Gamma-band synchronization in the neocortex: novel analysis methods and their application to sensory and motivational systems

Vinck, M.A.

Citation for published version (APA):
CHAPTER 1

General introduction

one man’s noise is another man’s signal - Edward Ng.
1.0 Introduction

The brain is a notoriously complicated organ that evolved to receive sensory data, model the state of objects in the world using this sensory data, and transform goals (which need to be developed based on interactions of the organism having innate behavioral predispositions with the environment) into actions based upon this model. To model the state of objects in the world, the brain possesses an exquisite apparatus of sensors, and it contains billions of specialized neurons devoted to perform pattern recognition on the received sensory data, and control actions based on internal representations. The brain distinguishes itself from other complex phenomena in nature in that a description of its emergent, complex behavior cannot be derived from the description of only one of its elements: Not only are its elements (neurons) highly specialized in function, but also, information can be represented by large-scale patterns of activity (Wilson & McNaughton, 1994; Buzsáki, 2006).

Advancement of our understanding of the brain relies on 1.) measuring the activity of many neurons simultaneously, given a controlled set of sensory inputs to the brain, 2.) identifying the type of neurons we are recording from (i.e., specific inhibitory interneuron or pyramidal cell types), and 3.) estimating the brain’s algorithmic (sensory, motor, cognitive) operations and dynamics based on these measurements. However, the nature of neuroscience data poses a large statistical problem to its observers. First, only very few measurements are available for a given stimulus or behavioral state (Chapters 2, 3, 5), which leads to a curse of dimensionality when we increase the scale of our recordings. For example, to estimate a functional interaction matrix for $10^6$ neurons, we need at least $10^{12}$ observations. However, only tens or hundreds of trials are typically available in neurophysiological experiments. Second, neuroscience data is very noisy, e.g. because electric currents spread to multiple measurement sensors (Chapters 3 and 4), but also because electric currents generated by the brain are often small compared to environment noise.

To further our understanding of the brain, both technological and statistical tools have improved significantly over the course of the past decades. Early functional studies of single neurons used only one or very few electrodes to map the responses of neurons to certain stimuli or actions. While these recordings provided invaluable information about the sensory features represented by single neurons, they did not address the question how information is encoded by a population of cells, nor did they reveal how information flows between different brain areas. The past two decades however have seen an increased interest in ensemble measurements. Some labs now use multi-area electrode arrays that allow for many parallel single unit measurements across brain areas (Salazar et al., 2012), or large-scale grids for intracranial EEG measurements (Rubehn et al., 2009; Bosman et al., 2012). These techniques have revealed invaluable insights into the functioning of the brain. For example, they have shown that highly specific activity patterns occur (and re-occur during sleep) in cortical cell assemblies (Wilson & McNaughton, 1994; Dragoi & Buzsaki, 2006; Lansink et al., 2009), and have suggested that the information flow between areas occurs in a frequency-specific and attention-dependent manner (Gregoriou et al., 2009; Bosman et al., 2012; Buschman & Miller, 2007). In Chapter 6, we will present data obtained with a new recording device called the ‘quad-drive’, which allows parallel measurement of well-isolated single units across four multiple brain areas.

Together with the increase in the scale of recordings, neuroscience has seen application of increasingly sophisticated statistical and signal processing tools to find patterns in brain data,
which is often inspired by the increased size of recording datasets, but also by the noisiness of brain data. In particular, neuroscience has seen increased application of sophisticated spectral estimation techniques (Mitra & Pesaran, 1999), advanced coherence metrics (Nolte et al., 2004a, 2004b), and improved information measures (Nemenman et al., 2004, 2001; Panzeri & Treves, 1996; Panzeri et al., 2007; Vinck et al., 2012b) (Chapter 5), and techniques to estimate causal links between neurons (Nolte et al., 2008; Dhamala et al., 2008; Haufe et al., 2012a) (Chapter 4).

In the methodological part of this thesis, we are primarily interested in the development of statistics that allow us to infer statistical relationships between random time series or variables. In particular, we are interested in (1) bivariate measures of phase-coupling or coherence, (2) bivariate measures of causality between time series, and (3) bivariate measures of information, i.e. the amount of reduction in uncertainty that knowledge of one random variable provides about another random variable. The concept of phase-coupling refers to the consistency of phase relationships between two signals. For example, if the relative phase between two signals at frequency \( f \) in trial \( k \) (\( K \) trials) is denoted \( \theta_k(f) \), then phase-coupling can be measured using the resultant vector length, 

\[
R = \frac{1}{K} \sum_{k=1}^{K} \exp(i\theta_k(f))
\]

Coherence can be understood as quantifying the amount of linear dependence in the frequency domain. More specifically, coherence is defined as the ratio of cross-spectral density over the square-root product of power spectral densities,

\[
C = \frac{S_{12}(f)}{\sqrt{S_{11}(f)S_{22}(f)}}
\]

where \( S_{12} \) is the cross-spectral density and \( S_{jj} \) the power spectral density of \( x_j \). Coherence, i.e. \( |C| \), can be understood as the extent to which the spectrum from one signal (at frequency \( f \)) can predict the spectrum of another signal. The phase of the coherency indicates the average phase-relationship between signals. An important relationship exists between coherency and the extent to which signals can linearly predict each other in the time domain. This relationship appears in the context of constructing an optimal noncausal (Wiener) filter. Suppose we have two signals \( x_1 \) and \( x_2 \) that are wide sensory stationary. Suppose we seek a filter kernel \( h(\tau) \) that minimizes the error \( E\{(x_2(t) - \hat{x}_2(t))^2\} \), with \( \hat{x}_2(t) = \sum_{\tau=+\infty}^{\tau=-\infty} h(\tau)x_1(t-\tau) \). The optimal filter kernel is given by \( h(\tau) \) with Fourier Transform \( H(\omega) = S_{21}(\omega)/S_{11}(\omega) \), such that the minimum error equals \( \epsilon_{\text{min}} = \int S_{22}(\omega)\left(1 - |C(\omega)|^2\right)d\omega \), where \( |C(\omega)|^2 \) plays a similar role as in linear regression analysis.

While coherence merely indicates whether one signal predicts the other signal, Granger-causality makes two critical extensions: 1) It indicates whether a first signal can predict a second signal after conditioning on the information that is already available from the second signal. Two signals can be coherent, but the first signal may not improve prediction of the second signal after predictions of the second signal by its own past are incorporated; 2) While linear prediction in the coherence framework incorporates both predictions from past to future and future to past values, Granger-causality merely considers the predictions of future values by a linear combination of past values. This allows one to derive inferences and hypotheses about causality from observations of time series (given some assumptions, Chapter 4). 3) While coherence is a symmetric function, Granger-causality is asymmetric, in the sense that the past values of the) first signal can predict the (future values of the) second signal, but not
Finally, Shannon Information Theory (Shannon, 1948) provides a powerful mathematical framework to capture relationships between random variables. The cornerstone of Information Theory is the concept of entropy, which can be understood as measuring uncertainty (for mathematical definition, see Chapter 5). Mutual information ($I$) can then be understood as the extent to which knowledge of one random variable ($X$) reduces uncertainty about another random variable ($Y$). Mutual information is a symmetric function, i.e. $I(X; Y) = I(Y; X)$. An advantage of the mutual information function is that it can capture all types of relationships between random variables, i.e. it does not make any assumptions about the probability distributions of $X$ and $Y$ and the nature (linear, non-linear) of their statistical association. In theory, it is therefore the most generic measure of association between two random variables. There has been a great deal of interest in using Information Theory to quantify the degree to which activation patterns of neuronal ensembles provide information about the features of sensory stimuli (Panzeri et al., 2007; Rieke et al., 1999). Yet, the mutual information function was originally developed for discrete variables whose probability distributions are a priori known, and its direct estimate suffers from very strong statistical bias, which makes application to neural data difficult (Panzeri et al., 2007; Nemenman et al., 2004).

1.1 Mechanisms and functions of gamma-band synchronization

*Information is a difference that makes a difference* - Gregory Bateson

We proceed by providing a broad introduction to the mechanisms and functions of gamma-band synchronization, as it is the main rhythmic process on which we are focused in this thesis. Following that, background information for the various areas from which we recorded will be given. Finally, we state our main aims and questions.

This subsection gives background knowledge for Chapters 6 to 8. The brain contains billions of excitatory (E) neurons that use the neurotransmitter glutamate and inhibitory (I) interneurons that use the neurotransmitter GABA. Interactions between E and I cells are a fundamental brain circuit mechanism underlying complex, precisely organized behaviors. Through these E-I interactions, collective oscillatory phenomena emerge that regulate the timing of action potentials (i.e., ‘spikes’), creating the brain’s internal clock mechanism. These oscillatory phenomena cover a wide range of frequencies, e.g. delta (1-4 Hz), alpha (8-12 Hz), beta (12-30 Hz) and gamma (30-90 Hz) oscillations. Oscillations in these separate frequency bands are associated with a rich repertoire of circuit mechanisms (Buzsáki & Draguhn, 2004; Wang, 2010). Here, we focus on one particular rhythm, gamma band oscillations, which tends to be observed when animals are in an active, attentive behavioral state. Many early human and animal EEG and LFP studies have revealed the presence of gamma band oscillations (Jasper & Andrews, 1938; Bressler & Freeman, 1980; Adrian, 1941; Boeijinga & Lopes da Silva, 1988; Freeman, 1959). The discovery of gamma-band synchronization in spiking output of the visual cortex (Gray et al., 1989) and the hippocampus (Bragin et al., 1995) has sparked intense research on its mechanic underpinnings, and its function

---

1 This section was partially adapted from the published book chapter Vinck et al. (2013)
in cortical computation. After many decades of research, the emerging picture is that gamma-band synchronization may play an important role in both information coding (König et al., 1995; Fries et al., 2007) and selective information transmission (Fries, 2005). In this section, we review evidence showing that: (i) A highly specialized machinery exists to generate cortical gamma-band oscillations; (ii) Gamma-band synchronization is a ubiquitous phenomenon in the cortex; (iii) Gamma-band synchronization likely bears strong network consequences, due to feedforward coincidence detection (Abeles, 1982), coherent phase-coupling across structures (Fries, 2005), and spike-timing-dependent-plasticity (Sejnowski & Paulsen, 2006; Fell & Axmacher, 2011; Jensen et al., 2007; Jutras & Buffalo, 2010); (iv) Gamma synchronization may be involved in selective attention, and long-term memory formation (Jensen et al., 2007; Jutras & Buffalo, 2010; Fries et al., 2001b; Sejnowski & Paulsen, 2006); (v) Gamma-band oscillations may serve as a temporal reference frame, allowing spike phases to convey stimulus information (Fries et al., 2007); (vi) Gamma-band synchronization may be an important determinant of the rate code itself (Womelsdorf et al., 2012).

### 1.1.1 Generation of gamma-band oscillations

The two main types of neurons in the mammalian brain are excitatory pyramidal cells and GABAergic interneurons. Interactions between excitation and inhibition are a critical determinant of patterns of neural firing, yet inhibition is provided by a rich diversity of inhibitory subtypes, characterized by different physiology, synaptic targets, and molecular markers. The division of labor among different inhibitory interneurons is still poorly understood (Monyer & Markram, 2004; Jonas et al., 2004; Whittington & Traub, 2003; Buzsáki, 2006; Gentet, 2012). There is a broad consensus that PV+ (parvalbumin expressing), FS (fast spiking) basket cells - providing perisomatic inhibition to pyramidal cells - play a pivotal role in generating cortical gamma-band synchronization (Bartos et al., 2007; Tiesinga & Sejnowski, 2009; Cardin et al., 2009; Sohal et al., 2009; Gulyas et al., 2010; Wang, 2010; Whittington et al., 2011).

The generation of gamma-band oscillations does not depend on remote pacemaker cell input, but is supported by the intrinsic components of the cortical microcircuit, i.e. excitatory and inhibitory cells (Cardin et al., 2009; Wang, 2010). Therefore, gamma-band oscillations may be a fundamental processing mode of cortical circuits (Fries, 2009). The gamma processing mode is triggered by sufficient excitatory input from pyramidal cells to FS basket cells, since the latter do not fire tonically without receiving recurrent excitatory inputs from pyramidal cells (of course, the pyramidal cells in turn need external input as well). FS basket cells provide perisomatic inhibitory inputs to pyramidal cells, which puts them in an ideal position to gate excitatory inputs from all dendritic compartments (Freund & Katona, 2007), and FS basket cells can strongly entrain pyramidal spiking activity (Lytton & Sejnowski, 1991; Cobb et al., 1995; Hasenstaub et al., 2005). Furthermore, FS basket cells are resonators - in the sense of being particularly sensitive to synaptic inputs or activation at certain frequencies (Hasenstaub et al., 2005; Pike et al., 2000; Cardin et al., 2009) - at gamma frequencies and are capable of firing more than one spike per gamma cycle, given that sufficient excitatory drive is provided (Pike et al., 2000; Klausberger et al., 2003; Hasenstaub et al., 2005; Cardin et al., 2009; Gulyas et al., 2010).

