A color-based visualization technique for multi-electrode spike trains

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Running title
Visualization of multi-electrode spike trains

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Abstract

Multi-electrode recordings of neuronal activity provide an overwhelming amount of data that is often difficult to analyze and interpret. Although various methods exist for treating multi-electrode datasets quantitatively there is a particularly prominent lack of techniques that enable a quick visual exploration of such datasets. Here, by using Kohonen self-organizing maps, we propose a simple technique that allows for the representation of multiple spike-trains through a sequence of color coded population activity vectors. When multiple color sequences are grouped according to a certain criterion, e.g. by stimulation condition or recording time, one can inspect an entire dataset visually and extract quickly information about the identity, stimulus-locking and temporal distribution of multi-neuron activity patterns. Color sequences can be computed on various timescales revealing different aspects of the temporal dynamics and can emphasize high-order correlation patterns that are not detectable with pairwise techniques. Furthermore, this technique is useful for determining the stability of neuronal responses during a recording session. Due to its simplicity and reliance on perceptual grouping, the method is useful for both quick online visualization of incoming data and for more detailed post-hoc analyses.
1. Introduction

The highly distributed architecture of the cerebral cortex enables neurons to constantly process patterns of spikes originating from multiple pre-synaptic sources (Douglas and Martin, 2004). Therefore, a proper characterization of brain dynamics has to rely on parallel recordings that enable the simultaneous sampling of the activity of many neurons (Buzsáki, 2004; Brown et al., 2004; Singer, 1999). Such recordings, however, generate large amounts of data that are multi-dimensional in nature and are thus, difficult to analyze. To handle such datasets, advanced quantitative methods are being developed (Perkel and Bullock, 1968; Abeles and Gerstein, 1988; Chapin and Nicolelis, 1999; Laubach et al., 1999; Grün et al., 2002; Puchalla et al., 2005; Schneidman et al., 2006; Yu et al., 2008; Pipa et al., 2008).

Progress has also been made in developing techniques of data visualization, which enable the experimenter to obtain a quick overview of multi-dimensional signals. These methods surpass the visualization of response properties by classical spike rastergrams or histograms, which, due to the arbitrary ordering of spike trains, provide little help in detecting multi-dimensional spike patterns (Toups and Tiesinga, 2006). Techniques for visualization of multi-neuron activity include the ‘gravitational’ method by Gerstein et al. (1985), which was designed for detecting correlations between multiple neurons, and a class of methods that visualize neuronal activity through a trajectory plotted in a reduced two- or three-dimensional state-space (Friedrich and Laurent, 2001; Galán et al. 2004; Brown et al., 2005; Bathellier et al., 2008). Two other methods enable the detection of groups of correlated neurons: the ‘correlation grid’ (Stuart et al., 2004) and plots using ‘parallel coordinates’ (Inselberg and Dimsdale, 1990; Stuart et al., 2001). However these latter ones do not provide means for visualizing the temporal dynamics of responses, but offer only static representations of correlations within neuronal groups. Also, most of the existing visualization
methods are limited to a relatively small number of neurons and thus, cannot deal with high-dimensional firing patterns.

Here, we propose a method that uses colors to represent the identity of multi-neuronal activity patterns and enables the quick visualization of large multi-electrode datasets, while preserving information about the structure (firing combination) of neuronal activity patterns. The method also allows for the inspection of activity patterns on various timescales and enables the estimation of the stationarity of cortical responses over time. Importantly, we will focus here only on the aspect of data visualization but will also discuss later some aspects of data classification, which is a different but related issue.

2. Material and methods

2.1. Experimental procedures and recording

Data were recorded from area 17 of 4 anesthetized and paralyzed adult cats, bred in the facilities of the Max-Planck Institute for Brain Research. Experiments were carried out in accordance with the European Communities Council Directive of 24 November 1986 (86/609/EEC). Anesthesia was induced with ketamine (Ketanest, Parke-Davis, 10 mg kg-1, intramuscular) and xylazine (Rompun, Bayer, 2 mg kg-1, intramuscular) and maintained with a mixture of 70% N2O and 30% O2 supplemented with halothane (0.5%–1.0%). After tracheotomy, the animals were placed in a stereotactic frame. A craniotomy was performed, and the skull was cemented to a metal rod. After completion of all surgical procedures, the ear and eye bars were removed, and the halothane level was reduced to 0.4%–0.6%. After assuring that the level of anesthesia was stable and sufficiently deep to prevent any vegetative reactions to somatic stimulation, the animals were paralyzed with pancuronium bromide (Pancuronium, Organon, 0.15 mg kg-1 h-1). Glucose and electrolytes were
supplemented intravenously and through a gastric catheter. The end-tidal CO2 and rectal temperature were kept in the range of 3%-4% and 37°C–38°C, respectively. Stimuli were presented binocularly on a 21 inch computer screen (HITACHI CM813ET) with 100 Hz refresh rate. To obtain binocular fusion, the optical axes of the two eyes were first determined by mapping the borders of the respective receptive fields and then aligned on the computer screen with adjustable prisms placed in front of one eye. All the experiments were conducted according to the guidelines of the Society for Neuroscience and German law for the protection of animals, approved by the local government's ethical committee and overseen by a veterinarian.

Data were recorded by inserting multiple silicon-based multi-electrode probes (16 channels per electrode) supplied by the Center for Neural Communication Technology at the University of Michigan (Michigan probes). Each probe consisted of four 3 mm long shanks that were separated by 200 µm and contained four electrode contacts each (1,250 µm² area, 0.3–0.5 MΩ impedance at 1,000 Hz, inter-contact distance 200 µm). Signals were amplified 10,000× and filtered between 500 Hz and 3.5 kHz and between 1 and 100 Hz for extracting multi-unit (MU) activity and local-field potentials (LFP), respectively. The waveforms of detected spikes were recorded for a duration of 1.2 ms, which allowed the later application of offline spike-sorting techniques to extract single units (SU). Spike-sorting was carried out with a custom-made software relying on principal component analysis of the waveforms. The software for visual stimulation was a combination of custom-made programs and a stimulation tool, ActiveSTIM (www.ActiveSTIM.com).

2.2. Datasets

**Dataset with drifting sinusoidal grating stimuli** (col05-e08). Sinusoidal gratings moving in 12 directions in steps of 30° were presented in trials of 4,800 ms duration (1,000 ms spontaneous activity, 3,500 ms stimulus, 300 ms OFF-response). Gratings spanned 12° of visual angle, had a
spatial frequency of 2.4° per cycle and drifted with a speed of 2° per second. Each direction was presented 20 times in a randomized order leading to the total of 240 presentations (trials). Dataset col05-e08 consisted of 26 simultaneously recorded SUs.

