Synthesis of Molecular Probes for Exploring the Human Consciousness, 5-HT₇ Ligands and Salvinorins

PÅR HOLMBERG
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Abstract


In this study, we have addressed the serotonergic and the opioid system within the CNS. Both systems are of outmost importance in the etiology of disease states, especially mental disorders.

In our investigation of the serotonergic system, we have synthesized novel enantiomerically pure 6-aryl-3-amino- and 8-aryl-3-aminochromans as ligands for the 5-HT, receptor. One reason for the lack of understanding of the physiological functionality of the serotonin 5-HT, receptor, the most recently discovered member of the serotonin receptor family, is the absence of partial agonists and agonists. In this series, we have identified partial agonists with more than 189 fold selectivity over the 5-HT, receptor and one agonist with 29 fold greater selectivity over the serotonin 5-HT, receptor. Thus the present series constitutes a starting point for developing highly selective ligands for the 5-HT, receptor.

In our investigation of the opioid system, our focus has been on the natural product salvinorin A, which is a highly selective kappa opioid receptor agonist. In the total synthesis of salvinorin A, we have accomplished the synthesis of a key intermediate, 6-(3-furyl)-4-methyl-5,6-dihydro-pyran-2-one via ring closing metathesis. Furthermore, synthetic methodologies have been developed as a part of the total synthesis. Several lipases have been screened for their ability to generate enantiomerically pure 1-(3-Furyl)-3-buten-1-ol via bio-catalyzed hydrolysis of the corresponding acetate. The lipase from Pseudomonas fluorescens was identified as having stereoselectivity high enough to generate a % ee value above 98%. We have also developed a route for the introduction of a hydroxyl functionality in the γ position of α,β-unsaturated cyclic ketones by the regioselective oxidation of 1-silyloxy-1,3-dienes using dimethyldioxirane. We have initiated the investigation of the pharmacophore responsible for the kappa opioid activity by synthesizing simplified analogues of salvinorin A. A synthetic route providing easy access to simplified analogues of salvinorin A have been established.

Keywords: serotonin system, opioid system, selective 5-HT7 agonist, 3-aminochromans, salvinorin A, selective kappa opioid receptor, mental disorders, consciousness, regioselective oxidation, enzymatic kinetic resolution

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IV. Holmberg Pär and Gogoll Adolf. Regioselective formation and oxidation of 1-silyloxy-1,3-dienes from Hagemann’s ester and other conjugated enones with dimethyldioxirane (DMDO) as oxidant. *Manuscript*. 
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### Abbreviations

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<thead>
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<th>Abbreviation</th>
<th>Description</th>
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<tr>
<td>5-HT</td>
<td>5-Hydroxytryptamine, serotonin</td>
</tr>
<tr>
<td>AC</td>
<td>Adenylyl Cyclase</td>
</tr>
<tr>
<td>ADMET</td>
<td>Acyclic Diene Metathesis Polymerization</td>
</tr>
<tr>
<td>cDNA</td>
<td>Complementary Deoxy Ribonucleic acid</td>
</tr>
<tr>
<td>CM</td>
<td>Cross Metathesis</td>
</tr>
<tr>
<td>CNS</td>
<td>Central Nervous System</td>
</tr>
<tr>
<td>DALY</td>
<td>Disability Adjusted Life Years</td>
</tr>
<tr>
<td>dppf</td>
<td>1,1'-Bis(diphenylphosphino)ferrocene</td>
</tr>
<tr>
<td>dppp</td>
<td>1,3-Bis(diphenylphosphino)propane</td>
</tr>
<tr>
<td>E</td>
<td>Enantiomeric Ratio</td>
</tr>
<tr>
<td>ee</td>
<td>Enantiomeric Excess</td>
</tr>
<tr>
<td>FDA</td>
<td>Food and Drug Administration</td>
</tr>
<tr>
<td>GABA</td>
<td>Gamma Aminobutyric Acid</td>
</tr>
<tr>
<td>GC</td>
<td>Gas Chromatography</td>
</tr>
<tr>
<td>gHMBC</td>
<td>Gradient Selected Heteronuclear Multiple Bond Coherence</td>
</tr>
<tr>
<td>gHSQC</td>
<td>Gradient Selected Heteronuclear Single Quantum Coherence</td>
</tr>
<tr>
<td>gNOESY</td>
<td>Gradient Assisted Nuclear Overhauser Enhancement Spectroscopy</td>
</tr>
<tr>
<td>G-protein</td>
<td>Guanine Nucleotide Binding Protein</td>
</tr>
<tr>
<td>Hippocampal CA</td>
<td>Hippocampal Cornu Ammonis</td>
</tr>
<tr>
<td>HIV</td>
<td>Human Immunodeficiency Virus</td>
</tr>
<tr>
<td>HPLC</td>
<td>High Performance Liquid Chromatography</td>
</tr>
<tr>
<td>IPA</td>
<td>Isopropyl Alcohol</td>
</tr>
<tr>
<td>KOR</td>
<td>Kappa Opioid receptor</td>
</tr>
<tr>
<td>LAH</td>
<td>Lithium Aluminum Hydride</td>
</tr>
<tr>
<td>LSD</td>
<td>Lysergic Acid Diethylamide</td>
</tr>
<tr>
<td>MCPBA</td>
<td>meta-Chloro perbenzoic acid</td>
</tr>
<tr>
<td>MIRC</td>
<td>Michael Initiated Ring Closure</td>
</tr>
<tr>
<td>mRNA</td>
<td>Messenger Ribo Nucleic Acid</td>
</tr>
<tr>
<td>MS</td>
<td>Mass Spectroscopy</td>
</tr>
<tr>
<td>NMR</td>
<td>Nuclear Magnetic Resonance</td>
</tr>
<tr>
<td>P.E. COSY</td>
<td>Primitive Exclusive Correlation Spectroscopy</td>
</tr>
<tr>
<td>Acronym</td>
<td>Description</td>
</tr>
<tr>
<td>---------</td>
<td>--------------------------------------</td>
</tr>
<tr>
<td>PCP</td>
<td>Phencyclidine</td>
</tr>
<tr>
<td>PLC</td>
<td>Phospholipase C</td>
</tr>
<tr>
<td>PNS</td>
<td>Peripheral Nervous System</td>
</tr>
<tr>
<td>RCM</td>
<td>Ring Closing Metathesis</td>
</tr>
<tr>
<td>REM</td>
<td>Rapid Eye Movement</td>
</tr>
<tr>
<td>ROM</td>
<td>Ring Opening Metathesis</td>
</tr>
<tr>
<td>ROMP</td>
<td>Ring Opening Metathesis Polymerization</td>
</tr>
<tr>
<td>SAR</td>
<td>Structure-Activity Relationship</td>
</tr>
<tr>
<td>SCN</td>
<td>Suprachiasmatic Nucleus</td>
</tr>
<tr>
<td>SPE</td>
<td>Solid Phase Extraction</td>
</tr>
<tr>
<td>SSRI</td>
<td>Selective Serotonin Reuptake Inhibitors</td>
</tr>
<tr>
<td>TOCSY</td>
<td>Total Correlation Spectroscopy</td>
</tr>
</tbody>
</table>
1. Introduction

The burden of mental disorders in the world has long been underestimated. The Global Burden of Disease Study in 1996, conducted by the World Health Organization, the World Bank, and Harvard University, revealed that mental illness, including suicide, ranks second in the burden of disease in established market economies (Table 1), when disability adjusted life years (DALY) was used as a measure.\(^1\)

<table>
<thead>
<tr>
<th>Illness</th>
<th>Percent of total DALYs(^a)</th>
</tr>
</thead>
<tbody>
<tr>
<td>All cardiovascular conditions</td>
<td>18.6</td>
</tr>
<tr>
<td>All mental illness(^b)</td>
<td>15.4</td>
</tr>
<tr>
<td>All malignant diseases (cancer)</td>
<td>15.0</td>
</tr>
<tr>
<td>All respiratory conditions</td>
<td>4.8</td>
</tr>
<tr>
<td>All alcohol use</td>
<td>4.7</td>
</tr>
<tr>
<td>All infectious and parasitic diseases</td>
<td>2.8</td>
</tr>
<tr>
<td>All drug use</td>
<td>1.5</td>
</tr>
</tbody>
</table>

\(^a\)DALY=disability adjusted life years. Sum of years of life lost because of premature death and years of life lived with disability. \(^b\)Including suicide.

The disability component of the measure DALY, is weighted for severity of disability. In the Global Burden of Disease Study\(^1\) disability caused by major depression was found to be equivalent in burden to paraplegia or blindness. Active psychosis as in schizophrenia was in this study equal to quadriplegia. By this measure, major depression alone ranked second only to ischemic heart disease in magnitude of disease burden. Depressive disorders, as a single diagnostic category were the leading cause of disability worldwide, with regards to years lived with the disability.\(^2\)

Future projections for global DALYs in the year of 2020 show a significant increase in the impact of noncommunicable diseases worldwide (Table 2). Unipolar major depression could become the second leading factor in the disease burden.\(^3\)

The Global Burden of Disease study has thus revealed that mental health issues has to be dealt with seriously, rather than relegating these issues to the margin of public health concerns. This neglect is probably a reflection of the
stigmatization of people with mental disorders, which has persisted through history.

Table 2. Global Burden of Disease projections for the year of 2020.1

<table>
<thead>
<tr>
<th>Rank</th>
<th>Cause</th>
<th>%a Rank</th>
<th>Cause</th>
<th>%a</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Lower respiratory infect. b</td>
<td>8.2</td>
<td>Ischemic heart dis. c</td>
<td>5.9</td>
</tr>
<tr>
<td>2</td>
<td>Diarrheal dis.</td>
<td>7.2</td>
<td>Unipolar major depression</td>
<td>5.7</td>
</tr>
<tr>
<td>3</td>
<td>Perinatal conditions</td>
<td>6.7</td>
<td>Road traffic accidents</td>
<td>5.1</td>
</tr>
<tr>
<td>4</td>
<td>Unipolar major depression</td>
<td>3.7</td>
<td>Cerebrovascular dis.</td>
<td>4.4</td>
</tr>
<tr>
<td>5</td>
<td>Ischemic heart dis.</td>
<td>3.4</td>
<td>Chronic obstructive pulmon. dis.</td>
<td>4.2</td>
</tr>
<tr>
<td>6</td>
<td>Cerebrovascular dis.</td>
<td>2.8</td>
<td>Lower respiratory infect.</td>
<td>3.1</td>
</tr>
<tr>
<td>7</td>
<td>Tuberculosis</td>
<td>2.8</td>
<td>Tuberculosis</td>
<td>3.0</td>
</tr>
<tr>
<td>8</td>
<td>Measles</td>
<td>2.7</td>
<td>War injuries</td>
<td>3.0</td>
</tr>
<tr>
<td>9</td>
<td>Road traffic accidents</td>
<td>2.5</td>
<td>Diarrheal dis.</td>
<td>2.7</td>
</tr>
<tr>
<td>10</td>
<td>Congenital abnormalities</td>
<td>2.4</td>
<td>HIV</td>
<td>2.6</td>
</tr>
</tbody>
</table>

%a = % total DALYs. b infect.= infection. c disease.

The human brain is by far the most complex structure and system ever investigated by science. The brain contains approximately 100 billion neurons (nerve cells) supported by many more glia cells. The ability of neurons to communicate with each other is the underlying fundamental of the workings of the brain. Communication occurs at synapses, which are junctions between two neurons. A synapse consists of two parts, a presynaptic structure on a terminal portion of the sending neuron (axon) which contains packets of signalling chemicals or neurotransmitters. The second is a postsynaptic structure on the dendrites of the receiving neuron, which has receptors for the neurotransmitters. In addition, there are presynaptic receptors which also interact with the neurotransmitters.

The communication proceeds via electrical signals that travel within the neurons, again giving rise to chemical signals (neurotransmitters) that cross synapses. This event gives rise to a new electrical signal in the postsynaptic neuron. The common neuron makes on the average 1,000 synaptic connections with other neurons. Purkinje cells make between 100,000-200,000 connections with other neurons. There may be between 100 trillion to a quadrillion synapses in the brain. The interconnection of nerve cells via synapses forms intricate ensembles or circuits. A single neuron may be part of more than one circuit. The circuits are organized in a way to enable the brain to process and analyze information in parallel. Ultimately these circuits are the matrix of behavior, mental life and in the end consciousness.

Superimposed on this structural complexity is the chemical complexity of the brain. As mentioned above, the chemical signals which elicit electrical signals are called neurotransmitters.

The neurotransmitters can either be small molecules such as dopamine and serotonin or peptides such as enkephalin and dynorphin. The chemical
The signal system in the brain is able to control the main functions of a human being across timescales ranging from milliseconds to years. The number of known chemical mediators responsible for this remarkable timescale is currently about 100 and this number is expected to grow.

- **Inhibitory amino acids**
  - GABA (gamma amino butyric acid)
  - Glycine
  - L-Glutamic acid
  - L-Aspartic acid

- **Excitatory amino acids**
  - Acetylcholine
  - Histamine
  - Adenosine

**Figure 1.** Important neurotransmitters in the central nervous system (CNS).

The neurotransmitter in question is made by a very small number of nerve cells clustered in a limited number of areas in the brain. For example, about 500,000 neurons, clustered in a few brain regions, of the hundred billions neurons, make dopamine.

It has to be noted that the brain is not static. It is always changing. Neurotransmission in itself not only contain current information but alters subsequent neurotransmission. This is a case where genetics never can give the full story. Brains are built in the interface between genes and environment.
Antidepressant and antipsychotic drugs acting on the central nervous system (CNS) are of major clinical and therapeutic importance. But the receptors for the neuromodulatory transmitters are also the target for drugs of abuse such as alcohol, caffeine in different forms, nicotine, cannabis, opioids, amphetamines etc. The relationship between the effects on the cellular and biochemical level produced by centrally acting drugs and the effect on the behavioural and functional level remains obscure.

1.1. The Serotonergic System

The chemical structure for the neurotransmitter serotonin (5-hydroxytryptamine, 5-HT) was determined in 1948 (Figure 2). The name serotonin originates from the last century, when an unknown vasoconstrictor substance was found in serum. The 5-HT receptors have been found in the CNS and peripheral nervous system (PNS), as well as in the gut and cardiovascular tissue. Serotonin is involved in controlling a variety of central functions such as mood, temperature regulation, pain perception, feeding, sleep, learning and memory.

The serotonin system has been implicated in the etiology of many disease states and may be particularly important in mental illness, such as depression, anxiety, schizophrenia, eating disorders, obsessive compulsive disorder and panic disorder. The serotonin system is also playing a major role in migraine. Many currently used medical treatments of these disorders are thought to act by modulating the serotonergic tone.

![Figure 2. Structure of the neurotransmitter serotonin or 5-hydroxytryptamine (5-HT).](image)

The family of 5-HT receptors is at present divided into 7 classes, based upon their pharmacological profiles, cDNA-deduced primary sequences and signal transduction mechanism (Figure 3). The classes are further divided into different subtypes. All 5-HT receptors belong to the superfamily of Guanine nucleotide binding protein (G-protein) coupled receptors, with the exception of the 5-HT3 receptor, which forms a ligand-gated ion channel. The most recent member of the serotonin receptor family is the 5-HT7 receptor.
Figure 3. The seven classes of 5-HT receptors, with their G-protein and signal transduction mechanism. AC=adenylyl cyclase. PLC=phospholipase C.

1.1.1. 5-HT₇ Receptors, Ligands and Pharmacology

The human and rat 5-HT₇ receptor was discovered in 1993 independently by three groups. The 5-HT₇ receptor has to date been cloned from many species, in addition to humans and rats, including mouse, guinea pig and pig. The mammalian 5-HT₇ receptor consists of 445-479 amino acids and is positively linked to adenylyl cyclase (AC). The receptor shows low (<40%) sequence homology to the other 5-HT receptors.

Isoforms of the 5-HT₇ receptor are known in man and rat. Alternative splicing of the 5-HT₇ receptor gene has given rise to 5-HT₇(a), (b), (c) in rat and 5-HT₇(a), (b), (d) in human. Recent studies suggest that the human 5-HT₇ receptor splice variants do not display any differences in respect to their pharmacological profile or functional coupling to adenylyl cyclase.

The 5-HT₇ receptor mRNA is expressed in both peripheral tissues and in the CNS. The mRNA in the brain has been found at particular high levels in the hypothalamus, thalamus and hippocampus. The expression pattern together with the discovery of certain atypical antipsychotic and antidepressant agents as highly potent and non-selective 5-HT₇ receptor antagonist (Figure 4) suggest that the 5-HT₇ receptor could be an important therapeutic target for the treatment of psychiatric disorders. Further evidence supporting this hypothesis was the finding that 5-HT₇ antagonists such as LY-215840 and 5-HT₁A agonists such as 8-OH-DPAT also exhibit affinity for the 5-HT₇ receptor (Figure 4).
In vivo and in vitro pharmacological studies suggest a possible role for the 5-HT7 receptor in the control of circadian rhythms. The suprachiasmatic nucleus (SCN) of the hypothalamus functions as the circadian pacemaker in mammals. Circadian rhythm disturbances have been suggested as contributory factor in schizophrenia and manic symptoms of bipolar disorders. In addition, several studies suggest a relationship between the processes underlying unipolar depression and disturbances in circadian rhythm and sleep. There is a close relationship between circadian rhythms and sleep architecture. A number of sleep abnormalities have been observed in unipolar depression, in particular a decreased rapid eye movement (REM) sleep latency and decreased REM density in depressed patients. It is known that most antidepressants, including the selective serotonin reuptake inhibitors (SSRI) increase REM latency and decrease REM density in patients diagnosed with unipolar depression.

