UC Berkeley

UC Berkeley Previously Published Works

Title

Relation of Olfactory EEG to Behavior: Spatial Analysis

Permalink

<https://escholarship.org/uc/item/55p7z3ff>

Journal

Behavioral Neuroscience, 101(3)

Authors

Freeman, Walter J, III
Baird, Bill

Publication Date

1987

Copyright Information

This work is made available under the terms of a Creative Commons Attribution License, available at <https://creativecommons.org/licenses/by/3.0/>

Peer reviewed

Relation of Olfactory EEG to Behavior: Spatial Analysis

Walter J. Freeman and Bill Baird
Department of Physiology-Anatomy
University of California at Berkeley

The hypothesis that odor-specific patterns of mitral cell activity during odor discrimination might be found in the corresponding spatial patterns of electroencephalogram (EEG) amplitude over a surgically accessible segment of the bulbar surface was tested in rabbits with chronically implanted electrode arrays. The spatial spectrum of the bulbar EEG was derived and compared with the spectrum predicted for the granule cell generator. Spatial filters were devised to identify, enhance, or remove the granule cell contribution to the EEG. Spatial deconvolution was applied to the filtered granule cell activity patterns to correct for distortion caused by volume conduction. The results indicated that the bulb generated odor-specific spatial patterns in rabbits trained to discriminate between two odors. The odor-specific information was not localizable to subsets of channels. This suggested that the discriminative output of the bulb involved the entire structure, even though the receptor input was delivered to limited subsets of mitral cells.

The aim of this study was to define the spatial passband of odor-specific information in the electroencephalogram (EEG) of rabbits trained to discriminate odors and to compare this with the passband of granule cells that are known to be the major contributors to the bulbar EEG. The aim was achieved through spatial spectral analysis of the olfactory EEG and the use of various spatial filters in the spatial frequency domain.

The neural activity of the bulb is determined mainly by interactions between the mitral (here including tufted) excitatory cells and inhibitory interneurons—the internal granule cells. Receptor input excites the mitral cells that in turn excite granule cells and are inhibited by them, giving rise to oscillatory activity in the range of 40 to 80 Hz. Each local region, corresponding to a glomerulus on the order of 0.1 to 0.2 mm in diameter with a mean center-to-center distance of 0.25 mm, contains many thousands of cells and can be regarded as an “oscillator.” The oscillators corresponding to the roughly 2,000 glomeruli in each bulb of the rabbit are coupled mutually by excitatory axon collaterals running between mitral cells in all directions under the bulbar surface. Receptor input during inhalation tends to destabilize this large set of coupled oscillators, resulting in a brief burst of oscillatory EEG activity. Although the activities of both the mitral and granule cells are involved in the production of the burst, it is the action potentials of the mitral cells that constitute bulbar output, and it is the synaptic currents of the granule cell dendrites that predominate in the EEG potentials.

However, these cells are not the sole contributors to the EEG. The extracellular tissue forms a common resistive path in which the potentials of currents sum from many nearby and some faraway neurons. These poorly defined contributions constitute “noise” in respect to the desired “signal” from granule cells, so that appropriately designed filters are needed. Further, each local subset of granule cells is oriented perpendicular to the bulbar surface; its synaptic currents spread outwardly in all directions and establish a bell-shaped field of potential at the surface, which is described by a “point spread function” in the terminology of optics. Each electrode in an array of electrodes on the bulbar surface detects the weighted sum of granule cell activity in an extended part of the bulb, so that each instantaneous spatial activity pattern in the set of granule cell is blurred in its electrical manifestation at the surface. The EEG pattern must be brought back into focus in order to resolve spatial pattern differences relating to odors. This operation, known as the “software lens” (Freeman, 1980), is exquisitely sensitive to noise at high spatial frequencies. The development and use of a low-pass spatial filter tailored to the granule cell characteristics is therefore needed for EEG spatial pattern measurement. These measurements are essential for the detection and characterization of odor-specific information in the EEG that is required for understanding how the bulb functions in its task of odor identification.

Method

Recording

Electrodes. For two-dimensional (2-D) spatial analysis, electrode arrays (8 × 8, 4 × 4 mm) were prefabricated with their connectors (Eastman, 1975). Individual electrodes were constructed of 0.25-mm-diameter stainless steel with platinized tips. The original center-to-center electrode distance averaged 0.5 mm. Due to the wires fanning back from the array face, grinding it for replatinizing prior to reuse increased average interelectrode distance to 0.57 mm. This larger spacing affected the interpretation of spatial frequency spectra in a

This article was supported by Grant MH06686 from the National Institute of Mental Health and Grant NS16559 from the National Institute of Health.

The authors gratefully acknowledge assistance from G. Wesley Davis in deriving spatial spectra, Ron Katznelson in interpreting them, G. Wesley Davis and Gonzalo Viana Di Prisco in data collection, Kamil Grajski in statistical analysis, and Brian Burke in system development and programming.

Correspondence concerning this article should be addressed to Walter J. Freeman, Department of Physiology-Anatomy, University of California at Berkeley, Berkeley, California 94720.

manner analogous to increasing the digitizing interval in temporal frequency analysis (Digital Signal Processing Committee, 1979).

For one-dimensional (1-D) spatial analysis, a circular array was devised. Two 15-cm-diameter gears each with 64 teeth were mounted in a rigid form 1 m apart. Stainless steel wires 0.1 mm in diameter were strung between the apposing pairs of grooves to form a cylinder. One gear was rotated so as to form two cones. The apices were embedded in dental cement, the two halves cut back, strung to connectors, and their faces ground and polished. Each array was 4.0 mm in diameter, which is optimal for surgical placement onto the lateral surface of the bulb. The 0.2-mm interelectrode distance gave sufficiently close spacing to evaluate the optimal interelectrode distance at which to prevent aliasing in the spatial spectrum. The shape of the electrode gave a periodic boundary condition in subsequent Fourier analysis.

Recording procedures. Surgical and recording protocols have been previously described (Freeman & Schneider, 1982; Viana Di Prisco & Freeman, 1985). Electroencephalographic (EEG) bursts were selected by editing from six second trials under appetitive and aversive conditioning (Viana Di Prisco & Freeman, 1985) and from records up to 4 min in length that were taken from satiated rabbits at rest. Up to 2,400 frames (4.8 s) were stored on disc for off-line presentation as density plots on an oscilloscope in the form of a movie. Additionally, sequential bursts were detected by a moving root mean square (rms) average of the ensemble time average of the 64 times series, and up to 1,000 bursts were stored for movie display. Polygraph records of respiration and EEG traces from representative channels were taken concomitantly with the 64 digitized records for off-line correlation.

Fourier Analysis

Recorded spatial patterns of EEG took the form

$$v(x, y, t),$$

where v represents the amplitude of the field potential at array location (x, y) at time t . For spectral analysis (Freeman, 1978, 1987; Nunez, 1981) the data were Fourier transformed into the frequency domain:

$$V(w_x, w_y, w_t) = \iiint v(x, y, t) \cdot \exp(-jw_x x) \exp(-jw_y y) \exp(-jw_t t) dx dy dt, \quad (1)$$

where w_x and w_y represent spatial frequencies in radians/mm in the two dimensions of the array and w_t represents the temporal frequency in radians/s (radians = 2 times frequency, in c/mm or c/s) (Childers, 1978). Due to constraints on computer time, the spatial and temporal integrations were done separately.

Decomposition of oscillation into several major component frequencies. Temporal spectral analysis showed that the EEG burst had a common waveform on all channels (Freeman & Viana Di Prisco, 1986b). Frequency components were extracted by successive identification of a temporal spectral peak of the ensemble average and subtraction of a fitted cosine at that frequency and phase on each channel until the final residual contained less than 3% of the temporal spectral energy of the burst. This procedure was itself a spatial filtering procedure, because activity in a subset(s) of channels at a given frequency that failed to sum to more than 3% of temporal spectral energy in the ensemble average remained in the residuals.