**Dataset with plaid and bar stimuli** (col08-e27). Eight stimuli were used including: two simultaneously moving bars, in one case with a dot at the intersection, two single bars moving at different speeds, two plaid stimuli with pattern or component motion, a grating, and a moving dot (see Fig. 3). The plaids and the grating had a rectangular change in luminance. Bars spanned 7° and gratings and plaids 12° of visual angle. Trials were 6,500 ms long with stimuli presented between 1,000 ms and 6,000 ms, and 20 times each. Analyses were performed on 32 simultaneously recorded, unsorted, multi-units (MUs).

**Dataset with natural stimuli** (cer01-a50). Three movies with natural images were presented to the cat (one recorded by the authors, two extracted from a documentary “The Greatest Places” by the Science Museum of Minnesota). The movies contained indoor and outdoor scenes with various image statistics (slow moving, fast moving, dark, light, etc). The movies were presented across the entire screen (41° × 31° of visual angle) with a resolution of 800x600 pixels. Each movie was 28 seconds long and was presented 20 times. Analyses were performed on 34 simultaneously recorded SUs.

**Datasets with center-surround stimuli** (col11-b44 and col11-b68). Sinusoidal gratings of three different sizes (small, medium and large) and two orientations (horizontal and vertical) were presented individually or superimposed. Gratings spanned visual angles of 7°, 14° and 21°, had a spatial frequency of 1° per grating cycle and were drifted at a speed of 1.5° per second, orthogonal to their orientation and in one direction only. Stimuli included 6 individual gratings, 4 superimposed gratings with a small central grating surrounded by an orthogonal medium or large
grating, and 4 superimposed gratings consisting of a small grating separated by a gray ring (3.5° wide) from a surrounding large grating of identical or orthogonal orientation. The resulting 14 stimuli were randomly presented 20 times each, leading to a total number of 280 trials. Trials were 6,000 ms long with the stimulus presented between 1,000 ms and 5,000 ms. After spike-sorting, from a total of 66 SUs only 12 SUs could be confidently tracked (matched) in both datasets and further used in the analyses.

Dataset with high spatial-frequency sinusoidal grating stimuli (cer01-a45). These stimuli were identical to those used in col08-e05 with the exception that gratings had a higher spatial frequency of 1.1° per cycle.

2.3. Visualization technique

The visualization technique includes three steps: low-pass filtering spike-trains, Kohonen mapping, and, finally, building color sequences (Fig. 1).

2.3.1. Low-pass filtering spike-trains

To handle multiple spike trains one must first find a robust multidimensional representation. Most previous studies have applied binarization in combination with binning to define multidimensional activation vectors (Grün et al., 2002). While being convenient for defining patterns as binary vectors (Schneidman et al., 2006), binning prevents a flexible manipulation of the timescale because, for large bin sizes, multiple spikes fall into the same bin. Replacing binarization with instantaneous firing rate (spike count per bin) does not solve the problem because, especially for large bin sizes, one cannot distinguish different firing patterns within the bin. Instead of binning, spike-trains can be first convolved with specialized kernels such as Gaussian (Richmond et al., 1990; Eskandar et al., 1992; Heller et al., 1995), sinusoidal (Sameshima and Baccalá, 1999) or
exponentially-decaying kernels (Baker and Gerstein, 2000; Häusler and Maass, 2007). We used the latter because such kernels are essentially causal low-pass filters (i.e. the values of the signal depend only on the preceding spikes). The convolution preserves information about all spikes and allows one to control the timescale of interest by manipulating the decay (integration) time constant ($\tau$). For each neuron $i$, a continuous signal, called activation, $a_i(t)$ was obtained using the formula:

$$a_i(t) = \begin{cases} a_i(t-1) + 1, & \text{if neuron } i \text{ has a spike at time } t \\ a_i(t-1) \cdot e^{-\frac{t}{\tau}}, & \text{otherwise} \end{cases}, \quad (1)$$

where $a_i(t)$ is the activation corresponding to neuron $i$ at time $t$, and $\tau$ is the decay (integration) time constant. For small time constants ($\tau = 1–5 \text{ ms}$), one extracts synchronous patterns of spikes, or joint-spike events. For large time constants ($\tau > 100 \text{ ms}$), one extracts firing-rate patterns. Unless specified otherwise, an integration time constant of 20 ms was used, because it is in the physiological range of membrane time constants of cortical neurons (Kasper et al., 1994; Spruston and Johnston, 1992; Lefort et al., 2009).

Low-pass filtering of spike trains (Fig. 1, steps 1 and 2) yielded multiple continuous traces which were then sampled (Sameshima and Baccalá, 1999) with a frequency of 1 kHz, and for each time step $t$ (with millisecond resolution), an activity profile, called activity vector, was defined as:

$$AV(t) = [a_1(t), a_2(t), \ldots, a_n(t)], \quad (2)$$

where $n$ is the number of analyzed neurons. A trial was then described as a succession of activity vectors. Note that an activity vector also contains information about the activity in the recent past of the corresponding neurons (Fig. 1, step 2).
2.3.2. Kohonen mapping

After integrating the original spike trains using exponentially-decaying kernels, the activity vectors sampled at each millisecond were presented, in random order, to a 3D Kohonen map (3DKM) for learning (Fig. 1, step 3). 3DKM is an extension of the classical 2D Kohonen self-organizing map (Kohonen, 2001) simply by adding one dimension (3DKM of size \( N \times N \times N \) is defined on an \( N \times N \times N \) lattice). Each element in the lattice contains a vector of dimension equal to the dimensionality of the input space, termed model vector. At each step \( k \) of the learning algorithm, the map learned an activity vector \( AV_k \) as follows: First, the most similar model vector to \( AV_k \) was found in the map and called best-matching unit (BMU); second, the BMU and its neighbors (Kohonen, 2001) were altered to make them more similar to \( AV_k \). The amount of change and the radius of the neighborhood were given by two monotonically decreasing functions: \( L(k) \) and \( R(k) \), respectively:

\[
L(k) = L_0 \cdot e^{-k \frac{\ln L_0 \cdot L_M}{M}},
\]

where \( L(k) \) is the learning rate, modulating the degree to which model vectors were changed at each training step, \( k \). \( L_0 \) and \( L_M \) are initial and final learning rates. We used \( L_0 = 1 \) and \( L_M = 0.01 \). The total number of training steps is denoted by \( M \).

\[
R(k) = \text{round} \left( R_0 \cdot e^{-k \frac{\ln (R_0 / 0.5)}{(g / 100) \cdot M}} \right),
\]

where \text{round} denotes the rounding to the nearest integer, \( R(k) \) specifies the neighborhood size around the BMU within which elements were allowed to learn at step \( k \). \( R_0 \) is the initial radius of the neighborhood. \( g \) is the percentage of \( M \) after which \( R \) becomes 0 (only the BMU is modified for \( R = 0 \)). We used \( R_0 = N/2 \) and \( g = 66 \) (66\% of steps were used to establish the topology of the map.
and the remaining 34% of the steps to fine-tune the representation of activity vectors in the map, i.e. only the BMU was changed).