The 5-HT7 receptor selective antagonists 1 (SB-656104-A) and 2 (SB-269970-A) in Figure 5 (A denotes the compounds as the hydrochloride salts) produced a qualitatively similar effect on REM sleep parameters. This suggests that 5-HT7 ligands might be therapeutically useful for treating affective disorders, such as unipolar depression.
The signaling of the 5-HT$_7$ receptor has also been linked to memory and learning processes. This involvement is supported by the 5-HT$_7$ receptor distribution (among other serotonin receptor subtypes) in brain areas such as the hippocampus, amygdala and cortex. There is also electrophysiological data suggesting that activation of the receptor leads to increased neuronal excitability in hippocampal cornu ammonis (CA) regions. In addition, 5-HT$_7$ receptor knockout mice have been reported to show both a selective impairment in contextual fear conditioning and a decrease in synaptic plasticity. Other evidence for the involvement of the receptor in learning and memory is that the 5-HT$_{1A}$/5-HT$_7$ agonist 8-OH-DPAT exhibit memory enhancing properties in animals. Rats were treated with 8-OH-DPAT, which facilitated memory consolidation of autoshaping. The selective 5-HT$_7$ receptor antagonist 6 (DR-4004) partially reversed this effect. Administration of 6 alone had no effect on autoshaping. The use of selective 5-HT$_7$ agonists will be important to confirm these results.

The receptor has also been linked to anxiety. Compounds 1 and 2 have been reported to significantly reduce vocalization time in a guinea pig maternal separation model of anxiety. A plausible explanation for this observation could be that the activity of the ascending dorsal raphe neurones is inhibited, as is known for example for the anxiolytic 5-HT$_{1A}$ agonist buspirone.

The localisation of 5-HT$_7$ receptors at the level of limbic structures together with the fact that antipsychotics such as clozapine and risperidone
exhibit high affinity for the receptor has led to speculations regarding the receptor as a molecular target for the treatment of psychotic disorders such as schizophrenia. One of the problems to validate this hypothesis has been the lack of selective pharmacological tools for the 5-HT\textsubscript{7} receptor. However, Pouzet et al. recently evaluated compound 3 (SB-258741)\textsuperscript{15} (Figure 5) in three models for positive symptoms of schizophrenia and in one model for negative symptoms. Compound 3 brought a positive outcome in one of the models for positive symptoms, where it normalised phencyclidine (PCP) disrupted prepulse inhibition. No beneficial effect in the model for negative symptoms was found.\textsuperscript{20} Further research in this area is needed to establish if there is any connection between treatment of schizophrenia and the 5-HT\textsubscript{7} receptor.

The pain of migraine may in part be derived from stimulation of the trigeminal sensory system. The 5-HT\textsubscript{7} receptor transcripts have been found to be expressed in human trigeminal ganglia.\textsuperscript{21} Also, 5-HT\textsubscript{7} receptors modulate the excitability of neurones important for nociception and sensory processing and is therefore thought to play a role in the pathophysiology of migraine in which hyperalgesic pain is a main component.\textsuperscript{22,23} Furthermore, 5-HT\textsubscript{7} receptor mRNA is expressed in vascular tissues, including intra- and extra-cranial blood vessels, mediating vasodilation. In addition, there is evidence that the receptor plays a role in mechanisms underlying neurogenic inflammation. Terron recently reported that the clinically active dose for a number of non-selective migraine prophylactic agents correlate with their affinity for the 5-HT\textsubscript{7} receptor,\textsuperscript{23} further strengthening the evidence for involvement of the receptor in migraine.

Thirteen years after the cloning of the 5-HT\textsubscript{7} receptor, the role of the receptor in both the CNS and PNS is far from being fully understood. Antagonists such as 1, 3, 4 (SB-691673),\textsuperscript{24} 6 and various aporphine derivatives (eg. 5)\textsuperscript{25} have appeared (Figure 5).

Still no selective partial agonists or agonists for the serotonin 5-HT\textsubscript{7} receptor have appeared, though non-selective agonists with good affinity have been published in recent years (Figure 6).

For example compound 7 (Figure 6) also has potent $\alpha_1$ and $\alpha_2$ binding activity, whereas compound 8 (Figure 6) has high affinity for the 5-HT\textsubscript{2A} receptor and moderate affinities for $\alpha_1$ and $\alpha_2$ receptors.
The development of selective agonists and partial agonists is thus essential for elucidating the functional role for this receptor and its potential as a drug target.

1.2. The Opioid System

The effects from preparations of the opium poppy *Papaver somniferum*, such as euphoria and analgesia, have been known to man since ancient times. In 1805 the German chemist Sertürner isolated a crystalline sample of the main constituent. The colorless crystals were obtained by extraction of opium with hot water, followed by precipitation with ammonia. The substance was named morphine (Figure 7) after the Greek god of dreams, Morpheus.

![Figure 7. Morphine, the first isolated alkaloid from a natural product.](image)

Morphine was the first alkaloid ever to be isolated and was later shown to be nearly entirely responsible for the pharmacological action of crude opium. The purification of morphine started the natural products chemistry resulting in the isolation of a range of alkaloids soon thereafter. As early as in 1827 Heinrich Emanuel Merck of Darmstadt began selling morphine resulting in the development of the famous company Merck.

It was first in 1973 that the molecular targets for opiates were identified independently by three laboratories. The evidence was compelling for endogenous opioid ligands and in 1975 the two pentapeptides Leu- and Met-enkephalin were discovered. These findings confirmed the 1954 suggestion of a specific “analgesic receptor” that recognizes morphine and highlighted the importance of an endogenous opioid system in the central nervous system. Further pharmacological studies led to the identification of three major types of opioid receptors, namely the delta (δ), kappa (κ) and mu (μ) receptors. All opioid receptors belong to the family of G-protein coupled receptors. The members of the opioid receptor family are homologous (~60% amino acid homology).

It is believed that all of the opioid receptors share common effector mechanisms. All of the three receptor types inhibit adenylyl cyclase. Other shared direct G-protein mediated effects are the activation of an inwardly rectifying potassium channel and inhibition of voltage operated calcium
channels. There is a range of other effects mediated via opioid receptor activation. The functional significance of these effects remains unclear and the biological functions of the opioid system are diverse and complex.

This neuromodulatory system has been implicated in the control of behaviours that are essentials for self and species survival, including responses to noxious information and stress, reward and motivation. Opioid peptides and their receptors further control autonomic functions, including respiration, thermoregulation and gastrointestinal motility. The opioid system even modulate immune responses.35

A paradox with the opioid system is that there are numerous endogenous ligands which activate a small set of receptors. In contrast other signalling pathways in the CNS, such as the serotonergic uses only one neurotransmitter. 5-HT activates a large number of different receptor subtypes which have different distribution of receptor densities in different areas of the CNS.

Akil et al.36 have suggested several explanations for this paradox, a pattern or profile of activation of multiple receptors by a ligand rather than the activation of a single receptor. For example, two ligands may activate the μ receptor equally but activate the κ opioid receptor (KOR) differently, leading to different biological responses. Another explanation could be that each opioid gene gives rise to multiple active peptides, each and every one having a unique profile of activity, giving rise to a very complex pattern able to respond in a finely tuned manner to various stimuli. These are just a few examples of the existing theories.

1.2.1. Opioid Kappa Receptors, Ligands and Pharmacology

The pharmacological effects mediated via the KOR and the other two opioid receptors are summarized in Table 3.4

<table>
<thead>
<tr>
<th>Functional effect</th>
<th>μ-receptor</th>
<th>δ-receptor</th>
<th>κ-receptor</th>
</tr>
</thead>
<tbody>
<tr>
<td>Analgesia</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Respiratory depression</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
</tr>
<tr>
<td>Reduced GI motility</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
</tr>
<tr>
<td>Euphoria</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Dysphoria</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>Sedation</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Diurese</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>Hallucinations</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
</tr>
</tbody>
</table>
There are several advantages in developing selective KOR agonists as analgesic drugs. As can be seen from Table 1 selective KOR agonists will have fewer side effects (e.g. respiratory depression, constipation, addiction) in comparison with the prototypical $\mu$-receptor agonists. However, undesired side effects associated with KOR agonists is hallucinations and dysphoria. Another interesting area, except analgesia, is neuroprotection. There are several reports that KOR agonists could have a beneficial effect in treating cerebral ischaemic injury.37,38

Synthetic selective KOR agonists belong to two chemical classes: benzomorphans, such as CCB and MPCB (Figure 8)39 and arylacetamides, such as U-50488 and U-69593 (Figure 8).40

![Figure 8. Selective KOR agonists.](image)

In 1987 the first KOR antagonist, norbinaltorphimine (nor-BNI) (Figure 9) was synthesized by Portoghese et al.41 Since then a small number of new KOR antagonists have been developed, e.g. GNTI in Figure 9.42 KOR antagonists like GNTI have been shown to have antidepressant-like effects in the forced swim test of rats.43 However, further investigations in this area are needed.

![Figure 9. Examples of selective KOR antagonists.](image)

Discovery of new selective opioid ligands will allow us to investigate the opioid system and how it mediates complex functions, such as the control of
pain and reward amongst others. There is a need to diversify the available chemical structures for both KOR agonists and antagonists, in order to fully elucidate the functional role of this receptor.
2. Aims of the Present Study

In this thesis we are investigating two systems within the CNS, the serotonergic system and the opioid system.

In the serotonergic system, we are addressing the 5-HT\textsubscript{7} receptor, the most recently discovered receptor subtype in the 5-HT receptor family. There are no published selective 5-HT\textsubscript{7} agonists and partial agonists yet available for disclosing the intricacies of this receptor.

Our aims were:
1. To identify and synthesize selective 5-HT\textsubscript{7} ligands based on the 3-amino chroman skeleton (Figure 10).
2. To elucidate the structure-activity relationship (SAR) for the 5-HT\textsubscript{7} receptor with differently substituted 3-aminochromans.

In the opioid system we are addressing the KOR, as a potential target for treating disorders characterized by perceptual disturbances.

Our aims were:
1. To initiate the total synthesis of the natural product salvinorin A (Figure 10), a highly selective KOR agonist.
2. To investigate the pharmacophore of salvinorin A by the synthesis of simplified analogues.

![Figure 10. The basic structure of 3-aminochroman and salvinorin A.](image-url)
3. Synthetic Methodologies

This thesis deals with two major topics in organic chemistry: the synthesis of enantiomerically pure compounds and the creation of C-C bonds.

The generation of enantiomerically pure compounds was accomplished by resolution. Resolutions of enantiomers have a long history in organic chemistry starting with Pasteur in the 19th century, and it still remains one of the more important methods to generate enantiomerically pure compounds.

Formation of C-C bonds falls into three major categories:
1. Addition of a carbanion to a carbonyl group.
2. The reaction of an enolate, derived from a carbonyl group, with an electrophilic carbon.
3. Transition metal mediated cross-coupling reactions.

In the thesis, two important transition metal mediated cross-coupling reactions have been used: the Suzuki-Miyaura cross-coupling reaction and the ring closing metathesis (RCM).

Below follows a short discussion on chirality and enantiomerically pure compounds followed by two short mechanistic descriptions of the Suzuki-Miyaura cross-coupling and the RCM.

3.1. Generation of Enantiomerically Pure Compounds

One of the foundations of the chemistry of life is chirality. The term "chiral" can be defined as an object not being superposable with its mirror image. The term can be applied to molecules, conformations as well as macroscopic objects, such as a human being. A human being is characterized by being chiral, i.e. not being superposable with its mirror image. Chirality (handedness) is the property of being chiral, implying the absence of any symmetry element.

Although enantiomers have identical physico-chemical properties, except for rotation of planpolarized light in opposite direction by equal amounts, they are from a biological point of view different. This is a consequence of their differential 3-D interaction with a chiral target such as a receptor or an ion channel.

In racemic drugs one enantiomer can be responsible for the activity of interest, while the paired enantiomer can function as an antagonist of the active enantiomer, have no activity or have activity of its own desirable or undesir-
able. Furthermore racemic drugs can cause problems not only in terms of differences in biological effects but also in the pharmacokinetics of the enantiomers. An example of this is perhexiline (Figure 11), which was marketed as a racemate and used to treat abnormal heart rhythms. The racemate killed a number of people who had accumulated gram quantities of one of the enantiomers that was more slowly metabolized.

Figure 11. Racemic Perhexelin was the cause of death for number of people.

In 1992 the Food and Drug Administration (FDA) started to regulate chiral drugs, with the publication of Policy Statement for the Development of new Stereoisomeric Drugs (http://www.fda.gov/cder/guidance/stereo.htm), which was closely followed 1994 in the European Union with the guidelines Investigation of Chiral Active Substances (http://pharmacos.eudra.org/F2/eudralex/vol-3/pdfs-en/3cc29aen.pdf). Applicants must now recognize the occurrence of chirality in new drugs, attempt to separate the stereoisomers, assess the contribution of the various stereoisomers to the activity of interest and make a rational selection of the stereoisomeric form that is proposed for marketing.

In addition the introduction of the so called chiral switch, have allowed the pharmaceutical companies to extend patent time for a compound. A chiral switch may be defined as the development of a single enantiomer from a previously marketed racemate. An example of a chiral switch is esomeprazole by AstraZeneca (Figure 12).

Figure 12. AstraZeneca’s synthesis of esomeprazole, by asymmetric oxidation.

The sales of single enantiomeric drugs were estimated to encompass 39% of the market worldwide in 2002, totaling US $152 billions. The available data for 15 FDA approved drugs in the period January-August 2003 shows
the following distribution: 64% single enantiomers, 14% racemates and 22% achirals. The technologies available to obtain optically pure compounds are asymmetric synthesis, classical resolution, chiral chromatography and enantioselective bio-catalysis. Commonly chemical methods do not obtain %ee values in the range of >99.5% and one has to resort to either resolution or chiral chromatography to improve the %ee value.

In this thesis we have used two of the available technologies to obtain enantiomerically pure compounds:
1. Classical resolutions, via formation of diastereomeric salts.
2. Enzymatic kinetic resolution, via enatioselective hydrolysis.

3.2. Metal-Catalyzed Carbon-Carbon Bond Forming Reactions

Transition metal mediated cross-coupling reactions are of outmost importance for creating carbon-carbon bonds. The most utilized transformations are probably the Heck, Stille and Suzuki-Miyaura reaction. One of the latest additions to this class of reactions is the olefin metathesis reaction. This reaction is mostly used in annulation transformations. The Heck, Stille and Suzuki-Miyaura reaction share a common mechanism, whereas the mechanism for the metathesis reaction is fundamentally different.

There is a wealth of protocols for these kind of reactions in the literature, which is indicative of their importance for the chemical community. In this thesis two types of transition metal mediated cross-coupling reactions have been utilized.

The first type of transition metal catalyzed reaction is the Suzuki-Miyaura cross-coupling, which is a reaction between an aryl or vinyl halide, triflate and an aryl borane catalyzed by palladium (Figure 13). Another variant of the Suzuki-Miyaura reaction between an alkyl borane and aryl or vinyl halide, triflate or enol phosphate and it is often referred to as the B-alkyl Suzuki-Miyaura reaction. One of the advantages of the Suzuki-Miyaura cross-coupling is the tolerable toxicity of the boronic coupling partner, as well as the tolerance of water. Addition of water is often a prerequisite for a successful reaction.

![Figure 13. The Suzuki-Miyaura cross-coupling reaction.](figure13.png)
The second type of transition metal catalyzed reaction is the RCM reaction between two olefins (Figure 13) catalyzed either by ruthenium or molybdenum. This transformation has emerged as a powerful tool for carbocyclizations.

RCM is one variant of olefin metathesis. Another common variant (Figure 14) is the ring opening metathesis (ROM). Below follows a short mechanistic description of the Suzuki-Miyaura cross-coupling and the olefin metathesis.

**Figure 14.** Five variants of olefin metathesis. 1. RCM = ring-closing metathesis. 2. ROMP = ring-opening metathesis polymerization. 3. ADMET = acyclic diene metathesis polymerization. 4. ROM = ring-opening metathesis. 5. CM = cross-metathesis.

### 3.2.1. Mechanism of the Suzuki-Miyaura Cross-Coupling

The Suzuki-Miyaura cross-coupling is a palladium catalyzed cross-coupling between a boron nucleophile, e.g. an aryl boronic acid, and an aryl or vinyl halide or triflate.

The catalytic cycle of the cross-coupling involves three distinct steps; oxidative addition, transmetalation and reductive elimination (Figure 15). The oxidative addition is often described as the rate limiting step. Aryl hal-
ides or triflates activated by electron-withdrawing groups are more reactive to the oxidative addition step than those with electron-donating groups.

The order of reactivity for the electrophilic component of the reaction has been established as follows: I >> Br > OTf >> Cl.

Figure 15. General catalytical cycle for the Suzuki-Miyaura cross-coupling.

Among the boron nucleophiles, the unhindered and electron-rich aryl or vinyl boronic acids are the most reactive coupling partners in the coupling reaction.

The ability of the boron atom to host an expanded coordination number and form “ate” complexes is taken advantage of in the reaction. The “ate” complex is more reactive as a nucleophile in the cross-coupling. The formation of this more reactive borate species is essentially the reason why a negatively charged base such as a hydroxide or a carbonate is necessary in the Suzuki-Miyaura cross-coupling. However, there are also other suggestions for the involvement of the base.

In recent years the number of catalysts used in the Suzuki-Miyaura cross-coupling has exploded. Catalysts have been developed even for the cross-coupling of alkyl boranes with aryl chlorides as by Buchwald et al. Still one of the most widely used catalysts is (PPh3)4Pd.

3.2.2. Mechanism of the Olefin Metathesis

Scrambling of double-bonds was first reported in the mid 1950’s by Euleute-rrio at Du Pont’s Polychemicals Department in his work with ring-opening
polymerization of cycloolefins and in the beginning of the 1960’s by Banks and Bailey, who employed various heterogenous catalysts for the disproportionation of olefins at high temperatures. But it was not until several years later, in 1967, that Calderon et al. showed that the products from a mixture of 2-butene and 2-butene occurred via redistribution of alkylidene moieties and coined the term “olefin metathesis”.

The word metathesis is derived from the greek word metatethe’nai which means place differently or to transpose (meta’=beyond, over and tithe’=to place, to set). Since the basic process is an alkylidene interchange, the word metathesis was appropriate to define the reaction.

The olefin metathesis reaction can be considered as a process in which all the carbon double bonds are cut and then rearranged in a statistical fashion (Figure 16).

