Parallel Coding Schemes of Whisker Velocity in the Rat’s Somatosensory System

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Abstract

The function of rodents’ whisker somatosensory system is to transform tactile cues, in the form of vibrissa vibrations, into neuronal responses. It is well established that rodents can detect numerous tactile stimuli and tell them apart. However, the transformation of tactile stimuli obtained through whisker movements to neuronal responses is not well-understood. Here we examine the role of whisker velocity in tactile information transmission and its coding mechanisms. We show that in anaesthetized rats, whisker velocity is related to the radial distance of the object contacted and its own velocity. Whisker velocity is accurately and reliably coded in first-order neurons in parallel, by both the relative time interval between velocity-independent first spike latency of rapidly adapting neurons and velocity-dependent first spike latency of slowly adapting neurons. At the same time, whisker velocity is also coded, though less robustly, by the firing rates of slowly adapting neurons. Comparing first- and second-order neurons, we find similar decoding efficiencies for whisker velocity using either temporal or rate-based methods. Both coding schemes are sufficiently robust and hardly affected by neuronal noise. Our results suggest that whisker kinematic variables are coded by two parallel coding schemes and are disseminated in a similar way through various brainstem nuclei to multiple brain areas.
INTRODUCTION

Our inferences about the brain mechanisms underlying perception rely on the mechanisms that make it possible for the brain to “reconstruct” a stimulus from information contained in the concerted activation of ensembles of neurons. A significant step towards uncovering the general principles underlying this process and its relevance to sensory perception would be made if the correspondence between neural ensemble activity and sensory stimuli could be determined. In the rodent whisker somatosensory system, tactile information is acquired by an array of whiskers on the facial pad. Using their whiskers, rodents can locate and distinguish objects in their immediate sensory environment and reliably detect small differences in surface texture (Diamond 2010; Jadhav and Feldman 2010; Diamond et al. 2008; Szwed et al. 2003; Mehta et al. 2007; Kleinfeld and Deschenes 2011; Petreanu et al. 2012; Xu et al. 2012). While it is well-established that interactions between whiskers and objects generate a complex pattern of whisker vibrations (Wolfe et al. 2008; Lottem and Azouz 2009; Adibi and Arabzadeh 2011; Pammer et al. 2013; Quist and Hartmann 2012; Bagdasarian et al. 2013), it is insufficiently understood which of the physical features of these vibrations are actually encoded, and what is the coding paradigm which serves as a basis for tactile perception.

Several features of whiskers’ vibrations have been proposed, each of which could underlie the perception of object identity, and each involving different kinds of computations. Two of the features, namely average whisker speed and power spectral density of whisker vibration, require the temporal integration of tactile signals, such as mean speed or total power of whisker vibrations (Arabzadeh et al., 2003), or the spectral frequency of whisker vibrations (Hipp et al. 2006). Other, kinematic features, such as whisker location, velocity and acceleration, on the other hand, require information regarding whisker transients (Arabzadeh et al., 2005; Jadhav et al., 2009; Wolfe et al., 2008). Rather than use these features in mutually exclusive ways, it is likely that rodents process several features simultaneously, depending on the complexity of both stimulus and behavioral tasks.
Finally, several studies have shown that rodents’ somatosensory system can measure the amplitudes of forces in the sensory follicles dedicated to tactile sensation, in order to estimate radial object distance (Bagdasarian et al. 2013; Pammer et al. 2013).

Another important question in the field of sensory physiology involves the coding paradigms that serve as the basis for tactile perception. It is hardly disputed that tactile features may be coded by firing rates (Adrian 1926; Romo and Salinas 2003; von Heimendahl et al. 2007). For instance, changes in firing rates have been proposed as the primary way in which somatosensory system neurons represent velocity (Arabzadeh et al. 2003; Gibson and Welker 1983; Pinto et al. 2000; Shoykhet et al. 2000; Zucker and Welker 1969). This, however, does not exclude the possibility that neurons may also represent tactile features by their spike timing. Substantial evidence from trigeminal ganglion (TG) neurons (Bale et al. 2013; Jones et al. 2004; Lottem and Azouz 2011; Shoykhet et al. 2000; Stutgen et al. 2006), from the somatosensory thalamus (Deschenes et al. 2003; Petersen et al. 2008), and from primary somatosensory cortex neurons (Arabzadeh et al. 2006; Arabzadeh et al. 2005; Panzeri et al. 2001; Petersen et al. 2002b; Pinto et al. 2000), have shown that neuronal responses in the somatosensory system are precise and reliable, and can follow the fine temporal features of the stimulus (Deschenes et al. 2003; Ewert et al. 2008), suggesting that relevant tactile information may be represented in neuronal temporal patterns.

In the present study, we examine the transformation of tactile features to transient kinematic variables of whisker vibrations and the coding paradigm that is being used as the basis for tactile perception.

We find that both the radial distance of an object and object velocity are transformed into whisker velocity. We show that whisker velocity is encoded in TG neurons primarily by latency coding, whereas in brainstem nuclei these signals are represented by two competing encoding schemes, latency and firing rate encoding. We discuss the implications of these results in the context of transmission of tactile information to various brain areas.
EXPERIMENTAL PROCEDURES

Recording and stimulation

Surgical procedures. Adult male Sprague Dawley rats (250-350 gm) were used. All experiments were conducted in accordance with international and institutional standards for the care and use of animals in research. Surgical anesthesia while recording from TG neurons was induced by urethane (1.5 gm/kg i.p.) and maintained at a constant level by monitoring forepaw withdrawal and corneal reflex; extra doses (10% of original dose) were administered as necessary. While recording from brainstem neurons, after initial anesthesia with ketamine (100 mg/kg) and xylezine (10%), a tracheotomy was made following local subcutaneous injection of lidocaine.

Rats were mounted in a stereotaxic device and respirated with a mixture of halothane (0.5% - 1.5%) and oxygen-enriched air. Atropine methyl nitrate (0.3 mg/kg i.m.) was administered after general anesthesia to avoid respiratory complications. Body temperature was maintained near 37°C using a servo-controlled heating blanket (Harvard, Holliston, MA). After placing subjects in a stereotaxic apparatus (TSE, Bad Homburg, Germany), an opening was made in the skull above the TG, and tungsten microelectrodes (2 MΩ, NanoBio Sensors, Israel) were lowered according to the known stereotaxic coordinates of the TG (1.5-3 mm ML, 0.5-2.5 mm AP (Shoykhet et al. 2000)), principal trigeminal nucleus (PrV) (2.9 mm ML, 0.3 mm posterior to interaural), spinal trigeminal nucleus interpolaris (SpVi) (2.4- 2.9 mm ML, 2.8-4.3 posterior to interaural).

To distinguish clearly between cell ensembles in the magnocellular and parvocellular divisions of the SpVi, all recordings were performed in either the rostral (11.8–12.2 mm AP) or caudal part (12.8–13.3 mm AP) of the nucleus. To verify these locations we used the rostral part of the PrV as a reference point and then moved back ~3.4 mm or ~4 mm. Once a neuron was encountered, its receptive field was mapped. The recorded signals were amplified (x 1000), band-pass filtered (1 Hz-10 kHz), digitized (25 kHz) and stored for off-line spike sorting and analysis. The data were
then separated to distinguish local field potentials (LFP; 1-150 Hz) from isolated single-unit activity (0.5-10 kHz).