Within the above defined neighborhood, model vectors further away from the BMU changed less than the ones closer to it by multiplying the learning rate with a 3D Gaussian envelope with a standard deviation of \( R(k) / 3 \):

\[
MV_{k}[x, y, z] = MV_{k-1}[x, y, z] + (AV_{k} - MV_{k-1}[x, y, z]) \cdot L(k) \cdot e^{\frac{(x_{BMU} - x)^2 + (y_{BMU} - y)^2 + (z_{BMU} - z)^2}{2(R(k)/3)^2}},
\]

where \( MV_{k}[x, y, z] \) is a model vector, at step \( k \) of the training, located within the neighborhood of the BMU (distance from BMU \( \leq R(k) \)) at position \((x,y,z)\) in the 3D lattice. \((x_{BMU}, y_{BMU}, z_{BMU})\) is the position of the BMU in the 3D lattice. \( AV_{k} \) is the activity vector that is learned at step \( k \), \( L(k) \) and \( R(k) \) are, respectively, the learning rate and the size of the neighborhood at step \( k \).

The map was initialized with values of 0. The convergence of the learning algorithm depends on the total number of steps \( M \). Typically ~100,000 steps are recommended, but in some cases, where “fast learning” is required, even 10,000 steps may be sufficient (Kohonen, 2001). In the data investigated here, depending on the number and length of trials, the number of activity vectors varied across recording sessions (ranging from 1,040,000 to 1,680,000). The number of steps used in our analyses was typically 3 times the number of activity vectors, i.e. each activity vector was learned three times by the map. Activity vectors were presented in random order to the 3DKM to avoid leaning temporal dependencies and to maximize the fidelity with which the clustering (model vectors) represented the activity vector space. In most cases we used maps that included 1,000 points (clusters) in the 3D lattice \( (N = 10) \), ensuring a fine grain representation of the activity vector space. Thus, for \( N = 10 \) there were 1,000 patterns (model vectors) that described a given dataset.
All activity vectors of a given dataset were learned using the 3DKM (Fig. 1, step 3). The Kohonen learning is essentially an ordered clustering, i.e. clustering and ordered mapping on the lattice (Kohonen, 2001). After learning, the lattice holds model vectors that are in fact clusters approximating the input space. They represent stereotypically appearing spatiotemporal activity patterns spanning a finite time window determined by the integration time constant. For simplicity, model vectors will be called patterns.

During training, the approximation error, i.e. the error between the activity vectors of the dataset and their corresponding model vectors, was computed as:

\[ E = \frac{1}{NA} \sum_{i} \| AV_i - BMU_{AV_i} \|^2, \]  

(6)

where \( E \) is the approximation error, \( \| AV_i - BMU_{AV_i} \| \) is the Euclidian distance between the activity vector and it’s corresponding BMU from the map, and \( NA \) is the total number of activity vectors for a dataset. The approximation error drops dramatically in the first steps and then converges to a low value towards the end of the training (Supporting Fig. S1A).

It can be checked whether model vectors represent faithfully the activity vectors also by computing the distribution of Pearson coefficients of correlation between the two. In the example provided in Supporting Fig. S1B, 82% of the coefficients were above 0.8, i.e. model vectors approximated activity vectors with a high degree of precision.

The computation time required for training the Kohonen map is fast to moderate, depending on the amount of data and the size of the 3DKM. For example, the map used to produce Fig. 2B was computed in a few minutes (< 15 min) on a personal computer with average performance. With modern parallel computing, the time to compute the map can be considerably reduced.
2.3.3. Building color sequences

For visualization, each dimension of the 3DKM lattice was considered to be one color dimension in the RGB space (Red, Green and Blue; Fig. 1, step 3). A model vector was then assigned a corresponding color, depending on its position in the lattice. The coloring strategy took advantage of the fact that, during learning, the Kohonen algorithm maps similar model vectors to nearby locations (i.e. ordered mapping). Due to the spatial (Euclidian) proximity of similar model vectors in the lattice (Kohonen, 2001), these were assigned similar colors. In addition, the matching cubic topologies of the 3DKM lattice and the RGB space ensured that, for each analysis, the entire spectrum of colors was used to represent the model vectors.

A trial could then be visualized as a sequence of colors, whereby each activity vector was labeled by the color of its corresponding model vector. This was done as follows: For each time bin, the activity vector was used to find its closest model vector in the 3DKM cube (its stereotypical model). At the respective time bin in the trial the color of the model vector was marked through a vertical color band (a painted rectangular with thickness = 1 ms; Fig. 1, step 4). A color sequence represents the activity of all neurons along a single recorded trial.

Collections of color sequences can be presented in an adjacent and time-aligned manner—usually one color sequence is shown below the other (e.g. Fig. 2B). For the detection of regularities, our presentation method relies on the Gestalt grouping principles of the human visual system. The regions of similar or near-similar colors blend into an easily detectable larger one-color region (e.g., a blob)(e.g. Fig. 2B and Fig. 3A and 3C). Thus, it is easy to detect the regions in which similar patterns (colors) occur consecutively along the trial and/or consistently at the same time points across different trials. The choice of the order in which the trials will be aligned (i.e., the grouping
order) determines to a large degree the type of information about neuronal dynamics that can be visually extracted.

Importantly, one can also chose to inspect individual color sequences carefully, tracking given color bands corresponding to some specific patterns and thus detecting the latter even without relying on Gestalt grouping. In such cases however, a global picture of the whole dataset is not possible to attain.

In the training process of the Kohonen map, activity vectors are presented in random order and, therefore, different runs of the algorithm yield maps with different structure. The algorithm converges such that learning is not affected by the random presentation of the samples, e.g. across 10 different runs for a drifting grating dataset, the coefficient of variation for the learning error was 0.000625 (Supporting Fig. S1A). However, because in different runs maps will have different structure, the assignment of colors to individual patterns is likely different. Therefore, colors obtained with one map cannot be directly matched with those obtained with other maps. A color assignment could be more salient than the other. However saliency can be improved by rotations of the Kohonen cube (i.e. changing the assignment of the Red, Green and Blue dimensions relative to the 3DKM).

