Mental arithmetic leads to multiple discrete changes from baseline in the firing patterns of human thalamic neurons.

Dept. of Neurosurgery, Johns Hopkins Hospital, Baltimore, MD. 21287-7713

Running Title: Altered human thalamic activity during arithmetic.

Key words: Attention, human thalamus, calculation, single neuron analysis, thalamic low threshold spike.

Address all correspondence and proofs to:
F. A. Lenz
Department of Neurosurgery
Meyer Building 7-113
Johns Hopkins Hospital
600 North Wolfe Street
Baltimore, MD  21287-7713

Telephone    410 955 2257
FAX          443 287 8044
E-mail     flenz1@jhmi.edu

Acknowledgements: Supported by grants to FAL from the Eli Lilly Corporation and the NIH (NS 383493, NS 40059). We thank L.Rowland for excellent technical assistance.
ABSTRACT

Primate thalamic action potential bursts associated with low threshold spikes (LTS) occur during waking sensory and motor activity. We now test the hypothesis that different firing and LTS burst characteristics occur during quiet wakefulness (spontaneous condition) versus mental arithmetic (counting condition). This hypothesis was tested by thalamic recordings during the surgical treatment of tremor.

Across all neurons and epochs, preburst interspike intervals (PBISIs) were bimodal at median values consistent with the duration of GABAa and GABAB IPSPs. Neuronal spike trains (117 neurons) were categorized by joint interspike interval distributions into those firing as LTS bursts (G, grouped), firing as single spikes (NG, non-grouped), or firing as single spikes with sporadic LTS bursting (I, intermediate).

During the spontaneous condition (46 neurons) only I spike trains changed category. Overall, burst rates (BR) were lower and firing rates (FR) were higher during the counting versus the spontaneous condition. Spike trains in the G category sometimes changed to I and NG categories at the transition to the counting condition, while those in the I category often changed to NG. Among spike trains which did not change category by condition, G spike trains had lower BRs during counting, while NG spike trains had higher FRs. BRs were significantly greater than zero for G and I categories during wakefulness (both conditions). The changes between the spontaneous and counting conditions are most pronounced for the I category which may be a transitional firing pattern between the bursting (G) and relay modes of thalamic firing (NG).
INTRODUCTION:

Thalamic low threshold spike (LTS) bursts which follow an inhibitory event have long been associated with behavioral states such as slow wave sleep (McCarley et al. 1983; Domich et al. 1986; Steriade et al. 1997a). More recent reports demonstrate that LTS bursts occur during wakefulness in humans (Zirh et al. 1997; Radhakrishnan et al. 1999; Jeanmonod et al. 1996) and in monkeys (Ramcharan et al. 2000a; Ramcharan et al. 2005). In addition to the association between LTS bursting and state, there is now evidence that thalamic LTS bursting can increase from baseline during sensory stimuli or motor events (Ramcharan et al. 1998; Ramcharan et al. 2000b; Ramcharan et al. 2001; Martinez-Conde et al. 2002; Lee et al. 2005). In monkeys, thalamic LTS bursts occur in neurons in the lateral geniculate nucleus following microscopic eye movements (microsaccades) which occur during visual fixation (Martinez-Conde et al. 2002; Lu et al. 1992). This activity may be transmitted to V1 cortex where bursts of action potentials are likewise related to microsaccades (Martinez-Conde et al., 2002; Martinez-Conde et al. 2000).

Microstimulation of somatic sensory thalamus with pulse trains like those of LTS bursts will evoke the sensations produced by stimuli which evoke LTS bursts (Lenz et al. 2004; Lee et al. 2005; Patel et al. 2006). Therefore, LTS bursts in primates may be related to sensation and to waking behaviors, as well as to behavioral states.

The effect of cognitive tasks upon spike train characteristics of primate thalamic neurons has not previously been studied, to our knowledge. In rabbits, random increases in the level of vigilance are associated with decreased thalamic neuronal burst rates and decreased responses of thalamic neurons to peripheral stimuli (Bezdudnaya et al. 2006). We now test the hypothesis that different firing and LTS burst characteristics occur during quiet wakefulness (spontaneous condition) versus mental arithmetic (counting condition).
The results demonstrate the presence of three types of spike trains including those with frequent LTS burst firing (G grouped category), or with tonic firing rate (NG non-grouped category). The third spike train category (I intermediate) was characterized by bursts interspersed between single spike firing with variable interspike intervals. The I category was more likely than the others to change category during both the spontaneous condition and the transition between the spontaneous and the counting condition. Among neurons which did not change their category between conditions increases in FRs and decreases in BRs were due to changes in the activity of NG and G neurons, respectively. The characteristic differences between neurons which do or do not change categories suggests that several discrete neuronal mechanisms contribute to the transition between the spontaneous and counting conditions.

MATERIALS AND METHODS

These studies were carried out at the Johns Hopkins Hospital (2003-2007) during the physiologic exploration of the thalamus which preceded implantation of deep brain stimulating (DBS) electrodes in Vim for treatment of essential tremor (ET)(Koller et al. 2001). ET is a monosymptomatic illness characterized by a 4 to 7 Hz postural or action tremor which is often familial, and which can be ameliorated by ingestion of alcohol (Deuschl et al. 1998). In this study, exclusions included patients who had neurologic symptoms in addition to the tremor which is characteristic of ET. The exclusions included patients with both essential and Parkinson’s tremor, or patients with the tremor characteristic of ET in addition to torticollis, or a cerebellar syndrome (Jankovic 2002). A neurologist specializing in movement disorders confirmed the diagnosis of ET and identified patients to be excluded. The protocol was reviewed and approved annually by the Institutional Review Board of the Johns Hopkins University. All patients signed an informed
consent prior to participation in these studies. All methods used here have been previously described (Lee et al. 2005; Ohara et al. 2007).

**Intraoperative procedures**

Prior to the procedure patients were been off all tremor medications for 18 hours. The thalamic exploration was performed as a stereotactic procedure using the Leksell frame. First, the frame coordinates of the anterior (AC) and posterior (PC) commissures were measured by magnetic resonance imaging or computed tomography. These coordinates were then used to estimate the nuclear locations. Physiological corroboration of nuclear location was then performed under local anesthesia (i.e. subject fully conscious) by both single unit recording and stimulation with a microelectrode. The microelectrode was advanced through a burrhole 2.5 cm off the midline just anterior to the coronal suture. The initial trajectories were always focused on Vc because the response of neurons in this area to somatosensory stimulation provides the most reliable landmark to guide the procedure (Lenz et al. 1995; Garonzik et al. 2002). Along these trajectories, stimulation sites with thresholds < 25 µA were correlated with the presence of neurons with tactile receptive fields, which are found in Vc (Lenz et al. 1988b; Lenz et al. 1994). Therefore, stimulation confirms the location of the anterior border of Vc as determined by recordings. Surface electromyographic (EMG) signals were recorded from flexors and extensors of the wrist and elbow. The signal from a foot pedal was used to indicate the timing of sensory stimuli or verbal commands.