*In vitro* pharmacological studies show that gamma-band oscillations in the hippocampus can be induced by the cholinergic agonist carbachol (Fisahn et al., 1998), by agonists of kainate receptors (Fisahn et al., 2004), or by metabotropic glutamate receptors (Whitting-
ton et al., 1995). In all cases, gamma-band oscillations are blocked by the GABAA receptor antagonist bicuculline. This supports a crucial role of inhibition in gamma rhythmogenesis, but does not exclude a potential role of other inhibitory interneuron classes. To directly test whether FS basket cells play a causal role in gamma rhythmogenesis, Cardin et al. (2009) and Sohal et al. (2009) used optogenetic tools to precisely control the firing of FS, PV+ interneurons. Activation of FS, PV+ interneurons through channelrhodopsin with broadband or gamma-rhythmic trains of light pulses strongly enhanced gamma-band oscillations in the LFP (Local Field Potential). Thus, upon activation, FS, PV+ interneurons rhythmically entrained pyramidal cells (Cardin et al., 2009), showing that their activation (both using narrow-band and broad-band activation patterns) is sufficient for the emergence of gamma-band oscillations. When gamma-band oscillations were induced by (optogenetic) activation of pyramidal cells, they were suppressed by the inhibition of FS, PV+ interneurons (Sohal et al., 2009), showing that their activation is not only sufficient, but also required for the emergence of gamma-band oscillations. However, these optogenetic studies do not exclude a potential role of FS, PV+ axo-axonic cells - i.e. cells that are also co-expressing Parvalbumin, but that inhibit the axon initial segment of pyramidal cells (Gentet, 2012). To address this issue, Gulyas et al. (2010) used opiates to selectively reduce GABAA release from FS basket cell terminals, leaving GABAA release from axo-axonic cells unaffected. This strongly reduced the carbachol-induced in vitro gamma-band oscillations, suggesting that the activation of FS basket cells is required, whereas the activation of axo-axonic cells is not sufficient for the generation of gamma-band oscillations.

Although a pivotal role of FS basket cells is implied by these findings, it remains difficult to dissect the precise dynamics of the gamma rhythm, since the network dynamics are not easily separable into cause and consequence. In particular, there is substantial debate about whether the precise timing of pyramidal cell firing plays an essential role in gamma rhythmogenesis (Bartos et al., 2007; Morita et al., 2008; Tiesinga & Sejnowski, 2009; Cardin et al., 2009; Wulff et al., 2009; Whittington et al., 2011). In the ING (Interneuron Network Gamma) model, the crucial factor producing gamma-band synchronization is the mutual inhibition between basket cells (that are activated by either synchronous or asynchronous pyramidal cell input) (Whittington et al., 1995; Wang & Buzsaki, 1996; Vida et al., 2006; Bartos et al., 2007): Upon activation, the FS basket cell network propels towards an attractor state where basket cells escape each other’s mutual inhibition through synchronization (Whittington et al., 1995; Wang & Buzsaki, 1996; Bartos et al., 2007). The gamma-synchronized FS basket cell network activity then rhythmically entrains pyramidal cells. In this scheme, the timing of pyramidal cells’ firing is a mere consequence of rhythmic inhibition, and the critical parameter determining the gamma frequency is the decay of GABAA receptor mediated inhibition (Whittington et al., 1995). Conversely, the timing of pyramidal cell spiking plays a central role in the PING (Pyramidal Interneuron Network Gamma) model. When pyramidal cells recover from inhibition, they cause an increase in feedback inhibition. This causes a decrease in pyramidal cell activity in turn, leading to a decrease in inhibition, until the pyramidal cells recover (Wilson & Cowan, 1972; Leung, 1982; Eeckman & Freeman, 1990; Hansel & Mato, 2003).

Both types of gamma-band oscillations can be induced in the cortex. Gamma-band oscillations that are induced in the CA1 and CA3 regions of the hippocampus by kainate induction or metabotropic glutamate receptor agonists are maintained in the presence of AMPA receptor antagonists (Traub et al., 1996; Whittington et al., 1996), consistent with ING-type models.
Gamma oscillations induced by cholinergic agonist activation are inhibited by AMPA receptor antagonists in both hippocampus (Fisahn et al., 1998) and neocortex (Buhl et al., 1998), consistent with PING-type models. However, converging evidence tentatively supports the conclusion that the typical gamma-band oscillations that have been observed in for example the hippocampus (Bragin et al., 1995; Csicsvari et al., 2003) and sensory cortices (Gray et al., 1989; Fries et al., 2001b) arise from a precise temporal interplay between excitation and inhibition: (i) PING-models predict a characteristic delay of several milliseconds between pyramidal cell activity and FS basket cell activity (Eeckman & Freeman, 1990). Consistent with this, extracellular in vivo recordings have shown that pyramidal cell activity has a gamma phase-lead of a few milliseconds over putative FS basket cell activity (Csicsvari et al., 2003; Hasenstaub et al., 2005; Tukker et al., 2007; Womelsdorf et al., 2008; van Wingerden et al., 2010a). This phase-relation is an important motif of the cortical microcircuit that also exists in the absence of gamma-band synchronization (Okun & Lampl, 2008). ING models on the other hand predict that pyramidal cells fire in phase with FS basket cells, since for both cell types the probability of firing is governed by the timing of inhibition (Traub et al., 1996). (ii) A problem with ING models is the robustness against heterogeneity in excitatory drive (Wang & Buzsaki, 1996). Strong, shunting interneuron-interneuron inhibition can make ING networks robust against such heterogeneities (Vida et al., 2006; Bartos et al., 2007). However, as argued by (Freund & Katona, 2007), the required depolarization induced by GABAa inhibition may be found only in the dentate gyrus, where the GABAa reversal potential is particularly close to action potential threshold. The fast kinetics of the mutual FS basket cell inhibition (Bartos et al., 2002) may rather support the genesis of fast oscillations (>100 Hz) instead of gamma-band oscillations (Brunel & Wang, 2003; Geisler et al., 2005). (iii) Cardin et al. (2009) induced gamma-band oscillations in vivo through depolarization of FS basket cells by activating channelrhodopsin with light pulses. Blocking AMPA and NMDA (see Box 1.1) receptors abolished gamma-band oscillations, even though the FS basket cells continued to receive external, channelrhodopsin mediated excitatory drive. Furthermore, pyramidal cells fired with a characteristic phase-lead with respect to FS basket cells Cardin et al. (2009), as predicted by PING-models. (iv) Genetically impairing the GABAa receptor in hippocampal PV+ interneurons, which abolishes the mutual inhibition mechanism, did not significantly affect the strength and frequency of gamma-band synchronization in awake mice (Wulff et al., 2009).

Box 1.1. In this box, we give a brief overview of the α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid receptor (AMPAR) and N-methyl-D-aspartate receptor (NMDAR). Both receptors are involved in excitatory synaptic transmission using the neurotransmitter glutamate, which is the most abundant neurotransmitter for excitatory synaptic transmission in the nervous system (Meldrum, 2000). The AMPA receptor is an ionotropic transmembrane receptor mediating relatively fast excitatory currents and permeable to sodium and potassium (Hestrin, 1992; Meldrum, 2000; Geiger et al., 1997; Kandel et al., 2000). The NMDA receptor is a ionotropic transmembrane receptor that mediates relatively slow excitatory currents. A unique property of the NMDA receptor is that its activation is both dependent on the binding of the ligand glutamate (besides the co-agonist glycine or d-serine), and the voltage of the transmembrane potential. If the transmembrane potential becomes more positive, then the Mg2+ block is released that
blocks the ion channels from the outside, making them permeable to Ca\(^{2+}\). The influx of Ca\(^{2+}\) in turns regulates synaptic plasticity e.g. through upregulation of the postsynaptic density of AMPARs (Meldrum, 2000; Kandel et al., 2000). This property plays an important role in regulating long-term potentiation (LTP), which refers to a long-lasting synaptic potentiation resulting from a particular combination of pre- and postsynaptic activity. (Note that LTP is not necessarily NMDAR-dependent). The NMDAR supports a form of Hebbian LTP, in the sense that presynaptic activation in combination with postsynaptic depolarization is required (Kandel et al., 2000; Hebb, 1949). In addition, long-term depression has been demonstrated in several forms. Prominent among these is a type of LTD that is induced by low-frequent stimulation and is NMDAR dependent. The NMDAR also supports a composite type of LTP-LTD called spike-timing-dependent-plasticity (STDP), where the synaptic strength is adjusted based on the precise timing of pre- and postsynaptic spikes (Markram et al., 1997; Dan & Poo, 1992). If the postsynaptic spikes occur after the presynaptic spikes, then a back-propagating action potential causes a larger postsynaptic calcium transient, giving rise to LTP. Long-term depression occurs when the postsynaptic neuron fires before the presynaptic neurons (Markram et al., 1997; Dan & Poo, 1992). A role of gamma-oscillations and coherence in modulating synaptic plasticity can be suggested based on the STDP rule, as they can precisely coordinate the timing of pre- and postsynaptic neurons (Sejnowski & Paulsen, 2006; Jensen et al., 2007; Fell & Axmacher, 2011; Jutras & Buffalo, 2010; Cassenaer & Laurent, 2007).

### 1.1.2 Incidence of gamma-band synchronization in the nervous system

If gamma-band oscillations emerge from an interplay between two basic components of the cortical microcircuit, pyramidal cells and FS basket cells, then it is to be expected that it is a widespread cortical phenomenon. Indeed, gamma-band oscillations have been documented in many areas and species, and under many behavioral conditions. Intense research on gamma-band oscillations commenced with the finding that neurons in primary visual cortex engage in strong gamma-band synchronization in response to visual stimulation (Gray et al., 1989). Since then, many labs have revealed gamma-band oscillations in anesthetized cat and monkey visual cortex (Livingstone, 1996; Yu & Ferster, 2010) and awake cat, monkey and human visual cortex (Fries et al., 1997, 2001b; Friedman-Hill et al., 2000; Maldonado et al., 2000; Rols et al., 2001; Hoogenboom et al., 2006; Womelsdorf et al., 2007; Gieselmann & Thiele, 2008; Chalk et al., 2010; Lima et al., 2010; Ray & Maunsell, 2010; Vinck et al., 2010a). Gamma-band synchronization in visual cortex is typically induced by the presentation of moving bars or gratings, but has also been induced by viewing stationary squares (Rols et al., 2001), smoothly changing shapes (Taylor et al., 2005) and by the free-viewing exploration of a stationary array of objects (Bichot et al., 2005). Gamma-band synchronization in visual cortex is stronger for salient stimuli, increasing with stimulus size (Gieselmann & Thiele, 2008), contrast (Henrie & Shapley, 2005) and spatial integrity/coherence (Zhou et al., 2008; Lima et al., 2010). Also, gamma-band synchronization in visual cortex is especially strong during active, awake states (Munk et al., 1996; Herculano-Houzel et al., 1999; Fries et al., 2001b; Rodriguez et al., 2004; Taylor et al., 2005; Womelsdorf et al., 2006; Gregoriou...
et al., 2009). In sum, while visual activation is required for the emergence of gamma-band synchronization in visual cortex, it is not a sufficient condition for strong gamma-band oscillations to emerge. Further, gamma-band synchronization in visual cortex (Buffalo et al., 2011) and somatosensory cortex (Roopun et al., 2006) is particularly strong in the superficial layers, whereas beta-band synchronization is particularly strong in infragranular layers. The dependence of gamma-band oscillations on these variables may explain why a minority of studies has failed to detect gamma-band synchronization in primary visual cortex (Lamme & Spekreijse, 1998; Nowak et al., 1999; Montemurro et al., 2008).

The gamma rhythm is also very prominent in rat and mouse hippocampal formation (Bragin et al., 1995; Csicsvari et al., 2003; Colgin et al., 2009). Given appropriate inputs, it can also be induced in primate auditory cortex (Brosch et al., 2002), human somatosensory cortex (Bauer et al., 2006), rat barrel cortex (Cardin et al., 2009) (see Chapter 6 in this thesis), the rat and locust olfactory system (Eeckman & Freeman, 1990; Wehr & Laurent, 1996), primate parietal cortex (Pesaran et al., 2002; Buschman & Miller, 2007; Medendorp et al., 2007), several nodes of the primate and rodent frontal cortex (Buschman & Miller, 2007; Gregoriou et al., 2009; Siegel et al., 2009; Sohal et al., 2009; Canolty et al., 2010; Sigurdsson et al., 2010; van Wingerden et al., 2010b) (see Chapter 7 in this thesis), rodent striatum (van der Meer & Redish, 2009; Kalenscher et al., 2010) and cat amygdala (Popescu et al., 2009).

Long-range gamma-band synchronization has been documented between spinal cord and motor cortex (Brown et al., 1998; Schoffelen et al., 2005), between different nodes of the visual system (Engel et al., 1991a,b; Womelsdorf et al., 2007; Bosman et al., 2012; Grothe et al., 2012; Jia et al., 2013; Roberts et al., 2013), visual cortex and parietal cortex (Roelfsema et al., 1997; von Stein et al., 2000; Saalmann et al., 2007; Bosman et al., 2012), V4 and FEF (Frontal Eye Fields) (Gregoriou et al., 2009), LIP (Lateral Intraparietal cortex) and FEF (Frontal Eye Fields) (Buschman & Miller, 2007), amygdala and striatum (Popescu et al., 2009), and hippocampus and prefrontal cortex (Sigurdsson et al., 2010).

It is relatively unknown what mechanisms support long-range gamma-band coherence. Long-range gamma-band synchronization at a phase delay that mirrors the synaptic delay (Gregoriou et al., 2009) may be supported by simple rhythmic entrainment (Fries, 2005; Börgers & Kopell, 2008; Gielen et al., 2010), in which pyramidal cells reset the phase of FS basket cell activity through long-range excitatory connections. Such a scheme is akin to a PING-type dynamics, in which local pyramidal cells control the timing of local FS basket cells. A more difficult problem is the generation of long-range, zero-lag rhythmic synchronization in the presence of substantial synaptic delays, as in Engel et al. (1991a). Computational models suggest that this type of long-range synchronization may be supported by mutual, long-range excitatory connections causing doublet spikes in FS basket cells (Traub et al., 1996; Kopell et al., 2000), a scheme which is stable up to conduction delays of 8-10 ms.

### 1.1.3 Consequences of rhythmic neuronal synchronization: Feedforward coincidence detection

Demonstrating that gamma-band synchronization plays a fundamental role in neural computation rests on two pillars. First, it needs to be demonstrated that rhythmic synchronization bears consequences in terms of neuronal interactions. Second, it needs to be demonstrated
that gamma-band synchronization varies across experimental conditions in a meaningful way, supporting the computations that the brain needs to perform (Salinas & Sejnowski, 2001; Fries, 2009). In the words of Gregory Bateson, for information to be contained in gamma-band synchronization, we need to show ‘a difference that makes a difference’. In this section, we will address the question whether differences in rhythmic synchronization indeed make a difference in terms of neuronal interactions.