All neurons could be driven by manual stimulation of one of the whiskers. Spike extraction and sorting was accomplished using a modified version of MClust (by A. D. Redish, available from http://redishlab.neuroscience.umn.edu/MClust/MClust.html ), which is a Matlab-based (Mathworks, Natick, MA) spike-sorting software. The extracted and sorted spikes were stored at a 0.2 msec resolution and peristimulus time histograms (PSTHs) were computed. The scale bars of the PSTHs indicate response probability. For each bin, the number of spikes was divided by the number of trials.

Artificial whisking (Fig. 1A; Brown and Waite 1974; Szwed et al. 2003) was induced by stimulating the buccolabialis motor branch of the facial nerve (Semba and Egger 1986). The nerve was cut, its distal end mounted on bipolar tungsten electrodes, and was kept moist. Bipolar rectangle electrical pulses (10-20 pulses of 100 µsec at 143 Hz for 70-105 msec) were applied through an isolated pulse stimulator (ISO-Flex; A.M.P.I. Israel) to produce whisker protraction, followed by a passive whisker retraction at frequencies in the range of 1 Hz. The stimulation magnitude was adjusted at the beginning of each recording session to the minimal value that reliably generated the maximal possible movement amplitude (50-200µA).

C3 Whisker displacements transmitted to the receptors in the follicle were measured by an infrared photo-sensor (resolution 1 µm; Panasonic CNZ1120) placed 2 mm from the pad. The voltage signals were digitized at 25 KHz and amplified (x500; FLA-01, Cygnus Technology, Delaware). During a “single whisker” experiments, other whiskers were trimmed off. We then converted the whisker movements to arc degrees.

**Histology.** To mark the recording sites in the brainstem, we induced electrolytic lesions by the recording electrodes. These were made by passing direct current (10-30 µA) for 4 sec at a depth that
corresponded to each recorded area. In some of the rats, brain tissues were also processed for CO histochemistry. The animals were perfused transcardially with 2.5% glutaraldehyde and 0.5% paraformaldehyde, followed by 5% sucrose, all in 0.1M PBS (Phosphate Buffered Saline). The animals’ brains were then placed in post-fixative solution of 30% sucrose at 4°C overnight. The next day, freezing microtome sections (120 μm) were prepared and incubated in a solution of 0.0015% cytochrome C (sigma), 0.05% diaminobenzidine in PBS for 20 - 50 minutes at 37°C. The reaction was terminated by washing the reagent solution with PBS. CO-stained sections were mounted on gelatin-coated slides, air-dried, and coverslipped.

**Whisker stimulation.** Receptive fields were initially determined by manually deflecting individual whiskers. Whiskers evoking detectable responses were then individually attached to a computer-controlled Galvanometer stimulator (Model 6210H Galvanometer Scanner, Cambridge Technology, Inc.). These scanners, once calibrated, have a resonant frequency well above the range of velocities used in the current study. Each stimulus was presented for one second and repeated 25-40 times in both the preferred and the null direction. A period of 2 sec separated each stimulus. Stimuli were delivered approximately 3mm from the mystacial pad. The whisker was attached firmly to the edge of the galvanometer lever. Accordingly, the angle of the whisker was given by: \( \theta = \tan^{-1}(x/3) \), where \( x \) is the whisker’s displacement in millimeters. Furthermore, individual whiskers were deflected by several types of stimuli which were randomly interspersed in time: first, ramp whisker deflections ranging from amplitudes of 100-2,000 μm with a duration of 20 msec, resulting in velocities 95 – 1,910 deg/sec; second, step stimuli (600, 1,200 μm). The galvanometer stimulator was calibrated using a calibrated, non-contact optical displacement measuring system (resolution: 1 μm; LD1605-2; Micro-Epsilon, Ortenburg, Germany).

**Data Analysis**

The significance of the differences between measured parameters was evaluated using one-way
analysis of variance (ANOVA). When significant differences were indicated in the F ratio test 
\( p<0.05 \), the Tukey method for multiple comparisons was used to determine those pairs of 
measured parameters that differed significantly from each other within a group of parameters 
\( P<0.05 \) or \( P<0.01 \). Below, averaged data are expressed as mean ± SE unless otherwise stated. 
Error bars in all the following figures indicate the standard error unless otherwise stated. To avoid 
cluttering in the graphs we use single-sided error bars.

Neuron classification into the three cell types was described in detail elsewhere (Lottem and 
Azouz 2011). Briefly, RA neurons respond to step stimuli with a phasic response. They do not 
show either directional selectivity at higher stimulus intensities, or any dependence of first spike 
latency on whisker deflection velocity. In contrast, within the SA neurons, namely those having 
phasic–tonic response, we identified two distinct subgroups, distinguished by their velocity 
thresholds (170 deg/s; Lottem and Azouz 2011) for firing: \( \text{SA}_{lt} \) (low threshold) and \( \text{SA}_{ht} \) (high 
threshold). These subtypes differ in other respects as well: (a) \( \text{SA}_{lt} \) neurons display large phasic 
responses followed by stochastic tonic firing, whereas \( \text{SA}_{ht} \) neurons exhibit slowly decaying 
periodic responses, followed by stochastic tonic firing, in response to step whisker deflections; (b) 
\( \text{SA}_{lt} \) neurons respond to low whisker velocities with a larger number of spikes than \( \text{SA}_{ht} \); (c) \( \text{SA} \) 
neurons display a dependence of first spike latency on whisker deflection velocity (100–56,000 
deg/s), which is expressed differentially by the two subtypes. The two SA subtypes are similar, 
however, in that all SA neurons show directional selectivity, and both subtypes show a clear 
dependence of initial firing rates on whisker velocity. (For further Distinguishing parameters 
between the cell types see Lottem and Azouz 2011).

Firing rates as a function of whisker velocity were calculated by dividing the number of spikes 
by stimulus duration (20 msec).

To determine the impact of whisker velocity on first spike latency, we first smoothed out the
response probability density function with a Gaussian of 0.5 msec STD. The latency was then
determined by finding the first maximum larger than 0.1. To quantify the dependence of 1/first
spike latency (first spike latency$^{-1}$) on whisker velocity, we fitted the points in Figs. 2 D, E with a
logistic sigmoid function:

\[\text{First spike latency}^{-1} (\text{Vel}) = \frac{\text{sat.}}{\text{Vel} - \text{Vel}_0} = \frac{1}{1 + e^{-\text{slope} \cdot \text{Vel}}}
\]

Where \text{Vel} is velocity, \text{sat.} is the function saturation value, \text{Vel}_0 is the function shift, and \text{slope} is
the rate of change.

Our analysis of relative spike timings relies on an onset signal, which is provided by RA
neurons. However, such an analysis might be sensitive to outliers, i.e., a single neuron that is faster
and more reliable than the rest of the population, could provide most or all of the onset spikes,
thereby masking a much larger spike-latency variability that may be present in the population.
Therefore we pooled responses from all of the RA neurons involved in our study, generating a
pseudo-population of responses treated as if they were recorded simultaneously (note that although
the existence of neuronal noise correlations might hamper such an analysis, the low response
variability and the lack of synaptic contacts between these neurons make such correlations very
unlikely). We then generated “pseudo-trials”, in which each neuron contributed one response to the
overall population response (trial assignments were drawn randomly and independently across the
population), and calculated the spike latency probability distributions of the first, second, etc.,
fastest spikes across all neurons. Finally, to extrapolate these data to any population size of onset
detectors, we expressed spike order in the form of percentiles. For example, the population onset
signal of the 10th spike is expressed as the 53rd percentile (10/19; 19 being the total number of
recorded RA neurons). Importantly, this procedure sequentially eliminates neurons from the dataset,
from fastest to slowest, such that the distribution of second-spike latencies, for instance, does not
include the fastest neurons on any given trial. Any outliers would be rapidly eliminated by this procedure, and the true nature of the onset signal in this population should be revealed.

