

# Supporting Information

Galuske et al., Relation between gamma-oscillations.....

## SI Materials and Methods

Eight adult cats bred at the institute's colony were used for this study. All experimental procedures were performed in accordance with the guidelines of the Society for Neuroscience and the European law for the protection of animals and were overseen by a veterinarian. In four animals, the orientation selectivity of cortical neurons was assessed both with optical imaging of intrinsic signals and multi-unit recordings from areas 17 and 18. In two cats, only optical imaging and in two only multi-unit recordings were performed. Anaesthesia was initiated by intramuscular injection of ketamine (10mg/kg) and xylazine (1mg/kg) and maintained after tracheotomy by artificial ventilation with a mixture of N<sub>2</sub>O (70%), O<sub>2</sub> (30%) and halothane (0.8%). In addition, we administered a systemic muscle relaxant (pancuronium, 0.8mg/h) to prevent eye movements. A craniotomy was performed between Horsley-Clarke coordinates A10 and P10 and a metal chamber was fixed on the skull with dental acrylic.

## Optical recording

Optical imaging was performed according to standard procedures as described previously (1) using a slow scan CCD camera system (Ora 2001, Optical Imaging Europe, Munich, Germany) and illumination of the cortex with red light at 620nm wave length. The images were sorted according to stimulation conditions and orientation preference maps were calculated by dividing the sum of the images from the same stimulation condition by the sum of all images ("cocktail blank"). Images were low-pass filtered using a Gaussian filter with a kernel size of 5 pixel. From the different single condition maps, preference maps were calculated for orientation and direction by pixel wise vectorial addition of signals derived from the single condition maps (2). For quantification of response selectivity we calculated the vector strength for each pixel. This value ranges from 0 to 1 and was calculated pixel wise by dividing the length of each individual vector by the sum of all activities. For display, orientation preference was color-coded and orientation selectivity was expressed by adjusting brightness according to the respective vector strength. For visualization images were clipped to their mean +/-1 standard deviation and scaled to 8 bit. For evaluation of changes, orientation preference maps acquired before and after conditioning were subtracted from each other. These procedures were performed using software developed in house on the basis of IDL (Harris Geospatial Solutions, Bloomfield, USA).

For the experiments illustrated in Fig. 5, we conditioned with a whole field grating but for the mapping of changes in the orientation map we stimulated with a small patch of drifting grating with the same parameters as the whole field gratings used for conditioning. The patch was located in the right lower quadrant and subtended only 10°x14° (degrees of visual

angle, its medial border having an eccentricity of 3° from the area centralis (n=2). These coordinates were determined not with RF mapping but with retinoscopy, using a fundus camera).

### **Electrical recording of neural responses**

For electrocorticogram (ECoG) recordings two to four silver ball electrodes were placed at the fringe of the craniotomy over the visual cortex and for multi-unit (MUA) and field potential (LFP) recordings 5 to 16 Teflon-coated platinum wires (25µm diameter) were implanted as floating electrodes under visual control into the visual cortex exposed for optical imaging. This allowed for direct comparison of optical signals with unit and LFP activity. For the electrical activation of the mesencephalic reticular formation (MRF) two concentric bipolar stimulation electrodes were placed bilaterally in the brainstem at Horsley-Clark coordinates A2/H8/L2. The exact position of these electrodes was controlled by testing the facilitating effects of stimulation on evoked cortical potentials that were induced by electrical stimulation of the optic chiasm as reported previously (29). For the assessment of the orientation tuning of units and the topology of orientation maps as well as for the induction of changes in cortical response properties, moving whole field square wave gratings (0.15 cycles/°, moving at 15°/s in both directions orthogonal to the stripe orientation) of four different orientations (0°, 45°, 90°, 135°) were binocularly presented on a 21"-computer screen (refresh rate 100Hz) at a distance of 57cm.

### **Experimental protocol and MRF stimulation**

For the initial assessment of response properties, which lasted ~50 minutes, stimuli were presented in pseudo-randomized order, each stimulus being repeated 64 times. Each stimulus presentation began with a 4 second long adaptation period, in which only the stationary grating was shown. Then the stimulus started to move for 4s and data were collected. Subsequently the screen went blank for 2s and the next stimulus appeared. For the induction of changes in orientation selectivity (conditioning), the same protocol was used except that only one of the 4 stimuli was presented 272 times (50min). In these conditioning sessions MRF was stimulated 100ms before the onset of stimulus movement (1 burst of 60ms duration at 100Hz, 100µs pulse width, 1-2mA). To assess the effects of light stimulation in the absence of MRF stimulation, the same sequence of light stimuli was applied as during conditioning but without MRF stimulation. Anaesthesia was maintained constant with halothane at 0.8% both during mapping and conditioning sessions.

