Role of Precise Spike Timing in Coding of Dynamic Vibrissa Stimuli in Somatosensory Thalamus

Marcelo A. Montemurro,1 Stefano Panzeri,1 Miguel Maravall,2 Andrea Alenda,2 Michael R. Bale,1 Marco Brambilla,1 and Rasmus S. Petersen1

1Faculty of Life Sciences, University of Manchester, Manchester, United Kingdom; and 2Instituto de Neurociencias de Alicante, Universidad Miguel Hernández–Consejo Superior de Investigaciones Científicas, Alicante, Spain

Submitted 25 May 2007; accepted in final form 30 July 2007

Montemurro MA, Panzeri S, Maravall M, Alenda A, Bale MR, Brambilla M, Petersen RS. Role of precise spike timing in coding of dynamic vibrissa stimuli in somatosensory thalamus. J Neurophysiol 98: 1871–1882, 2007. First published August 1, 2007; doi:10.1152/jn.00593.2007. Rats discriminate texture by whisking their vibrissae across the surfaces of objects. This process induces corresponding vibrissa vibrations, which must be accurately represented by neurons in the somatosensory pathway. In this study, we investigated the neural code for vibrissa motion in the ventroposterior medial (VPM) nucleus of the thalamus by single-unit recording. We found that neurons conveyed a great deal of information (up to 77.9 bits/s) about vibrissa dynamics. The key was precise spike timing, which typically varied by less than a millisecond from trial to trial. The neural code was sparse, the average spike being remarkably informative (5.8 bits/spike). This implies that as few as four VPm spikes, coding independent information, might reliably differentiate between 106 textures. To probe the mechanism of information transmission, we compared the role of time-varying firing rate to that of temporally correlated spike patterns in two ways: 93.9% of the information in the spike trains could be decoded even if temporal correlations were ignored. Taken together, these results suggest that the essence of the VPm code for vibrissa motion is firing rate modulation on a submillisecond timescale. The significance of such a code may be that it enables a small number of neurons, firing only a few spikes, to convey distinctions between very many different textures to the barrel cortex.

INTRODUCTION

Rats can distinguish objects that differ only in micron-scale surface texture using their mystacial vibrissae (Carvell and Simons 1990). Texture discrimination behavior is mediated by barrel cortex (Guic-Robles et al. 1992), implying that neural responses at each stage of the subcortical somatosensory pathway must preserve a great deal of information about the complex vibrissa vibration patterns (“kinetic signatures”) that distinguish different textures (Arabzadeh et al. 2005; Neimark et al. 2003).

The ventroposterior medial nucleus (VPM) of the thalamus is the principal gateway by which these signals reach the barrel cortex. The VPM is composed of barreloids, each of which contains approximately 250 neurons (Haidarliu and Ahissar 2001; Land and Simons 1985; Sugitani et al. 1990; van der Loos 1976). Because, in some cases, rats can discriminate between objects using only a single vibrissa (Carvell and Simons 1995; Hutson and Masterton 1986), the implication is that each VPm neuron (or a substantial fraction of them) must use a powerful neural code, capable of transmitting a considerable amount of texture information.

What is the nature of this thalamic neural code? The simplest possibility is that it is based on spike count (the number of spikes evoked in a time interval of, for example, 100-ms duration). In such a code, many different temporal patterns of spikes consist of the same number of spikes, but such differences are assumed to reflect random variation rather than coding. Alternatively, the VPm code may be based on precise spike timing. Because the number of possible spike patterns increases exponentially with spike timing resolution, a spike timing code can transmit information at a much higher rate than a spike count code (MacKay and McCulloch 1952; Rieke et al. 1997). This suggests that a spike timing code might be better suited to the demands of VPm texture coding.

That precise spike timing may be important for coding is consistent with known anatomy and physiology. The projection from the trigeminal complex appears to be optimized for temporal fidelity. Trigemino-thalamic fibers make multiple synaptic contacts onto the proximal dendrites or somas of relay cells (Spacek and Lieberman 1974; Williams et al. 1994). As a result, temporally isolated vibrissa deflections evoke large, fast, and reliable excitatory postsynaptic potentials (Brechtt and Sakmann 2002; Castro-Alamancos 2002; Deschenes et al. 2003), which trigger phasic firing with low trial-to-trial variability in timing (Diamond et al. 1992; Simons and Carvell 1989).

The aim of our study was to investigate the hypothesis that VPm uses a spike timing code at the functional level. To this end, we recorded responses of VPm neurons to white noise vibrissa movement. The advantage of this stimulus is that it efficiently presents an unbiased sample of many different vibrissa movement sequences. We then investigated the neural code using a new information theoretic estimation technique (Montemurro et al. 2007). This approach permits the information that spike times convey to be quantified in a very general way, without ad hoc assumptions about precisely which stimulus features (position, velocity, etc.) drive the neuronal response. In this way, we quantified and compared the information...
tion available in the precise timing of spikes with that available in the spike count.

To gain additional insight into the neural code in VPm, we also examined the mechanisms used to convey information. The simplest possibility (rate coding) is that different vibrissa movements evoke different firing rates (Ahissar et al. 2000; Bialek et al. 1991; Panzeri et al. 2001; Petersen et al. 2001; Richmond et al. 1987). However, thalamic neurons have well-known physiological properties (burst firing and refractoriness) that introduce temporal correlations into their spike trains. It has been suggested that correlated spike patterns are crucial for neural coding (Abeles et al. 1993; Gray et al. 1989). We compared the relative roles of firing rate and temporal correlations using an information theoretic approach (Nirenberg et al. 2001; Pola et al. 2003; Schneidman et al. 2003).

We found that VPm neurons conveyed information about vibrissa motion at remarkably high rates, for which submillisecond precision spike timing was crucial. We found the time-dependent firing rate to play an important role in the neural code, whereas we did not find a significant role for temporal correlations. Taken together, our results indicate that the essence of the VPm code for vibrissa motion is firing rate modulation on a submillisecond timescale.

