Supplementary Material

Comparing recording radii of tetrodes in cat area 17:

Gray et al. (1995) and the current study

to

Three-dimensional Localization of Neurons in Cortical Tetrode Recordings

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Summary

The first, and probably most referenced, estimates of the recording radius of a tetrode (Gray et al., 1995; Maldonado et al., 1997) represent an important scientific context to our study. Because the Gray et al. estimate was obtained by triangulation, a method that, unlike dipole localization, is not based on a physical source model, comparison demands an analytic treatment that bridges the difference in methods, which we carry out below. First we provide the mathematical formalism of triangulation. A crucial part of this is the calculation of the form factor that characterizes the tetrode. Then, using these tools, we show that the differences between our results and Gray et al.'s estimate of the recording radius can be explained by a difference in probe size and not by a difference in methods. Moreover, we show here that Gray et al.'s different method, when re-analyzed, lend further support for the dipole source/local arbor interpretation.

Triangulation

Gray et al., along with several other groups (Gray et al., 1995; Maldonado et al., 1997; Bartho et al., 2004; Buzsaki, 2004; Seshagiri and Delgutte, 2007), have estimated the recording radius of their tetrodes using a "triangulation method".

The method is based on a phenomenological approximation of the dependence of the magnitude of the extracellular action potential (EAP) amplitudes on distance from the neuron, rather than on a physical source model. In order to make our comparison as transparent as possible, we formalize the procedure of triangulation. The starting point is to assume that the potential V varies as a function of distance r according to

$$V(r) = \exp(-r/\lambda), \qquad \text{Eq.(1)}$$

where r is measured from the cell and λ is the space constant of decay. The recording radius is then defined as the range over which an n-fold decay of V takes place. The number n, an approximate signal-to-noise ratio, is defined by

$$n = V_{\max} / V_{\min} \,, \qquad \text{Eq.(2)}$$

where $V_{\max} \approx V_{source}$, the largest observed EAP amplitude (e.g., nearest to the source), and $V_{\min} \approx V_{noise}$, the smallest observed EAP amplitude, similar in amplitude to the multiunit background. Combining equations 1 & 2 leads to an estimate of the recording radius,

$$R = \lambda \log(n) \,. \qquad \text{Eq.(3)}$$

Gray et al., the first authors to use this method (Gray et al., 1995) set $V_{\max}/V_{\min} = 10$ because they never observed a ratio larger than this; other authors may have adopted the same value without further verification.

The exponential space constant, λ, is estimated from the voltages recorded at all pairs of contacts. Specifically, let V_i and V_j denote the mean EAP amplitudes recorded from a single unit by the *i*th and *j*th tetrode contacts of the tetrode, one of the 6 possible such pairs. For each cell, the 6 voltage ratios V_i/V_j are calculated, in each case, ordering i and j so that $V_i/V_j \leq 1$. This ratio is then averaged for all cells and contact pairs to obtain the average contact-pair potential attenuation (a number <1):

$$A = \left\langle V_i / V_j \right\rangle . \qquad \text{Eq.(4)}$$

Similarly, average contact-pair path difference,

$$P = \left\langle \Delta r_{ij} \right\rangle \qquad \text{Eq.(5)}$$

is calculated, where $\Delta r_{ij} = \left| r_i - r_j \right|$ is the difference of the distances r_i and r_j measured from a given cell to the *i*th and *j*th tetrode contacts, respectively, and the average is taken over all cells and contact pairs. With these quantities, The exponential space constant, λ, is estimated from

$$A = \exp\left(-P/\lambda\right), \qquad \text{Eq.(6)}$$

The average contact-pair potential attenuation, A, can be directly measured from the voltage data. The average path difference, though, requires an indirect strategy, since cell location is not known. (It is impossible to localize each cell from the measurement of EAP amplitudes on the 4 tetrode contacts by triangulation because fitting the exponential approximation requires 5 parameters—the 3 location coordinates and the intensity of the source in addition to the space constant—more than the available 4 data points.) Assuming that recorded cells lie in random directions from the tetrode, it can be shown that the average path difference, P, is determined by the product of the contact separation, Δs, and a scalar form factor, c_T, i.e.,

$$P = c_T \Delta s \ . \qquad \text{Eq.(7)}$$

Combining the above equations, we obtain the formula for the recording radius that Gray et al. implicitly used (but did not explicitly write out):

$$R = -\Delta s\ c_T \log\left(n\right)/\log\left(A\right). \qquad \text{Eq.(8)}$$

The form factor, c_T, is defined below.