### **Signal filtering, digitization and pre-processing**

Data recorded with the ECoG electrodes were low pass filtered (1-100Hz, 3dB/octave), digitized at 1kHz and analysed according to their frequency content by computing normalized power spectra (Spike2, CED, Cambridge, UK). Signals recorded from the intracortical electrodes were band pass filtered from 1 to 3 kHz for MUA recordings and 0.1 to 200Hz for LFPs. We estimated signal power of the LFP in multiple frequency bands between 1 and 100Hz by applying windowed FFT (Spike2, CED, Cambridge, UK), summing coefficients of 1Hz bins in a 2s window starting 500ms after stimulus motion onset. Frequency bands were defined as follows: delta 1-3.5Hz, theta 3.5-7.5Hz, alpha 7.5-13.5Hz, beta 13.5-20Hz, gamma 20-70Hz, omega 70-120Hz. Interference from the 50Hz line frequency was eliminated by analysing separately a low and a high gamma frequency band at 20-48Hz and 52-70Hz and adding up the respective power values. Spikes were discriminated with a Schmitt-trigger whose threshold was set 2 times above noise amplitude and stored as time stamps. MUA responses to the different moving gratings were evaluated in the same 2s-window starting 500ms after stimulus motion onset and after subtraction of the spontaneous activity of the units before stimulus onset. Subsequently, data were averaged over the 16 stimulus presentations. For the quantification of orientation preference and selectivity, the same vector analysis was applied as for the optical imaging data. All statistical analysis was performed using StatView™ 5.0 (SAS, Cary, USA).

### **SI Supplementary results**

Comparison of changes in the different frequency bands: 1. As reported earlier (3), the power of frequencies below the gamma band, here in particular in the theta band [3.5-7.5Hz] was also modulated by MRF stimulation but in the opposite direction of power changes in the gamma band (20-70Hz) [see Suppl. Fig. 4]. 2. Activity changes in the lower beta band [13.5-19.5Hz], addressed as the “beta 1 band” in the EEG literature, were positively correlated to changes in the low gamma band [20-48Hz] but they were less prominent than changes in the gamma band. In individual conditioning trials the increase in gamma power was most prominent during the first 500ms of the responses while the beta enhancement occurred only during later response phases. A similar trend was observed in the course of the conditioning sessions. Beta power increased over the 50min long conditioning period. This could reflect conditioning dependent changes in network dynamics. As beta synchrony is typically observed when large populations of neurons engage in synchronous oscillations or when synchronisation occurs over larger distances (4) it is conceivable that this beta increase is due to conditioning dependent recruitment of additional neuron populations. 3. Changes in the alpha and delta band were highly variable and not correlated with the effects of conditioning.

## Literature

1. R.A.W. Galuske, K.E. Schmidt, R. Goebel, S.G. Lomber, B.R. Payne, The role of feedback in shaping neural representations in cat visual cortex. *Proc. Natl. Acad. Sci. U.S.A.* 99, 17083-17088 (2002).
2. E. Batschelet. *Circular Statistics in Biology*, Academic Press (1981).
3. S. Herculano-Houzel, M.H. Munk, S. Neuenschwander, W. Singer, Precisely synchronized oscillatory firing patterns require electroencephalographic activation. *J. Neurosci.* 19, 3992-4010 (1999).
4. N. Kopell, G.B. Ermentrout, M.A. Whittington, R.D. Traub, Gamma rhythms and beta rhythms have different synchronization properties. *Proc. Natl. Acad. Sci. U.S.A.* 97, 1867-1872 (2000).

