PERGAMON

Vision Research 39 (1999) 2485–2497

Vision Research

Synchronous oscillations in the cat retina

Sergio Neuenschwander *, Miguel Castelo-Branco, Wolf Singer

Max-Planck-Institut für Hirnforschung, Deutschordenstraße 46, 60528-Frankfurt am Main, Germany

Received 10 August 1998; received in revised form 1 February 1999

Abstract

Retinal ganglion cells exhibit oscillatory responses which are precisely synchronized over large distances. Here we examined, with multi-electrode recordings, the time course of synchronization during spontaneous and stimulus-driven oscillatory activity. Spontaneous discharges showed synchronized oscillations at ~ 30 Hz, which were occasionally associated with slower superimposed oscillations in the range of 1–5 Hz. Stationary stimuli or moving gratings induced synchronous oscillations at higher frequencies (mean of 79.0 ± 20.0 Hz for OFF- and 91.7 ± 11.7 Hz for ON-responses), with time lags of a few milliseconds. At response onset, the first few oscillatory cycles were occasionally time locked to the stimulus. Thereafter, synchronization became independent of stimulus coordination and was exclusively due to neuronal interactions. Oscillatory modulation emerged rapidly and was sustained throughout the responses while oscillation frequency decreased gradually. This periodic patterning of responses persisted despite brief and local occlusion of stimuli, suggesting that synchronous oscillations emerge from population dynamics and entrain cells even if they are intermittently silenced.

Keywords: Correlation oscillatory synchronization; Temporal code retina

1. Introduction

Since the discovery of stimulus induced synchronization of visual responses, increasing attention has been given to the time structure of neuronal activity. Synchronization of responses with a precision in the millisecond range has been observed at different levels of processing, in various sensory modalities and in a variety of species (for a review see Singer, Engel, Kreiter, Munk, Neuenschwander & Roefselma, 1997). In the visual system, synchronization of cortical responses reflects global stimulus properties, supporting the hypothesis that correlated firing serves to establish relationships among distributed responses and plays a role in the binding of local features.

In the retina, precise oscillatory patterns in the spiking activity of ganglion cells have been discovered both in vertebrates and invertebrates already in the early functional investigations (Fröhlich, 1914; Adrian & Matthews, 1928; Granit, 1933). Later, several studies have described oscillatory activity in the retina of cats and monkeys in response to sudden luminance changes (Kuffler 1953; Doty & Kimura, 1963; Laufer & Verzeano, 1967; Rodieck, 1967; Ariel, Daw & Rader, 1983). These oscillations covered a broad range of frequencies, from 3 to 150 Hz, and occurred with and without anesthesia. It has been shown that retinal oscillations depend on stimulus size and contrast, pointing to the involvement of center-surround mechanisms (Ariel et al., 1983; Przybyszewski, Lankheet & van de Grind, 1993). The oscillatory patterning of activity is not restricted to ganglion cells but also present in the graded responses of horizontal and amacrine cells (Foerster, van de Grind & Grusser, 1977; Sakai & Naka, 1990).

Laufer and Verzeano (1967) first suggested that periodic activity in retinal networks may result from coupling among large cell populations. Recording simultaneously from more than one cell in the cat retina, the authors observed that discharges of single cells may show constant phase relations with their oscillating neighbors. Later studies employing cross-correlation analysis have shown that the activity of neighboring cells with overlapping receptive fields may

* Corresponding author. Tel.: +49-69-96769336; fax: +49-69-96769327.

E-mail address: neuenschwand@mpih-frankfirt.mpg.de (S. Neuenschwander)

0042-6989/99/$ - see front matter
PII: S0042-6989(99)00042-5

indeed exhibit strong correlations (Arnett, 1978; Arnett & Spraker, 1981; Mastronarde, 1989; Sakai & Naka, 1990; Meister, Lagnado & Baylor, 1995; Brivanlou, Warland & Meister, 1998). Recently we have demonstrated that synchronization of responses may also occur over surprisingly large distances ($>20°$) provided that the stimuli used for the activation of the cells are continuous (Neuenschwander & Singer, 1996). This correlated firing was always associated with a strong periodic modulation, suggesting that synchronization was mediated by oscillatory activity.

In studying stimulus-driven neuronal interactions it is important to determine to what extent correlations arise simply from stimulus coordination. In our previous investigations we obtained evidence that the oscillatory patterning of responses that was associated with response synchronization was not phase locked to the onset of stationary stimuli or to the periodic activation caused by drifting gratings. This contrasts with the claim that the temporal patterning of retinal and thalamic responses is precisely time-locked to stimulus onset (Wörgötter & Funke, 1995; Reich, Victor, Knight, Ozaki & Kaplan, 1997). However, in our previous studies, we have not examined stimulus-locked synchronization in the early phase of the responses, since we computed average correlations over the sustained components of the responses (Neuenschwander & Singer, 1996; Castelo-Branco, Neuenschwander & Singer, 1998; but see Rager & Singer, 1998). Here we extend this analysis to examine a possible contribution of stimulus-locking to the development of synchronization. We have made multiple-electrode recordings from retinal ganglion cells and employed a sliding window correlation analysis to study the time course of synchronous oscillations during spontaneous and visually-driven activity.

2. Methods

2.1. Recordings

Simultaneous intra-ocular recordings from retinal ganglion cells were performed in 11 anesthetized and paralyzed cats. Anesthesia was induced with ketamine (10 mg kg^{-1}, i.m.) and xylazine (2 mg kg^{-1} i.m.), and maintained with administration of N_2O and O_2, 70:30%, supplemented with 0.5–1.2% halothane. The left eye was immobilized by suturing the conjunctiva to a ring fixed to the stereotaxic frame (Peichl & Wässle, 1979). After surgery, the pupil was dilated with atropine (2%) and the animal paralyzed with intravenous pancuronium bromide (0.1 mg kg^{-1} per h).

Multi-unit and occasionally also single cell activity was recorded from a total of 150 sites (68 pairs of recordings sites) with two to three tungsten electrodes (1.0–2.0 MΩ) inserted into the eye through a guide tube. The electrodes could be moved independently and placed under visual control at the desired location. Signals were amplified ($\times 10\,000$), band-pass filtered from 0.3 to 3 kHz and spikes were detected with an amplitude discriminator (resolution, 0.1 ms). All surgical procedures were performed according to the German guidelines for animal experimentation.

2.2. Visual stimulation

Stationary light stimuli were generated with an optical bench (contrast, 0.75; background luminance ~ 0.4 cd m^{-2}), and front-projected onto a tangent screen. Since the projection light was provided by a DC powered source, the stimuli were free of any oscillatory component. Drifting gratings (spatial frequency, 0.2–1.5 c deg^{-1}; temporal frequency, 1–6 Hz) were generated on a monitor with a refresh rate of 100 Hz (contrast, 0.50). The responses of a photo-diode were used to measure the onset jitter of stimuli generated by the optical bench and the computer screen. The scatter of onset latencies was below one millisecond for both stimulus conditions.

2.3. Data analysis

Response histograms were computed with a resolution of 25 ms, and a 1–2 s analysis window was placed over an epoch of strong coactivation for correlation analysis. Auto- and cross-correlograms were calculated between single trial responses within these windows with a resolution of 1.0 ms and subsequently averaged over 20 stimulus repetitions. Shift predictors were routinely computed to control for correlations caused by stimulus-locking (Perkel, Gerstein & Moore, 1967). Responses were considered as synchronized by intraretinal interactions if a modulation persisted in the correlograms after subtraction of the shift predictor controls from the raw correlograms (see evaluation criteria in Castelo-Branco et al., 1998). To quantify the strength, frequency and phase shift of synchronous oscillations, we have taken the parameters of a damped-cosine function fitted to the correlograms, as described by König (1994). Correlation strength was assessed from the relative modulation amplitude index, i.e. the ratio of the amplitude of the central peak of the fitted function over its offset. The amplitude of the correlograms was normalized by the geometric mean of the number of spikes for each cell of a pair.

A sliding window analysis was used to follow the time course of synchronization. To this end, a 100–250 ms analysis window was moved over the responses in successive 50–100 ms steps. The correlograms obtained from each of those windows were displayed in a two-dimensional plot (see example in Fig. 3), where the y-axis

denotes the time lag of the correlation function and the x-axis the time course of the responses.