To calculate the relative timing between RA and SA neuronal discharges, we convolved SA neuron first spike latency distribution with the inverted RA neuron first spike latency distribution (Fig. 5A). The relative timing was measured on positive and negative differences, i.e., when RA spikes precede or follow SA, respectively.

To classify brainstem neuronal ensembles according to their functionality, we first calculated a first spike latency range (maximum first spike latency to minimum first spike latency) for each neuron. We then established a first spike latency range, in which all the neurons that had a first spike latency range smaller than or equal to an arbitrary latency threshold were considered shorter latency neurons. Neurons with a first spike latency range higher than the threshold as well as having a minimal spike latency later than the earliest onset spike were considered longer latency neurons.

The precision measure (which is based on Mainen and Sejnowski 1995) gives a measure of the temporal “jitter” of spike latency. Briefly, we detected in each PSTH (bin size=1 msec) peaks which exceeds a certain threshold. This threshold was calculated for each neuron. Threshold calculation was based on the mean ± SD of peak values of the PSTH. We then calculated the standard deviation (SD) of the latency of the spike corresponding to each event. Finally, the SD for all the events was averaged. The precision measure, referred to as the “jitter,” corresponds to this average SD, and therefore increases in the jitter (i.e. increased SD) are associated with decreases in the precision of action potential timing measurement.

**Receiver operating characteristics analysis**

We used signal detection theory (receiver operating characteristics, ROC analysis, Green and Swets 1974), to compute the probability that an ideal observer could accurately determine the differences among whisker velocities based on neuronal activity. For each measured whisker velocity pair, an
ROC curve was constructed. The ROC curve is a two-dimensional plot of hit probability on the ordinate against false-alarm probability on the abscissa. To transform raw data into a measure of discriminability, we analyzed the distributions of several neuronal response characteristics across trials. The distributions also included trials in which there were no responses. Each point on an ROC curve shows a pair of hits and false alarms, representing the proportion of trials in which the first velocity response exceeded a criterion level, plotted against the proportion of trials in which the other velocity response exceeded the same criterion (Figure 3B). The entire ROC curve was calculated by sweeping the criterion from the minimal level to a value greater than the highest level in the two distributions. Improved separation between the two whisker velocity response distributions leads to an increased deflection of the ROC away from the diagonal. Green and Swets (1966) showed that the area under the ROC curve (AUC) corresponds to the performance expected of an ideal observer in a two-alternative, forced-choice paradigm, such as the one used in the present analysis. The ROC curve was calculated for four neuronal response characteristics: (a) the firing rate of a single neuron as a function of whisker velocity; (b) first spike latency; (c) first inter-spike interval; and (d) time interval between RA and SA response.

To assess the discrimination within all velocity pairs across these four neuronal characteristics, we calculated the AUC for all velocity pairs. This is shown in Fig. 3B for first spike latency. Each value in this matrix represents an AUC value for a velocity pair. To reduce the dimensionality of our analysis, we averaged all matrix values that belonged to the same diagonal (average global AUC, or discriminability plot; Fig. 3C), which illustrates the overall discrimination for all velocity differences. It should be noted that this presentation has some limitations, since the number of elements per diagonal decreases as the difference between the velocities increases.

To measure the significance level of P(correct) in the ensemble of TG neurons, we took all possible whisker velocity comparisons ($\frac{20 \times 19}{2}$) for all SA neurons ($n = 37$) and shuffled the trials
across the different stimuli. We then repeated this procedure 500 times to have a distribution of 500 x 190 x 37 AUCs. The significance level was set at 90% of this population, namely 0.625 (Fig. 4A).

A similar procedure was used for the brainstem nuclei, where we found that the significance level for the PrV was 0.59, SpVi – 0.585.
RESULTS

Encoding of whisker velocity in TG neurons plays a major role in rats’ perception of their tactile environment. We employed several stimulus paradigms to examine the role of whisker velocity in the representation of tactile stimuli: (a) We used objects moving at different velocities to touch a whisker at a constant radial distance (approximately in the middle of the whisker; Fig. 1A). The figure shows that whisker velocity changes as a function of object velocity. (b) We examined whether an object moving at a constant velocity (575 mm/sec), touching a whisker at different radial distances (Fig. 1B), has an impact on whisker velocity. We used normalized values to indicate distance from the pad to the tip of the whisker (smaller values indicate points closer to the pad). We found that the farther from the pad an object touches the whisker, the slower the whisker moves. (c) Finally, we induced artificial whisking in anaesthetized rats by facial motor nerve stimulation, and monitored the movement of whiskers against objects located at different radial distances ($n = 3$). An example of whisker trajectories against objects located at different distances is shown in Fig. 1C. Quantification of the impact of object distance on transient whisker velocity is shown on the right-hand side. Radial object location is translated to whisker velocity. We found that the farthest the object was from the pad, the faster the whisker moved. These stimulus velocity changes occur within a range of approximately 20 msec (shaded area).

Taken together, our results indicate that transient whisker velocities may serve as one of the physical parameters representing tactile features, and that active object contacts show qualitatively different velocity profiles, compared with passive ones.

To characterize the rules governing the transformation of whisker velocity into spike generation, and the transmission of these signals from primary afferents to brainstem neurons, we employed ramp stimuli of various velocities. We probed neuronal response properties in 56 TG neurons
obtained from 14 adult rats, using both step and constant velocity ramp stimuli. An example of the
time courses of RA \((n = 19)\) and SA \((n = 37)\) neurons to step and ramp stimuli is shown in Figs. 1D, E.

We further divided SA neurons into two distinct subgroups, defined according to their velocity
threshold for firing (the distinction between the two subtypes is described thoroughly in Lottem and
Azouz 2011 and in the Methods section herein): \(SA_{lt}\) (low-threshold, \(n = 26\)) and \(SA_{ht}\) (high-
threshold, \(n = 11\)).

Despite the simple time course of the stimulus, the PSTHs of the different neuronal types have a
complex temporal waveform (Fig. 1E). At least three distinct aspects of the response can be
observed: first spike latency, which is defined as the time interval between stimulus onset and the
beginning of the neural response; first inter-spike interval; and firing rate. In SA neurons, the
decrease in latency can be observed by examining the column of PSTHs in Fig. 1B and noting that
in these neurons, the onset of responses becomes progressively earlier as velocity increases. This
phenomenon has been reported in several studies in rodents and humans (to name but a few,
Johansson and Birznieks 2004; Kwegyir-Afful et al. 2008; Shoykhet et al. 2000). The same applies
also to the first inter-spike interval, which becomes progressively shorter. Finally, the growing
number of PSTHs’ peaks as velocity goes up indicates an increase in neuronal firing rate, which has
also been reported previously (Arabzadeh et al. 2003; Gibson and Welker 1983; Pinto et al. 2000;
Shoykhet et al. 2000; Zucker and Welker 1969). An example of these characteristics in SA neurons
is shown in Fig. 2A. The shaded areas indicate the borders of our measurements of first spike
latencies and first inter-spike intervals, while colored solid squares on the right hand side indicate
the normalized firing rate of the neuron.