Formation of "spatial patterns" of amplitude and phase of all channels at each component frequency. The sum of five cosines at the identified frequencies and phases was fitted to each of the 64 traces in a burst by nonlinear regression. The residuals on channels averaged 20% of EEG energy. The resultant amplitude and phase patterns

were noisy estimates of their putative "true" values conforming to the individual components. The spatial patterns of amplitude and phase of each component conformed to the focus or "signature" pattern (Freeman & Viana Di Prisco, 1986a) characteristic for each subject.

Spatial spectral analysis (1-D and 2-D). Spatial frequency spectra (1-D and 2-D) were calculated for individual components. The temporal Fourier transform was taken at the five identified frequencies and phases on each channel:

$$V_i(x, y, w_t) = \int v(x, y, t) \exp(-jw_t t) dt; \quad i = 1, \dots, 5. \quad (2)$$

The spatial transform was performed on the real and imaginary parts of the temporal spectrum:

$$\begin{aligned} V_i(w_x, w_y, w_t) &= \iint \text{Re } V_i(x, y, w_t) \exp(-jw_x x) \exp(-jw_y y) dx dy \\ &+ \iint \text{Im } V_i(x, y, w_t) \exp(-jw_x x) \exp(-jw_y y) dx dy. \end{aligned} \quad i = 1, \dots, 5. \quad (3)$$

In eventual routine use, the transform was applied only to the dominant component, that is, $i = 1$, with major savings in computer time.

For detailed analysis of the 2-D spatial spectrum, each 8×8 data set was embedded in a matrix (32×32) of zeroes, a Hamming window applied, and the data translated to bring the zero spatial frequency values to the center of the 2-D plot of power (Gonzalez & Wintz, 1977). Inspection of density plots showed no significant departures from radial symmetry. The 2-D power spectrum was radially integrated to give $V_i(w_r, w_t)$, where

$$w_r = (w_x^2 + w_y^2)^{0.5}, \quad i = 1, \dots, 5. \quad (4)$$

The values for power were divided by the power at the lowest accessible frequency (0.04 c/mm) as the best available estimate of zero-frequency power, and the natural logarithm of normalized power was plotted as a function of spatial frequency in c/mm.

Spatial filtering. For spatial filtering, each data set was embedded and translated, but Hamming was omitted in order to avoid nonlinearities (Katznelson, 1982; Nunez, 1981). The real and imaginary parts were multiplied by the 2-D spatial filter of choice, and the filtered data were transformed back to the spatial domain by the inverse 2-D Fourier transform. This method was preferable to convolution in the spatial domain, because it tied the design of the spatial filters directly to the spatial spectral properties of the EEG.

Theoretical Simulation of EEG for Prediction of Spatial Spectra Due to Granule Cells Only

Analysis of spatial power spectra was undertaken by comparison with theoretically derived spectra based on the following assumptions and conditions (Freeman, 1975): (a) The elemental generator was the granule cell, which sustained a current source-sink pair at an average depth from the surface of 0.58 mm., separated by ± 0.17 mm along a line perpendicular to the surface. (b) Neural current flowed in a homogeneous resistive medium bounded by the nonconducting electrode face on one side. (c) The electric dipole point charges that were equivalent to generator sources and sinks lay in two planar surfaces under and parallel to the array face.

Modeling of granule cell activity and noise. Granule cell activity was modeled by a 2-D matrix of dipoles (25×25) 0.2 mm apart. In position, magnitude varied with a symmetric bivariate Gaussian distribution centered under the array. In time, magnitude varied sinusoidally at 60 Hz with unit peak amplitude, zero mean, and zero

phase. This spatiotemporal activity constituted the signal. Additive noise was generated independently for each channel by a Gaussian random number generator. The signal and noise were each normalized to zero mean and unit standard deviation. The noise was multiplied by a fixed value for the entire simulated burst to give a desired signal to noise ratio.

Modeling of array recording. Array recording at the bulbar surface was modeled by calculating a potential for each channel of a simulated electrode located 0.58 mm above the dipole matrix. The surface potential was calculated from the sum of the reciprocals of distance for that point to the sites of the two point charges in each dipole, corresponding to a current source-sink pair (Freeman, 1975). The inverse procedure whereby a surface potential is "refocused" to a specified depth below the array is termed *spatial deconvolution* (Freeman, 1980). The simulated surface recording was normalized to unity at its maximum at the epicenter. Digitizing was simulated by truncation of values. The 64 time series at 0.5 mm spacing, each with 38 points at 2 ms intervals, were stored, digitally smoothed, detrended, and processed in the same way as EEG bursts.

Computational Procedures

Linear and nonlinear regression with least squares deviation were computed in accordance with procedures described by Golub and Pereyra (1973). The signal-to-noise ratio (S:N) was given by the ratio of the sum of squares of the fitted amplitudes to the sum of squares of the residual for each channel. The implementation of each procedure by computer program was tested by processing simulated EEG bursts for a range of S:N values.

Cluster analysis was performed on sets of up to 120 bursts per subject. Each channel on the array was treated as a euclidean dimension. A multidimensional scaling algorithm projected the 64 space to two dimensions while preserving relative distances between points (Sammon, 1969). Clustering in the reduced space expressed the degree of similarity of burst spatial patterns.

Results

Overview of "Signature" Spatial Amplitude Patterns

Four-minute series of 2-D rms amplitude patterns. Up to 1,000 bursts were recorded and edited seriatim for 4 min in the rest state (no odor or other stimulation) from each of 14 subjects and stored as 8×8 matrices of rms amplitudes. Averaged rms amplitude matrices were displayed as density plots on an oscilloscope. These showed that each subject had its characteristic signature pattern of relatively high amplitude in a contiguous portion of the array window, which appeared as a "focus." In 7 of the subjects, a maximum of the focus lay within the window. In the other 4, it was at or beyond the edge. In 3 subjects, there were two foci within the window, separated by 2 mm or more.

Individual bursts recurred at rates up to 10/s. Sequential display of rms patterns showed that their foci were restricted within the domain indicated by the signature pattern, but with variable location, size, shape, amplitude, and number of local maxima (see Figure 1). These variables could not be predicted from preceding bursts or from the amplitude and shape of the accompanying respiratory wave. Comparison of burst density plots with polygraph traces of selected channels showed only that burst duration decreased with increasing respiratory rate.

Successive single frames (8×8 samples at the digitizing interval of 2 ms) of the EEG activity between bursts showed chaotic activity. On average, however, the signature pattern was reproduced. Bursts were seen to emerge within the indicated domain. The emergent waves reversed polarity sequentially at 40 to 80 Hz in place. At the available recording resolution, the waves appeared to be nonpropagating. The term *standing* was inappropriate owing to lack of a reflective boundary for each focus.

Comparison of signature of two animals by cluster analysis. Cluster analysis of representative subsets of 120 bursts, 60 from each of 2 subjects, showed clear separation of the two sets. Within each set, there was a single mode for the distribution from each subject (see Figure 2, upper panel). In those subjects having a double maximum in the focus, successive bursts were labeled according to whether one or the others' location had the larger amplitude; also the upper and lower 5% of cases with respect to overall amplitude were identified with separate plotting symbols. The results (Figure 2, lower panel) showed that the several types of bursts fell into different parts of the unimodal distribution and that overall amplitude had little influence on location within the distribution. By these tests, within-subjects and within-sessions sets of control bursts could be treated as random samples from a common larger population.

Temporary shift of signature pattern. An exception to unimodality was observed once in 2 out of 3 subjects that were being familiarized with the recording box but had not undergone training. During the course of the recording session, but not within a 4-min recording period, an apparently spontaneous arousal occurred manifested by a sustained increase in respiratory rate with exploratory movements of the head and ears. Comparison of burst sequences and averages from 4-min periods before and after the onset of arousal showed that the spatial pattern had changed to an extent that the differences were as great as those previously seen under training (Freeman & Schneider, 1982). Although not reproducible, the changes were evidence for the capacity of the pattern of bulbar activity to shift in seconds or perhaps a fraction of a second from one stable state to another one. In general, familiarization minimized such adventitious changes and stabilized the EEG spatial patterns.