3. Results and discussion

3.1. Synchronous oscillations during spontaneous activity

Retinal ganglion cells often showed an oscillatory modulation of the sustained discharges that occur during darkness or constant levels of illumination. Such oscillations typically started several seconds after a change in ambient illumination and then gradually increased in strength. Oscillation frequencies were in the range of 25–35 Hz and tended to decrease over time. Occasionally, oscillatory patterns of different frequency could coexist. Fig. 1 shows a case of spontaneous oscillatory activity that was correlated across two sites. Here fast oscillations around 30 Hz are superimposed onto slower oscillations at 2–5 Hz. In this example, multi-unit recordings were obtained from two sites

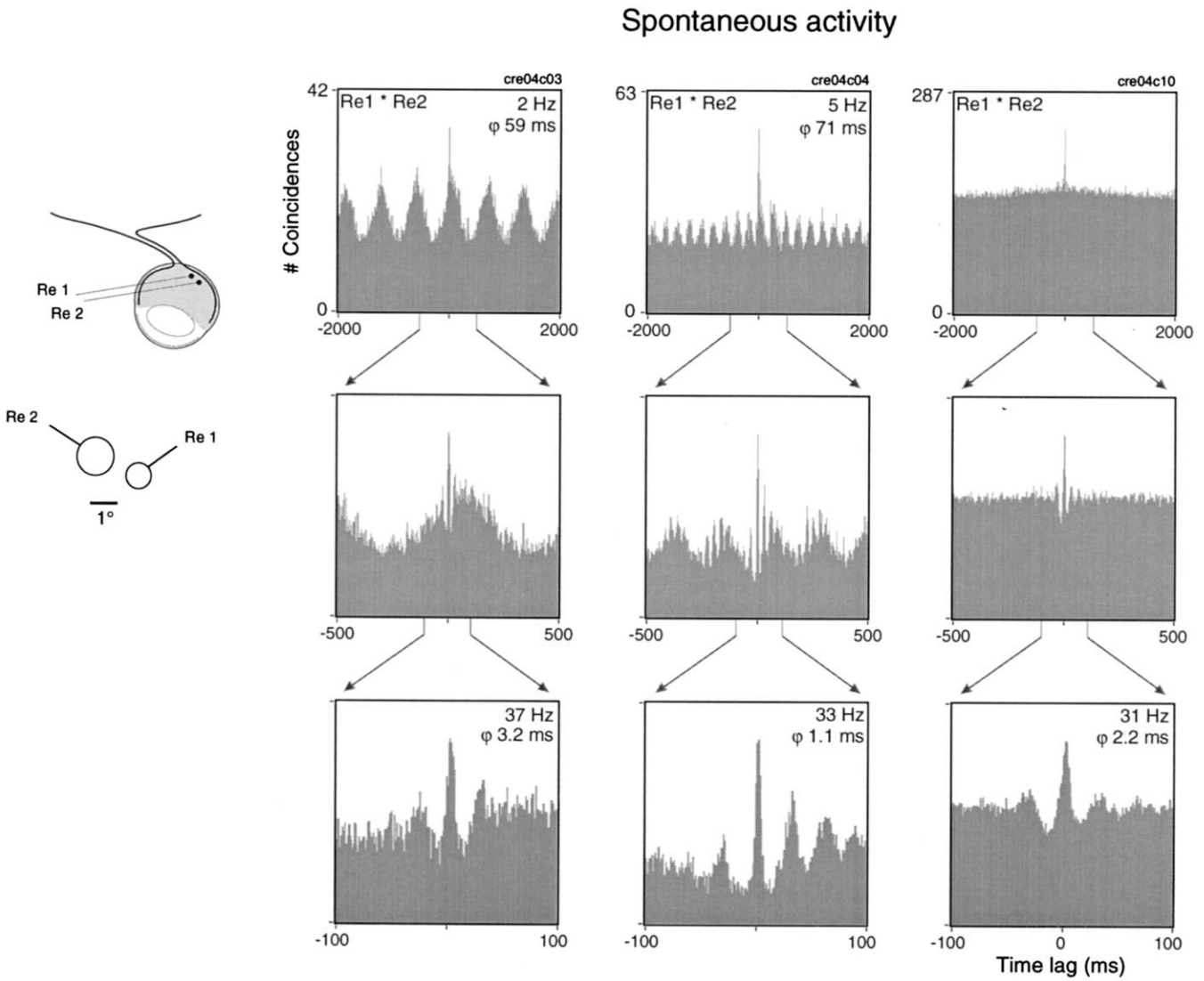

Fig. 1. Synchronous oscillations in spontaneous discharges of retinal ganglion cells. Cross-correlation functions were obtained for the ongoing activity of two cell clusters (multi-unit recordings, Re1 and Re2) at different epochs and various levels of adaptation. The correlograms shown in the upper row were successively expanded in the lower rows, as indicated by the arrows. Left and middle columns; the slow 2–5 Hz oscillatory pattern visible in the upper correlograms reveals a fast component above 30 Hz at higher time resolutions. The superposition of these oscillatory patterns is readily visible in the correlograms at the center. Right column; only the fast component is present. Notice that synchronization of the fast oscillatory component is highly precise (phase shifts, 1–3 ms) and occurs independently of the slow oscillatory patterns. Oscillation frequency and phase shifts (φ) were computed after fitting a damped cosine function to the correlograms (not shown). Adaptation levels for recordings in left and right columns, mesopic ($\sim$200 lux); middle column, scotopic. Recording sites were located close to the area centralis. Inset, circles, receptive fields; center-to-center distance, 2° of visual field. Re1 mean firing rates, 4.3, 4.0, 50.7; Re2, 125, 169, 101 spikes s^{-1} for each case, respectively.