METHODOLOGIES

Electrophysiology

All experiments were conducted in accordance with international and institutional standards for the care and use of animals in research. Adult Wistar rats (n = 16) were anesthetized with urethane (1.5 g/kg body weight) and placed in a stereotaxic instrument. A craniotomy was made 2.0–4.5 mm posterior to bregma, 1.5–4.0 mm lateral to bregma, and the dura reflected. A tungsten microelectrode (8 MΩ at 1 kHz; FHC, Bowdoin, ME) was lowered vertically into the cerebrum (mean subpial depth 5,400 μm, SD 260 μm) using a customized piezoelectric motor (Lambda Photometrics, Harpenden, UK). Extracellular signals were preamplified, digitized (sampling frequency 24.4 kHz), band-pass filtered (second-order Butterworth, 300–3,000 Hz), and continuously stored to hard disk for off-line analysis (TDT, Alachua, FL).

At each recording site, the center receptive field (CRF) was identified by deflection of the individual vibrissae. Only units with CRFs including one or more of E1–4, D1–4, C1–4, γ, and δ were considered. Vibrissae contralateral to the recorded hemisphere (E1–4, D1–4, C1–4, γ, and δ) were cut to 10-mm length and individually placed into the holes of a Plexiglas grid, glued to a piezoelectric multilayer bender (Lambda Photometrics). The grid was positioned 3 mm from the skin. Motion of the actuator was in the ventrodorsal direction.

The stimulus was a 15-s sequence of pseudorandom white noise with Gaussian amplitude distribution, low-pass filtered by convolution with a Gaussian kernel (SD 1.6 ms) to restrict stimulus power to frequencies less than the resonant frequency of the mechanical stimulator (300 Hz). The stimulus was generated at a sampling frequency of 12.2 kHz and was repeated 100 times.

Stimuli were converted to a 0- to 10-V signal by a digital signal processor (TDT) and amplified to 0–60 V by an amplifier designed to drive piezoelectric loads (Lambda Photometrics). The resulting dynamic range of the stimulator was 400 μm. We verified that the piezoelectric bender accurately reproduced the white-noise stimulus by measuring its motion using a custom-built LED-phototransistor circuit.

Location within VPm was verified electrophysiologically during the experiment (Diamond et al. 1992) and checked by histological identification of the recording site (Yu et al. 2006). Recording sites were marked by electrolytic lesions: 5–10 μA for 20 s at 50 kHz (Adams and Horton 2006). After perfusion with 10% formalin, sites were identified by staining 50-μm coronal sections with cresyl violet.

Data analysis

Neural events were detected by thresholding the microelectrode signal: 1- to 2-ms segments of the signal were extracted around the time of each threshold crossing. Spikes corresponding to a given single unit were isolated by clustering in the space of two to four principal components either by maximum likelihood fitting of a Gaussian mixture model or by using the SAC algorithm (Shoham et al. 2003). Only units whose interspike interval and autocorrelation statistics exhibited a refractory period were considered for further analysis.

Jitter analysis

It is apparent from the raster plots of Fig. 1 that the neuronal response consisted of a series of well-defined episodes in which spikes were fired at very similar times on different trials. To quantify the timing precision of these episodes, we measured the trial-to-trial variability in spike timing (“jitter”) for each single unit as follows. First, we divided the 15-s time course of the stimulus into 15,000 1-ms time bins and averaged across trials to compute the firing rate in each bin. Firing episodes corresponded to local peaks in the firing rate. We identified a unit’s maximum firing rate and selected all peaks whose rate was at least a fraction f of the maximum. For each such peak, we extracted all spikes fired within ±2 ms of the time of the peak and computed spike times relative to that (Fig. 2, A and B, inset). We pooled these time differences across all trials and peaks and defined the neuron’s jitter as their SD. The distribution of time differences was approximately Gaussian (Fig. 2, A and B). We found that jitter results were robust to changes in f in the range 0.4–0.6. The values in the text are for f = 0.5.

Experimental limits on spike timing precision

Because we found spike timing in VPm to be very precise, it is interesting to consider how the observable precision was constrained by experimental conditions—in particular, by the low-pass filtering and sampling procedures. Under our conditions, most of the power of the action potential waveform was at around 1,000 Hz, below the cutoff of the low-pass filter (3,000 Hz). Thus action potential shape was only weakly affected by low-pass filtering. To be more precise, we computed the impulse response function of our Butterworth filter. The resulting deviations in spike timing due to filtering and sampling were summarized by measuring its motion using a custom-built LED-phototransistor circuit.

For each unit, we computed a normalized autocorrelogram averaged over the 100 trials as follows. First, for each trial, we binned the spike count into 0.04-ms intervals. In sum, we conservatively estimate that the joint effect of sampling and filtering under our conditions was a spike timing error of about 0.1 ms.

Autocorrelation analysis

For each unit, we computed a normalized autocorrelogram averaged over the 100 trials as follows. First, for each trial, we binned the
spike train at resolution $\Delta t = 0.25$ ms, yielding the binary response function $n(t)$, where $t$ is the time after onset of trial $\eta$; $n(t) = 1$ if there was a spike in the interval $(t, t + \Delta t)$ on trial $\eta$ and $n(t) = 0$ otherwise. Second, we constructed a shifted response function $n(t) = n^{\eta}(t)$. We then computed the autocorrelation function $C(\tau)$ using the following expression 

$$C(\tau) = \frac{\langle n(t) n(t + \tau) \rangle - \langle n(t) \rangle \langle n(t + \tau) \rangle}{\langle n(t) \rangle \Delta t},$$

Here the averages $\langle \cdot \rangle$ and $\langle \cdot \rangle$ are over all possible time bins and trials, respectively. The first term in the numerator is the average number of spike pairs separated by a time lag $\tau$ ms. The second term (the symmetrized shift predictor) is the average number of spike pairs, separated by a lag $\tau$, predicted to occur purely due to firing rate modulation. Defined in this way, the autocorrelation has units of spikes/s.

The burstiness of a cell is associated with positive peaks in the autocorrelation (compare the tonically firing cell of Fig. 4A to the bursty cell of Fig. 4B). To quantify the degree of bursting exhibited by a given cell, we integrated its autocorrelation over lags in the range $t_1$–$t_2$ ms. Formally

$$i_{\text{burst}} = \int_{t_1}^{t_2} C(\tau) d\tau$$

where $i_{\text{burst}}$ is expressed in units of spikes. For the data reported in RESULTS, $t_1 = 2$ ms and $t_2 = 4$ ms. The mean of $i_{\text{burst}}$ across units was 0.019 ± 0.040 (means ± SE) spikes. This means that, during the 2- to 4-ms period after a spike, 0.019 spikes were fired, on average.