The Experimental EEG Spatial Spectrum Compared With the Simulated EEG Spatial Spectrum

1-D spectra of rms amplitudes. A 1-D array burst recording yielded a pattern of rms amplitudes exemplified by the solid dots in Figure 3 (upper panel). The location of maximal amplitude was set arbitrarily at $x = 0$. The lower frame shows the rms amplitude of a simulated burst centered at $x = 0$ mm with $SD = 1$ mm and S:N = 4.0 (see Methods). The dotted curve in the inset shows a linear cross section of the bivariate Gaussian distribution, which defined the spatial variation of activity in the dipole matrix used to simulate granule cell activity. The inset solid curve shows the falloff in density during measurement owing to the curvature of the array centered at $x = 0$. The depth and point separation in the generator matrix are indicated to scale in mm, shown below

Figure 1. Spatial patterns of root mean square (rms) amplitude from a rabbit for 40 consecutive bursts (left to right in successive rows) concurrent with inhalations at 4.2/s. (The 7-level darkness scale of printed symbols is [$\#$ $+$ $=$ $-$ $\cdot$], in which the darkest is highest amplitude. Activity peaked most commonly just to the right of the center of the array; less commonly it peaked in the lower left corner over the ventroposterior lateral surface of the left bulb. The purpose of the study was to find invariants with respect to odor that were embedded in these data.)

the abscissa. The predicted potential in the absence of noise is shown in each frame by the solid curve.

1-D spectra of component frequency amplitudes. The 1-D fast Fourier transform (FFT) of the amplitudes of the dominant cosine, fitted to the 64 EEG traces at identified frequency and phases, gave a spectrum (see Figure 4, upper panel) that

about 0.4 c/mm and remained near a fixed level or asymptote for frequencies up to 2.5 c/mm. Simulation gave the same pattern (lower panel). The spatial spectrum gave three elements of information about the burst. (a) The level of the asymptote at higher spatial frequencies decreased with increasing S:N ratio. (b) The rate of decrease in power with increasing spatial frequency decreased with increasing S:N ratio.

Figure 2. Upper panel: Sets of control bursts from 2 rabbits (A and B) expressed as points in 64-space in the coordinates of root mean square (rms) channel amplitudes. (The two axes with the largest variances in arbitrary units were selected for a 2-D projection of these points. Then by the nonlinear scaling algorithm of Sammon [1969], the projection was modified iteratively so as to optimize the clustering of bursts most similar in spatial pattern as represented by the distance between them. The successful outcome is shown by the separation of bursts from 2 rabbits each with its "signature" pattern. The points for each rabbit were unimodally distributed. Lower panel: Control bursts from the same rabbits as those in Figure 1 but labeled as to whether the peak lay to the right (solid dots) or the left (open dots) of the center of the array, and whether the mean rms amplitude was in the top 5% or bottom 5% of the 120 bursts. (The distribution of points in arbitrary units was unimodal, which indicated that the whole set could be treated as random samples from a common larger population. This was a premiss of major importance in further statistical analysis.)

related to the width of the active focus expressed by its SD . (c) The break point of the spectrum and the reciprocal of the greatest linear dimension of the array defined the accessible spectral passband of the signal (in the 1-D example shown,

0.08 to 0.40 c/mm, and in the 2-D case to be discussed, 0.17 to 0.50 c/mm).

Parameter sensitivity. The simulation also showed that the spatial spectrum was relatively insensitive to changes in the depth and separation of the generator. For each 0.1 mm of increasing depth from 0.40 mm to 0.80, the slope of the low-frequency spectrum increased as if the width of the focus increased by 0.1 mm. Variation of dipole separation by ± 0.10 from 0.17 had negligible effects.

The resistive boundary at the surface had no effect on normalized surface measurements. The curvature of the array in itself did not affect the results significantly. A shift of the fixed SD Gaussian epicenter to a point within the array caused an over estimate of burst focal width (see Figure 5). With the circular array, there was no way to locate the epicenter of a focus and therefore no way to estimate width. The main value of the configuration was to show the absence of spectral peaks above 0.5 c/mm and thereby to validate the continued use of interelectrode distances on the order of 0.5 mm.

Comparison with other theoretical predictions. The form of the 1-D spectrum for activity in a planar dipole layer recorded above its surface has been predicted by Katznelson (1982),

Figure 3. Upper panel: The root mean square (rms) amplitude (solid dots) for a single representative burst recorded from a circular array 2 mm in radius of 64 electrodes 0.2 mm apart. (The channel with peak amplitude was set at $x = 0$.) Lower panel: Computed simulation. (The points below the abscissa show a cross-section of a 2-dimensional [2-D] array of point sources and sinks [dipoles] at intervals of 0.2 mm and 0.58 mm below the surface [abscissa], with point separation of ± 0.17 mm. These simulated the granule cell current generators. The inset dotted curve shows the dipole moment density as a normal density function with $SD = 1$ mm. The solid inset curve is the density under the circular array at the surface passing through the epicenter of the density function. The solid curve in each frame is the potential at the surface along the array. The curve dots in the lower panel show the effect of adding "noise" to the individual channels at the overall indicated signal:noise [S:N] ratio, in order to simulate the EEG data in the upper panel.)

Figure 4. Upper panel: The ensemble average of spatial spectrum by the 1-dimensional (1-D) Fourier transform over 20 control bursts ($\pm$ SE) in the coordinates of cycles/mm and normalized natural logarithm of power. Lower panel: Simulation. (The dots show the M and SE of the spatial spectra of 10 "bursts" with $SD = 1$ mm and signal:noise [S:N] = 4. The four curves show the effect of shifting the array eccentrically from the epicenter of the simulated focus in steps of 0.5 mm [see inset in Figure 5]. The mean level of power over spatial frequencies exceeding 0.6 c/mm defined an asymptote A . The level of A was found to vary monotonically with simulated S:N by the relation

$$S:N = \exp(0.83 - 1.08 A). \quad (6)$$

This equation was used to estimate the S:N of EEG spectra as well [upper panel].)

under the assumptions that (a) the total energy density of the distributed dipole layer is uniform and (b) the local dipole vectors are randomly oriented in all directions, except for a domain of coherence, the size of which is evaluated by a "correlation distance" (equivalent here to the SD measure). The predicted rate of decrease in normalized log_e power with increasing spatial frequency conforms to a Bessel function, that is, near-linear with a slope that increases with increasing correlation distance (dashed lines in Figure 5). Comparison of these predicted curves with the spectra derived from the EEG and from the simulation showed that the latter pair displayed an upward convexity over the low-frequency domain. This discrepancy showed that the assumptions of uniform energy density and varying dipole orientation did not hold in the bulb. The same discrepancy was found in 2-D spectral analysis (discussed below). The falloff of log_e power was predicted by the Katznelson model to be linear with spatial frequency, but the EEG spectrum showed an upward convexity.

2-D experimental spatial spectra. In 2-D spatial analysis, sets of bursts were taken from 14 rabbits under the control conditions described above. Representative averages of 2-D power spectra are shown in Figure 6. Power decreased mon-

Figure 5. Comparison between two models to predict the EEG spatial spectrum. (The dashed curves show the prediction from the Katznelson [1982] model, which assumes uniform energy density in the neural population and a varying distance of coherence of a focus of aligned dipoles within a distribution of dipoles randomly varying in orientation. The solid curves show the prediction from the model that assumes fixed dipole orientation and nonuniform energy distribution, in which the width of a focus is described by the SD of an equivalent 2-dimensional [2-D] Gaussian focus of activity. Owing to the large SE of the EEG data, the 1-dimensional [1-D] array did not support the distinction. The 1-D array did not serve to locate the peak of a focus, nor could the width of a focus be estimated. Its main value was in its small interelectrode distance that enabled the search for activity at high-spatial frequencies.)

otonically from 0.04 c/mm to about 0.5 c/mm. Generally, there was an asymptote between -2 and -7 over spatial frequencies above 0.8 c/mm. When the asymptote was sufficiently low, a peak at 0.85 c/mm was occasionally observed.