The mapping on the Kohonen cube ensures that all colors are used to represent a dataset. However, the color similarity between model vectors cannot be appreciated with respect to the absolute distance between them. Depending on the structure of the datasets, a map might assign more similar colors to some vectors, while for another dataset it might assign more different colors to the same vectors. In other words, the spread of the data might be relevant to understanding how color similarity should be interpreted with respect to absolute distance between vectors. A possibility to estimate the spread of vectors is to compute the distribution of all pairwise Euclidean distances
between model vectors in the Kohonen map. In addition, the distances have to be normalized to the dimensionality of the vectors (i.e. neuron number) in order to have results comparable across datasets with different number of neurons. Example distributions for two different datasets are shown in Supporting Fig. S1C. For a dataset recorded with drifting sinusoidal gratings (see Datasets), the distribution is shifted towards lower values, indicating that model vectors are rather close to each other. On the contrary, for a dataset recorded with plaid and bar stimuli (see Datasets), the distribution is shifted towards larger values, indicating that model vectors are farther apart from each other (Supporting Fig. S1C).

2.4. Artificial datasets

To study the ability of the method to detect patterns evolving on different timescales, we generated two artificial datasets of random non-stimulus related spiking and embedded/inserted within them additional spikes that represented stimulus-specific activity. The datasets consisted of 25 spike trains, simulated as homogenous Poisson processes (Cox and Isham, 1980) with a rate of 10 Hz. These were segmented in 3,000 ms long trials, which were distributed among 10 hypothetical stimulation conditions, 20 trials per condition. In one dataset we inserted stimulus specific joint-spike events (JSEs) (Pipa et al., 2008), consisting of synchronized spikes across 5 neurons. In the other dataset we added stimulus specific rate covariations (RCs), consisting of simultaneous rate increases, lasting 100 ms, in 3 neurons. In this later case, firing rates increased within the first 25 ms from 10 Hz to 50Hz, and stayed then constant for the next 50 ms, falling back to 10 Hz during the last 25 ms of the RC (Lerchner et al., 2006). For each stimulus condition we randomly assigned different combinations of neurons participating in JSEs and RCs. JSE and RC events occurred 2-3 times per trial and always at the same timepoint for a given stimulation condition.
2.5. Pattern coefficient of variation

We defined a coefficient of variation for patterns \([PCV_j(t)]\), which gives a normalized, \textit{time resolved measure} of the variability of all patterns across trials of stimulus \(j\). This measure can be used to quantify how variable is the occurrence of patterns across different trials of the same stimulus. A high PCV indicates unreliable responses across trials, while a low PCV indicates that the same patterns are produced on repeated presentations of the same stimulus. For a single pattern \(p\) (model vector \(MVp\)), the classic coefficient of variation \((CV_{p,j})\) in a sliding window \([t-h,t+h]\), across trials of stimulus \(j\), was defined as the ratio between the standard deviation of the pattern counts and the average pattern counts in the window, across trials:

\[
CV_{p,j}(t) = \frac{SD_{p,j}(t)}{r_{p,j}(t)}
\]

\[\quad \text{where } SD_{p,j} \text{ is the standard deviation of counts for pattern } p \text{ across trials of stimulus } j, \]  
\[\quad \text{and } T_j \text{ is the number of trials for stimulus } j.\]

Since multiple patterns can occur in a given window, we defined the average coefficient of variation for a stimulus \(j\), in the sliding window centered at \(t\), as:

\[
r_{p,j}(t) = \frac{\sum_l r_{l,p,j}(t-h,t+h)}{T_j}, \quad l = 1,T_j
\]

\[
SD_{p,j}(t) = \sqrt{\frac{\sum_l [r_{l,p,j}(t-h,t+h) - r_{p,j}(t)]^2}{T_j - 1}}, \quad l = 1,T_j
\]
where $2h+1$ is the size of the window, $\overline{r_{p,j}(t)}$ is the average count of pattern $p$ in window $[t-h,t+h]$ (Equation 8) for stimulus $j$, and $P_j$ is the number of distinct patterns that occur in the window across trials of stimulus $j$.

Patterns within a window can have different degrees of similarity to each other. To consider this, we defined an average pair-wise distance between patterns, as follows:

$$\overline{D_j(t)} = \frac{\sum_{u \neq v} \sum_u \|MVp_u - MVp_v\| \cdot \overline{r_{p_u,j}(t)} \cdot \overline{r_{p_v,j}(t)}}{\sum_{u \neq v} \sum_u \overline{r_{p_u,j}(t)} \cdot \overline{r_{p_v,j}(t)}}, \quad u = 1, P_j \quad v = 1, P_j$$

(Equation 11)

where $\|MVp_u - MVp_v\|$ is the Euclidean distance between patterns $p_u$ and $p_v$, $\overline{r_{p_u,j}(t)}$ and $\overline{r_{p_v,j}(t)}$ are average pattern counts of patterns $p_u$ and $p_v$ in window $[t-h,t+h]$ across trials of stimulus $j$, and $P_j$ is the number of distinct patterns that occur in the window. If $\overline{D_j}$ is small, these patterns are highly similar.

Finally, the coefficient of variation for patterns ($PCV_j$) corresponding to stimulus $j$, was:

$$PCV_j(t) = \overline{CV_j(t)} \cdot \overline{D_j(t)}$$

(Equation 12)

$PCV_j(t)$ is 0 when only one pattern occurs in the window at $t$ across all trials of stimulus $j$ and it can increase to different values, depending on the structure of the dataset. Note that PCV cannot be compared to the classic CV due to the weighing with the distance, which can take arbitrary values.

In the analysis shown in Fig. 5 we averaged $PCV_j(t)$ across all stimuli $j$. It is important to emphasize
that each $PCV_j$ was computed per stimulus and only then taken into the average. Thus, a small average PCV does not mean that patterns corresponding to different stimuli were similar but rather that patterns were similar across multiple trials belonging to the same stimulus.

2.6. Pattern Specificity Index

To determine how specifically a pattern $p$ occurs for a stimulus $s$, we defined the Pattern Specificity Index (PSI) as the fraction of occurrences of pattern $p$ for stimulus $s$ with respect to the total number of occurrences of pattern $p$ across all stimuli:

$$\text{PSI}_{p,s} = \frac{\text{count}(p|\text{stim} = s)}{\sum_j \text{count}(p|\text{stim} = j)} \quad (13)$$

The PSI of pattern $p$ for stimulus $s$ has a value of 1 when the pattern occurs exclusively for that stimulus and a value of 0 when the pattern never occurs for that stimulus.