![Figure 16. Rearrangement of double bonds catalyzed by a transition metal.](image)

The mechanism for the reaction has been a subject of controversy, but the commonly accepted mechanism today is the one originally suggested by Chauvin and Herrison. They proposed that the mechanisms involve a [2+2] cycloaddition between the olefin and a metal alkylidene, forming a metallacyclobutane intermediate (Figure 17).

![Figure 17. Mechanism for olefin metathesis proposed by Chauvin and Herrison.](image)

This metallacyclobutane intermediate breaks up providing a new metal alkylidene and olefin. This process is repeated and eventually an equilibrium mixture of olefins and metal alkylidenes is obtained.

RCM has emerged as a powerful tool for constructing rings. The method has found wide applications especially for macrocyclizations in the synthesis of natural products. These reactions are essentially driven by the gain in entropy upon bisecting the diene (Figure 18).
Since ethylene is volatile the reaction can be driven to completion. However dimerizations and oligomerizations can be problems in the synthesis of medium-sized to large rings. The reaction temperature and concentration of the precursor can be manipulated to control these events.55
4. Enantiomerically Pure 3-Aminochromans

Originally various derivatives of 3-aminochroman were prepared as oxygen isosters of 2-aminotetralins since, for example, 8- and 5-hydroxy-2-(dipropylamino)tetrains (8-OH-DPAT and 5-OH-DPAT) were well characterized as 5-HT_{1A} and D_{2} receptor agonists respectively (Figure 19).^{56,57}

![Figure 19. Numbering scheme of 2-aminotetralins and 3-aminochromans respectively.](image)

In 1999 Caldirola et. al. described the preparation and pharmacological evaluation of a series of 8-aryl and 8-aroyl substituted derivatives of 3-(dipropylamino)chromans.\textsuperscript{58}

The affinities of the compounds were evaluated for central 5-HT_{1A} and dopamine D_{2A} receptors and several of the compounds displayed high affinity and selectivity for 5-HT_{1A} receptors over D_{2A} receptors. These compounds were further evaluated for central 5-HT_{7} receptors and (R)-8-(2-methoxyphenyl)-3-(dipropylamino)chroman 9\textsubscript{b} (Figure 20) was found to display high affinity and about 4 times fold selectivity for 5-HT_{7} receptors over 5-HT_{1A} receptors. The interaction with the receptor was also stereo-specific, favoring the (R)-chroman over the (S)-chroman with a ratio of 130:1 (Table 4).

![Figure 20. (R)-8-(2-methoxyphenyl)-3-(dipropylamino)chroman as lead structure for selective 5-HT_{7} receptor agonists.](image)
These results initiated the search for selective 5-HT\textsubscript{7} ligands based on the structural motif of \((R)-8\)-aryl-3-aminochromans as shown in Figure 20.

![Aryl substituents in 6- or 8-position](image)

**Figure 21.** Objectives for the investigation of SAR for \((R)-8\)-aryl-3-aminochromans and \((S)-6\)-aryl-3-aminochromans.

We explored SAR for chromans at the 5-H\textsubscript{7} receptor, by studying different \(N\)-alkyl substituents, by inverting the stereochemistry in position 3 and moving the aryl substituent to the C6-position of the 3-aminochroman skeleton (Figure 21).

4.1. \((R)-8\)-Aryl-3-aminochromans as Serotonin 5-HT\textsubscript{7} Receptor Agonists (Paper I)

In this paper we describe the synthesis and the resolution of 8-methoxy-3-(dibenzylamino)chroman. Enantiomerically pure 8-methoxy-3-(dibenzylamino)chroman is a valuable key intermediate in the sense that it would be possible to generate the primary amine and from there investigate the significance of different \(N\)-alkyl substituents for the interaction with the 5-HT\textsubscript{7} receptor. The synthesis of the target compounds 20\textsubscript{a} and 20\textsubscript{b} is also described.

4.1.1. Synthesis of 8-Methoxy-3-(dibenzylamino)chroman

Initially we attempted to synthesize 8-methoxy-3-aminochroman 11 via the pathway in Scheme 1. A ring-closing condensation between 2-hydroxy-3-methoxybenzaldehyde (\(o\)-vanillin) and 2-nitroethanol yielded the unsaturated nitro-derivative 10.\textsuperscript{59}
Compound 10 was efficiently reduced to the primary amine 11 with the BH$_3$•THF and a catalytical amount of NaBH$_4$.

Attempts to use lithium aluminum hydride (LAH) as a reducing agent afforded complex product mixtures. Unfortunately, when the condensation reaction was attempted in a 40g scale, extensive polymerization occurred and the yield was less than 5%. Due to this problem, we surveyed the literature for other ways to synthesize the chroman.

The synthetic route utilized for the preparation of 8-methoxy-3-(dibenzylamino)chroman 16 is shown in Scheme 2 and starts with a conjugate addition of $\alpha$-vanillin to acrylonitrile. The reaction mechanism of this transformation is not completely certain and the reaction could just as well start with a DABCO catalyzed addition of the aldehyde to acrylonitrile at the $\alpha$-position (Baylis-Hillman reaction), followed by addition of the phenolate ion to the unsaturated nitrile. Either way, this step was followed by a ring-closing condensation affording the unsaturated nitrile 12 in 73% yield. The nitrile was hydrolysed in 10% aqueous NaOH to afford the carboxylic acid 13. Compound 13 was subjected to a Curtius rearrangement giving the enamine, which was, in situ, hydrolysed with 6 M HCl providing the ketone 14 in 84% yield. Reductive amination with benzylamine in THF gave the monobenzylated chroman 15, which was further $N$-alkylated with benzylbromide to give the dibenzylated compound 16. The overall yield for the synthetic sequence was 30%. The reported yields in the scheme are from the scale up experiments with amounts of starting material in each step varying from 20g-200g. Thus, the sequence provided us with a robust synthesis of 8-methoxy-3-(dibenzylamino)chroman 16 affording the desired compound in preparative useful amounts.

The enantiomers were obtained from the racemate by fractional recrystallization. The dibenzyl derivative 16 was fractionally recrystallized with 0.5 equiv. L-dibenzoyl tartaric acid or D-dibenzoyl tartaric acid in a two phase system with water and 1,2-dichlorehane/CHCl$_3$, affording the enantiomers in good yields with an %ee value of $>99$. The process for resolving the antipodes are described in detail in section 4.1.2.
4.1.2. Resolution of 8-Methoxy-3-(dibenzylamino)chroman

Previously, several attempts to resolve 8-methoxy-3-aminochroman 11 had been made by the preparation of diastereomeric amides. The diastereomeric mandelic acid derived amide had been separated by flash column chromatography, but cleavage of the amide bond failed. Different reagents for the cleavage of the amide bond such as Et$_3$BH, HCl and NH$_2$NH$_2$ were tried. HCl resulted in the elimination of the amido moiety, otherwise the amide bond was totally inert.

These disappointing results prompted us to investigate carbamates as an alternative to amides. The general pathway for the synthesis of the carbamates is outlined in Scheme 3.
We abandoned this route after some unsuccessful initial attempts with this route. There are several disadvantages with this strategy. The foremost being the cost of the enantiomerically pure alcohols, which in a larger scale (>20g) was troublesome. Taken together there were no advantages in pursuing this path, other than as a last resort, compared with a classical resolution by crystallization of diastereomeric salts.

Resolutions are often tedious and can be hard to succeed with. It is an empirical method, but Wilen et al. has described a systematic approach to optical resolutions.\textsuperscript{64} The requirements are a reasonably large number of resolving agents and a way to monitor the progress of the resolution. The monitoring can be done by measuring the optical rotation or by chiral HPLC. The solvent selection for the salt formation is critical for success. A solvent as polar as possible should be chosen since the success of the resolution depends on the formation of salt pairs and their differences in physicochemical properties. Problems may arise with for example water because of solubility problems for the substrate or the resolving agent.

We chose ten enantiomerically pure acids as resolving agents. All of them at reasonable prices, except for (R)-(-)-1,1'-binaphthyl-2,2'-diyl hydrogen phosphate. The acids were screened against the primary amine 11, the monobenzylated 15 and the dibenzylated amine 16. To monitor the result we initially measured the optical rotations on the free amines. Six different solvents or solvent mixtures were used (Scheme 4).
The screening was performed with 100 mg of amine, 1 equiv. of resolving acid and various amounts of solvent, starting with 1mL then adjusting the volume. The proportion between EtOH and H₂O was also varied from a few drops of H₂O to a 1:1 ratio.

There was no induction of any crystal formation with the primary amine. In some cases for the monobenzylated and dibenzylated amine crystals were formed, but they were in all cases racemic. Different volumes and proportions of solvent mixtures were tried.

One of the factors affecting the solubility differences between the salts is the pH value. Wittig et al. in his resolution of chiral triarylphosphines used 0.5 equiv. of resolution agent. The rationale behind is to obtain a pH maximum for the resolution, since the pH for the optimum resolution does not coincide with the pH of a reaction mixture containing the reactants in equal amounts.

Performing the screening 0.5 equiv. of the resolving agent did not improve the situation. The crystals were in all cases virtually racemic. However, in an interesting article from 1985 by Ács et al. they describe an improved resolving technique, using two immiscible solvents and a half equivalent of resolving agent. This method combines several advantages in one system. Solubility problems are avoided and an optimal pH is reached. The pH increases with one unit compared with the pH of a homogenous aqueous solution.

Screening with a two phase system consisting of 1,2-dichlorethane/H₂O (1:1) and 0.5 equiv of the enantiomerically pure acids above finally gave result. A modest %ee value of 72 was obtained with L-dibenzoyl tartrate. This was not sufficient for our purposes as SAR investigation typically re-

---

**Scheme 4.** Screening procedure for resolution.

<table>
<thead>
<tr>
<th>Acid</th>
<th>Solvent 1</th>
<th>Solvent 2</th>
<th>Solvent 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dibenzoyl-L-tartaric acid</td>
<td>H₂O</td>
<td>i-PrOH</td>
<td>EtOH</td>
</tr>
<tr>
<td>D(-)-mandelic acid</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(+)-dianisoyl-D-tartaric acid</td>
<td>EtOH/</td>
<td>CH₃CN</td>
<td>Benzene</td>
</tr>
<tr>
<td>2-nitro-L-tartranic acid</td>
<td>H₂O</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(1R, 3S)(+)-camphoric acid</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(+)-tartaric acid</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>di-p-toluoyl-L-tartaric acid</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(R)-(-)-1,1’-binaphthyl-2,2’-diyl hydrogen phosphate</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(1S)-(-)-10-camphorsulfonic acid</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(R)-(-)-α-methoxyphenylacetic acid</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

---

36
quires %ee >98. Other organic solvents than 1,2-dichloroethane were tried, such as toluene but with disappointing results. However a mixture of chloroform/1,2-dichloroethene (1:9) improved the %ee to ~85. The crystallizations were finalized as shown in Scheme 5. The ee values were determined by chiral HPLC of the free amines.

![Scheme 5](image-url)

The first step, resolution of 29 g of the racemic amine 16 with L-dibenzoyl tartaric acid, results in two phases: crystals and an organic phase. The crystals were isolated, and after liberating the amine (R)-16 a %ee of 86 was
obtained. The amine isolated from the organic phase contained the other enantiomer (S)-16 with a %ee of 68 (Scheme 5).

The liberated amine from the L-dibenzoyl tartaric acid salt, was then further resolved using 0.8 equiv. of L-dibenzoyl tartaric acid. (R)-16 were obtained from the L-dibenzoyl tartaric acid salt with %ee value of 96. The organic phase contained (R)-16 with a %ee of 35. The obtained organic phase was evaporated and resolved in the same manner with 0.5 equiv of L-dibenzoyl tartaric acid yielding (R)-16 with a %ee of 91. The mother liquor contained (S)-16 with a %ee value of 14.

The other enantiomer was resolved in a corresponding way, using D-dibenzoyl tartaric acid as resolving agent. To obtain enantiomerically pure (S)- and (R)-16 phases were pooled containing the appropriate enantiomer and resolved with different amounts of the corresponding resolving agent as described in Scheme 5 until %ee value >99 was obtained. This protocol provided us with enantiomerically pure (S)- and (R)-16 in high yields.

4.1.3. Synthesis of Target Compounds

The synthesis of the target compounds (R)-8-(2,6-Dimethoxyphenyl)-3-(dipropylamino)chroman 20a and (R)-8-(2,6-dimethoxyphenyl)-3-(dimethylamino)chroman 20b were performed via the route in Scheme 6.

Palladium catalyzed N,N-debenzylation in refluxing isopropyl alcohol (IPA) with ammonium formate as hydrogen source provided the primary amine (R)-11. Compound (R)-11 was then reductively alkylated with either aqueous formaldehyde or propionaldehyde, giving the dialkylated amines 17a and 17b in good yields.

Demethylation of 17a and 17b with BF₃·Me₂S yielded the phenols 18a and 18b, respectively, which were treated with triflic anhydride at –78°C to give the triflates 19a and 19b, respectively. The target compounds 20a and 20b were obtained by microwave facilitated Suzuki-Miyaura cross-coupling of the triflates. For a discussion of the Suzuki-Miyaura cross-couplings in the synthesis of (R)-8-aryl- and (S)-6-aryl-(dimethylamino)chromans see section 4.2.3.
4.2. (S)-6-Aryl-3-aminochromans as Serotonin 5-HT<sub>7</sub> Receptor Ligands (Paper II)

In this paper we describe the synthesis of (S)-6-bromo-3-(dimethylamino)chroman 26 and its utilization in the Suzuki-Miyaura cross-couplings for the preparation of target molecules.

4.2.1. Synthesis of (S)-6-Bromo-3-(dimethylamino)chroman

The synthesis of key intermediate (S)-6-bromo-3-(dimethylamino)chroman is outlined in Scheme 7.

The partially resolved starting material (S)-2-amino-5-methoxychroman 21 was obtained as a gift from AstraZeneca AB and contained about 10% of the (R)-isomer. The enantiomerically pure compound 22 was obtained by recrystallization of the D-(-)-tartrate of 21 in H<sub>2</sub>O.
The enantiomeric excess was determined, indirectly on the corresponding Mosher amides, by chiral HPLC to be >99%. The primary amine 22 was then reductively alkylated with formaldehyde and NaCNBH₃ to give the dimethyl derivative 23. Demethylation of the methoxy group in aqueous HBr, kept at reflux, yielded the phenol. The phenol was treated with triflic anhydride to afford the triflate 24. Palladium catalyzed reduction of 24 with formic acid and Et₃N as hydrogen source provided 25. A remarkable increase in both yield and reaction rate was observed when the palladium ligand 1,3´-bis(diphenylphosphino)propane (dppp) was changed to 1,1´-bis(diphenylphosphino)ferrocene (dppf). Bromination in acetic acid afforded the key intermediate 26, which subsequently was used in the palladium cross-coupling reactions to give the target compounds.

4.2.2. Synthesis of Target Compounds

The Suzuki-Miyaura cross-coupling was also here facilitated by microwave heating. The target compounds 27a, 27c and 27d were obtained in good yields (Scheme 8): 76%, 75% and 73% respectively. 2,6-Dimethylphenylboronic acid provided the Suzuki-Miyaura cross-coupling product 27b in a somewhat lower yield (57%). The lower yield can be explained by greater sterical hindrance in 2,6-dimethylphenylboronic acid compared to that of 2,6-dimethoxyphenylboronic acid. In addition, 2,6-dimethylphenylboronic acid is less electron rich than the 2,6-dimethoxy derivative, making the 2,6 dimethylphenyl boronic acid less prone to undergo the cross-coupling reaction.

Scheme 7. Synthesis of the key intermediate 26 (S)-6-bromo-3-(dimethylamino)chroman.
Scheme 8. Suzuki-Miyaura cross-coupling of 26. Compound 27e was obtained by demethylation of the dimethoxy derivative 27a.

The dimethoxyphenyl derivative 27a was O,O-didemethylated by refluxing in 48% aqueous HBr to give the resorcinol 27e in reasonable yield (Scheme 8).

4.2.3. Notes on the Suzuki-Miyaura Cross-Couplings

There were notable differences between the Suzuki-Miyaura cross-couplings performed at the C6 and C8 position of the 3-aminochromans (Scheme 9), but there were also some similarities.

It was considerably harder to perform the coupling at the C8-position than at the C6-position. One explanation might be that the C8-position is more electron rich than the C6-position, as a result of the presence of the ortho alkoxy group. The C8-position is also more sterically hindered.

Attempts were made to increase the reactivity in the 8-position by synthesizing the corresponding boronic ester 28 from triflate 19a, via the two routes in Scheme 10.69-71 The results were not encouraging. The synthesis of the pinacol boronate 28 was always accompanied by deoxygenation of the substrate 19a to the reduced chroman 29 in a 1:1 relationship. In addition the product was not stable, rendering any attempts to purify the reaction mixture impossible.

Scheme 10. Attempts to synthesize boronic esters from aryltriflates.

The synthesis robot Chemspeed was used in an attempt to screen a larger number reactions. The bases (KOAc and Et3N), the three ligands in Figure 2250,72,73 combined with Pd(OAc)2 and Pd2dba3 as the palladium source were screened with bis(pinacolata)diboron as the boronic source.

Figure 22. Palladium ligands used in the screening to obtain pinacol boronates.

The reactions were heated over night in dioxane, under inert conditions. No work-up of the reactions were made before samples were subjected to 11B
NMR and $^1$H NMR spectroscopy. The results were in all cases complex reaction mixtures and the conclusion was that this path had to be abandoned.

The Suzuki-Miyaura cross-coupling at the C8-position in 19a was accompanied by a substantial amount of reductive deoxygenation to the reduced chroman 29. The amount of deoxygenation was proportional to the amount of protic solvent present. On the other hand it was necessary to include water in the solvent mixture for both triflate 19a and 26 in order for the reaction to work. In the coupling of the (S)-6-bromo-derivative 26 we used a solvent system published by Gronowitz et al., consisting of DME/H$_2$O/EtOH (62.5:25:12.5). No deoxygenation could be detected. Applying the same solvent system to the coupling of the triflate 19a, gave a high proportion of the deoxygenated product. It was experimentally determined that EtOH could be excluded, minimizing the amount of the deoxygenation. A solvent system consisting of DME/H$_2$O (9:1) was found to be optimal. Less water did not give full conversion of the starting material and the amount of the deoxygenated product increased. Other solvents like DMF gave no reaction at all or in THF only partial conversion accompanied by deoxygenation took place.

