random subset of 7 parameters is selected and the model error is minimised with respect to this subset. This is repeated many times with different randomly selected subsets all minimised from the same initial set of parameters. The subset of parameters that produces the lowest error is then used to modify the parameter values to produce a new initial set. This process is then repeated until minimisation of all subsets no longer reduces the error.

Minimising over small subsets of parameters has a two-fold advantage. First, it allows convergence of the simulated annealing procedure within a reasonable time frame. Second, where coordinated parameter changes are required for effective error minimisation, searching over multiple random subsets encourages these to be quickly found. It is difficult to assess the final parameters attained by the minimisation procedure as there is little data with which to compare (particularly in the dendrites). Maximum conductance values are within general biologically realistic regions as a consequence of the upper and lower bounds placed on them in the search procedure. However, we could compare peak isolated currents in the model with equivalently recorded currents. For example, in the final model, the maximum proximal calcium current density is -85µA cm⁻² using a voltage clamp setup (calculated from the soma and dendritic trunks within 50µm). This yields a peak current of -1.6nA which is smaller yet comparable to recordings of peak calcium currents in dissociated STN neurons using a similar protocol (~ -2.4nA, Song et al., 2000). However, this can only be considered as an illustration of the parameters being generally biologically plausible. Making such comparisons is difficult due to the limited availability and nature of the data.

All simulations were performed using the NEURON simulator (Hines and Carnevale, 1997) with the CVODE numerical integration system.

3 Results

The model reproduces the primary features of STN physiology that were used in the construction of the error space (Figure 3, with final parameters given in Appendix A). With
the same parameters it also exhibits a number of key STN properties not included in the error set. These include the slow depolarising (plateau) potential and the slow action potential (see Nakanishi et al., 1987). Using an error set that contains no rhythmic bursting properties the model demonstrates robust rhythmic bursting in the presence of a hyperpolarising current and simulated apamin. Apamin is simulated with a uniform 90% reduction over the neuron morphology of the sKCa conductance. When bursting properties are added to the error set, the final model is found to exhibit two bursting modes: a low frequency mode with frequencies less than 1Hz and a high frequency mode with frequencies between 4-7Hz. The model also reproduces the short and long post-hyperpolarising rebound responses observed in vitro (Bevan et al., 2002; Hallworth et al., 2003). We now characterise these and other key behaviours of the model and highlight the active channel interactions that underlie these behaviours.

### 3.1 The Action Potential and Spontaneous Rest Activity

**Key channels:** Na, KDR, NaP, Kv31

Spontaneous resting activity exhibited in the model (5-15Hz) is critically dependent on the persistent sodium channel (NaP). The persistent sodium current is a major component of the slow ramp depolarisation preceding an action potential. This is consistent with the findings in rat subthalamic projection neurons in vitro (Bevan and Wilson, 1999; Do and Bean, 2003).

The characteristic action potential exhibited by the model (Figure 3A) captures key features of the STN action potential. However, the half height width in the model is overly large (0.98 msec compared with 0.65±0.03 as recorded by Beurrier et al., 1999). This has the consequence of slowing the model down when firing fast. The Kv31 channel plays an important role in reducing the width of the action potential (Figure 3A, dotted line), yet the width is not reduced sufficiently.

### 3.2 The Short and Long Post-Hyperpolarisation Response

**Key channels:** CaT, CaL, sKCa

As mentioned in the introduction, some rat STN neurons exhibit a short post-hyperpolarising rebound response while others exhibit a long rebound burst (Hallworth et al., 2003). We can use the model to explore this diversity.
During the parameter search procedure the reproduction of a rebound response was required to reduce the model error yet the extent of the response was not used as a constraint. The model produced a significant rebound that may be characterised as a long response (>100 msec, see Figure 3D).

Short Rebound Responses

Three channel specific currents play a determining role in this rebound behaviour: the Ca,3.- low voltage activated calcium current (CaT), the Ca,1.2-1.3 high voltage activated calcium current (CaL) and the small calcium activated potassium current (sKCa). It is the balance of the channels’ spatial distributions and conductance levels that determine the nature of the rebound. For example, a reduction (20%) in the CaL-type channel conductance produces a short rebound response (Figure 4A,B). The primary current underlying this short rebound is mediated by the CaT channel (Figure 5A). The shape and time course of the short response is defined by the time course of the CaT current. As the CaT channel is activated from hyperpolarised potentials and inactivates at depolarised potentials, this yields a short post-hyperpolarisation rebound current.

Long Rebound Responses

It is possible to convert a short rebound burst (e.g. Figure 4A) into a long burst by a reduction in sKCa channel conductance (Figure 4C,B) or by an increase in the CaL channel conductance (Figure 4D,B). In all cases the CaT current is necessary for the generation of a rebound response (Figure 5B, diamonds) which is consistent with the calcium dependent nature of the burst. The generation of long bursts under reduced sKCa conductance conditions is comparable with the observation that apamin can extend burst duration in neurons which exhibit short bursts (Hallworth et al., 2003). In the model there is an associated increase in resting firing rate with the simulated apamin condition. This is uncharacteristic of STN neurons where, in the presence of apamin, an increase in coefficient of variation is often observed.

The dendritic CaL current primarily underlies long bursts (Figure 5B). At the soma a transient L-type current occurs during the action potential (Figure 5B, blue dotted line). In the dendrites there is sufficient calcium entry at the initiation of the burst (via the combined CaT and CaL currents) to generate a depolarisation that facilitates prolonged CaL channel activation and an associated depolarising current. This leads to a positive feedback where,
on average, CaL channels remain activated for long periods. The prolonged dendritic depolarisation sustains the long rebound burst (Figure 5B, blue solid line).