Figure S1:

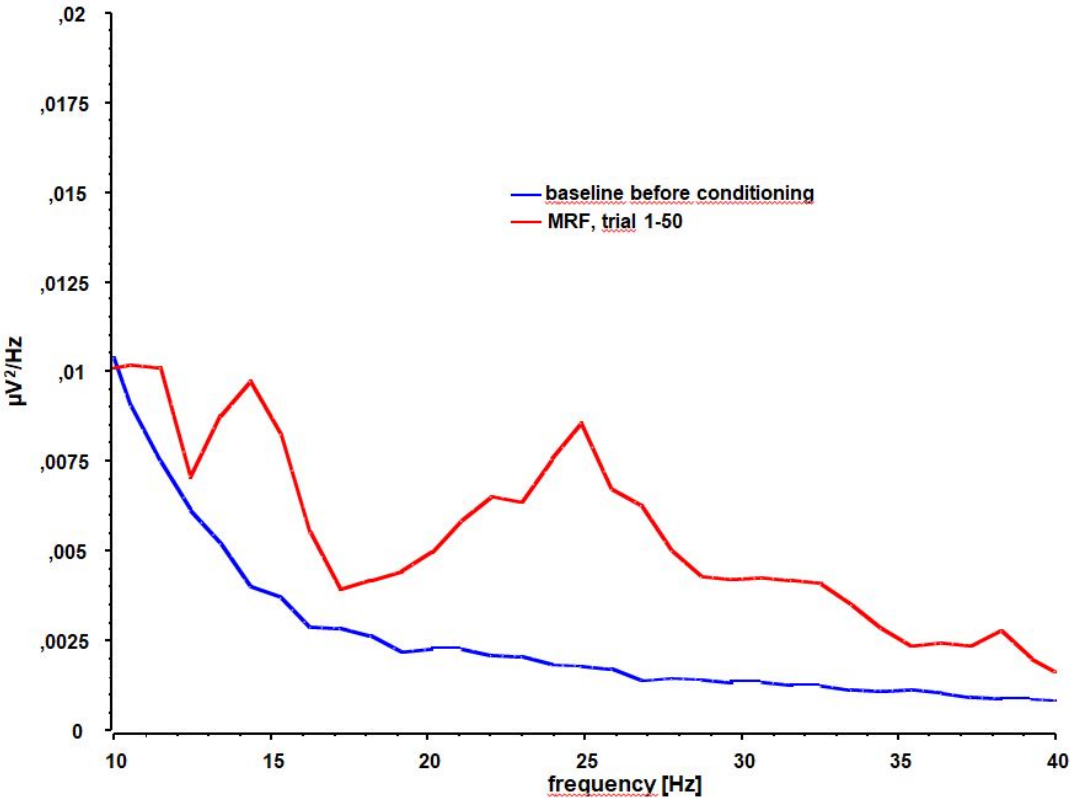
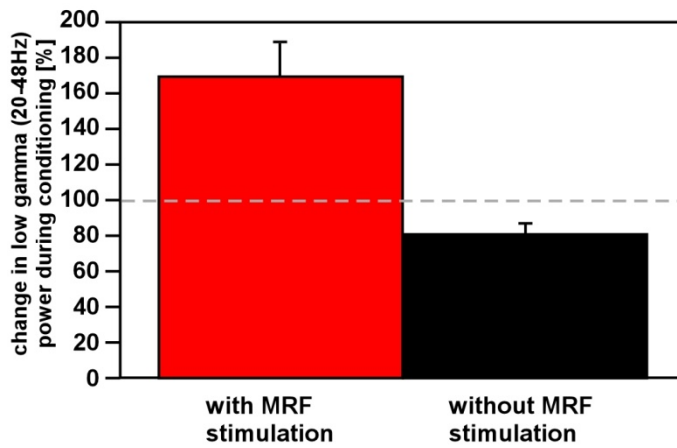