Form factor

The form factor is defined as the mean contact-pair path difference, averaged over all possible cell-probe configurations, normalized by mean contact separation. Specifically, we average path differences for cells placed uniformly within a volume of radius r, and take the limit as r approaches infinity. This approximation is valid because the dependence of path length difference on absolute cell-probe distance is very weak, especially at the cell-probe distances that is typical of isolated cells.

Below, we give the form factor for typical tetrode geometries, and for two kinds of cell distributions: uniform in the plane of the tetrode, and uniform in space. In all cases, c_T is substantially smaller than 1. As equation 8 shows, the triangulation estimate of the recording radius depends on using an accurate value for this quantity.

For an idealized planar wire tetrode with contacts that are centered on the corners of a square of size Δs , c_T can be determined analytically. For random sampling in 2 dimensions (the plane of the contacts), it is

$$c_{T\square,2D} = 2\left(2+\sqrt{2}\right)\Big/\left(3\pi\right) \approx 0.72\,; \qquad \text{Eq.(9)}$$

for random sampling in 3-dimensions, c_T it is somewhat smaller:

$$c_{T\square,3D} = \left(\pi/4\right)c_{T,2D} \approx 0.57\,. \qquad \text{Eq.(10)}$$

Numerical simulations show that among all tetrode configurations in the class of planar rectangles (blue curves in Figure 1.), the square configuration (vertical dotted line) maximizes the form factor. For distortions of the square into a rhombus (a typical result of wire splaying (Jog et al., 2002; Chelaru and Jog, 2005)) that becomes progressively more elongated, the form factors in both 2D and 3D gradually decline to their minima of

$$c_{T,2D} = 2/\pi \approx 0.63 \qquad \text{Eq.(11)}$$

$$c_{T,3D} = 0.5 \qquad \text{Eq.(12)}$$

at maximal distortion of the rhombus (corresponding to elongation index ±2 in Figure 1.). In both extremes, the contact rectangle collapses into a linear (1D) configuration where the pair of contacts defining the small diagonal of the rhombus collapses into one.