**Information theoretic analysis**

The main goal of this study was to analyze how thalamic neurons encode complex vibrissa motion. Our approach was to quantify and compare the information conveyed about the stimulus by different possible coding mechanisms. We did this by applying information theory. First, we describe how we estimated the total information a neuron conveyed about the stimulus. Then we describe how we estimated the role of specific coding mechanisms (firing rate modulations and spike correlations).

The aim of the analysis was to quantify the extent to which different sensory stimuli (vibrissa trajectories) evoked distinct neuronal responses. To this end, on each trial, we measured the response $r$ of a neuron within time windows of length $T$ ms. At each time $t = 0, T, 2T, \ldots$ (trial onset was $t = 0$), the spike count response was the number of spikes fired in the window $(t, t + T)$. To obtain the spike timing response at time resolution $\Delta t$ ms, the window was subdivided into $L = T/\Delta t$ bins of duration $\Delta t$. These parameters were selected to make the number of bins at most 12 (see Sampling bias). Here, $r = [r_1, r_2, \ldots, r_L]$, where $r_i$ is the number of spikes evoked in $i$th bin. The set of possible spike timing responses was therefore a set of “words” of length $L$.

We defined $P(r | t)$ as the probability of response $r$ at time $t$, for each possible $r$, and $P(r) = \sum_i P(r | t_i)$, where $\langle \cdot \rangle$ denotes averaging over time. For a stimulus-sensitive neuron, the time-conditional distribution $P(r | t)$ varies greatly with time. For a stimulus-insensitive neuron, $P(r | t)$ does not vary with time and is therefore equal to $P(r)$.

Mutual information (called “information” in the following) quantifies how stimulus sensitive a neuron is by measuring how much, on average, $P(r | t)$ differs from $P(r)$ (de Ruyter van Steveninck et al. 1997; Shannon 1948)

$$I = \sum_r P(r | t) \log_2 \frac{P(r | t)}{P(r)}.$$  

For a randomly firing neuron, $P(r | t) = P(r)$ for all $t$, the argument of the logarithm is 1 and $I$ is therefore 0. In contrast, when $P(r | t) \neq P(r)$, the argument is nonzero and $I$ is positive. $I$ has units of bits. Its value depends on both the bin size $\Delta t$ and the response time window size $T$. It is important to note that the information we compute concerns all possible kinetic attributes that may activate a neuron and makes no assumption about which particular attributes to which it might be selective.

We also measured the “information rate,” defined as the limit of $I(T)$ for $T$ going to infinity, measured in bits per second. In practice, we first computed $I$ as a function of $T$ and checked that the increase was linear with time. Our measure of information rate was then the linear coefficient (slope).

Different spike patterns occur at different times not only because different stimuli elicit different responses but also because of noise (fluctuations in the response to repetitions of the same stimulus). The total variability of the response due to both sources is quantified by the response entropy $H(R)$

$$H(R) = - \sum_r P(r) \log_2 P(r) \quad (2a)$$

The variability arising specifically from noise is quantified by the noise entropy $H(R | S)$

$$H(R | S) = - \sum_r P(r | t) \log_2 P(r | t) \quad (2b)$$

Information is defined as the amount of response variability left over after noise has been taken into account: $I = H(R) - H(R | S)$.

**Information conveyed by temporally correlated spike patterns**

The aim of the present study was not simply to quantify the information transmitted by a neuron but also to measure how much could be attributed to different coding mechanisms. A simple type of neural code is one where all stimulus information is conveyed by modulations of firing rate and temporal correlations between spikes are irrelevant. In general, however, correlated spike patterns can play a significant role.

To discriminate between these possibilities, we first define the concept of correlation between spikes. A (spike) timing response $r$ is “uncorrelated” if $P(r | t) = P(r)$ for all $t$. If this is not the case, the response is “correlated.” Correlations in this sense have been termed “noise correlations” because they describe correlations in the variability of the responses to repeated presentation of the same stimulus (Gawne and Richmond 1993).

To assess whether temporal correlations increase or decrease the information available in the neuronal response, we compared the information conveyed by the spike train of each recorded neuron to that of a hypothetical neuron with identical time-varying firing rate but no noise correlations (Hatsopoulos et al. 1998; Nirenberg and Latham 1998; Pola et al. 2003; Schneidman et al. 2003). The latter quantity, $I_{\text{rand}}$, is measured by substituting $P_{\text{rand}}(r | t)$ for $P(r | t)$ in Eq. 1.

$$I_{\text{rand}} = \sum_t P_{\text{rand}}(r | t) \log_2 \frac{P_{\text{rand}}(r | t)}{P_{\text{rand}}(r)} \quad (3)$$

$H_{\text{rand}}(R)$ and $H_{\text{rand}}(R | S)$ are defined by applying the same substitutions to Eqs. 2a and 2b. The difference between $I$ and $I_{\text{rand}}$—termed $I_{\text{corr}}$—quantifies whether the presence of noise correlations increases or decreases the information available in the neuronal response, compared with the case where such correlations are absent but the time-dependent firing rate is the same

$$I = I_{\text{rand}} + I_{\text{corr}} \quad (4)$$

In terms of entropies, $I_{\text{corr}}$ can be expressed as
It is also useful to analyze the role of correlations by explicitly considering the perspective of a decoder. The measure $\Delta I$ (Nirenberg et al. 2001)

$$\Delta I = \sum_t P(r(t) \mid S) \log \frac{P(r(t) \mid S)}{P(r(t) \mid R)}$$

also known as $I_{\text{corr-dep}}$ (Pola et al. 2003), is an upper bound on the information lost if a downstream decoder ignores correlations (Latham and Nirenberg 2005). For discussion of the relationship between the different information theoretic measures of correlation, see Latham and Nirenberg (2005), Pola et al. (2003), and Schneidman et al. (2003).