The mechanism of feedforward coincidence detection entails that spikes have more impact on a postsynaptic target neuron if they coincide within a narrow temporal window (Abeles, 1982; Bernander et al., 1991; Softky, 1994; König et al., 1996; Azouz & Gray, 2000; Galarreta & Hestrin, 2001; Salinas & Sejnowski, 2001). Coincident firing implies that there exists a peak near $t = 0$ in the cross-correlogram between spike trains, i.e. zero-lag synchronization. If spiking activity is asynchronous, then the cross-correlogram between spike trains will be flat. The general concept of synchronization should be distinguished from the more specific concept of rhythmic or oscillatory synchronization. Zero-lag synchronization merely implies coincident firing in the time domain, and can occur without any oscillatory preference. However, coincident firing typically arises as a consequence of coherent oscillatory activity, which focuses spikes within a narrow temporal window. Zero-lag synchronization can also be caused by common inputs or co-variation of stimulus-locked rate modulations (Brody, 1998).

The question whether neurons are coincidence detectors or temporal integrators should not be framed as an absolute one. Obviously, excessive temporal integration would limit the brain’s capacity to track fast changes in sensory inputs. Enhanced synchrony between presynaptic inputs may only increase the firing rate of a postsynaptic target up to a certain point (Murthy & Fetz, 1994; Bernander et al., 1994): if a packet of coincident spikes causes the postsynaptic neuron to fire, then the surplus spikes that were not needed for threshold potential crossing are lost in the postsynaptic neuron’s refractory period. Consequently, excessive coincident firing in the presynaptic population causes, ceteris paribus, low firing rates in postsynaptic cells.

Viewpoints on coincidence detection have changed substantially during the past decade, and effective temporal integration times may in fact be much shorter than was previously appreciated (Shadlen & Newsome, 1994; Koch et al., 1996). First, the classic idea is that feedforward coincidence detection depends on the membrane time constant, which co-determines the decay rate of EPSPs (Excitatory Postsynaptic Potentials). Shadlen & Newsome (1994) argued that integration times are effectively limited by membrane time-constants of 8-20 ms, values that are typically obtained from in vitro slice recordings. Thus, coincident firing on a faster time-scale would not significantly impact neuronal interactions, and cause many presynaptic inputs to be lost in the postsynaptic neuron’s refractory period (Shadlen & Newsome, 1994). Consequently, gamma-band synchronization of spiking activity with a cycle duration of 12-30 ms would not enhance impact on a postsynaptic target, since spikes occurring at preferred and non-preferred gamma phases would effectively be temporally integrated.

However, membrane time constants depend on both neuron type and cortical state, and are shorter when the slice is at physiological temperatures. When an animal is in an awake state, neurons are bombarded by synaptic background activity that is composed of balanced excitation and inhibition. This synaptic background activity causes the membrane potential to be closer to threshold than the resting potential is, and induces a several fold increase in the membrane leak conductance. The membrane leak conductance is inversely proportional
to the membrane time constant. It follows that the several fold increase in membrane conductance causes a proportional decrease in the membrane time constant in comparison to in vitro slice recordings. This decrease in the membrane time constant corresponds to a proportional decrease in the temporal window of integration (Bernander et al., 1991; Borg-Graham et al., 1998; Hirsch et al., 1998; Destexhe et al., 2003; Kuhn et al., 2004; Leger et al., 2005; Hasenstaub et al., 2007; Kumar et al., 2008). In addition, inhibitory interneurons operate more like coincidence detectors than pyramidal cells, since they have shorter membrane time constants, and respond with rapidly decaying EPSPs to pyramidal cell inputs (Galarreta & Hestrin, 2001; Geiger et al., 1997; Cardin et al., 2007, 2010).

Second, several studies have shown that spiking activity is not merely determined by the level of depolarization of the membrane potential. In addition, a strong determinant of firing is the first derivative of the membrane potential (Azouz & Gray, 2000; Henze & Buzsaki, 2001). Rapid depolarizations of the membrane potential Vm lower the action potential threshold. Computational models show that this effect is caused by the activation of voltage-gated sodium channels (Azouz & Gray, 2000; Cardin et al., 2010; Farries et al., 2010). On the other hand, while slow membrane potential depolarizations increase the probability that fast fluctuations cause a crossing of the action potential threshold, this effect is to some extent counteracted by the inactivation of sodium channels, which increases action potential threshold, and by an increase in membrane leak conductance, which decreases the amplitude and duration of incoming EPSPs (Petersen et al., 2003; Kuhn et al., 2004; Leger et al., 2005). In fact, a much stronger correlation of firing rate with the membrane potential derivative dVm/dt than with the membrane potential Vm has been observed in vivo (Azouz & Gray, 2000, 2003). Consequently, orientation tuning in V1 is mainly driven by tuning of fast fluctuations of the membrane potential (20-80 Hz), rather than by tuning of the mean or slow fluctuations of the membrane potential Vm (Azouz & Gray, 2003). The integration times based on dVm/dt are on the order of the time-course of the capacitive membrane current, which has a time-constant that is even faster than the excitatory postsynaptic current at resting level (about -70 mV). The latter time-constant is on the order of a few milliseconds for the AMPA receptor on pyramidal cells (Hestrin, 1992) and there is a sub-millisecond decay constant for the AMPA receptor on FS basket cells (Geiger et al., 1997; Galarreta & Hestrin, 2001). Thus, temporal integration of synaptic inputs separated by a longer time delay occurs on the basis of Vm depolarization only: The positive rates of depolarization are not added across inputs separated by more than a few milliseconds, even though the levels of depolarization are. Given a neuronal refractory period of several milliseconds, it follows that multiple spikes from the same neuron do not give rise to a larger rate of depolarization than single spikes. Consistent with the dVm/dt spiking mechanism, cross-correlation peaks between pre- and postsynaptic neurons are very narrow (i.e. 2-3 ms) and typically precede the peak of the EPSP (Knox, 1974; Fetz & Gustafsson, 1983; Alonso et al., 1996; Matsumura et al., 1996; Bruno & Sakmann, 2006). During awake states, the integration period based on the Vm change induced by EPSPs may typically not be longer than 10 ms (Leger et al., 2005). Thus, spikes from opposite phases of the gamma cycle, which are separated by 6-15 ms, will be only weakly integrated based on an adaptive threshold mechanism, and will only be weakly integrated based on the addition of their EPSP depolarizations.

Finally, neurons possess mechanisms that do not only render them sensitive to synchronous inputs, but also to specific temporal sequences of dendritic inputs. A series of sequential synaptic inputs that traverse from the dendritic branch towards the soma are more
effective in triggering neuronal firing than a series of inputs that traverse from the soma towards the dendritic branch (Branco et al., 2010). Further, the membrane potential is tuned to the velocity by which synaptic inputs traverse from dendritic branch to soma (Branco et al., 2010). Hence, dendrites do not act as linear integrators, but render neurons sensitive to the arrival order of synaptic inputs. Thus, neurons may detect regular sequences of synaptic inputs that are occurring across different phases of the gamma cycle (König et al., 1995; Vinck et al., 2010a).

Given the existence of these feedforward coincidence detection mechanisms that are based on biophysical properties of neurons, the prediction follows that increasing the amount of firing coincidences in a presynaptic population increases the impact on a postsynaptic target. Several modeling and experimental studies directly support the notion that neurons are indeed highly sensitive to synchronous inputs. Salinas & Sejnowski (2000) investigated the effects of increasing the zero-lag synchrony between excitatory inputs in a simple conductance-based integrate-and-fire model with uncorrelated, yet balanced excitatory and inhibitory inputs (similar to Shadlen & Newsome (1994)). Enhanced synchrony between synaptic inputs did not cause a change in the mean membrane potential, but causes increased membrane potential fluctuations. This strongly increased the neuron’s output firing rate. Leger et al. (2005) have shown that during UP states, temporal integration times are twice as short as during DOWN states, likely due to an increased membrane leak conductance. EPSPs that were triggered through microstimulation only summated when they arrived within a window of 10 ms (Leger et al., 2005). Interestingly, assuming that a neuron receives a large number of synaptic Poisson inputs (10000) with a high average firing rate (4.5 Hz), then the probability of obtaining the required coincidences to trigger spiking is very small (Leger et al., 2005). Thus, sparse, synchronous spiking activity may be required to cause threshold crossings of the membrane potential. Similarly, Stevens & Zador (1998) found that sparse, synchronous events are needed to produce the typical statistics of neuronal spike trains. Alonso et al. (1996) have shown that LGN (Lateral Geniculate Nucleus) spiking activity is often synchronized with <1 ms precision. These synchronized spikes have supra-linear effects on V1 cells, showing that not only coincidence detection takes place, but also supralinear integration. The temporal integration of LGN input spikes by V1 neurons decays at a fast rate of $1/e$, with $e = 2.5$ ms (Usrey et al., 2000).

In sum, there is abundant evidence that synchrony substantially increases the impact of synaptic inputs, and that neurons are particularly sensitive to synaptic inputs coinciding in a window of few milliseconds. Thus, observed differences in synchronization between experimental conditions likely bear strong network consequences.

### 1.1.4 Balanced excitation and feedback inhibition shape synaptic integration

We have reviewed several biophysical mechanisms that enable neurons to detect coincident synaptic inputs. In this section, we review another factor that shapes the integration of synaptic inputs, namely the temporal interplay between excitation and inhibition. Excitatory and inhibitory synaptic inputs are not only balanced in terms of amplitude, as in Shadlen & Newsome (1994), but are also tightly coordinated in time (Csicsvari et al., 2003; Wehr & Zador, 2003; Hasenstaub et al., 2005; Fries et al., 2007; Okun & Lampl, 2008; Atallah & Scanziani, 2009; Cafaro & Rieke, 2010; Cardin et al., 2010; Zhou et al., 2010). A balance and tempo-
eral coordination of excitation and inhibition may serve several functions. A critical function of fast inhibitory feedback is to prevent runaway excitation of the network (Abeles, 1982; Buzsáki, 2006; Pouille et al., 2009; Moore et al., 2010), allowing strong recurrent connections that facilitate fast responses to external network input (van Vreeswijk & Sompolinsky, 1996). Further, balancing excitation by global inhibitory feedback has been proposed as a mechanism to remove output noise correlations (Renart et al., 2010). Such a mechanism would explain why noise correlations are very weak in auditory and primary visual cortex, despite the abundance of common inputs (Ecker et al., 2010; Renart et al., 2010). An enhancement of gamma-band synchronization may correspond to a reduction in slow noise correlations through an upregulation of the temporal coordination between excitation and inhibition. Attention enhances V4 gamma-band synchronization, and decreases low-frequency synchronization (Fries et al., 2001b, 2008), effectively equating a reduction in noise correlations (Mitchell et al., 2009). Balancing excitation and inhibition can also sharpen neuronal selectivity by canceling out noisy fluctuations in excitatory inputs (Cafaro & Rieke, 2010).

A delay of inhibitory activity relative to excitatory activity is a fundamental motif of network activity, and has been shown in extracellular in vivo recordings in hippocampus (Csicsvari et al., 2003; Tukker et al., 2007) and orbitofrontal cortex (van Wingerden et al., 2010a) from awake rats, and awake monkey (Womelsdorf et al., 2008). In addition, the delay has been demonstrated by intracellular recordings from anesthetized animals in several areas, including the ferret prefrontal cortex (Hasenstaub et al., 2005), the CA3 field of the rat hippocampus (Atallah & Scanziani, 2009), the rat barrel cortex (Wilent & Contreras, 2005; Okun & Lampl, 2008), the rat primary auditory cortex (Wehr & Zador, 2003), and the cat primary visual cortex (Cardin et al., 2010). The delay between excitation and inhibition can be expressed in the frequency domain as a phase lag in the gamma cycle (Csicsvari et al., 2003; Hasenstaub et al., 2005) or in the time domain as a temporal delay (Okun & Lampl, 2008), and is thought to arise from recurrent feedback inhibition or feedforward inhibition (Eeckman & Freeman, 1990; Pouille & Scanziani, 2001; Wehr & Zador, 2003; Wilent & Contreras, 2005).

An important consequence of the delay between balanced excitation and inhibition is that it can strongly reduce the temporal window of integration (König et al., 1996; Pouille & Scanziani, 2001; Thorpe et al., 2001; Fries et al., 2007). Balanced, delayed inhibition can effectively quench slow fluctuations in excitatory inputs (Cafaro & Rieke, 2010; Renart et al., 2010). Consequently, only strongly synchronized excitatory inputs may effectively trigger spiking. For example, in Pouille & Scanziani (2001), blocking the GABAa receptor on pyramidal cells increased maximum temporal integration times from a few milliseconds to about 30 ms.

The delay between excitation and inhibition is not a fixed quantity, but may be an important gating device to flexibly modulate the gain of excitation (Wehr & Zador, 2003; Wilent & Contreras, 2005; Fries et al., 2007; Kremkow et al., 2010; Zhou et al., 2010). An extreme example of such gating is the case of feedforward inhibition that precedes feedforward excitation, which occurs in so called “silent” neurons in L6 of primary auditory cortex. The temporal lead of feedforward inhibition causes these neurons to be suppressed by sensory stimulation (Zhou et al., 2010). Conversely, “normal” regular spiking neurons, for which inhibition lags excitation, are activated upon sensory stimulation (Wehr & Zador, 2003; Zhou et al., 2010).
1.1.5 Consequences of rhythmic neuronal synchronization: Rhythmic gain modulation

The impact of excitatory synaptic inputs onto a postsynaptic target neuron is modulated by inhibitory inputs. Gamma-band oscillations entail rhythmic fluctuations in inhibitory inputs. Synaptic inputs may have a larger effect on the firing rate of a postsynaptic target during gamma phases of low inhibition (Burchell et al., 1998; Fries, 2005). Neuronal interactions between rhythmically active groups should therefore depend on the phase relationship between their gamma cycles (Fries, 2005). This mechanism makes selective coherence between sender’s and receiver’s gamma-band oscillations a potentially very powerful mechanism for the flexible routing of signals in the nervous system (Fries, 2005) (“Communication-Through-Coherence” hypothesis).