3. Results

3.1. Visualization of large datasets

We first analyzed a dataset recorded from cat area 17, evoked with drifting sinusoidal gratings and consisting of 26 simultaneously recorded SUs (see Materials and Methods, Dataset with drifting sinusoidal grating stimuli). When the activity of all 26 neurons is represented as spike rastergrams grouped by trials belonging to 4 different stimuli (Fig. 2A), one can detect only the onset (ON) and offset (OFF) responses and a certain degree of rate modulations between these two events. Spike rastergrams do not easily reveal the specific firing patterns (spiking combinations) that are evoked by different stimuli. By contrast, when color sequences are computed on the same data and are grouped according to the corresponding stimulus, specific activity patterns and their occurrence in
time become visible (Fig. 2B). Color sequences reveal spontaneous activity (first 1,000 ms), onset and offset of stimuli (1,000 ms and 4,500 ms, respectively), and modulation of responses as the gratings pass the receptive fields (RFs) of neurons. Unlike spike rastergrams, color sequences show stimulus-specific patterns, as certain colors predominate in the responses to certain stimuli. The expression of these colors is modulated by the passage of the grating over the receptive fields, reminiscent of phase-sensitive ‘simple’ cells that participate in the multidimensional firing patterns (Hubel and Wiesel, 1959). Such modulatorly effects are not visible in spike rastergrams when phase-insensitive ‘complex’ cells (Hubel and Wiesel, 1962) dominate the activity (Fig. 2A, first stimulus) because in such cases spikes corresponding to ‘simple’ cell activity are embedded in the sustained activity of ‘complex’ cells. However, because activity patterns represented by color sequences consist of both simple and complex cells, these patterns will be always modulated due to the contribution of ‘simple cells’. The color sequences can therefore always reveal the modulation of multi-neuronal firing patterns caused by ‘simple’ cell activations (Fig. 2B, first stimulus). For the complete set of color sequences, across all 12 directions of the drifting grating, see Supporting Fig. S2.

Different colors in the color sequences represent different firing patterns of neurons. In Fig. 2C, left, two portions of color sequences are shown, corresponding to two different stimuli from Fig. 2B, for which two distinct classes of colors were expressed. These colors correspond to different classes of activity patterns (model vectors) that can be traced down in the Kohonen map (Fig. 2C, 3D Kohonen Map and Pattern). We further computed pattern-triggered spike-raster histograms (PTSRH), i.e. sum of spike-rasters before each occurrence of a given pattern (Fig. 2C, Spike Histogram). When the same pattern occurred multiple times consecutively, due to the decay of the integration kernel, only the first occurrence of the pattern was taken as a reference. In Fig. 2C two PTSRHs are shown for a pattern with three and a pattern with two active neurons (neurons that
define the pattern by firing a spike are referred to as ‘active’), corresponding to colors green and red, respectively. The time of pattern occurrence is taken as a reference at 0 ms in the PTSRH. The bright stripes in the bottom PTSRH shown in Fig. 2C indicate that active neurons of the pattern emit spikes frequently participating in bursts. That is, spikes in the pattern were often preceded by other spikes of the same neurons, at inter-spike intervals < 8 ms (DeBusk et al., 1997). The model vectors (Fig. 2C, Pattern), represented as vectors with grayscale coded entries (black = active, white = inactive), were very similar to the spiking patterns in the PTSRHs. Thus, colors from the color sequences have corresponding model vectors in the 3DKM, which faithfully represent the actual firing patterns of the recorded neurons. These firing patterns can be traced down in the 3DKM and subjected to analyses also with quantitative methods (e.g. pattern occurrence rate, stimulus specificity etc).

Color sequences become richer when a larger variety of stimuli is used in the experiment. In Fig. 3A color sequences are shown for multiunit activity simultaneously recorded from a set of 32 electrodes, under 8 conditions that varied considerably in the spatial extent and complexity of the used stimuli (see Materials and Methods, Plaid and bar stimuli). It is evident that i) similar stimuli elicit similar responses (respectively, stimuli 1 and 3, 2 and 4, 5 and 6), ii) the spatiotemporal evolution of stimuli is reflected in the temporal expression of patterns (stimuli 2 and 4: the speed of the moving bar determines the temporal activation profile of the ‘green’ and ‘purple’ patterns), iii) some stimuli produce clear onset (ON) (stimuli 1, 3, 5, 6 and 7, at 1,050-1,060 ms) or offset (OFF) responses (stimuli 5, 6 and 7).

Another advantage of the present analysis is that the succession of patterns in time can be conceived as a trajectory in a multidimensional phase-space (Friedrich and Laurent, 2001; Galán et al., 2004; Brown et al., 2005; Bathellier et al., 2008). Given that model vectors were mapped on the 3DKM lattice, these trajectories could also be visualized in a 3D space. The successions of patterns in Fig.
3A are shown as trajectories in 3D space in Fig. 3B. To this end, we first computed the average pattern within non-overlapping, 50 ms-long sliding windows, across all the trials of a given stimulus. For each position of the sliding window, we marked the location of the average pattern in the 3DKM lattice and then connected successive points with lines. Consistently with the similarity of the color sequences in Fig. 3A, the 3D phase-spaces in Fig. 3B were also similar for stimuli 1 and 3, 2 and 4, and 5 and 6, respectively. Thus, one can also construct three dimensional phase space representations of multi-neuron activity using the 3D Kohonen map. However, the 3D trajectory representation is less informative than color sequences and represents only an added value to the method, not its main purpose.

For more realistic stimuli, comprising natural scenes, neuronal responses can become more complex (Fiser et al., 2004). In Fig. 3C color sequences are depicted for a dataset consisting of 34 SUs with responses evoked by 3 movies with natural scenes, each in length of 28 s (Materials and Methods, Dataset with natural stimuli). The grouping by stimulus reveals preferential expression of particular patterns at different moments in time. In some cases, patterns were reliably stimulus-locked across different trials (e.g. Fig. 3C, stimulus 1, around 6,500 ms), while in other cases they were not. Moreover, the color sequences associated with natural movies contain sometimes fewer color blobs than for simple stimuli, and this varies across different movies. This property is expected given the more sparse responses of neurons to natural scenes (Vinje and Gallant, 2000). Also, an important factor determining the structure of color sequences could be the degree to which the elements of the movies stimulate the receptive fields of the investigated neurons. Finally, the color sequence representation permits a quick and comprehensive inspection of the entire dataset and selection of periods of interest for further quantitative analyses. One can, for example, subsequently determine whether some patterns appear preferentially for given stimuli, and evaluate
their stimulus-locking properties. Color sequences for the entire duration of the movies are shown in Supporting Fig. S3.