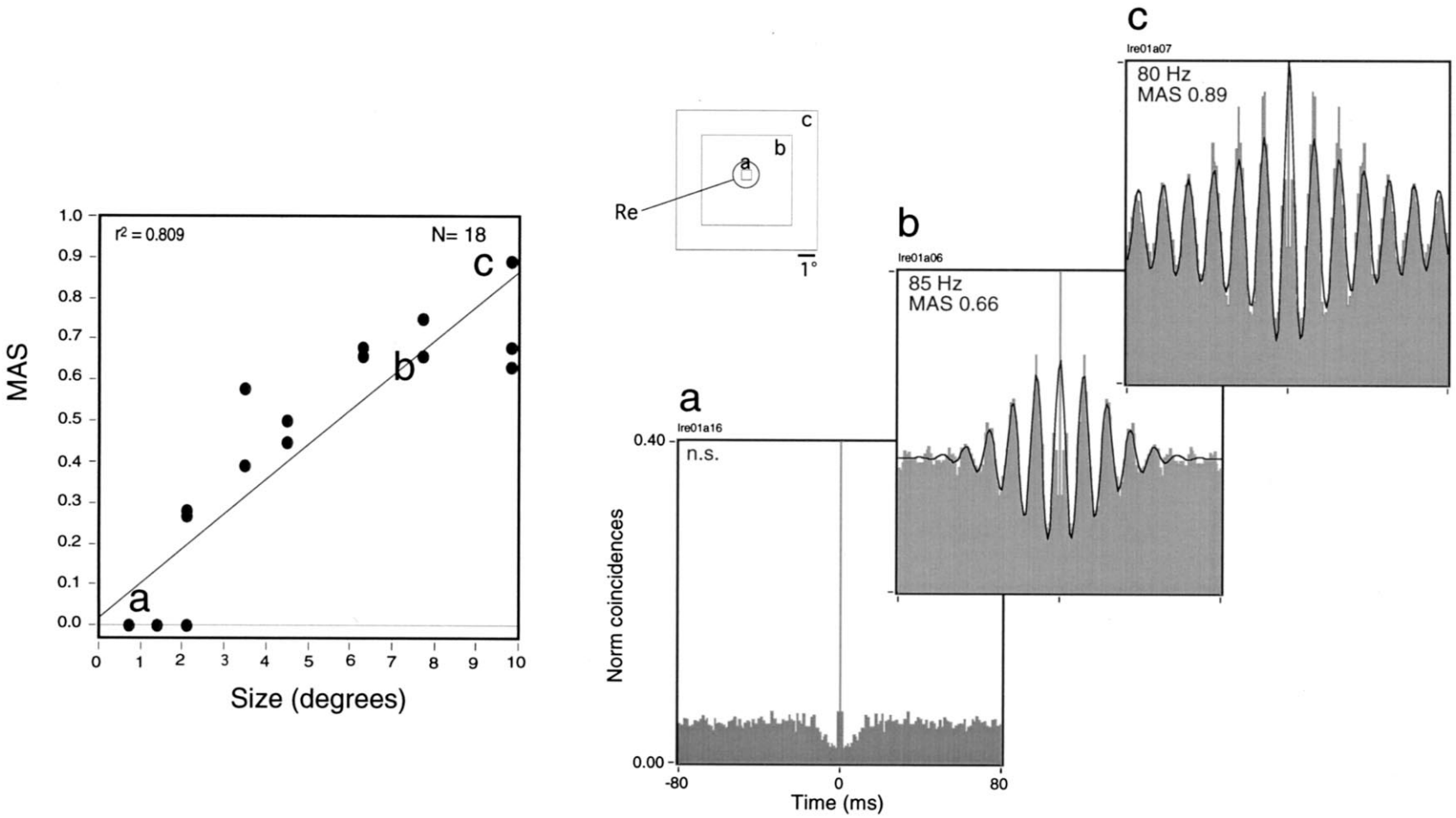

Fig. 2. Effects of stimulus size on strength of oscillatory modulation. The plot depicts the modulation amplitude of the first satellite peak as a function of the size of the stimulus. Multi-unit responses were obtained to light squares of different sizes flashed over the center of the RF (inset). Autocorrelograms corresponding to points a, b and c in the plot are shown to the right. The amplitude of the correlograms was normalized by the geometric mean of the number of spikes for each cell.

at different epochs, after prolonged adaptation at either mesopic or scotopic levels. Cross-correlograms were computed over several seconds. The deep modulations of the correlograms indicate that the oscillatory patterning of each of the cell groups was very stable and precisely synchronized. Accordingly, both autocorrelation functions were also deeply modulated (not shown). The fast oscillations were not related to the slow oscillations. The latter were erratic and appeared and disappeared spontaneously with a time course of several minutes. In contrast, the fast oscillations were sustained and regular, and changed only when changes in ambient illumination caused a sharp increase in oscillation frequency.

The most striking feature of the spontaneous fast oscillations in the retina is the precision of their synchrony. As shown in Fig. 1, the center peaks in the correlograms are only a few milliseconds wide, indicating precise phase-locking of the fast oscillatory components (peak widths, 7–20 ms; time lags, 1–3 ms). In contrast, the broad peaks associated with the slow oscillations often deviate from zero phase lag, indicating that large phase rotations may exist for the slow oscillatory activity. These large differences in the phase rotations suggest independent mechanisms involved in the generation of the slow and fast oscillations.

In the vertebrate retina, precise synchronization of the spontaneous discharges of ganglion cells has been reported in several cross-correlation studies (Arnett, 1978; Arnett & Spraker, 1981; Mastronarde, 1989; Brivanlou et al., 1998). In the cat, simultaneous recordings from neighboring cells revealed strong correlations between cells with overlapping receptive fields and same center polarity, which were attributed to reciprocal interactions (Mastronarde, 1983, 1989). These correlations showed no sign of an oscillatory patterning. Recently, similar findings were reported for ganglion cells of the salamander under dark adaptation (Brivanlou et al., 1998). The synchronization patterns were heterogeneous, differed in the degree of temporal precision, and were shown to depend on different mechanisms. Broad correlations depend on chemical synaptic transmission, probably reflecting common input from photoreceptors. The more precise correlations (peak widths of 10–50 ms or less than 1 ms) are due to electrical coupling both between amacrine and ganglion cells and among ganglion cells (Brivanlou et al., 1998). Interestingly, our data show the same multimodal distribution of the width of correlation peaks as described for the salamander. Thus, it is possible that the slow and fast oscillations that were associated with high and low precision synchronization in the present study reflect interactions at different levels of retinal processing.

The question remains why oscillatory phenomena in the frequency ranges above 2 Hz have been seen only rarely in the isolated retina (Meister et al., 1995; Brivanlou et al., 1998) and in vivo (Mastronarde, 1983; Robson & Troy, 1987) when single unit activity was recorded, but are commonly observed in multi-unit recordings (Doty & Kimura, 1963; Laufer & Verzeano, 1967; Rodieck, 1967). Robson and Troy (1987) present in their single cell study inter-spike interval distributions with broad peaks, suggestive of oscillatory activity. The most likely reason for this discrepancy is that individual cells, even though their discharges are time locked to the global oscillatory activity of the populations, reflect the rhythmic activity only poorly because they do not discharge in every cycle, missed cycles occurring in irregular sequences (see Laufer & Verzeano, 1967; Buzsáki, 1996).

3.2. Stimulus-driven synchronization

Upon visual stimulation, ganglion cells often engage in strong oscillatory activity, which becomes precisely synchronized over large distances, depending on spatial and temporal properties of the stimulus (Neuenschwander & Singer, 1996). Stationary light stimuli or moving gratings can induce long-range synchronization, provided that contiguous regions in the retina are activated. One critical determinant for the strength of the oscillatory patterning is the size of the stimulus (Fig. 2). Punctiform stimuli restricted to the center of the receptive fields elicit only weak oscillations while large stimuli covering both the center and surround regions commonly induce a strong oscillatory modulation of the responses, whereby these effects on oscillations are independent of changes in firing rates.

Of the 150 recording sites, 115 exhibited oscillatory responses for at least one stimulus condition (81 ± 17 Hz, mean $\pm$S.D.). Oscillation frequencies differed markedly from those observed during spontaneous activity (~ 30 Hz). For stationary stimuli, ON-responses oscillated at significantly higher frequencies than OFF-responses (mean of 91.7 ± 11.7 Hz for ON- and 79.0 ± 20.0 Hz for OFF-responses; $P < 0.0001$, ANOVA, 1 d.f., $F = 55.83$). For responses to moving stimuli there was an interesting relationship between velocity and oscillation frequency (Fig. 3). For a moving bar sweeping over the receptive field, oscillation frequency increased monotonically with the velocity of the stimulus (slope, ~ 0.5 Hz deg^{-1} per s; tested range, 1.7–70 deg s^{-1}; three recording sites). A similar relation was found between stimulus velocity and correlation strength when two sites were coactivated by a common moving stimulus (either a single bar or gratings). The relative modulation amplitude of the center peak increased with stimulus velocity. Even short responses (120 ms) evoked by fast moving stimuli exhibited a strong oscillatory modulation, indicating that retinal oscillations have a fast onset, and may follow brisk changes in the visual scene.