The counting condition involved mental arithmetic during which the patient carried out a serial subtraction task. The patient first subtracted the number 7 repeatedly from 100, i.e. 100, 93, 86, etc. If the patient was unable to carry out the serial subtraction task with the number 7, then a task based upon 6 was attempted and so on through the series 7, 6, 4, 3, 2, and 1 until the patient’s
error rate was less than approximately 10%. This is a complex cognitive task which engages attention, working memory and calculation (Lezak 1995).

The protocol for each cell began with an epoch of the spontaneous period lasting between 60 – 180 seconds (average approximately 80 seconds), followed by a counting epoch of approximately the same duration, followed by another spontaneous epoch, and so on to a maximum of 8 epochs. Therefore, a series of 8 epochs would have a duration of approximately 640 sec. The analysis of spontaneous periods was carried out between epochs separated by a counting epoch. Analysis of firing and bursting activity for a single cell including stationarity was carried out between these spontaneous epochs as well as between spontaneous and counting epochs in a complete spike train record.

The behavioral state is well known to change thalamic bursting activity so that LTS bursting increases during drowsiness or sleep (Domich et al. 1986; Steriade et al. 1997b). Therefore we observed the subjects’ behavior and measured scalp EEG or thalamic local field potentials (LFP) or both intraoperatively during spontaneous activity and multiple cognitive tasks, in addition to the present task. Any change in these measures of state from wakefulness to drowsiness or sleep is very rare in our experience (Lenz et al. 1993; Lenz et al. 1994; Zirh et al. 1997). This may be due to the stress of an awake operation and to regular stimulation by the anesthesiologist, the surgeon, the monitoring nurses, and the monitoring devices (blood pressure cuff and the pulse oximeter).

**Study of Thalamic Activity:**

When a neuron was first isolated, spontaneous activity was recorded briefly. The activity of that neuron was then studied to identify neurons responding to light touch, tapping or pressure to skin (cutaneous sensory neurons), as well as deep stimuli such as pressure to muscles or ligaments.
and passive joint movement (deep sensory neurons). These neurons were characterized by a neuronal receptive field (RF). The activity of neurons was also examined as patients carried out movements such as making a fist, flexing or extending the wrist and elbow, pointing, etc. We carried out microstimulation along each trajectory.

As previously described, the anterior and inferior borders of Vc were identified by the most anterior and the most inferior deep or cutaneous neuron in the region where the majority of neurons responded to either deep or cutaneous stimuli (Ohara and Lenz 2003; Lee et al. 2005). These points and the location of the ACPC line were used to overlay the map of the recording data with a standard, sagittal atlas map (Schaltenbrand and Bailey 1959). The nuclear location of any recorded neuron was determined from this overlay. All neurons were located in one of the following thalamic nuclei: ventral oral (Vo) ventral intermediate (Vim), ventral caudal (Vc) and lateral dorsal (LD).

**Data Collection and Analysis:**

The signals recorded on a digital system (Neuroguide, Alpha-Omega, Nazareth, Isreal) during the procedure included: the foot pedal indicating events during the examination, the microelectrode signal, electromyograms (EMGs), and the audio channel describing instructions to the patient, application of stimuli, etc. Digitally recorded microelectrode signals were analyzed using Spike2 software (CED, Cambridge, UK) by template-matching of action potential waveforms. Subsequent analyses of spike characteristics such as burst detection, firing rates and duration of identified interspike intervals were carried out using MATLAB (Mathworks, Natick, MA). Analysis of bursting was carried out on spontaneous activity with eyes open and with the arm
at rest. The absence of tremor was confirmed by visual inspection of both the limbs and the EMG signals.

**Analysis of Burst Activity**

As a bias free measure of neuronal bursting activity, we constructed plots of the interspike interval before an action potential in a spike train (n) versus the interspike interval after (n+1), designated as a n versus n+1 plot. This plot provides a graphical display of the characteristics of the spike train overall as reflected by clusters of points in the plot. In the n versus n + 1 plot the cluster at the lower right indicates a long interspike interval (approximately 300 to 400ms, see Figure 1B upper) followed by a short interspike interval (< approximately 5 ms), the defining characteristic of the first spike in an LTS burst. The cluster at the upper left indicates a short interspike interval (< approximately 16 ms) followed by a long interspike interval (approximately 100ms), so that the short interspike interval is the final interspike interval in an LTS burst. The lower left and upper right clusters indicate the interspike intervals within and between bursts, respectively. This plot indicates spike train characteristics overall; it does not allow the identification of individual LTS bursts, an example of which is shown in the inset of Figure 1A (G spike train).

Individual LTS bursts in the spike train were identified by criteria used in studies of awake cynomologous monkeys which have been confirmed by intracellular studies in thalamic slices from the same species (Ramcharan et al. 2000a;Ramcharan et al. 2000b). The criteria (50-6-16) to select these bursts from the spike train were as follows: the interspike interval preceding the first action potential in the burst had a duration > 50 ms, the interspike interval following the first action potential in the burst had a duration of < 6 ms, and all following action
potentials are considered as part of the burst if their interspike interval increased by no more than 2 ms for each succeeding action potential, up to maximum interspike interval of 16 ms. All bursts meeting these criteria, including bursts composed of two action potentials, were included in the burst rate. While the n versus n+1 plot is a bias free way to assess a long spike train it is less accurate than the template method for identification of individual bursts. Therefore the template method is used to determine burst parameters, but not to classify neuronal spike trains. We have repeatedly validated, illustrated and published these techniques and these burst criteria (Lee et al. 2005; Lenz et al. 1998; Lenz et al. 1994).

We also measured the primary event rate (PR) which includes all spikes occurring outside bursts plus the first spike in each burst defined by the criteria above. Therefore, the PR is a measure of the rate of spikes occurring between bursts (McCarley et al. 1983; Cox and Lewis 1966). We calculated burst rate/primary event rate (BR/PR) as an indicator of the relationship between burst firing and ongoing neuronal firing, as reflected by the PR. If the difference in burst rates between two neuron types is lost when the ratio of BR/PR is calculated, then the difference in bursting is dependent on the PR, and perhaps on the degree of membrane depolarization. A measure of neuronal firing which is related to the BR/PR is the percentage of action potentials occurring in bursts (BP). The number of action potentials occurring in bursts included bursts of two action potentials and all action potentials in a burst, including the first and last action potentials.

As indicators of the preburst inhibition, we measured the preburst interspike interval (PBISI) and the ratio of the PBISI/ PRISI (inverse of the primary event rate). The PBISI is an indicator of the duration of the inhibitory event preceding a burst, while the ratio is an indicator of
the strength of the preburst inhibition. All firing, burst and inhibitory indexes were compared by locations in the thalamus and between spontaneous and counting conditions.