One of the goals of the current study was to juxtapose these three aspects of neuronal response
and examine their pros and cons in terms of reliability, robustness, and resilience. We calculated the
distribution of firing rates, first spike latencies, and first inter-spike intervals of TG neurons as a
function of whisker velocity. An example of these characteristics in an SA neuron is shown in Fig. 2A. The shaded areas indicate the border of our measurements of first spike latencies and first inter-spike interval, while colored solid squares on the right hand side indicate the normalized firing rate of the neuron. Velocity-dependence is confirmed by the different scalar measures of the neuronal responses, plotted against velocity. Our results show that increased whisker velocity leads to a decrease in both first spike latency (Fig. 2B) and first inter-spike interval (Fig. 2I), as well as an increase in discharge rates of SA neurons (Fig. 2F). In contrast, RA neurons do not show any apparent dependence of either first spike latency or firing rate on whisker velocity (Fig. 2B, F lower panels). The minor velocity-dependent changes in the firing rate of RA neurons are mostly due to an addition of a few spikes within a short period (e.g. the increase of firing rate to 50 Hz was due to an addition of a single spike during the 20 msec interval).

We further examined the impact of whisker velocity on the three neuronal response characteristics in the same cell types. SA\textsubscript{lt} neurons start responding at lower velocities, and their first spike latencies show a clear yet modest dependence on whisker velocity (Fig. 2C, upper panel). In contrast, SA\textsubscript{ht} neurons start responding at higher velocities and their first spike latency shows a strong dependence on whisker velocity. RA neurons are activated at a wide range of velocities, and once they respond, they hardly show any dependence of first spike latency on whisker velocity (Fig. 2B, lower panel).

To quantify the influence of whisker velocity on first spike latency and first inter-spike interval in a continuous manner, we plotted first spike latency \(^{-1}\) and first inter-spike interval \(^{-1}\) as a function of whisker velocity. These relations were then fitted to a logistic function, in which “no responses” are set to zero. Fig. 2D shows the normalized fits of all neurons in our sample. First spike latency \(^{-1}\) in RA neurons is practically step-like, whereas in the two types of SA neurons, first spike latency \(^{-1}\) varies more smoothly as a function of whisker velocity. Thus, single RA neurons are less suited to
velocity discrimination than SA neurons. Quantification of firing rates as a function of whisker velocity produces very similar results, in which the firing rates of SA neurons are better suited to velocity coding than single RA neurons (Fig. 2F-H). The smooth increase in first spike latency and firing rate as a function of velocity in the averaged plots (Figs. 2C, G) reflects different firing thresholds for each RA neuron, as shown by a shift in the plots.

To determine the velocity threshold for neuronal activation, we examined the proportions of the different cell types activated at each velocity. This analysis revealed that at lower velocities, SA neurons tend to dominate. As whisker velocity increases, more neuronal types become relevant (Fig. 2E). Furthermore, these results indicate that SA and RA neurons can represent whisker velocity by the proportion of active neurons within each subpopulation. All in all, our results indicate that single RA neurons convey little information about stimulus properties (i.e. velocity), other than indicating its onset. In contrast, SA neurons may code for stimulus characteristics by their first spike latency, yet they convey ambiguous information about its onset. The difference in the slopes of the two SA subtypes indicates a larger dynamic range for SA neurons, i.e. better discriminability between velocities (Fig. 2D).

Our results challenge the observation that RA neurons change their latency as a function of stimulus velocity (Shoykhet et al. 2000). However, in that study a piezoelectric element was used to move the whiskers. Due to a mechanical limitation of the piezo, the stimulus had to be filtered. Not knowing which type of filters these authors used, we chose a conservative low-pass filter (“filtfilt” filter in Matlab; low-pass filter: cut-off - 250 Hz; slope = 2) in 3 TG neuron. When we compared unfiltered and filtered stimuli, we found that this filter causes a latency dependence on velocity. Thus, RA neurons hardly show any velocity-dependent changes in their first spike latency (Fig. 3B).

*The Encoding mechanisms that underlie velocity discrimination*
To examine the possible coding mechanisms underlying tactile discrimination, we used ROC analysis (Green and Swets 1974) in order to assess the discriminative power and robustness of neuronal response features. The ROC curve for the three neuronal characteristics described in Fig. 2A is shown in Fig. 4A. The ROC curve in the panel, which was used to discriminate between two stimuli (859 deg/sec and 1,050 deg/sec; see arrows in Fig. 2A), shows that both first inter-spike interval and first spike latency are better at discriminating between the two velocities than the firing rate.

An assessment of the predictive performance of each of the response characteristics can be obtained by computing the AUC. The AUC can then be used to assess the discriminatory powers of the various response features. It approaches 0.5 for a chance level discrimination, and equals unity (or zero) for a perfect discrimination. To assess the amount of stimulus-related information conveyed by the different neuronal response characteristics, we calculated AUCs for all velocity pairs. This is shown in Fig. 4B for first spike latency. Each colored patch in this matrix represents an AUC value for a pair of velocities (the black square corresponds to the AUC of the first spike latency ROC curve in panel A of the figure). We then constructed a discriminability plot (Fig. 4C), which illustrates the overall discrimination for all velocity differences; it shows that as \( \Delta v \) increases, so does discriminability.

Figure 5A compares the discrimination curves of all SA neurons for all three neuronal features (firing rate, first spike latency and first inter-spike interval). The panel shows that first inter-spike interval and first spike latency are better at velocity discrimination than neuronal firing rates. To examine the role the different neuronal types in velocity discrimination, we created discriminability plots for each of neuronal response features and for each of the three neuronal types. Our results indicate that SA\(_{ht}\) neurons are superior to both SA\(_{lt}\) and RA neurons in velocity discrimination for all response features. With respect to firing rate, SA\(_{ht}\) and RA neurons show comparable
discriminability, whereas $SA_h$ neurons’ temporal response properties discriminate various velocities as well (Fig. 5 C, D).

“When” and “what” signals in TG neurons

Stimulus identity can be evaluated from SA first spike latency only if *stimulus onset time* is known. We have shown that first spike latency in single RA neurons does not depend on stimulus velocity, and therefore may serve as the stimulus onset signal. To determine whether RA and SA neurons responses can serve as “when” and “what” signal, respectively, we examined velocity discrimination using the relative timing between RA and SA neuronal discharges (Fig. 6A, gray area).

To determine the robustness of the “when” signal, we took two approaches: First, we pooled the first spike latencies from an ensemble of RA neurons ($n = 19$; see Methods section). To determine whether a population of RA neurons can provide an accurate and reliable onset signal, we examined the impact of whisker velocity on the mean first spike latency of the 5 (1st), 32 (6th), and 53 (10th) percentiles of first spikes across the ensemble (the numbers in parentheses are actual spike numbers). Our results show that these neurons hardly change their latency and can signal stimulus onset for all velocities (Fig. 6B).

Second, we averaged all the points on the various discriminability plots (using the time interval between RA and SA neuronal discharge) for different RA spike percentiles. The results show that even if we discard the more reliable half of the RA population, velocity discrimination using relative timing deteriorates just slightly and stays well beyond the significance threshold (Fig. 6C). Thus, RA neurons can provide the brain with a robust and reliable onset signal.