3.2. Multiple timescales and higher-order correlations

We next investigated the ability of the method to detect firing patterns that evolve on different timescales. In order to have a precise control over the timescale of patterns, we generated two artificial datasets containing patterns evolving on either fast, i.e. synchrony, or slow timescales, i.e. firing rate (see Materials and Methods, Artificial Datasets). By manipulating the integration time constant ($\tau$) when computing the activity vectors one biases the analysis to various timescales. In the color sequences from Fig. 4A, corresponding to the dataset containing synchronous events (joint-spike events: JSEs), JSEs could be clearly detected when a small time constant ($\tau = 5$ ms) was used, but could not be detected with a large time constant ($\tau = 50$ ms). When a too large time constant is used, the activity vectors corresponding to JSEs are contaminated by the spikes preceding the JSEs. Conversely, for the dataset containing slow rate covariation patterns, using a small time constant hampers the detection of the specific firing rate combination because not enough information is integrated. Therefore, activity vectors fail to properly represent the firing rate profile (Fig. 4B, top). A larger $\tau$ on the other hand permits the proper integration of the firing rate profile across different neurons and thus enables the representation of the specific rate pattern by the activity vectors and, for that matter, by color sequences (Fig. 4B, bottom). Thus, by manipulating the integration time constant and plotting color sequences at different timescales one can identify the expression of multi-neuronal firing patterns across multiple timescales. Moreover, one can also infer the characteristic timescale on which a given firing pattern occurs and this is expected to be especially useful in studies that investigate dynamical stimulus encoding over multiple timescales (Butts et al., 2007).
To illustrate the ability of the method to detect high-order spike correlations (e.g. JSEs) not extractable with pairwise methods we generated an additional dataset with neurons firing homogenous Poisson spike trains at 10 Hz. We chose 20 trials of 30 s length and inserted in each trial a single JSE with 5 active neurons. Supporting Fig. S4 shows that although cross-correlation histograms between all the pairs of active neurons in the JSE fail to reveal a clear central peak, the visualization method can successfully identify the JSEs in the data. Importantly, given the color-richness of the color sequences, one can easily detect the JSEs if they are time-locked to the same position in the trial. When the JSEs are not clearly stimulus-locked, the method probably provides little help in visually detecting them and more advanced quantitative techniques may be required. A possibility to solve this problem in presented in section 3.4.

3.3. Detection of non-stationary responses and cortical state-changes

Non-stationary responses can occur during the experiment due to unstable recording setup, cortical state changes (Arieli et al., 1996), or other factors (Gur and Snodderly, 2006; Sannita, 2006). Such non-stationary responses can be quickly identified using color sequences. An example is depicted in Fig. 5A, where color sequences corresponding to 12 SUs recorded in response to 14 stimuli (see Materials and Methods, Dataset with center-surround stimuli) are shown in the order the trials were recorded (trials of different stimuli were presented in random order). Responses to the same stimuli were recorded from the same neurons in two different sessions, about 26 hours apart. Session 1 (Fig. 5A, left) exhibited marked non-stationary responses for successive trials along the recording. ON- and OFF-responses were poorly defined. Periods with relatively stable responses were interrupted by periods of weak and unreliable responses. By contrast, in session 2 (Fig. 5A, right), cortical responses were more locked to input and stable over time, ON- and OFF-responses were reliable and precise. We further quantified these observations by computing the coefficient of variation for the patterns (PCV; see Materials and Methods), in a time-resolved fashion, and
averaging it across stimuli. A large PCV in a time window denotes a large variability in the identity of patterns expressed across the trials corresponding to the stimulus at that moment in time. In other words, the larger the PCV the more unreliable the multidimensional response pattern is across repeated presentations of the same stimulus. Average PCV’s were clearly larger for session 1 than session 2 (Fig. 5B), indicating a marked difference in the reliability of pattern expression over time.

When computing autocorrelation histograms for the responses in session 2, we found oscillatory activity of ~ 27 Hz, which is in the beta-high band (20-30 Hz) (Fig. 5C, top-right), but did not find oscillations in session 1 (Fig. 5C, top-left). We quantified oscillatory response properties of all neurons by computing their oscillation score (Mureșan et al., 2008) in the beta-high/gamma-low bands (20-40 Hz). Significantly higher oscillation scores were found for session 2 than for session 1 (p < 0.001; Mann-Whitney U test; Fig. 5C, bottom), confirming that neurons in session 2 exhibited robust oscillations, while the ones in the session 1 did not. This result shows that differences observed in the color sequences and computed quantitatively on patterns, were associated with a change in the oscillatory behavior of neurons, for the investigated case. The result is also consistent with reports that firing rate responses are less variable across repeated trials in brain states with strong gamma oscillations (Rodriguez et al., 2004).

In another example, responses evoked by high spatial-frequency sinusoidal grating stimuli (Materials and Methods, Dataset with high spatial-frequency sinusoidal grating stimuli), exhibited also marked non-stationarities during the experiment (Supporting Fig. S5).

### 3.4. Identifying meaningful patterns

Color sequences reveal all patterns that evolve on a given timescale. However, not all patterns are evoked by the presentation of the stimulus and sometimes similar patterns are evoked by multiple stimuli. As a consequence, it is useful to determine how specifically given patterns occur for given
stimuli. To this end, we developed a measure, called *Pattern Specificity Index* (PSI), that reflects the proportion of occurrences of a pattern for a given stimulus out of the total occurrences of the pattern throughout all stimuli (see Materials and Methods). The PSI can be used to threshold color sequences: For a given stimulus, only color bands are painted corresponding to patterns with PSI larger than the threshold. In Fig. 6A, we show color sequences for a dataset recorded with sinusoidal grating stimuli and two thresholded color sequences with thresholds of 0.25 (Fig. 6B) and 0.5 (Fig. 6C). For higher thresholds, fewer but more specific patterns are revealed.

The thresholding may come in handy when specific patterns are not stimulus-locked and therefore their discrimination is hindered by the color richness of color sequences. A high threshold eliminates most colors and keeps only those corresponding to patterns with high specificity. Therefore, these become detectable regardless of their stimulus-locking properties.

4. Discussion

The Kohonen self-organizing map provides an ordered clustering (Kohonen, 2001) that creates a discrete approximation of the activity vector space. This approximation allows for a flexible representation of the occurring multi-dimensional activity patterns through a limited set of representative model patterns. As we have shown, by introducing a color-based representation of these patterns, one can visually inspect large datasets and draw conclusions on the temporal distribution of classes of firing patterns. Importantly, the ordered mapping provided by Kohonen maps enables the consistency of the color representation. Thus, two different segments of color sequences sharing similar colors represent similar activity patterns of the recorded neurons. The visualization technique we have introduced is relatively straightforward to implement and conceptually simple.
The present method represents advancement over previous PCA-based visualization techniques (Galán et al. 2004; Friedrich and Laurent, 2001; Bathellier et al., 2008). First, the Kohonen mapping is non-linear and can map the structure of the input space onto the 3D lattice more flexibly while preserving local similarity relations (Lee and Verleysen, 2007). This is not the case with linear techniques such as PCA, which can introduce large approximation errors if the structure of the input space cannot be linearly decomposed. Second, instead of emphasizing a state-space trajectory representation (Friedrich and Laurent, 2001; Galán et al. 2004; Brown et al., 2005; Bathellier et al., 2008), albeit possible with our method as well, we introduced a representation based on color bands. This technique offers an important benefit to the visualization, especially when non-similar activity patterns appear in a fast succession. In that case, a state-space trajectory will appear noisy and difficult to interpret. By contrast, the color bands in the color sequence can be inspected in detail, one by one, being perceptually associated through their color with the actual activation pattern of the underlying neurons. Moreover, the precise occurrence time of patterns within the trial is also straightforward to estimate in color sequences. Therefore, the latter provide a more salient and flexible visual description of neuronal dynamics than trajectories in a reduced state-space.