**Statistical analysis**

Parametric measures of the spike train were compared between different groups and categories by an ANOVA, with post hoc testing including a correction for multiple comparisons (Tukey honestly significant difference, HSD). Parametric data that were not normally distributed were tested by Mann-Whitney or Kruskal-Wallis, and descriptive statistics were expressed as the median and 5 - 95%ile of the distribution (Confidence interval of the median, CI). Paired analysis was carried out using the nonparametric Wilcoxon matched pair test (Snedecor and Cochran, 1967). Differences between proportions were tested with a Chi square or Fisher test, as appropriate. The null hypothesis was rejected at P<0.05 in all statistical tests.

As a test of the reliability of visual classification of neuronal spike train categories, the Calinski statistic was used to estimate the number of clusters (k) for a given spike train (Weingessel et al. 1999). For any number of clusters (k) large values of the Calinski statistic are evidence for that number of clusters, where this statistic is similar to an F statistic in an analysis of variance. The value of the k statistic is measured for a number of values of k, and the largest k value indicates the number of clusters.

**RESULTS.**

**Thalamic neuronal firing patterns:**

These results were obtained in 28 patients who were between 41 and 79 years old, and who were comprised of 12 women and 16 men. Surgery was carried out on the left thalamus in 18
patients; the right in 10. For each patient, between 1 and 6 trajectories were explored (mean 3.3). A total of 117 neurons (Lateral Dorsal 13, Vo 36, Vim 42, Vc 26) were studied during periods of up to 749 sec.

Each row in Figure 1A shows examples of the firing of one neuron in each category, as labeled. Each row in 1B shows four examples of n versus n+1 plots for each of the three categories, with rows corresponding to categories as labeled in panel A. The left n versus n+1 plot in each row of 1B is derived from the spike train shown in the corresponding row of panel A. Neuronal spike trains were classified by the category of the first spontaneous epoch.

In Figure 1A, note that the dots above the digitized G and I category spike trains indicate the occurrence of an LTS burst as identified by the 50-6-16 criteria (see methods). In Figure 1B top row, the cluster of dots in the G category at the lower right of the n versus n+1 plot indicate a long interval (> 100ms) followed by a short interval (< 6 ms), the defining characteristic of the first spike in an LTS burst. The cluster at the upper left of the n versus n+1 plot (Figure 1B, upper row) indicates a short interspike interval (< 16 ms) followed by a long interspike interval (> 100ms), characteristic of the final interspike interval in an LTS burst. The lower left and upper right clusters of the n versus n+1 plot (Figure 1B, upper row) indicate the interspike intervals within and between bursts, respectively. Therefore, spike trains in the G category (Figure 1A and B: top row) were identified from n versus n+1 plots by the presence of well-defined clusters at the four corners of the n versus n+1 plot.

Spike trains in the NG category showed high frequency spontaneous firing during the spike train (Figure 1A, middle row) resulting in n versus n+1 plots characterized by a central cluster (Figure 1B, middle row). This distribution is consistent with the lack of LTS bursts as indicated by the small number of clusters at the four corners of the n versus n+1 plot, particularly the cluster the
lower right (Figure 1A, right middle row). Nevertheless some of these plots do show data points at
the lower right consistent with doublet LTS bursts, e.g. Figure 1B, Row 2, third and fourth plot
from the left.

Spike trains in the I category had irregular spike firing, and common LTS bursts (Figure 1A, 
bottom row). Spike trains in the I category were identified by two or more clusters (Figure 1B, 
bottom row) which were less well defined than, but consistent with, those found in the G category 
(Figure 1B, top row). These clusters in the I category (Figure 1B, bottom row) were often in the
lower right, or the lower left, or both, approximately consistent with the clusters of the G category.
The I category spike trains (Figure 1B, bottom) often had a central cluster, consistent with that 
found for the NG category (Figure 1B, middle row). Neuronal spike trains in the G and NG 
category always maintained their category across all epochs of spontaneous activity (see Results:
Spike train properties during the spontaneous condition), while I category spike trains often
did not change category between spontaneous epochs (Figure 4).

In categorizing these results we relied on visual assessment of the n versus n+1 plots. 
This assessment was checked against the results of cluster analysis by employing the Calinski
statistic (see Methods: Statistical analysis). Six representative neuronal spike trains, identified 
visually as being in the G category, were studied by cluster analysis. Of these, five were found
to have four clusters. One of these neuronal spike trains had approximately equal likelihood of 
four or five clusters (spike train: 490hl65newsp2), with the possible fifth cluster in the center of
the four clusters characteristic of the G category. The BP for this cell was 39% which is greater
than the BP for 95% of I cells but is at the median value for G cells, confirming visual
identification of this spike train in the G category.
Four I spike trains were studied by cluster analysis and were found to have three or four clusters, including a central cluster with 2 or 3 peripheral clusters. These were usually located the lower right and left of the n versus n+1 plot.

The Calinski statistic was calculated for four distributions in which there were possible second and third clusters just peripheral to a central cluster. The central cluster was in a location similar to the G category (spike train: XS148013). In none of these cases was the visual classification changed by cluster analysis. Overall, cluster analysis of a sample of spike trains in this dataset confirmed the results of visual classification.

**Distributions of Burst Parameters.**

In order to study the properties of individual bursts we identified the LTS bursts by the 50-6-15 criteria during the spontaneous condition (117 neurons)(Methods: Analysis of Burst Activity). Along the diagonal in Figure 2 are histograms of burst parameters expressed as logarithms to the base e (natural logarithms, log). These histograms show the histogram of log values for the measures of firing (FR, BP), the bursting (BR, BR/PR), and inhibitory events (PBISI, PBISI/PRISI). Note that the distributions seem to be bimodal for the log FR, log BR/PR, log PBISI, and log BP, while the distributions seem to be unimodal for log BR, log PBISI/PRISI.

In order to test the possibility that these distributions were best described by more than one peak, we used the technique of the Bayesian information criterion (BIC)(Schwarz 1978). BIC analysis was used to identify whether these distributions are actually a mixture of two different components. We considered the possibility that these variables were described by a single distribution (1 component model), a mixture of two distributions (2 component model), a mixture of
3 distributions, and so on. For any two estimated models, the one with the highest value of the BIC is identified as the optimal number of components contributing to the histogram. The analysis provides the estimated mean (and standard deviations) for each identified component. For distributions with two components these measures indicate the probability that the two components are separate, and that a single observation belongs to the first group or the second group.

This analysis was carried out on all epochs from all neurons and demonstrated that the distribution of log(mPBISI) had two peaks (Abbreviations defined in Methods: Analysis of Bursting activity). The two peak values of the PBISIs were 97ms and 403ms. These are approximately consistent with the duration of GABAa and GABAb IPSPs for thalamic relay neurons (Andersen and Sears, 1964; Roy et al. 1984; Soltesz et al. 1989; Sherman and Guillery 2001).

Graphs of each variable against the others are shown for individual neurons in the G category (red circles), NG category (blue crosses) and I category (green triangles) along the diagonal in Figure 2. The G and I categories had long log PBISIs which were associated with the high BRs (Figure 2, second row from the top, fourth graph from the left). The long log PBISIs were also associated with high percentages of interspike intervals in bursts (BP), which is characteristic of the G and I categories (Figure 2 fourth row from the top, right plot).

