# Summary

This thesis describes the role that pharmacological 'challenge'-models could have in the development of new drugs for psychosis and schizophrenia. A pharmacological challenge consists of the administration of a pharmacologically active compound, such as a drug, with the purpose of influencing an underlying biological system. Subsequently, the changes on certain outcome measures (this could be hormones, but also neuropsychological tests) are observed. The relation between these outcome measures and the amount of administered drug (or the changes in drug concentrations in plasma) provides information on the function of the underlying system.

## *Chapter 1: Introduction*

The first drug that was used for the treatment of psychosis is chlopromazine (market under the name 'Largactil' in Europe and 'Thorazine' in the United States). The effectiveness of this drug in treating psychosis was first investigated in 1952 in a Parisian mental asylum. The discovery of this drug is sometimes referred to as 'the French revolution of 1952', because of the unmistakable change it brought about in psychiatry. It was also the beginning of psychopharmacology.

In the late 1960s the dopamine hypothesis of schizophrenia was formed. This hypothesis described schizophrenia as the result of a disturbance of the neurotransmitter dopamine. It was formulated when it became known that the new drugs for the treatment of psychosis blocked the dopamine receptors in the brain and that the administration of amphetamine (which causes an increase in dopamine levels) induces symptoms that resemble psychosis.

The development of clozapine (marketed as Leponex) in 1966 led to a new change in the treatment of psychosis. Clozapine acts on many other receptors (such as serotonin) and has a relatively low affinity for dopamine receptors. This led to the development of many 'atypical' antipsychotics. Clozapine remains the only drug with a proven superior efficacy in the treatment of psychosis, but it is also associated with rare but severe side-effects (i.e. agranulocytosis).

Even though dopamine is not the only neurotransmitter involved in schizophrenia, dopamine does have a major role. All currently available drugs have a dopaminergic mechanism of action (to some extent), which is related to their antipsychotic potencies. Available animal models of psychosis have mainly been validated with dopamine antagonists. In this way, positive effects of dopaminergic antipsychotics in dopaminergic animal models have become self-comfirmatory. This thwarts the development of drugs with other mechanisms of action.

The use of pharmacological challenge models could constitute a solution. There are different compounds, with different mechanisms of action that can induce psychomimetic effects (resembling psychosis). These compounds are (also) often used as recreative drugs, like amphetamine (dopaminergic), LSD (serotonergic), ketamine (glutamatergic) and THC (cannabinoid). These compounds can be used to induce psychomimetic symptoms in healthy volunteers. These drug-induced symptoms could constitute a model for psychosis.

### *Chapter 2: Does olanzapine inhibit the psychomimetic effects of THC?*

In this chapter, THC (the active ingredient of cannabis) is used to induce psychomimetic symptoms in healthy volunteers. Previous studies with this model have shown that THC can be used to achieve this effect, and that it can be reversed by the 'classic' antipsychotic haloperidol. The most important aim of this study was to investigate whether the 'atypical' antipsychotic olanzapine was also able to reduce these psychomimetic effects. As a positive control for the histaminergic effects of olanzapine (i.e. somnolence) diphenhydramine was added to the study design.

Forty-nine healthy male volunteers participated in this study. The study consisted of five study days that were similar, except for the combination of drugs that were administered. These combinations were: (1) only THC, (2) THC preceded by 10 mg of olanzapine, (3) THC preceded by two doses of

15 mg diphenhydramine, (4) only 10 mg of olanzapine, and (5) only placebo. The study was blinded and placebo-controlled, which means that nobody was aware of the order of treatments during the execution of the study and that a placebo was used if a certain treatment was not given. Baseline measurements were performed in the morning, after which the oral medication (olanzapine, diphenhydramine or placebo) was administered. In the afternoon three balloons filled with THC or placebo (subsequently 2 mg, 4 mg and 6 mg with a 90 minute interval) were administered. In the afternoon many outcome measures were tested again to measure the effect. The most important outcome measure was the PANSS, an interview that is being used to quantify the severity of symptoms in patients with schizophrenia.