The spatial location of these channels over the model neuron is shown in Figure 5C. Each channel has a linearly increasing density with distal location. This spatial relationship also plays a role in the nature of the rebound response. Potassium currents mediated via the sKCa channels limit the rebound burst duration by repolarising the dendrites when internal calcium levels are elevated. This is particularly potent in distal locations where the CaL feedback system underlying extended dendritic depolarisations can significantly increase intracellular calcium levels near the membrane (intracellular calcium concentration and buffering is modelled within a volume immediately below the membrane, see Appendix A). Artificially lowering the sKCa conductance levels leads to a reduced potassium current and longer rebound bursts (Figure 4B, triangles). When the distribution of sKCa channel conductance is shifted to a less distal location, the repolarising effect of the potassium current is again reduced. Although the total level of sKCa channel conductance is constant, there is less conductance concentrated in the distal areas where higher levels of intracellular calcium accumulate. The sKCa and CaL distributions move out of balance and, in this case, the rebound response becomes longer (Figure 4B, squares).

The occurrence of a CaL mediated dendritic depolarisation underlying the long rebound burst is an all-or-none event. The feedback nature of the system generates robust initiation and termination of the dendritic CaL current (Figure 5B). A trigger depolarisation is required for the initiation of the response (mediated here by the CaT current). The termination of the sustained depolarisation occurs when it drops below a critical level necessary for CaL activation (consequently breaking the feedback). As we have seen, different factors can influence when this occurs (such as the level or location of the sKCa potassium current).

### 3.3 Mechanisms of Slow Rhythmic Bursting

**Key channels:** CaT, CaL, sKCa

In the presence of a uniform reduction in the sKCa conductance (simulating the application of apamin) and constant hyperpolarising current injection the model generates slow rhythmic bursting (Figure 6B). Two primary modes of rhythmic bursting are exhibited: a high frequency mode (e.g. Figure 6A) where the frequency lies between 4-7Hz and a low frequency mode (e.g. Figure 6B) yielding a bursting frequency less than 1Hz. A difference
in the form of a single burst is also observed between the two modes. In the high frequency mode, bursts are elevated above the inter-burst potential whereas in the low frequency mode there is no elevation (Figures 6A,B).

The interaction of the CaT and CaL conductances determines the presence and nature of the rhythmic bursting. The sKCa conductance is less involved as it is significantly reduced due to the effects of simulated apamin. Similarly to the post-hyperpolarisation rebounds (Section 3.2), the presence of a sufficient CaT conductance is necessary for the generation of individual bursts. Uniformly reducing the CaT conductance can eliminate rhythmic bursting. Increases in this conductance can lead to a jump from the low frequency to the high frequency mode. This is illustrated in Figure 6C, where bursting is prevented by decreasing CaT levels by 12% and a jump from low to high frequency modes occurs with a 16-20% increase. The role of the CaL current is similar to that in the short/long rebound burst behaviour. In the low frequency mode, increasing the CaL conductance increases the burst duration (Figure 6D, solid line). There is also a decrease in the inter-burst interval (Figure 6D, dashed line) which leads to an overall increase in bursting frequency as the conductance density is increased.

**High Frequency Mode**

The high frequency bursting mode arises from a dominant CaT current. During the inter-burst hyperpolarisation the CaT channels become available for activation. Activation of a small fraction creates a weak current that begins the depolarisation leading to burst initiation (Figure 7B, insert). In a similar manner to long post-hyperpolarisation rebounds, upon sufficient depolarisation the CaL channels become active and generate a depolarising plateau in the dendrites sustaining the burst (Figure 7B). Burst termination occurs once the CaL mediated depolarisation drops below a critical threshold for sustaining the CaL activation feedback (see Section 3.2). The membrane becomes hyperpolarised again as a result of the constant current injection. This allows the CaT channels to become deinactivated, eventually becoming available for activation and the burst cycle repeats. In this mode, the bursting frequency is highly influenced by the dominant role of the CaT channel. Channel characteristics, such as the deinactivation time course (here based in the kinetic description of Wang *et al.,* 1991) influence the resulting frequency (4-7Hz).
Low Frequency Mode

The CaT current plays a different role in the low frequency bursting mode. It is still necessary for rhythmic bursting to occur. However, it does not initiate bursts as in the high frequency mode. The CaL current is responsible for burst initiation while retaining its role in sustaining the burst (and mediating burst duration) via a depolarising plateau. At the end of a burst the membrane potential begins to hyperpolarise due to the constant current injection (Figure 7C). There remains a small but influential CaL current in the dendrites (Figure 7D, insert) as the hyperpolarising current injection is not large enough to completely reverse the dendritic depolarising plateau. Together with a small CaT current (resulting from the fraction of CaT channels that deinactivate during the post-burst hyperpolarisation) this leads to a further depolarisation of the distal dendritic membrane. This is not sufficient to initiate another burst. Instead, a very gradual increase in the CaL current occurs leading to a slowly increasing dendritic depolarisation (Figure 7D, insert). This continues until the majority of CaL currents activate and lead to the initiation of another burst.

Mixed Mode

From the low frequency mode it is possible to jump to a high frequency mode by increasing the amount of CaT current via an overall increase in CaT conductance (as shown in Figure 6C). It should also be possible to increase the post-burst CaT current by further hyperpolarising the inter-burst potential. This would lead to a larger pool of deinactivated CaT channels after a burst and consequently a larger CaT rebound current. Increasing the constant hyperpolarising current injection is one method of increasing the inter-burst hyperpolarisation and thus indirectly increasing the CaT current. This can indeed lead to mode changes in the rhythmic bursting (Figure 8A). For reduced CaL conductance levels no rhythmic bursting is generated with medium current injections (-0.25 – -0.30nA). For larger hyperpolarising currents (e.g. -0.35nA) a high frequency bursting mode emerges. Default and larger values of CaL conductance generate robust low frequency rhythmic bursting from medium current injection levels (Figure 8A, circles and triangles). The frequency of bursting slows as the current injection is lowered (compare frequencies of -0.25 and -0.3nA in Figure 8A). This results from slowing the gradual increase in CaL mediated depolarisation (see Figure 7D insert). As the stimulus hyperpolarises the membrane further the model cell can jump between high and low frequency modes (Figure 8B). The lowered bursting frequency (<4Hz) for -0.35nA current injection in Figure 8A is an artifact.
of mixed modes of bursting. This is reflected in much larger coefficient of variation of burst frequency.