**Sampling bias**

Information measures typically suffer from bias due to the probabilities $P(r \mid t)$ being sampled from a limited number of trials $N$ (Panzeri and Treves 1996). To estimate the spike count information, we corrected for bias using an extrapolation procedure (Strong et al. 1998). To estimate the spike timing information, we used the bias-reduction procedure of Montemurro et al. (2007). Most of the bias comes from $H(R \mid S)$ rather than $H(R)$, given that $H(R \mid S)$ depends on many more parameters (Eqs. 2a and 2b). The Montemurro et al. method reduces the bias of $H(R \mid S)$ as follows. The quantity $H_{\text{ind}}(R \mid S)$ is computed, as defined earlier, through estimation of $P_{\text{ind}}(r \mid t)$. However, an estimate of $H_{\text{ind}}(R \mid S)$ can also be obtained by a shuffling procedure where, at each $t$, pseudoresponse vectors $r$ are created by randomly permuting bins across trials. The resulting estimate $H_{\text{sh}}(R \mid S)$ has the same value as $H_{\text{ind}}(R \mid S)$ for infinite $N$, but has much higher bias for any given finite $N$. In fact, the bias of $H_{\text{sh}}(R \mid S)$ is similar to that of $H(R \mid S)$, but slightly larger (Montemurro et al. 2007). This observation motivates the following formula for estimating $I$

$$I_{\text{sh}} = H(R) - H_{\text{ind}}(R \mid S) + H_{\text{sh}}(R \mid S) - H(R)$$

For infinite $N$, $H_{\text{ind}}(R \mid S) = H_{\text{sh}}(R \mid S)$, so $I_{\text{sh}} = I$ (Eqs. 1 and 2). For finite $N$, the bias of $I$ is reduced due to cancellation between the bias of $H(R \mid S)$ and that of $H_{\text{ind}}(R \mid S)$. Moreover, because the bias of $H_{\text{sh}}(R \mid S)$ is (more) negative than that of $H(R \mid S)$, the residual bias of $I_{\text{sh}}$ is negative. Finally, the bias was further reduced by quadratic extrapolation to infinite $N$ (Strong et al. 1998). The bias of $\Delta I$ was reduced (and made slightly negative) in a similar way (Montemurro et al. 2007). The SE of $\Delta I$ was estimated by a bootstrap procedure.

We tested the reliability of the information estimates by simulation, systematically varying both word length and bin size. For the present data set with $N = 100$, reliable information estimates could be obtained for response words of up to length $L = 12$. For any given $L$, there was a mild tendency for reliability to increase with bin size. The reason was that information increased with bin size, whereas bias, which depends mostly on $L$, varied little (at fixed $L$). Thus the information/bias ratio was greater for larger bins. Performance was accurate across the entire range of bin widths considered in this study (0.5–10 ms).

**Reliability analysis**

To study the effect of temporally correlated spike patterns on response reliability, we conducted the following analysis. First, we measured the spike count response on every trial in windows ($t$, $t + T$) for $t = 0, T, 2T, \ldots$ as described earlier. To quantify the reliability, for each $t$, we computed unbiased estimates of the variance of the response across trials $V(t)$ and of its mean $M(t)$. For a neuron that fires each spike independently (Poisson process), the Fano factor $V(t)/M(t) = 1$ (see Rieke et al. 1997) and, in a scatterplot of $V(t)$ against $M(t)$ as in Fig. 4, C and D, the points will follow the main diagonal, albeit with a degree of scatter due to finite sampling.

When the response is measured in a time window short enough so that $M(t) \ll 1$, it can be proved that $V(t) > M(t)$ implies positive autocorrelation (averaged over the time window $T$) and $V(t) < M(t)$ implies negative autocorrelation (Panzeri et al. 1999).

As reported in results, we typically found many points for which $V(t) < M(t)$. Because the most evident type of negative autocorrelation in our data was at time lags of 0–2 ms, the likely source of this effect was refractoriness. To test this, for each recorded neuron, we simulated the response of a matched, hypothetical unit that fired spikes independently except that it was constrained to have an absolute refractory period equal to that of the real neuron (typically between 1 and 1.5 ms). We did this (similar to Berry and Meister 1998; Reich et al. 1998) by generating spikes according to an inhomogeneous Poisson process with firing rate equal to that of the actual neuron, modified so that, after each spike, the firing rate was set to zero for a period equal to the refractory period. In this way, the simulated unit had a time-varying firing rate identical to that of the real one and the only source of noise correlations in its spike train was refractoriness. To compare the simulated data to the recorded data, we computed the bias factor in each time window, as defined earlier, for both real and simulated responses and compared them using a two-sample $t$-test.

**RESULTS**

Figure 1, B and C shows the spikes fired by two representative single units in response to 100 repetitions of the white-noise whisker stimulus shown in Fig. 1A. Both units were highly responsive. Their activity consisted of brief episodes of high firing rate whose constituent spikes were precisely aligned across trials. Two such episodes are shown in Fig. 2, A and B (inset). To identify the spike timing precision, we measured the differences in spike time across trials within each episode (METHODS). For unit 1, the SD of these differences (“jitter”) was
0.36 ms (Fig. 2A); for unit 2 it was 0.35 ms (Fig. 2B). The mean jitter across our sample of 31 neurons and across all high firing rate episodes was 0.43 ms (SD 0.05 ms). These data indicate that VPm relay cells can respond reliably to white-noise vibrissa deflection and that the spikes can be timed with submillisecond precision.

Spike timing versus spike count

The potential significance of precise spike timing is that it permits a neuron to transmit information efficiently, at a high rate (MacKay and McCulloch 1952; reviewed by Rieke et al. 1997). To test whether this mechanism might operate in VPm, we compared the stimulus information conveyed by the times of spikes (spike timing code) to that conveyed simply by the number of spikes (spike count code). For each trial, we measured the neuronal response in windows of duration \( T \) ms starting at \( t = 0, T, 2T, \ldots \) ms post stimulus onset. For the spike count code, the response was the number of spikes fired within the window. For the spike timing code, we subdivided the window into bins of length \( \Delta t \) ms and registered whether each of the \( T/\Delta t \) bins contained a spike. The number of bins was varied in the range 1 to 12. To quantify the stimulus information conveyed by each type of code, we used Shannon’s \textit{mutual information} (see Methods). Figure 3A shows information as a function of window size for the two units illustrated in Fig. 1 (\( \Delta t = 5 \) ms). When the response window was narrow (\( T = 5 \) ms, one bin), the spike timing and spike count information were equal (0.062 bits for unit 1, 0.060 bits for unit 2). As the response window widened, the information conveyed by spike timing remained approximately constant, but that conveyed by spike timing steadily increased. For 60-ms windows (12 bins), spike timing conveyed 10.5 times more information than spike count for unit 1 and 5.2 times more for unit 2.