Two peaks were identified for the log FR with mean (standard deviation) frequencies of the two peaks being at 1.29/s and 2.73/s (Figure 2 left top histogram, Table 1). For the PR two peaks were apparent in the BIC analysis at log PR at 1.00/s and 2.71/s. The lower FR are associated with longer log PBISIs (Figure 2, top row, fourth graph from the left), and are consistent with the properties of G and I spike trains (Table 1).
The log BR was unimodal and had substantial overlap of the distributions for the different categories (Figure 2). A unimodal distribution with overlap of categories was also observed for the ratio of log PBISI/PRISI. These results demonstrate that the rate of inhibitory events (LTS burst rate), and the intensity of preburst inhibition (ratio PBISI/PRISI) consist of a single distribution for the different categories of spike trains.

**LTS bursting by 50-6-15 criteria in different categories.**

In this analysis, bursts identified by the 50-6-15 criteria were compared statistically between the three categories in the same set of neurons (n=117) as described above. This analysis confirms the presence of high burst rates in the G and I categories. In addition, some spike trains in the NG category have bursts of two action potentials; which is reflected in this analysis. As shown in Figure 3, FRs were significantly higher in NG category than either G or I category (both P<0.00001, Kruskal Wallis corrected for multiple comparisons); the PR which was also higher for the NG category than for G or I categories (both P<0.00001).

- place figure 3 about here -

BRs were significantly higher in G than in NG (P<0.0000001) or I categories (P<0.04). Analysis of the BR/PR did not lead to any change in the rank in categories with respect to BRs or the statistical significance for G versus NG or I categories. Therefore, the BRs were not dependent on a high FR, which could lead to identification of bursts resulting from random clustering of short interspike intervals. The 5th percentile of the confidence interval of the median of the BR was greater than zero for the G and I categories, but not for the NG category (Figure 3) which indicates that BRs were significantly greater than zero for G and I categories but not for the NG category.
PBISIs were significantly longer in G and I categories than in NG category (both $P<0.0000001$), although bursts did not occur commonly in the latter category (see above Results section: Thalamic neuronal firing … ). PBISIs were not significantly different between the G and I categories ($P>0.05$). The duration of PBISIs were greater than 100 ms for all categories: G (362 – 500 ms, 5 – 95% confidence interval of the median), NG (267 – 519), and I (99 – 136), consistent with latencies required to deinactivate the LTS (Sherman and Guillery 2001; Steriade et al. 1990).

The longer PBISIs might reflect slower firing rates between bursts rather than preburst inhibition. This possibility was assessed by examining the ratio of the PBISI/PRISI which will be greater than 1 if there is preburst inhibition. The confidence interval of the ratio was greater than 1 for I and NG neurons suggesting that the LTS bursts are the result of preburst inhibitory events. The ratio was significantly greater in the NG than in the G category ($P<0.0000001$) or I categories ($P<0.03$), which indicates that the occasional bursts during NG spike trains clearly follow an inhibitory event. The median of the confidence interval of this ratio was greater than 1 for G spike trains which have a very low FR. In this situation, bursting may result from tonic inhibitory drive or inhibitory events occurring in the presence of a relatively hyperpolarized resting membrane potential (Figure 3).

**LTS properties of neurons located in different thalamic nuclei.**

The properties of spike trains in different nuclei were studied during the spontaneous condition for 117 neurons. The 5% confidence interval of the median demonstrated that neurons in all nuclei had burst rates significantly greater than zero, i.e. all 5%ile confidence intervals > 0 (Table 2). The number of spike trains in the G category was significantly higher in the case of
neurons in Vc (13/26) than in Vim (5/42, P<0.002, Fisher) or Vo (7/36, P<0.001), but not Lateral Dorsal (4/13, P=0.31).

The number of spike trains in the NG category was significantly lower in the case of neurons in Vc (7/26) than in Vim (32/42, P<0.002, Fisher) or Vo (20/36, P=0.033), but not Lateral Dorsal (5/13, P=0.49, Fisher). The number of spike trains in the I category was not significantly different (P>0.31, Fisher or Chi Square) in the case of neurons in Vc (6/26) than in Vim (5/42) or Vo (9/36), and Lateral Dorsal (3/13). Therefore, the majority of neuronal spike trains in Vc were category G while the majority in the Vim and Vop were category NG and spike trains in category I were equally as common in Vc, Vim and Vo.

- place Table 2 about here -

Neurons in the G category had lower FRs and PRs than the NG or I category. Therefore, it is not surprising that FR were significantly higher in Vim, than in the Dorsal Lateral (P<0.04, Fisher), Vo (P<0.001) and Vc (P<0.005), with more spike trains in the G category. PRs were significantly higher in Vim than in Vo (P<0.005), and Vc (P<0.0001). The FRs in Vo were uniformly less than 50/sec (cf (Smith and Sherman 2002)).

Neurons in Vc had higher burst rates that those in Vo (P<0.0001, Kruskal Wallis) and Vim (P=0.044) but not LD (P=0.54). Neurons in Vc had burst rate/primary event rate ratios which were significantly higher than among those in Vim and Vo (P<0.005 in both cases). The PBISI were significantly (P<0.05) shorter in Vim than in Vc, perhaps consistent with the higher firing rates in Vim. The PBISI was short enough to be in the GABAa range for Vim (148 ms) while the PBISI was in the GABAb range for the rest of the nuclei (range 323 to 435 ms)(Andersen and Sears 1964;Roy et al. 1984;Soltesz et al. 1989;Sherman and Guillery 2001).
The PBISI/PRISI was smaller in Vim than in Vc (p<0.05). The CI of the median demonstrated that the PBISI was significantly greater than 100ms for neurons in all nuclei which is consistent with the length of the inhibition required to deinactivate the LTS (Jahnsen and Llinas 1984). The ratio of PBISI/PRISI was significantly higher in Vim than Vc. The median of the confidence interval was > 1 for the majority of neuronal trains in all nuclei, which indicates the presence of a significant preburst inhibition.

Differences in the number of interspike intervals within a burst (NSPK) were not significant between nuclei (P=1 for all comparisons between nuclei). The greatest number of interspike intervals per burst was found in Vc (2.6, 2.3 – 2.9: median: 5% - 95%ile), and the least in Vim (2.5, 2.2 – 2.7). The number of action potentials in a LTS burst is an indicator of the size of the LTS and the magnitude of the preburst inhibition (Steriade et al. 1990).

The number of neurons in the G category among nuclei in the lateral group containing a majority of cells with receptive fields (Vc) is uniformly greater than in the nuclei in which the majority of cells do not have a receptive field (Vim and Vo) (Lenz et al. 1988a). Among spike trains of all neurons with receptive fields the proportion of spike trains in the NG category (14/32 Fisher) was not significantly different from the proportion in I category (2/7, P=0.44, Fisher) and G category (2/14, P=0.092, Fisher) (Results section: Neurons which did not change Categories … ). Therefore, among cells with RFs 14 were in the NG category and 2 were in each of the G and I category. Since these results are obtained during spontaneous activity, these results suggest that the G pattern predominates in sensory nuclei and neurons in the absence of stimulation (Lee et al. 2005; Ramcharan et al. 2000b). Furthermore, prior studies demonstrate that somatic simulation uniformly results in increased firing and bursting rates (Lee et al. 2005).
Stationary behavior of spike train during the spontaneous condition.