Channel types other than the CaT, CaL or sKCa have less influence on the nature of rhythmic bursting or on post-hyperpolarising rebound responses. Large variations in the distributions and densities of other major channels introduce only relatively small changes in key rhythmic and rebound properties (Figure 9A,B). Other model parameters, such as the time constant of the intracellular calcium buffering model or the maximum calcium inactivation of the CaL channel also play limited roles in these properties (Figure 9A,B, white and solid circles).

3.4 Slow Action and Depolarising Potentials

Key channels: CaT, CaL, sKCa

Both the slow action potential (Figure 10A) and the slow depolarising potential (Figure 10C, left) are caused by the same channel interactions underlying the short and long rebounds (Section 3.2) and rhythmic bursting behaviours (Section 3.3). The CaT current primarily underlies the slow action potential (Figure 10B). CaT channels are made available for activation by the hyperpolarising current injection and activated when there is a reduction in the current injection (see Nakanishi et al., 1987 for the protocol used in the generation of the slow action potential). The short time course of the potential follows the time course of inactivation of the CaT channel. A larger constant hyperpolarising current injection and brief depolarising stimulus is used for the slow depolarising or plateau potential (Nakanishi et al., 1987). This generates a significantly larger CaT current, which is sufficient to trigger a CaL mediated depolarising plateau sustaining the longer depolarising potential (Figure 10D). Membrane hyperpolarisation is important in the generation of these behaviours. Reducing the level of hyperpolarisation eliminates the plateau (Figure 10C, right). This reveals a hyperpolarisation threshold for plateau generation that is consistent with in vitro observations (Otsuka et al., 2001).

4 Discussion

Our model provides insight into the mechanisms underlying the physiology of rat subthalamic nerve cells. A large number of parameter values, such as morphology, passive properties and some channel kinetic properties are obtained from measurements from the rat STN. Other channel kinetic parameter values are obtained from measurements
from other brain regions. This produces a model with a restricted parameter set and generic channel behaviours. The remaining parameter values are selected using an error space defined over a range of rat STN recordings, from different laboratories and under different experimental conditions. Some of the channel interactions may reflect similar interactions underlying the dominant STN behaviours. The use of this type of model allows us to identify key channels and their cooperation in emergent behaviours to provide predictions of similar arrangements within STN projection neurons.

The parameter search procedure yields a model that captures a remarkable array of STN characteristics. The choice of parameters produces dendritic distributions of the high voltage activated Ca_{1.2-1.3} (L-type) channels and the small calcium activated potassium channels. A dendritic distribution of the low voltage activated Ca_{3.-} (T-type) calcium channel is imposed on the model (following observations of Song et al., 2000). This spatial arrangement creates a dendrite-bound mechanism that plays a key role in the generation of a wide range of behaviours. In particular, it is responsible for the CaL dependent depolarising plateau. Long depolarising plateaus are observed in STN neurons (Nakanishi et al., 1987; Otsuka et al., 2001). A sufficiently large and long trigger depolarisation can initiate a CaL current that sustains the initial depolarisation. This membrane depolarisation in the dendrites in turn extends the activation of CaL channels by holding the voltage above their activation threshold, creating a positive feedback. As CaL channels are modelled with a slow calcium mediated inactivation (Meuth et al., 2002), the resulting depolarising plateau is able to sustain long bursts of action potentials at the soma. The model exhibits a similar mechanism underlying the plateau to that described by Otsuka et al. (2004). In our model, the primary mechanism is located in the dendrites as a consequence of imposing a dendritic constraint on the location of the CaT channels. In the model of Otsuka et al. (2004), a single compartment is used enforcing co-localisation of the key channel types (CaL, CaT, sKCa). We use a systematic parameter selection procedure within a multi-compartmental framework which creates a similar co-localisation in the model presented. In addition, our model demonstrates the role that a depolarising plateau may play in a wide range of characteristic STN behaviours, from rebound responses to rhythmic bursting.

**Interactions between the CaT, CaL and sKCa channel types**

The balance in location and density between the three channel types CaL, CaT, and sKCa is critical in determining the presence and nature of this depolarising plateau. For example, the variability in the observed nature of post-hyperpolarisation rebound bursts (short or long) in the rat STN could reflect differences in channel location and/or density.
The CaT channel is required for the rebound burst to occur and provides the link between hyperpolarising stimuli, which are necessary in many of the observed behaviours, and the range of emergent responses. The dendritic location of this channel type makes its influence on the dendrite-bound plateau particularly potent. In the model it is extremely difficult to generate a plateau from localised depolarisation alone (either in the soma or dendrites). However, under hyperpolarising conditions, depolarisation, or release from the hyperpolarisation, can easily elicit a plateau. This is consistent with plateaus elicited from hyperpolarised conditions demonstrated by Otsuka et al. (2001). The dendritic and dispersed nature of the CaL mediated mechanism makes plateau generation resistant to localised excitatory initiation. The co-localisation of these two channels facilitates robust plateau initiation under hyperpolarising conditions. Sufficiently large CaT mediated currents can provide a trigger depolarisation to initiate a dendritic CaL mediated plateau. If it is not sufficiently large, the CaT mediated response may remain the dominant observation (as seen in the “slow action potential”). Increasing CaT or CaL channel densities, reducing sKCa density, and shifting the sKCa or CaL channel distributions can all facilitate the emergence of depolarising plateaux. Large reductions in the density of CaT channels can abolish rebound responses completely. Together these three channels provide a sufficient (although not necessarily unique) parameter set to explain the diversity of rebound responses seen in the STN (Hallworth et al., 2003). Figure 11 illustrates the key channel interactions.