Figure 6. Bulbar 2-dimensional (2-D) spatial spectrum of the EEG. (The solid dots show the mean $\pm$ 2 SE for 20 control bursts from 6 rabbits. The open dots show the 2-D FFT of a uniform amplitude distribution the size of the array after Hamming. This defined a limit on spectral analysis with the 8×8 array. The solid curves show spectra predicted for simulated foci having the form of the bivariate normal density function. The curve labeled $W = 0$ is for a spatial delta function, that is, the activity of one granule cell or a focus less than $W = 0.25$ mm. The indicated width of EEG foci is between 1 and 1.5 mm. In these coordinates the falloff in log power with increasing spatial frequency is linear from $P = 0$ by the Katznelson [1982] model. All subjects showed an upward convexity in their spectra.)

In about 1 in 20 bursts, the asymptote was relatively high (between 0 and $-2 \log_e$ power), and in rare instances, it exceeded zero.

Simulated 2-D spatial spectra. Simulations were undertaken as before with bivariate activity density functions at various SD values to generate artificial "bursts." These were "recorded" and processed in the same manner as EEG bursts from the 8×8 arrays with 0.5-mm spacings (see Methods). The rates of falloff in power with representative values for SD in the absence of noise are shown as solid curves in Figure 6. Simulated noise based on random numbers was added as before. This introduced an asymptote at the higher spatial frequencies as described for the 1-dimensional case. There was a monotonic relation between the mean power in the asymptotic domain (from 0.75 c/mm to the maximal estimated frequency at 1.18 c/mm) and the S:N ratio. The small peak near 0.85 c/mm was an artifact dependent on the spacing of the array.

Conclusions. Four conclusions were drawn: (a) there was no significant departure from radial symmetry for the observed EEG foci. (b) there was no evidence for periodicity at any spatial frequency of the EEG. (c) the width of foci expressed as SD of a bivariate normal density function varied among subjects between 1 and 1.5 mm, and (d) the modal frequency value of the inflection point serving to define the upper boundary of the spatial frequency signal passband was estimated to be 0.5 c/mm. Attempts were made to evaluate the focal width and S:N ratios of individual bursts from the slopes and asymptotes of their spectra. These attempts were unsuccessful owing to three factors. First, the number of points available in the spectra were too few to give unbiased slope estimates. Second, the asymptotic value of normalized spectra was sensitive to poorly defined estimates of the gain at 0.04 c/mm value, the lowest frequency available. Third, there was no analytic curve with which to fit individual burst spectra. Estimates of focal width and S:N ratios computed for averaged spectral slopes and asymptotes served as controls for the results of other procedures of measurement.

Design and Application of Spatial Filters Based on Spectral Analysis

An exponential filter (Gonzalez & Wintz, 1977) was generated by the equation

$$A(f) = \log_e[2^{-0.5}](f/f_0)^n, \quad (5)$$

where f_0 represents the cut-off frequency (down 3 dB) and $A(f)$ represents attenuation at each frequency f in c/mm in polar coordinates. The exponent was $n = 4$ for the low-pass filter (see Figure 7) and $n = -2$ for the high-pass filter.

Low-pass spectral domain. The low-pass spatial filter found its first use in more detailed analysis of the spectral domain between 0.1 and 0.5 c/mm. Three spectra were generated from simulated data to serve as standard conditions. One was the spectrum of a uniform rectangle passed through the Hamming window; this gave the maximal observable rate of decrease in spectral energy (see Figure 8). Another was the spectrum of a spatial delta function (Figure 5, $SD = W = 0$)

Figure 7. Rates of attenuation by a low-pass spatial filter (Equation 5, $n = 4$) for 4 values of the cut-off frequency (at power down 3 dB).

for which the potential defined the point spread function of a single granule cell or a cluster less than 0.25 mm in diameter. A third standard spectrum was that of a simulated granule cell focus with SD radius of 1 mm, for which the potential approximated the modal width of EEG foci in the bulb (Figure 8).

In noise-free simulations, these spectra fell to very low values with increasing spatial frequency; for example, the rate of falloff for the 0 mm focus was 10 fold for each 0.5 c/mm increment in frequency. A useful technique for observing the spectral domain of interest was to subtract the Hamming window spectrum from the EEG spectrum (Figure 8). This was feasible only if the EEG spectrum was filtered beforehand with the low-pass filter ($f_0 = 0.5$ c/mm).

Examples are shown in Figure 9 (Panel A) of the mean spectral differences (Figure 8) over 20 "bursts" from simulated foci of selected radii; the inset shows the density plot of the measured amplitudes for the focus of width $\sigma = 1$ mm (SD). Panels B-F show mean spectral differences (Figure 8) over 20 EEG bursts from 5 rabbits. The points show the means and standard errors of the spectral differences in $\log_e$ power. The dark segmented solid curves show the simulated spectra of the sums of two components in the spatial domain: one focus with radius (SD) $\sigma_1 = 0$ mm and a second focus with σ_2 as indicated within each frame. The light segmented curves are from Panel A. The insets show the density plots of mean rms amplitudes (centroids). The locations of the two epicenters were of no significance, provided they were both within the frame; the amplitude proportions of the broader components V_2 to the narrow components $V_1 = 0$ mm were determined by trial and error.

The results showed that each spectrum could be simulated in detail by these two components. The interpretation was that the spectrum was determined in part by the width of a "smoothed" focus and in part by irregularities in the shape of the focus, but that after low-pass spatial filtering, the entire spectrum could be accounted for by the potentials generated by the granule cells, given their known spectral properties.

Fitting of surfaces to amplitude focus. Low-pass filtering also made it possible to fit surfaces to the amplitude and

Figure 8. Two standard simulated spectra and their differences, spatially filtered (D) and unfiltered (S-H). (Most of the behaviorally relevant information in the EEG was below 0.5 c/mm, but it was difficult to visualize in logarithmic coordinates. It was made easier to observe by subtracting the log_e power H of the uniform square transformed under the Hamming window from the log_e power of the data. In the example here, the data were from a simulated focus 1 mm in SD radius. No filtering was needed in the noise-free case, S-H, but for work with noisy cases and with the EEG, a low-pass filter, F, was necessary to observe the differences, D.)

phase patterns of the 64 curves fitted to the EEG. Without such filtering, the nonlinear regression failed to converge in 30% or more of bursts. A bivariate normal density function was fitted to the amplitude of 60 bursts from each of 5 rabbits as an approximation to the bell-shaped distribution of potential in the foci. The fitted curve incorporated on the average over 80% of the variance in amplitude (see Table 1). The geometric mean of the half-amplitude radii in the x and y dimensions provided an estimate for the half-amplitude radius x_h of the focus for each rabbit. This was used to estimate the SD radius of a granule cell focus, that is, an equivalent normal distribution of activity density under the bulbar surface, by an equation previously derived (Freeman, 1975) and reconfirmed in this study,

$$SD = x_h - z_o/2, \quad (7)$$

where $z_o = 0.58$ mm was determined by depth recording

(Freeman, 1975) and by postmortem measurement of the depth of the mitral cell layer in this and previous studies (Freeman, 1978, 1980; Freeman & Viana Di Prisco, 1986a). Three rabbits with single-peaked foci gave estimates near 1 mm; 2 rabbits with bimodal foci gave estimates for SD near 1.5 mm.

Fitting of surfaces to spatial phase pattern. After low-pass filtering, the phase data from the dominant component of individual bursts were fitted with a plane, which incorporated on the average about 55% of the variance. A closer fit (65% of the variance) was obtained by fitting phase data in accordance with an approximation of the bulb by a sphere of radius 2.5 mm (see Table 2). By fitting a cone in spherical coordinates, phase gradients took the form of concentric spread. Further, the sites of phase convergence and divergence were localized. The mean phase gradient (radians/mm) at each burst peak frequency in radians/s was used to estimate velocity (m/s), giving an average velocity of 1.72 ± 0.39 m/s. The apex of the cone usually lay outside of the electrode array window; the angles and distances of locations were plotted in a radial projection centered in the array (see Figure 10), with sites of divergence (phase "source") noted by solid dots and convergence (phase "sink") noted by open dots. About half of the residual variance of phase (Table 2) was accounted for by errors of measurement at the S:N levels derived from these bursts (Freeman & Viana Di Prisco, 1986b, Figure 5).