A total of 46 neurons in 12 subjects were studied during multiple epochs of the spontaneous and counting task, so that each epoch had a duration of > 80 sec. Analysis of stationarity of LTS burst rates by median runs tests was carried out on a subset of 28 G and I spike trains during spontaneous and counting tasks (Snedecor and Cochran 1967). NG spike trains were not included in analysis of stationarity of the burst rate because they rarely fire in LTS bursts. The spike trains during the spontaneous condition were nonstationary for burst rates over 10 sec intervals in a distinct minority of spike trains (5/28, p=0.0005, Binomial). The number of nonstationary burst rates within spike trains was significantly less during epochs composed of spontaneous tasks exclusively (5/28, P<0.05, Fischer) than in those composed of both the spontaneous and counting task (13/28).

The stationarity of FRs over 5 second intervals was tested among all three categories. This analysis demonstrated that spike trains were commonly stationary (24/38) across spontaneous epochs. All nonstationary epochs in both FR and BR variables were characterized by fewer transitions from below to above the median, or visa versa, than expected at random. We next examined changes in category during the spontaneous condition among the spike trains with the greatest duration.

Changes in category during the spontaneous condition.

The n versus n+1 plots of spontaneous neuronal activity were studied over multiple epochs of at least 40 sec each, extracted from 21 spike trains recorded over periods of 6 to 12 minutes (369 to 749 s), which contained both spontaneous and counting conditions. This is a subset of the 46 neuronal spike trains (12 patients) that were studied during the spontaneous and counting conditions
over intervals of > 80 sec combined. In the G category, none of the eight spike trains changed their
category from that in the first spontaneous epoch. Five out of these eight neurons showed constant
n vs n+1 plots across the sampling period for spontaneous activity as in the figure 4, left. Three
neurons showed variability in the dispersion of the top right cluster, corresponding to variability
within intraburst interspike intervals (Figure 4A, second and third from the top). The four clusters
were seen clearly in all samples for each neuron. Therefore, the spike trains in the G category had
the same category during quiet wakefulness over intervals, spanning both conditions, of up to 12
minutes.

In the NG category, nine neuronal spike trains were studied, all of which were classified in
the NG category over all epochs of the spontaneous task across the complete spike train. For five
of these eight neuronal spike trains the n versus n+1 plot remained constant. In three, the large
central cluster migrated toward the origin (Figure 4B, middle and lower panel), and then away from
the origin of the plot. These changes were consistent (Figure 4C, top) with changes in the mean
firing rate (neurons 7929.2, 7929.1, 6734).

- place figure 4 about here -

Four neurons were classified in the I category, based on the first epoch of the spontaneous
condition of clusters in lower left and right corners of the n versus n+1 (Figure 4C upper and
middle) (see Results: Thalamic neuronal firing patterns). This pattern was found in all spontaneous
across the complete spike train for two neurons (378od11b.2, 540sz11.1). For the other two spike
trains in the I category, the lower left and right clusters (Figure 4 upper and lower) disappeared so
that the distribution assumed the appearance of the NG category (neuron 534cn8.2, see Figure 4C
lower, and 540sz11.2)(Lee et al. 2007). These changes were correlated with the numbers of bursts
in the spike trains which decreased in Figure 4B from the upper panel, to the middle and the lower
panel. Therefore, two of the I neuronal spike trains maintained their category and had a constant FR and BR, while two others changed their category.

The incidence of changes in spike train category was significantly higher for the I category (2/4) versus the G and NG categories combined (0/17, \( P=0.0286 \) Fisher). Therefore, the I category had greater variability of the firing activity than did NG or G categories as measured by median runs tests on FR and BR, and by changes in category between spontaneous categories in the complete spike train.

**Changes in Firing and Burst Rates at the transition between in Spontaneous and Counting Conditions Overall.**

Spontaneous versus counting condition were studied in the same 46 neuronal spike trains as described above. Figure 5 shows examples of spike trains recorded over epochs including both spontaneous and counting periods. The neuronal spike train shown in the upper panel was categorized as NG based on a single central cluster on the \( n \) versus \( n+1 \) plot. The lower panel in Figure 5 shows an example of a neuron categorized G based on the presence of the four clusters, one at each corner of the plot (Figure 1). After epochs had been classified into categories based on \( n \) versus \( n+1 \) plots, the burst parameters were calculated for bursts as identified by the 50-6-16 template. Note that the burst rate of the G spike train category decreased during counting, and the firing rate of neurons in the NG category increased during counting. We first examined these differences in LTS burst indexes among neurons overall.

- place Figure 5 about here -

Differences between LTS burst indexes were tested by a non-parametric analysis between spontaneous and counting conditions (Wilcoxon matched pair test)(Snedecor and Cochran 1967)).
The results demonstrate that the burst rate and percentage of interspike intervals occurring in bursts (or BR/PR) were less during the spontaneous conditions by approximately half (P<0.004, Table 3). The increase in firing rate of approximately one quarter during the counting condition showed a trend toward significance (P=0.09) overall. There were no significant changes in indices of inhibitory LTS associated events (PBISI and PBISI/PRISI). We next examined changes in the spikes train within neurons which changed their spike train category with the condition.

- place Table 3 about here -

**Spike Trains which changed Categories at the transition between Conditions.**

The upper panel of Figure 6 shows an example of a neuronal spike train categorized as I during the first counting epoch, based on poorly defined clusters including an absence of a right lower cluster on the n versus n+1 plot. This spike train changed to the G category for the first spontaneous period and the subsequent counting task period. The evidence of this change is based on the presence of well defined clusters at the four corners of the n versus n+1 plot, and particularly on the emergence of a right lower cluster (see Results: Thalamic neuronal firing patterns). In the second epoch of the spontaneous condition the peripheral clusters were less defined and a poorly defined central cluster emerged, leading to classification of this epoch in the I category.

The numbers of neuronal spike trains changing their category by the condition were significantly lower among neuronal spike trains categorized during the spontaneous condition as NG (0/18) than in those categorized as G (6/22, P=0.0243 Fisher) and I (4/6, P<0.002). Differences were not significant between spike trains categorized as G and those categorized as I (P=0.147 Fisher).

- Place Figure 6 about here -
Among neuronal spike trains initially categorized as G (n=22) during the spontaneous condition, two changed to the NG category, and four changed to the I category at the transition to the counting condition. Some of the neuronal spike trains categorized as I during the spontaneous condition changed to the NG category (4/6). In contrast, none of the spike trains in the NG category changed their category (0/18). Therefore all changes in the category of spike train activity between the two conditions were in the direction of categories with lower BR and higher FR e.g. G changes to NG or I. Assuming that either rate during the counting condition will be greater than that during the spontaneous condition with a P=0.5, the odds that the FR will always go up and the BR will always go down for any change in category P=0.25. Therefore, uniformity of these changes across neurons in these rates is much greater than expected at random (10/10, P=0.000001, Binomial).