The amount of deoxygenation could also be a result of different heating methods. In both cases microwave heating was necessary for the reactions to proceed. In the case for the cross-coupling at the C6-position in 26 a much higher energy input was used (145 W, 4 min). However for the intermediate 19a the heating period was set to 20 min at 140°C with the Smith Microwave Synthesizer. Heating at lower temperatures in oil baths increased the amount of deoxygenation.

The choice of base was also critical. In the cross-coupling of (S)-6-bromo-derivative 26 the weak base Na$_2$CO$_3$ was enough for full conversion but not in the case of triflate 19a. Here the stronger base Ba(OH)$_2$ had to be used in order to obtain full conversion. Other bases such as KF, CsCO$_3$ did not achieve this objective.

Finally, it has to be mentioned that by coincidence our first selected catalyst for the cross-coupling of 19a, (PPh$_3$)$_4$Pd proved to be the most active. Other catalysts, such as the catalysts published by Buchwald, Nolan and Hermann, proved to be much less effective in this case.

In conclusion, the Suzuki-Miyaura cross-coupling reaction is an efficient method to create new C-C bonds. However, optimizing reaction conditions for substrates such as 19a is largely empirical. For success it is necessary to have large sets of catalysts, bases and solvents or solvent systems at hand.
5. SAR of the Novel 3-Aminochromans

The SAR of novel \((R)\)-8-aryl-3-aminochromans at the serotonin 5-HT\(_7\) receptor were evaluated in comparisons with different enantiomerically pure 2-aminotetralins and previously synthesized 3-aminochromans. The \((S)\)-6-aryl-3-aminochromans were evaluated and compared with \((R)\)-8-aryl-3-aminochromans for biological activity. The synthesis of the 2-aminotetralins, compounds \(30a-30f\) and of the previously synthesized 3-aminochromans, \(9a-9c\), are described in Paper I.

5.1. SAR of \((R)\)-8-Aryl-3-aminochromans (Paper I)

The novel compounds were evaluated for their affinity to the 5-HT\(_7\), 5-HT\(_{1A}\) and D\(_{2A}\) receptors, respectively and compared with 2-aminotetralins.

The compounds were tested for their ability to displace \([\text{\textsuperscript{3}H}]\)-raclopride from mouse fibroblast (Ltk\(^{-}\)) cells expressing the human D\(_{2A}\) receptor. Affinity for the 5-HT\(_{1A}\) receptor was determined in CHO cells expressing the human receptor or in rat hippocampal cells, using \([\text{\textsuperscript{3}H}]\)-8-OH-DPAT as the ligand. The affinity of the compounds for the rat 5-HT\(_7\) receptor was measured in CHO- or Sf9-cells using \([\text{\textsuperscript{3}H}]\)-5-HT as the ligand. Efficacy at the rat 5-HT\(_7\) receptor was obtained by measuring the effect of cAMP production in CHO cells in relation to the effect elicited by 5-HT\(^{-}\). Compounds \(1, 4 \text{ and } 5\) and the corresponding literature data are included for comparison (Table 4).

The dimethylaminotetralins and the dipropylaminotetralins as well as the corresponding chromans, gave ligands with high affinity for the 5-HT\(_7\) receptor, whereas the primary amines are of lower potency.

The interaction of the novel ligands with the 5-HT\(_7\) receptor is stereospecific favouring the \((R)\)-chromans ((S)-tetralins) over the corresponding enantiomers, with the ratio of approximately 130:1 for \((R)-9b\) vs. \((S)-9b\).

The introduction of a single \textit{ortho} methoxy substituent reduces the affinity for the 5-HT\(_{1A}\) receptor 8-13 times while the affinity for the 5-HT\(_7\) receptors is unchanged (compare tetralins \(30a\) and \(30b\) and chromans \(9a\) and \(9b\)). Further introduction of a second \textit{ortho} methoxy substituent (\(30d\) and \(20a\)) substantially reduces the affinity for the 5-HT\(_{1A}\) receptor while only marginally affecting the binding to the 5-HT\(_7\) receptor. It thus seems as the \textit{ortho} substituents, by forcing the 5- (or 8-) aryl group out of the plane of the
tetralin (or chroman) ring system, reduces favourable interactions with the 5-HT$_{1A}$ receptor.

Table 4. *In Vitro* Binding Affinities of (R)-8-aryl-3aminochromans and (S)-5-aryl-2-aminotetralins to 5-HT$_7$, 5-HT$_{1A}$ and D$_{2A}$ Receptors and effects on 5-HT$_7$ mediated stimulation of cAMP production.

<table>
<thead>
<tr>
<th>Compd R$'$ Ar Struct.</th>
<th>5-HT$_7$</th>
<th>5-HT$_{1A}$</th>
<th>D$_{2A}$</th>
<th>K$<em>i$-Ratio 5-HT$</em>{1A}$/5-HT$_7$</th>
<th>Efficacy$^b$ (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>30a Pr Ph A</td>
<td>3.38±0.53</td>
<td>0.87±0.28</td>
<td>NT$^c$</td>
<td>0.26</td>
<td>100</td>
</tr>
<tr>
<td>30b Pr 2-MeOPh A</td>
<td>1.73±0.28</td>
<td>11.7±1.7</td>
<td>NT</td>
<td>6.8</td>
<td>100</td>
</tr>
<tr>
<td>30c Pr 4-MeOPh A</td>
<td>12.4±0.6</td>
<td>1.49±0.36</td>
<td>NT</td>
<td>0.12</td>
<td></td>
</tr>
<tr>
<td>30d Pr 2,6-(MeO)$_2$Ph A</td>
<td>7.9±0.42</td>
<td>347±198</td>
<td>1210±10</td>
<td>44</td>
<td>100</td>
</tr>
<tr>
<td>30e H 2,6-(MeO)$_2$Ph A</td>
<td>78.7±5.1</td>
<td>&gt;1000</td>
<td>NT</td>
<td>&gt;12.7</td>
<td></td>
</tr>
<tr>
<td>30f Me 2,6-(MeO)$_2$Ph A</td>
<td>2.55±0.45</td>
<td>1420±210</td>
<td>241±46</td>
<td>257</td>
<td>Antag.</td>
</tr>
<tr>
<td>(R)-9b Pr Ph B</td>
<td>2.92±1.11</td>
<td>1.2±0.05</td>
<td>106±11</td>
<td>0.42</td>
<td>100</td>
</tr>
<tr>
<td>(S)-9b Pr 2-MeOPh B</td>
<td>2.7±0.35</td>
<td>10.3±1.6</td>
<td>512±110</td>
<td>3.8</td>
<td>100</td>
</tr>
<tr>
<td>9c Pr 4-MeOPh C</td>
<td>36±37</td>
<td>680±110</td>
<td>2110±440</td>
<td>1.9</td>
<td></td>
</tr>
<tr>
<td>20a Pr 2,6-(MeO)$_2$Ph B</td>
<td>12.6±2.3</td>
<td>1.3±0.15</td>
<td>741±200</td>
<td>0.10</td>
<td>91</td>
</tr>
<tr>
<td>20b Me 2,6-(MeO)$_2$Ph B</td>
<td>6.44±1.39</td>
<td>174±15</td>
<td>NT</td>
<td>&gt;189</td>
<td>28±3</td>
</tr>
<tr>
<td>4$^e$ 1.99</td>
<td>562</td>
<td>-</td>
<td>282</td>
<td>Antag.</td>
<td></td>
</tr>
<tr>
<td>4$^e$ 2.24</td>
<td>479</td>
<td>234</td>
<td>214</td>
<td>Antag.</td>
<td></td>
</tr>
<tr>
<td>5$^f$ 3.79</td>
<td>142</td>
<td>498</td>
<td>37</td>
<td>Antag.</td>
<td></td>
</tr>
</tbody>
</table>

$^a$The K$_i$-values are means ± standard errors of 2-3 experiments. The binding studies were performed essentially as described in Johansson *et al*.$^{79}$ $^b$cAMP production in CHO-cells in % of 5-HT stimulated production. Antag.=antagonist. $^c$NT = not tested. $^d$From ref. 14 $^e$From ref. 24 $^f$From ref. 25

The 4-methoxyphenyl (30c and 9c) and phenyl (30a and 9a) derivatives are equipotent at the 5-HT$_{1A}$ receptors while their affinity for 5-HT$_7$ receptors is reduced 4 times. Thus, the ligand binding site of the 5-HT$_7$ receptor appears to be more restricted for substituents in the para position of the 5- (or 8-)aryl group than the corresponding site in the 5-HT$_{1A}$ receptor.

Decreasing the size of the N-alkyl-substituents from propyl to methyl increases the selectivity for the 5-HT$_7$ receptor 6-7 fold. This increase is at least for the chromans due to the drop in affinity for the 5-HT$_{1A}$ receptor supporting earlier studies suggesting the existence of a propyl pocket at this receptor.$^{80}$ Not only the selectivity is affected by varying the alkyl substitution of the amino group, but also the signal transduction seems to be influenced by the choice of N-alkyl groups. While the 2,6-dimethoxyphenyl substituted 2-,(dipropylamino)-tetralin (30d) and 3-(dipropylamino)chroman (20a) are agonists at the 5-HT$_7$ receptor, the corresponding dimethylamino
substituted derivatives 30f and 20b are antagonist or weak partial agonist, respectively. The chroman derivatives appear to have a higher efficacy than the corresponding tetralin derivatives. The dipropylamino derivatives are all full agonists whereas compounds with a dimethylamino group show significantly lower efficacy. The increase in volume and lipophilicity of the N-substituents induces a dramatic change in intrinsic activity without affecting affinity, indicating that occupation of a propyl pocket is needed for G-protein activation of the 5-HT7 receptor.

5.2. SAR of (S)-6-Aryl-3-aminochromans (Paper II)

The ability of the novel (S)-6-aryl-3-aminochromans to bind to serotonin 5-HT7 receptors and in two cases to 5-HT1A receptors was studied. The affinities of the compounds for cloned rat 5-HT7 receptors expressed in Sf9-cells and labeled by [3H]5-HT as well as for cloned 5-HT1A receptors expressed in CHO-cells and labeled by [3H]8-OH-DPAT were determined in vitro. Efficacy of 27b at the 5-HT7 receptor was determined by measuring its effect on cAMP production in CHO cells in relation to the effect elicited by 5-HT.

The results are presented in Table 5 and compounds 1, 20a and 20b are included for comparative purposes.

Table 5. Biological Data of the Novel Derivatives 27a-27e.

<table>
<thead>
<tr>
<th>Compd</th>
<th>Ar</th>
<th>$K_i$ (nM)</th>
<th>$[^3H]5$-HT (5-HT1A)</th>
<th>$[^3H]5$-HT (5-HT7)</th>
<th>Ratio 5-HT1A/5-HT7</th>
<th>Efficacy</th>
</tr>
</thead>
<tbody>
<tr>
<td>27a</td>
<td>2,6-diMeOPh</td>
<td>367 ± 11</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>27b</td>
<td>2,6-diMePh</td>
<td>75.1</td>
<td>13.4 ± 15</td>
<td>6</td>
<td>76±11</td>
<td></td>
</tr>
<tr>
<td>27c</td>
<td>Ph</td>
<td>&gt;1000</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>27d</td>
<td>1-MeO-2-Naphthyl</td>
<td>594</td>
<td>75.9 ± 15</td>
<td>8</td>
<td></td>
<td></td>
</tr>
<tr>
<td>27e</td>
<td>2,6-diOHPh</td>
<td>&gt;1000</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1a</td>
<td></td>
<td>562</td>
<td>1.99</td>
<td>282</td>
<td>antagonist</td>
<td></td>
</tr>
<tr>
<td>20a</td>
<td></td>
<td>174 ± 15</td>
<td>6.44 ± 1.39</td>
<td>27</td>
<td>154 ± 11</td>
<td></td>
</tr>
<tr>
<td>20b</td>
<td></td>
<td>&gt;1000</td>
<td>5.29 ± 0.09</td>
<td>&gt;189</td>
<td>28 ± 3</td>
<td></td>
</tr>
</tbody>
</table>

*From ref.14  bFrom ref.81
The C6-phenyl derivative 27c does not bind to 5-HT\textsubscript{7} receptors, while the corresponding 2,6-dimethylphenyl derivative 27b has high affinity for the 5-HT\textsubscript{7} receptor and is a highly efficacious 5-HT\textsubscript{7} receptor partial agonist.

The affinity for the 5-HT\textsubscript{7} receptor decreased 27 times by changing the ortho substituents from dimethyl to dimethoxy as in compound 27a. This is in contrast to the 8-aryl-3-aminochroman series where the dimethoxy derivatives were identified as having high affinity for the receptor.

The corresponding resorcinol derivative 27e is devoid of affinity to the 5-HT\textsubscript{7} receptor. The 1-methoxy-2-naphthyl derivative 27d showed moderate affinity for the 5-HT\textsubscript{7} receptor and some selectivity over the 5-HT\textsubscript{1A} receptor.

A requisite for agonism in the 8-aryl-3-aminochroman series appears to be dipropyl substitution at the nitrogen as in compound 20a, while the dimethylsubstituted 20b is a weak partial agonist. In this series, the N,N-dimethylamino derivative 27b is a highly efficacious partial agonist.

To rationalize the difference in stereoselectivity and activity between 20b (the (R)-isomer) and 27b (the (S)-isomer) the two regioisomers can be overlaid as shown in Figure 23. We performed a Monte Carlo search in Macromodel using the MM2 force field to identify low-energy conformations of the selective 5-HT\textsubscript{7} ligands 20b (partial agonist) and 27b (partial agonist). The lowest energy conformations of 20b and the second lowest energy conformation of 27b were used to compare the two structures. In this fit the 8-aryl and 6-aryl substituents overlap.

![Figure 23](image)

Figure 23. Best fit of the 5-HT\textsubscript{7} partial agonists 20b (grey) and 27b (black). Mean distance between fitted atoms (centroids in the two aromatic rings, N, N-electron pair) is 0.67 Å.

The nitrogen atom and the nitrogen lone-pair also overlap and can interact with the same hypothetical binding sites in the receptor. However, the position of the oxygen in the chroman ring is different between the two isomers and may to a certain extent explain the differences observed between the
8- and 6-aryl series. There are other alternatives to overlay these structures. However, the present overlay rationalizes the difference in stereochemistry as well as having the common bulk of the compounds in the same region.

The present series of compounds, although limited, seems to constitute an interesting starting point for further structure-activity relationship (SAR) studies at 5-HT7 receptors.
6. Salvinorin A, a New Kappa Opioid Receptor Agonist

_Salvia divinorum_ (labiate) is a rare Mexican plant of the mint family. It has been used by the Mazatecs of Oaxaca in divinatory rites, but also as a panacea for a variety of disorders such as diarrhea, headache, rheumatism and anemia etc.\textsuperscript{82} The main active ingredient in the plant is the neoclerodane diterpene salvinorin A, which was isolated and characterized in 1984 by Valdes et al., together with the closely related diterpene Salvinorin B (Figure 24).\textsuperscript{83}

![Salvinorins](image.png)

**Figure 24.** The different salvinorins isolated from _Salvia divinorum._

Salvinorin C was isolated in 2001 by the same group (Figure 24).\textsuperscript{84} Both salvinorin B and salvinorin C are devoid of activity.

It was not until 2002 that salvinorin A was demonstrated to be a selective KOR agonist.\textsuperscript{85} Interestingly, salvinorin A had no action on the 5-HT\textsubscript{2A} receptor, which is considered to be the main target for the classical hallucinogens.\textsuperscript{85} The effective dose in humans has been determined to be in the 200-1000 μg range when smoked. The effects are intense hallucinatory experiences lasting several minutes to an hour. Salvinorin A is thus the most potent naturally occurring hallucinogen, rivalling the semi-synthetic hallucinogen lysergic acid diethylamide (LSD) in potency.

Salvinorin A is not only structurally distinct from naturally occurring hallucinogens such as psilocybin, \(N,N\)-dimethyltryptamine and mescaline, but also from other KOR agonists. One unique structural feature of salvinorin A
is the lack of nitrogens. All other known hallucinogens and KOR agonists contain a basic nitrogen. The discovery of salvinorin A as a potent, selective KOR agonist implicates the KOR as a novel molecular target for diseases characterized by perceptual distortions. KOR selective antagonists could thus be novel psychotherapeutic agents for diseases such as schizophrenia, dementia, bipolar disorders and Alzheimer disease amongst others.

6.1. Retrosynthetic Analysis

Salvinorin A is a synthetic challenge, with its tricyclic system and seven stereocenters adding to the complexity. The sensitivity of the furan ring was another factor to take in consideration, when planning the pathway for the total synthesis.

We chose a convergent synthesis, since ring A and C of salvinorin A contain all the functional groups. A Michael initiated ring closing reaction (MIRC)\textsuperscript{86-89} would then form ring B as the final step (Scheme 11). An advantage of this approach was that we could send the simplified parts A and C for pharmacological testing and start to evaluate the structural features of importance for the binding of salvinorin A to the KOR.

![Scheme 11. MIRC reaction as the final step.](image)

A structure similar to building block A, but lacking the methyl group, have previously been synthesized, with control of the stereochemistry.\textsuperscript{90,91} In the synthesis of building block A the final step is a RCM reaction just as in the previously published procedure (Scheme 12). The stereochemistry can be
controlled either via Brown´s allylboration of 3-furaldehyde\textsuperscript{92} or via enzymatic kinetic resolution of the homoallylic alcohol.\textsuperscript{93}

![Scheme 12. Retrosynthetic analysis of building block A.](image)

For the synthesis of building block C two routes are possible (Scheme 13), either using Hagemann’s ester 31 or 4-oxo-cyclohexanecarboxylic acid methyl ester (32) as the starting material.