The linear distribution of conductances that is assumed only allows a loose assessment of the coupling of different channel types. The linear simplification itself biases toward distal distributions. For the minimisation procedure to generate peaks of channel concentration it can only shift concentrations proximally or distally. The CaT constraint biases toward the distal shift. However, the parameter optimisation procedure maintains a high level of coupling or co-localisation of the sKCa, CaL and CaT channels. For example, the co-localisation of the sKCa and CaL channels is consistent with the observations that nifedipine (an L-type channel antagonist) has little effect on small rebound responses and significantly reduces long rebound responses (Hallworth et al., 2003). In the model, small rebound responses are generated largely by the CaT-type channel and we would expect nifedipine to have little effect on this response. In the long rebound responses, nifedipine would be expected to seriously disrupt the CaL mediated dendritic plateau by indirectly increasing the dominance of the co-localised sKCa channel and consequently reduce the response.
Rhythmic Bursting

Rat STN neurons can rhythmically burst with very low frequency (Beurrier et al., 1999). This is observed in vitro under hyperpolarising conditions and in the presence of apamin (Wilson et al., 2004; Hallworth et al., 2003). The model robustly exhibits slow rhythmic bursting in the presence of simulated apamin and hyperpolarising current injection (Figure 6B). It can also generate a high frequency bursting mode with frequencies ranging from 4-7Hz. Unlike the high frequency mode, where the time constants of the CaT deinactivation are a key determinant of burst frequency, there are no time constants in the equations of the model that are of the correct order to underlie the low frequency bursting. There is a different dynamic in the interaction of these key channels in this low frequency mode. The CaL channel not only sustains the depolarising plateau, but also is responsible for burst initiation. If the hyperpolarising influence (e.g. from the current injection) is not sufficiently effective in the dendrites, a dendritic sub-threshold depolarisation can be maintained in the inter-burst interval. This creates a slow and creeping feedback system. The resulting dendritic depolarisation is small and only able to activate a small proportion of CaL channels. However, their activation adds to the depolarisation and the depolarising process continues slowly. At a critical threshold this dendritic depolarisation initiates a somatic burst. This slow inter-burst feedback process is robust to parameter variability (e.g. see Figures 6C,D and 9B) and can last for many seconds (with bursting frequencies <0.1 Hz). The dendritic nature of the CaL distribution consequently has a significant impact on low frequency bursting. The separation of the dendritic depolarisation from the soma facilitates the extended interval between somatic bursts by delaying engagement of the sodium mechanisms concentrated at the soma.

Deviations from experimental observations

Some behaviours exhibited by the model differ from experimental observations. For example, the width of a single action potential is notably larger than recorded experimentally (see Section 3.1). This may arise from a non-optimal parameter selection (e.g. it may be possible to reduce the action potential width via changes in the Kv3.1 channel density or distribution) or, alternatively, the channel kinetic description may be inaccurate. Another deviation from experimental observation is a small sub-threshold membrane oscillation observed during low frequency inter-burst intervals (compare with Beurrier et al., 1999). This occurs immediately after a burst and is generated from a CaT current rebound arising from the post-burst hyperpolarisation. Similarly to the action po-
tential form above, it may be possible to adjust parameters to reduce or eliminate this phenomenon. Such deviations are expected given the constraints imposed to reduce the number of free parameters (e.g. fixing kinetic descriptions).

As mentioned above, the linear distribution of channel densities places a distal bias on the location of the mechanisms underlying the plateau potentials. Plateau potentials can be observed in dissociated STN cells (Do and Bean, 2003). The linear constraint can only reveal clustering and co-localisation of channel types and not the physical location due to this bias.

The demonstration of strong coupling between N-type calcium channels and sKCa channels (using the N-type antagonist ω-conotoxin GVIA, see Hallworth et al., 2003) is not observed in the model. This is likely to be due to the error space used in the selection of parameters insufficiently constraining the N-type channel distribution.

Predictions

The model provides a number of decisive predictions for the nature of these cells. It has been experimentally demonstrated that modification of the sKCa or CaL channel potency can influence the length of post-hyperpolarising rebound responses. Increases in CaL or decreases in sKCa effective densities should extend short responses. Moreover, in the model the coupling of these two channel types is also related to the length of rebound response. If it is the distribution, rather than density, that governs the observed rebound variations in the rat STN neuron population, the model predicts that STN neurons that exhibit short responses should also exhibit a stronger coupling between sKCa and CaL channel types. Conversely, STN neurons exhibiting long rebound responses have a weaker coupling between these channels.

The CaT channel is necessary in the model as a trigger of many of the behaviours. This is compatible with the majority of behaviours examined emerging from hyperpolarising stimuli. Blocking or disrupting the CaT current should mute the emergence of rebound responses and rhythmic bursting. Conversely, increasing the CaT potency should lead to the emergence of a high frequency bursting mode (4-7Hz). This range of rhythmic bursting frequency is not readily observed in in vitro arrangements, but can be observed in STN populations in animal models of Parkinson's disease. It may be possible to generate such a rhythmic bursting mode in vitro with large hyperpolarising stimuli and reduced (although not eliminated) CaL channel potency.

In summary, we have presented a model built on basic principles and assumptions that provides an explanation for some of the key behaviours exhibited in rat STN projec-
tion neurons. The behaviours emerge from only a small set of generic channel interactions which together can bring about a dendrite-bound depolarisation capable of sustaining long bursts. Variations in these channel densities and distributions may underlie the variations in stimulus responses observed within the rat STN population. The nature and construction of the model framework is readily extensible and able to accommodate new data from the rat STN as it is available. The feedback from experimental data and testing of model predictions is a vital component of the model development process.
References


Shink E, Bevan M, Bolam JP, and Smith Y. The subthalamic nucleus and the external pallidum: two tightly interconnected structures that control the output of the basal ganglia in the monkey. *Neuroscience* 73: 335-357, 1996.


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A Model Specification

The voltage of each compartment, $j$, with membrane surface area $A_j$, is given by Equation 1. This includes the sum of all ionic currents passing through the membrane ($I_{ionj}$) as specified in Equation 3. Equations 1 and 3 are reproduced from the main text:

$$c_{mj} \frac{dv_j}{dt} = \frac{v_{j-1} - v_j}{r_{j-1,j}} - \frac{v_j - v_{j+1}}{r_{j,j+1}} - I_{ionj} - I_{sj}$$

(1)

$$I_{ionj} = I_{Na_j} + I_{KDR_j} + I_{Kv31j} + I_{skCa_j} + I_{h_j} + I_{CaT_j} + I_{CaL_j} + I_{CaN_j} + I_{L_j}$$

(3)

The kinetics of each ionic current component are defined from data of the associated channel types in specific cells. The origin of the data, equations, parameters, and $Q_{10}$ temperature adjustments are all listed here. $Q_{10}$ values are used to modify the rate of kinetic equations (see Hille, 2001, for a good description of $Q_{10}$ and its origin).