During gamma-band oscillations, the output of pyramidal cells is entrained by gamma-rhythmic inhibition (Hasenstaub et al., 2005). Inhibition has two effects. First, it drives the membrane potential towards the reversal potential of the GABAa receptor, which lies well below action potential threshold. Consequently, more EPSPs are needed to breach action potential threshold. Second, perisomatic inhibition increases membrane conductance, which decreases the amplitude and duration of incoming EPSPs, i.e. shunting inhibition. Thus, excitatory synaptic inputs will be less likely to trigger firing in the postsynaptic neuron when it receives strong inhibitory inputs. Consequently, postsynaptic spikes are often preceded by a drop in inhibition (Softky, 1994; Hasenstaub et al., 2005; Azouz & Gray, 2008). Thus, the gamma cycle may represent a repetitive transition between a window of opportunity and a window of depression for communication (Fries, 2005). An advantage of sparse windows of opportunity may be to spare the energy of being in an excitable state all the time (Buzsáki, 2006), at the same time preventing noise from impacting neurons all the time (König et al., 1996; Buzsáki, 2006), although setting up rhythmic GABAergic inhibition may be likewise costly (Buzsaki et al., 2004).

The core prediction of the Communication-Through-Coherence hypothesis is that a good gamma-phase relationship between sender and receiver improves their interactions. To directly test this hypothesis, Womelsdorf et al. (2007) recorded simultaneously from spatially separate sites in the visual cortex from awake cat and monkey. Upon visual activation, sites engaged in sustained gamma-band synchronization. Consistent gamma phase relationships were observed between different sites. Phase-relationships were not completely consistent however, because there was some variation around the mean phase-relationship. The mean phase-relationship was hypothesized to be the ‘good’ gamma-phase relationship (although this hypothesis does not have a clear a priori motivation), suberving strongest interactions. As predicted by the principle of rhythmic gain modulation, the gamma-rhythm strengths in spikes and LFPs at the separate recording sites co-fluctuated strongly when there was a good gamma-phase relationship, and only weakly when there was a ‘bad’ phase relationship. These co-fluctuations were measured as the correlation between trial-by-trial power estimates of two signals, and can be measured independently from the phase relationship. Cross-correlation analysis revealed that ‘good’ gamma-phase relationships preceded strong interactions by about 5 ms, suggesting a causal relationship between the two variables, although, in general, causation cannot be inferred from the asymmetry of the cross-correlation function, as will be shown in Chapter 4. Importantly, the dependence of interactions on gamma phase held true for both short and long-range interactions, and also for the gamma-
band cortico-muscular coherence as observed by (Schoffelen et al., 2005).

Not only the effectiveness of neuronal inputs, but also the effectiveness of stimulus inputs depends on the ongoing phase of the gamma rhythm. Cardin et al. (2009) showed that the magnitude of evoked responses to whisker deflections depends on the ongoing phase of the pre-stimulus gamma rhythm. Azouz & Gray (1999) demonstrated that visually evoked membrane potentials depend strongly on the depolarization of pre-stimulus membrane potentials. Fries et al. (2001a) showed that there exist strong latency co-variations between separate LFP sites in visual cortex. Latencies of activation were especially early if the LFP was in a falling phase (i.e., depolarization) before the stimulus onset. Further, latencies were especially correlated if the LFPs engaged in gamma-band oscillations before the stimulus onset. Thus, the ongoing gamma phase of a neuronal group may modulate the impact of a sensory stimulus.

1.1.6 Consequences of rhythmic neuronal synchronization: STDP

In the previous section, we have made the case that changes in local synchrony or inter-areal coherence may have network consequences in terms of regulating the effective synaptic efficacy. This is a form of synaptic regulation that is dynamic, i.e. it does not require any persistent physical change on the synapse in terms of e.g. receptor density. Gamma synchronization may also be involved in regulating synaptic plasticity, i.e. synaptic potentiation or depression through a relatively long-lasting change in the properties of the synapse Sejnowski & Paulsen (2006). This idea will be explored in this subsection.

Cajal (1894) already proposed that learning may rely on strengthening the connection between neurons. Hebb (1949) expanded on this basic idea by proposing that a repeated contribution of one neuron’s firing (say neuron A) to another neuron’s firing (say neuron B) results in some growth process or metabolic change that changes the connection strength from neuron A to B. The capacity of synapses to show changes in strength is commonly referred to as synaptic plasticity. When these changes are relatively long-lasting, potentiating and result from a specific activation pattern of the pre- and postsynaptic cell, they are referred to as long-term potentiation (LTP). Long-term potentiation was first discovered in the anesthesized rabbit by Bliss & Lomo (1973). In their experiments, they stimulated the perforant path of the hippocampal formation and measured the population response in the dentate gyrus, using the LFP. They showed that after stimulation with ‘conditioning trains’, i.e. repetitive stimulation of the perforant path with single pulse currents at a high frequency (e.g. at a frequency of about 15 Hz for about 10 seconds), the population response of the dentate gyrus to subsequent stimulation was strongly enhanced - a phenomenon referred to as potentiation. This enhancement was observed to last for many hours (long-term potentiation). Conditioning trains can also lead to a depotentiation, however. Long-term depression (LTP) in CA1 can be induced by repetitive stimulation of the Schaffer collaterals with low-frequency conditioning trains (1 Hz) for extended periods of time (Bear & Abraham, 1996).

It has been shown that LTP or LTD (see Box 1.1) can also occur as a function of the precise timing of the pre- and postsynaptic spikes. When the spikes of the presynaptic cells (or EPSPs in the postsynaptic cell) occur slightly before the spikes of the postsynaptic cells in time, then synaptic connections are strengthened (LTP). If however the spikes of the postsynaptic cell precede the spikes of the presynaptic cells, then synaptic connections are depressed (LTD) (Markram et al., 1997; Dan & Poo, 1992). This phenomenon is commonly referred to as spike-timing-dependent-plasticity (STDP).
LTP and LTD refer to the phenomenon that synaptic connections are strengthened and weakened, respectively, as a function of the activation pattern of the pre- and postsynaptic cells. A large variety of physiological mechanisms may contribute to LTP and LTD, however (Malenka & Bear, 2004). The most well-known mechanism for LTP relies on activation of the NMDA receptor (see Box 1.1). A role of synchronization in regulating LTP is implied because of two reasons. First, synchronous activation of a presynaptic cell population leads to a larger depolarization of the postsynaptic cell, enabling NMDAR-mediated synaptic potentiation given a sufficiently depolarized membrane potential. Second, neural coherence between pre- and postsynaptic cells corresponds to a typical relationship between the timing of their respective action potentials, tapping into the NMDAR-mediated STDP mechanism. This suggests that rhythmic synchronization may play a role in the storage of long-term memories, an hypothesis which will be explored further below (Sejnowski & Paulsen, 2006; Jensen et al., 2007; Fell & Axmacher, 2011; Jutras & Buffalo, 2010).

1.1.7 Functions of the gamma rhythm: Attention

Gamma synchronization has been linked to many different functions (Sejnowski & Paulsen, 2006). When discussing possible functions of the gamma rhythm, it is important to distinguish two different levels of functions. First, a network or circuit level, e.g. functions like regulating spike-timing-dependent-plasticity or communication between areas (see above). Second, a cognitive level, e.g. functions like attention (Fries et al., 2001b) or working- and long-term memory (Jensen et al., 2007; Fell & Axmacher, 2011; Jutras & Buffalo, 2010; Sejnowski & Paulsen, 2006). Circuit-level functions may contribute to different cognitive level functions, and higher-order level functions may be subserved by different first-level functions. As for network-level functions, we already discussed the potential role of the gamma rhythm in regulating feedforward communication and inter-areal communication in the previous subsections. In this subsection, we will review evidence that gamma synchronization plays a role in selective attention.

The brain needs to achieve two important tasks in sensory processing. First, it needs to reliably encode, classify, and integrate sensory stimuli. It is commonly accepted that neurons represent much information through reliable firing rate changes, although temporal codes may play an important role as well. Neurons can store and represent information because the weights of specific synaptic inputs are slowly adjusted over time through long-term potentiation (Hebb, 1949; Bliss & Lomo, 1973; Markram et al., 1997). Second, the brain needs to select a subset of available information for further processing and for action selection. The selection of information requires that routing of signals between neuronal groups is flexibly adjusted on a timescale much faster than the timescale at which long term synaptic potentiation takes place. Selective routing of signals between neuronal groups may be implemented through selective coherence between their ongoing rhythms (Fries, 2005), although it may alternatively rely on the actions of neuromodulators like acetylcholine (Harris & Thiele, 2011).

In the visual hierarchy, connectivity patterns are highly convergent, with many upstream neurons providing convergent input into a given downstream neuron. Accordingly, receptive field sizes strongly increase along the visual hierarchy. This design principle has several advantages: First, having large receptive fields allows for the invariant representation of objects, independent of e.g. object position. Second, it is economical, since higher order representations are capable of covering large portions of the visual field. The disadvantage of this
design is that multiple objects can populate the large receptive field of downstream visual neurons, which confuses information from different objects. While this problem might be solvable by population coding, it might also be resolved at the level of individual neurons by using attention to select and process single objects from the multitude of available objects, effectively shrinking the receptive field size of higher visual neurons (Moran & Desimone, 1985). When attention is directed to one of the stimuli, then the neurons in higher visual areas respond as if only the behaviorally relevant stimulus was presented (Moran & Desimone, 1985; Desimone & Duncan, 1995; Reynolds et al., 1999; Reynolds & Chelazzi, 2004). That is, there is competition between converging inputs, which can be biased by attention (Desimone & Duncan, 1995).

Fries (2005) proposed that synaptic inputs that are strongly gamma-rhythmic and coherent with the intrinsic gamma-rhythm of the downstream receiver have an advantage over competing synaptic inputs, a mechanism that could be employed to implement biased competition, although the mechanism that regulates gamma-coherence between distant neuronal groups is not further specified in Fries (2005). In support of this hypothesis, several studies have shown a strong enhancement of gamma-band synchronization with attentional modulation in V4 (Fries et al., 2001b, 2008; Bichot et al., 2005; Taylor et al., 2005; Gregoriou et al., 2009; Chalk et al., 2010). An enhancement of visual gamma-band synchronization may have strong behavioral consequences, since it corresponds to reduced reaction times and/or more accurate responses (Taylor et al., 2005; Womelsdorf et al., 2006; Hoogenboom et al., 2010). Importantly, induced gamma-band oscillations in V4 emerge before top-down modulations on firing rate arise (>50 ms) (Fries et al., 2001b), and attentional enhancement of gamma-band oscillations occurs as soon as an attentional cue is presented, already before stimulus onset (Fries et al., 2008). Enhanced gamma-band synchronization in V4 may enhance impact onto IT (Inferior Temporal cortex) neurons through feedforward coincidence detection, or may reflect increased coupling of oscillatory rhythms across visual areas. Consistent with the CTC-hypothesis, attentional modulation is correlated to an increase in gamma-band synchronization between V4 and FEF (Gregoriou et al., 2009), LIP and FEF (Buschman & Miller, 2007), V1 and V4 (Bosman et al., 2012) and MT (Medial Temporal Area) and LIP (Saalmann et al., 2007). Interestingly, attention does not increase gamma-band synchronization in V1 but often even mildly decreases it (Chalk et al., 2010; Herrero et al., 2013), suggesting that neurons in V1 might not enhance their synaptic gain onto V4 target neurons by increased gamma-band synchronization among the V1 neurons.

While these findings are supportive for a role of gamma synchronization in selective attention, some critical remarks are in place at this point. (1) It is presently unknown what mechanisms cause enhanced gamma-band synchronization in area V4, and between different brain areas (V1-V4, LIP-FEF, V4-FEF). (2) Alternative explanations for the finding of enhanced inter-areal gamma coherence between brain areas with selective attention are not excluded. In particular, it is possible that there is another mechanism to regulate synaptic efficacy dynamically, e.g. the actions of neuromodulators. Enhanced synaptic efficacy might subsequently increase the extent to which signals can predict each other (i.e. coherence) at the frequencies at which communication ‘typically’ takes place, explaining an increase in observed neural coherence. (3) Gamma synchronization is quite dependent on sensory stimulation (Gieselmann & Thiele, 2008; Ray & Maunsell, 2010). It is therefore questionable whether gamma-band synchronization can provide a mechanism for selective attention under all sensory circumstances. (4) Thus far, there has been only correlational (i.e., not causal)
evidence for the hypothesis that selective attention is subserved by neuronal gamma synchronization. It remains an open question whether the changes in synchrony induced by the gamma rhythm, and the changes in the timing between pre- and postsynaptic activity induced by neural coherence, are of sufficient strength to have impact on synaptic efficacy. For example, neural coherence values between V1 and V4 attain values of about 0.2 with attention (Bosman et al., 2012). However, it should be noted that this pertains to coherence between population mass signals. As shown by Zeitler et al. (2006) and Vinck et al. (2012b), squared coherence between spikes and LFPs increases linearly with the number of spikes. Similarly, one can show that coherence between spike trains increases linearly as a function of the number of spikes in both signals. As the LFP represents the activity of many (tens of thousands) local neurons (Buzsaki et al., 2012), it is to be expected that coherence between V1 spikes and V4 membrane potential fluctuations (in the gamma range) is much smaller, i.e. a factor of $\frac{\sqrt{n}}{n}$ smaller, where $n$ is on the order of the number of neurons constituting the LFP in V1. Indeed, the modulation of spikes in V1 by the LFP gamma phase in V2 has been shown to be quite weak (Jia et al., 2013). The critical consideration here is that rhythmic gain modulation should increase the average impact of individual spikes, which means that spike-field coherence is the crucial quantity to consider, not field-field coherence.