There are several advantages in using Hagemann’s ester 31 as starting material. First of all it is commercially available. Furthermore, The conjugate addition of vinylmagnesium bromide in presence of cuprous iodide and the anti-Markovnikov hydrohalogenation of the double bond generating building block C are published procedures.\textsuperscript{94-96} The $\alpha'$-oxidation was considered to be the most problematic part in this scheme, partly because of the presence of additional acidic protons. In addition the stereochemistry will be a problem, since Hagemann’s ester 31 is racemic. However there are procedures for the preparation of derivatives of Hagemann’s ester 31 with a high %ee value.\textsuperscript{97,98}

![Scheme 13. Retrosynthetic analysis of building block C.](image)
The other route starts with the prochiral compound 32, which is oxidized in the α-position to the ketone. Dehydroisilylation of the silyl enol ether, followed by dehydroisilylation coupled with conjugate addition of organocuprate to the α,β-unsaturated ketone in the presence of trimethylchlorosilane, yields 33. The last two steps in this scheme are identical to the two last steps in the scheme using Hagemann’s ester 31. Stereochemistry is also a problem in this route, although the enolacetate of 32 can be subjected to enzymatic kinetic resolution. We anticipated the generation of the α,β-unsaturation as the major obstacle.

Initially we decided to leave the stereochemistry aside and see if the proposed mechanisms could achieve the desired transformations.

6.2. Isolation of Salvinorin A and Synthesis of Intermediates

The isolation of salvinorin A and the synthesis of various intermediates are described in the sections below.

6.2.1. Isolation of Salvinorin A from Salvia divinorum

In our initial attempts we tried to isolate salvinorin A by steeping the dried leaves from Salvia divinorum in diethylether over night, then filtering off the solids. The volatiles were evaporated. The residue was dissolved and subjected to solid phase extraction (SPE) with a reversed phase C-18 column, eluted with methanol to give salvinorin A in a very low yield.

Valdes et al. reported that they removed the nonpolar compounds by partitioning between hexanes and 90% aqueous methanol. No product was detected when Soxhlet extraction of the leaves, over 24 h, was tried followed by continuous extraction of an aqueous methanol phase with hexane over 24 h. Thus it is likely that salvinorin A rapidly decomposes in the presence of heat. This problem has been described in the recrystallization of salvinorin A from alcohols.

Another procedure is to steep the leaves (50g) in acetone (3x15 min), then evaporate the combined volatiles. The residue is then purified by chromatography and eluted with acetone through a column containing celite/activated charcoal, to yield an oil. In the original procedure the oil is triturated with diethylether, but in our hands methanol works better, providing salvinorin A as fine crystals. The extraction of 50 g of dried leaves yields ~90 mg of salvinorin A.
6.2.2. Synthesis of Building Block A and Analogues

The synthesis of building block A was achieved by treating 3-furaldehyde with 3-bromo-2-methyl-propene and Zn in a 9:1 solvent mixture of aqueous NH₄Cl and THF (Scheme 14). The homoallylic alcohol was used without any further purification in the next step. Williamson’s ether synthesis with allylbromide provided 35 in good yield. RCM, using Grubb’s 2nd generation catalyst furnished the desired 3,6-dihydro-2H-pyran ring 36 in excellent yield. Allylic oxidation with CrO₃/3,5-dimethylpyrazine at –50°C gave the building block A in low yield. Presumably the low yield is explained by the facile oxidation of the furan ring. For a discussion on the use of RCM and different catalysts for the synthesis of building block A and the other analogues see section 6.2.3.

![Scheme 14. Synthesis of building block A](image-url)

The analogues 39 and 41 were synthesized in a similar fashion, starting with the Barbier reaction with allylbromide, followed by Williamson’s ether synthesis or acetylation with acryloyl chloride providing the intermediate needed for the RCM (Scheme 15).
6.2.3. Notes on the RCM Reaction

An overview of the RCM reactions with the different substrates and the different catalysts are made in Scheme 16. For the mechanism of the RCM reaction see section 3.2.2.

Electron poor alkenes, such as acrylates, are known to be poor substrates for RCM, especially when ruthenium complexes are used. The ruthenium catalysts suffer from limited thermal stability and RCM to trisubstituted olefins have only been reported in special cases.\(^{106}\) RCM forming tertiary substituted olefins are unknown with catalysts.

Replacement of one of the phosphine ligands in Grubb’s 1st generation catalyst with a carbene ligand, such as in \(\text{ii,}\) leads to increased thermal stability and increased ring closing activity toward tri- and tetrakisubstituted substrates.

The molybdenium based catalysts such as Schrock’s catalysts \(\text{iii,}\) are more active than the ruthenium based catalyst systems,\(^{107,108}\) but suffers from being highly air and moisture sensitive. Another problem is the incompatability with many functional groups.

We started the investigation to use RCM with substrate \(\text{40,}\) in entry 4 (Scheme 16) using a published procedure.\(^{90}\) The acrylate was unreactive...
towards catalyst \(i\) in refluxing dichloromethane. Heating in toluene gave the same result. Adding Ti(O-iPr)_4 to the system gave 40 in a 60% yield, comparable to the yield (67%) obtained with the more active catalyst \(ii\) of the 2nd generation.

The RCM of the diene-ether substrate 38 in entry 3 (Scheme 16) yielded the desired product 39 in comparable yields under the same reaction conditions as above. This time Ti(O-iPr)_4 was not necessary as an additive. The yields were somewhat higher with catalyst \(ii\).

On the other hand, the substrate 35 in entry 2, giving a trisubstituted olefin as the product (Scheme 16), was totally unreactive toward catalyst \(i\), even in the presence of Ti(O-iPr)_4. The reaction proceeded smoothly in refluxing dichloromethane with catalyst \(ii\).

Substrate 42 in entry 1 (Scheme 16) was totally unreactive. Grubb’s 2nd generation catalyst \(ii\) failed, even at elevated temperatures in toluene, \(p\)-cymene and 1,2-dichloroethane. The reaction was also tried in a Smith Microwave Synthesizer with no success.

Schrock’s catalyst \(iii\) has been reported to be less sensitive to sterically hindrance. Treating 42 with 0.1 equiv of catalyst \(iii\), in dichloromethane at room temperature, provided, according to GC-MS, a product with the expected
mass. Further analysis with $^1$H NMR spectroscopy showed that the furan ring protons had disappeared and the presence of polymers. The low reactivity of 42 to undergo RCM transformation is likely a result of the combination of being electron poor and the sterical hindrance round the olefin. The direct synthesis of building block A via RCM of substrate 42 was abandoned and the indirect approach in Scheme 14 was used instead.

This small investigation highlights some aspects of RCM and the different reactivities of commercially available catalysts in the synthesis of heterocycles.

6.3. Synthesis of Simplified Analogues

The investigation of the pharmacophore of salvinorin A can be done in several ways. Synthesis of advanced intermediates in the total synthesis, direct chemical transformations or degradation studies of the natural product or by synthesizing different structural motifs contained in the original structure.

Using the isolated natural product as source for chemical transformations was not an option, since the cost of obtaining enough raw material to isolate salvinorin A was too high. Another disadvantage is the presence of different functional groups in the native structure, often rendering the desired chemical manipulations impossible to perform. This approach was used in a recent publication of Munro et al. They showed in this study that it is possible to transform the structure of Salvinorin A, while retaining affinity, functionality and selectivity. However the number of chemical transformation is limited.

Our approach was to mix the first and last options, in the sense that reactions used to synthesize the structural motifs must directly be applicable in the total synthesis scheme.

There is a simple model of the pharmacophore for salvinorin A in the 2002 article by Roth et al. disclosing salvinorin A as a KOR agonist, where the interaction with receptor mainly is thought to be mediated via hydrogen bonds.

We decided on two types of simplified analogues, both retaining the functional groups responsible for the potential hydrogen bonds (Figure 25).
Ring A and B is removed in the “free rotating chain analogue” 43, allowing the free rotation around the single bonds. Another feature of this analogue is the simplification of the stereochemistry. Only three stereocenters is remaining, of the original seven stereocenters in salvinorin A. The synthesis of 43 can be accomplished as shown in Scheme 17.

The utilized reactions in the two main steps (Michael addition and α-oxidation) are identical to the ones in the total synthesis. Michael addition to methyl acrylate as acceptor provides 45, which is hydroxylated in the α-position to 46. Protection of the hydroxy functionality, ester hydrolysis followed by esterification and acetylation would then give the target compound 43.

In the “two ring analogue” 44 the rings A and C are kept intact, while ring B is removed. Again the stereochemistry is simplified with five remaining stereocenters. The synthetic sequence is identical to the one above (Scheme 18). In this case we will have full control of the stereocenter in ring A. 93
However, the Michael addition to 41 will be considerably harder to perform, compared with the Michael addition in Scheme 17.

Both these schemes open up the possibility of synthesizing analogues with different functional groups. For example, it would be possible to make the Michael addition to acrylonitrile, reduce the nitrile or hydrolyze it to the amide or in both scenarios exchange the furan ring against some other heteroaromatic.

The schemes become even more interesting in the light of the recent published studies of the salvinorin A pharmacophore, where it is shown that it is possible to transform some of the functional groups while retaining affinity and functionality.110

6.3.1. The Michael Reaction

The Michael addition, in its original meaning, is the addition of an enolate derived from a carbonyl (donor) to an electrophilic carbon that forms a part of a conjugated system (acceptor). The original paper appeared in 1887, by A. Michael,111 and has since then been one of the major reactions for forming new C-C bonds. It has to be observed that the Michael reaction is a reversible process and can as such cause problems under certain circumstances.112

There are two major limitations in reactions of the Michael type (Figure 26):

1. The necessity of using a strong base to transform the carbonyl compound into its anion.
2. Proton transfers between the initially generated product and the enolate.
Metal enolates derived from ketones and esters typically function as Michael donors and \(\alpha,\beta\)-unsaturated ketones and esters are often used as Michael acceptors. Four types of combinations between donors and the acceptor can arise. The reaction between a ketone enolate and a \(\alpha,\beta\)-unsaturated ester, as in our case, have hardly been reported, unless there is a subsequent step such as a cyclization contributing to the stabilization of the product. This can be explained by the fact that the generated ester enolates are more labile than their counterpart, the ketone enolate.

The problem with a charged enolate anion was solved by the pioneering work of Stork with enamines as non-charged enolates. Enamines had a unique status as non-charged enolate equivalents until the 1970’s when silyl enol ethers were introduced as enolate equivalents. Still the enamines are better nucleophiles than the silyl enol ethers, due to the lower electronegativity of the nitrogen. Another advantage is the simple path for proton transfer in the product, leading to the neutral molecule (Figure 27)."}

Silyl enol ethers on the other hand typically requires the addition of a Lewis acid to increase the reactivity. The advantage with silyl enol ethers is the possibility of controlling the formation of the thermodynamic vs the kinetic silyl enol ether, in the case of unsymmetrical ketones.

Neutral reaction conditions are necessary in instances when there are additional acidic protons in the substrate. In our hands the use of silyl enol ethers completely failed, while the enamine approach worked fine. Attempts to use the hydrazone derivative of acetone as the Michael donor for the addition to compound 41 (Scheme 19) also failed.
6.4. Synthetic Studies for Simplified Analogues

The enamine 47 was formed by refluxing the commercially available ethyl 4-oxocyclohexanecarboxylate with pyrrolidine in benzene under azeotropic conditions (Scheme 20) for 14 h. The volatiles were evaporated and the crude enamine 47 was heated with 0.9 equiv. methyl acrylate in anhydrous EtOH to 50°C for 2 h followed by hydrolysis by H2O (1.5 equiv.).

Kugelrohr distillation afforded ester 48 in 70% yield accompanied by the diester in 15% yield. It was of outmost importance to use “dry” ethanol, even small amounts of H2O in the EtOH had a deteriorating effect on the reaction. Other solvents, such as THF provided no product. Furthermore, reaction temperatures above 50°C drastically decreased the yield to less than 30%.

Treating the ester 48 with triethylsilyl triflate (TESOTf) and Et3N, followed by oxidation with dimethyldioxirane (DMDO) and hydrolysis with triethylamine trihydrofluoride provided a mixture of the secondary alcohol 50a and the tertiary alcohol 50b (Scheme 22). Only the secondary alcohol was silylated when the mixture of the alcohols 50a and 50b was treated with tert-butyldimethylsilyl chloride (TBDMSCl), imidazole and a catalytic amount of 4-dimethylaminopyridine (DMAP),123 thus allowing a convenient separation of the two regioisomers.
Scheme 21. α-Oxidation of compound 49 and the separation of the regioisomers 50a and 50b.

The epoxidation of the silyl enol ether with DMDO was highly stereoselective, providing only the two diastereomers 51a and 51b of the four possible (Table 6).

Table 6. Obtained diastereomers after oxidation of 49.

<table>
<thead>
<tr>
<th>Diastereomeric relationship of oxidation product 51a</th>
</tr>
</thead>
<tbody>
<tr>
<td>3 : 2 : 0 : 0</td>
</tr>
</tbody>
</table>

Table 6. Obtained diastereomers after oxidation of 49.

The full characterization and determination of the diastereomeric relationship was performed by NMR spectroscopy, using gHMBC, gHSQC, gNOESY, e-COSY and TOCSY experiments.

Both the diastereomers 51a and 51b contain the α-substituents in a cis relationship. Thus the relative stereochemistry is correct compared to salvinorin A. An explanation for the observed stereoselectivity is that the orientation of the bulky tert-butyl dimethyl silyl group in the silyl enol ether depends on the corresponding α-substituent, favouring the formation of the epoxide cis to the same α-substituent.

The study described here provided the desired compound 51 with the correct relative stereochemistry. Further investigations focusing on the separation of the initial cis/trans isomers and improving the ratio of the kinetic vs. the thermodynamic silyl enol ether in the epoxidation sequence is ongoing.
6.5. Enzymatic Kinetic Resolution (Paper III)

We required enantiomerically pure 1-(3-furyl)-3-buten-1-ol ((S)-37 or (R)-37) for the synthesis of simplified analogues of salvinorin A (Figure 28).

![Salvinorin A together with the enantiomers of compound 37.](image)

The enantiomers of 37 have previously served as an important building block in the total synthesis of several natural products and have been synthesized either via Brown allylation of 3-furaldehyde or by enzymatic kinetic resolution of 37. Two obvious reasons made the enzymatic kinetic resolution approach attractive for us:

1. The tedious laboratory protocol for the Brown allylation. We anticipated problems, such as filtration under inert conditions and maintaining a reaction temperature of –100°C, in the multigram scale we wanted to perform.

2. The reported %ee of the procedure is 93-96 with no chance of further improvement. For useful SAR purposes a %ee of >98 is normally needed.

6.5.1. The Enantiomeric Ratio, $E$

An attractive feature of enzymatic kinetic resolutions is the possibility of calculating the enantiomeric ratios $E_s$ and $E_p$. $E_s$ relates the extent of conversion of starting material to product, $c$, to the enantiomeric excess of the remaining starting material, $ee_s$, while $E_p$ relates $c$ to the enantiomeric excess of the product $ee_p$. While $ee$ is a property of the product or starting material, $E$ is the characteristic of a process.

$E$ describes the enantioselectivity under a set of particular physical conditions (solvent, temperature, pH etc) with a certain substrate and a specific enzyme. Thus calculation of the $E$ value facilitates the rapid evaluation of an enzyme’s enantioselective property in a screening procedure. The enantiomeric ratios for the substrate $E_s$ and for the product $E_p$ were calculated us-
ing Sih’s method,\(^ {133}\) equation (1) and (2), assuming irreversibility for the biocatalytical hydrolysis (Figure 29).

\[
E_s = \frac{\ln[(1 - c)(1 - ee_s)]}{\ln[(1 - c)(1 + ee_s)]} \quad (1)
\]

\[
E_p = \frac{\ln[1 - c(1 + ee_p)]}{\ln[1 - c(1 - ee_p)]} \quad (2)
\]

Figure 29. Sih’s equations for calculation of the enantiomeric ratios, \(E_s\) and \(E_p\).

The course of the kinetic resolution is different for the product and the substrate. Therefore it is important to distinguish between the enantiomeric ratio of the substrate \((E_s)\) and that of the product \((E_p)\). For the substrate, a high enantiomeric purity value \((ee_s > 98\%)\) can be obtained even with an \(E_s\) value as low as 5. However, for the product the \(ee_p\) value initially equals \((E-1)/(E+1)\), then at \(c \sim 50\%\), the \(ee_p\) value rapidly diminishes ending up as the racemic product at \(c = 100\%\). Only systems with \(E_p > 100\) can be used to obtain \(\%ee_p > 98\). This kind of enantioselectivity is often not obtainable\(^ {132}\) and other strategies such as product recycling\(^ {133}\), amongst others, have to be considered in order to obtain the desired \(ee_p\).\(^ {134}\)

6.5.2. Synthesis and Screening

In 2002 Bierstedt \textit{et al.}\(^ {93}\) published the enzymatic kinetic resolution of 37. The highest \(E\) value was achieved via enantioselective hydrolysis of 53 using a lipase from the \textit{Alcaligines} species with an \(E_s\) value of 80. However, the value appears to have been miscalculated, since recalculation using equation (1)\(^ {133}\) gave an \(E_s = 29\). Unfortunately most of the enzymes used in their work were from Boehringer Mannheim and are no longer commercially available. Also, even if enzymes are from the same organism different fermentation and purification methods produce enzymes with different characteristics (probably due to different composition of isoforms). In order to obtain gram quantities of enantiomerically pure 37 we have reassessed the screening procedure using new sets of lipases obtained from Fluka (Lipase basic kit) and from Europa Bioproducts Ltd (\textit{Alcaligines} spp12 and \textit{Alcaligines} spp20).

The synthesis of 53 (Scheme 22) is straightforward and high yielding. 3-Furaldehyde was subjected to a Barbier reaction\(^ {103}\) with allylbromide and Zn powder in a saturated solution of ammonium chloride and THF providing the racemic alcohol 37 in 99% yield. The alcohol was then acetylated with
Scheme 22. Synthesis of racemic 3, the substrate for the enzymatic kinetic resolution.

acetic anhydride to afford 53, the starting material for the enzymatic kinetic resolution.