We next combined this onset signal (RA first spike latency distribution) with each of the SA neurons first spike latency to get a relative timing$^{-1}$ distribution for each velocity (*see* the Methods
section). As in our previous analysis, we created from these distributions ROC curves for all velocity pairs and calculated a discriminability plot. Our results indicate that while relative timing signals carry less information about whisker velocity than first spike latency of SA neurons, both are superior to firing rate (Fig. 6D) in this respect. Although this is true for both types of SA neurons, SA_{ht} neurons show superior velocity discrimination in their relative timing (Fig. 6E, right panel). Brought together, our results show that the relative first impulse timing in an ensemble of TG neurons accurately and reliably reflects whisker velocity. We showed that stimulus onset time can be evaluated using stimulus-independent first spike latency of RA neurons, whereas stimulus identity can be gauged by stimulus-dependent relative timing between RA and SA neuronal discharge.

Transmission of velocity signals through the brainstem

Tactile signals from TG neurons are transmitted through brainstem nuclei to multiple brain areas through several parallel pathways (Haidarliu et al. 2008; Veinante et al. 2000; Yu et al. 2006). To determine which response features are disseminated through brainstem nuclei, we recorded from neurons in the parallel pathways in the PrV and SpVi obtained from 9 adult rats (Fig. 7A). Figs. 7B, C show typical responses of PrV neurons to step and ramp stimuli with various velocities. We recorded from 78 PrV neurons, 68 of which were significantly responsive (3 SD above spontaneous activity). Out of these neurons, 55 showed consistent velocity dependence of their first spike latency and firing rate. We grouped these neurons according to their responses to step stimulus to SA (n = 46) and RA (n = 8) neurons (Fig. 7B). In contrast to TG neurons, PrV neurons did not show as clear a difference in their responses to various velocities. Both types of neurons exhibit a varying degree of velocity dependence in their first spike latencies (Fig. 7B right panels).

We also recorded from 109 SpVi neurons, 100 of which were significantly responsive. Of these neurons, 83 showed consistent velocity dependence of both first spike latency and firing rate. We
found that rostral (SpVi_r; \( n = 34 \)), intermediate (\( n = 21 \)), and caudal (SpVi_c; \( n = 28 \)) neurons respond differently to step stimulus as well. In the SpVi_r, the majority of the neurons were RA (Fig. 7C, RA: \( n = 21 \); SA: \( n = 13 \)), whereas in the SpVi_c, most neurons were SA (Fig. 7C, RA: \( n = 8 \); SA: \( n = 20 \)) (as reported also by Chiaia et al. 1987). Same as in the PrV, SpVi RA and SA neurons did not show a clear difference in their responses to various velocities. Both types of neurons exhibited a varying degree of velocity dependence in their first spike latency.

To ascertain which signals are transmitted through second-order neurons in brainstem nuclei, we charted a discriminability plot for the three different features for each neuron in the different areas (the data from SpVi_c and SpVi_r were lumped together since we could not find any significant differences between them). Our results indicate that the transmission of the three neuronal signals, as reflected by velocity discrimination, is degraded in comparison to TG neurons. The deterioration in velocity discrimination is apparent in PrV (compare Fig. 7D with 6D). To quantify this deterioration we averaged all the points on each plot to get a value that reflects overall discriminability for each feature in each area. We then calculated the ratios between the discriminability values obtained in PrV and brainstem to quantify the differences between the two areas (Fig. 7E). The figure shows that the deterioration in firing rate signal was minimal (light gray), whereas the largest deterioration in velocity discrimination occurred in relative timing signals (dark gray). Nevertheless, despite this worsening of temporal coding, the ratios between the coding paradigms were only 0.93 and 0.96 in PrV and SpVi, respectively (Fig. 7E, black bars). These results indicate that although transmission of tactile information does degrade in the brainstem, the discriminability of whisker velocity remains high.

Neuronal responses in the somatosensory system are precise and reliable, and can keep track of the temporal structure of the stimulus (Deschenes et al. 2003; Ewert et al. 2008). However, our results show that some deterioration in velocity discrimination occurs between first- and second-
order neuron. To examine the mechanisms responsible for this deterioration, we measured the jitter in first spike latency in all three brain regions. We quantified this jitter by measuring the SD of first spike latency, and found that first spike latency jitter increases from first- to second-order neurons (Fig. 8A, B), which may underlie the use of temporal coding in velocity discrimination. Moreover, this jitter is dependent on whisker velocity in SA neurons, but not in RA neurons. To eliminate the possibility that this deterioration is due to the nature of our stimulus, we compared first spike latency jitter in response to ramp and complex stimuli in the same neurons. We used filtered white noise stimulus with a velocity equivalent of 900 deg/sec (low-pass filter, 250 Hz); an example of TG and PrV neuronal responses to complex stimuli is shown in Figs. 8C, D. To compare response jitter between the two brain regions and between the two stimuli, we similarly calculated the first spike latency jitter in response to complex stimuli. We found that the first spike latency jitter in TG neurons is stimulus-independent (Figs. 8E, F, see horizontal lines). In general, however, SA neurons in PrV and SpVi show significantly less deterioration in their first spike latency jitter when responding to complex stimuli, compared with ramp stimuli (Fig. 8E). These results suggest that a temporal jitter in first spike latency may be the main cause for the deterioration of velocity discrimination. Moreover, this deterioration is less evident when complex stimuli are used.

In the preceding analysis, the “when” and “what” signal were predefined using the adaptation profile of the studied neurons (RA vs. SA). However, an alternative way to transmit an onset signal would be to divide the neurons functionally according to their velocity dependence. That is, we set a latency range criterion in which neurons with a velocity dependent latency range that is lower than the criterion are assigned to convey the onset signal. We used a dynamic latency threshold in which we chose a subset of the neurons (“velocity-independent” neurons) with a predefined latency range in each area (the range of first spike latency in response to the full range of whisker velocity). We considered these neurons to be the carriers of onset signal and calculated the relative timing between this subset of neurons and each of the remaining (“velocity-dependent”) neurons. For each
latency threshold, we charted the relative timing discriminability plot and compared it to first spike latency and firing rate discriminability plots. An example of discriminability plots for both brainstem areas at a latency range threshold of 4.4 msec is shown in Fig. 9A. The plots indicate that all three features are nearly equally effective in velocity discrimination.

To quantify the transmission of the three signals using our functional classification, we calculated the ratio between relative timing and firing rate discriminability plots for all latency thresholds. Fig 9B shows that at almost all latency ranges, the ratio is close to one. To see whether these two functionally-defined groups of neurons correspond to the previously described RA and SA neurons, we calculated in both areas and at different time ranges the proportions of RA neurons out of the “velocity-independent” neuronal population. We found that in the PrV region, RA neurons play a minor role, whereas in SpVi, most “velocity-independent” neurons are RA neurons (Fig. 9C). Thus, the different brainstem areas may have a different degree of “shuffling” of RA and SA populations (Sakurai et al. 2013). All in all, our results suggest that stimulus onset time can be estimated using velocity-independent first spike latency of a subset of the neurons, while stimulus identity can be gauged by velocity-dependent relative timing between these neurons and stimulus-independent neuronal discharge.

The impact of noise on “when” and “what” signals

One of the major caveats in using the temporal features of the neuronal response to code for whisker velocity is the response’s sensitivity to both intrinsic and extrinsic noise. These types of noise may cause spike time jitters and additions or subtractions of spikes, which could have a detrimental effect on the quality of the temporal code. It is therefore essential to test the noise-sensitivity of “when” and “what” signals. More specifically, spontaneous firing, if high enough,
could be detrimental to the coding of an onset signal.