1.1.8 Functions of the gamma rhythm: Long-term memory

Another function to which gamma synchronization has been linked is long-term memory. The idea that gamma synchronization is involved in the storage of long-term memories is derived from the basic idea that rhythmic synchronization entails precise timing relationships between neurons, which has consequences for synaptic plasticity mediated by the STDP mechanism, as reviewed above (Jensen et al., 2007; Jutras & Buffalo, 2010; Fell & Axmacher, 2011; Sejnowski & Paulsen, 2006). Thus, increases in local gamma synchronization and interareal gamma coherence may lead to a potentiation or depression of synapses, potentially contributing to the formation of long-term memories.

Indeed, studies in humans have shown that gamma synchronization in neocortex and hippocampus during encoding predicts formation of long-term memories (Osipova et al., 2006; Jensen et al., 2007; Fell & Axmacher, 2011). This has also been shown, for recognition memory, in the primate hippocampus Jutras et al. (2009). Gamma phase-synchronization between rhinal and hippocampal areas has also been shown to predict successful formation of long-term memories (Fell & Axmacher, 2011). Most studies examining the role of gamma synchronization in the formation of long-term memories examined power of and neural coherence between population mass signals (Jensen et al., 2007; Fell & Axmacher, 2011; Jutras & Buffalo, 2010). However, the storage of memories is likely specific to the neurons that are activated by the to-be-encoded stimulus. One mechanism for the specific formation of memories may be found in the precise timing of neuronal firing relative to the gamma cycle. We have recently shown, in area V1 of the awake monkey, that neurons that are optimally activated by a visual stimulus fire at earlier phases in the gamma cycle than neurons that are activated by non-preferred stimuli (Vinck et al., 2010a). The consequence of this phenomenon, coined gamma phase shifting, may be enhanced potentiation between the early-firing neurons and the rest of the gamma-rhythmic network, due to STDP (Vinck et al., 2010a).

Again, we would like to make several critical remarks. (1) It is not excluded that enhanced gamma power and coherence during encoding - predicting formation of long-term memories
is a by-product of another physiological process, e.g. the overall excitability or activity of the network. (2) A causal demonstration that gamma synchronization is indeed critical for the formation of long-term memories has not yet been given. (3) As above, it remains an open question whether the changes in synchrony induced by the gamma rhythm, and the changes in the timing of pre- and postsynaptic activity induced by neural coherence, are of sufficient strength to have substantial impact on the formation of long-term memories.

1.1.9 Functions of the gamma rhythm: Coding by phase-of-firing

Finally, we consider a role of the gamma rhythm in the formation and read-out of sensory representations. It is well established that much information about sensory data is encoded by the firing rate. Yet, it is highly debated to what extent the temporal structure of spiking activity serves as a coding space. Temporal codes can, in theory, carry much more information than the firing rate (MacKay & McCulloch, 1952). A fundamental problem with temporal coding is that spike times can carry information only in relation to other neuronal events, i.e. the brain needs to measure spike times relative to a temporal reference frame. Using the onset of sensory stimuli as a reference frame is problematic, since the brain cannot obtain independent information about the timing of stimulus onsets, and sensory input typically forms a temporal continuum where distinct stimulus onsets are absent (VanRullen et al., 2005; Fries et al., 2007; Panzeri et al., 2010). Using the onset of saccades or microsaccades as a temporal reference frame would effectively limit the temporal resolution of sensory systems to the saccade or microsaccade rates, which are only about 3-4 Hz (Maldonado et al., 2008; Otero-Millan et al., 2008; Bosman et al., 2009). A more plausible solution is that the brain defines the timing of a spike relative to the activity of a local group or population of neurons. Endogenously generated oscillations can serve as a reliable, internal clock to define the timing of spikes, constituting a phase coding space for sensory data (O’Keefe & Recce, 1993; Hopfield, 1995; Wehr & Laurent, 1996; Fries et al., 2007; Nadasdy, 2010; Panzeri et al., 2010; Tiesinga & Sejnowski, 2010) and assembly formation (Singer, 1999; Buzsáki, 2010). In such a coding scheme, a group of neurons is entrained by one rhythm, while the spike times of individual neurons are phase shifting within the rhythm’s cycle as a function of the neuron-specific inputs.

The first experimental evidence for the existence of phase coding in the central nervous system was obtained in the rat hippocampus. When a rodent moves through a place field of a given place cell, then the cell’s phase of firing advances in a monotonic fashion relative to the hippocampal theta rhythm, a phenomenon called phase precession (O’Keefe & Recce, 1993; Harris et al., 2002; Mehta et al., 2002; Huxter et al., 2003; Harvey et al., 2009; Schmidt et al., 2009). Phase precession relative to the hippocampal theta rhythm also occurs in prefrontal cortex (Jones & Wilson, 2005) and ventral striatum (van der Meer & Redish, 2011). The mechanisms underlying phase precession are still highly debated (Harris et al., 2002; Mehta et al., 2002; Huxter et al., 2003; O’Keefe & Burgess, 2005; Harvey et al., 2009; Romani et al., 2010). An elegant proposal is that phase precession arises from an interaction between somatic inhibition and dendritic excitation. Stronger dendritic excitation would allow a neuron to overcome inhibition earlier in the theta cycle. Correspondingly, a theta-phase advance would be observed concurrently with higher firing rates. Indeed, earlier theta phases correspond to higher firing rates when a rat moves through the first half of a place field (Harris et al., 2002; Mehta et al., 2002). Importantly, the same relationship between phase
and rate holds during other behavioral states, namely wheel running and REM sleep (Harris et al., 2002). One problem with this model however is that firing rates start to decrease when a rat has moved beyond the center of the place field, while the spike’s theta phases continue to advance (Huxter et al., 2003; O’Keefe & Burgess, 2005). This discrepancy has lead others to the proposal that the theta phase of firing advances because an activated place field is driven by theta-rhythmic inputs that have a slightly higher frequency than the LFP population theta-rhythm (O’Keefe & Burgess, 2005). However, intracellular recordings revealed that membrane potential depolarization is asymmetric, i.e., continues to rise when a rat moves beyond the center of a place field (Harvey et al., 2009). This asymmetric tuning of the membrane potential may explain why the phase continues to advance beyond the center of the place field (Mehta et al., 2002). However, the observed intracellular profile does not necessarily rule out oscillatory interference (O’Keefe & Burgess, 2005) or network models (Romani et al., 2010). Nevertheless, the soma-dendritic model offers the simplest explanation of rate-to-phase conversion under different behavioral states (Harris et al., 2002).

The short duration of the gamma cycle makes it especially suited to organize the phase-dependent coding of sensory information. The alpha, theta and delta rhythm also imply synchrony of spike discharges, but their peaks of synchronized activity only repeat themselves every 100-1000 ms. If sensory representations are built up and broken down along the oscillatory cycle, then these rhythms are too slow to be matched to typical sensory reaction times. The decoding of spike phase requires a waiting or updating time on the order of the cycle duration. For slower rhythms, this means that the system has to wait 100 to 1000 ms before it can update its representations again. This updating time may be sufficient for representing the low-frequency characteristics of stimuli (Montemurro et al., 2008; Kayser et al., 2009; Panzeri et al., 2010). However, the visual system can distinguish distinct objects at a high sampling rate (<100 ms) (Thorpe et al., 1996; VanRullen & Koch, 2003). The gamma cycle (40-80 Hz) allows for sensory updates every 12-20 milliseconds, which is well matched to these processing requirements.

First evidence for gamma phase-coding of visual stimuli was obtained by König et al. (1995). Primary visual cortex of anesthetized cat was activated by a drifting bar stimulus. A single stimulus activated separate MUAs (Multi Unit Activities) recorded on separate electrodes. Upon visual activation, the MUAs displayed synchronous firing, i.e., there was a peak in the cross-correlogram between their spike trains. The extent to which a MUA was driven by the bar stimulus depended on the stimulus’ orientation and spatial frequency. If a stimulus was driving two MUAs equally strongly, then zero-lag synchrony was observed. When a stimulus activated one MUA more strongly than another MUA, the firing of the strongly activated MUA preceded the firing of the weakly activated MUA. While these results suggest that the relative timing of spikes can code for stimulus features, they do not demonstrate directly that spikes shift relative to the LFP gamma-band oscillation, since the effects were not studied in the frequency-domain.

We tested directly whether gamma phase-shifting exists in area V1 (Vinck et al., 2010a). Awake monkeys were passively viewing drifting gratings, which elicited strong locking of spiking activity to the ongoing LFP gamma-band oscillations. When an isolated single unit was stimulated by its preferred grating orientation, spikes were on average advanced in the gamma cycle (by a maximum of about 50°). In general, when the local spike density was high around the time of spiking, then the gamma phase of firing was also advanced (this also occurred within a single trial). The phase difference between high and low spike densities
was on average 2-3 ms, a difference that should be detectable by a feedforward coincidence detection mechanism.

We hypothesized that gamma phase-shifting is especially strong when spiking activity is weakly constrained by the gamma rhythm. This hypothesis follows from a Hopfield-type model (Hopfield, 1995), in which higher excitation allows the neuron to overcome gamma-rhythmic inhibition earlier in the gamma cycle, leading to a phase advance. If gamma-rhythmic inhibition is strongly constraining spike timing, then an equal increase in excitation should lead to a smaller phase advance. Indeed, neurons whose firing was strongly constrained by the gamma-rhythm displayed smaller gamma-phase shifts (Vinck et al., 2010a). In addition, we found gamma phase-shifts to be larger when LFP gamma-band power was weaker, and during the fixation baseline condition. These findings may explain why phase shifts were larger in König et al. (1995) than in Vinck et al. (2010a): The former experiment used anesthesized cats, while the latter experiment used awake monkeys and large stimuli, causing spikes to be strongly constrained by the gamma rhythm. However, smaller yet reliable gamma-phase shifts may be more informative than larger yet unreliable gamma phase shifts.

The relationship between local firing rate and phase points to a shared mechanism between theta phase-precession and gamma phase-shifting. However, if stronger excitation allows a neuron to overcome inhibition earlier in the gamma cycle, then it should also allow the neuron to overcome inhibition for a longer period in the gamma cycle. This would not cause a phase shift, but a decrease in phase locking. However, neurons are more strongly locking to the gamma rhythm when they are activated by their preferred stimulus orientation (Friedman-Hill et al., 2000; Volgushev et al., 2002). Thus, a negative feedback mechanism, such as local inhibitory feedback or a refractory period, is required in addition to the interaction between excitation and global gamma-rhythmic inhibition.

Gamma phase-coding has also been demonstrated outside visual cortex, for a two-object short-term working memory task in lateral prefrontal cortex (Siegel et al., 2009). Monkeys were trained to remember the presentation order of two serially presented visual stimuli. Spikes were strongly locked to the ongoing LFP gamma-band oscillation at around 32 Hz, especially during the delay periods following the visual stimuli. These gamma-band oscillations were not phase locked to trial events. During the second delay period, when monkeys had to represent both objects in working memory, firing rates carried information about the identity of both visual stimuli. Astonishingly, information about the two objects was carried at separate gamma phases. Information about the first object was maximal at a gamma phase that was about $60^\circ$ (i.e., about 5 ms) earlier than the gamma phase at which information about the second object was maximal. These results suggest that the gamma cycle allows for the representation of different objects to be segregated by phase, thereby linking the activity of members of the same neuronal assembly by firing at the same gamma phase (Singer, 1999; Buzsáki, 2010).

### 1.1.10 Relationship between firing rate coding and rhythmic synchronization

Rate coding and rhythmic synchronization have traditionally been described as two separate processes that may serve complementary functions (Singer, 1999; Fries, 2005). However,
changes in the rate code itself may be a consequence of changes in rhythmic synchronization. Differences in rhythmic synchronization at the presynaptic side may be converted into differences in rhythmic synchronization at the postsynaptic side through feedforward coincidence detection (Abeles, 1991; Aertsen et al., 1996; Fries et al., 1997, 2001b) or rhythmic entrainment (Fries, 2005). However, rhythmic synchronization of presynaptic neuronal activity likely affects postsynaptic firing rates as well. Azouz & Gray (2000) showed that the output firing rate of V1 neurons can be better predicted by the amplitude of membrane potential gamma-band fluctuations than by the average or the slow component of the membrane potential. Consequently, gamma-synchronized spiking activity has a prominent role in shaping V1 orientation tuning (Azouz & Gray, 2003). In concordance, the most gamma-synchronized spikes are also the most orientation tuned (Womelsdorf et al., 2012), and the gamma-band LFP power is more tuned to orientation than the power of the low-frequency LFP components (Frien et al., 2000). A similar relationship between firing rate selectivity and gamma-band synchronization likely holds for other stimulus features as well. In V1, the gamma-band component of the LFP is most strongly tuned to contrast (Henrie & Shapley, 2005; Ray & Maunsell, 2010). In area MT (Medial Temporal), the LFP gamma-band power is more strongly tuned to speed and direction than low-frequency power is (Liu & Newsome, 2006). Rotermund et al. (2009) showed that the gamma-band power spectrum of V4 electrodes provides much more information about stimulus shape than was contained in low-frequency LFP oscillations. Finally, a positive attentional modulation of V4 firing rates corresponds to an increase in LFP gamma-band power, but a decrease in the power of low-frequency LFP oscillations (Fries et al., 2008), suggesting that the attentional modulations in firing rate are partially driven by changes in gamma-band synchronization.