It follows from the discussion of $E$ values above, that it is of outmost importance to have the means to, with great accuracy, monitor the conversion and the enantiomeric excesses of both substrate and product in the enzymatic kinetic resolution. These means has to be tested and evaluated before the actual screening procedure begins.

We chose $^1$H NMR spectroscopy to follow the conversion ($c$) with an internal standard added. There are many advantages to use $^1$H NMR spectroscopy compared with other methods such as GC or HPLC. The foremost being that no work-up before or other manipulations is required before the sample is run. The internal standard was added for the calculation of the actual yield in the hydrolysis (Scheme 23).

The other parameter to monitor is the $ee$ value. The separation of the ester 53 was obtained by chiral GC and the corresponding alcohol 37 was separated by chiral HPLC.

The screening procedure for the hydrolysis in Scheme 23 was performed with 11 different lipases in 10 mL of a 9:1 mixture of a phosphate buffer (pH 7) and DMSO, at 40°C, using 100 mg of substrate for each lipase. A control experiment, without any enzyme, was performed to assure that the substrate was not subjected to any competing non-catalyzed background reactions.

The results are presented in Table 7 and entry 14 is included for comparison. The lipase from Candida cylindracea gave a reasonable high $E_s$ value of 22, but showed only a 29% conversion to the alcohol over 5 days.

The $E_s$ values for Alcaligines spp20 from Europa Bioproducts Ltd and the lipase from the same organism, from Boehringer Mannheim (entry 14), used
in the investigation of Bierstedt et al. were comparable, $E_s = 24$ and $E_s = 29$, respectively. It would indeed be possible to change the commercial sources of lipases from *Alcaligines* species, while retaining the enantioselectivity. The most efficient lipase identified in our screening was *Pseudomonas fluorescens*, with $E_s = 38$ and $E_p = 58$.

To gain scale-up experience with these enzyme reactions, we chose to use the lipase from *Pseudomonas cepacia*. This lipase gave a reasonably good $E_s$ value of 13 and is considerably cheaper compared with *Pseudomonas fluorescens*. The scale-up experiment was performed with 793 mg of substrate, affording, a similar $E_s$ value (13) and conversion (63%) as in the screening procedure ($E_s = 12$ and 66% conversion).

**Table 7. Enzyme Catalyzed Hydrolysis of (+)-53.**

<table>
<thead>
<tr>
<th>Entry</th>
<th>Enzyme source</th>
<th>Time (h)</th>
<th>$c$ (%)</th>
<th>$ee_s$ (%)</th>
<th>$ee_p$ (%)</th>
<th>$E_s$</th>
<th>$E_p$</th>
<th>$[\alpha]_D^{19}$</th>
<th>$[\alpha]_D^{19}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Aspergillus</td>
<td>96</td>
<td>81</td>
<td>84</td>
<td>19</td>
<td>3</td>
<td>3</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>2</td>
<td>Candida antarctica</td>
<td>120</td>
<td>$&lt;10$</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>3</td>
<td>Candida cylindracea</td>
<td>120</td>
<td>29</td>
<td>36</td>
<td>48</td>
<td>22</td>
<td>3</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>4</td>
<td>Mucor miehei</td>
<td>144</td>
<td>$&lt;10$</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>5</td>
<td><em>Pseudomonas cepacia</em></td>
<td>72</td>
<td>67</td>
<td>98</td>
<td>72</td>
<td>12</td>
<td>n.c.</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>6</td>
<td><em>Pseudomonas fluorescens</em></td>
<td>72</td>
<td>51</td>
<td>89</td>
<td>89</td>
<td>38</td>
<td>58</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>7</td>
<td>Rhizopus arrhizus</td>
<td>120</td>
<td>$&lt;10$</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>8</td>
<td>Rhizopus niveus</td>
<td>144</td>
<td>$&lt;10$</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>9</td>
<td>Hog pancreas</td>
<td>144</td>
<td>$&lt;10$</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>10</td>
<td><em>Alcaligines spp</em>12</td>
<td>192</td>
<td>19</td>
<td>3</td>
<td>61</td>
<td>1</td>
<td>4</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>11</td>
<td><em>Alcaligines spp</em> 20</td>
<td>72</td>
<td>57</td>
<td>96</td>
<td>85</td>
<td>24</td>
<td>n.c.</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>12</td>
<td>Control</td>
<td>144</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>13</td>
<td><em>Pseudomonas cepacia</em></td>
<td>72</td>
<td>63</td>
<td>96</td>
<td>74</td>
<td>13</td>
<td>n.c.</td>
<td>$-25$&lt;sup&gt;(S)&lt;/sup&gt;</td>
<td>$20$&lt;sup&gt;(R)&lt;/sup&gt;</td>
</tr>
<tr>
<td>14&lt;sup&gt;p&lt;/sup&gt;</td>
<td><em>Alcaligines</em></td>
<td>7</td>
<td>55</td>
<td>95</td>
<td>94</td>
<td>29&lt;sup&gt;p&lt;/sup&gt;</td>
<td>n.c.</td>
<td>$-31$&lt;sup&gt;(S)&lt;/sup&gt;</td>
<td>$26$&lt;sup&gt;(R)&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a</sup>Determined by $^1$H NMR. $^{ee_s}$ = enantiomeric excess of substrate. Determined by GC on a Supelco ß-Dex 120 column. $^{ee_p}$ = enantiomeric excess of product. Determined by HPLC on a Chiracel OD-H column. $^{E_s}$ = enantiomeric ratio of substrate, calculated by eq. (1). $^{E_p}$ = enantiomeric ratio of product, calculated by eq. (2). $^{[\alpha]_D}$ = [\alpha]_D for the substrate, measured on the alcohol (c 1.0, CH2Cl2), see footnote m. $^{[\alpha]_D}$ = [\alpha]_D for the product (c 1.0, CH2Cl2). $^{Fluka lipase basic kit.}$ = = not determined. $^{n.c.}$ = not possible to calculate with eq. (2). $^{Lipases from Europa Bioproducts Ltd.}$ $^{Same reaction conditions, but without enzyme.}$ $^{Scale up experiment with 793 mg of acetate.}$ $^{The ester was hydrolysed with 2M NaOH, then the $ee_s$ was determined for the alcohol by HPLC on a Chiracel OD-H column.}$ $^{Absolute configuration determined indirectly by measuring the [\alpha]_D value and comparing with ref. 5.}$ $^{See ref. 5.}$ $^{Value from ref 5.}$ $^{Value from ref 5.}$ $^{Value from ref 5.}$ $^{Value from ref 5.}$
Interestingly Sih’s equation (2) failed to calculate $E_p$ for *Pseudomonas cepacia* (entry 5) and *Alcaligines* spp20 (entry 11) and for *Alcaligines* in entry 14. In all three cases the $ee_p$ values were too high for equation (2) to handle.

A plausible explanation for the failure to use Sih's equations might be that not only is the reaction reversible but also enantioselective in the synthetic direction towards the ester.

### 6.5.3. Conclusions

*Pseudomonas Fluorescens* (Fluka) was found to give the highest enantiomeric ratio of the 11 lipases screened. At 51% conversion the $ee$ value ($ee_p$) for the product was found to be 89%, giving an enantiomeric ratio ($E_p$) of 58, while the $ee$ value ($ee_s$) for the substrate was 89%, giving an enantiomeric ratio ($E_s$) of 38. These values are higher than for the previously reported but no longer commercially available lipases, making *Pseudomonas Fluorescens* a suitable substitute to obtain the desired %$ee$ of >98.

### 6.6. Formation of 1-Silyloxy-1,3-dienes from Hagemann’s Ester and other Cyclic Conjugated Enones, and Their Oxidation with Dimethyldioxirane (Paper IV)

The background for this paper is an extensive search for the proper reaction conditions to obtain the $\alpha'$-hydroxylated derivate 55 (Scheme 25), which appears to be accessible from compound 31. The commercially available Hagemann’s ester 31 (Scheme 24 and 25) was first synthesized in 1893. Compound 31 and various analogues have since then served as valuable building blocks in the synthesis of complex molecules such as terpenes, taxanes and steroids.

Even assymetric methodology using hydrazones (Scheme 24), developed by Enders et al. was tried in the attempts to obtain compound 55. It was not the possibility of performing the $\alpha'$-hydroxylation in a stereoselective manner that made us interested in this approach, but the change of the protic properties in Hagemann’s ester 31. However, the attempts to abstract the proton in the $\alpha'$-position were futile, even though the hydrazone was conveniently generated under azeotropic conditions.
In an attempt to obtain the precursor 55 using triethylsilyl chloride (TESCl) and lithium diisopropylamide (LDA) (Scheme 25), we instead observed the formation of the two possible 1-silyloxy-1,3-diene regisomers. Oxidation of the 1-silyloxy-1,3-dienes with DMDO generated the γ-hydroxylated derivatives. In this paper (Paper IV) we present a convenient way of generating 1-silyloxy-1,3-dienes from cyclic conjugated enones containing a primary methyl group in the β-position and the regioselective oxidation of silyloxy-dienes with DMDO.

6.6.1. Formation of 1-Silyloxy-1,3-dienes

Selective formation of 2-silyloxy-1,3-dienes from conjugated enones is commonly achieved by α'-deprotonation with LDA followed by quenching with a trialkyl silyl chloride such as trimethyl silyl chloride (TMSCl). However, applying these conditions at −78°C gave a 1:1 mixture (determined by 1H NMR spectroscopy) of the two possible regioisomers of 1-silyloxy-1,3-dienes, 54a and 54b (scheme 1).

Several bases and silylating agents were screened and the results are summarized in Table 8. Compound 54a (entry 1b) was formed with no trace of the regioisomer 54b when the base was changed from LDA to lithium hexamethyldisilazide (LHMDS). The reaction was clean and proceeded with 100% conversion as determined by 1H NMR spectroscopy.
Compound 54b could be formed, accompanied by a small amount of the regioisomer 54a, when 31 was treated with TESOTf and Et3N (entry 1c). Because of the instability of these compounds, resulting in decomposition during purification attempts (distillation or chromatography), yields were determined by NMR quantification against added 1,4-dimethoxy benzene. The reaction was completely regioselective for 54b when the more sterically hindered base DIEA was used instead of Et3N (entry 1d). There was no trace of the corresponding 2-silyloxy-1,3-diene 54c (Scheme 25) in any of the reactions.

Table 8. Regioselective Formation of 1-Silyloxy-1,3-Diene Derivatives.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Base</th>
<th>Subst.</th>
<th>Conditions</th>
<th>Time</th>
<th>Product</th>
<th>Yield (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1a</td>
<td>LDA</td>
<td>31</td>
<td>TESCl (2 eq), THF, -78°C</td>
<td>15 min</td>
<td>1:1</td>
<td>ND</td>
</tr>
<tr>
<td>1b</td>
<td>LHMDS</td>
<td>31</td>
<td>TESCl (1.3 eq), THF, -78°C</td>
<td>15 min</td>
<td>0:1</td>
<td>85b</td>
</tr>
<tr>
<td>1c</td>
<td>Et3N</td>
<td>31</td>
<td>TESOTf (1.5 eq), CH2Cl2, 0°C</td>
<td>30 min</td>
<td>1:1</td>
<td>ND</td>
</tr>
<tr>
<td>1d</td>
<td>DIEA</td>
<td>31</td>
<td>TESOTf (1.5 eq), CH2Cl2, 0°C</td>
<td>30 min</td>
<td>1:0</td>
<td>80b</td>
</tr>
<tr>
<td>1e</td>
<td>TBAF</td>
<td>31</td>
<td>ETSA² (1.2 eq), THF, -78°C</td>
<td>1 h</td>
<td>NR</td>
<td></td>
</tr>
</tbody>
</table>

To confirm the importance of steric hindrance of the base, we subjected 3-methyl-2-cyclohexen-1-one 56 to the same reaction conditions as in entry 1d (DIEA and TESOTf), and obtained exclusively the 1-silyloxy-1,3-diene 57b (entry 2a). In contrast, the use of the stronger base LDA has been reported to give the kinetic product 57c, and the weak base Et3N resulted in the formation of a regioisomer mixture of 57b and 57c (entries 2b and 2c). Thus, this method is a compliment to existing synthetic methodology. It is also interesting that no trace of the isomeric 2-silyloxy-1,3-dienes 54c and 57c were formed.
6.6.2. Oxidations of Silyloxy-dienes

Hydroxylations of 1-silyloxy-1,3-dienes are not very common.\textsuperscript{144} They occur usually at the 2-position.\textsuperscript{144,145} Suryawanshi et al., in their study of the total synthesis of the antileukemia agent Bruceantin hydroxylate exclusively at the γ position.\textsuperscript{146} Few examples of hydroxylation at the γ position in 1-silyloxy-1,3-dienes exist. The oxidations are mostly performed with \textit{m}-chloroperbenzoic acid (MCPBA),\textsuperscript{144} but other oxidants have been used, such as potassium peroxymonosulfate.\textsuperscript{146}

Oxidation of 54a with freshly purified MCPBA was accompanied by extensive hydrolysis of the silyl enol ether, resulting in unsatisfactory yields. The neutral oxidant 2-benzenesulfonyl-3-phenyloxaziridine\textsuperscript{147} did not react with 54a at room temperature. DMDO is another neutral oxidant\textsuperscript{148,149} that has been used to epoxidize silyl enol ethers, enabling the isolation and characterization of these labile epoxides.\textsuperscript{150,151}

Thus, the tert-butyl dimethyl silyl enol ether 54d of Hagemann’s ester 31 (Scheme 26) was prepared with the method described in entry 1d (Table 8). After being cooled to –78°C, a –20°C acetone solution of DMDO was added. The reaction mixture was stirred for 30 min and then hydrolyzed with triethylamine trihydrofluoride. Standard work-up procedures afforded the γ-hydroxylated derivative 58 of Hagemann’s ester, in 45% isolated yield (entry 1, Table 9). This is comparable with the published yield of 50%.\textsuperscript{152}

Scheme 26. Regioselective formation and epoxidation of the 1-silyloxy-1,3-dienes of Hagemann’s ester.

We wanted to further explore DMDO as a general oxidant for silyloxy dienes. Entry 2 shows the exclusive oxidation of 1-silyloxy-1,3-diene 54b at the γ-position. The formation of compound 59 can be explained by rearrangement of the initially formed exocyclic epoxide during evaporative concentration of the product, as has been described for related epoxides derived from silyl enol ethers.\textsuperscript{150}
**Table 9. Regioselective Oxidations of 1-Silyloxy- and 2-Silyloxy-1,3-Dienes with DMDO.**

<table>
<thead>
<tr>
<th>Entry</th>
<th>Substrate</th>
<th>Product</th>
<th>Ratio trans/cis</th>
<th>Yield (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>OTBDMS COOEt</td>
<td>54d</td>
<td>OHO</td>
<td>45(^*)</td>
</tr>
<tr>
<td>2</td>
<td>OTES COOEt</td>
<td>54b</td>
<td>COO EtOTES O</td>
<td>65(^*)</td>
</tr>
<tr>
<td>3(^c)</td>
<td>TESO</td>
<td>60</td>
<td>3:1</td>
<td>53(^*)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>2:3(^d)</td>
<td>80(^d)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>2:2:1(^d)</td>
<td>59(^d)</td>
</tr>
</tbody>
</table>

The full characterization of the compounds were performed by NMR, using gHMBC\(^{124}\), gHSQC\(^{125}\), gNOESY\(^{126}\), P.E-COSY\(^{127}\) and TOCSY\(^{128}\) experiments. NMR on crude reaction mixtures was performed using the Wet pulse sequence\(^{141}\). \(^*\)Isolated yield. \(^\text{b}^\circ\) 1H NMR yield. \(^c\)Et\(_3\)N used as the base, instead of DIEA, in the silylation procedure. \(^d\)Included for comparison, ref. 5. \(^e\)Included for comparison, ref. 21.

Silylation of (+)-4-cholesten-3-one (entry 3) followed by oxidation with DMDO of the not isolated silyloxy diene 60 resulted in 53% isolated yield of the 2-hydroxy derivative 61 based on the steroid. This is in the same range as previously obtained using triphenyl phosphite ozonide.\(^{153}\) A crude product yield of 80% using MCPBA has also been reported.\(^{138}\) However, DMDO shows a higher stereoselectivity (2α:2β = 3:1) than triphenyl phosphate ozonide (2α:2β = 2.2:1)\(^{153}\) and MCPBA (2α:2β = 2:3)\(^{138}\).

In conclusion, silylation of α,β-unsaturated cyclic ketones using DIEA/TESOTf followed by oxidation with DMDO appears to be an interesting method for the preparation of unsaturated α- or γ-hydroxyketones.
7. Concluding Remarks

Two systems within the CNS were addressed in this thesis, the serotonergic and the opioid system. In the part dealing with the serotonergic system we have evaluated SAR of novel 3-aminochromans for the 5-HT_7 receptor. During the course of this study we have:

1. Synthesized and identified **20b** as a >189 fold selective serotonin receptor 5-HT_7 partial agonist over the 5-HT_{1A} receptor.
2. Synthesized and identified **20a** as a serotonin receptor 5-HT_7 agonist with 27 fold selectivity over the serotonin 5-HT_{1A} receptor.
3. Identified **27b** as a highly efficacious partial agonist for the serotonin receptor 5-HT_7 by moving the aryl-substituent from the C8 position to the C6 position of the 3-aminochroman.
4. Performed a Monte Carlo search to identify the low-energy conformations of the partial agonists **20b** and **27b** providing a rationale for the observed differences between the 8-aryl and 6-aryl series.