Locations of phase extrema. Plots of the locations of phase extrema were examined from each subject for groups of orderly control bursts (C), CS+ and CS- odor bursts, and disorderly bursts. The locations by projection from the array were broadly scattered over the bulb, with some tendency for increased density in the middle third (from anterior to posterior) but with no predilection for or against the area covered by the array. The locations were seldom extrapolated posteriorly to the array and thereby to areas outside of the bulb. No large part of the bulb appeared to be excluded. The maxima and minima occurred in roughly equal numbers in all subjects and groups of bursts. Each extremum had its opposite at its antipodal point, which suggests an ambiguity in description; this was resolved for those extrema located anterior to the array, because their antipodes lay posterior to the bulb. If the criterion was accepted that a valid extremum must lie within the bulb, then for an extremum in the anterior bulb, and by extrapolation in the entire bulb, the likelihood of its being a minimum or a maximum was about equal. Finally, the locations of the phase extrema bore no relation to the locations of the amplitude maxima of the signature patterns for any subject or group of bursts or to the control and odor conditions.

Fitting of cone to phase pattern of second frequency component. The cone was also fitted to the phase matrices of the secondary component of the bursts, thereby incorporating 53% of its variance on the average. The temporal frequencies of the two components differed on the average by 29 Hz with a range from 12 to 68 Hz, but the phase extrema were found to be located near each other. The signs (lead or lag) agreed in 95.3% of bursts; the correlation coefficient of the pairs of x and y coordinate values averaged 0.79; and the mean distance between dominant and secondary extrema averaged 0.80 ± 0.22 mm. When the two measurements were treated

Figure 9. A: A family of curves showing the differences in log. power between the spectrum from each of eight simulated EEG foci with the indicated SD radius and the spectrum of a square the size of the 8×8 array under Hamming. (The inset shows a density plot of the root mean squares [rms] amplitude for $SD = 1$ mm and signal:noise [S:N] = 1.0. B-F: The dots show the mean ($\pm SE$) of the differences between the spectra of the EEG ($N = 20$) and the square. The insets show the density plots of the rms ensemble averages. The lower curve in each panel shows the selected curve from A that best fitted the spectral difference between 0.04 and 0.25 c/mm. The upper curve in each panel shows the spectral difference from the two foci; one at the indicated SD and the other at $SD = 0$ mm. The relative energy V_1 and V_2 in the two superimposed foci was determined by trial and error. The results showed that the EEG spectra could be explained as the activity of granule cells acting in concert in irregularly shaped foci with radii up to 3 mm plus activity at higher spatial frequencies from other sources, here removed by low-pass filtering. All of the spectra were low-pass filtered at 0.5 c/mm.)

as two experimental estimates of the location of a single site, the standard error was 0.28 mm, which was half the mean interelectrode distance of the arrays. The estimated mean velocity of the secondary components, 1.87 ± 0.43 m/s, agreed with that of the dominant components. The same results held for both orderly and disorderly bursts. These findings further supported the concept of the burst as a common mode of oscillation with a spatially phase-locked waveform irrespective of the high or low degree of concentration of its energy into narrow temporal spectral bands.

Optimization of Filters By Their Improvement of the Correlation of EEG Patterns With Behavior

Introduction. A "behavioral assay" as described in the preceding report (Freeman & Viana Di Prisco, 1986a, 1986b, Figures 12 & 13) was used to find optimal parameters and

procedures for low- and high-pass filtering, spatial deconvolution, and normalization of data. After optimization of the procedure, confidence intervals for the assay were derived by a data resampling procedure. The efficacy of classification with subsets of channels either randomly or systematically selected was measured with the behavioral assay. The results to be described here showed that all channels were equally important within the SE for the classification of odor bursts.

Behavioral assay. The behavioral assay measured the efficacy of data derived from EEGs to classify bursts correctly with respect to odor stimuli: the difference in the percentage of correct classification of CS+ and CS- odor bursts minus the percentage of correct classification of the two groups of control bursts, C+, C-. The assay was based on the premiss that control bursts from randomly interspersed CS+ and CS- trials should not differ. On the basis of data screening with this assay, bursts were selected by four criteria for analysis.

Table 1
Data on Spatial Amplitude Variation

Subject	Location		Width		Radius of focus	Variance %
	<i>x</i>	<i>y</i>	<i>SD_x</i>	<i>SD_y</i>		
1						
C	1.40 ± .56	-.51 ± .22	1.67 ± .48	1.01 ± .12	1.22 ± .22	77.6
CS+	1.33 ± 1.07	-.58 ± .14	1.62 ± .57	.98 ± .12	1.18 ± .32	74.2
2						
C	1.33 ± .05	-1.07 ± .13	1.02 ± .07	1.12 ± .03	.97 ± .05	97.3
CS+	1.23 ± .91	-1.08 ± .26	1.32 ± .76	1.08 ± .14	1.08 ± .42	92.8
3						
C	.50 ± .28	-.27 ± .25	1.48 ± .24	1.46 ± .13	1.43 ± .10	86.2
CS+	.50 ± .35	-.33 ± .42	1.49 ± .45	1.46 ± .18	1.42 ± .21	86.5
4						
C	1.25 ± .09	1.05 ± .22	1.10 ± .24	1.17 ± .18	1.04 ± .22	78.6
CS+	1.20 ± .28	.93 ± .26	1.11 ± .22	1.16 ± .14	1.04 ± .20	73.9
<i>M</i>			1.35 ± .25	1.18 ± .19	1.17 ± .18	83.4

Note. *M* and *SEs* for 4 rabbits for the locations (in mm) in the *x* (horizontal) and *y* (vertical) directions of the array; *SDs* of the fitted bivariate Gaussian surface; mean radius of the active granule cell population (also expressed by a bivariate but symmetric Gaussian distribution under the surface) from Equation 7; and percentage of the total variance of the data that was incorporated by the fitted surface. The data were the 64 amplitude values of the dominant component after low-pass spatial filtering. *N* = 30 for each set. The 4 rabbits showed behavioral evidence for odor discrimination. Data from control (C), CS+, and CS- (not shown) subjects did not show significant differences in these measurements.

(a) Data were omitted from 1 of the 5 rabbits that failed to show behavioral evidence of discrimination. (b) Only those trials were included on which a correct response occurred to the CS+ (licking on 74% of trials) or to the CS- (sniffing on 42% of trials). (c) Bursts with peak frequencies less than 55 Hz (approximately 30%) were omitted (Freeman & Viana Di Prisco, 1986a). (d) Only the last two bursts in each trial (T2 and T3) were included, along with the first control bursts (C1). The latter met the frequency condition about 2.5 times more often than did the test bursts; optimal separation of odor bursts occurred with approximately equal numbers of control and odor bursts. This selection process reduced the number of usable bursts to about one fifth of the 1,800 originally recorded in the three session under analysis.

Low-pass filter optimization. The optimal low-pass filter parameter gave maximal separation of normalized (see below)

CS+ from CS- bursts at a cut-off frequency of 0.5 c/mm. The results are shown in Figure 11 (right-hand curve) for 4 rabbits pooled. With the low-pass filter fixed at a cut-off of 0.5 c/mm, a range of high-pass filter values was explored. Minimal separation occurred at a cut-off frequency of 0.17 c/mm. This value was in the passband for the focus of granule cell activity seen in density plots of burst rms amplitude but, more important, at or near the minimal boundary of the array passband.