To test whether this result was typical, we repeated the comparison for all units. Figure 3B shows information as a function of response window duration, averaged over all units in our sample. Because the timing information increased with window size, whereas spike count information remained approximately constant, the timing advantage increased considerably with window size (Fig. 3B). At \( T = 60 \) ms, on average, spike timing conveyed 9.3 times more information than spike count (\( \Delta t = 5 \) ms). These data indicate that, by precise spike timing, considerable stimulus information is available in the responses of VPm neurons.

The preceding analyses were performed at a fixed bin size of 5 ms. However, use of finer temporal resolution has the potential to increase dramatically the capacity of neurons to transmit information. Therefore our next aim was to assess the temporal precision of the code. To do so, we varied the bin size. Figure 3A illustrates that information increased linearly with time: the rate of increase was 12.3 bits/s for unit 1 and 12.8 bits/s for unit 2 (at \( \Delta t = 5 \) ms). Because this “information rate” was independent of both time window duration and the number of bins, it was a useful way of quantifying the response. We consistently found information to increase linearly with time for all bin sizes examined (0.5–10 ms); Fig. 3, C and D shows results for 0.5-ms bins. We found that information rate was strongly dependent on bin size (Fig. 3, E and F). For unit 1, information rate increased from 6.2 bits/s at 10-ms-bin resolution to 35.8 bits/s at 0.5-ms-bin resolution; for unit 2 from 7.8 to 26.9 bits/s (Fig. 3E). This behavior was consistent across our sample (Fig. 3F): information rate continued to increase with bin resolution right up to 0.5 ms (the highest we could reliably measure). The combination of high spike timing precision, high instantaneous firing rate, and a linear increase of information with time meant that the neurons conveyed information at a remarkably high rate. The maximum information rate across the sample was 77.9 bits/s; the mean, 26.5 bits/s (SD 19.3 bits/s). Given that the mean neuronal firing rate was 5.8 spikes/s (SD 3.6 spikes/s), these data imply that each spike conveyed a considerable amount of information—the mean was 5.3 bits/spike (SD 2.6 bits/spike). Thus spike timing differences of as little as 0.5 ms affect the amount of stimulus information encoded by VPm neurons, and the precision of the spike timing code is submillisecond. The significance of such a high precision temporal coding scheme may be that it enables a relatively small number of thalamic neurons to accurately encode complex sensory signals.

Firing rate versus temporal correlations

What might be the coding mechanism by which precisely timed spikes convey stimulus information? The simplest possibility consistent with the preceding results is that neurons transmit information by modulating their firing rate on a fast timescale. This implies that each spike in a spike train is fired independently. However, it is well established that this is not generally the case. Refractoriness prevents the occurrence of interspike intervals of \( \leq 2 \) ms and, when thalamic neurons fire in burst mode, the rate of spike intervals of about 4 ms is enhanced. These phenomena were evident in neuronal autocorrelation functions (Fig. 4, A and B). Both units 1 and 2 showed strong negative autocorrelation at \( 0–2 \) ms, reflecting refractoriness. The degree of bursting varied across cells. Unit 1 is an example of a neuron recorded in tonic mode (Fig. 4A). Unit 2 is an example recorded predominantly in burst mode.
exhibiting a clear peak of positive autocorrelation at about 4 ms (Fig. 4B).

The presence of such temporal correlation structure in the spike trains shows that firing rate by itself is an incomplete description of how VPm neurons respond to dynamic vibrissa stimulation. This raises the possibility that correlated spike patterns, such as those induced by refractoriness and/or bursting, may be important to how neurons transmit sensory information. Our next aim was to test this temporal correlation coding hypothesis.

A fundamental aspect of a neural code is its reliability—the variability of a neuron’s response over repeated presentations of the same stimulus. Theory suggests that reliability can be strongly influenced by correlations between spikes (METHODS). If a neuron fires spikes independently (a Poisson process), the variance of the spike count in a given time window will be equal to its mean. Such behavior has been reported in visual cortex (see Gershon et al. 1998). Spike correlations can make the neuronal response either more or less variable than a Poissonian one (Abbott and Dayan 1999; Oram et al. 1998; Panzeri et al. 1999). Under the conditions of low spike count observed here, negative correlations tend to decrease variability (spike count variance less than spike count mean) and positive correlations to increase it (variance greater than mean) (METHODS).

We measured both the mean and variance of the spike count in 20-ms windows starting at times 0, 20, 40, . . . ms with respect to the onset of the white-noise stimulus. For unit 1, the spike count variance tended to be lower than the mean (Fig. 4C). The simplest explanation is that refractoriness induced negative correlations between spikes, and that this promoted reliability. To test this, we simulated spikes from a hypothetical unit with identical time-varying firing rate to unit 1 and a refractory period of 1.5 ms (METHODS). If the variance–mean relationship of the simulation matched that of the real data, this would suggest that refractoriness is a sufficient explanation for the sub-Poisson variability of unit 1. Consistent with this, we found the simulated variance–mean relationship (Fig. 4E) to be similar to that of the real data (Fig. 4C) and the means of their respective Fano factor distributions to be statistically indistinguishable (t-test, $P = 0.34$).

In addition to negative correlations at 0–2 ms, the autocorrelation function of unit 2 exhibited positive correlations at about 4 ms. Thus one might expect the unit’s responses to exhibit a mixture of super-Poisson and sub-Poisson variability. Consistent with this, the spike count variance of unit 2 was sometimes less than the mean but, at other times, greater than the mean (Fig. 4D). The sub-mean variance could be accounted for by a simulated refractory neuron (Fig. 4F), but the super-mean variance could not. For unit 2, this resulted in the means
of the simulated and actual Fano factor distributions being significantly different \((P < 10^{-6})\). These data indicate that temporally correlated spike patterns have a significant impact on VPm response reliability. Refractoriness induced negative correlations into a neuron’s spike train, and this tended to reduce the variability of its firing. For neurons such as unit 1, this appeared to be the major type of correlation effect. For other neurons, typified by unit 2, there were also positive correlations due to bursting; here, correlated spike patterns had a more complex effect on response variability.