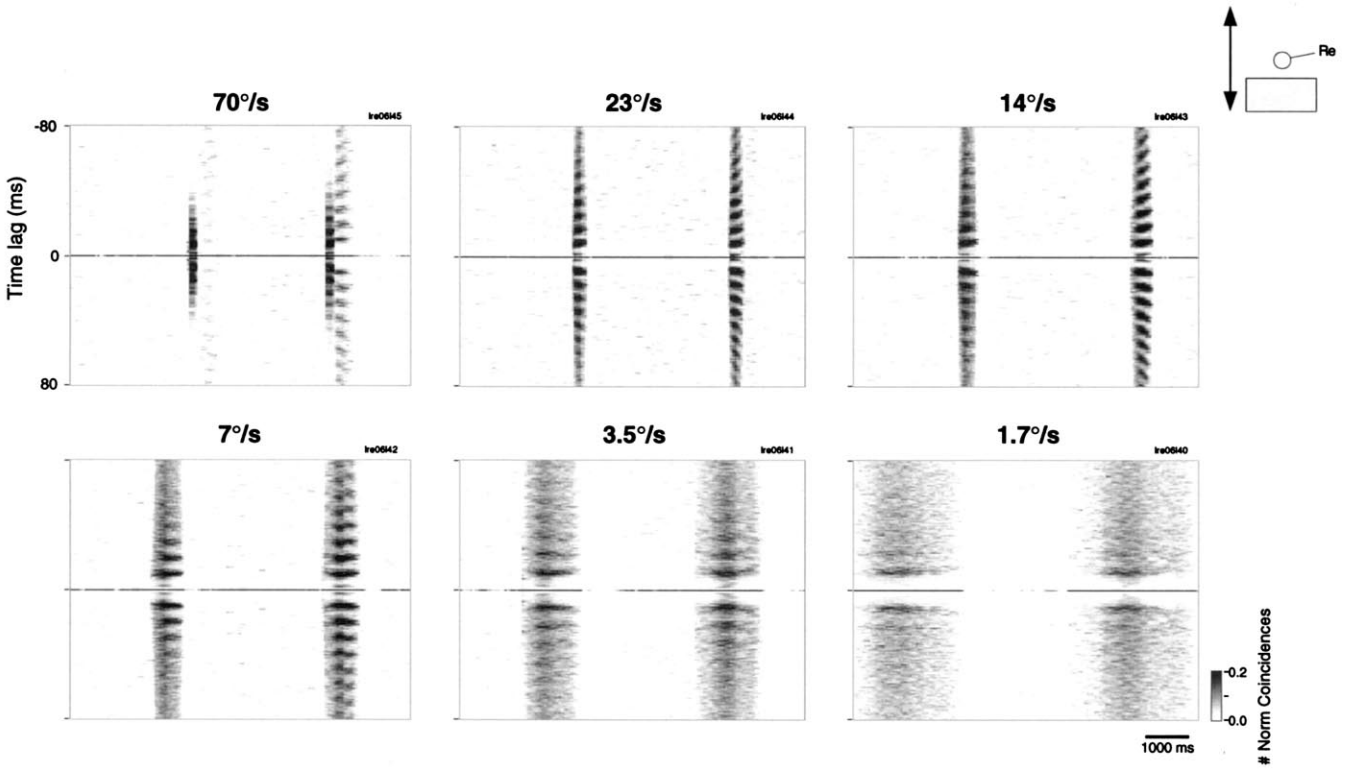

Fig. 3. Effects of stimulus velocity on strength of oscillatory modulation. Series of sliding-window auto-correlation analysis of the response of a single ON-X-cell to a bar sweeping over the receptive field at various speeds (Inset: circle, receptive field center; the arrow indicates the direction of forward and backward movement of the bar over the receptive field). Number of stimulus repetitions, 10. The oscillatory strength increases from 0.3 to 4.1, and the oscillation frequency from 87 to 123 Hz (average auto-correlograms, not shown). Correlograms have normalized amplitudes.

This stimulus-induced synchronization had several characteristic features: (i) if it occurred over large distances it was always associated with oscillatory patterning, suggesting that synchronization and oscillatory activity in the retina are mutually dependent processes (Fig. 4); (ii) it was very precise, the mean phase rotation being $26.0 \pm 36°$ and the mean phase lag 1.8 ± 1.2 ms (S.D.); (iii) the correlations were strong (mean modulation amplitude, 0.8 ± 0.7 S.D.) and exhibited little fluctuations throughout the responses. Both, the oscillation frequency and the phase angle of synchronization remained remarkably constant within and across trials.

Ghose and Freeman (1992, 1997) have proposed a model which postulates a retinal origin for oscillatory activity in the cortex. In the model, cortical oscillations are explained by the integration of oscillatory inputs arising from spontaneous discharges in the retina transmitted reliably through the thalamo-cortical pathway. A basic assumption of the model is that individual retinal ganglion cells behave as independent oscillators. As shown above, our results point to a different scenario. Synchronization of oscillatory responses in the retina is very precise, phase lags being restricted to only a few milliseconds. Moreover, synchronization occurs in a stimulus-dependent manner and increases with stimulus size. With large stimuli, more cells are recruited into synchronously oscillating ensembles, whereas the individual phase differences remain rather small. This explains why retinal oscillations are readily visible in multi-unit recordings. By contrast in the model of Ghose and Freeman addition of more oscillatory cells results in a decrease of oscillation strength of the net outputs. Moreover, our results show that even during spontaneous activity, correlations occur with only small phase delays and the associated oscillations occur in a frequency range (~ 30 Hz) that is well below the range postulated by the model of Ghose and Freeman (1997). Taken together, our findings fail to support the hypothesis that ganglion cells oscillate independently, suggesting instead that oscillatory phenomena in the retina arise from large-scale network interactions.

3.3. Are synchronous oscillations in the retina stimulus-locked?

We have shown previously that the sustained oscillatory responses in the retina are not phase-locked to the stimulus onset (Neuenschwander & Singer, 1996; Castelo-Branco et al., 1998). A key finding was the dependence of long-range synchronization on stimulus

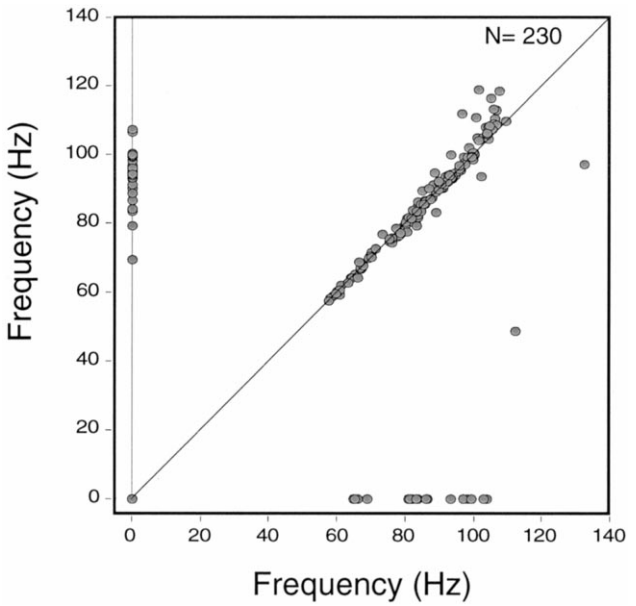

Fig. 4. Comparison of oscillation frequency for the two sites of each recording pair exhibiting synchronization. The large majority of pairs showed oscillatory responses with similar frequencies at both recording sites (points cluster along the diagonal). The points lying on the x- or y-axis correspond to the cases in which one or both cells showed no oscillatory responses.

configuration. Two simultaneously flashed stimuli induced precise synchronization of oscillatory responses only when they were contiguous. However, when the same stimuli were repositioned so that they were separated by a gap, synchronization was disrupted (see Fig. 4b of Neuenschwander & Singer, 1996). These findings indicate clearly that synchronization cannot be due solely to stimulus-locking but results from neuronal interactions. If synchronization resulted from phase-locking to the stimulus onset, it should persist regardless of the spatial stimulus configuration. In our previous studies, however, correlograms were computed within 1–2 s windows, in general beginning 100–300 ms after stimulus onset to exclude the initial phasic component of the responses. Here we have re-examined the question of stimulus locking by means of a sliding window analysis, including also the early response component.

This refined analysis occasionally revealed tight phase-locking to the stimulus onset for the first few cycles of the oscillatory responses. Thereafter, synchronization persisted independently of stimulus coordination. A simple way to examine stimulus-locked synchronization is to compute response histograms with a resolution of 1 ms. If individual oscillatory responses exhibit negligible phase jitters in relation to stimulus onset, deep modulations should become visible in the histograms. Fig. 5 shows an example of strong phase-locking to stimulus onset that is restricted to the initial component of the responses. Multi-unit recordings were obtained from three spatially separated sites in the retina, that had non-overlapping receptive fields. At all sites, the cells showed strong oscillatory responses to a large stationary stimulus flashed over the receptive fields (oscillation frequencies, ~ 87 Hz; number of stimulus repetitions, 100). The response histograms computed at high resolution exhibit multiple peaks, that are precisely aligned across recording sites for the first 100 ms of the responses. This oscillatory modulation in the responses appears as early as 20 ms after response onset and rapidly decays over the course of the responses. This decay is attributable to a decrease of stimulus locking which in the averages leads to smearing of the oscillatory modulation. Accordingly, at the very beginning of the responses, the shift predictor controls are modulated as strongly as the raw correlograms (indicated by the arrows in Fig. 5) while they become flat thereafter. This indicates that the oscillatory modulation of the early response is phase locked to the stimulus while the synchronization of the later oscillations is due to intraretinal coordination.