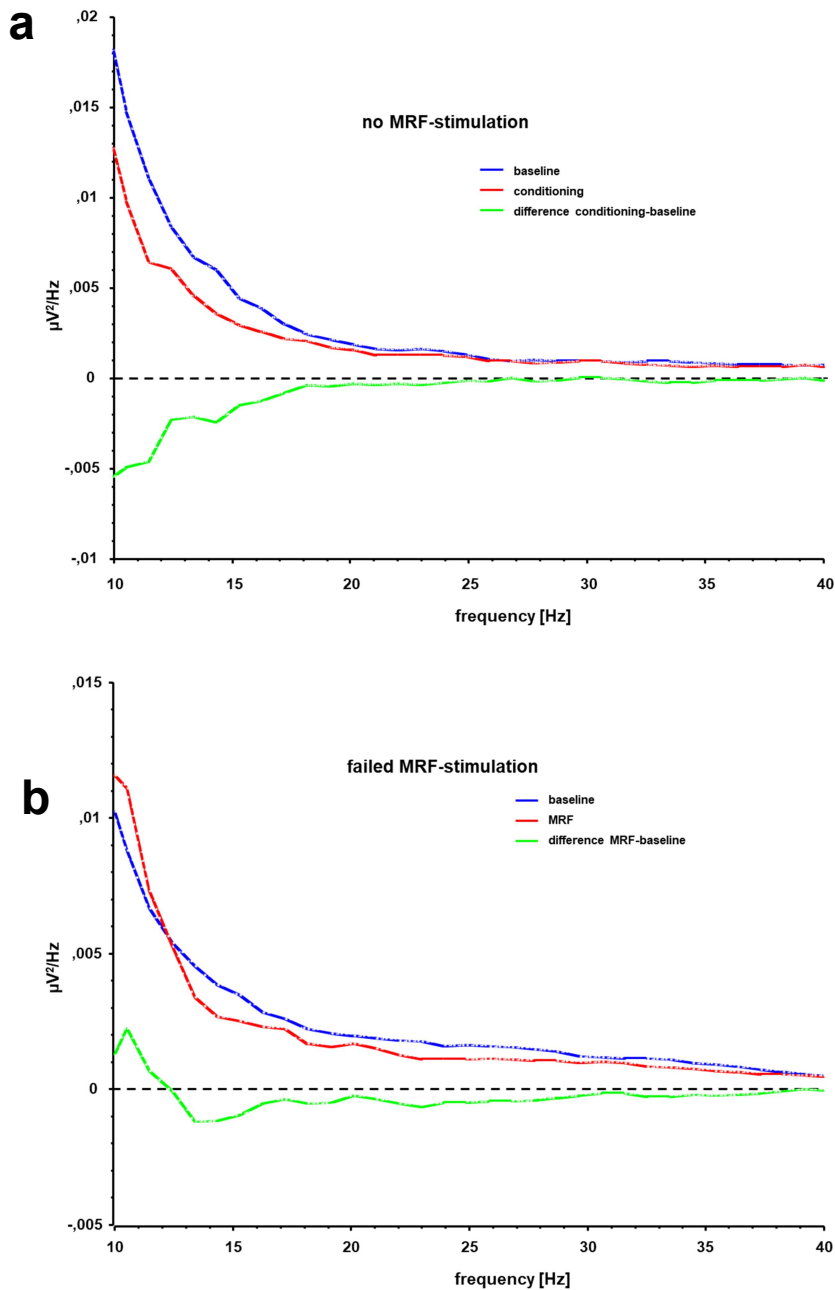
Figure S1: Examples of frequency spectra of ECoG-activity recorded before and during conditioning. Spectra were calculated over a time window of 1100ms starting 200ms after trial start. The plot shows the spectra in the frequency band between 10 and 40Hz averaged over the first 50 conditioning trials. The blue line corresponds to the baseline before conditioning, the red line refers to the conditioning trials with MRF. Frequencies below 10Hz are cut off to enhance resolution in the high frequency band.

**Figure S2:**



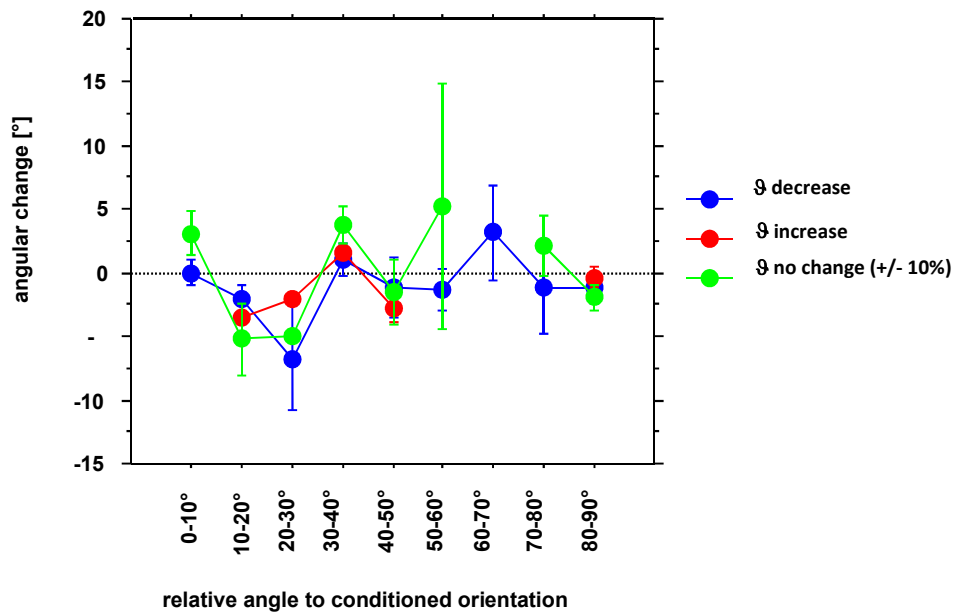
**Figure S2:** Relative changes in the lower  $\gamma$ -band activity (20-48Hz) during conditioning with and without MRF stimulation as compared to the gamma band activity in the preceding mapping session. Note that pairing of visual and MRF stimulation (red) resulted on average in significant increases of gamma power by 70% (n=49,  $\gamma$ -increases in 43 cases, no change in 3 cases,  $\gamma$ -decrease in 3 cases), while without concomitant MRF stimulation (n=8, black) repetitive visual stimulation led to decreases in  $\gamma$ -oscillations up to 36% in 6 out of 8 cases, in 2 cases  $\gamma$ -power remained unchanged. Error bars indicate the standard error of the mean.