The neuronal spike trains which changed category between conditions tended to be different between nuclei. Neuronal spike trains of any category which changed category at the transition from the spontaneous to the counting condition tended to be less common in Vo (3/30, Fisher), than in other nuclei including: Vc (2/4, P=0.093), Vim (3/7,P=0.068), but not LD (2/5, P=0.14). Similarly, there was no difference among neuronal spike trains which changed their category from I category, or from G category at the transition to the counting task. We next focused on the remaining spike trains which did not change their category with condition.

**Neurons which did not change Categories between Conditions.**

Figure 5 and the lower panel in Figure 6 show examples of neuronal spike trains which did not change category by condition. Among the 22 such spike trains categorized as NG there were significant increases in FR (P<0.001, Wilcoxon matched pair test), and PR from spontaneous to
counting task periods (Table 4, P=0.003). Burst rates were lower for the counting condition overall (P=0.004, Table 3). Among the G category which did not change at the transition between conditions there was a strong trend toward decreased BRs during the counting versus the spontaneous task (P=0.054, Table 4). None of the other firing, burst or inhibitory indexes was different between spontaneous and counting conditions for neurons in the G and NG categories. Only two spike trains in the I category did not change their category by condition, so that spike train measures could not be studied in this category. Sensory properties of neurons were not related to spike train categories during spontaneous condition, or counting condition, or at the transition between spontaneous and counting.

- place Table 4 about here -

**DISCUSSION:**

We have tested the hypotheses that human thalamic activity changes between quiet wakefulness versus the condition produced by mental arithmetic. The results demonstrate changes during the counting versus the spontaneous condition in spike train category, or in firing and burst rates within a category. Overall the counting condition is characterized by higher FRs and lower BRs. The G spike trains which did not change category between conditions had lower BRs during the counting condition while the FRs and PRs did not change between these spike trains (Table 4). In the case of spike trains which changed between conditions, the G category spike trains changed to I or NG (Figure 3), and the I spike trains changed to NG. The parameters of inhibitory events were not different between conditions for these G and NG neurons (PBISI and PBISI/ PBISI)(Table 4). The constancy of the FR and PR in the G spike trains between conditions suggests that the resting membrane potential is unchanged, while the decrease in the BR suggests that there is a
decrease in the rate of inhibitory events in the counting condition. These results suggest that thalamic activity can be dramatically altered by a cognitive task and that the mechanisms governing human thalamic activity during cognitive tasks engage many different neuronal processes.

**Methodologic Concerns**

In identifying LTS burst activity we have used the n versus n+1 plot, a bias-free method which demonstrates the presence of LTS bursts in our data. However, in order to be able to quantify the indices which describe LTS burst, it is necessary to identify individual bursts. Therefore, we adopted the spike burst template (50-06-16, see Methods: Analysis of thalamic …), which has been used to identify LTS bursting in studies of behaving monkeys (Ramcharan et al. 2000a; Ramcharan et al. 2000b). In one of these reports, a parallel study in monkey thalamic slice demonstrated the validity of this template in identifying the burst pattern associated with primate LTS (Ramcharan et al. 2000a). This template was used in the present recent study because there are no human thalamic intracellular studies (Lee et al. 2005).

It is possible that an LTS can produce one spike without a burst, which is defined by two or more action potentials following a long interspike interval (see Figure 1 in (Ramcharan et al. 2000a)). Some of the points in the upper right of the n versus n+1 plots may represent such events. It is also possible that these extracellular recordings and analysis will identify bursts which are not associated with underlying LTS, although the burst fits the template. We cannot identify either of these two potential sources of error without intracellular recordings, and therefore we account for these potential errors by increased variance within the statistical analysis.

The present study is complicated by the demonstration that the rate of LTS bursting is increased in a number of neurologic diagnoses and symptoms including chronic pain (Jeanmonod et
al. 1996; Lenz et al. 1994), Parkinson’s tremor, dystonia, tinnitus and some psychiatric diagnoses (Jeanmonod et al. 1996). The presence of LTS bursts during normal wakefulness is suggested by its’ presence in all these diagnoses, since it seems unlikely that the same pathology would be observed in these very different conditions. The normal presence of LTS bursts during waking is also consistent with the present results in two waking conditions, and with studies in healthy monkeys (Ramcharan et al. 2000a; Ramcharan et al. 2005; Ramcharan et al. 1998; Ramcharan et al. 2000b; Ramcharan et al. 2001; Martinez-Conde et al. 2002; Lee et al. 2005).

Although thalamic surgery is often carried out for treatment of ET (Koller et al. 2001), to our knowledge LTS bursting has not been reported to be altered in ET. We studied subjects with ET because it can be characterized as a mono-symptomatic illness without cognitive symptoms, and without the complex physiologic/clinical abnormalities of other neurologic diseases treated with thalamic surgery, e.g. Parkinson’s Disease (see Methods and (Deuschl et al. 1998)). Finally, these results focus on differences between spike trains recorded from the single neurons during both spontaneous and counting conditions, so that disease related effects are unlikely to account for the differences between conditions.

**Differences in Thalamic LTS Bursting between Nuclei:**

These results suggest that LTS bursting and significant pre-burst inhibition occur in these first order (Vc, Vim, Vo) and higher order thalamic nuclei (LD) which is consistent with studies of awake rhesus monkeys (Ramcharan et al. 2005). The FRs and PRs are higher in Vim than in any other nucleus studied, while the LTS burst rate, the burst rate/primary event rate, and the proportion of G category spike trains are higher in Vc. The PBISI is shorter in the Vim (148 ms) versus Vc (327 ms), which approximately compatible with the duration of IPSPs mediated by GABAA versus
GABAb receptors respectively (Andersen and Sears 1964; Roy et al. 1984; Soltesz et al. 1989; Steriade et al. 1997b). These results suggest that inhibitory events in Vc are more frequent and more intense than those in Vim. Although Vo receives inhibitory (pallidal) inputs it has a significantly lower rate of LTS bursts than the other nuclei, and the FR in Vo is lower to the point that neurons in Vo may not respond to inhibitory inputs (Smith and Sherman 2002).

Neurons in the thalamic sensory nuclei have demonstrated that prolonged hyperpolarization in the range of 0.5 sec results from inhibition evoked phosphorylation of T channels and results in increased LTS burst firing (Leresche et al. 2004). The latency of this effect is similar to the median of PBISIs in Vc (Table 2), suggesting that this effect might explain the characteristics of preburst inhibition and LTS bursting in this sensory nucleus.