To quantify the contribution of stimulus coordination to the synchronization of oscillatory responses, we have made the following analysis. For all cases of synchronous oscillations, response histograms were computed with a resolution of 1 ms and two consecutive 100 ms analysis windows were placed over the initial phase of the responses for correlation analysis. We then compared the raw cross-correlograms with their corresponding shift predictor controls. Even within the first window, shift predictors were less strongly modulated than the raw correlograms in 52% of the cases ($N = 191$ cross-correlograms), suggesting a substantial contribution of internal synchronizing mechanisms already during the first 100 ms of the response. In the remaining 48% of the cases, synchronization could be fully explained by stimulus coordination. Interestingly, the precision of synchronization achieved by the internal mechanism was as high as that caused by stimulus locking. The phase shifts for stimulus locked correlations were as small (mean 0.93 ms) as those for non-stimulus locked correlations (0.53 ms, Mann–Whitney, $P > 0.5$). In the second window, 92% of the correlograms remained strongly modulated after subtraction of the shift predictor, indicating that stimulus-locking is very brief and cannot account for sustained synchronization of the responses.

In the experiments described above, the stimuli were generated with an optical bench, and except for the initial transient component, they were lacking any temporal structure. However, when drifting gratings were used, the stimulus contained an oscillatory component of 100 Hz caused by the monitor retrace. Because retinal ganglion cells can follow high frequency flicker (Reich et al., 1997), we wondered whether this temporal modulation influenced the oscillatory patterns in the

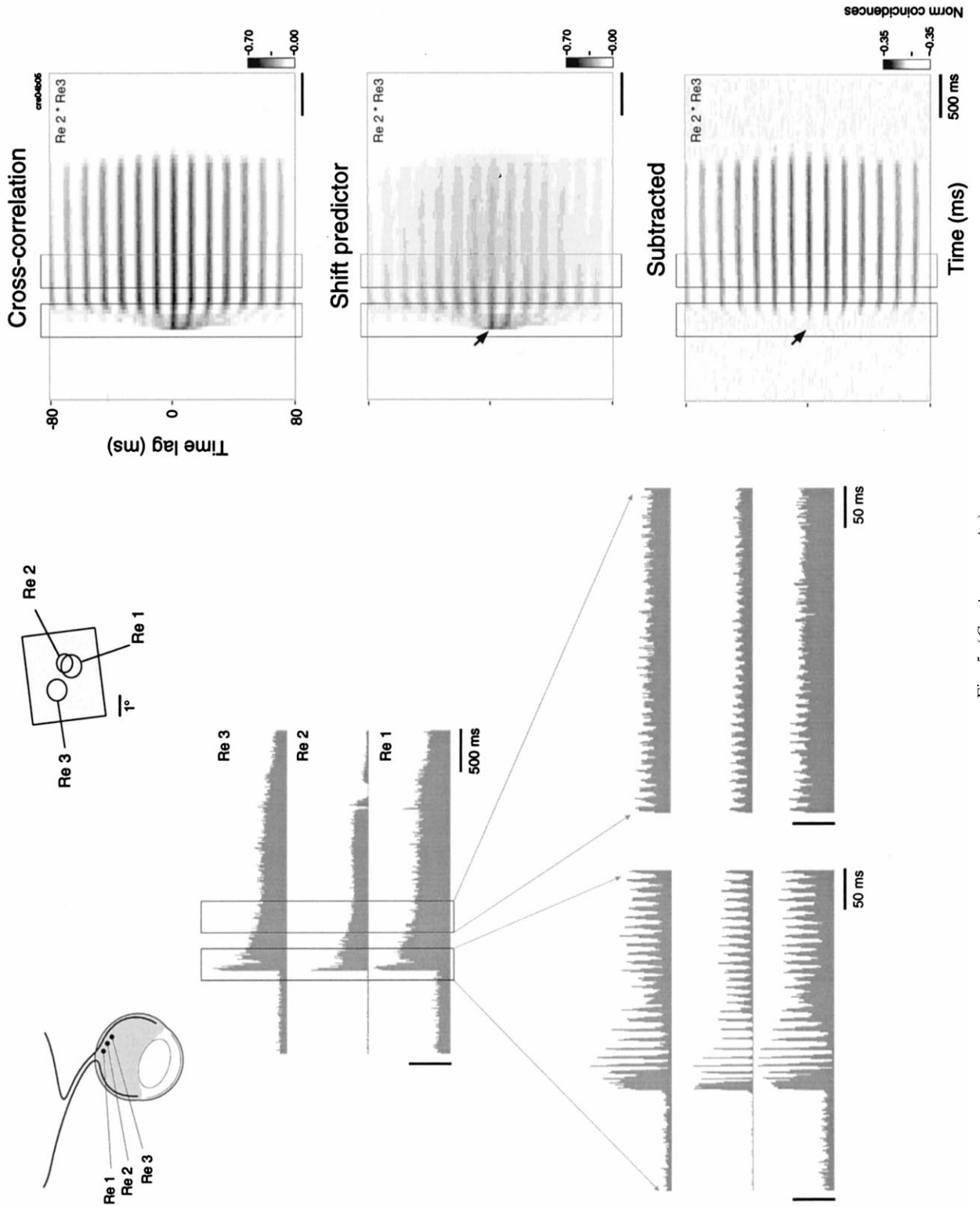

Fig. 5. (*Caption opposite*)

responses. In a small fraction of sites (two out of 150), the cells were indeed able to follow the monitor retrace. This is exemplified in Fig. 6 for two independently recorded Y-cells that were activated with high contrast gratings that suddenly appeared on a dark display, covering an area of $\sim 10°$ of visual angle. As seen in the response histograms, both cells strongly responded with modulated discharges to the gratings. The respective autocorrelations show an oscillatory modulation of the responses which persists throughout the whole response and is superimposed on the much slower modulation of discharge rate caused by the grating. The only difference in the response patterns of the two cells is the frequency of the fast oscillatory modulation (93–80 Hz and 100 Hz, respectively). However, the shift predictors reveal dramatic differences. In the first case, the oscillatory modulation in the raw correlogram persists after subtraction of the shift predictor, except of the transient at the beginning of the response (Fig. 6A). In the second case subtraction of the shift predictor leads to a near complete abolition of the oscillatory modulation, indicating that the oscillatory patterning was precisely locked to the stimulus flicker throughout the whole response. As expected in this case, oscillation frequency was 100 Hz and hence matched precisely the frequency of the monitor retrace.

The fact that a cell is able to follow with high precision the temporal modulation of a stimulus does not preclude its participation in internally generated oscillations. Y-cells which were able to follow the monitor retrace exhibited strong oscillatory responses to stimuli lacking temporal modulation.

A few autocorrelation studies have shown clear evidence for stimulus-locking of oscillatory responses in the retina and lateral geniculate (Ariel et al., 1983; Ghose & Freeman, 1992; Wörgötter & Funke, 1995). Our results extend these observations in at least two respects. First, our sliding window analysis reveals that although precise locking to stimulus onset may occur initially, it does not explain the synchronization along the course of the responses. Second, spatial properties of the stimulus seem to be more important in establishing internal phase-locking of the responses than the relative timing of the stimulus. Moreover, with respect to precision, there was no difference between internal and external phase-locking. This is important considering the impact of precisely synchronous spikes arriving at a target cell (Alonso, Ursey & Reid, 1996). At the cortical level, both internal and external coordination in the relative timing of the inputs may be used for further, more complex, linking operations. Recently it has been demonstrated that binding is affected by external synchrony in the visual stimulus (Alais, Blake & Lee, 1998; Leonards & Singer, 1998; Usher & Donnelly, 1998). Precise phase-locking to stimulus onset may be used for perceptual grouping as it has been shown that features that are correlated in time tend to be grouped over space (Alais et al., 1998).