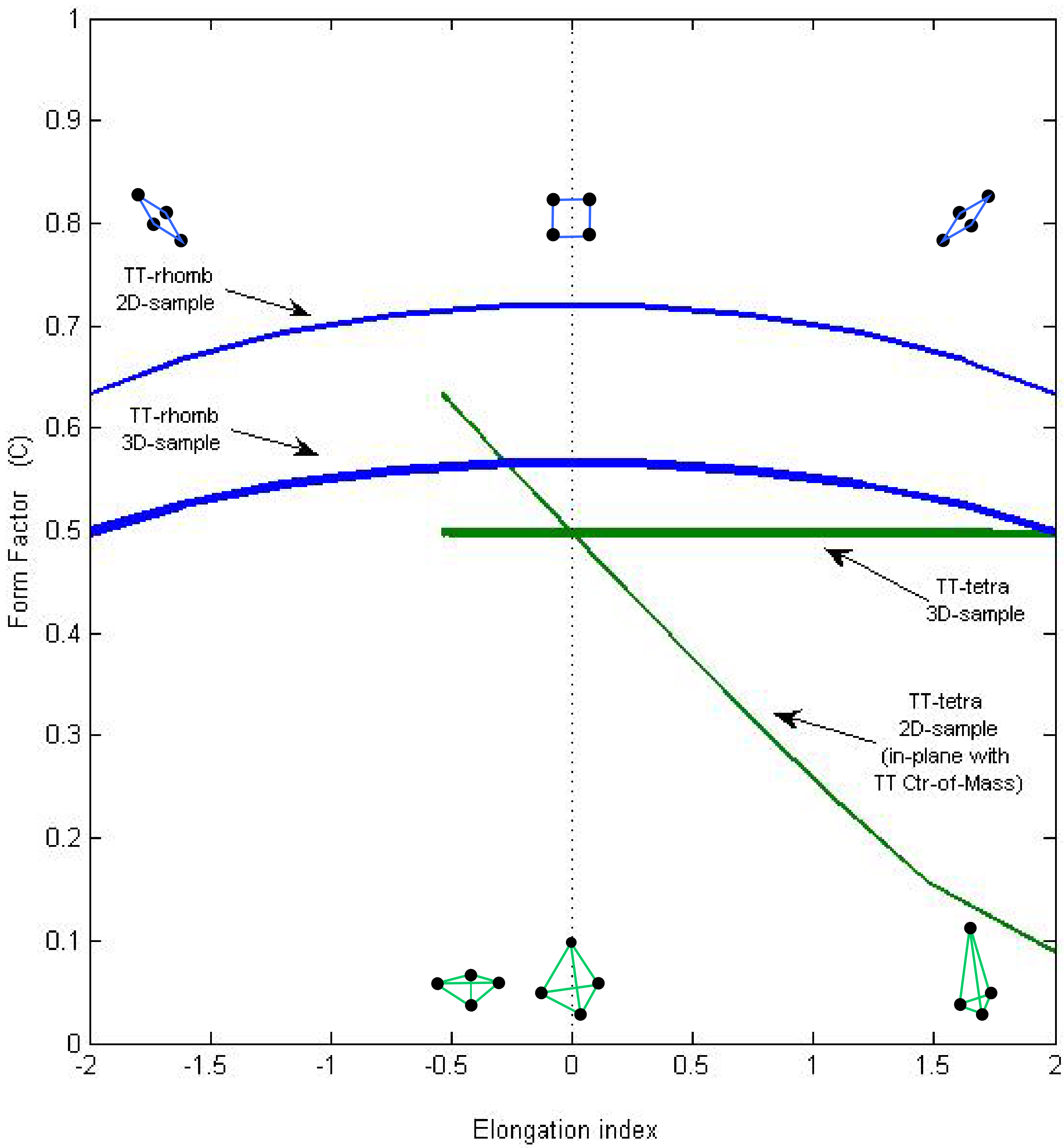

Figure 1

The form factor is used to quantify the spatial sensitivity of tetrodes in tetrahedral (3D) and rhombus (2D) configuration as a function of their elongated shape and whether cell sampling is in 2D or 3D. Form Factor is defined as the mean contact-pair path difference, normalized by mean contact separation. Elongation index here is defined by a difference in characteristic contact separations.

It is the diagonalA - diagonalB difference for the rhombus configuration (all edges = 1).

It is the lateral edge - basal edge difference for the tetrahedron configuration (mean edge = 1).

Correspondingly, numerical simulation indicate that the form factor of the 3D tetrode, whose contacts are arranged in a tetrahedral contact configuration by design, is a constant

$$c_{T\blacktriangle,3D} = 0.5, \qquad \text{Eq.(13)}$$

independent of the elongation of the tetrahedron (green flat line in Figure 1.). At the right extreme (elongation index equals 2), the green curve is continuous with the blue curve. This is not surprising: here the tetrahedron, as the rhombus, is reduced into a 1D configuration (in this case, 3 contacts collapse into 1). The support for the tetrahedron configuration extends on the left only to the abscissa where the elongation index assumes the value of $-\sqrt{3}/3 \approx -0.58$, at which point the tetrahedron collapses into a flat 3-pointed star configuration.