**Spike trains in the spontaneous condition.**

The results of this study (Figures 4, 5, and Results: Stationary behavior of spike train during the spontaneous …) show that the FRs and BRs are relatively constant during spontaneous conditions, even though the epochs of spontaneous condition were interrupted by epochs of the counting condition in the complete spike train. The majority of spike trains were characterized by the stationary BR and FR during epochs of the spontaneous condition. This result suggests that the timing of action potentials is random as measured by FRs and BRs during the spontaneous condition. By the median runs test, all nonstationary epochs were characterized by fewer transitions in FR and BR than expected at random. In the spontaneous condition, spike trains initially categorized the G and NG category never changed category. Therefore, there was relatively little variability of spontaneous neuronal activity in these two categories.
On the other hand, spike trains in the I category frequently changed category during the spontaneous period. In this category, all spike trains are composed of both single spikes and LTS bursts, unlike the G and NG category spike trains which are largely composed of LTS bursts and single action potentials, respectively (Figures 1 and 4). These characteristics of the I category are compatible with high variability of membrane conductance, as suggested by modeling studies and studies of a thalamic slice (Wolfart et al. 2005; Debay et al. 2004). These proposed changes in conductance could result from corticothalamic connections which provide a dense excitatory input both to the distal dendrites of thalamocortical neurons, and to the inhibitory interneurons projecting to thalamocortical neurons (Ahlsen et al. 1982; Contreras and Steriade 1995; Liu et al. 1995; Steriade et al. 1997b). During the spontaneous condition and at the transition to the counting condition, spike trains in the I category have a significantly greater propensity to change categories than the G and NG categories. These changes suggest that the I category may be a transitional firing pattern between the thalamic burst mode (G category) and the relay mode (NG).

**Spike trains in the spontaneous versus the counting condition.**

Among G neuronal spike trains which did not change category with the counting condition, it seems likely that decreased BRs result from decreased rates of inhibitory events. The spike trains which changed from the G category to I or NG will also result in fewer inhibitory events, as reflected in the decreased BRs. NG spike trains have shorter PBISI than I and G spike trains (Figure 3 and Table 4) so that the inhibitory input may switch from GABA\(_b\) to GABA\(_a\) receptors at the transition from G or I to NG. This change in inhibitory processes could be the result of anatomic distribution of receptors, or of activity changes in pathways innervating the neuron (Figure 3) (Roy et al. 1984; Soltesz et al. 1989; Sherman and Guillery 2001; Bal et al. 1995a).
It seems likely that some of these changes in inhibitory processes during the cognitive task are mediated through inputs to the thalamus from the cortex. Inhibition of thalamic relay neurons may be mediated through cortical inputs to the thalamic reticular nucleus (TRN) or local interneurons or both. For example, excitatory inputs to the thalamic reticular nucleus are highly correlated with LTS bursts in thalamocortical neurons, probably as a result of inhibitory input from the TRN to thalamocortical neurons, which is mediated through GABAa and GABAb receptors (Jones 2002) (Bal et al. 1995b).

In summary, BRs were significantly greater than zero for G and I categories during both (waking) conditions. Decreased BR of G spike trains during the counting versus the spontaneous condition may be due to decreases in corticothalamic inputs to TRN or local interneurons, that lead to IPSPs in thalamocortical neurons. The increased FRs of NG spike trains in the absence of burst firing may be due to depolarization mediated through excitatory cortical input to thalamocortical neurons. Increased variance of membrane conductance leading eventually to membrane depolarization may explain the transformation from G category spike trains, first to I, and then to NG category spike trains, so that the I category is a transitional state between the thalamic bursting (G category) and relay modes (NG category)(Wolfart et al. 2005;Debay et al. 2004). Since bursting leads to increased efficacy of thalamocortical synapses, to expanded cortical activation, and to altered thalamocortical responses to peripheral stimuli. the spike trains in the I category may facilitate cortical plasticity (Guido et al. 1995;Swadlow and Gusev 2001;Beierlein et al. 2002).
FIGURE LEGENDS

Figure 1. Examples of spike trains recorded during the spontaneous condition.  A. Spike trains of spontaneous activity are shown as the raw microelectrode signal on the left, and digitized spike trains on the right. In panel A, row G the inset is a raw signal showing an LTS related burst of action potentials. Dots above the digitized spike train indicate the occurrence of LTS bursts as identified by the 50-6-15 criteria (see Methods: Analysis of thalamic … ). These data are shown in A for one each of Grouped (G), Nongrouped (NG) and Intermediate (I) categories as labeled from the top down.  B. The n versus n+1 plot for the G spike train in A (top row) is the left plot the top row in B ; similarly the n versus n+1 plot for NG spike train in A is shown at the left end of the middle row of B, and so on for I. From the top down these rows show 4 examples each of the G, NG and I spike trains in the same order as in A. An alphanumeric identifier is shown with the results from each neuron.

Figure 2. Logarithms (Log, base e) of different variables of bursts identified by 50-6-15 criteria for spike trains of the G, NG, and I categories. These variables are described in detail in the Methods (Analysis of Burst Activity), and include variables describing firing (firing rate - FR), bursting (Burst rate - BR, BR/PR, burst percentage - BP), and inhibitory events (including Preburst interspike interval - PBISI, and PBISI/Primary event interspike interval - PBISI/PRISI). Histograms for these variables are given along the diagonal, as labeled. The vertical and horizontal axis of each graph is labeled along the corresponding axis of the whole figure. For example, the fourth graph from the left (top row) plots log FR on the vertical axis versus log PBISI along the
horizontal axis. Each red ○ indicates the variable for a G spike train, while blue + and green Δ indicate NG and I spike trains, respectively (inset right lower graph).

Figure 3. Box and whisker plots for rates, bursting and inhibitory measures of spike trains. The heavy horizontal line is the median value while the bottom and top of the box indicate the 25th and 75th percentile, and the whiskers indicate the 5th and 95th percentile. Dots indicate values outside the 95th percentile.

Figure 4. Spike trains and n versus n+1 distributions measured from spike trains over long samples of spontaneous activity. The spike train in the G category is shown in the left column, the NG in the right column upper, and I in the right column lower. Intervals below each raster indicate the duration of the epoch of the spontaneous epoch from which the raster was taken.

Figure 5: Spike trains during epochs containing both spontaneous and counting conditions, as labeled. A, upper panel shows the spike train of NG neuron (left) as the raw spike train (inset, shape of the discriminated action potential), and the n versus n+1 plot (right). The lower panel shows the histogram of the neuronal firing rate. B shows the results for a spike train in the G category. Conventions are as in A except that the burst rate is plotted above the firing rate histogram.

Figure 6: Spike trains epochs initially in the I (upper) and G (lower) category across epochs including the spontaneous and counting conditions as labeled. Dots above the spike train indicate
spikes forming a burst (left). Burst rates are shown in the graph above the corresponding n versus n+1 plots. In the lower panel lines through the raster indicate the epochs of the counting condition.
Reference List.