**Na, NaP** fast acting sodium channel and persistent sodium.

$$I_{Na_j} = (g_{Na_j} m(v_j)^2 h(v_j) + g_{NaP_j})(v_j - E_{Na})$$

(4)

Equations for the activation and inactivation functions ($m(v_j)$ and $h(v_j)$) respectively are given in Traub et al. (1991) and reproduced below. They are modified for temperature alignment using a $Q_{10}$ of 1.98 and 1.5 respectively. The equations are based on electrophysiological data from Sah et al. (1988b). Persistent sodium is modelled as above, with voltage dependent activation variants taken from data of Do and Bean (2003).

$$\dot{m}(v_j) = a_m(v_j)(1 - m(v_j)) - \beta_m(v_j)m(v_j) \quad \dot{h}(v_j) = a_h(v_j)(1 - h(v_j)) - \beta_h(v_j)h(v_j)$$

$$a_m(v_j) = 0.32 \frac{(13.1 - v_j)}{\exp((13.1 - v_j)/4) - 1} \quad a_h(v_j) = 0.128\exp((17 - v_j)/18)$$

$$\beta_m(v_j) = 0.28 \frac{(v_j - 40.1)}{\exp(v_j - 40.1) - 1} \quad \beta_h(v_j) = \frac{4}{\exp((40 - v_j)/5) + 1}$$

**KDR** potassium delayed rectifier.

$$I_{KDR_j} = g_{KDR_j} n(v_j)(v_j - E_K)$$

(5)

Equations for the activation function ($n(v_j)$) are given in Traub et al. (1991) and reproduced below. They are modified for temperature alignment using a $Q_{10}$ of 1.2.

$$\dot{n}(v_j) = a_n(v_j)(1 - n(v_j)) - \beta_n(v_j)n(v_j)$$
\[
\alpha_n(v_j) = \frac{0.016(35.1 - v_j)}{\exp((35.1 - v_j)/5) - 1}
\]
\[
\beta_n(v_j) = 0.25\exp((20 - v_j)/40)
\]

**Kv31** Kv3.1 fast rectifier.

\[
I_{Kv31j} = g_{Kv31j}p(v_j)(v_j - E_K)
\]
\[
\dot{p}(v_j) = \frac{p_\infty(v_j) - p(v_j)}{\tau_p(v_j)}
\]

The activation and time constant functions (given below) are based on data from Wigmore and Lacey (2000). Temperature alignment modifications using a \(Q_{10}\) of 1.7 were then made.

\[
p_\infty(v_j) = \frac{1}{1 + \exp(-(v_j + 5)/9)}
\]
\[
\tau_p(v_j) = \frac{18.71}{\exp(-(v_j + 28)/6) + \exp((v_j + 4)/16)}
\]

**sKCa** small calcium activated potassium channel.

\[
I_{sKCa} = g_{sKCa}w(v_j)(v_j - E_K)
\]
\[
\dot{w}(v_j) = \frac{w_\infty(v_j) - w(v_j)}{\tau_w(v_j)}
\]

The activation and time constant functions (given below) are based on data from Hirschberg et al. (1998). Temperature alignment modifications using a \(Q_{10}\) of 1.5 were then made.

\[
w_\infty(v_j) = \frac{0.81}{1 + \exp(-\log([Ca^{2+}]) + 0.3)/0.46}
\]
\[
\tau_w(v_j) = 40
\]

**Ih** HCN channel.

\[
I_{ih} = g_{ih}f(v_j)(v_j - E_{HCN})
\]

Equations and data for the activation function \(f(v_j)\) are given in Huguenard and McCormick (1992) and reproduced below. They are modified for temperature alignment using a \(Q_{10}\) of 2.0.

\[
f_\infty(v_j) = \frac{1}{1 + \exp((v_j + 75)/5.5)}
\]
\[
\tau_j(v_j) = \frac{1}{\exp(-14.59 - 0.086v_j) + \exp(-1.87 + 0.0701v_j)}
\]

\textbf{CaT} Low voltage activated calcium channel.

\[ I_{\text{CaT}j} = g_{\text{CaT}j} r(v_j)^3 s(v_j) \xi(v_j) \]  \hspace{2cm} (9)

where \( g_{\text{CaT}j} \) is calculated as an effective conductance (from the uniform membrane permeability \( \rho_{\text{CaT}j} \) of compartment \( j \)) and \( \xi(v_j) \) is derived from the Goldman-Hodgkin-Katz (GHK) equation:

\[ \xi(v_j) = \frac{v_j [\text{Ca}^{2+}]_i - [\text{Ca}^{2+}]_o \exp(-zv_j F / RT)}{[\text{Ca}^{2+}]_o \left( 1 - \exp(-zv_j F / RT) \right)} . \]  \hspace{2cm} (10)

\( z \) is the ion valence (2 in this case), \( F \) is Faraday’s constant, \( T \) is temperature (in Kelvin), \( R \) is the gas constant. The relationship between \( \rho_{\text{CaT}j} \) and \( g_{\text{CaT}j} \) is given by

\[ g_{\text{CaT}j} = \rho_{\text{CaT}j}^2 \frac{F^2}{RT[\text{Ca}^{2+}]_o} . \]  \hspace{2cm} (11)

This specification of \( I_{\text{CaT}j} \) reduces to the GHK current equation (modified by \( r(v_j)^3 s(v_j) \) in the Hodgkin and Huxley manner), however is convenient for comparing conductance levels.