Not only the strength of rhythmic synchronization, but also its phase may affect rate coding substantially. V1 neurons that are strongly driven by a visual stimulus, spike early in the gamma cycle (König et al., 1995; Vinck et al., 2010a). These early spikes may, through feedback inhibition, suppress the firing rates of neurons that are less strongly driven by the same stimulus (Fries et al., 2007; de Almeida et al., 2009). This could serve to suppress firing rates for non-preferred stimuli, thereby sharpening firing rate selectivity. The critical parameter in such a scenario is the delay between excitation and inhibition. If a neuron fires early in the gamma cycle, then it may have either received excitatory inputs that fall relatively early in the gamma cycle, or it may trigger early excitatory inputs in co-tuned neurons, thereby escaping the rise of global FS basket cell inhibition. Small changes in the delay between excitation and inhibition strongly affect output firing rates (Wehr & Zador, 2003; Wilent & Contreras, 2005; Kremkow et al., 2010). In auditory and barrel cortex, the delay between monosynaptic feedforward excitation and disynaptic feedforward inhibition is a crucial parameter that shapes the resulting rate code (Wehr & Zador, 2003; Wilent & Contreras, 2005).

An important function of gamma-band synchronization may be to act as a mechanism to filter out irrelevant information (König et al., 1996; Buzsáki, 2006). Noisy background activity that is not synchronized to the gamma rhythm may on average fail to exert strong impact on output firing rates, since it is likely quenched by gamma-rhythmic inhibition. At the same time, the nervous system may focus the most information rich cortical computations at the phase in the gamma cycle where pyramidal cells escape inhibition. Indeed, in V1, there is enhanced orientation tuning at this gamma phase, independent of the number of spikes (Womelsdorf et al., 2012). Similarly, synchronous LGN spikes carry more stimulus
information (Dan et al., 1998) and also have more impact on V1 cells (Alonso et al., 1996; Usrey et al., 2000). Thus, the gamma cycle may act as a filter that focuses information rich processes at the phase of low inhibition, at the same time quenching the majority of noise background inputs.

1.2 Anatomy and function of the orbitofrontal cortex (OFC)

We briefly describe anatomical and functional properties of the two main brain areas that will be studied in Chapters 6 to 8.

1.2.1 Anatomy of the OFC

We first describe the anatomical location, divisions, and connectivity profiles of the rat OFC. The rat OFC is located on the ventral surface of the prefrontal cortex, like primate OFC, and is exclusively agranular (Ongur & Price, 2000; Wallis, 2012). There exist some notable differences and similarities between rat OFC and primate OFC: In terms of connectivity profile and anatomical location, the OFC is comparable to the primate OFC (Ongur & Price, 2000; Wallis, 2012), however, primate OFC consists of agranular, dysgranular and granular cortex, whereas rat OFC consists only of agranular cortex; thus, one should be cautious with extrapolating findings from rat OFC to primate OFC and vice versa (Wallis, 2012). In what follows, we focus on the anatomy and function of the rat OFC, unless stated otherwise. Rat OFC is subdivided into the medial (MO), ventral (VO), ventrolateral (VLO), and lateral (LO) OFC, with MO receiving fewer inputs from sensory areas than the other subdivisions (Palomero-Gallagher & Zilles, 2004; Reep et al., 1996; Hoover & Vertes, 2011). Rat OFC receives inputs from many neocortical areas, namely cingulate cortex, Fr2 (dorsomedial PFC), PPC (posterior parietal cortex), insular cortex (primary cortical site devoted to taste processing, i.e. gustatory cortex), S1, S2 (primary and secondary somatosensory cortex), Oc2M, Oc2L (medial and lateral visual association area), piriform (primary olfactory cortex), perirhinal, postrhinal and enthorinal cortex (Datiche & Cattarelli, 1996; Palomero-Gallagher & Zilles, 2004; Reep et al., 1996; Kondo & Witter, 2013; Ongur & Price, 2000). Connections with these areas are reciprocal (Reep et al., 1996; Hoover & Vertes, 2011; Datiche & Cattarelli, 1996). Reep et al. (1996) did not observe projections from auditory areas to OFC, although these have been reported for medial PFC and monkey OFC (see Groenewegen & Uylings (2000) for an overview). OFC also holds reciprocal connections with thalamic nuclei, in particular the submedial and medial dorsal nuclei (carrying noxious, cutaneous, visceral and limbic inputs) (Reep et al., 1996) and the amygdala (Reep et al., 1996; Ongur & Price, 2000). OFC also makes disynaptic reciprocal connections with the CA1 field of the hippocampus via the nucleus reuniens (McKenna & Vertes, 2004). OFC cells also project to the ventral striatum (Ongur & Price, 2000). Thus, OFC can be described as a multisensory integration area with extensive connections to fronto-parietal, medial temporal lobe and subcortical regions. Yet, its sensory representations are somewhat limited, as there is no evidence that rat OFC (vs. monkey OFC) receives direct projections from auditory areas, although OFC has been reported to project to auditory cortex (Reep et al., 1996; Hoover & Vertes, 2011). Rolls (2008) has hypothesized that the sensory representations of the OFC (in the monkey) are primarily concerned with the representation of edible items. This hypothesis is to some degree
supported by the nature of the afferents to the OFC (Ongur & Price, 2000). For example, in the monkey, the somatosensory inputs to the OFC derive from hand and face regions of somatosensory cortex (Carmichael & Price, 1995a). As for the rat OFC, it is plausible that auditory inputs are least important when it comes to the representation of food: One can obviously smell, taste, feel and see food, but the concept of ‘hearing food’ appears somewhat strange. Interestingly, OFC neurons are highly selective for olfactory inputs, to a similar or higher degree than cells in the piriform cortex (primary olfactory cortex), and OFC lesions impair rats’ odor discrimination abilities (Schoenbaum & Roesch, 2005; Eichenbaum et al., 1983, 1980; Roesch et al., 2007a). These information-rich olfactory representations in the OFC likely rely on inputs from the medial dorsal nucleus of the thalamus and piriform cortex (Schoenbaum & Eichenbaum, 1995). Nevertheless, more understanding is required of the precise information content of the multisensory representations and afferents in the OFC, and likewise the projection fields of OFC neurons to sensory areas. A final and important note with respect to the anatomy of the OFC is that many of the recordings that are typically made in OFC (also in Chapter 7 and 8) do not sample exclusively from OFC proper (especially the ventro-lateral part), but also sample partially from the agranular insular cortex. The insular cortex receives strong gustatory inputs, especially the granular part of the insular cortex, which has less dense connectivity with the OFC than the agranular insular cortex however, Palomero-Gallagher & Zilles (2004); Reep et al. (1996). Thus, the term ‘OFC’ as often used in the electrophysiological literature should sometimes rather be interpreted as OFC/AI.

1.2.2 Function of the OFC

There is no consensus about the function of the rat (or the monkey) OFC (Schoenbaum et al., 2009; Pennartz et al., 2011), and it is doubtful whether the concept of ‘the functions’ readily applies to a structure such as the OFC. One way to inquire the function of the OFC is to examine the behavioral effects of lesions. OFC lesions impair the performance on olfactory discrimination tasks (Eichenbaum et al., 1983), indicating an important role of the OFC in olfactory processing. Furthermore, OFC lesions do not lead to impairments in the acquisition of stimulus-outcome associations, but do lead to strong impairments in reversal learning, i.e. when animals learn a change in stimulus-outcome associations (when the S+ stimulus becomes the S- stimulus and vice versa) (Stalnaker et al., 2007; Schoenbaum et al., 2009, 2003a; Bohn et al., 2003b). Electrophysiological studies have shown diverse responses of neurons in the OFC. These include discriminative sensory responses, e.g. to an olfactory cue or delivered fluid, but also anticipatory responses, i.e. responses that predict the probability, magnitude and timing of different outcomes (van Duuren et al., 2008, 2009; Schoenbaum et al., 1998; Schoenbaum & Eichenbaum, 1995; Roesch et al., 2006). These predictive responses build up gradually over the course of learning, and are modified gradually upon an experimental change in stimulus-outcome contingencies (Schoenbaum et al., 2009).

Both OFC and piriform cortex contain many cells that carry olfactory information in their firing rate (Schoenbaum & Eichenbaum, 1995). It is therefore highly revealing to examine in what aspects OFC firing differs from firing in piriform cortex. Few important differences can be highlighted. (1) OFC cells are more predictive of trial outcome (based on stimulus-reward associations) than cells in piriform cortex (Schoenbaum & Eichenbaum, 1995; Roesch et al., 2007a), suggesting a role for OFC in generating predictions that surpass the olfactory domain. (2) While cells in piriform cortex often change firing rate selectivity for the S+/S-
stimulus after a reversal, their S+/S- selectivity does not reverse, as it does, for a subset of about 20-25% of neurons, in the OFC (Roesch et al., 2007a; Schoenbaum et al., 2003a). This finding indicates that OFC cells do not only carry stimulus-information, but mix stimulus representations with predictions about associated trial outcomes. (3) OFC contains neurons that are especially active both during the odour cue sampling and outcome delivery, whereas the piriform cortex does not (Roesch et al., 2007a). Thus, OFC cells add an extra ingredient on top of its immediate sensory signals, an ingredient that is typically interpreted in economical terms as being a ‘value’ signal. Value in the economical sense builds on the notion of exchangeability of goods, where a certain amount of good X equates a certain amount of good Y, and is derived from the subjective preferences of the actors, in that the value of a good is indicated by the extent to which it is preferred by the actor. Neurons in the monkey OFC have been shown to be selective for value, in tasks where both the type and magnitude of offered goods was varied (Padoa-Schioppa & Assad, 2006). Nevertheless, many other kinds of responses (e.g. selective responses for taste, or quantity) can be found in the monkey OFC (Padoa-Schioppa & Assad, 2006). However, it should be noted that in the rat OFC, neurons do not seem to integrate different aspects of a trial (delay, magnitude) outcome into one unified value representation (Roesch et al., 2006); it therefore remains to be seen to what extent rat OFC represents values.

1.2.3 OFC and predictive coding

A unified perspective on the function of the OFC is that the OFC generates prediction signals about expected outcomes that are used in other structures, such as amygdala and striatum, to generate prediction errors, which can then be used to drive associative learning in these and other structures (Schoenbaum et al., 2009). If the function of OFC is indeed to generate predictive signals, then the central question is what the nature of these predictions is, and how they are used. Schoenbaum et al. (2009) hold that the OFC generates predictions about expected ‘outcomes’: “Specifically, we suggest that the OFC signals the predicted characteristics - such as sensory properties (size, shape, texture and flavour) and perhaps even contextual cues such as particular timing or likelihood - and unique value of specific outcomes that an animal expects given particular circumstances and cues in the environment.”. However, Schoenbaum et al. (2009) do not give a formal definition of what constitutes an ‘outcome’; the concept of ‘outcome’ is merely used to refer to artificial laboratory settings where there is a predictive stimulus and a subsequent outcome with some valence attached to it. It is unknown however whether the predictive nature of OFC extends beyond these ‘outcomes’, and what the function of these predictions is.

An interesting link can be made with theories of predictive coding (e.g., see Bastos et al. (2012); Friston (2008); Rao & Ballard (1999)). These theories hold that the brain compares the sensory input data with an internal predictive model about that sensory input data, and updates the predictions about new sensory input data based on the prediction error between the incoming sensory input data and the predicted sensory input data. The advantage of such a predictive coding scheme is that sensory input data is often noisy, and that more accurate representations of the state of objects (and actors) can be constructed when combining sensory data with prior knowledge about the behavior of real-world objects (Friston, 2008). A good engineering example is a GPS system: It combines noisy input data (GPS measurements) with predictions about the physical trajectory of the car based on previous position estimates.
Chapter 1

1.2.4 The domain of OFC predictions

We propose here the following functional account of the OFC: The OFC serves to make state predictions of objects in the immediate spatio-temporal surrounding of the actor; these predictions are derived from first- and higher-order associations (priors) between different sensory input streams, combined with information from the motor system and medial temporal lobe, encoded by an associative, plastic neural network; the state predictions have a continuous character in that the spikes subserving the state estimate are only weakly entrained by rhythmic processes such as licking and sniffing, in contrast with gustatory and olfactory sensory areas; the predictions makes critical contributions to multimodal (conscious) perception and sensory discrimination; the functional role of the OFC predictions is multipurpose: it serves to guide decision making, to overrule and guide predictions based on first-order associations - implemented by the older subcortical brain systems such as the amygdaloid complex - by higher-order associations; but it also serves to make coding in earlier sensory areas more efficient and sparse, through reciprocal connections with sensory areas.

There are three main aspects of OFC predictions to consider in this predictive coding framework: (1) the input domain, (2) the priors, and (3) the valorization of predictions. We discuss each of these factors in the following subsections. As for the first aspect, the sensory and motor data that the OFC predictions are made for - these encompass a certain input domain. For example area IT may generate top-down predictions that only concern the visual domain. The OFC may generate predictions that extend beyond the limited concept of ‘outcomes’ having some value; predictions may be made about all input (sensory and motor) data that reaches the OFC, regardless of whether the object generating that data is valuable or not. For example, the OFC may also contain predictive signals about the olfactory cue itself carrying outcome-predictive information. The specific nature of sensory inputs to the rat OFC, i.e. its strongly selective olfactory and gustatory signals (but weak auditory inputs, see above), supplemented by visceral and noxious information, suggests that sensory predictions are primarily generated about objects with which the rat directly interacts, and which will often carry a certain value (e.g., food, or another rat), but also about the internal (visceral, noxious) and motor state of the agent, as objects have bodily consequences for the actor. Nevertheless, future studies are required to elucidate the precise nature of the sensory inputs that the rat OFC receives; furthermore, the idea that the OFC encompasses a certain input domain as a whole might neglect the existence of domain gradients within the OFC, or across several prefrontal structures.