As expected, administration of THC resulted in a significant increase on the positive subscale of the PANSS (a measure for hallucinations and delusions) of 20.6%. If this was preceded by administration of olanzapine, THC only increased the positive PANSS by 13.7%. This was a statistically significant reduction in psychomimetic symptoms ($p < 0.001$). This reduction is slightly smaller than previously found for haloperidol. The study confirmed that THC can be used as a model for psychosis and that this model is sensitive for the effects of antipsychotic drugs with (partly) different mechanisms of action.

### *Chapter 3: Profiling the subjective effects of THC using VAS-scales.*

The study described in Chapter 2 also showed that measuring psychomimetic effects is complicated by non-linear responses, multiple dimensions and non-responders. Subjective drug effects are often measured with different visual analogue scales (VAS), which typically consist of a 10 cm line, with a subjective feeling on both sides (for example 'alert' and 'drowsy'). Other more unsual subjective effects are measured on unipolar VAS-scales, where the intensity is rated from absent (0 mm) to extreme (100 mm). A subject is asked repeatedly throughout a study day to indicate his or her feelings on

these lines. During the analysis described in this chapter, the underlying structure in the reaction on two often-used sets of VAS-scales after administration of THC was examined.

To this end, the data from ten studies, performed at CHDR, were used. In all these studies, THC was administered to healthy volunteers (217 in total). There were 29 different VAS-scales: 16 describing mood, alertness and calmness and 13 describing psychedelic effects. Different statistical methods were employed to discover underlying patterns in the response on these VAS-scales. Each method has its own advantages and this analysis focussed on the similarities between these methods.

This resulted in three effect clusters that describe the subjective effect of THC: 'perception', 'relaxation' and 'dysphoria'. The cluster 'perception' includes the 'high' feeling of THC, changes in time perception and the control of one's own thoughts. 'Relaxation' describes the mental relaxation that is experienced after administration of THC. The cluster 'dysphoria' consists of more negative reactions that occurred in a small subset of subjects. This includes the hearing of voices, suspicion or delusions. These symptoms were, when present, of mild severity. These VAS-scales probably did not include all subjective effects of THC, and may for instance have missed feelings like hunger or craving, or aspects of anxiety and panic. Still, the composite scales encompass a range of distinguishable THC-effects which cover most feelings associated with psychotic-like states. These clustered scales can thus be used to investigate factors that are associated with psychomimetic propensity, or effects of antipsychotic drugs.

### *Chapter 4: The influence of personality on the sensitivity to subjective effects of THC.*

Using the three subjective effect clusters identified in the previous chapter, the sensitivity of mild cannabis users to THC was investigated. This chapter once again used data collected in previous studies. Data of seven previous studies, with administration of THC by inhalation, were combined.

Previously a pharmacokinetic model, describing the course of THC-concentrations in plasma, was made using these data. This model was extended by describing the concentrations of the active metabolite (11-OH-THC). Also, a mathematical model was developed to describe the relationship between THC plasma concentrations and the three clusters of subjective effects.
This mathematical model can be used to estimate individual parameters that describe the sensitivity to THC for each subject. This measure of sensitivity was then compared to scores on different subscales of a personality questionnaire (the TCI) using multiple linear regression. It was found that the cluster 'perception' is influenced by the personality trait 'harm avoidance' and the cluster 'dysphoria' by 'self-transcendence'. According to the underlying psychobiological model of personality the personality trait 'harm avoidance' is regulated by the neurotransmitter serotonin. Given the effects of serotonergic drugs like LSD and psilocybin (the active compound of magical mushrooms), it is not surprising that this trait is related to changes in 'perception'. The endocannabinoid system regulates and fine-tunes the action of several neurotransmitter systems, including serotonin. Higher scores on 'self-transcendence' are associated with schizotypy and it is therefore not surprising that people with higher scores are more sensitive to the dysphoric effects of THC. These results provide insight into the neuropharmacological systems underlying personality traits.

### *Chapter 5: Optimizing the glutamatergic challenge model of psychosis, using S(+)-ketamine to induce psychomimetic symptoms in healthy volunteers.*

Ketamine is an NMDA-receptor anatagonist that is mainly used in anaesthesiology. Since the drug has psychomimetic side-effects at lower concentrations, ketamine is also used as a psychosis model. The studies describing this model use different doses of ketamine, only measure the

effect on a single timepoint (at different times relative to administration) and do not measure ketamine concentrations in plasma. Therefore, this study further explored the ketamine model of psychosis, with the aim of describing the relations between ketamine concentrations in plasma, and the psychomimetic effects on one hand and the undesirable effects (that affect tolerability) on the other hand.