Unlike PCA, Kohonen mapping is not a dimensionality reduction technique per se, but a topological mapping technique. In fact, the 3DKM is an ordered clustering (Kohonen, 2001) and for that reason it has the important advantage of ensuring both an approximation of activity vectors (clustering) and a topological mapping on the 3D lattice that enables the coloring technique we have described. The 3DKM preserves the original dimensionality of the data through model vectors (cluster centers) and therefore one has immediate access to classes of firing patterns that occur in the data and are segregated through clustering during the training of the Kohonen map. In the model vectors, the identity of individual neurons from the original recording is preserved. In addition, both silent and active neurons determine the identity of the pattern (model vector), i.e. silent neurons also
matter. On the contrary, PCA reduces the dimensionality of the data and the univocal relation to the original, high-dimensional firing patterns is therefore lost, the contribution of individual units being difficult to recover.

Our method is related to the gravitational technique of Gerstein et al. (1985) in that it can flexibly manipulate the timescale to detect multi-dimensional activation patterns evolving over different timescales. However, in Gerstein et al., although computations are performed in a multi-dimensional space (multiple particles coalesce when neurons fire in a correlated fashion), the visualization provided therein can only represent pair-wise distances between particles corresponding to neurons (Gerstein et al. 1985; Gerstein and Aertsen 1985; Lindsey et al. 1997) and therefore that method differs from the one introduced here with respect to visualization. Nevertheless, some parallels may be drawn to the gravitational technique. For example, a high-dimensional correlated firing pattern could be represented by a color in our method and by a decrease in the pair-wise distances between the corresponding particles in the method of Gerstein et al. In general, the two methods are complementary and could be combined to explore multi-neuronal correlation structures in more detail.

Color sequences are especially useful when multiple recorded trials are available for the same stimulus. When the evoked activity patterns are stimulus-locked and color sequences are grouped according to their corresponding stimulus, the visualization technique can provide especially salient information. Similar colors would be grouped in larger portions of color sequences and will pop-out from the background. Relatively small temporal jitter of patterns across trials is also detectable, as we have shown in the examples with natural movie stimuli. Furthermore, one can estimate the reliability of pattern occurrence over multiple trials. When patterns are not locked to stimulus onset but exhibit a large range of time shifts relative to the stimulus, their detection is visually less salient, but nevertheless possible by careful inspection or by thresholding the color sequences according to
the specificity of patterns. The same applies when only one single recorded trial is available and therefore there is a single color sequence.

Because activity vectors integrate information over a controllable timescale, color sequences also reflect the snapshot of the activity patterns in the data at a given timescale. The temporal dynamics of cortical networks and the characteristic timescale on which information is encoded has been thoroughly debated, especially for the visual system (Shadlen and Newsome, 1998; Singer, 1999). Color sequences can reveal specific activity patterns that evolve on various timescales, and hence, they may become handy in detecting the characteristic timescale of stimulus-specific multi-neuronal firing patterns (Butts et al., 2007).

The multidimensional framework relying on activity vectors is a generalization of population coding. In the limit, for very large integration time constants, one obtains firing rate vectors as patterns. These firing rate vectors can represent a population vector similar to that described by Georgopoulos et al. (1986). As the time constant is decreased, the emphasis is shifted towards synchronous rate co-variantions and finally to synchronous patterns with millisecond precision. In fact, the multidimensional vectors can be interpreted as correlated fluctuations of firing, on the timescale corresponding to the integration time constant. Therefore, this multidimensional representation of the spiking signals is general, as has already been described in other studies that have combined exponentially-decaying kernels with multidimensional analysis (Gerstein et al. 1985; Nikolić et al., 2007).

Activity patterns detected by the method reflect high-dimensional correlations between recorded neurons, and therefore, as we have shown here, one can extract information that is not accessible to pair-wise techniques such as the cross-correlation (Perkel et al., 1967). Activity vectors are integrated with realistic, exponentially-decaying kernels that mimic synaptic currents (Häusler and
Maass, 2007; Mureșan and Savin, 2007) and exhibit fading memory (exponential decay). Hence, the activity vectors can also represent serial (temporal) correlation in addition to the high-dimensional spatial correlation (multiple neurons). This transcends the representational power of the frequently employed binning and binarization (Puchalla et al., 2005; Schneidman et al., 2006; Yu et al., 2008), which is inaccurate for large bin sizes and fails to consider serial (temporal) correlation (Roudi et al., 2009).

Importantly, the method described here does not distinguish between multi-neuronal activation patterns that arise either by chance or are due to some internal coordination process (Singer, 1999). The purpose of the method is merely the representation of multiple spike-trains in a format appropriate for visualization. Nevertheless, activity vectors can also be used in quantitative analyses and issues regarding their statistical properties, stimulus-specificity, stimulus-locking, etc, can be further investigated. Activity vectors represent a snapshot of the input currents that a hypothetical post-synaptic neuron would receive from the recorded neurons at a given moment in time (Nikolić et al., 2007). The visualization through colors helps identifying these patterns quickly, albeit with some degree of loss in precision.

One aspect that needs to be considered is that color perception is dependent on context (Purves and Lotto, 2003). The distribution of colors in the sequences can either facilitate or hinder the discrimination of colors similar to each other. From this point of view, the visualization method is limited by the fact that, given a certain context, two similar but distinct patterns, may not be distinguished in the color sequences because their similar colors are visually indistinguishable. Therefore, color sequences alone cannot provide accurate quantitative details and may need to be complemented by a further inspection of the model vectors in the 3DKM. Color sequences compress a large amount of multidimensional information onto a limited color space such that there is a tradeoff between the global visualization capability and the limited precision that the visual
representation can offer. Nevertheless, the ordered mapping provided by the 3DKM ensures that confounded patterns are similar to each other and one can always go back to model vectors to make a precise quantitative estimate of the underlying neuronal activity patterns.

Kohonen maps have also been employed before for data classification (Nicolelis et al., 1999). However, data classification, although related to data visualization, has a notably different purpose. While in visualization the emphasis is on finding an intuitive representation for a human observer, in data classification an artificial classifier is used in order to discover stimulus-specific structure in the recorded data. In both cases, the data may be transformed in non-trivial ways, such that it provides to humans or artificial classifiers features that are easy to learn and discriminate. Here, we have shown how Kohonen mapping can be successfully used to enable such a transformation, facilitating visualization by a human observer.