3.4. Cooperative interactions in the retina

The sliding window analysis revealed a time-dependent change in the oscillatory patterning of the responses. Oscillation frequency decreased progressively along the course of the responses, whereby the decrease in frequency was steepest immediately after response onset. This trend was invariably seen for responses to flashed stationary light stimuli or moving gratings and at all recording sites, except for the few cases in which oscillatory patterning was not generated internally but due to the retrace flicker (Fig. 6B).

In Fig. 7, we examined the development of oscillatory responses of a single ON-cell to moving gratings of different contrasts. As in the examples shown in Fig. 6, the gratings suddenly appeared on a previously dark screen. In these conditions, the display of gratings with zero contrast was equivalent to a large flashed stimulus of average luminance. As seen in the autocorrelation functions, strong oscillatory patterning developed immediately after stimulus onset, with gradual decrease in oscillation frequency along the responses. The increase in contrast induced a deep modulation in the firing rates, without affecting however the overall time course of the oscillatory discharges. The modulation of the oscillatory activity is present right from the beginning of the responses to each of the successive cycles of the gratings. Notice also that the progressive change in oscillation frequency follows the same time course for

Fig. 5. (*Opposite*) Stimulus-locking of oscillatory responses in the retina. Responses to a flashed light stimulus were obtained from three simultaneous multi-unit recordings. Left panels, response histograms computed with a 1 ms resolution. Two distinct epochs in the course of the responses are expanded below (enclosed by the boxes, length of 400 ms). A strong oscillatory modulation is visible in the early phase of the responses (left box), fading completely within a few hundred milliseconds (right). The peaks seen in the histograms are precisely aligned in the first few cycles, indicating a strong phase-locking to stimulus onset. Right panels; cross-correlation functions obtained through a sliding window analysis (shown for only one recording pair). Notice that the shift predictor control explains entirely the correlation pattern seen in the transient phase of the response (left box, as pointed by the arrows). Thereafter, the modulation persists after subtracting the shift predictors from the raw correlograms, indicating that synchronization is no longer stimulus-locked, reflecting neuronal interactions. Number of stimulus repetitions, 100. Response histograms, scale bar 150 spikes s^{-1}. Sliding window analysis, step 50 ms, analysis window 200 ms; normalized amplitude. Top inset, circles, receptive fields; square, light stimulus. Receptive fields were located in the central visual field. The stimulus was generated by an optical bench and therefore it was free of any oscillatory component.

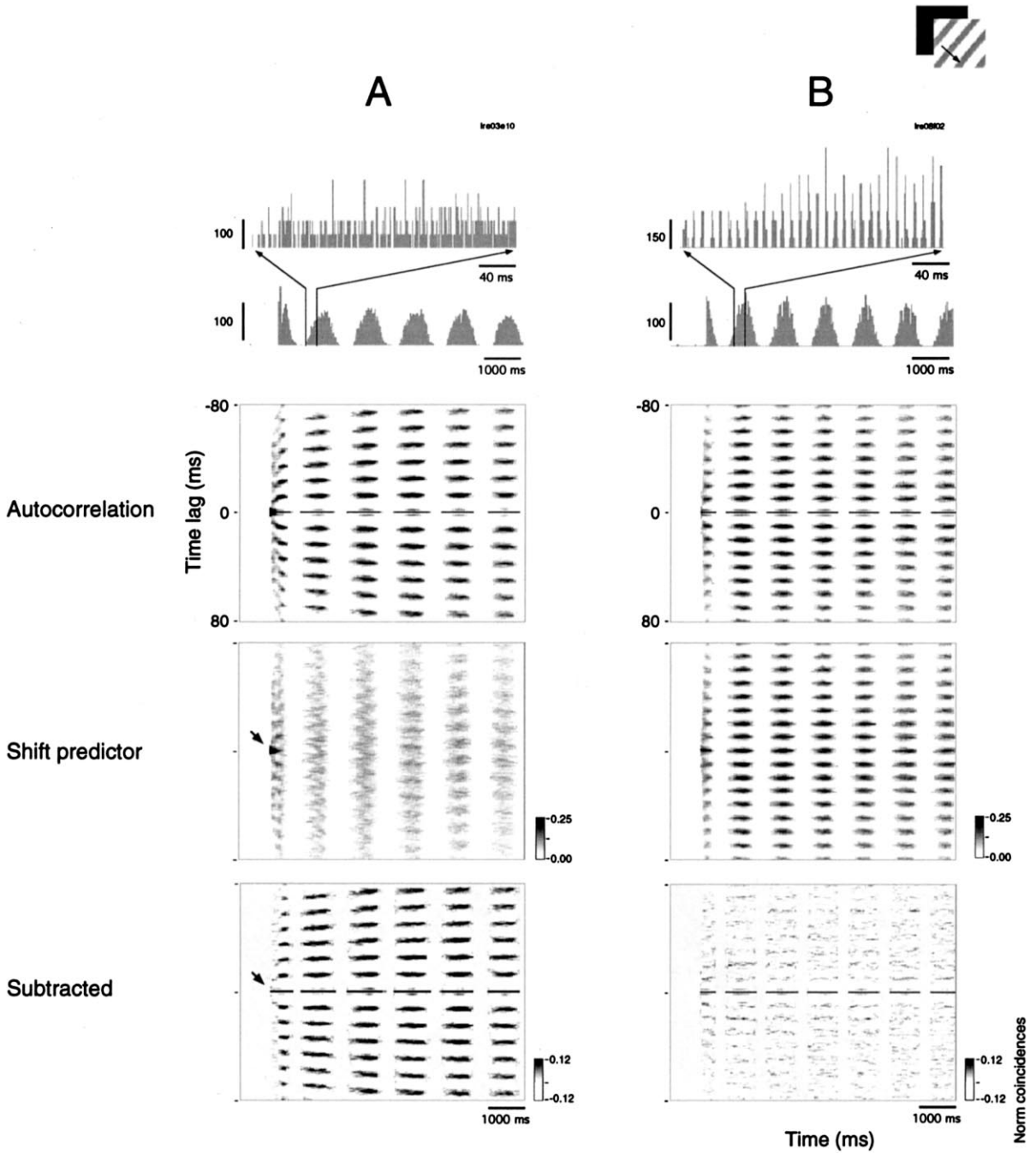

Fig. 6. Oscillatory responses of Y-cells to moving gratings displayed on a computer monitor. (A) Oscillatory responses of a cell which is not able to follow the 100 Hz flicker inherent to the stimulus. Sliding window autocorrelation analysis shows a strong oscillatory modulation at 82 Hz. The modulation in the correlogram persists after subtraction of the shift predictor, indicating that there was no phase-locking of the oscillatory processes to the onset of the stimulus. Observe, however, that the initial phasic component of the response is precisely locked to the stimulus onset (indicated by the arrows). (B) In contrast to (A), this particular cell is able to follow the retrace of the computer monitor. The shift predictor explains the quasi totality of the modulation in the raw autocorrelation function and therefore no structure remains in the subtracted correlogram. Oscillation frequency was identical to the monitor refresh rate (100 Hz). Notice that the slight decrease in oscillation frequency over time seen in (A) is absent in (B). Response histograms (bin width, 25 ms) were aligned to the sliding window autocorrelograms (step 50 ms, analysis window 200 ms, normalized amplitude). Response histograms computed with a 1 ms resolution are shown above. Gratings were presented on a dark screen (contrast, 0.50; background luminance, 0.4 cd m^{-2}). Stimulus repetitions, 20.

the periodic responses to the grating or for the sustained responses for a large flashed stimulus (grating of zero contrast). A possible interpretation is that upon the sudden luminance change associated with the appearance of the gratings, a large ensemble of retinal elements engages in synchronous oscillatory activity

that is sustained across the pauses in firing and assures the continuity of the oscillatory patterning of the responses independently of rate modulations of individual cells.

This possibility was investigated using the following paradigm. Two cells were activated by a single large stimulus and subsequently one of them was silenced by covering its receptive field with a small stimulus of opposite polarity. In Fig. 8, an example is shown for a pair of OFF-cells. The offset of a light stimulus induced a strong synchronous oscillatory patterning, with a smooth decrease in frequency over time. Intermittent silencing of one of the cells did not introduce any discontinuity in the overall pattern of synchronization between the two cells. Right after the epochs of silencing, the first spikes were already synchronized between the two cells. Notice that if the oscillatory responses were locked to the onset of the stimulus, we should expect a resetting of the oscillatory pattern for each new response. Thus, during the silent epochs, the inactive cell had apparently remained exposed to subthreshold influences from the oscillatory population of surrounding cells. We propose that it is this coupling mechanism that organizes the retinal network into large ensembles of cooperating neurons that can synchronize their discharges over long distances if activated by a continuous stimulus.