Optimization of spatial deconvolution. Spatial deconvolution compensated for distortion by volume conduction of granule cell activity patterns recorded at the surface of the bulb (Freeman, 1980). An inverse of the procedure whereby the surface potential was calculated in the model was applied at a specified value of generator depth below the surface, 0.58 in the model. The optimal value for the depth parameter, as

Table 2
Data on Spatial Phase Variation

Subject	Location		Number		Velocity m/s	SD of phase		Variance %
	<i>x</i>	<i>y</i>	Source	Sink		Total	Residuals	
1								
C	3.19 ± 4.70	-.04 ± 4.27	17	27	1.83 ± .30	.397	.249	55.9
CS+	3.34 ± 5.57	-1.23 ± 2.78	10	5	2.14 ± .44	.325	.232	54.7
2								
C	2.68 ± 2.09	.84 ± 2.01	25	32	1.54 ± .33	.446	.190	71.4
CS+	2.85 ± 2.90	1.91 ± 5.96	10	9	1.46 ± .38	.548	.371	53.1
3								
C	1.47 ± 1.49	2.98 ± 3.14	22	42	2.36 ± .41	.278	.141	74.3
CS+	1.63 ± 1.69	2.42 ± 2.85	12	12	1.93 ± .43	.363	.235	64.3
4								
C	2.98 ± 2.23	-.37 ± 2.57	27	37	1.41 ± .21	.491	.286	65.9
CS+	2.96 ± 1.91	-.58 ± 2.11	8	12	1.55 ± .36	.512	.269	73.9
<i>M</i>			16	22	1.72 ± .39	.420	.247	64.6

Note. Measurement of the locations (in mm) from the center of the array in its *x* and *y* coordinates of the maxima (sources) and minima (sink), the velocity, the average *SDs* of the phase values before curve-filtering and of residuals, and the percentage of variance of the phase incorporated by a fitted surface. The data were orderly control (C) and CS+ bursts from 4 behaviorally discriminating rabbits. The fitted surface was a cone in spherical coordinates. The *SDs* of the residuals were about twice those predicted from errors of measurement (Freeman & Viana Di Prisco, 1986b, Figure 5).

Figure 10. Upper panel: Example of the locations of phase maxima (solid dots) and minima (open dots) projected into the flattened surface of the bulb of a rabbit. (The 64 phase values from each burst [after low-pass filtering of the real and imaginary parts from the amplitude and phase values of the dominant component] were fitted with a cone in spherical coordinates. The sketch is a projection of the outline of the bulb as it would appear on looking through the bulb onto the surface of the array. A representative set of isophase contours is shown at 0.25 radians/mm. The locations of the apices of the cone are shown in polar coordinates as angle and distance from one pole of the sphere (2.5 mm in radius) at the center of the array to the antipodes, which takes the form of the large circle in this display. The square is the outline of the 8×8 array. Many of the phase values could be fitted as well with a plane, the direction of which varied from burst to burst, which indicated that the degree of confidence concerning the location of the conic apex diminished with increasing distance from the array. The standard error of each point was estimated to be twice the radius of its plotting symbol. The apex was rarely extrapolated to locations outside of the bulb; those values were interpreted as replaceable by their antipodes.) Lower panel: Locations of amplitude maxima and one minimum. (The conic surface was also fitted to the 64 amplitude values. The peak of the focus was often

Figure 11. Results of spatial filtering as measured by the behavioral assay. (The behavioral assay was applied to sets of amplitude values of the dominant components of bursts from each of 4 rabbits after low-pass filtering at the values indicated on the abscissa. Optimal classification was obtained with the cut-off at 0.5 c/mm [right curve]. The procedure was repeated after fixing the low-pass filter but applying the high-pass filter. Minimal classification was found with this filter at 0.17 c/mm. The order $n = 4$ of the filter was set in Equation 5.)

measured by the assay, ranged between 0.49 and 0.54 mm for the 4 rabbits (see Figure 12). Postmortem measurement of the depth of the mitral cell layer under the array in frozen sections of the bulb gave averaged values ranging from 0.67 to 0.89 mm across subjects. There was no correlation across animals between anatomical depth and optimal depth parameter. Figure 13 shows the mean and standard error of the

Figure 12. The optimal focal depth as evaluated by repeating the behavioral assay on each subject after spatial deconvolution at the indicated depths of focus. (The resulting curve is pooled over the 4 subjects showing behavioral evidence for odor discrimination.)

extrapolated to a location outside of the array, but the distribution of the apices was relatively narrow, which demonstrates that the phase and amplitude patterns were independent of each other.)

Figure 13. The average spatial spectrum ($\pm SE$) computed after spatial deconvolution at a focal depth of 0.49 mm of the amplitudes of the dominant component (ordinate scale at right). (The behavioral assay was applied to the data as in Figure 11, again showing that the odor-specific information in the EEG was concentrated in the domain from 0.1 to 0.5 c/mm.)

spatial spectrum for the 4 rabbits after spatial deconvolution. The bar graphs show the effects on the assay when either a low- or high-pass filter was applied to the data following spatial deconvolution.

The effects of deconvolution were assessed on the distribution of the phase values summarized in Figure 14. As expected, the distribution *SDs* increased under deconvolution. The distributions of phase values within the inner 6×6 array did not differ significantly from those in the outer electrodes at the edge of the array, which revealed a lack of edge effects. The plane and conic surfaces fitted to each burst after deconvolution showed that the phase gradient was largely removed from the data by deconvolution.

Normalization and derivation of confidence intervals. Normalization was used after data selection, filtering, and deconvolution. Four procedures were available to standardize the data to zero mean and unit *SD*: (a) by the entire set, (b) by individual bursts, (c) by channel, or (d) by both b and c. Optimal results as measured by the assay occurred with channel normalization. The values for each channel on all bursts of the four groups (CS+, CS-, C+, C-) for each rabbit were transformed to *z* scores by subtracting the channel mean and dividing by channel *SD*. The operation removed the signature pattern from the data for each rabbit.

Confidence intervals for the behavioral assay were derived with a jackknifing procedure. For each of 40 runs per subject, single bursts were removed and an assay value calculated. The *SDs* of the assay values ranged from 2.93% to 5.12% over the 4 subjects, $M = 4.90\%$. The pooled values ($N = 157$) were normally distributed. Values of the assay exceeding 10% for a given subject and condition were inferred to be significantly above zero percentage.

Tests for spatial localization of odor information. With 64 electrodes, it was possible that some subset of channels could classify bursts as well as or better than all 64 together. That

Figure 14. The distributions of phase values before and after spatial deconvolution and, for the latter, for the inner 6×6 matrix of channels in the 8×8 array.

is, for each odor, there might be a subset of bulbar neurons that were maximally activated or suppressed by either the control (background) odors or the two test odors. The remaining channels might have obscured classification by contributing uncorrelated activity, that is, noise. Three procedures were used to test this hypothesis.

Systematic deletion of 8, 16, ..., 56 channels by several criteria showed that the correct classification assay decreased with deleted channels. Figure 15 shows the results of deleting channels having the lowest amplitude prior to normalization. Comparable results were found with deletion of channels with higher amplitudes, with high *SDs*, and with most or least average change in amplitude in transition from control to odor state. On average, the subjects' goodness-of-burst classification decreased with channels deleted.

Random subsets of channels were deleted in groups of 8, 16, ..., 56 by selection with a random number generator. The procedure was repeated 40 times for the data set of each subject. There was a decrease in success of classification with channel deletion as shown in Figure 16. The large standard errors for each point reflected the sensitivity of the assay to the choice of channels.