A further implication of these data is that the response of VPm neurons to dynamic vibrissa deflection cannot be fully described by temporal modulations of the firing rate—temporally correlated spike patterns also have an effect. However, what is the relative importance of firing rate and correlations to coding stimulus information? We examined this question by quantifying the effect of temporal correlations in two ways. First, we measured the information \((I)\) that each VPm neuron conveyed about the white-noise stimulus (METHODS) and compared it to the information conveyed by a hypothetical neuron with identical time-varying firing rate but no correlations between spikes \((I_{\text{ind}})\). If \(I\) were substantially greater than \(I_{\text{ind}}\), this would show that the presence of temporal correlations increases the information conveyed by the neural code. To this end, we measured the difference \(I_{\text{cor}} = I - I_{\text{ind}}\).

For unit 1 (Fig. 5A), the firing rate information \(I_{\text{ind}}\) was very similar to the spike train information (12.4 vs. 12.3 bits/s); \(I_{\text{cor}}\) was negligible (−0.1 bit/s). For unit 2 (Fig. 5B), the firing rate information was also large (11.8 vs. 12.9 bits/s), although the unit did exhibit a significant, albeit still small, \(I_{\text{cor}}\) (1.1 bits/s). On average (Fig. 5C), firing rate accounted for 93.9% of the spike train information, \(I_{\text{cor}}\) for 6.1%. For 42% of neurons \(I_{\text{cor}}\) was <5% of \(I\); for 23% of neurons \(I_{\text{cor}}\) was >10% of \(I\). Thus the presence of correlated spike patterns made the neuronal response more informative than it would have been in their absence, although the difference was small. This is strong evidence against the hypothesis that the functional role of temporal correlations is to substantially increase information and suggests that firing rate modulations are the key coding mechanism.

A second way to quantify the effect of spike correlations is to consider the perspective of the downstream barrel cortical circuits. How much stimulus information is lost by a decoder that considers only firing rate and ignores temporal correlations? The measure \(\Delta I\) is an upper bound on this information loss (Latham and Nirenberg 2005). For unit 1, we found that \(\Delta I\) was statistically indistinguishable from zero (−0.08 bits/s, SE...
0.26 bits/s); for unit 2, it was 0.73 bits/s, SE 0.17 bit/s. The very slight negative value of $\Delta I$ for unit 1 was a residual effect of bias correction; see METHODS.) On average, $\Delta I$ was $0.85 \pm 0.15$ bit/s, corresponding to 6.6% of the total information ($T = 60$ ms, $\Delta t = 5$ ms). The implication of this result is that a downstream neural system can decode most of the information available in the afferent spike train without taking correlations into account. Thus barrel cortical neurons might decode the VPM signal in a fairly simple, yet efficient, manner based purely on time-dependent firing rate.

The preceding results were robust to the precise timescale of the analysis: we computed $\Delta I$ and $I_{\text{cor}}$ in the range $\Delta t = 0.5$–8 ms and always found them to be small (data not shown). We also found a strong correlation relationship between $I_{\text{cor}}$ and $\Delta I$ across cells (Pearson correlation coefficient 0.95). Thus for our data, the two measures of the impact of correlations on information transmission were consistent. Taken together, these results indicate that the neural code for vibrissa motion in VPM depends on temporal modulations of the firing rate rather than on temporally correlated spike patterns.

To test whether the amount of information conveyed by correlated spike patterns was related to bursting, we compared the degree of each cell’s burstiness to the impact of correlations on the information. Burstiness was quantified by the index $I_{\text{burst}}$ (METHODS); the correlation effect by the ratio $I_{\text{cor}}/I$. We found a statistically significant relationship between them (Pearson correlation coefficient 0.48; $P = 0.007$). This suggests that a significant fraction of the effect of temporal correlations on information was due to bursting.

Why did temporal correlations have a clear effect on response reliability (Fig. 4) yet contribute only a small amount of information (Fig. 5)? The amount of information that a neuron can transmit is limited not only by the reliability of its response but also by its dynamic range (that is, the range of different spike patterns that it can emit). Both properties can be quantified using information theory (METHODS). Dynamic range is quantified by the “response entropy” $H(R)$, reliability by the “noise entropy” $H(R | S)$: a reliable neuron has low $H(R | S)$, an unreliable one high $H(R | S)$. Information ($I$) is the amount of a neuron’s dynamic range that is not used up by noise: that is, $I = H(R) - H(R | S)$.

We investigated how both these entropies were affected by temporal correlations in the spike train. To do so, for each unit, we measured $H(R)$ and $H(R | S)$ together with the corresponding quantities $H_{\text{ind}}(R)$ and $H_{\text{ind}}(R | S)$ for a hypothetical unit with identical time-varying firing rate but no temporal spike train correlations. Because correlations constrain the set of possible spike patterns that can occur, $H(R | S)$ must be less than or equal to $H_{\text{ind}}(R | S)$, implying that the actual response is at least as reproducible as the correlation-free response. Quantitatively, we found that $H(R | S)$ was 1.1 bits/s less than $H_{\text{ind}}(R | S)$ ($T = 50$ ms, averaged across units; Fig. 6, +). Therefore consistent with the effect of negative correlations on response variance reported earlier, spike correlations did increase reliability. However, we also found that $H(R)$ was 0.5 bit/s less than $H_{\text{ind}}(R)$ ($T = 50$ ms, averaged across units; Fig. 6, C): the actual response had lower dynamic range than the correlation-free response. The net effect on information was therefore a combination of two factors pulling in opposite directions: the increase in reliability was partially cancelled out by a decrease in dynamic range. The net result was the small, positive correlation effect reported earlier.