4. Conclusion

Our results show that retinal ganglion cells may interact over long distances depending on the spatial (Neuenschwander & Singer, 1996) and temporal prop-

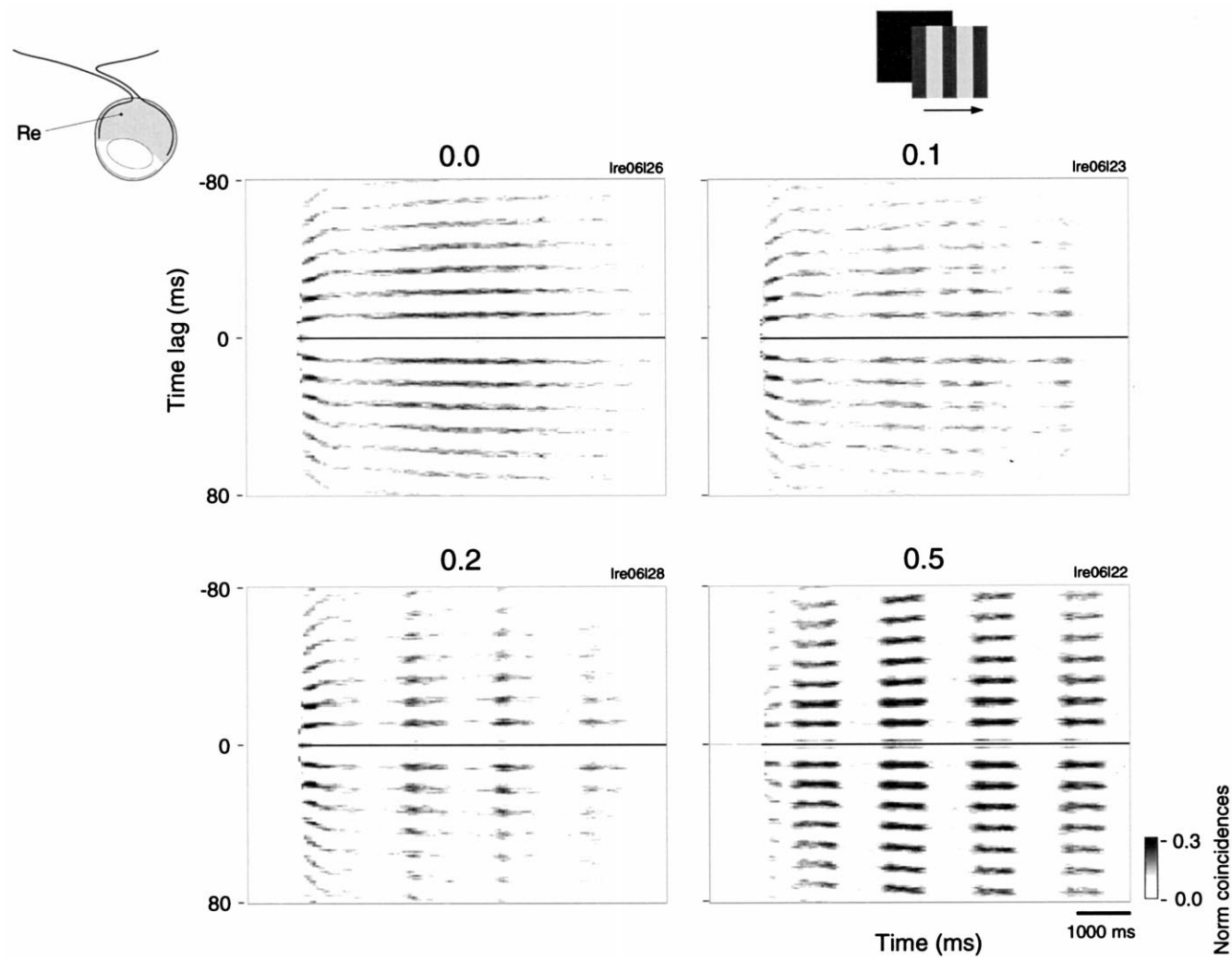

Fig. 7. Oscillatory patterning in responses to moving gratings of different contrasts. Recordings were obtained from a single OFF-retinal ganglion cell. Contrast of the stimulus was changed from 0.0 to 0.5, as indicated at the top of the sliding window autocorrelation panels. Notice that the overall oscillatory pattern is maintained regardless of changes in rate modulation of the responses. Gratings were presented on a dark screen (background luminance, 0.4 cd m^{-2}). Stimulus repetitions, 20. Average luminance was constant for the various stimulus conditions. Sliding window analysis step 50 ms, analysis window 200 ms; normalized amplitude.