It is important to mention that the visualization technique can be implemented using methods other than Kohonen maps. In principle, any topological mapping method that ensures topology preservation of the input space, combined with a way to assign colors based on topology, can substitute the 3DKM (e.g., Multidimensional Scaling, Generative Topographic Mapping, Locally Linear Embedding etc). We chose Kohonen maps because they are able to preserve the topology of the input space (activity vector space), cope well with large numbers of input samples (no need to compute distance matrices), and their predefined topology in the low-dimensional space can be mapped directly onto a 3D color space. The non-linearity of the Kohonen map is another advantage. For example, between two patterns located at the extremes of the 3DKM cube one does not find a smooth transition from the first pattern to the second but can recover notably different patterns. Only the local similarity is ensured in the Kohonen map and this enables an efficient, less redundant use of the 3D lattice for representing the stereotypical firing patterns.
High-dimensional datasets are difficult to analyze and especially difficult to understand and explore graphically. The visualization technique we have introduced here can provide graphical/visual information about the activation patterns of all neurons across all trials of a given experimental session and can help guide subsequent quantitative analyses. In the future, this visualization technique may be applied to other types of multidimensional signals such as EEG, MEG or local field potential data.

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Software package

Free source code and libraries are available at: http://www.ovidiu.jurjut.ro/sources/visualization/.

References


Figure Legends

Figure 1. From spikes to color sequences. Steps required to construct a color sequence from spike trains: simultaneously recorded spike trains (1), low-pass filtering by convolution with a decaying exponential function (2), clustering activity vectors with a 3D Kohonen map (3), assignment of colors to activity vectors based on the position of their corresponding models in the Kohonen map (4).

Figure 2. Stimulus specific activity patterns revealed by color sequences in a dataset recorded from cat visual cortex in response to drifting sinusoidal gratings. A: Rastergrams of trials (26 simultaneously recorded neurons) in response to 4 different stimuli, shown for 20 repetitions (trials) per stimulus. B: Color sequences of the responses from A. C: Tracking of colors down to neuronal activity patterns (Pattern) that correspond to the actual firing pattern of recorded neurons (Spike Histogram). $\tau = 20$ ms.

Figure 3. Exploring large, multidimensional datasets recorded from cat visual cortex. A: Color sequences for a dataset with 8 stimulation conditions and 20 trials per condition. The length of the red arrow indicates the speed of the moving stimuli. B: Trajectories computed on the dataset from A, mapped on the 3D lattice of the Kohonen map. C: Color sequences for a dataset with three natural movie stimuli (28 seconds long). Only 6,500 ms are shown. $\tau = 50$ ms for A and B and 20 ms for C.

Figure 4. Detection of stimulus-locked activity on different timescales in artificial datasets. A: Color sequences of one stimulation condition from a dataset containing stimulus-specific synchronous spikes across 5 out of 25 neurons (joint-spike events). B: Color sequences corresponding to a stimulation condition from a dataset containing stimulus-specific rate covarianations in 3 out of 25 neurons. In both A and B color sequences were computed with two
different integration time constants of 5 and 50 ms. Occurrences of joint-spike and rate covariation events are marked by black arrows.

**Figure 5.** Analysis of brain-state fluctuations (non-stationarities). A: Color sequences shown in the order the trials were recorded, for two different recording sessions (about 26 hours apart) evoked by the same stimuli, recorded from the same animal, same electrodes, and same neurons (SUs). B: Time-resolved coefficient of variation computed on the patterns corresponding to the two sessions in A. Error bands on the curves represent s.e.m. C: Autocorrelation histograms for the activity of one single-unit recorded in the two sessions (top two panels) and comparison of oscillation scores in the beta-high/gamma band for all neurons in the two sessions (bottom panel: mean and s.d. shown). Blue and red colors in all plots correspond to the recording sessions 1 and 2, respectively. OS and CS denote oscillation score and confidence score, respectively (Mureșan et al., 2008). \( \tau = 20 \) ms.

**Figure 6.** Finding meaningful patterns through pattern specificity. A: Color sequences of responses to 12 sinusoidal gratings. B: Thresholded color sequences from A, PSI \( \geq 0.25 \). C: Thresholded color sequences from A, PSI \( \geq 0.5 \).
Figure 1

1. Spikes

2. Activity Vectors

3. 3D Kohonen Map

4. Color Sequence
Figure 2

A

Stimulus

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Time [ms]

B

Stimulus

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Time [ms]

C

Stimulus

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<th>3D Kohonen Map</th>
<th>Pattern</th>
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20 trials

250 ms
Figure 3

A

Stimulus On

Stimulus Off

B

C

Stimulus On

Stimulus Off
Figure 4

A. Synchronous Spikes

B. Rate Covariations

\( \tau = 5 \text{ ms} \)

\( \tau = 50 \text{ ms} \)
Figure 5

A. Trial presentation order

B. Pattern CV

C. # Coincidences / s

Session 1 Session 2

Stimulus presentation

OS: 4.01 CS: 0.72
OS: 18.7 CS: 0.88

Session 1 (A.left) Session 2 (A.right)
Figure 6

A. Stimulus On $\text{PSI} = 0$ Stimulus Off

B. $\text{PSI} = 0.25$

C. $\text{PSI} = 0.5$
Figure S1

A: Error of approximating the activity vectors with the model vectors from the Kohonen map, computed at different steps. Error bars represent SDs over 10 maps.

B: Histogram of Pearson correlation coefficients ($r$) between activity vectors and their corresponding model vectors.

C: Histogram of Euclidean distances between pairs of model vectors belonging to a Kohonen map, computed for two datasets. Distances are normalized to the number of vector elements (# neurons).
Figure S2. Complete set of color sequences for a dataset consisting of 26 neurons from cat area 17. Neural activity was recorded in response to drifting sinusoidal gratings moving in 12 different directions (steps of 30°). There are 20 color sequences grouped for each stimulus (20 trials per stimulation condition). Integration time constant, $\tau = 20$ ms.
Figure S3. Full-length color sequences for a dataset consisting of 34 neurons from cat area 17. Neuronal activity was recorded in response to three long movies with natural scenes (28 seconds in length). There are 20 color sequences grouped for each stimulus (20 trials per stimulation condition). Integration time constant, $\tau = 20$ ms.
Figure S4. Detection of higher order joint-spike events (JSEs) that are not observable with pairwise techniques. A: Color sequences showing the occurrence of a JSE with 5 active neurons out of 25 (N6, N12, N15, N22, N24). B: Cross-correlation histograms between the pairs of active neurons of the JSE fail to reveal significant central peaks. Trials were 30 seconds long and each contained one single JSE occurrence at the same location, marked by a black arrow in A.
Figure S5. Analysis of brain-state fluctuations (non-stationarities) in a dataset recorded in response to sinusoidal gratings of high spatial frequency. A: Color sequences are shown in the order in which the trials were presented during the experiment (temporal order). B: Color sequences are grouped by the stimulus. Marked non-stationarity of responses is revealed in both cases. Integration time constant, $\tau = 20$ ms.