It is known from theoretical work that a small value of the measure $I_{\text{cor}}$ can occur in different ways (Latham and Nirenberg 2005; Panzeri et al. 1999). As detailed in METHODS (Eq. 5), $I_{\text{cor}}$ is the difference between two factors: the effect of correlations on dynamic range, $H(R) - H_{\text{ind}}(R)$ and their effect on reliability, $H(R | S) - H_{\text{ind}}(R | S)$. Thus one way for $I_{\text{cor}}$ to be small is if both correlation effects are small. However, $I_{\text{cor}}$ can also be small even if the correlation effects are large, providing that both are of similar magnitude. In the latter regime, it is possible for correlations to affect neuronal responses strongly yet for this effect not to be shown by the $I_{\text{cor}}$ measure. To discriminate between these possibilities, we computed $H(R) -
As reported earlier, we found both the dynamic range effect to be small (0.9, averaged across units) and the reliability effect to be small (2.4, averaged across units). Thus our finding that $I_{cor}$ was small reflected the weak effects of temporal correlations on $H(R)$ and $H(R | S)$. As reported earlier, we found both the dynamic range effect to be small (0.9, averaged across units) and the reliability effect to be small (2.4, averaged across units). Thus our finding that $I_{cor}$ was small reflected the weak effects of temporal correlations on $H(R)$ and $H(R | S)$ rather than strong cancellation effects.

Collectively, these results show that VPm neurons convey information about dynamic vibrissa stimuli at a remarkably high rate. The key to the neural code is the submillisecond precision of spike timing. Temporal correlations do not increase the information rate significantly and almost all the information in the spike train can be decoded in their absence. Thus temporal modulation of the firing rate appears to be the crucial variable used to represent and transmit information about vibrissa motion.

**Discussion**

A key question in neural coding is how to “read” the response of a neuron. Is the essential information-bearing unit of a spike train the firing rate or a correlated spike pattern? Is its timescale fast (spike timing) or slow (spike count)? There is evidence from previous research that both vibrissa location and deflection velocity affect the firing rate of VPm neurons (Armstrong-James and Callahan 1991; Ito 1988; Simons and Carvell 1989; Waite 1973) and therefore that firing rate modulation is important to the neural code in VPm. Our study builds on this work by presenting a quantitative comparison of alternative information-bearing units. To take into account all temporal stimulus features (position, velocity, acceleration, etc.) that might affect the neural response, we used a white-noise stimulus that thoroughly explores the space of possible temporal features and an information theoretic formalism that quantifies the importance of a putative coding unit, without making assumptions about which stimulus features might be encoded. By using a new information estimation technique (Montemurro et al. 2007), we were able to obtain reliable results at a higher temporal resolution than would previously have been possible.

Our results show that robust encoding of dynamic vibrissa stimuli occurs in the VPm, shedding the following light on the neural code. We found that 1) VPm neurons convey considerably more information by precisely timed spike patterns than by the spike count; 2) information increases with spike timing precision up to at least 0.5 ms; 3) information increases linearly with time; 4) 93.9% of the information encoded by a neuron is accounted for by a hypothetical neuron with the same time-dependent firing rate but no correlations between spikes; and 5) ≥93.4% of the information in a VPm spike train can be decoded even if temporal correlations are ignored.

**Role and significance of precise spike timing**

Coding at fine timescales is possible only if repeated presentations of the same stimulus elicit spikes at very similar times on each trial. Published single-unit poststimulus time histograms (PSTHs) recorded from both trigeminal ganglion (Fig. 9 of Arabzadeh et al. 2005; Jones et al. 2004) and VPm (Fig. 5 of Armstrong-James and Callahan 1991; Fig. 5 of Diamond et al. 1992; Fig. 6 of Ito 1988; Fig. 5 of Simons and Carvell 1989) consistently exhibit peaks a few milliseconds or less wide. Such firing rate peaks can occur only if the associated spikes are timed with millisecond precision. Thus these data are consistent with those of the present study. However, to play a significant role in coding, spike timing must be sufficiently reliable to afford stimulus discrimination on a single-trial basis. The PSTH is an average across trials and does not answer this question. The principal contribution of our investigation is to demonstrate, by information theoretic analysis, that submillisecond spike timing does indeed code a large amount of information. This implies that spike timing may indeed play a crucial role in the VPm neural code for texture-induced vibrissa motion.

The number of different messages a spike train can transmit increases exponentially with spike timing precision (MacKay and McCulloch 1952). Thus the potential significance of spike timing is that it makes for a powerful neural code. In the present case, with 0.5-ms precision, we observed information rates of up to 77.9 bits/s (26.5 bits/s on average). To interpret such information rates, it is useful to consider the context of a rat performing a texture-discrimination task. Because we found that information increased linearly with time, the amount of information available to downstream cortical circuits is strongly limited by how long the rat spends doing sensory processing before making its decision. Assuming the available processing time is one whisking cycle, roughly 100 ms, the 77.9 bits/s neuron would convey 7.8 bits; the average neuron, 2.7 bits. Given that 1 bit of information allows a decoder to narrow down its uncertainty about which stimulus occurred by a factor of 2, 2 bits by a factor of $2^2 = 4$, and so on, this means that observing one VPm neuron for 100 ms can narrow down the stimulus uncertainty by a factor of up to $2^{7.8}$. Supposing the rat is able to differentiate between $10^6 = 2^{19.9}$ different textures and assuming that neurons code information independently, this implies that as few as three neurons (or eight average ones) could represent which of the million textures had occurred. In this way, our data illustrate the power of a precise spike timing code and its utility for a demanding, high-capacity skill such as texture discrimination. Precise spike timing potentially enables a small ensemble of thalamic neurons to transmit a great deal of surface texture information.

In principle, a neuron can convey the same amount of information either by a diffuse code, involving many, weakly informative spikes, or by a sparse code, involving a few, highly informative spikes. We found that the white-
noise stimulus elicited an average firing rate of 5.8 spikes/s but conveyed 5.3 bits of information per spike. This implies that the average VPm spike narrows down the stimulus uncertainty by a factor of $2^{5.3} = 39.4$. To interpret this number, it is again useful to consider the illustrative problem of a rat discriminating between a million different textures. Assuming that spikes convey independent information, this means that as few as four spikes could support reliable discrimination. Thus not only do small numbers of neurons convey a great deal of information, but they do so by small numbers of spikes. Because action potentials require significant energy to produce and transmit across synapses (Laughlin 2001), this type of sparse code might convey the same information as a diffuse code but at much lower metabolic cost. An additional advantage of the sparse VPm code may therefore be energy efficiency. These functional benefits of spike timing may explain why the trigemino-thalamic synapse has specific morphological features (Spacek and Lieberman 1974; Williams et al. 1994) that facilitate accurate propagation of signals from the periphery.