We tested for the impact of noise in both TG and brainstem neurons. In TG neurons, we introduced noise in two different ways: (a) we added noise to the stimulus (Fig. 10A; this could be thought of as emulating naturally occurring small whisker movements caused by the wind, for instance); (b) we superimposed the stimuli on a DC whisker offset, which resulted in neuronal discharges in SA neurons. The introduction of these two types of noise resulted in spontaneous neuronal discharges (24±1.2 Spikes/Sec; Fig. 10A, lower panel). To determine the effect of these two types of noise on the “when” and “what” signals, we repeated the previous analysis (Figs. 5, 6) to create discriminability plots.

In order to eliminate the bias which could arise from the fact that stimulus onset was known to us, our trials began 15 msec prior to actual stimulus presentation (Fig. 10A). Under these conditions, some trials had “negative” first spike latencies, signifying that neurons fired before stimulus onset. We saw that in TG neurons, unlike first spike latencies, relative timing and firing rate signals maintained the level of velocity discrimination in the system (Fig. 10B). This is due to the fact that RA neurons, which relay the onset signal, are much less affected by noise: not firing during both static whisker offset (as their name implies) and low-velocity noise. In brainstem neurons, using lightly anaesthetized rats, we chose neurons that discharge spontaneously (PrV: n = 46, 72%; firing rates = 5.08±0.75 spikes/sec; SpV_{ir}: n = 13, 38%; firing rates = 1.89±1.0 spikes/sec; SpV_{ic}: n = 20, 60%; firing rates = 7.53±1.69 spikes/sec). We found that in brainstem neurons, only mild deterioration appeared in relative timing and first spike latency signals, while no degradation was observed in firing rate signal. These results were consistent across all brainstem nuclei (Fig. 10C). Put together, our results indicate that within the noise limits we tested, relative timing and firing rate signals are transmitted with the same reliability and efficiency through the brainstem.
The present study shows that in the rats’ whisker somatosensory system, both the speed and the radial distance of a moving object are translated into changes in whisker velocity during transient movements. Moreover, it has already been shown that during artificial whisking, the radial distance of a stationary object is translated into variations in whisker velocity (Bagdasarian et al. 2013). These observations, supported as they are by previous studies, suggest that whisker velocity may be used by rats to detect and discriminate isolated, punctate whisker deflections (Ito 1985; Pinto et al. 2000; Simons 1978). Thus, rodents can evaluate whisker kinematic variables, as well as the amplitudes of forces in the sensory follicles for tactile sensation. Such multiple variables as bending moment, axial force, lateral force as well as velocity, can reliably represent the coordinates of a contacted object’s position (Knutsen et al. 2008; Pammer et al. 2013).

A range of coding schemes for whisker velocity available at the first- and second-order networks was investigated using passive whisker stimulation in anesthetized rats. The feasibility of these schemes lies at the heart of an ongoing debate about the way the brain represents sensory information and the role of single neurons in this encoding (Averbeck et al. 2006; Meister and Berry 1999; Stanley 2013; Zador 1997). In the current study, we asked whether these neurons transmit tactile information by their firing rate or by the relative timing of their action potentials. The velocity sensitivity of neurons in the whisker somatosensory system has been measured in a number of previous studies (Ito 1985; Pinto et al. 2000; Shoykhet et al. 2000; Simons 1978). Most of the work done in this area has focused on neuronal firing rates in response to fast transient or ramp-and-hold deflections, reporting a monotonic dependence of firing rates on deflection velocity, which is consistent with our present results. However, we showed also that the relative timing of the first spikes in ensembles of first-order sensory neurons contains a wealth of information about the stimulus. Specifically, the relative timing of the first spikes contains reliable information about whisker velocity. In these ensembles, RA neurons apparently covey stimulus onset, whereas SA
neurons seem to transmit stimulus identity. Furthermore, first spikes code this information more efficiently than firing rate, which traditionally was thought to represent information in primary sensory neurons.

As tactile information is transferred from first-order neurons to various brainstem nuclei, whisker afferent arbors in different subnuclei of the same axon tend to have different sizes, shapes and bouton numbers (Shortland et al. 1996), which indicates that second-order neurons in each brainstem subnucleus receive a different sensory information. However, we showed that while the different subnuclei contain different proportions of RA and SA neurons (Fig. 7C), the transmission of whisker velocity information (based on our stimulus) through the various response features varies only slightly (Fig. 7E). More surprisingly, at the PrV level, the “when” signal, represented by RA neurons, is reduced by a large amount. This was indicated by measuring a sub-ensemble of neurons, which did not show any velocity dependence on the “what” signal (Fig. 9C). These results argue in favor of a “shuffling” of RA and SA populations, as was also predicted by anatomy (Sakurai et al. 2013).

All in all, our results suggest that temporal and firing rates codes provide the brain with independent information about whisker velocity. Given that numerous parallel pathways are used to process tactile information (Haidarliu et al. 2008; Veinante et al. 2000; Yu et al. 2006), it is conceivable that these encoding paradigms are used by the rest of the brain for different processes and at different time scales, depending on context and conditions. Each of these codes provides significant information about whisker velocity and thus, in principle, may guide sensory perception in behaving rodents.

In many situations, the rodents actively move their whiskers, thereby setting whisker speed. The resulting neuronal responses can serve as both representations of the tactile environment and feedback signals that may convey the actual whisker velocity to the system, during either whisking in the air or contact with an object. These conjectures are consistent with studies which suggest that
somatosensory cortex responses are transient, and are the most sensitive to whisker velocity (Pinto et al. 2000), and that spiking on the ascending pathway is precise and carries information about detailed features of the trajectory (Hipp et al. 2006; Jones et al. 2004; Petersen et al. 2008).

Our findings support the notion that the whisker somatosensory system is able to extract transient kinematic events, in addition to the total power and spectral frequency of whisker vibrations (Arabzadeh et al. 2003; Hipp et al. 2006). These findings fit well with several papers by both the Schwarz group (Gerdjikov et al. 2010; Schwarz et al. 2010; Stuttgen et al. 2006; Waiblinger et al. 2013), and Adibi et al. (Adibi et al. 2012), which linked a high level of psychophysical performance in rodents to the presence of transient kinematic signals.

Methodological considerations

To explore the transformation of tactile features into transient kinematic variables of whisker vibrations, we used artificial whisking, which generates whisking-like movement of the vibrissae. However, it should be borne in mind that the actual displacement generated by electrical whisking is not the same as naturally-occurring whisking (Berg and Kleinfeld 2003). Yet although whisking movements during artificial and natural conditions may differ in detail, artificial whisking does induce mechanical interactions between whiskers and objects which are within similar ranges under both conditions, as well as in head-fixed untrained rats (Bagdasarian et al. 2013; Lottem and Azouz 2009; Ritt et al. 2008). More importantly, several studies have shown that the somatosensory system of rodents can use the amplitudes of forces in the sensory follicles for tactile sensation to estimate radial object distance (Bagdasarian et al. 2013; Pammer et al. 2013). Therefore, it is possible that whisker velocity may be one of several variables used to sense the environment.

Our study relied on a set of applied stimuli (brief, individual deflections with different velocities), whereas in many situations, the system operates with continuous dynamic whisker
stimulation or self-induced whisker movements. We suggest however that the results of the current study are applicable to numerous situations in which discrimination of whisker velocity is required (Fig. 1).

Other studies have reached very different conclusions regarding the roles of RA and SA neurons, when more complex stimuli were used. For example, Jones et al. (Jones et al. 2004) have shown that RA neurons are as efficient in encoding velocity as SA neurons when stimulated with naturalistic noise waveforms. In contrast, RA neurons were shown to be less suited to velocity discrimination than SA neurons in response to constant stimuli since, as their name implies, they emit so few spikes in response to such stimuli.