<table>
<thead>
<tr>
<th>Variable</th>
<th>Optimal number of normal distributions</th>
<th>Mean (SD) for distribution 1</th>
<th>Mean (SD) for distribution 2</th>
<th>Mean (SD) for distribution 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>ln(mPBISI)</td>
<td>2</td>
<td>4.57 (0.29) 96.5 ms</td>
<td>6.00 (0.58) 403 ms</td>
<td></td>
</tr>
<tr>
<td>ln(FR)</td>
<td>2</td>
<td>1.29 (0.51) 3.63/s</td>
<td>2.73 (0.51) 1.53/s</td>
<td></td>
</tr>
<tr>
<td>ln(PR)</td>
<td>2</td>
<td>1.00 (0.52) 2.72/s</td>
<td>2.71 (0.52) 15.0/s</td>
<td></td>
</tr>
<tr>
<td>BP</td>
<td>3</td>
<td>1.48% (1.32) 8.55% (4.77)</td>
<td>47.05% (21.64)</td>
<td></td>
</tr>
</tbody>
</table>

Table 1: Indicates the optimal number of peaks as characterized by the mean value (SD) which describe the underlying distribution for each of the variables expressed as the base of the natural logarithms. The antiln value of the mean is below the mean, units as indicated. We used a model selection criteria known as the Bayesian Information Coefficient (BIC).
Table 2. Firing, Burst, and Preburst Parameters for neurons in Four Different Thalamic nuclei. Each cube includes the median and 5%-95%ile confidence interval of the median. Units of each parameter are given in the top row. Variables for nuclei with median and CI when bold and underlined are outliers which are significantly different from nuclei with median and CI shown in bold, e.g. burst rates in Vc are higher than those in Vo and Vim.

<table>
<thead>
<tr>
<th></th>
<th>Firing Rate (/sec)</th>
<th>Primary Rate (/sec)</th>
<th>Burst rate (/sec)</th>
<th>Burst rate/primary rate</th>
<th>Preburst ISI (msec)</th>
<th>Ratio of PBISI/inverse PR</th>
</tr>
</thead>
<tbody>
<tr>
<td>LD</td>
<td>4.2, 3.2-8.0</td>
<td>3.5, 2.6-10.2</td>
<td>0.34, 0.15–0.46</td>
<td>0.085, 0.03-0.21</td>
<td>435, 192-586</td>
<td>1.53, 1.23-1.82</td>
</tr>
<tr>
<td>Vo</td>
<td>4.4, 3.0-9.4</td>
<td>3.2, 2.5-8.9</td>
<td>0.12, 0.05-0.20</td>
<td>0.035, 0.01-0.09</td>
<td>323, 131-433</td>
<td>1.19, 1.12-1.57</td>
</tr>
<tr>
<td>Vim</td>
<td>12.9, 8.7-18.7</td>
<td>12.3, 7.1-8.4</td>
<td>0.21, 0.14-0.34</td>
<td>0.020, 0.01-0.03</td>
<td>148, 119-219</td>
<td>1.99, 1.53-2.28</td>
</tr>
<tr>
<td>Vc</td>
<td>5.2, 3.7-9.5</td>
<td>2.9, 2.2-5.7</td>
<td>0.67, 0.47-0.86</td>
<td>0.26, 0.08-0.41</td>
<td>327, 222-422</td>
<td>0.96, 0.84-1.35</td>
</tr>
<tr>
<td></td>
<td>Spontaneous Task</td>
<td>Count task</td>
<td>Probability</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>--------------------------</td>
<td>------------------</td>
<td>-----------------</td>
<td>-------------</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Firing rate (/sec)</td>
<td>4.24(3.37 – 5.31)</td>
<td>5.33(3.36 – 6.33)</td>
<td>0.09</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Primary event rate</td>
<td>2.96(2.55 – 3.67)</td>
<td>3.23(2.59 – 4.96)</td>
<td>0.671</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(1/sec)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Burst Rate (/sec)</td>
<td>0.23(0.16 – 0.35)</td>
<td>0.13(0.08 – 0.20)</td>
<td>0.004</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Burst rate/primary</td>
<td>0.12(0.05 – 0.19)</td>
<td>0.06(0.03 – 0.10)</td>
<td>0.004</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>event rate</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>PBISI (ms)</td>
<td>372.1(318.1 – 450.8)</td>
<td>338.9(269.0 – 433.5)</td>
<td>0.886</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>PBISI-inverse of</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>primary event rate</td>
<td>1.19(1.12 – 1.28)</td>
<td>1.16(1.04 – 1.29)</td>
<td>0.975</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Burst percentage</td>
<td>13.7(6.35 – 27.35)</td>
<td>6.7(2.47 – 18.41)</td>
<td>0.011</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Table 3: Thalamic Spike Train Characteristics during spontaneous and counting conditions. The units of the indices are shown in the column to the left. Each cube gives the median, 5 - 95%tile confidence interval of the median. The units of each row are given in brackets in the left cube of each row.
<table>
<thead>
<tr>
<th></th>
<th>G Spontaneous</th>
<th>G counting</th>
<th>NG Spontaneous</th>
<th>NG counting</th>
</tr>
</thead>
<tbody>
<tr>
<td>FR</td>
<td>3.3 (2.6-4.3)</td>
<td>3.0 (2.2-3.9)</td>
<td>9.1 (6.5-13) **</td>
<td>15 (10-18)</td>
</tr>
<tr>
<td>PR</td>
<td>2.2 (1.8-2.8)</td>
<td>2.1 (1.6-2.8)</td>
<td>10 (6.5-16) **</td>
<td>14 (9.2-17)</td>
</tr>
<tr>
<td>BR</td>
<td>0.61 (0.45-0.85) *</td>
<td>0.44 (0.36-0.62)</td>
<td>0.01 (0.00-0.04)</td>
<td>0.04 (0.02-0.08)</td>
</tr>
<tr>
<td>BR/PR</td>
<td>0.33 (0.22-0.36)</td>
<td>0.26 (0.17-0.31)</td>
<td>0.01 (0.00-0.011)</td>
<td>0.01 (0.00-0.01)</td>
</tr>
<tr>
<td>mPBISI</td>
<td>461 (422-637)</td>
<td>469 (380-600)</td>
<td>97 (80-125)</td>
<td>100 (85-120)</td>
</tr>
<tr>
<td>PBISI/PRISI</td>
<td>1.1 (1.0-1.2)</td>
<td>1.1 (1.0-1.2)</td>
<td>1.3 (0.97-1.8)</td>
<td>1.5 (0.89-2.0)</td>
</tr>
<tr>
<td>BP</td>
<td>49 (37-61)</td>
<td>48 (48-55)</td>
<td>0.3 (0.0-1.3)</td>
<td>0.8 (0.40-1.40)</td>
</tr>
</tbody>
</table>

Table 4: Firing and Burst indices during spontaneous and counting conditions for G and NG cells which did not change their spike train category between tasks. Each result is given as Median (5-95 percentile of the median). Results with significant differences between conditions for a variable are underlined and the level of significance is indicated by ** (P<0.003), or a trend toward significance (P=0.054, Wilcoxon paired) is indicated by *. The I category was not included because only two neurons did not change category.