This study consisted of three study days, during which a constant S(+)-ketamine concentration in the plasma was obtained (a high target concentration, a low target concentration and placebo respectively). This was accomplished by calculating an infusion scheme using a previously described pharmacokinetic model. In the first part of the study the target ketamine concentrations were 360 ng/mL, 180 ng/mL and 0 ng/mL. The high concentration in particular led to many side-effects and the study was temporarily halted to revisit the target concentrations. In the first part of study six male subjects were dosed. After the temporary halt, the study was continued with target concentrations of 240 ng/mL, 120 ng/mL and 0 ng/mL. This revised infusion regimen also incorporated the accumulation of the active metabolite norketamine, which has roughly a third of the activity of the parent compound. Thirty subjects (15 males and 15 females) participated in this second part of the study. As was done in Chapter 2, many different measurements were performed before, during and after the infusion and the PANSS was again selected as main outcome measure.

A dose-dependent, robust increase in psychomimetic symptoms was seen on all subscales of the PANSS. The positive PANSS was increased by 43.7% for the 120 ng/mL concentration and by 70.5% for the 240 ng/mL concentration relative to placebo. Most other outcome measures also showed a dose-dependent increase in effect. Based on the occurrence of side-effects (and thus tolerability) a ketamine concentration of about 200 ng/mL is advised for the use of ketamine as a psychosis model. The concentration-range is sufficient to induce psychomimetic symptoms and provides enough possibility to reduce the effects with antipsychotic drugs.

### *Chapter 6: Development and evaluation of a new VAS for psychedelic effects.*

As was already discussed in Chapter 3, VAS-scales are often used to quantify subjective feelings. However, there are several limitations to these VAS-scales. For example, psychedelic effects are absent under normal conditions, which leads to a score of 0 (without much variation) during placebo treatment. Furthermore, there appears to be a 'threshold' in psychedelic effects. For these reasons, a new set of VAS-scales has been developed to measure psychedelic effects. It was decided not to measure one effect per scale (ranging from 'not at all' to 'extremely'), but to measure two opposing subjective effects (e.g. from 'I feel sad' to 'I feel happy').

This new set of VAS-scales was measured during the study described in Chapter 5, together with the original set and several comparable questionnaires. During the evaluation, it was first examined which items responded to the ketamine challenge. Thereafter all items that responded were analysed for underlying patterns of effect, which resulted in two subscales. These subscales were not superior in the discrimination between the different target concentrations of ketamine. Furthermore, these subscales were more focussed on mental and physical sedation and not as much on the typical psychomimetic effects, which the scales were intended to measure. It is therefore not recommended to use these new VAS-scales in clinical trials.

### *Chapter 7: Subjective effects of ethanol, morphine, tetrahydrocannabinol and ketamine following a pharmacological challenge are related to functional brain connectivity.*

To investigate which areas of the brain are involved in the origin of psychomimetic effects of a pharmacological challenge, the data of previous studies with a pharmacological challenge with ethanol (alcohol), morphine, THC and ketamine were compared. In all these studies, changes over time were measured using an MR-scanner during rest. These measurements in

the MR-scanner can be used to describe 'functional connectivity', which is a measure for the synchronicity of activation or deactivation of different brain areas. All these studies also measured the subjective effects using VAS-scales, following the pharmacological challenges.

For each drug, data of 12 healthy, male participants were available. The changes in functional connectivity were related to the changes in the three subjective clusters described in Chapter 3 ('perception', 'relaxation' and 'dysphoria'). The analysis resulted in a cluster, located in the posterior cingulate gyrus and the precentral gyrus, with a statistically significant relation between changes in functional connectivity and changes in subjective 'perception'. This area of the brain was previously found to show a relation between functional connectivity and positive psychotic symptoms (e.g. hallucinations and delusions) in patients with schizophrenia.