Finally, we used anaesthetized animals. This procedure has its advantages, beside several disadvantages. While the magnitudes and correlations of cortical neuronal activity are significantly different from behaving animals, certain characteristics of cortical sensory processing, such as texture encoding, are consistent across various brain states, including anesthesia (Arabzadeh et al. 2005). Moreover, it has been shown that the transmission of tactile information in anaesthetized rodents can be very reliable in response to varying stimuli (see multiple references in the Introduction and Discussion sections). Hence, we believe that anesthesia does not play a major role in brainstem neuronal fidelity.

**Latency encoding**

The study reported herein is one of several studies that show a clear correspondence between cellular functionality and tactile information transmission. Specifically, RA neurons convey stimulus onset times and SA neurons communicate stimulus identity. First spike latency has been shown to transmit information in a variety of sensory modalities, including the auditory (Furukawa
and Middlebrooks 2002), visual (Gawne et al. 1996; Reich et al. 2001), somatosensory (Johansson and Birznieks 2004; Panzeri et al. 2001), and olfactory (Junek et al. 2010) systems.

However, it is still unclear whether the different sensory modalities can actually acquire an accurate measure of first spike latencies, since latency determination requires an estimation of stimulus onset from which to extract latency. Thus, a major feature of the temporal encoding scheme in general, and latency encoding in particular, is the requirement that in order to obtain stimulus identity (“what”) from the neural responses, *stimulus onset* times (“when”) must be used (Arabzadeh et al. 2006; Gawne et al. 1996; Meister and Berry 1999; Panzeri et al. 2001; Reich et al. 2001; VanRullen et al. 2005). However, with the exception of one clear instance (Johansson and Birznieks, 2004), an internal neural representation of the onset time remains unclear, a fact considered by many to be a major drawback of the temporal coding strategy (Arabzadeh et al. 2006; Chase and Young 2007; Phillips 1998; VanRullen et al. 2005). To overcome this obstacle, a number of possible mechanisms have been proposed, in which first spike latency information could be achieved by comparing the response times of a sub-ensemble of neurons to the mean response times of the entire population (Zohar et al. 2011). Other studies have used relative spike latencies or rank order codes to estimate the time of stimulus onset (Furukawa and Middlebrooks 2002; Gollisch and Meister 2008).

Neuronal noise and temporal encoding

Our results suggest that a population code based on spike latencies can be a powerful mechanism for the rapid and reliable transmission of tactile information. Neurons responsible for early sensory processing have been shown to evoke reliable and temporally precise patterns of spikes (Buracas and Albright 1999; Petersen et al. 2002a). These first-order spikes are transmitted through highly efficient synapses to the thalamus and the cortex (Deschenes et al. 2003). Thus, our results mesh
well with the literature and suggest that temporal response properties of the various neuronal subtypes can be utilized to code for whisker velocity.

However, as we have shown, “when” and “what” signals may be prone to errors. Onset estimation in RA neurons may deteriorate because of several factors: (a) Detection errors. As whisker velocity decreases, the proportion of activated RA neurons decreases as well, and eventually may fail to signal the presence of a stimulus (Fig. 1E). We have shown that even at the lowest velocity examined, a significant proportion of RA neurons still discharge (Fig. 2E). (b) The temporal fidelity of the estimation. The onset signal has to be independent of stimulus identity. We have shown, using the slope of the plot of RA first spike latency dependence on velocity, that first spike latency in most TG RA neurons is independent of whisker velocity (Fig. 1E, 2D), while first spike latency estimation in brainstem nuclei is noisier (Fig. 7). However, by using a population of RA neurons or velocity-independent neurons in first- and second-order neurons, we were able to reduce the small jitter in the onset signal. The introduction of low-level noise did not result in TG and brainstem RA neuronal firing, showing them to be robust and reliable onset detectors. In contrast to the onset signal, the deterioration in velocity discrimination is mostly influenced by the increase in the “jitter” of first spike latency signals of SA neurons (Fig. 7), which defines the basic temporal encoding strategy.

A major finding of this study is that a large deterioration of velocity discrimination occurs at the second stage, both for relative timing and firing rate signals, making them coequal candidates for velocity discrimination (Fig. 7). This large deterioration in relative timing signal contradicts previous studies, which have shown that the neuronal responses in the somatosensory system are precise and reliable throughout, and can faithfully follow the temporal structure of the stimulus (Deschenes et al. 2003; Petersen et al. 2008). To resolve this discrepancy we compared the responses to both stimuli and found that the deterioration of temporal encoding is dependent on
whisker velocity in SA neurons (Fig. 8A). The transmission between first- to second-order neurons plays a critical role in this deterioration (Fig. 8B). Using naturalistic noise stimuli significantly reduced first spike latency jitters, indicating that temporal encoding is stimulus-dependent and therefore more suited for the representation of continuous whisker signals (Buracas and Albright 1999; Petersen et al. 2002a). The mechanisms underlying these differences lie beyond the scope of the present study. However, several findings in the literature suggest that inhibitory interneurons may play a role in reducing spike jitter (Gabernet et al. 2005; Pouille and Scanziani 2001; Wilent and Contreras 2004), and that this inhibitory control may be stimulus-dependent.

**Readout Mechanisms**

Several decoding mechanisms that rely on determining precise relative timing of first spikes may require the integration of “when” and “what” signals. One of them involves neurons that can be “tuned” to respond to a wide range of specific durations (Hooper et al. 2002). Through this mechanism, the temporal sum of the synaptic inputs of such a neuron corresponds to its firing rate (Loewenstein and Sompolinsky 2003). Within this framework, neurons can also integrate the two signals through mechanisms of coincidence detection. Owing to both their intrinsic and synaptic characteristics, such neurons could be sensitive to particular temporal patterns in their inputs (for example, the time interval between two afferent inputs), thereby converting a temporal representation of the stimulus into a firing-rate-based one (Azouz and Gray 2003; Polsky et al. 2004).

A second class of mechanisms that can recognize specific temporal firing patterns may be implanted through supervised learning rules in which a neuron is able to distinguish between different temporal input patterns. This is accomplished through the “tempotron” learning rule, which adjusts the synaptic efficacy of the different inputs. Through such learning rules, neurons can
“learn” to categorize a broad range of input classes, as characterized by the latencies of single spikes (Gutig and Sompolinsky 2006).