Equations for the activation (\( r(v_j) \)) and inactivation (\( s(v_j) \), \( d(v_j) \)) functions and electrophysiological data are given in Wang et al. (1991) and Coulter et al. (1989) and reproduced below. Temperature alignment modifications using a \( Q_{10} \) of 1.52 were then made.

\[ r(v_j) = \alpha_r(v_j)(1 - r(v_j)) - \beta_r(v_j)r(v_j) \]
\[ s(v_j) = \alpha_s(v_j)(1 - s(v_j)) - \beta_s(v_j)s(v_j) \]
\[ d(v_j) = \beta_d(v_j)(1 - s(v_j)) - \alpha_d(v_j)d(v_j) \]

\[ \alpha_r(v_j) = \frac{1}{1.7 + \exp(-(v_j + 28.2)/13.5)} \]
\[ \beta_r(v_j) = \frac{\exp(-(v_j + 63)/7.8)}{1.7 + \exp(-(v_j + 28.8)/13.5)} \]
\[ \alpha_s(v_j) = \exp(-(v_j + 160.3)/17.8) \]
\[ \beta_s(v_j) = (\sqrt{0.25 + \exp((v_j + 83.5)/6.3)} - 0.5)\exp(-(v_j + 160.3)/17.8) \]
\[ \alpha_d(v_j) = \frac{1 + \exp((v_j + 37.4)/30)}{240(0.5 + \sqrt{0.25 + \exp((v_j + 83.5)/6.3)})} \]
\[ \beta_d(v_j) = (\sqrt{0.25 + \exp((v_j + 83.5)/6.3)} - 0.5)\alpha_d(v_j) \]

\textbf{CaN, CaL} High voltage activated calcium channels.
\[ I_{CaL} = g_{CaL}(v_j)^2 h([Ca^{2+}]) \xi(v_j) \]  
\[ I_{CaN} = g_{CaN}(v_j)^2 u(v_j) \xi(v_j) \]  

where \( \xi(v_j) \) is defined in Equation 10 above, and \( g_{CaL} \) and \( g_{CaN} \) are interpreted in a similar manner to \( g_{CaTj} \) above. Common activation kinetics (\( q(v_j) \)) are given in Brown et al. (1993), CaN inactivation (\( u(v_j) \)) is from (Fox et al., 1987) (given below). The calcium mediated inactivation (\( h([Ca^{2+}]) \)) of the CaL channel is based on data from (Meuth et al., 2002) and given by

\[
\begin{align*}
\dot{h}([Ca^{2+}]) &= \frac{h_{\infty}([Ca^{2+}]) - h([Ca^{2+}])}{\tau_h([Ca^{2+}])} \\
\tau_h([Ca^{2+}]) &= 0.53 + \frac{0.47}{1 + \exp((-0.7)/0.15)} \\
h_{\infty}([Ca^{2+}]) &= 1220
\end{align*}
\]

A model Q_{10} of 1.95 was applied.

\[
\begin{align*}
\dot{q}(v_j) &= \frac{q_\infty(v_j) - q(v_j)}{\tau_q(v_j)} \\
\dot{u}(v_j) &= \frac{u_\infty(v_j) - u(v_j)}{\tau_u(v_j)} \\
q_\infty(v_j) &= \frac{1}{1 + \exp(-24.6 - v_j)/11.3} \\
\tau_q(v_j) &= \frac{1.25}{\cosh(-0.03(v_j + 37.1))} \\
u_\infty(v_j) &= \frac{1}{1 + \exp((v_j + 60)/12.5)} \\
\tau_u(v_j) &= 98 + \cosh(0.021(10.1 - v_j))
\end{align*}
\]

[Ca^{2+}], Intracellular calcium.

Intracellular calcium levels were modelled in a sub-membrane shell with buffering and diffusion modelled as an exponential decay.

\[
[Ca^{2+}]_i = (I_{CaL} + I_{CaNj} + I_{CaTj}) c \left[ \frac{[Ca^{2+}]_i - [Ca^{2+}]_0}{\tau_{Ca}} \right]
\]

where \( c \) is the conversion constant, \( \tau_{Ca} \) is the time constant of the decay and \([Ca^{2+}]_0 \) is the basal intracellular calcium level. A value of 185.7 msec for \( \tau_{Ca} \) was used in the simulations as determined from the parameter search procedure.

