Summary

Olfactory deficits in Parkinson's disease (PD) were first empirically documented in 1975 by Ansari and Johnson. Over the ensuing years it has become clear that most PD patients have olfactory disturbances that are not restricted to a single functional modality. Even in early stage, untreated PD patients, deficits in olfactory function have been demonstrated, which is supported by recent neuropathological studies demonstrating that the olfactory bulb and anterior olfactory nucleus structures may be among the induction sites of PD pathology. In later pathological stages, the olfactory bulb and tract are among the brain regions where Lewy bodies and Lewy neurites, the characteristic neuropathological features of PD, are particularly abundant. Since impairments in the sense of smell may even precede the development of overt motor symptoms, olfactory testing could prove valuable in establishing an early diagnosis of PD when other clinical (motor) symptoms are not apparent yet. Also in the early clinical motor stages of PD, olfactory testing may be useful as a diagnostic tool, both for distinguishing between PD patients and controls, and in differentiating between PD and other neurodegenerative disorders. Furthermore, the pathophysiology underlying the olfactory deficits in PD is far from being elucidated.

The following research questions were addressed in this thesis:

- What is the prevalence and nature of impairments in the different specific olfactory modalities in PD and how do they relate to other (motor and non-motor) disease characteristics?
- Which (combination of) olfactory test(s) is best in discriminating PD patients from control subjects?
- Is it possible to explore the neurophysiological basis of olfactory (dys)function by means of magnetoencephalography (MEG) in healthy controls and PD patients?

Prevalence and nature of olfactory deficits in PD

The "Sniffin' Sticks" is a multimodal olfactory test battery that can be used to assess three different aspects of olfactory function: odour identification, discrimination and detection, each consisting of 16 items.

In **chapter 1**, we provided age-specific normative values for the Dutch population (over 45 years of age) for the two culture-dependent components of the "Sniffin' Sticks" test battery: odour identification and odour discrimination. In **chapter 3,** we used these age-dependent normative values to study the prevalence of deficits on the odour identification and discrimination task in a large population of Dutch PD patients from two university medical centres. The prevalence of an olfactory deficit in PD patients on any of the two tasks in this study was 73%. In **chapter 2**, we assessed the prevalence of olfactory

deficits (odour identification, discrimination and detection threshold) in PD in a large sample of PD patients from three populations in Australia, Germany, and the Netherlands. When we applied age-independent criteria for hyposmia, only 3.3% out of a total of 400 PD patients were normosmic. However, when applying age-specific criteria, as we did for the Dutch cohort in **chapter 3,** 25.5% of patients were normosmic. From these data we concluded that, apparently, a significant minority of PD patients does not suffer from olfactory dysfunction.

The results in **chapter 3** further demonstrate an impairment in odour identification in 65% of PD patients relative to the performance of controls, and an impairment in odour discrimination in 42% of patients. The results described in **chapter 5** indicate that PD patients have a slight impairment of odour recognition memory that appears to be fully accounted for by an increase in odour detection threshold. Taken together, these findings argue against the notion that the olfactory impairments in PD would be based on a single common underlying deficit, such as an increased odour detection threshold, but suggest that olfactory dysfunction in PD entails a disturbance of multiple, but not all, olfactory modalities.

Relationship between olfactory (dys)function and other disease characteristics

Since approximately 25% of PD patients do not appear to have olfactory deficits (see above), olfactory function might contribute to the phenotypic characterization of PD patients. Therefore, we wanted to determine the relationship between the different olfactory modalities and other PD characteristics.

The results described in **chapter 3** show that odour identification performance in PD is related to age and sex, but independent of disease duration or stage. By contrast, odour discrimination performance was found to decrease with disease duration in PD.

Chapter 4 addresses the relationship between olfactory impairment and other aspects of phenotypical heterogeneity among PD patients. Apart from the above-mentioned association between odour discrimination deficits and disease duration, there were no other significant correlations between olfactory function and motor or (other) non-motor symptoms in PD, such as cognitive status, psychiatric complications, sleep or autonomic function. Moreover, there were no significant differences in olfactory test scores (either measured as a combined test score or each of three olfactory modalities separate) between patients with different motor phenotypes (tremor-dominant, akinetic-rigid, postural instability gait difficulty or mixed (**chapters 2 and 4**).

Diagnostic value of olfactory testing in PD

The results of the studies described in **chapters 2 and 3** show that odour identification is more frequently impaired in PD than odour discrimination and odour detection, and that an odour identification test allows a better differentiation between patients and controls.

Odour recognition memory, was not independently impaired in PD (**chapter 5**), and is therefore not useful as a diagnostic tool to differentiate between PD patients and control subjects.

In **chapter 6**, we used extended versions of the odour identification and discrimination parts of the "Sniffin' Sticks" and found that adding more items within a single olfactory modality does not improve the diagnostic accuracy of these tests. By contrast, combining different olfactory modalities did increase diagnostic accuracy. A combination of an odour identification and a detection threshold task turned out to be the best in differentiating between PD patients and control subjects.

Neurophysiological studies of olfactory function

In **chapter 7**, we determined the number of chemosensory stimuli needed to obtain an optimal signal-to-noise (S/N) ratio for studying olfactory event-related responses by means of an olfactometer and electroencephalography (EEG) in healthy controls. The S/N ratio of olfactory and trigeminal event-related potentials significantly improved up to 60-80 stimuli, mainly due to a reduction of the noise level. However, in a pilot study involving both healthy controls and PD patients, applying our EEG results to MEG, we were unable to obtain consistent olfactory event-related magnetic fields (*unpublished observations*).

Therefore, we changed focus towards time-series analyses of MEG data instead, as a means to gain more insight in the neurophysiological aspects of olfactory information processing in healthy controls and the pathophysiology of olfactory dysfunction in PD. **Chapter 8** describes the results of a study in which we were able to show for the first time that time-series analysis of MEG data, including spectral power and synchronization likelihood (a general measure of functional connectivity between brain areas), can be used to detect odour-induced changes in brain activity in healthy subjects. In addition, we found differences in odour-induced changes in brain activity between PD patients and controls using analysis of functional connectivity, but not of spectral power. These differences in functional connectivity may reflect abnormal olfactory information processing in PD patients that leads to the clinically observed olfactory impairments.

General discussion

In the general discussion, the data presented in the various chapters of this thesis were combined and a consideration of the potential implications as well as future research perspectives was provided. The most striking observations from the first two sections of this thesis are A) that apparently approximately 25% of PD patients do not suffer from olfactory dysfunction, B) that the impairment of olfactory function in PD entails a disturbance of multiple, but not all, olfactory modalities, and C) that a combination of an odour detection threshold test and an identification test is the best in distinguishing PD patients from controls. Furthermore, differential characteristics of the odour identification

and discrimination deficits in PD suggest that these olfactory modalities involve at least partly differential components of the olfactory information processing system.

From the last section, we can conclude that time-series analysis of MEG data is a suitable method to study odour-induced changes in brain activity. In addition, differences in odour-induced functional connectivity were found between PD patients and controls. The results obtained may be used in future olfactory neuroimaging studies to further investigate the pathophysiology of olfactory dysfunction in PD, in particular moving beyond the mere administration of odorants to the use of more complex tasks, such as odour identification or discrimination.