In the part dealing with the opioid system we have initiated the total synthesis of salvinorin A and developed a method for the synthesis of simplified analogues. During the course of this study we have:

1. Synthesized building block **A**, a key intermediate in the total synthesis of salvinorin A.
2. Resolved alcohol **37** via enzymatic kinetic resolution, providing a route to enantiomerically pure building block **A** and analogues.
3. Developed a method for the regioselective formation of 1-silyloxy-1,3-dienes from cyclic conjugated enones.
4. Developed a method for the regioselective oxidation of 1-silyloxy1,3-dienes with DMDO, providing easy access to a hydroxyl functionality at the γ-position in α,β-unsaturated ketones.
8. Acknowledgments

This investigation was carried out at Organic Pharmaceutical Chemistry, Department of Medicinal Chemistry, Faculty of Pharmacy, Uppsala University. The years at the department have been both creative and enjoyable, thanks to all present and former colleagues.

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Mina barn, Moses, Simon och Saga. Ni är helt fantastiska och det käraste jag har.

Maria, min kära och bästa vän här i livet. För att du alltid finns vid min sida.
9. References


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10. Appendix

10.1. Supplementary Information Paper I

Pär Holmberg and Anette M. Johansson.

Experimental

Melting points were determined on a Büchi SMP-20 apparatus. $^1$H and $^{13}$C NMR spectra were recorded at ambient temperature on a Varian Unity 400 MHz, Varian MercuryPlus 300 MHz, or a JEOL JNM-EX 270 MHz instrument. Chemical shifts are referenced indirectly to tetramethylsilane (0.0 ppm) via the (residual) solvent signals. Coupling constants are given in Hertz, and the splitting patterns are designated as follows: s, singlet, d, doublet, dd, doublet of doublets, t, triplet, q, quartet, quint, quintett, hept, heptet, sext, sextet, oct, octett and app for apparent. Infrared spectra (IR) were obtained on a Perkin-Elmer 1605 FT-IR spectrophotometer. Thin layer chromatography was performed on silica gel (Merck) mounted on aluminium cards or glass plates and with a fluorescent indicator (254 nm). Preparative chromatographic separations were achieved by using silica gel (Merck) or alumina (Merck). Elemental analyses (C,H,N) were performed by Mikro Kemi AB, Uppsala, Sweden or by Analytische Laboratorien, Gummersbach, Germany. The values were within ±0.4% of theoretical if not otherwise indicated.

8-Methoxy-2H-chromene-3-carbonitrile (12). 2-Hydroxy-3-methoxybenzaldehyde 200 g (1.3 mol), acrylonitrile 349 g (6.6 mol) and 1,4-diazabicyclo[2.2.2]octane (“DABCO”) 37 g (329 mmol) were refluxed for 26 h. The reaction mixture was diluted with 500 mL Et$_2$O and washed with 1 M NaOH and 1 M HCl. The organic phase was dried over K$_2$CO$_3$ and concentrated to afford an oil which was flash column chromatographed [SiO$_2$, CH$_2$Cl$_2$] to yield 178 g (73%) of 12 as crystals.

$^1$H NMR (CDCl$_3$) δ 7.16 (m, 1H), 6.94-6.86 (m, 2H), 6.73 (m, 1H), 4.85 (d, 2H), 3.87 (s, 3H); $^{13}$C NMR (CDCl$_3$) δ 148.1, 143.2, 140.0, 122.3, 120.8, 120.4, 116.4, 115.3, 103.6, 64.6, 56.2.
8-Methoxy-2H-chromene-3-carboxylic acid (13). A solution of 12 (27.5 g, 147 mmol) in 400 mL of 10% aqueous NaOH was refluxed for 4 h under N₂. The cooled solution was washed with 3x200 mL Et₂O, followed by acidification with conc. HCl. The precipitate was filtered off and recrystallized in EtOH/H₂O to yield 13 (23 g, 77%) as colorless needles.

\(^1\)H NMR (DMSO-D₆) δ 12.87 (s, 1H), 7.41 (d, 1H), 7.02-6.97 (m, 1H), 6.92-6.86 (m, 2H), 4.87 (s, 2H), 3.75 (s, 3H); \(^{13}\)C NMR (DMSO-D₆) δ 166.1, 148.1, 144.0, 132.0, 124.1, 122.1, 122.0, 121.3, 115.6, 64.6, 56.2.

8-Methoxy-chroman-3-one (14). To a solution of 13 (20 g, 97 mmol) in 15 mL of Et₃N and 200 mL of CH₂Cl₂ was added diphenylphosphoryl azidate (26.7 g) in 80 mL toluene added dropwise while the reaction mixture was heated to distill off CH₂Cl₂. 200 mL Toluene was added when 100 mL CH₂Cl₂ had been distilled. The reaction mixture was heated for 3 h at 80°C, then 160 mL 6 M HCl was added dropwise and the mixture refluxed for 1 ½ h. The phases were separated and the organic phase washed with saturated 3x100 mL NaHCO₃, 3x100 mL brine and dried over MgSO₄. The volatiles were evaporated to afford an oil which was further purified by filtering through Al₂O₃ with CHCl₃/MeOH (98:2) as eluent to yield 15.1 g (84%) of 14.

\(^1\)H NMR (CDCl₃) δ 6.99 (t, 1H), 6.84 (m, 1H), 6.71 (m, 1H), 4.44 (s, 2H), 3.88 (s, 3H), 3.59 (s, 2H); \(^{13}\)C NMR (CDCl₃) δ 207.5, 149.4, 143.7, 123.5, 122.8, 120.6, 110.9, 73.2, 56.1, 48.8.

8-Methoxy-3-(benzylamino)chroman (15). Acetic acid (60 mL) was added to a solution of 14 (39.2 g, 220 mmol) and benzylamine (47 g, 439 mmol) in 300 mL THF. The solution was allowed to cool to RT, then NaBH₃CN (41.4 g, 659 mmol) was added dropwise at a rate to maintain reflux. The solution was cooled to RT and stirred overnight. The reaction was quenched with 300 mL 1M NaOH and extracted with 3x200 mL CH₂Cl₂. The extracts were pooled and washed with 3x100 mL brine, dried with K₂CO₃ and concentrated. The residue was subjected to flash column chromatography [CHCl₃/MeOH (97:5:2,5), followed by CHCl₃/MeOH (95:5) and finally by CHCl₃/MeOH (91:1)] to yield 48.3 g (82%) of 15 as an oil.

\(^1\)H NMR (CDCl₃) δ 7.39-7.20 (m, 5H), 6.84-6.63 (m, 3H), 4.29 (m, 1H), 4.03 (m, 1H), 3.91 (d, 2H), 3.85 (s, 3H), 3.17 (m, 1H), 3.01 (m, 1H), 2.71 (m, 1H); \(^{13}\)C NMR (CDCl₃) δ 148.01, 143.4, 139.8, 128.4, 128.0, 126.0, 121.8, 121.0, 120.1, 109.0, 68.8, 55.6, 50.9, 49.0, 31.8.

8-Methoxy-3-(dibenzylamino)chroman (16). A solution of 15 (29.4 g, 109 mmol), benzyl bromide (112.3 g, 656 mmol), KI (54.5 g, 328 mmol) and K₂CO₃ in (45.4 g 328 mmol) 55 mL CH₃CN was heated to 70°C for 20 h. The reaction mixture was filtered and the filter rinsed with several portions
of CH₂Cl₂. The filtrate was poured into a separatory funnel and 200 mL 1M NaOH was added. The layers were partitioned and the aqueous phase extracted with 3x100 mL CH₂Cl₂. The combined organic phases were dried with K₂CO₃ and concentrated under vacuum. The resulting solids were recrystallized from 2-PrOH to yield 29.5g (75%) of 16 as a white powder.

**1H NMR (CDCl₃)** δ 7.42-7.15 (m, 10H), 6.82-6.62 (m, 3H), 4.44 (m, 1H), 4.7-3.92 (m, 1H), 3.80 (s, 3H), 3.73 (s, 4H), 3.26 (m, 1H), 3.06-2.84 (m, 2H);

**13C NMR (CDCl₃)** δ 148.2, 143.6, 139.7, 128.3, 128.2, 126.9, 122.4, 121.8, 120.1, 108.8, 67.6, 55.6, 54.3, 51.1, 27.3.

**Resolution of 8-methoxy-3-(dibenzylamino)chroman (16).** A solution of 16 g (42.5 mmol) of L-dibenzoyl tartaric acid was prepared by heating in a mixture of 400 mL of 1,2-dichloroethane/CHCl₃ (9:1) and 400 mL H₂O until a solution was obtained then poured into a 2 L Erlenmeyer flask, containing a solution of 16 (30.5 g (85.1 mmol) in 400 ml of 1,2-dichloroethane/CHCl₃ (9:1) (this solvent system is from here referred to as “solvent”). The resulting mixture was heated with stirring until a solution was obtained. The heating mantle was turned off and the solution allowed to cool to r.t. Stirring was maintained throughout. No crystals had been formed after 18h and the flask was put in the refrigerator. After 1h the flask was taken out from the refrigerator and stirred at r.t for ½ h. The flask was then put in an ice bath for 1h, followed by vigorously stirring at r.t for 1h. The Erlenmeyer flask was once more left in the refrigerator, whereas a white powder started to precipitate. The process was allowed to continue for 2h. After this period of time the crystals of the 16•L-dibenzoyl salt were filtered off. The salt was dissolved in 1M aqueous NaOH and extracted with CH₂Cl₂ (3x150 mL). The combined extracts were dried over K₂CO₃ and evaporated to yield 12.9 g (42%) of (R)-16 with a %ee value of 86.5 see Step A for further resolution. The phases in the mother liquor was allowed to separate. The water phase was extracted with CH₂Cl₂ (2x200 mL). The pooled organic phases were dried over K₂CO₃. The volatiles were evaporated providing of (S)-16 (16.2 g, 53%) with a %ee value of 68 see Step B.

**Step A**

The 12.9 g (36 mmol) of (R)-16, from the crystals in the first resolution, dissolved in 169 mL solvent was subjected to another resolution with 10.8g (29 mmol) of L-dibenzoyl tartaric acid, 169 mL solvent and 169 mL H₂O, using the procedure in the introduction. However, after 1h of stirring at r.t. the flask was put in the refrigerator, providing crystals within 2h. The work up procedure was performed as in the introduction for crystals, yielding (R)-16 (10 g, 78%) with a %ee value of 96, see Step I for further resolution of (R)-16 derived from the L-dibenzoyl tartaric salt. The mother liquor was treated as above providing 2.5 g (20%) (R)-16 with a %ee value of 35, see Step C for further resolution.
Step B
A solution of (S)-16 (16.2 g, 45 mmol), from the crystals in the first resolution, in 225 mL solvent was subjected to another resolution with D-dibenzoyl tartaric acid (8.5 g, 29 mmol) in 225 mL solvent and 225 mL H₂O. Vigorous stirring was demanded for precipitation to occur, otherwise the procedure in step A. Work up of the crystals yielded (S)-16 (7.9 g, 49%) with a %ee value of 96, see Step F for further resolution of (S)-16 derived from the D-dibenzoyl tartaric salt. Work up of the mother liquor provided (S)-16 (7.8 g, 48%) with a %ee value of 39, see Step D for further resolution.

Step C
A solution of (R)-16 (2.6 g, 7.2 mmol), from the mother liquor in step A, in 35 mL solvent was subjected to another resolution with 1.3 g (3.6 mmol) of L-dibenzoyl tartaric acid 35 mL solvent and 35 mL H₂O, using the procedure in Step A. Work up of the crystals yielded (R)-16 (1.1 g, 44%) with a %ee value of 91, see Step I for further resolution of (R)-16 derived from the L-dibenzoyl tartaric salt. Work up of the mother liquor provided (S)-16 (1.4 g, 53%) with a %ee value of 14, see Step E for further resolution of amine (S)-16 from the mother liquor.

Step D
The 7.8 g (22 mmol) of (S)-16, from the mother liquor in step B, in 110 mL solvent was subjected to another resolution with 4.1 g (11 mmol) of D-dibenzoyl tartaric acid 110 mL solvent and 110 mL H₂O, using the procedure in Step A. Work up of the crystals yielded 3.4 g (44%) (S)-16 with a %ee value of 85, see Step F for further resolution of (S)-16 derived from the D-dibenzoyl tartaric salt. Work up of the mother liquor provided 3.8 g (49%) (R)-16 with a %ee value of 3, see Step E for further resolution of amine (R)-16 from the mother liquor.

Step E
The isolated amines from the mother liquors in step C and D, (S)-16 and (R)-16 (5.1 g, 14 mmol) was dissolved in 71 mL solvent and subjected to another resolution with L-dibenzoyl tartaric acid (4.1 g, 11 mmol) of 71 mL solvent and 71 mL H₂O, using the procedure in Step A. Work up of the crystals yielded (R)-16 (2 g, 40%) with a %ee value of 80, see Step G for further resolution of (R)-16 derived from the L-dibenzoyl tartaric salt. Work up of the mother liquor provided (S)-16 (2.8 g, 64%) with a %ee value of 64, see Step N for further resolution of amine (R)-16 from the mother liquor.

Step F
The isolated (S)-16 (11.4 g, 32 mmol), derived from the D-dibenzoyl tartaric salt in step B and D, was dissolved in 120 mL solvent and subjected to another resolution with D-dibenzoyl tartaric acid (8.1 g, 22 mmol) in 120 mL
solvent and 120 mL H₂O. Work up of the crystals yielded (S)-16 (8.3 g, 72%) with a %ee value of 96, see Step J for further resolution of (S)-16 derived from the D-dibenzoyl tartaric salt. Work up of the mother liquor provided (S)-16 (3 g, 27%) with a %ee value of 74, see Step H for further resolution of amine (S)-16 from the mother liquor.

Step G
The isolated (R)-16 (2 g, 6 mmol) from the L-dibenzoyl salt in step E, in 30 mL solvent was subjected to another resolution with L-dibenzoyl tartaric acid (1.7 g, 4.6 mmol) in 30 mL solvent and 30 mL H₂O, using the procedure in Step A. Work up of the crystals yielded (R)-16 (1.5 g, 72%) with a %ee value of 96, see Step I for further resolution of (R)-16 derived from the L-dibenzoyl tartaric salt. Work up of the mother liquor provided (R)-16 (0.5 g, 22%) (with a %ee value of 35, see Step N for further resolution of amine (R)-16 from the mother liquor.

Step H
The isolated (S)-16 (3 g, 8.5 mmol), derived the D-dibenzoyl tartaric salt in step F, in 42 mL solvent was subjected to another resolution with D-dibenzoyl tartaric acid (2.2 g, 5.9 mmol) in 42 mL solvent and 42 mL H₂O, using the procedure in Step A. Work up of the crystals yielded (S)-16 (2 g, 66%) with a %ee value of 98, see Step J for further resolution of (S)-16 derived from the D-dibenzoyl tartaric salt. Work up of the mother liquor provided (S)-16 (1 g, 34%) with a %ee value of 33, see Step N for further resolution of amine (S)-16 from the mother liquor.

Step I
The isolated (R)-16 (12 g, 35 mmol) from the L-dibenzoyl salt in step A, C and G in 175 mL solvent was subjected to another resolution with L-dibenzoyl tartaric acid (12 g, 31 mmol) of in 175 mL solvent and 175 mL H₂O, using the procedure in Step A. Work up of the crystals yielded (R)-16 (11 g, 87%) with a %ee value of 99, see Step K for further resolution of (R)-16 derived from the L-dibenzoyl tartaric salt. Work up of the mother liquor provided (R)-16 (1.4 g, 11%) with a %ee value of 70, see Step N for further resolution of amine (R)-16 from the mother liquor.

Step J
The isolated (S)-16 (9.4 g, 27 mmol), from the D-dibenzoyl tartaric salt in step F and H, in 135 mL solvent was subjected to another resolution with of D-dibenzoyl tartaric acid (9 g, 24 mmol) in 135 mL solvent and 135 mL H₂O. Work up of the crystals yielded 8.2 g (84%) (S)-16 with a %ee value of >99. Work up of the mother liquor provided 1.4 g (15%) (S)-16 with a %ee value of 83, see Step M for further resolution of amine (S)-16 from the mother liquor.
Step K
The isolated \((R)-16\) (11 g, 30 mmol) of derived from the L-dibenzoyl salt in step I in 150 mL solvent was subjected to another resolution with L-dibenzoyl tartaric acid (11 g, 29 mmol) in 150 mL solvent and 150 mL H₂O. Work up of the crystals yielded 9.9 g (91%) \((R)-16\) with a %ee value of >99. Work up of the mother liquor provided 700 mg (6%) \((R)-16\) with a %ee value of 89, see Step L for further resolution of amine \((R)-16\) from the mother liquor.

Step L
The isolated \((R)-16\) (2.1 g, 5.8 mmol) of derived from the mother liquors in step I and K in 30 mL solvent was subjected to another resolution with 1.5 g (4.1 mmol) of L-dibenzoyl tartaric acid in 150 mL solvent and 150 mL H₂O. Work up of the crystals yielded \((R)-16\) (1.4 g, 64%) with a %ee value of 96, see Step M for further resolution of \((R)-16\) derived from the L-dibenzoyl tartaric salt. Work up of the mother liquor provided \((R)-16\) (600 mg, 30%) with a %ee value of 29, see Step N for further resolution of amine \((R)-16\) from the mother liquor.

Step M
The isolated \((R)-16\) 1.4 g (3.8 mmol) derived from the L-dibenzoyl salt in step L in 20 mL solvent was subjected to another resolution with L-dibenzoyl tartaric acid (0.9 g, 3.4 mmol) in 20 mL solvent and 20 mL H₂O. Work up of the crystals yielded \((R)-16\) (1.2 g, 89%) with a %ee value of 99, see Step O for further resolution of \((R)-16\) derived from the L-dibenzoyl tartaric salt. Work up of the mother liquor provided \((R)-16\) (97 mg, 7%) with a %ee value of 29, see Step N for further resolution of amine \((R)-16\) from the mother liquor.

Step N
The isolated \((S)-16\) 5.9 g (16 mmol) derived from the mother liquors in step E, G, H, J, L and M in 83 mL solvent was subjected to another resolution with D-dibenzoyl tartaric acid (3 g, 3.1 mmol) in 83 mL solvent and 83 mL H₂O. Work up of the crystals yielded \((S)-16\) (2.8 g, 47%) with a %ee value of 94, see Step P for further resolution of \((S)-16\) derived from the D-dibenzoyl tartaric salt. Work up of the mother liquor provided \((R)-16\) (2.9 g, 49%) with a %ee value of 3, see Step R for further resolution of amine \((R)-16\) from the mother liquor.