The third test was made by summing the assay value on each run from the above in a tally for each channel, whenever it was randomly selected over 40 runs at each of seven selection combinations (56, 48, ..., 8). If a given channel was particularly important, then whenever it was selected it would contribute a large value to that tally. Concurrently in

Figure 15. The effect on the behavioral assay of deleting amplitude values from channels in sets of eight beginning with those having the lowest amplitudes prior to normalization. (The results were pooled over 4 subjects; the SE [upper and lower lines] was derived by jackknifing. Similar results occurred by other criteria, including highest amplitude, lowest or highest channel variance, and greater change from control to odor states.)

each run, the average assay value was tallied. The 64 differences between the two sets of 64 numbers was divided by the averaged assay value for each subject when no channels were deleted. For all subjects, the grand mean difference was 0.057;

Figure 16. Demonstration with the behavioral assay that the odor-specific information was broadly distributed among all channels of the array. (Channels were selected for deletion by a random number generator on 40 trials at each number of deletions for each rabbit. The M and SE are shown for each set. This procedure provided the basis for a search for individual channels that consistently contributed to high- or low-classification values; no preeminent channels were found.)

subject means ranged from -0.019 to 0.115 . The 64 normalized differences were normally distributed with SD s ranging from 0.009 to 0.035 . In other words, the grand mean percentage difference for the behavioral assay over the 7×40 runs for each subject, averaging 32 channels/run, was 10.6%; the average channel difference from that mean was 0.60%; and the 99% upper confidence limit was 1.26%. There was no evidence for any secondary mode or tail that would indicate the presence of channels with unusually high or low effects on classification. By this test, all channels were equally important to within a few percent for the classification of odor bursts.

Discussion

The basic hypothesis of this study is that between the times of onset of a discriminative CS and a correct CR, an odor-specific space-time pattern of neural activity must exist somewhere in the bulb as basis for the response. The accepted view since the work of Adrian (1950) has been that a subset of mitral cells receiving input from selectively sensitive receptors is activated by each odor and that the mitral cell subsets differ between odors either in their locations, their temporal firing patterns, or both. Direct tests of this view have been unsuccessful, because it is not technically feasible to record simultaneously from sufficiently large numbers of mitral cells. An alternative is to measure the spatial patterns of the dendritic potential fields of the granule cell interneuronal activity that, owing to feedback connections, is closely correlated temporally and spatially with the activity of mitral cells and to search for EEG activity patterns that relate to the CS odors.

The nature of those patterns being unknown at the outset of this study, the procedure was to measure the EEG patterns in space and time with as much precision as the data allowed, and then to partition the results of measurement by selection and by filtering. Each fraction from partitioning was evaluated with respect to its power correctly to classify control, CS⁺, and CS⁻ bursts, in the manner of testing the potency of a compound undergoing chemical purification. A crucial step in this process was the spatial deconvolution of the EEG patterns in order to correct for distortion of the granule cell activity patterns by volume conduction. Each local subset of granule cells contributed broadly to an array of electrodes at the surface so that activity patterns became blurred and indistinct in the EEG. Yet, deconvolution could not be used in the presence of high spatial frequency activity, because that component was amplified by the transform (Freeman, 1980). In turn, the design of a low-pass spatial filter required an understanding of the spatial spectral properties of the point spread function of the granule cells and of their temporal passband into the EEG. In brief, the proper use of the bulbar EEG for testing the data against behavior required spatial and temporal filters that retained granule cell contributions and rejected extraneous activity.

It was also apparent that at least in the area of the array on the bulb, in about 20% of control bursts and 50% of test bursts the bulbar mechanism failed to converge to a stable and reproducible spatiotemporal pattern. These disorderly events occurred at relatively low temporal frequencies. At the

present level of understanding, they were simply identified and removed from the sample as outliers.

The results at this stage of analysis can be summarized as follows. Control, CS+, and CS- bursts can be correctly classified in numbers significantly exceeding chance levels on the basis of the 64 amplitude values of the dominant oscillatory component of the bursts. Overall burst amplitude plays a role in separating control from odor bursts but not in separating CS+ from CS- bursts. The frequency and phase properties play no role, nor do the amplitude properties that determine the characteristic signature pattern for each animal. The peak locations and average widths of foci fluctuate erratically but not in relation to odor condition.

Anatomical Basis of Oscillation

These spatial and temporal properties of bulbar activity that are revealed by the EEG give some clues to the dynamics of the bulb. The basic oscillation in the gamma range from 35 to 90 Hz is generated by negative feedback between the mitral and granule cells. The dense synaptic coupling of mitral apical dendrites from receptor axons and with each other can be expected to lead to some degree of coordination among the set of 75 to 100 mitral (and tufted) cells having apical tufts within each glomerulus. They and the 20,000 to 30,000 granule cells with which they form synaptic negative feedback loops can be regarded as a neural oscillator.

There are roughly 2,000 glomeruli in each bulb of the rabbit. The instantaneous frequency of oscillation is the same throughout the bulb at all times. The receptor input cannot drive the bulb with this high degree of synchrony; there is no known mechanism for or evidence of the requisite coordination among receptor cells. The activity is not driven by centrifugal input, because it survives transection of the bulbar stalk. The coordination arises within the bulb by coupling between oscillators. Either of three anatomical features may help to explain for this. One is formed by the basal dendrites of the mitral and tufted cells, which radiate from the cell bodies to distances exceeding 1 mm in directions parallel to the surface but with no preferential orientation in the external plexiform layer. The second is the set of stellate cells of Golgi, Cajal, and Blanes, whose axons ramify broadly through the external plexiform and granule cell layers. Although their sign of action is unknown, they may provide the link of mutual inhibition among granule cells that is postulated to exist on theoretical grounds (Freeman, 1975). The third is formed by axon collaterals of the mitral and tufted cells found mainly in the internal plexiform layer, which extend at least as far as the basal dendrites do (White, 1965) and that couple mitral and tufted cells by mutual excitation. These are thin myelinated axons; their conduction velocity has not been measured directly. It is known that the conduction velocity of the fine terminals of the mitral axons in the prepyriform cortex is between 1.5 and 2 m/s (Freeman, 1975). The mean estimate of velocity (1.72 m/s) derived from measurements of EEG phase gradients suggests that the degree of synchronization of oscillatory bulbar activity might be limited by the conduction velocity of these axon collaterals. A difficulty with this interpretation is that the axon collaterals from any one neuron do

not extend over the entire bulb, and successive synaptic delays are not accounted for. That is, the phase velocity should be less than the axonal conduction velocity. The conic form of the phase gradient might suggest that a "pacemaker" exists at the apex analogously to an ectopic focus in the myocardium. This seems implausible because the location varies seemingly at random, it rarely lies near the site of maximal amplitude of oscillation, and it may be either a phase maximum or phase minimum. Further studies are needed, particularly simulations with solutions of differential equations that incorporate the requisite time delays (Martinez & Freeman, 1984).

Significance of High-Amplitude Focus of Activity

Focal, high-amplitude EEG activity and intense unit activity are maintained in the midlateral and ventromedial regions of the olfactory bulb of waking rabbits and cats with and without odorant stimulation (Freeman, 1975). The lateral focus has been shown to undergo subtle changes in shape under conditioning with odorants (Freeman & Schneider, 1982). High-metabolic activity in these regions has been revealed by 2-deoxyglucose studies (e.g., Lancet et al., 1982). Together these results suggest that the foci of high-amplitude EEG activity are specialized locations of intense bulbar activity but are not crucial to the machinery for odor discrimination. The present findings that argue against the significance of the high-amplitude focus in odor discrimination are (a) the commonality of waveform over the entire array and indeed throughout the bulb (Bressler, 1984), (b) the phase gradient, (c) the optimal classification obtained after channel normalization which removes the signature pattern, and (d) the optimal classification using all available data without regard to prenormalization amplitude. The genesis of these high-amplitude foci appears to have an anatomical basis. Meisami and Emamian (1985) have shown that the midlateral and ventromedial regions are characterized by smaller glomeruli and higher packing densities in the glomerular, mitral, and granular layers. In analogy with the retina, these regions may constitute "olfactory foveas." Whether and how these features might lead to high-metabolic activity and to EEG foci are questions that remain to be answered.

Distributed Character of Information Processing in the Bulb

The most important result is the finding that the odor-related information in the EEG cannot be localized to any subset of channels. On the one hand, deletion of channels chosen either at random or by several criteria decreases the power of the remaining channel data to classify bursts correctly. On the other hand, in 2 rabbits, subsets of as few as 16 channels randomly selected have the power to classify bursts correctly at better than chance levels, which suggests that this information is broadly distributed over the bulb.