### *Chapter 8: Discussion*

Different pharmacological challenge models can be used as model for psychosis and as model for antipsychotic drug activity. These pharmacological challenge models act through different neurotransmitter systems. The psychomimetic symptoms induced by all these models can be reduced by antipsychotic drugs. It is important to note that these different neurotransmitter systems do not operate independently, but they are related through complicated interactions. Different hypotheses on the pathophysiology of schizophrenia have been formulated that unite the role of these different neurotransmitter systems. The role of dopamine in these hypotheses is different, but it is clear that other neurotransmitters play a role in the development of schizophrenia.

Professor van Gerven (my doctoral supervisor) described five requirements for pharmacological challenges during his inaugural address (latin: oratio).

First of all, a good model of a disease should resemble the symptoms of the disease. The psychomimetic symptoms that were induced by THC and

ketamine have unmistakable similarities with the symptoms of psychosis. There are also differences between the two. For example, subjects remain insight in the development of psychomimetic symptoms, whereas patients loose insight when they become psychotic. If a subject is not explained that they receive a psychomimetic drug, the presentation is similar to the clinical presentation of (acute) schizophrenia. Also, the relative frequency of acoustic and visual hallucination is different for subjects and patients, which might be explained by underreporting of hallucinations by patients. The fact the psychomimetic symptoms of a pharmacological challenges can be reduced by antipsychotic drugs is a strong argument in favour of the validity of the pharmacological challenge as model for psychosis. The finding that the brain area that shows a relation between functional connectivity and psychomimetic symptoms also shows a relation between functional connectivity and positive psychotic symptoms in patients with schizophrenia, further supports the role of pharmacological challenges as model for psychosis.

The second requirement for a pharmacological challenge is a known pharmacological mechanism of action. Both THC and ketamine have known but different mechanisms of action and pharmacokinetic models have been described. This thesis also presents a pharmacokinetic / pharmacodynamic model for the subjective effects of THC, which is an indication that these subjective effects are pharmacologically related to (endo-)cannabinoid activity. Such a PK-PD model has not yet been developed for ketamine, although Chapter 5 provides the necessary data for such a model.

The third requirement is that the evoked effect should be quantifiable. In this thesis, much emphasis was placed on this aspect of challenge test development. Psychosis is a complex condition, which includes shifts of normal aspects of consciousness and behaviour to abnormal extremes, as well as the occurrence of novel subjective experiences that do not normally occur in healthy subjects. This mix of quantitatively and qualitatively abnormal mental states creates difficulties for measurement instruments suitable for research. It is an important requirement for useful psychomimetic challenge tests that subjects remain aware of their altered state of mind.

Because subjects retain insight, it is easier for them to share their experiences with the investigator and perform different measurements to quantify the evoked effect. The PANSS appeared to be a suitable method to quantify psychomimetic effects. As this is a clinically validated scale, the evoked symptoms are easily related to the symptoms of schizophrenia. The VAS-scales are also suitable to measure subjective effects of a pharmacological challenge. This thesis further substantiates this claim by the introduction of three clusters of subjective effects, which have also been described using a pharmacokinetic / pharmacodynamic model. An attempt was made to create an improved VAS for psychedelic effects, which was unsuccessful.

The fourth requirement for a pharmacological challenge test is the presence of a dose- or concentration-dependent effect. For the effects induced by THC and ketamine a relation with drug concentration was established.
The fifth requirement is that a pharmacological challenge should be safe and tolerable. At the doses of THC used during the different studies, THC was found safe and tolerable. In the first part of the study with the ketamine model, the highest target concentration was no longer tolerable. After an adjustment of the dose, a clear range in concentrations could be identified in which ketamine administration is tolerable, while producing the desired psychomimetic effects. Therefore, both models also meet this requirement.

## *Conclusion*

Pharmacological challenges with psychomimetic drugs may not fully represent the complexity of (chronic) schizophrenia, but constitute a useful model for psychosis and antipsychotic drug action. This thesis describes the use of THC and ketamine as models and explores different outcome measurements that can be used to quantify psychomimetic effects, in particular VAS-scales and FMRI. The phenomenological and pharmacological basis of psychomimetic challenge models were discussed. Well-controlled circumstances and repeated measurement of drug concentrations and effect greatly benefit the use of pharmacological challenges in drug development.