Although millisecond precision spike timing has also been shown to play an important role in other neural systems, including the lateral geniculate nucleus of the thalamus (Reinagel and Reid 2000) and invertebrate visual/audiovisual systems (de Ruyter van Steveninck et al. 1997; Rokem et al. 2006), it is not universal. In the vibrissa system, for example, the timing of spikes fired by neurons in the paralemniscal posterior complex (POm) is much more variable than that of neurons in the VPm (Fig. 5 of Diamond et al. 1992). Consequently, POm neurons appear constrained to code on a substantially slower timescale. This intriguing difference may reflect substantially different functional roles for the two structures (Ahissar et al. 2000). Our data indicate that the VPm is well suited to the high-capacity demands of texture coding. POm neurons have been proposed to encode the whisking rhythm (Yu et al. 2006), which may be an intrinsically lower capacity task.

**Role of firing rate compared with temporal correlation**

It is well established that sensory neurons do not fire spikes independently, but that spike trains exhibit significant correlations (Mastronarde 1983). In the temporal domain, two important sources of correlation are refractoriness and bursting. The implication is that firing rate is, in general, only a partial description of the neuronal response: correlated spike patterns may also be significant. There is direct evidence from previous studies that both refractoriness and bursting influence neural coding. Refractoriness can increase the temporal precision of neuronal firing and thereby boost response reliability (Berry and Meister 1998; Chacron et al. 2001; Miller and Mark 1992). Bursting can convey stimulus information beyond that available from single spikes (Guido et al. 1995; Kepcs and Lisman 2003; Reinagel et al. 1999). Consistent with these findings, we observed that refractoriness lowered response variance and thus increased response reliability. Moreover, we found that temporal correlations in the spike train contributed to both encoding and decoding. However, their effect was small: 6.1% according to the $I_{corr}$ measure, 6.6% according to the $\Delta I$ measure. This reflected a partial cancellation between a correlation-induced decrease in noise entropy and decrease in response entropy. Thus the foundation of the high VPm information transmission rate was temporal variations in firing rate.

Temporally correlated spike pattern information arises partly from burst firing, which in the thalamus depends on current flow through T-type Ca$^{2+}$ channels. The $I_T$ current is inactivated by depolarization and requires about 100 ms of hyperpolarization before this is alleviated (Jahnsen and Linás 1984). Thus thalamic bursting is particularly prevalent when each stimulus is temporally well isolated (Sherman 2001). However, under our experimental conditions, vibrissae were continuously deflected and prolonged periods of hyperpolarization likely to be rare. Consistent with the “wake-up call” theory of burst function (Sherman 2001), it may be that VPm bursts are important for informing the rat about sudden, unexpected whisker contact but that single spikes are the main channel for coding detailed texture structure in VPm.

**Implications for cortical decoding of thalamic spike trains**

To influence behavior, VPm signals must typically be propagated to barrel cortex (Guic-Robles et al. 1992; Hutson and Masteron 1986) and decoded by barrel cortical circuits. There is evidence that cortical neurons respond to temporally localized peaks in the firing rate of their thalamic afferents (Pinto et al. 2000) and therefore that it is the temporally modulated VPm firing rate that barrel cortical neurons decode. In general, decoding based purely on firing rate may neglect important information contributed by correlated spike patterns. However, because we found that neglecting temporal correlations led to little loss of information, in this case, decoding by firing rate may be an efficient strategy. Due to the sensitivity of information to precise spike timing, a significant implication of our findings is that the timescale of the decoding is likely to be critical. If, for example, cortical neurons counted spikes on a 50-ms timescale, $<10%$ of the thalamic information concerning the white-noise stimulus would get through.

In conclusion, the essence of the VPm code for vibrissa motion appears to be firing rate modulation on a submillisecond timescale. Such a code allows distinctions between very many different textures to be accurately conveyed to the barrel cortex in a metabolically efficient manner. The next step in our investigation will be to identify exactly which stimulus features these precisely timed spikes convey (Brambilla et al., unpublished observations).

**Acknowledgments**

We thank D. Adams, E. Ahissar, M. E. Diamond, and J. Schnupp for valuable discussion, S. Shoham for sharing code for the SAC program, J. Simpson for workshop assistance, and S. Shelmerdine for histological assistance.

**Grants**

This work was supported by the Royal Society through an International Joint Project grant to R. S. Petersen and M. Maravall and research grants to S. Panzeri and R. S. Petersen; by the European Commission through Marie Curie International Reintegration Grant MIRG-CT-2004-511273 to M. Maravall; by the Human Frontier Science Program through a Long-term Research fellowship to M. A. Montemurro and Program Grant RG0043/2004-C to M. Maravall; by the United Kingdom Medical Research Council through a Research fellowship to M. A. Montemurro; and by the Spanish Ministry of Education and Science through a grant to M. Maravall BFU2006-04791/BFI, co-funded by the European Regional Development Fund.
Sherman SM. Tonic and burst firing: dual modes of thalamocortical relay. 

Shoham S, Fellows MR, Normann RA. Robust, automatic spike sorting 
using mixtures of multivariate t-distributions. *J Neurosci Methods* 127: 

Simons DJ, Carvell GE. Thalamocortical response transformation in the rat 

Spacek J, Lieberman AR. Ultrastructure and three-dimensional organization 
of synaptic glomeruli in rat somatosensory thalamus. *J Anat* 117: 487–516, 
1974.

Strong SP, Koberle R, de Ruyter van Steveninck R, Bialek W. Entropy 

Sugitani M, Yano J, Sugai T, Ooyama H. Somatotopic organization and 
columnar structure of vibrissae representation in the rat ventrobasal com-

van der Loos H. Barreloids in mouse somatosensory thalamus. *Neurosci Lett* 
2: 1–6, 1976.

Waite PM. The responses of cells in the rat thalamus to mechanical move-

Williams MN, Zahm DS, Jacquin MF. Differential foci and synaptic organ-
ization of the principal and spinal trigeminal projections to the thalamus in 

Yu C, Derdikman D, Haidarliu S, Ahissar E. Parallel thalamic pathways for 