The tests by deletion are based on the premiss that the informational contribution of each channel is independent of the others. The likelihood remains that some weighted combination of channels might prove to have classification power

superior to that of all 64 channels, it if could be found. Its existence and significance appear unlikely for the following reason. The information is contained in the low-spectral passband of 0.15 to 0.25 c/mm, corresponding to the typical form of a bulbar EEG focus, even after spatial deconvolution. This implies that an effective combination of channels would have to be formed from a contiguous subset in the array and that the underlying activity of mitral subsets would have to be coordinated within the bulb in order to give rise to a reproducible pattern. Coordination must either be imposed from outside the bulb or it must arise from within the bulb by cooperative interaction. The burst oscillation is a property of the bulb, not of its sensory or centrifugal input. Cooperativity must be either local (for local mitral subsets to combine separately from their surrounds) or global. The commonality of EEG waveform implies the latter.

The results from channel normalization and the low-spatial passband suggest that the granule cell activity patterns that contain odor-specific information extend well beyond the limits of the array window of observation. The phase gradient suggests that they involve the entire main bulb. Electroencephalograph records from arrays of electrodes implanted within the bulb (Bressler, 1984) and designed to sample activity from the greater part of it show that the commonality of temporal waveform already found for the 4×4 mm surface array (Freeman & Viana Di Prisco, 1986a, 1986b) appears to hold for the whole bulb. This interpretation is consistent with the cytoarchitecture of the bulb: broadly ramifying axons and dendrites in the internal and external plexiform layers interconnecting all of the neurons in a neuropil bounded only by the edges of the main bulb. It is also consistent with the nonlinear dynamics of the bulb leading to the emergence of cooperative activity through large-scale interaction of large numbers of neurons. It conforms with the properties of the outflow path of the bulb: the lateral olfactory tract has little or no topographic order in its projection onto the olfactory cortex, so that each neuron in the cortex that receives tract input must carry out a spatiotemporal integration over a large region of the bulb.

It remains likely that odor-specific receptor input is delivered by primary nerve axons to limited subsets of mitral cells. In the absence of learning by an animal to identify a particular odor, there is no evidence to support the outcome that bulbar activity might be globally convergent. However, during learning to identify an odor, it appears that a nerve cell assembly is formed by strengthened connections among mitral cells that are coactivated by the odor under reinforcement (Freeman, 1979, 1983; Gray, Freeman, & Skinner, 1986). Thereafter, the arrival of the odor may lead to formation of a stereotypic spatial pattern over the entire bulb as a necessary albeit insufficient condition for correct response to that odor. On this interpretation, the bulbar output is not localized but is truly global; every neuron participates in every learned odor response but in differing degrees for different odors (Freeman & Viana Di Prisco, 1986a).

If this hypothesis is correct, then the spatial pattern for each CS+ and CS- should be invariant over sessions in which the response contingencies are stable. This aspect is taken up in a following report (Freeman & Grajski, 1987) on factor analysis of the data, wherein a more extensive statistical analysis is

reported to validate the results given here based on the classification assay (Freeman & Viana Di Prisco, 1986a).

References

- Adrian, E. D. (1950). The electrical activity of the mammalian olfactory bulb. *Electroencephalography and Clinical Neurophysiology*, 2, 377-388.
- Bressler, S. L. (1984). Spatial organization of EEGs from olfactory bulb and cortex. *Electroencephalography and Clinical Neurophysiology*, 57, 270-276.
- Childers, D. G. (1978). *Modern spectrum analysis*. New York: IEEE Press.
- Digital Signal Processing Committee (Eds.). (1979). *Programs for digital signal processing*. New York: IEEE Press.
- Eastman, C. (1975). Construction of miniature electrode arrays for recording cortical surface potentials. *Journal of Electrophysiological Techniques*, 5, 28-30.
- Freeman, W. J. (1975). *Mass action in the nervous system*. New York: Academic Press.
- Freeman, W. J. (1978). Spatial frequency analysis of an EEG event in the olfactory bulb. In D. A. Otto (Ed.), *Multidisciplinary perspectives in event-related brain potential research* (pp. 531-546). Washington, DC: U.S. Government Printing Office.
- Freeman, W. J. (1979). EEG analysis gives model of neuronal template-matching mechanism for sensory search with olfactory bulb. *Biological Cybernetics*, 35, 221-234.
- Freeman, W. J. (1980). Use of spatial deconvolution to compensate for distortion of EEG by volume conduction. *IEEE Transactions on Biomedical Engineering*, 27, 421-429.
- Freeman, W. J. (1987). Analytic techniques used in the search for the physiological basis of the EEG. In A. Gevins & A. Remond (Eds.), *Handbook of electroencephalography and clinical neurophysiology* (Vol. 3A, Pt. 2, chap. 18). Amsterdam: Elsevier.
- Freeman, W. J., & Grajski, K. A. (1987). Relation of olfactory EEG to behavior: Factor analysis. *Behavioral Neuroscience*, 101.
- Freeman, W. J., & Schneider, W. S. (1982). Changes in spatial patterns of rabbit olfactory EEG with conditioning to odors. *Psychophysiology*, 19, 44-56.
- Freeman, W. J., & Viana Di Prisco, G. (1986a). EEG spatial pattern differences with discriminated odors manifest chaotic and limit cycle attractors in olfactory bulb of rabbits. In G. Palm & A. Aertsen (Eds.), *Brain theory* (pp. 97-119). Berlin: Springer-Verlag.
- Freeman, W. J., & Viana Di Prisco, G. (1986b). Relation of olfactory EEG to behavior: Time series analysis. *Behavioral Neuroscience*, 100, 753-763.
- Golub, G. H., & Pereyra, V. (1973). The differentiation of pseudo-inverses and nonlinear least squares problems whose variables separate. *SIAM Journal on Numerical Analysis*, 10, 413-432.
- Gonzalez, R. C., & Wintz, P. (1977). *Digital image processing*. Reading, MA: Addison-Wesley.
- Gray, C. M., Freeman, W. J., & Skinner, J. E. (1986). Chemical dependencies of learning in the rabbit olfactory bulb: Acquisition of the transient spatial pattern change depends on norepinephrine. *Behavioral Neuroscience*, 100, 585-596.
- Katznelson, R. D. (1982). *Deterministic and stochastic field theoretic models in the neurophysics of the EEG*. Unpublished doctoral dissertation, University of California, San Diego.
- Lancet, D., Greer, C. A., Kauer, J. S., & Shepherd, G. M. (1982). Mapping of odor-related neuronal activity in the olfactory bulb by high-resolution 2-deoxyglucose autoradiography. *Proceedings of the National Academy of Science (US)*, 79, 670-674.
- Martinez, D. M., & Freeman, W. J. (1984). Periglomerular cell action on mitral cells in olfactory bulb as shown by current source density analysis. *Brain Research*, 308, 223-233.
- Meisami, E., & Emamian, S. (1985). Cytoarchitectural diversity in

- the main olfactory bulb: Are there olfactory foveas? *American Chemosensory Society VII (Abstract)*.
- Nicoll, R. A. (1971). Recurrent excitation of secondary olfactory neurons: A possible mechanism for signal amplification. *Science*, 171, 824-825.
- Nunez, P. L. (1981). *Electric fields of the brain*. New York: Oxford University Press.
- Rall, W., & Shepherd, G. M. (1968). Theoretical reconstruction of field potentials and dendrodendritic synaptic interactions in olfactory bulb. *Journal of Neurophysiology*, 31, 884-915.
- Sammon, J. W. (1969). A nonlinear mapping for data structure analysis. *IEEE Transactions on Computers*, 18, 401-409.
- Viana Di Prisco, G., & Freeman, W. J. (1985). Odor-related bulbar EEG spatial pattern analysis during appetitive conditioning in rabbits. *Behavioral Neuroscience*, 99, 964-978.
- White, L. E., Jr. (1965). Olfactory bulb projections of the rat. *Anatomical Record*, 152, 465-480.

Received October 6, 1985

Revision received May 20, 1986 ■

Statement of APA Posting Conditions

This article may not exactly replicate the final version published in the APA journal. It is not the copy of record.

<http://content.apa.org/pubmed/bne/101/3/393>