Another important aspect of predictions - besides the input domain - is their temporal extent, i.e. how far the OFC’s predictions look ahead. If the OFC serves to make top-down predictions about incoming sensory data, then it is sensible that it primarily makes predictions about events occurring in the near future. This highlights an important distinction with
Schoenbaum et al. (2009)’s theory: Schoenbaum et al. (2009) holds that the OFC codes for the expected value of outcomes. However, outcomes can be located quite far in the future - when does the OFC stop coding for the predicted outcome? We believe it is more plausible that the OFC’s state predictions are focused on the objects that are giving rise to sensory data in the near future. If OFC is bombarded with complex sensory inputs, then it will allocate most of its processing resources to estimate and predict the state of objects giving rise to this data; if however the OFC does not receive much sensory input, e.g. when a rat moves from an odor sampling port to a fluid delivery well, then its neurons may express predictions about distant future events more intensely. In the odor sampling period of an odor-discrimination task, the most relevant variable to estimate is the identity of the odor that is currently being presented. It is therefore to be expected that most cells discriminate the identity of the odor, rather than the outcome that is associated with the odor, as has been shown by Schoenbaum & Eichenbaum (1995). The OFC’s top-down predictions of odor identity may then lead to a prediction of the sensory inputs arriving via the medial dorsal nucleus of the thalamus and the piriform cortex, areas that are reciprocally connected with the OFC. While the OFC’s state estimates and predictions should be continuous in time, allowing them to be continuously broadcast to other areas, olfaction suffers from the problem that odor sampling is highly rhythmic (about 8 Hz) due to sniffing. Thus, sensory inputs only arrive in bouts, implying that state predictions need to fill in the temporal gaps in sensory data, akin to our visual system filling in the position of a driving car when it temporarily disappears behind a tree (Uchida et al., 2006). Interestingly, cells in the olfactory bulb and piriform cortex are very strongly entrained by the sniffing rhythm, providing stimulus information only in bouts of spiking outputs occurring during a certain phase of the sniffing cycle (Shusterman et al., 2011; Miura et al., 2012). In the OFC, on the other hand, theta locking and LFP theta power during the odour sampling period are only weakly elevated (Chapter 7, van Wingerden et al. (2010b,a)), suggesting that OFC cells fill in the temporal gaps within the sniffing cycle and create a continuous state estimation of the world. Such a continuous state estimate may be a sine qua non for conscious perception. In fact, in humans, OFC damage can lead to a complete loss of conscious odor perception (Li et al., 2010), resembling the impairment in olfactory discrimination ability in rats after OFC lesions (Eichenbaum et al., 1980, 1983). A similar ‘discrete to continuous’ conversion may occur for taste perception: Cells are strongly theta phase locked to LFP theta oscillations before reward delivery in the OFC (van Wingerden et al., 2010b), which likely reflects an interaction between reward predictions and rats’ licking behavior (van Wingerden et al., 2010b; Gutierrez et al., 2006). However, van Wingerden et al. (2010b) show that theta locking decreases towards the fluid delivery and disappears almost completely after fluid delivery, a period during which rats lick most vigorously. A plausible explanation for this finding is that the OFC cells make continuous state estimates of the fluid identity after fluid delivery, such that locking to the licking cycle is actively suppressed, whereas cells are free to track and predict rhythmic motor behavior before fluid delivery. The topic of rhythmicity in the OFC will be addressed in Chapters 7 and 8.

The finding that in the odor cue sampling period only few neurons care about the predicted task outcome, in the sense that only about 20-25% of cells slowly reverse their S+/S- selectivity in the cue period (Schoenbaum et al., 2003a; Roesch et al., 2007a; van Wingerden et al., 2012), may be easily explained if the OFC is not too much concerned with the state of the world in a few seconds, but is primarily concerned with making a correct state estimate for the objects that are presently giving rise to sensory data. However, some neurons may
care about a more distant sensory event - perhaps because they actively anticipate the disappearance of the sensory stimulus, or because they actively sustain stimulus associations, since prediction presupposes memory. van Wingerden et al. (2010a) (Chapter 7) have shown that cells firing right after the cue sampling period, i.e. in the movement period, are even inhibited during odor sampling, which is also reflected by an increase in gamma-rhythmic entrainment. This may reflect a suppression of a prediction network that should only become active after the present stimulus disappears. In fact, reward predictions are more intense immediately before reward delivery, and reward state estimates (in terms of reward magnitude) are most accurate after reward delivery (van Duuren et al., 2008), which is predicted if the OFC serves to generate predictions about objects in its immediate spatio-temporal surroundings, rather than about some distant value-carrying outcome.

Evidence for a role of the OFC in generating immediate olfactory state predictions has been provided by Schoenbaum & Eichenbaum (1995), who trained rats on an olfactory discrimination task in which the sequence of odor presentations was predictable. The authors showed that cells’ firing rates distinguished different odours, but that they were also strongly influenced by whether or not a certain odour was predicted or unpredicted (as was behavior). A similar modulation was found in the piriform cortex though, so it is unknown whether this predictive signal indeed derives from the OFC (Schoenbaum & Eichenbaum, 1995). This question might be addressed by a double-dissociation study examining the effects of OFC / piriform cortex lesions on coding in the unlesioned area. The generation of OFC prediction based on different types of memory (declarative, episodic, short-term, long-term) likely depends on interactions with the parahippocampal (with which the OFC holds direct connections, see above) and hippocampal formation (with which the OFC holds disynaptic connections, see above). Ramus et al. (2007) discuss the differential roles of OFC and perirhinal cortex in distinct types of memory. In DNMS (delay nonmatching to sample tasks) tasks, an animal is required to remember a ‘sample’ stimulus and decide whether a ‘choice’ stimulus, which is presented at a delay, matches the ‘sample’ stimulus. Rats with perirhinal cortex lesions fail to perform this task at larger delays. Interestingly, rats with lesions of the OFC are impaired on the performance of these tasks on shorter delays (Ramus et al., 2007). Ramus & Eichenbaum (2000) have shown that OFC responses depend on whether a presented ‘choice’ odor matches the ‘sample’ stimulus or not, indicating that sensory responses in OFC are affected by memory. Ramus et al. (2007) also inquired neuronal OFC correlates in an 8-odor sequence learning task. They showed that many OFC neurons fired in anticipation of the presented odor (about 200-400 ms before odor onset), i.e. generated sensory predictions. These anticipatory responses were not observed when the sequence of odors was random (Ramus et al., 2007). The required memory for generating these immediate sensory predictions may be partially derived from hippocampal inputs, as hippocampal lesions impair the learning of these odor sequences (Fortin et al., 2002). Together, these findings are congruent with the hypothesis that the OFC subserves immediate predictions of the incoming sensory data based on associative memory, and that predictions are not restricted to value-carrying outcomes.

1.2.5 The priors of OFC predictions

Another important aspect of predictions lies in the priors and memory that the system uses to generate predictions. Predictions are generated based on a memory of past events that is used to infer the future, but also on innate prior knowledge about the regularity of the physical
world that was shaped by evolution. The ‘memory priors’ can be short-lived or long-lived, they may have certain temporal integration constants, and they may contain knowledge of high-level or only first-order associations. For example, simple cue-response conditioning is a way of generating predictions, but it may fail if context or other rules need to be taken into account. The role of OFC in reversal learning suggests that the OFC has a clue about the validity of a certain rule given other rules, indicating that it represents not only first-order, but also higher-order associations. OFC may also use information about the current and predicted rat’s position from the parahippocampal formation to generate sensory and motor predictions (which conforms to its connectivity profile), and inputs from other frontal and parietal regions (that are involved in direct motor planning, which generates sensory and motor predictions in turn). For example, planning a movement of a hand that holds food towards the mouth region generates a prediction signal about the taste of food. Or, passing by a location where food is typically located, e.g. a pizza restaurant (derived from e.g. parahippocampal information) generates a sensory prediction of food (olfactory, gustatory, visual), but not if one knows that the restaurant is closed on Mondays (second-order association), unless one knows that it is a special Monday (e.g., because it is Christmas) (third-order association).

1.2.6 The utilization of OFC predictions

A final aspect of predictions to consider is the way that predictions are utilized by the system. In the predictive coding framework, the goal of the predictive system is to generate a state estimate that is as accurate as possible. If OFC generates a state estimate, then this state estimate can be used by other areas in different ways. It can be used to directly guide goal-directed behavior; it can be used by sensory areas to make coding sparse and efficient (Bastos et al., 2012; Rao & Ballard, 1999; Friston, 2008), and it can be used to drive learning in subcortical structures such as the amygdala and the striatum (Schoenbaum et al., 2009; Takahashi et al., 2011). In Schoenbaum et al. (2009)’s framework, the OFC predictions merely serve to drive stimulus-outcome associations in subcortical areas. The sensory predictions of the OFC might serve another important function though: They might be used to generate top-down predictions of sensory data that are used by sensory areas to generate sensory prediction errors (Bastos et al., 2012; Friston, 2008). This serves to generate new sensory predictions in the OFC, but it also serves to render sensory coding energy efficient, as unsurprising (low entropy) events do not need extensive updating of the representation of the state of objects, whereas surprising (high entropy) events lead to increases in activity as extensive updating of representations is required (Bastos et al., 2012; Friston, 2008). A large body of research has shown that sensory cortices show higher activity when the sensory input data is surprising (Bastos et al., 2012). Such a coding scheme is efficient from the perspective of Information Theory (Shannon, 1948). Thus, one strong prediction is that OFC lesions should disrupt signals of sensory prediction errors in structures such as gustatory and olfactory cortex. To our knowledge, this prediction has not been tested yet, although it is consistent with the strong reciprocal connections between OFC and sensory areas (see above). Another prediction is that while OFC lesions do not affect the acquisition of stimulus-outcome associations (Schoenbaum et al., 2009), they should affect the efficiency of coding in sensory cortices (especially gustatory and olfactory). This prediction is consistent with the finding that OFC lesions impair olfactory discrimination abilities (Eichenbaum et al., 1983, 1980).

Yet, an open question is why OFC cells, upon an experimental reversal of stimulus-
outcome contingencies, take longer to reverse their expected S+/S- outcome representations than cells in subcortical structures such as amygdala and striatum (Stalnaker et al. (2007); Schoenbaum et al. (2009, 2003a), cf. (Pasupathy & Miller, 2005)). That is, OFC cells are ‘wrong’ for a longer period of time than behavior or subcortical predictions are (Schoenbaum et al., 2009). There are three possible accounts for this phenomenon. First, it may be possible that the OFC needs to generate ‘incorrect’ predictions because its primary function is to enable the generation of prediction errors in subcortical areas like striatum and amygdala, which can be used to train the representations in these areas (Schoenbaum et al., 2009). This proposal is supported by the finding that OFC lesions impair reversal learning (Schoenbaum et al., 2003a). This would mean that the function of OFC is in fact to generate erratic sensory predictions, which seems odd at first glance. This account is not compatible with a function of the OFC in predictive coding: In predictive coding, higher areas try to generate a correct model of the state of objects, which is then updated according to the prediction errors that are generated by early sensory areas (Friston, 2008; Rao & Ballard, 1999). The goal of this predictive scheme is to generate an estimate of the current state of the world which is as accurate as possible.

A second possibility is that the OFC does not generate ‘incorrect’ predictions, but merely has a dominant prior in updating its predictions based on sensory data. The generation of predictions relies on the predictable behavior of physical objects and agents in the external world, making use of innate or acquired priors. In reversal learning experiments, the correct prior would be that stimulus-outcome contingencies can change suddenly and permanently, such that one should only consider the last few trials to generate correct predictions. The OFC may simply have the wrong prior about the physical laws of the laboratory experiment, e.g., the prior that associations do not suddenly and permanently change in the real world, but change gradually, which might be the right prior for the evolutionary environment in which rats evolved. Thus, the finding that the OFC generates ‘incorrect’ predictions might be due to a discrepancy between its priors of how the physical world works, and the priors that would generate correct predictions in the used experimental settings. The observation that subcortical structures adapt their predictions more rapidly (Stalnaker et al., 2007; Schoenbaum et al., 2003a, 2009; Pasupathy & Miller, 2005) may relate to these structures being more important for the control of immediate behavior, or because their predictions do not take uncertainty into account, i.e. they might threshold a slightly imbalanced uncertainty function to generate all-or-nothing predictions. This leads to the fundamental question that needs to be addressed in order to understand the function of the OFC, namely: Which priors does it use to generate sensory predictions? Does it generate ‘incorrect’ predictions (upon reversal learning), or does it use some priors that happen to be incorrect in the typical reversal learning experimental settings? To the author’s knowledge, this question has not been directly addressed to date. Yet, this account suffers from the problem that the OFC is supposed to encode higher-order associations, while it sticks to an ‘incorrect’ prior even after many experimental sessions.

A more plausible account is that the OFC neurons simply do not have to reverse their S+/S- representations literally, because most of its neurons might not have a fixed sensory receptive field, as the connections with their peers are highly plastic and associative. A given OFC neuron might simply not have the function to represent stable features of the world, as is implicitly assumed in Schoenbaum et al. (2009), but may rather be part of an associative network that adaptively changes the feature selectivity of its members. This idea is supported by the rather non-topographical nature of OFC representations (see Chapter 7). The question
is therefore not whether individual OFC neurons reverse their selectivity for the $S+/S-$ stimuli (representing a stable feature), but whether their ensemble responses discriminate between the $S+$ and $S-$ condition; a decoding/ensemble approach as in van Duuren et al. (2008, 2009) is therefore a critical analysis step. In fact, in Chapter 8 (van Wingerden et al., 2012) we will show that even single neuronal responses during the early reversal phase discriminate well between the $S+$ and $S-$ condition. It is therefore perhaps nonsensical to claim that the OFC representations are ‘incorrect’ after task reversal; they are only ‘incorrect’ insofar we assume that OFC neurons have a fixed feature selectivity. Feature selectivity can be a dynamic quantity if the OFC ‘knows’ how to broadcast its predictions to other sensory, frontal, and subcortical areas. Stable feature selectivity is not necessarily required for this broadcasting: the OFC may effectively use population coding to broadcast predictions ‘correctly’ to other sensory areas. In addition, OFC neurons do not merely care about the future, but part of the OFC cells might also carry information about immediate past (and hence fixed) events and would thus not reverse; another part of the OFC cells might merely be affected by the present input data, and not actively generate predictions about more distant future events at all. Obviously, these ideas require further modeling and investigation.