Conductance levels for each channel are specified below. Each channel is described by four parameters specifying the distribution over the cell morphology (see Figure 2). Absent parameter values in the table indicate they were not modelled and excluded from the parameter search procedure. For example, constant rather than linear distributions were modelled for the sodium channels.
<table>
<thead>
<tr>
<th>Conductance</th>
<th>Soma (mS.cm⁻²)</th>
<th>Tree (mS.cm⁻²)</th>
<th>Basal (mS.cm⁻²)</th>
<th>Proximity</th>
</tr>
</thead>
<tbody>
<tr>
<td>( g_{Na} )</td>
<td>1.48e-2</td>
<td>.</td>
<td>1.00e-7</td>
<td>.</td>
</tr>
<tr>
<td>( g_{NaP} )</td>
<td>1.11e-5</td>
<td>.</td>
<td>8.10e-6</td>
<td>.</td>
</tr>
<tr>
<td>( g_{KDR} )</td>
<td>3.84e-3</td>
<td>9.32e-5</td>
<td>4.22e-5</td>
<td>-0.05</td>
</tr>
<tr>
<td>( g_{KV31} )</td>
<td>1.34e-2</td>
<td>1.00e-3</td>
<td>8.91e-4</td>
<td>0.80</td>
</tr>
<tr>
<td>( g_{sKCa} )</td>
<td>6.84e-5</td>
<td>3.92e-5</td>
<td>.</td>
<td>-0.52</td>
</tr>
<tr>
<td>( h )</td>
<td>1.01e-3</td>
<td>5.10e-4</td>
<td>.</td>
<td>-0.39</td>
</tr>
<tr>
<td>( g_{CaT} )</td>
<td>0.00</td>
<td>1.67e-3</td>
<td>1.17e-3</td>
<td>-0.01</td>
</tr>
<tr>
<td>( g_{CaN} )</td>
<td>1.15e-3</td>
<td>4.79e-4</td>
<td>.</td>
<td>0.50</td>
</tr>
<tr>
<td>( g_{CaL} )</td>
<td>9.50e-4</td>
<td>1.87e-3</td>
<td>1.21e-4</td>
<td>-0.57</td>
</tr>
</tbody>
</table>
Figure 1: Final morphological specification used in the simulation of the rat subthalamic projection neurons. This includes a shrinkage factor which scales the diameters of the soma and each branch calculated during the fitting procedure for passive properties.
Figure 2: A schematic illustration of the four parameters (shown here as A-D) used to specify the density and distribution of a single channel type over the model neuron. The first parameter, A, gives the density at the soma. The second parameter, B, gives the overall density to be distributed across the dendritic trees. Parameter C specifies the proportion of this density that is uniformly distributed across the trees. Parameter D ranges from -1 to +1 and specifies how the remaining density is distributed. +1 indicates an extreme proximal distribution, -1 an extreme distal distribution. The value zero for this parameter yields a uniform distribution over the tree. The example above approximates a value of 0.2 for this parameter which generates a gentle proximal distribution.
Figure 3: Simulation of the main STN cell behaviours used in the error space. A. Properties of a single resting action potential (AP) and afterhyperpolarising potential (AHP). Properties were compared with and aCSF conditions from Beurrier et al. (1999). The dashed line shows an action potential with no Kv31 conductance. B, C. Spontaneous rest activity at 25 °C and 35 °C respectively. Simulations were run under the aCSF conditions of and properties were compared with Bevan and Wilson (1999). D. Generation of the post-hyperpolarisation response (induced with a 500msec -0.25 nA soma current injection). Simulation conditions from and properties compared with Bevan et al. (2002). E. Passive properties. In this simulation the model includes the simulated soma shunt derived from the passive data of Nakanishi et al. (1987).
Figure 4: Short and long post-hyperpolarising rebound responses. A 500msec hyperpolarising current injection of -0.25nA is simulated to generate the post-stimulus rebound burst in all simulations. A. Short rebound response (burst duration <100 msec). In this simulation dendritic linear CaL conductance is reduced by 20%. B. The influence of changes in the dendritic CaL conductance density (-50% to +50%) on rebound burst duration (circles). Due to the scale of the plot the very short rebound (<20 msec) is not visible at < -30% reduction in CaL density. With a uniform reduction in sKCa conductance (20%) there is an increase in burst duration over all CaL density variations (triangles). A change in the distribution of the sKCa channel (30% less distal) also leads to increased rebound burst duration (squares). A uniform reduction (50%) in the CaT conductance eliminates all rebound bursting (diamonds). C. Parameters as in A (i.e. to generate a short rebound response) produce a long (150 msec) rebound burst in the presence of simulated apamin (uniform 40% reduction in sKCa channel conductances). D. Long rebound response (burst duration 460msec). In this simulation dendritic linear CaL conductance is increased by 50%.
Figure 5: A. Currents underlying the short post-hyperpolarising rebound response (simulated experimental setup as in Figure 4B, with 50% reduction in CaL conductance to generate a short rebound burst). Records begin 1msec before termination of the hyperpolarising stimulus. Distal (solid lines) and soma (dotted lines) specific membrane currents mediated through the CaL (blue), CaT (red), and sKCa (green) channel types are shown. The locations of current recordings are indicated by arrows in the schematic morphology in C (distal - solid arrow, soma - dotted arrow). B. Currents underlying the long rebound response (simulated experimental setup and parameters as in Figure 4D). Colours and line types are the same as in A. C. Three schematics of the model neuron morphology showing in a colour hotscale the distributions of CaT, CaL, and sKCa channel densities (hotscale units S/cm²).
Figure 6: Rhythmic bursting. A. High frequency rhythmic bursting. The model with a 12% reduction in dendritic linear CaL conductance and a hyperpolarising current injection of -0.35nA. In this example burst frequency is 4.8Hz. In all rhythmic bursting the same simulated apamin conditions are maintained (90% reduction in sKCa conductances). B. Low frequency bursting exhibited by the model with a hyperpolarising current injection of -0.25nA. In this example the frequency is 0.53Hz. C. The influence of changes (-30% to 30%) in the low threshold calcium CaT conductance on the rhythmic bursting frequency (note, the frequency is plotted on a logarithmic scale). Constant hyperpolarising current used -0.25nA. D. The influence of dendritic CaL conductance changes on the burst lengths (solid line) and inter burst lengths (dashed line) during low frequency rhythmic bursting. In C and D the greyed region for each line shows the coefficient of variation of the rhythmic bursting at each point (with a scale bar insert).
Figure 7: Mechanisms underlying the high and low frequency rhythmic bursting. **A.** Two bursts in a high frequency rhythmic bursting pattern generated as in Figure 6A. **B.** Currents underlying the high frequency rhythmic bursting shown in A. Distal dendritic CaL currents are shown in blue and CaT currents in red. The dendritic location is the same as used in Figure 5. Soma currents of each are shown with dotted lines using the same colour code. The insert rescales the currents at a single burst (the insert time bar is 50 msec). The shaded background highlights events during the burst. **C.** Low frequency rhythmic bursting. A current injection of -0.25nA and simulated apamin are used in this example. **D.** Currents underlying the low frequency rhythmic bursting. Colours and line types as in B. The insert rescales the currents during a burst (the time bar is 250msec) with shaded areas as in B.
Figure 8: Rhythmic bursting. A. The influence of changes in the dendritic CaL conductance density (-15% to 15%) on burst frequency. The frequencies at three hyperpolarising current injections are shown (-0.25nA circles, -0.30nA triangles, -0.35nA diamonds). The greyed region for each line shows the coefficient of variation of the rhythmic bursting at each point (with a scale bar insert). B. Intermixed high and low frequency modes. The model has a 10% increase in dendritic linear CaL conductance and a hyperpolarising current injection of -0.35nA.
Figure 9: The effects of changes in major parameters on rebound and bursting properties. The parameters include the time constant of exponential intracellular buffering, densities and distributions of the CaN, Ih, and Kv31 channels and the maximum levels of CaL calcium inactivation. Each case shows the effects of varying the parameter values between -50% and +50% of its default value. **A.** Effects on rebound burst duration. The rebound is generated as described in Figure 4. **B.** Effects on rhythmic bursting frequency. The rhythmic bursting is generated as described in Figure 6B. Note, in the comparison with Figures 6C and 8A, the scale here is not logarithmic in order to differentiate the lines. The key shown in **A** is common to both plots.
Figure 10: Slow action and depolarising potentials. A. An all-or-none slow action potential is generated from a reduced constant hyperpolarising current (Nakanishi et al., 1987). B. CaL (blue), CaT (red) and sKCa (green) currents during the generation of the slow action potential. Distal dendritic currents are shown in solid lines and soma currents in dotted lines. The locations of current recordings are indicated by arrows in Figure 5C. C. Upon brief stimulation during a constant hyperpolarising current pulse the model exhibits a slow plateau potential which long outlasts the stimulation (Nakanishi et al., 1987). Adjacent is a demonstration of the threshold hyperpolarisation level for the plateau to be induced (Otsuka et al., 2001). D. The same currents as in B recorded during the generation of the slow depolarising (plateau) potential.
**Figure 11**: Schematic of key dendritic channel interactions. Solid colours illustrate channel activity. Red is the CaT channel, green the sKCa channel and blue the CaL channel. **A.** CaT dominated response. The voltage overlay plot is an example of the slow action potential (fast action potentials truncated). The sKCa channel activity prevents the resulting CaT current from initiating a CaL mediated plateau. **B.** CaT current triggering the CaL mediated plateau. In this example, the sKCa channel activity is reduced (e.g. by the application of apamin). The voltage overlay is an example of a burst in the high frequency rhythmic bursting mode.
Table 1: Examples of Recorded Passive Properties.