The triangulated recording radius of wire tetrodes in Gray et al (1994)

With the above method of triangulation, Gray and coworkers estimated a recording radius of $R_{\exp} \approx 65\,\mu\text{m}$ for twisted wire tetrodes in cat V1 (Gray et al., 1995; Maldonado et al., 1997). To reproduce their calculations, we used $c_{T\square,3D} = 0.57$ for random 3-dimensional cell sampling, and substituted the values that Gray et al. reported, (the nominal $\Delta s = 15\,\mu\text{m}$ contact separation, $n = V_{\max}/V_{\min} = 10$, and their measured $A = 0.59$) in equation (8). The resulting $R = 37$ µm radius is puzzlingly smaller than the $R \approx 65$ µm Gray et al. reported, by a ≈ 0.57 factor, suggesting that Gray et al. erroneously omitted the c_T form factor carrying out their calculation. Alternatively, the reported radius reflects an effective average contact separation ($\Delta\tilde{s} \approx 25\,\mu\text{m}$) that is $1/c_T = 1.7$ times larger the reported nominal value. An almost equal enlargement of the contact separation caused by splaying of the wire tips has been documented in these tetrodes (Jog et al., 2002; Chelaru and Jog, 2005).

The triangulated recording radius of Thomas tetrodes in our study

To compare the triangulation and dipole localization methods directly, here we use the triangulation approach to derive a recording radius for the Thomas tetrode in cat V1. As indicated in the main text, the mean contact separation on the tetrode we used to record from the cat V1 neurons was $\Delta s = 45$ μm, and the recording radius estimated by dipole localization (and defined by the distance of the farthest of the 10 recorded cell in lieu of 95-percentile of the localized sample), was $R_{cat} \approx 124$ μm.

For the triangulation calculation, we used $c_T = 0.5$ (Eq. 13), because the contacts on the Thomas tetrode formed a nearly perfect tetrahedron. In our cat sample, the average contact-pair attenuation was somewhat weaker ($A = 0.64$), and the ratio of the largest signal amplitude ($V_{\max} \approx 157$ μV) and the smallest at background level ($V_{\min} \approx V_{noise} \approx 20$ μV) was somewhat smaller ($n = V_{\max}/V_{\min} \approx 8$). With the substitution of these values into Eq. (8), the triangulated recording radius was $R_{\exp,cat} \approx 106$ μm, about 20% smaller than determined via dipole localization ($R_{cat} \approx 124$). With Gray's larger nominal signal-to-noise ratio ($n = 10$), the triangulated recording radius was $R_{\exp,cat} \approx 117$ μm, within a few percent of the value determined by dipole localization.

Triangulation rule of thumb for dipole regime

Here we show how the quantities determined in the course of carrying out the triangulation measurement can be re-interpreted to indicate the nature of the source model (i.e., monopole, dipole, or quadrupole).

The basic idea is that an exponential decay, $\exp\left(-r/\lambda\right)$, can be fit, locally, as a power law r^{-k}. The best-fitting power law exponent, k, changes with distance, according to $k \sim r/\lambda$. Thus, at the distance of the recording radius, R, the best-fitting power law exponent is given by

$$k \approx R/\lambda . \qquad \text{Eq.(14)}$$

But, according to Eq. (3) above, this ratio is also estimated by the log signal-to-noise ratio:

$$R/\lambda = \log(n). \qquad \text{Eq.(15)}$$

Substituting eq.(15) and Eq.(2) in Eq.(14), yields

$$k \approx \log(V_{\max}/V_{\min}). \qquad \text{Eq.(16)}$$

That is, the triangulation approximation of the best fitting exponent offers a simple phenomenological rule of thumb for the best fitting kind of equivalent source model:

$3 \le V_{\max}/V_{\min} < 5$ implies $1 \le k \le 1.5$, i.e., monopole. Eq.(17)

$6 \le V_{\max}/V_{\min} \le 10$ implies $1.75 \le k \le 2.25$, i.e., dipole Eq.(18)

$15 \le V_{\max}/V_{\min} \le 25$ implies $2.75 \le k \le 3.25$, i.e. quadrupole Eq.(19)

In particular, the observed values of the ratio $V_{\max}/V_{\min}$ (10 in Gray et al., 8 in our data) offer further support for the appropriateness of the dipole model.

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