Figure S3:



**Figure S3:** Examples of frequency spectra of ECoG-activity recorded before and during conditioning in cases where MRF stimulation failed to enhance gamma power. **a)** Conditioning sessions in which the visual stimuli were not accompanied by electrical activation of MRF. **b)** Recordings of spectra in a case where MRF was paired with visual stimulation but failed to increase  $\gamma$ -power. The blue lines show the baseline spectra before conditioning, the red lines the spectra during conditioning and the green lines the difference between conditioning and baseline. Note that without MRF activation (a) there is a global decrease in power over most frequencies, while with MRF activation frequencies tended to increase in the  $\beta$ -range while no changes occurred in the  $\gamma$ -range (b), suggesting that increases in the  $\gamma$ - rather than in the  $\beta$ -band were responsible for the changes of the orientation maps in the successful conditioning sessions.

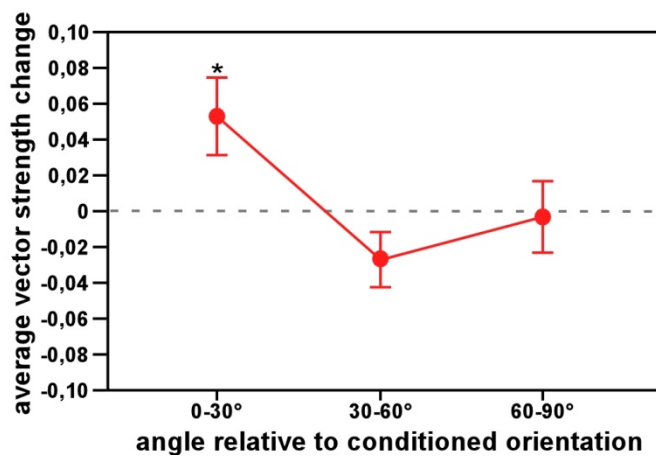
Figure S4:



**Figure S4:** Angular changes in orientation preference in all recorded units (n=171), grouped according to their initial distance to the conditioned orientation and as a function of the power changes in the theta band (3.5–7.5Hz) during conditioning. Red: Power increases of more than 10%; blue: decreases above 10%; green: changes  $<\pm 10\%$ . Error bars indicate the SEMs. When all cells with initial preferences in the range of 5-30° were grouped together, the changes in angular preference were significant ( $p=0.0414$ , one sample sign test) when theta - activity decreased by more than 10% while no significant changes occurred for groups differing by 0-5°, 30-60° and 60-90°, respectively. When theta - power remained unchanged or increased during conditioning, no significant changes occurred in any of the different preference groups.



Figure S5:



**Figure S5:** Changes in vector strength of MUA-responses after conditioning with MRF stimulation for conditioning sessions associated with an increase in gamma power of at least 10% above the level of the previous mapping session. Note the increase in vector strength in units differing by less than 30° from the conditioned orientation (n=44, p=0.035, one sample sign test), the decrease in vector strength in units differing 30-60° from the conditioned orientation (n=60) and the lack of changes in units differing by more than 60° from the conditioned orientation (n=51).