Where does this integration occur? Based on Figure 7 it seems likely that PrV neurons code mostly SA signals (i.e., velocity), and that SpVi neurons contain both RA and SA signals. If so, the integration is likely to occur downstream to these neurons. Several findings in the literature and our own observations show that TG neurons are sensitive to stimulus kinetics such as velocity, position, and acceleration (Bale et al. 2013; Petersen et al. 2008), and that these neurons correspond to SA_{lt}, SA_{ht}, and RA neurons, respectively (manuscript in preparation). These channels remain mostly separated in the thalamus (Petersen et al. 2008), suggesting that the signals were generally kept separate up to the thalamus. We conjecture that upon arrival at the cortex, these channels converge, since numerous barrel cortical neurons are sensitive to a combination of multiple kinetic features (Maravall et al. 2007). An alternative approach was shown by (Chagas et al. 2013), in which each TG neuron contains a combination of several kinetic features, suggesting that this integration may occur at earlier stages.
Figure Legends

Figure 1 The effects of object distance and velocity on kinematic variables in passive and artificially whisking by anesthetized rats. (A) An object touching the whisker at a constant distance and moving at different velocities. The middle panel shows the angular position of a single whisker during object-touching by the whisker at different velocities (the shaded area represents the 20 msec borders). The right-hand side shows the effect of object velocity on whisker velocity. (B) A grid of object positions. The arrow indicates the direction of each object’s motion. The middle panel shows the angular position of a single whisker during object-touching at different locations. The right-hand side shows the effect of object location on whisker velocity. (C) A grid of object positions. The curved arrow indicates the direction of whisker protraction. The middle panel shows the angular position of a single whisker during a single whisk in free air and against an object located at different radial distances. The right panel shows the influence of object location on whisker velocity. (D) Responses of the three types of neurons to step stimuli. TG neurons are divided into SA_{lt}, SA_{ht}, and RA types. (E) Responses of RA, SA_{ht}, SA_{lt} neurons to increasing whisker velocity (bottom panel). The right panel shows raster plots and the left one, PSTHs of the three neuronal subtypes. The scale bars in the PSTHs indicate response probability (see Methods section). Each PSTH row corresponds to a different velocity. Averaged data are expressed as mean ± SE.

Figure 2. The effect of whisker velocity on first spike latency, firing rate, and first inter-spike-interval in our three neuronal types. (A) PSTHs of a sample SA neuron’s responses to increasing whisker velocities. The gray and purple borders mark first spike latency and inter-spike interval, respectively. The color bar on the side indicates the mean firing rate of this neuron. (B) The effect of whisker velocity on first spike latency in our three neuronal subtypes. The dots indicate the lowest whisker velocity each neuron responded to (see Methods section). Thick colored lines
represent the medians of the neurons in each group. (C) The effect of whisker velocity on first spike latency $^{-1}$ in our three neuronal types in Figs. 1D, E. (D) Sigmoid fits for all neurons normalized to their maximum value. (E) The cumulative proportion of active units responding to the different velocities. (F) The same as B for neuronal firing rates. (G) The same as C for neuronal firing rates. (H) The same as D for neuronal firing rates. (I) The same as B for first inter-spike-interval. (J) The same as C for first inter-spike-interval$^{-1}$. (K) The same as D for first inter-spike-interval$^{-1}$.

**Figure 3.** First spike latency of RA neurons is dependent on whisker velocity when the stimulus is filtered. (A) Sample PSTHs in response to three whisker velocities (unfiltered on the left; filtered on the right). (B) The effect of whisker velocity under the same conditions.

**Figure 4.** Velocity discrimination using ROC analysis. (A) An ROC curve used to discriminate between the two velocities marked by arrows in Fig. 2A (859 deg/sec and 1050 deg/sec). The different colored curves above the diagonal show the curves corresponding to each of the neuronal features. (B) AUC values, using first spike latency $^{-1}$, for all pairs of velocities (the black square corresponds to the AUC of the first spike latency ROC in B); (C) Averaging all diagonals results in a discriminability plot.

**Figure 5.** Velocity discrimination using the neuronal response features in our three neuronal types. (A) Discriminability plot for the three neuronal features (First inter-spike interval,; First spike latency; Firing rate) for SA neurons. (B-D) Discriminability plots for these three features, further divided among the three neuronal types. Averaged data are expressed as mean ± SE.
Figure 6. Velocity discrimination using relative timing between SA and RA discharges. (A) Sample probability distributions of RA and SA first spike latency for each velocity. The shaded area marks the relative time interval between the two distributions. (B) The effect of whisker velocity on first spike latency when using the 5<sup>th</sup>, 32<sup>nd</sup>, and 53<sup>rd</sup> RA percentiles (see Methods). (C) The effect of ensemble RA spike percentile on discriminability. (D) A discriminability plot using the different neuronal features of all SA neurons. (E) A discriminability plot using the various neuronal features of the two neuronal SA subtypes. Averaged data are expressed as mean ± SE. The scale bars in the PSTHs indicate response probability.

Figure 7. Brainstem neurons, divided into SA and RA types. (A) Lesions along the electrode track (arrows) in the different brainstem areas. (B) Responses of RA and SA neurons to step stimuli and increasing whisker velocity. The scale bars indicate response probability. Each PSTH row on the left corresponds to a different velocity. (C) Proportions of the different neuronal subtypes for TG and brainstem neurons. (D) The effect of whisker velocity on first spike latency in the two neuronal subtypes of the PrV. (E) An estimate of velocity discrimination deterioration in brain stem nuclei based on a calculation of the ratios between the discriminability values of relative timing and firing rate in brainstem nuclei and TG neurons. Relative timing – RT; Firing rate – FR. Averaged data are expressed as mean ± SE. The scale bars in the PSTHs indicate response probability.

Figure 8. Deterioration in first spike latency jitter. (A) The effect of whisker velocity on first spike latency jitter in SA neurons in the TG, PrV and SpVi areas. (B) First spike latency jitter in RA neurons in the same areas. (C-D) Responses of two representative TG and PrV neurons to white noise stimuli. The scale bars indicate response probability. (E) First spike latency jitter in SA neurons in the same areas in response to white noise stimuli. Horizontal lines indicate the values
taken from the same neurons responding to a ramp stimulus. (F) First spike latency jitter in RA neurons in the same areas in response to white noise stimuli. Horizontal lines indicate the values taken from the same neurons responding to a ramp stimulus. Averaged data are expressed as mean ± SE. The scale bars in the PSTHs indicate response probability.

**Figure 9.** Velocity discrimination in the brainstem with the functional classification of “when” and “what” signals. (A) A discriminability plot using the different neuronal features in SA neurons in PrV, and SpVi areas. The threshold latency range for the “when” neurons was set to 4.4 msec. (B) The ratio between the discriminability plot of relative timing and firing rate of SA neurons in PrV and SpVi neurons against the latency range threshold. (B) The proportion of RA neurons out of functionally classified “when” neurons as a function of threshold and area. Averaged data are expressed as mean ± SE.

**Figure 10.** Velocity discrimination using relative timing is resistant to noise. (A) A probability distribution for each velocity of RA and SA first spike latency during control (upper panel) and during the addition of filtered noise whisker vibrations (lower panel). (B) A discriminability plot using the various neuronal features in all SA neurons under control conditions (continuous) and during the introduction of noise (dashed) in TG neurons. (C) The ratios between the discriminability plot of control and noise conditions of the three neuronal features in the recorded areas. Averaged data are expressed as mean ± SE. The scale bars in the PSTHs indicate response probability.
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**Wolfe J, Hill DN, Pahlavan S, Drew PJ, Kleinfeld D, and Feldman DE.** Texture coding in the


A.

Regular stimuli

Filtered stimuli

B.

Latency (ms)

Velocity (deg/sec)

- Regular stimuli
- Filtered stimuli

- 757 (deg/sec)
- 1123 (deg/sec)
- 1514 (deg/sec)
Figure A shows the discriminability of PrV and SpVi neuronal responses to different velocity changes. The discriminability is plotted against velocity (Δ velocity, 10^3 deg/sec) with error bars indicating variability. The latency range threshold for both PrV and SpVi is 4.4 ms.

Figure B compares the relative timing and firing rates of PrV and SpVi. The relative timing/firing rates are plotted against latency threshold (ms) with increasing latency thresholds (from 0 to 14 ms).

Figure C shows the proportion of RA (relative activity) over latency threshold (ms) with a peak at a latency threshold of 2 ms before decreasing.