**Input Resistance ($R_{in}$)**

<table>
<thead>
<tr>
<th>Resistance (MΩ)</th>
<th>[N=7, range=9:28]</th>
<th>rat in vitro</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>18±6</td>
<td></td>
<td></td>
<td>Kita et al. (1983b)</td>
</tr>
<tr>
<td>146±48</td>
<td>[N=26]</td>
<td></td>
<td>Nakanishi et al. (1987)</td>
</tr>
<tr>
<td>200.2±6.8</td>
<td>[N=88]</td>
<td></td>
<td>Beurrier et al. (1999)</td>
</tr>
</tbody>
</table>

**Membrane Time Constant (τ)**

<table>
<thead>
<tr>
<th>Time Constant (ms)</th>
<th>[N=7, range=4:9]</th>
<th>rat in vitro</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>6±2</td>
<td></td>
<td></td>
<td>Kita et al. (1983b)</td>
</tr>
</tbody>
</table>
**Table 2**: Final calculated passive properties

<table>
<thead>
<tr>
<th>Property</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Membrane time constant</td>
<td>12.8 ms</td>
</tr>
<tr>
<td>Capacitance</td>
<td>1.0 $\mu$F cm$^{-2}$</td>
</tr>
<tr>
<td>Membrane resistance</td>
<td>12,753 $\Omega$cm$^2$</td>
</tr>
<tr>
<td>Input resistance</td>
<td>146.5 M$\Omega^*$</td>
</tr>
<tr>
<td>Cytoplasmic resistance</td>
<td>150.2 $\Omega$cm</td>
</tr>
</tbody>
</table>

* Calculated using soma shunt estimated from the Nakanishi *et al.* (1987) data.
**Table 3:** Active channel properties and their descriptions, the brain area where the properties were recorded and temperatures used in the experimental setup. Additional abbreviations, LGN lateral geniculate nucleus, TCR thalamocortical relay, DRG dorsal root ganglion. See Appendix A for a fuller specification of the modelling of the active channels.

<table>
<thead>
<tr>
<th>Current</th>
<th>Family</th>
<th>Description/Origin</th>
<th>Area</th>
<th>°C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Na</td>
<td>Na,1.-</td>
<td>Fast acting sodium channel (Traub <em>et al.</em>, 1991; Sah <em>et al.</em>, 1988b)</td>
<td>CA1</td>
<td>22-24</td>
</tr>
<tr>
<td>NaP</td>
<td>Na,1.-</td>
<td>Persistent sodium channel (Bevan and Wilson, 1999)</td>
<td>STN</td>
<td>25</td>
</tr>
<tr>
<td>KDR</td>
<td>K,2.1</td>
<td>Delayed rectifier (Traub <em>et al.</em>, 1991; Sah <em>et al.</em>, 1988a)</td>
<td>CA1</td>
<td>22-24</td>
</tr>
<tr>
<td>Kv31</td>
<td>K,3.1</td>
<td>Fast rectifier (Wigmore and Lacey, 2000)</td>
<td>STN</td>
<td>32</td>
</tr>
<tr>
<td>sKCa</td>
<td>K_{Ca}2.1-2</td>
<td>Small conductance calcium activated potassium channel (Hirschberg <em>et al.</em>, 1998)</td>
<td>STN</td>
<td>22-24</td>
</tr>
<tr>
<td>Ih</td>
<td>HCN</td>
<td>Hyperpolarisation-activated cation channel (Huguenard and McCormick, 1992)</td>
<td>LGN</td>
<td>35.5</td>
</tr>
<tr>
<td>CaT</td>
<td>Ca,3.-</td>
<td>Low voltage activated calcium channel (Wang <em>et al.</em>, 1991)</td>
<td>TCR</td>
<td>22-24</td>
</tr>
<tr>
<td>CaN</td>
<td>Ca,2.2</td>
<td>High voltage activated calcium channel (Brown <em>et al.</em>, 1993; Fox <em>et al.</em>, 1987)</td>
<td>DRG</td>
<td>22</td>
</tr>
<tr>
<td>CaL</td>
<td>Ca,1.2-1.3</td>
<td>High voltage activated calcium channel (Brown <em>et al.</em>, 1993; Fox <em>et al.</em>, 1987; Meuth <em>et al.</em>, 2002)</td>
<td>DRG/LGN</td>
<td>22</td>
</tr>
</tbody>
</table>