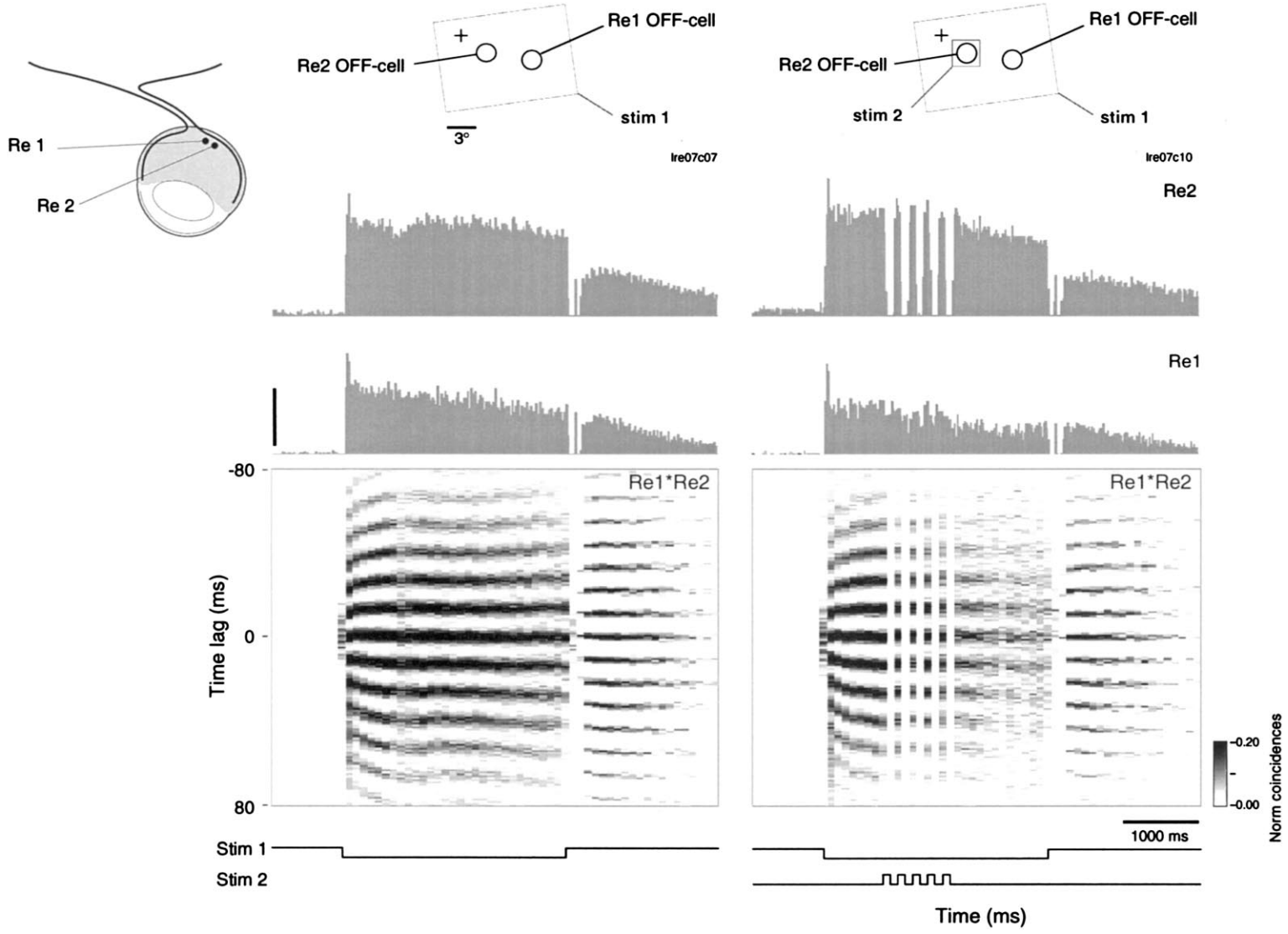

Fig. 8. Synchronous oscillatory patterning is not affected by brief silencing of cell responses. Left panels, responses from two OFF-cells to the offset of a large light rectangle centered over the receptive fields (stimulus 1). Sliding window cross-correlation analysis reveals a strong oscillatory pattern with a progressive decrease in frequency over the course of the response. Right panels; one of the cells (Re2) was silenced intermittently by a second stimulus flashed over its receptive field (small light square, stimulus 2). As seen in the response histograms, the second stimulus caused a complete silencing of this cell, without affecting the other. Notice that the absence of spiking from one cell during brief epochs introduces no discontinuities in the overall course of synchronization. Response histograms, scale bar 100 spikes/s. Sliding window analysis step 100 ms, analysis window 100 ms; the amplitude of the correlograms was normalized (see Section 2). Each stimulus condition was repeated in blocks of 20 trials. Inset; cross denotes the projection of area centralis.

erties of the stimulus. Synchronization of oscillatory responses was observed for continuous stimuli and was shown to be non phase locked to the stimulus, suggesting an internal mechanism for synchronization. Synchronous oscillations persisted despite brief and local occlusion of stimuli, suggesting that synchronization emerges from population dynamics. Strong phase-locking to the stimulus onset was observed in the very early phasic component of the responses. We proposed that external phase-locking serves for binding in the temporal domain (two stimuli appearing at the same time are linked together, Leonards & Singer, 1998) and internal phase-locking serves for binding in the spatial domain.

References

Adrian, E. D., & Matthews, R. (1928). The action of light on the eye. Part III. The interaction of retinal neurones. *Journal of Physiology*, *65*, 273–298.

Alais, D., Blake, R., & Lee, S. H. (1998). Visual features that vary together over time group together over space. *Nature Neuroscience*, *1*, 160–164.

Alonso, J. M., Ursey, W. M., & Reid, R. C. (1996). Precisely correlated firing in cells of the lateral geniculate nucleus. *Nature*, *334*, 348–350.

Ariel, M., Daw, N. W., & Rader, R. K. (1983). Rhythmicity in rabbit retinal ganglion cell responses. *Vision Research*, *23*, 1485–1493.

Arnett, D. W. (1978). Statistical dependence between neighboring retinal ganglion cells in goldfish. *Experimental Brain Research*, *32*, 49–53.

Arnett, D. W., & Spraker, T. E. (1981). Cross-correlation analysis of the maintained discharge of rabbit retinal ganglion cells. *Journal of Physiology*, *317*, 29–47.

Brivanlou, I. H., Warland, D. K., & Meister, M. (1998). Mechanisms of concerted firing among retinal ganglion cells. *Neuron*, *20*, 527–539.

Buzsáki, G. (1996). The hippocampo-neocortical dialogue. *Cerebral Cortex*, *6*, 81–92.

Castelo-Branco, M., Neuenschwander, S., & Singer, W. (1998). Synchronization of visual responses between the cortex, LGN and retina in the anesthetized cat. *The Journal of Neuroscience*, *18*, 6395–6410.

Doty, R. W., & Kimura, D. (1963). Oscillatory potentials in the visual system of cats and monkeys. *Journal of Physiology*, *168*, 205–218.

Foerster, M. H., van de Grind, W. A., & Grusser, O. J. (1977). Frequency transfer properties of three distinct types of cat horizontal cells. *Experimental Brain Research*, *29*, 347–366.

Fröhlich, F. W. (1914). Beiträge zur allgemeinen Physiologie der Sinnesorgane. *Psychologische Physiologie Sinnesorganische II. Abteilung Sinnesphysiologische*, *48*, 28–164.

Ghose, G. M., & Freeman, R. D. (1992). Oscillatory discharge in the visual system: does it have a functional role? *Journal of Neurophysiology*, *68*, 1574–1588.

Ghose, G. M., & Freeman, R. D. (1997). Intracortical connections are not required for oscillatory activity in the visual cortex. *Visual Neuroscience*, *14*, 963–979.

Granit, R. (1933). The components of the retinal action potential in mammals and the relation to the discharge in the optic nerve. *Journal of Physiology*, *77*, 207–239.

König, P. (1994). A method for the quantification of synchrony and oscillatory properties of neuronal activity. *Journal of Neuroscience Methods*, *54*, 31–37.

Kuffler, S. W. (1953). Discharge patterns and functional organization of mammalian retina. *Journal of Neurophysiology*, *16*, 37–68.

Laufer, M., & Verzeano, M. (1967). Periodic activity in the visual system of the cat. *Vision Research*, *7*, 215–229.

Leonards, U., & Singer, W. (1998). Two segmentation mechanisms with differential sensitivity for color and luminance contrast. *Vision Research*, *38*, 101–109.

Mastronarde, D. N. (1983). Interactions between ganglion cells in cat retina. *Journal of Neurophysiology*, *49*, 350–365.

Mastronarde, D. N. (1989). Correlated firing of retinal ganglion cells. *Trends in Neurosciences*, *12*, 75–80.

Meister, M., Lagnado, L., & Baylor, D. A. (1995). Concerted signaling by retinal ganglion cells. *Science*, *270*, 1207–1210.

Neuenschwander, S., & Singer, W. (1996). Long-range synchronization of oscillatory light responses in the cat retina and lateral geniculate nucleus. *Nature*, *379*, 728–733.

Peichl, L., & Wässle, H. (1979). Size, scatter and coverage of ganglion cell receptive field centres in the cat retina. *Journal of Physiology*, *291*, 117–141.

Perkel, D. H., Gerstein, G. L., & Moore, G. P. (1967). Neuronal spike trains and stochastic point process. II. Simultaneous spike trains. *Biophysics Journal*, *7*, 419–440.

Przybyszewski, A. W., Lankheet, M. J., & van de Grind, W. A. (1993). Non-linearity and oscillations in X-type ganglion cells of the cat retina. *Vision Research*, *33*, 861–875.

Rager, G., & Singer, W. (1998). The response of cat visual cortex to flicker stimuli of variable frequency. *European Journal of Neuroscience*, *10*, 1856–1877.

Reich, D. S., Victor, J. D., Knight, B. W., Ozaki, T., & Kaplan, E. (1997). Response variability and timing precision of neuronal spike trains in vivo. *Journal of Neurophysiology*, *77*, 2836–2841.

Robson, J. G., & Troy, J. B. (1987). Nature of the maintained discharge of Q, X, and Y retinal ganglion cells of the cat. *Journal of the Optical Society of America* [*A*], *4*, 2301–2307.

Rodieck, R. W. (1967). Maintained activity of cat retinal ganglion cells. *Journal of Neurophysiology*, *30*, 1043–1071.

Sakai, H. M., & Naka, K. I. (1990). Dissection of the neuron network in the catfish inner retina. V. Interactions between NA and NB amacrine cells. *Journal of Neurophysiology*, *63*, 120–130.

Singer, W., Engel, A. K., Kreiter, A. K., Munk, M. H. J., Neuenschwander, S., & Roefselma, P. R. (1997). Neuronal assemblies: necessity, signature and detectability. *Trends in Cognitive Sciences*, *1*, 252–261.

Usher, M., & Donnelly, N. (1998). Visual synchrony affects binding and segmentation in perception. *Nature*, *394*, 179–182.

Wörgötter, F., & Funke, K. (1995). Fine structure analysis of temporal patterns in the light response of cells in the lateral geniculate nucleus of cat. *Visual Neuroscience*, *12*, 469–484.