1.3 Anatomy and function of the barrel cortex

Rats have whiskers on their snouts that serve to explore nearby objects in their surroundings. The barrel cortex, i.e. S1BF (primary somatosensory cortex, barrel field), processes information about these whiskers and is an important model system for sensory processing because of its anatomical organization. In particular, each whisker is represented by a discrete structure in Layer 4 of the barrel cortex. These discrete structures have the appearance of a barrel - hence the name ‘barrel cortex’ (Woolsey & Van der Loos, 1970). These barrels are somatotopically arranged in a way that is similar to the physical topography of whiskers on the snout (Petersen, 2007; Diamond et al., 2008). Two further reasons for the popularity of the barrel cortex as a model system are that (1) it allows easy manipulation of sensory inputs, by removal of one or more whiskers, but also by stimulation of the whiskers, and (2) that the whisking system is an ‘active’ sensory system, in the sense that rats make sweeping movements with their whiskers to explore their environment, making the whisking system an excellent model system to study sensorimotor integration and the influence of motor planning on sensory representations. Responses of cells in the barrel cortex are quite different from responses in the trigeminal ganglion at the periphery: (1) They show higher trial-by-trial variability, probably driven by interactions with spontaneous cortical activity; (2) ‘spatial’ receptive fields are broader in barrel cortex, with units often responding to movements of nearby vibrissae (Petersen, 2007; Diamond et al., 2008). In fact, voltage sensitive dye imaging shows that receptive fields are quite restricted 10 ms after whisker deflection, but that responses propagate to nearby barrel columns in the subsequent milliseconds (Ferezou et al., 2006). Thus, a primary function of barrel cortex may be to form a coherent object representation based on tactile information from multiple whiskers (Petersen, 2007). It should be noted though that subthreshold responses (membrane potentials) have broader receptive fields than suprathreshold responses (spikes), which may reflect a tip-of-the-iceberg phenomenon (Petersen, 2007).

Rats make characteristic whisking movements with a speed of about 3 to 20 whisks per
second. Interestingly, carnivores (cats, dogs) have whiskers as well, but do not make sweeping whisking movements, perhaps because of their superior vision capacities, and absence of subterranean behavior. Many behavioral studies have shown that macrovibrissal information can be used to localize objects (Brecht, 2007; Diamond et al., 2008). In particular, rats can perform gap measurement, gap jumping, distance perception, and can distinguish the orientation of objects. A major function of the whisking system may thus be to perceive depth and determine object location and shape (Brecht, 2007; Diamond et al., 2008). Another line of research has shown that the macrovibrissae can be used for texture discrimination, in the same way as humans use their fingertips. This has been shown by using discrimination tasks with rough and smooth textures (von Heimendahl et al., 2007; Diamond et al., 2008). In fact, the tactile acuity of rats is comparable to humans, and neurons in the barrel cortex carry much information about texture (von Heimendahl et al., 2007).

Whole-cell recordings in awake animals have shown that active whisking has a major impact on the intracellular membrane potential characteristics of barrel neurons. In particular, during quiet wakefulness, membrane potentials show relatively large, low-frequency fluctuations. During whisking, the membrane potential characteristics change completely, with neurons depolarizing and membrane potentials having a smaller variance (Crochet & Petersen, 2006; Gentet et al., 2012). However, neuronal firing rates do not increase dramatically (Crochet & Petersen, 2006). The cortical state during whisking periods can be compared to what has been called an ‘active’ state, in which there is network desynchronization, as opposed to a state of quiet wakefulness or slow wave sleep, which is often referred to as a synchronized state (Harris & Thiele, 2011). An example of an active state in primates is the cortical state during active vision, in which LFP fluctuations are desynchronized together with the emergence of fast gamma oscillations (Harris & Thiele, 2011). During quiet wakefulness, large amplitude alpha fluctuations occur. While membrane potentials desynchronize during active whisking (Crochet & Petersen, 2006), it is unknown whether gamma oscillations occur during the ‘active’ state in the barrel cortex. This question will be addressed in Chapter 6.

A recent anatomical connectivity study has shown that there are prominent reciprocal connections between S1BF and secondary somatosensory cortex, motor cortex, perirhinal cortex, and thalamus. In addition, the barrel cortex projects to many subcortical structures (Aronoff et al., 2010). Projections to the perirhinal cortex are bilateral, but weaker to the contralateral hemisphere. This projection to the perirhinal cortex - which has direct connections with CA1 and the subiculum (Naber et al., 1999) - may underlie the integration of tactile information in higher (parahippocampal and hippocampal) level structures involved in memory and object representation. Indeed, tactile responses occur in the hippocampus (Pereira et al., 2007; Itskov et al., 2011). It is unknown though what the functional connectivity - in terms of neuronal coherence - is between S1BF and perirhinal cortex. Interestingly, strong tactile driven responses occur in rat V1 (Vasconcelos et al., 2011; Iurilli et al., 2012). These may rely on known disynaptic (with few monosynaptic) anatomical projections between S1BF and V1 in the rat cortex or thalamic afferents (Paperna & Malach, 1991; Avanzini et al., 1980). Again, the functional connectivity - in terms of neuronal coherence - between S1BF and V1 is unknown.
1.4 Aims of thesis

This thesis has two main aims.

Aim 1—the first aim of this thesis is to address four methodological problems that are central to neuroscience, and important for our experimental applications, which will be presented in the second part of this thesis. First, we address the problem that measures of spike-EEG or EEG-EEG (or MEG-MEG) coupling can suffer from strong statistical bias (Chapters 2 and 3). Statistical bias refers to a systematic misestimation of a statistic when the number of independent observations (sample size) is small. Chapter 2 will deal with this problem in the context of quantifying the strength of spike-EEG phase-coupling. Chapter 3 will deal with this problem in the context of quantifying the strength of EEG-EEG or MEG-MEG phase-coupling. The solution to this problem opened new possibilities to study rhythmic synchronization in areas OFC (Orbitofrontal cortex) and S1BF (Barrel Field of the Primary Somatosensory Cortex) (Chapters 6 to 8). Second, we study the problem that measurement noise, in particular instantaneously mixed noise (‘volume conduction’), can strongly affect EEG-EEG (or MEG-MEG) connectivity measures. Chapter 3 studies the impact of noise on connectivity measures (which do not allow causal inference) like coherence, phase locking value (Lachaux et al., 1999), the imaginary component of the coherence (Nolte et al., 2004), and the phase lag index (Stam et al., 2007), and constructs a new measure of coherence based on the imaginary component of the cross-spectrum (WPLI) that is claimed to be more robust to noise. This technique is applied to study rhythmic synchronization in area S1BF (Chapter 6). Third, we study the problem that measurement noise can strongly affect measures of causal directionality (Nalatore et al., 2007; Newbold, 1978; Nolte et al., 2008). We evaluate the validity of some new measures of causal inference like phase slope index (Nolte et al., 2008), and show the impact of independent and dependent measurement noise on measures of causality. We propose a new criterion on the cross-covariance function that can strongly reduce the expected number of false positive detections of causal directionality (Chapter 4). Fourth, we consider the problem that estimators of the Shannon entropy and Shannon mutual information (Shannon, 1948) can be strongly biased by sample size, which is a major obstacle in applying the powerful mathematical framework of Information Theory (Shannon, 1948) to neuroscience data. This problem will be addressed in Chapter 5.

Aim 2—the second aim of this thesis is to apply these statistical tools to our own experimental data in order to further our understanding of several neurophysiological problems. In addition, we aim to perform large-scale recordings from multiple brain areas simultaneously (Chapter 6). In particular, we are interested in the mechanisms of multisensory integration, hippocampal-neocortical communication, and the mechanisms and functions of oscillatory processes in the brain. Chapter 6 investigates function and mechanisms of gamma-band oscillations in the barrel cortex, hippocampus, visual, and perirhinal cortex, and investigates long-range coherence between these brain areas in the gamma-frequency range. Chapter 7 investigates the functional role and mechanisms of gamma-band oscillations in the OFC, an area in which gamma-band oscillations had not yet been investigated. Chapter 8 investigates the functional role of the NMDA receptor in controlling OFC firing rate representations and oscillations, which can provide further insight into the mechanisms and functions of rhythmic synchronization.
1.5 Questions addressed in the experimental chapters

We will now outline the questions that we address in the experimental chapters.

- A first and basic question: Is gamma-band synchronization indeed a widespread phenomenon in cortex? Most studies concerned with gamma-band synchronization and local spiking have focused on the hippocampus (Bragin et al., 1995), visual cortex (Gray et al., 1989) and olfactory system (Eeckman & Freeman, 1990; Wehr & Laurent, 1996). In Chapters 6 to 8, we ask whether band-limited gamma synchronization occurs in area S1BF (Primary somatosensory cortex, Barrel Field), OFC (Orbitofrontal cortex), and perirhinal cortex (area 35/36). It has not been demonstrated previously that band-limited gamma synchronization occurs in these structures. This question is critical from both functional and mechanistic points of view. Mechanistically, the hypothesis that activation of FS PV+ basket cells leads to the generation of gamma oscillations predicts that gamma oscillations are a widespread phenomenon in the cortex that can be generated in any structure given the right set of inputs. Functionally, the hypothesis that gamma oscillations are a fundamental component of cortical computation (Fries, 2009) predicts that gamma oscillations play a functional role in all cortical structures. If not, it would point to a more specialized role for gamma oscillations in a subset of cortical structures.

- To what extent is the integration of visual-tactile information subserved by long-range gamma synchronization? Gamma coherence has been proposed to be a generic mechanism for communication between brain areas (Fries, 2005), and has been hypothesized to play a role in multi-modal integration (Pennartz, 2009; Engel et al., 2012; Singer, 1999). We recorded from areas that have known mono- and disynaptic anatomical connections to S1BF (V1M, Perirhinal, CA1). There exist mono-synaptic connections between S1BF and perirhinal cortex (Aronoff et al., 2010; Naber et al., 2000), which may underlie tactile responses in the CA1 area of the hippocampus (Pereira et al., 2007), which has mono- and disynaptic connections to the perirhinal cortex in turn (Naber et al., 1999). Further, tactile driven responses occur in rat V1 (Vasconcelos et al., 2011), and there are known disynaptic (with few mono-synaptic) anatomical projections between S1BF and V1 in the rat cortex (Paperna & Malach, 1991). Our data show that S1BF gamma is not phase synchronized with gamma oscillations in these other areas, at least not in the behavioral setting that we studied.

- What is the role of gamma synchronization in decision making and learning of stimulus-outcome associations? The OFC has been hypothesized to play a critical role in these functions (Schoenbaum & Roesch, 2005). In contrast to sensory processing and attention, gamma oscillations have been barely studied in the domain of emotional processing and valuation of sensory signals and actions. Thus, we investigated gamma oscillatory activity in the rat orbitofrontal cortex (OFC), a prefrontal structure implicated in making behavioral adjustments to changing reward contingencies (Schoenbaum et al., 2009) and control over prepotent, impulsive responses to incentive stimuli (Jones & Mishkin, 1972; Chudasama et al., 2003; Winstanley et al., 2004; Man et al., 2009), but see Chudasama et al. (2007). OFC neurons have been shown to flexibly encode representations of stimulus-outcome and action-outcome associations (Schoenbaum et al.,)
1998; Tremblay & Schultz, 1999; Wallis & Miller, 2003; Padoa-Schioppa & Assad, 2006; van Duuren et al., 2008, 2009). Although gamma oscillatory activity has been previously reported for human, monkey and rat OFC (Nishida et al., 2004; Liu et al., 2005; Sun et al., 2006) the question arises whether gamma oscillations exert similar functions in emotional decision-making and valuation as suggested by studies on sensory processing. The existence of extensive connections of the OFC with other brain areas suggests that rhythmic synchronization is an interesting candidate to coordinate communication with other brain areas. Also, learning of stimulus-outcome associations may imply rhythmic synchronization, being implicated in the potentiation of synapses through spike-timing-dependent-plasticity (Jensen et al., 2007; Sejnowski & Paulsen, 2006; Fell & Axmacher, 2011; Jutras & Buffalo, 2010; Vinck et al., 2010a).

- What is the role of the NMDAR receptor in regulating rhythmic synchronization in OFC, and in regulating the coding of stimulus-outcome associations in OFC? Gamma synchronization is generated by an interaction between inhibitory and excitatory neurons. A prominent excitatory current is mediated by the NMDA receptor. However, the role of this current in sustaining gamma-band synchronization is only poorly understood. Furthermore, gamma synchronization may cause synaptic potentiation mediated by the NMDA receptor, as reviewed above. Finally, the formation and adaptation of stimulus-outcome associations may rely heavily on the NMDA receptor. We will explore these aspects in Chapter 8.

- To what extent are different neuron types engaged in the gamma rhythm? To understand the mechanisms and functions of gamma synchronization, it is critical to establish which cell types are involved in their generation, and which cell types exhibit gamma-synchronized behavior. In awake recordings, this question has thus far only be addressed in the hippocampal formation (Csicsvari et al., 2003). Changes in behavioral state have powerful effects on cortical dynamics and brain rhythms (Buzsáki, 2006), and changes in the temporal dynamics of inhibitory interneurons are a prominent candidate mechanism to explain state-dependent cortical dynamics (Buzsáki, 2006). We address this question by separating different neuron types according to action potential waveform characteristics, and examining the extent to which cells are entrained by the gamma rhythm (Chapters 6 to 8). Neurons in the mammalian brain can be subdivided into excitatory and inhibitory cells. A subset of these inhibitory cells, fast spiking, parvalbumin expressing basket cells and somatostatin-expressing cells, has been shown to have narrow action potential waveforms, while excitatory pyramidal cells have broad waveforms (Gentet et al., 2012) (see Chapter 6 for an in-depth overview of the relationship between action potential waveform characteristics and cell classes). We will use this distinction to classify cells based on extracellular data.