Step O
The isolated \((R)-16\) (1.2g, 3.38 mmol) of derived from the L-dibenzoyl salt in step M in 14 mL solvent was subjected to another resolution with L-dibenzoyl tartaric acid (0.9 g, 2.5 mmol) in 17 mL solvent and 17 mL H₂O.
Work up of the crystals yielded (R)-16 (820 mg, 83%) with a %ee value of >99. Work up of the mother liquor provided (R)-16 (120 mg, 7%) with a %ee value of 83, see Step R for further resolution of amine (R)-16 from the mother liquor.

**Step P**
The isolated (S)-16 (2.8 g, 7.8 mmol) of derived from the D-dibenzoyl salt in step N in 40 mL solvent was subjected to another resolution with 2.6 g (7.05 mmol) of D-dibenzoyl tartaric acid in 40 mL solvent and 40 mL H₂O. Work up of the crystals yielded (S)-16 (2.4 g, 85%) with a %ee value of 99, see Step R for further resolution of (S)-16 derived from the D-dibenzoyl tartaric salt. Work up of the mother liquor provided (R)-16 (340 mg, 12%) with a %ee value of 59.

**Step Q**
The isolated (S)-16 (2.4 g, 6.7 mmol) from the D-dibenzoyl salt in step P in 33 mL solvent was subjected to another resolution with D-dibenzoyl tartaric acid (2.3 g, 6.3 mmol) in 33 mL solvent and 33 mL H₂O. Work up of the crystals yielded (S)-16 (2.2 g, 93%) with a %ee value of >99. Work up of the mother liquor provided (S)-16 (170 mg, 6%) with a %ee value of 86.

**Step R**
The isolated (R)-16 (2.9 g, 8.3 mmol) derived from the mother liquors in step N and O in 41 mL solvent was subjected to another resolution with 1.5 g (4.2 mmol) of L-dibenzoyl tartaric acid in 41 mL solvent and 41 mL H₂O. Work up of the crystals yielded (R)-16 (1.2 g, 42%) with a %ee value of 93, see Step S for further resolution of (R)-16 derived from the L-dibenzoyl tartaric salt. Work up of the mother liquor provided (S)-16 (1 g, 38%) with a %ee value of 59.

**Step S**
The isolated (R)-16 (1.2 g, 3.4 mmol) derived from the L-dibenzoyl salt in step R in 17 mL solvent was subjected to another resolution with L-dibenzoyl tartaric acid (1.1 g, 3.1 mmol) in 17 mL solvent and 17 mL H₂O. Work up of the crystals yielded (R)-16 (1 g, 84%) with a %ee value of 98, see Step T for further resolution of (R)-16 derived from the L-dibenzoyl tartaric salt. Work up of the mother liquor provided (S)-16 (120 mg, 10%) with a %ee value of 41.
The isolated (R)-16 (1g, 2.9 mmol) derived from the L-dibenzoyl salt in step S in 15 mL solvent was subjected to another resolution with L-dibenzoyl tartaric acid (990 mg, 2.6 mmol) in 15 mL solvent and 15 mL H2O. Work up of the crystals yielded (R)-16 (870 mg, 83%) with a %ee value of >99. Work up of the mother liquor provided (S)-16 (120 mg, 12%) with a %ee value of 85. The total yield was for (R)-16 11.5 g (38%), with a %ee value of >99; [α]D18 = -76.4 (c=1, CH2Cl2). The total yield was for (S)-16 10.5 g (35%), with a %ee value of >99; [α]D18 = -76.2 (c=1, CH2Cl2). The yield for collected and pooled rests was 2.6 g (9%) containing (S)-16 with a %ee value of 46%.

(R)-8-Methoxy-3-chromanamine (R)-11. A mixture of 5.3 g (14.8 mmol) of (R)-16, 3.7g (59 mmol) of NH4HCOO and 1.1 g Palladium, 10wt% on activated carbon, in 50 mL 2-PrOH, was refluxed for 18 h. The reaction mixture was filtered through Celite and the filter cake rinsed with several portions of 2-PrOH and CH2Cl2. The filtrate was concentrated to yield 2.6 g (97%) of (R)-11.

1H NMR (CDCl3) δ 6.63-6.56 (m, 1H), 6.51-6.47 (m, 1H), 6.45-6.42 (m, 1H), 4.00-3.95 (m, 1H), 3.64-3.57 (m, 4H), 3.12-3.04 (m, 1H), 2.81-2.74 (m, 1H), 2.32 (dd, 1H); 13C NMR (CDCl3) δ 148.1, 143.2, 122.0, 121.1, 120.2, 109.2, 71.7, 55.7, 43.9, 34.3.

(R)-8-Methoxy-3-(dipropylamino)chroman (17a). To a solution of 321 mg (1.8 mmol) of (R)-11, 1mL (14.3 mmol) of propionaldehyde and 60 mg of Montmorillonite K-10 in 10 mL MeOH was 900 mg (14.3 mmol) NaCNBH3 added portionwise. The reaction was run for 22 h at room temperature. The reaction was quenched with 40 mL 1 M NaOH and extracted with 3x10 mL CH2Cl2. The organic phases were dried over K2CO3 and evaporated. Flash chromatography [SiO₂, CHCl3/MeOH (98:2)] afforded 350 mg (74%) of 17a.

1H NMR (CDCl3) δ 6.85-6.76 (m, 1H), 6.74-6.62 (m, 2H), 4.47-4.39 (m, 1H), 3.90-3.78 (m, 2H)3.25-3.10 (m, 1H), 2.93-2.77 (m, 2H), 2.60-2.43 (m, 4H), 1.45 (sext, 4H), 0.92-0.80 (m, 6H).

(R)-8-Methoxy-3-(dimethylamino)chroman (17b). 2.8 g (44 mmol) NaCNBH3 was added to a solution of 1.3 g (7.3 g) of (R)-11 and 3.3 mL of 37% aqueous formaldehyde in 40 mL MeOH at 0°C. The resulting mixture was stirred for 1 h, then 330 mg of Montmorillonite K-10 was added. Stirring at 0°C was continued for 1 h. The ice bath was removed and stirring was continued overnight at room temperature. The reaction mixture was filtered through Celite and the filtercake rinsed with several portions of MeOH. The
volatiles were evaporated and the residue partitioned between 30 mL CH₂Cl₂ and 30 mL 1 M NaOH. The aqueous phase was additionally extracted with 2×30 mL CH₂Cl₂. The pooled extracts were washed with 30 mL brine, dried over K₂CO₃ and concentrated to yield (98%) 17b.

**1H NMR (CDCl₃)** δ 7.27-7.19 (m, 1H), 7.13-7.03 (m, 2H), 4.91-4.80 (m, 1H), 4.41-4.19 (m, 4H), 3.39-3.03 (m, 3H), 2.79 (s, 1H); **13C NMR (CDCl₃)** δ 147.3, 142.8, 121.1, 120.8, 119.4, 108.3, 67.3, 66.4, 56.5, 54.8, 53.4, 41.7, 28.2.

**1H NMR (CDCl₃)** δ 7.27-7.19 (m, 1H), 7.13-7.03 (m, 2H), 4.91-4.80 (m, 1H), 4.41-4.19 (m, 4H), 3.39-3.03 (m, 3H), 2.79 (s, 1H); **13C NMR (CDCl₃)** δ 147.3, 142.8, 121.1, 120.8, 119.4, 108.3, 67.3, 66.4, 56.5, 54.8, 53.4, 41.7, 28.2.

**(R)**-8-Hydroxy-3-(dipropylamino)chroman (18a). To a solution of 200 mg (0.8 mmol) of 17a in 2 mL dry CH₂Cl₂, under an inert N₂ atmosphere, at –20°C was 800 μL (7.6 mmol) of BF₃·Me₂S added dropwise. The reaction mixture was allowed to warm to room temperature and stirred for 22 h. The reaction was quenched with 5 mL H₂O and the aqueous phase made basic (pH~10) with NH₄OH. The basic aqueous phase was extracted with 3×5 mL CH₂Cl₂. The extracts were dried over Na₂SO₄ and evaporated to yield 118 mg (61%) of 18a.

**1H NMR (CDCl₃)** δ 6.78-6.69 (m, 1H), 6.63-6.56 (m, 2H), 4.41-4.30 (m, 1H), 3.89-3.78 (m, 1H), 3.27-3.14 (m, 1H), 2.89-2.79 (m, 2H), 2.59-2.41 (m, 4H), 1.45 (sext, 4H), 0.88 (t, 6H); **13C NMR (CDCl₃)** δ 144.7, 141.5, 122.0, 120.8, 120.6, 112.3, 68.25, 53.4, 52.6, 27.5, 21.6, 11.6.

**(R)**-8-Hydroxy-3-(dimethylamino)chroman (18b). To a solution of 734 mg (3.5 mmol) of 17b in 10 mL dry CH₂Cl₂, under N₂ atmosphere, at –10°C was 3 mL (28 mmol) of BF₃·Me₂S added dropwise. After the addition was complete the reaction was stirred at room temperature for 22 h. The reaction was quenched with H₂O, made basic (pH~10) with NH₄OH and extracted with 3×15 mL CH₂Cl₂. The organic phases were dried over Na₂SO₄ and evaporated to afford (95%) of 18b.

**1H NMR (CD₃OD)** δ 6.61-6.52 (m, 3H), 4.42-4.34 (m, 1H), 3.99-3.91 (m, 1H), 3.03-2.71 (m, 3H), 2.37 (s, 6H); **13C NMR (CD₃OD)** δ 146.6, 143.7, 122.5, 122.0, 121.7, 114.5, 68.3, 59.2, 42.8, 29.4.

**(R)**-3-(Dipropylamino)-8-trifluoromethanesulfonyloxychroman (19a). To a solution of 314 mg (1.3 mmol) of 18a and 263 μL (1.9 mmol) of Et₃N at -78°C in 20 mL CH₂Cl₂ was added dropwise 390 mg (1.4 mmol) of triflic anhydride. The reaction mixture was kept under N₂ atmosphere and stirred for 16 h, then quenched with 10 mL 1 M NaOH. The resulting mixture was poured onto Hydromatrix and the product eluted with 40 mL CH₂Cl₂ to afford 293 mg (59%) of 19a.

**1H NMR (CDCl₃)** δ 7.11.695 (m, 1H), 6.88-6.79 (m, 2H), 4.44-4.38 (m, 1H), 3.92-3.85 (m, 1H), 3.29-3.11 (m, 1H), 2.91-2.85 (m, 2H), 2.58-2.42 (m,
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(R)-3-(Dimethylamino)-8-trifluoromethanesulfonyloxychroman (19b).
To a solution of 307 mg (1.6 mmol) of 18b and 664 μL (4.7 mmol) of Et3N at -78°C in 20 mL CH2Cl2 was added dropwise 493 mg (1.7 mmol) of triflic anhydride cooled to -18°C. The mixture was kept under N2 atmosphere and stirred for 2h. The volatiles were evaporated and the residue taken up in 20 mL CH2Cl2. The organic phase was washed with 2x10 mL brine, dried over K2CO3 and concentrated. The oil was further purified by filtering through Al2O3 with Et2O as eluent to yield 315 mg (61%) of 19b.

1H NMR (CDCl3) δ 7.11-6.93 (m, 2H), 6.89-6.79 (m, 2H), 4.49-4.35 (m, 1H), 4.04-3.89 (m, 1H), 3.05-2.69 (m, 3H), 2.39 (s, 6H); 13C NMR (CDCl3) δ 146.3, 137.7, 129.6, 124.0, 120.2, 120.1, 118.5 (q), 68.0, 57.6, 42.4, 28.7.

(R)-8-(2,6-Dimethoxyphenyl)-3-(dipropylamino)chroman (20a).
(R)-3-Dipropylamino)-8-(trifluoromethanesulfonyloxy)chroman (19a) (50 mg, 0.13mmol), tetrakis(triphenylphosphine)palladium (15 mg, 13 μmol), Ba(OH)2×8H2O (165 mg, 0.52 mmol) and (2,6-dimethoxyphenyl)boronic acid (95 mg, 0.52 mmol) in 3 mL of degassed DME/H2O (9:1) were mixed in a heavy walled Pyrex tube under N2 followed by irradiation in a microwave cavity (Smith synthesizer) to 140°C for 20 min. After cooling, the reaction mixture was diluted with water and poured onto a reservoir containing Hydromatrix and eluted with several portions of CH2Cl2. The volatiles were evaporated and the residue was purified twice by flash column chromatography [silica gel, CHCl3/MeOH (98:2) and silica gel, EtOAc/i-Hex (7:3) followed by EtOAc] followed by kugelrohr distillation (165 °C at 7 mbar) to yield 20a (31.1 mg, 65%), which was converted into the corresponding oxide 20a×C2H2O4. mp 70-75 °C; 1H NMR (CD3OD) δ 0.99 (t, 6 H), 1.76 (sext, 4H), 3.52-3.38 (m, 3H), 3.52-3.38 (m, 1H), 4.02-3.89 (m, 1H), 4.24 (dd, 1H), 4.42-4.36 (m, 1H), 6.70-6.67 (m, 2H), 7.01-6.90 (m, 2H), 7.17-7.14 (m, 1H), δ 7.28 (t, 1H); 13C NMR (CD3OD) δ 28.8, 52.6, 53.3, 55.9, 67.9, 104.1, 104.2, 115.9, 119.8, 121.8, 122.3, 128.6, 129.4, 129.8, 152.4, 157.9, 164.7. Anal. (C23H31NO3×C2H2O4×1.5H2O) C, H, N.

(R)-8-(2,6-Dimethoxyphenyl)-3-(dimethylamino)chroman (20b).
(R)-3-(Dimethylamino)-8-(trifluoromethanesulfonyloxy)chroman (19b) (100 mg, 0.3 mmol), Ba(OH)2×8H2O (300 mg, 1.2 mmol), tetrakis(triphenylphosphine)palladium (71 mg, 61 μmol) and 2,6-(dimethoxyphenyl)boronic acid (223 mg, 1.2 mmol) were mixed with degassed DME/H2O (9:1) (3 mL) in a heavy walled Pyrex tube under N2. The mixture was irradiated in a microwave cavity (Smith synthesizer) to 135 °C for 20 min and then filtered through basic Al2O3 [CHCl3/MeOH (95:5)]. The filtrate was concentrated.
and the residue was dissolved in 20 mL Et₂O and 30 mL HCl (1M). The mixture was stirred for 20 min, the phases were separated and the acidic aqueous phase was further washed with Et₂O (2x20 mL). The aqueous phase was alkalinized with solid NaOH to pH~12 and extracted with Et₂O 3x20 mL. The extracts were dried (K₂CO₃), filtered and concentrated. The resulting oil was purified by flash chromatography [SiO₂, CHCl₃/MeOH (95:5)] followed by kugelrohr distillation (165 °C at 7 mbar), to yield the amine (53.7 mg, 57%) which was converted to the corresponding oxalate $\text{C}_2\text{H}_2\text{O}_4$. $\text{C}_2\text{H}_2\text{O}_4$: mp 62 °C; $[\alpha]_D^{15} = +6.7^\circ$ (c=1, MeOH); $^1$H NMR (CD₃OD) $\delta$ 2.90 (s, 6H), 3.24-3.20 (m, 1H), 3.50-3.38 (m, 1H), 3.65 (s, 6H), 3.83-3.78 (m, 1H), 4.19 (dd, 1H), 4.44 (ddd, 1H), 6.68-6.65 (m, 2H), 6.98-6.93 (m, 2H), 7.14-7.10 (m, 1H), 7.29 (t, 1H); $^{13}$C NMR (CD₃OD) $\delta$ 25.2, 40.5, 54.9, 55.0, 58.6, 63.7, 103.8, 104.0, 115.3, 119.0, 121.0, 123.7, 128.4, 128.9, 130.8, 151.8, 157.9, 164.7. Anal. (C₁₉H₂₃NO₃×C₂H₂O₄×H₂O) C, H, N.

10.2. Supplementary Information Paper II

Pär Holmberg and Lars Tedenborg.

**General Methods.** $^1$H NMR and $^{13}$C NMR SPECTRA were recorded at ambient temperature on a JEOL JNM-EX270 (270 MHz $^1$H, 67.8 MHz $^{13}$C) spectrometer. Chemical shifts are given in ppm referenced internally to CD₃OD (residual) solvent peaks at $\delta$ 3.31 ppm ($^1$H) and $\delta$ 49.18 ppm ($^{13}$C). Coupling constants are given in Hz. Optical rotations were recorded with a Perkin-Elmer 241 polarimeter. Melting points were obtained with open glass capillary tubes in an Electro-thermal melting point apparatus and are uncorrected. Elemental analyses were performed by Mikro Kemi AB, Uppsala. The values were within ±0.4% of theoretical unless indicated otherwise. Microwave heating was carried out with a MicroWell 10 single-mode microwave cavity operating at 2450 MHz from Labwell AB, Uppsala. The reactions were performed in heavy-walled Pyrex tubes with a special screw-cap allowing the release of pressure if overpressurization occurred.

**Formation and recrystallization of the D-tartaric salt of (S)-3-amino-5-methoxycroman (22)-tartrate.** To a well stirred solution of NaOH (47.7 g, 1.19 mol) in 600 mL H₂O was added the D-tartaric salt of (S)-3-amino-5-methoxycroman (196.2 g, 596 mmol). The resulting solution was extracted with Et₂O (3x150 mL) and dried over K₂CO₃. The volatiles were evaporated under vacuum to yield the free base (100.7 g, 562 mmol, 94%).
D-(-)-Tartaric acid (84.4g, 562 mmol) was added to the oil, followed by H₂O (2.8 L) under stirring. The mixture was heated until a solution was obtained. The heating mantle was turned off and the crystals allowed to form, under gentle stirring, overnight. The formed crystals were filtered off and washed with some Et₂O. The crystals were put in a desicator under vacuum to remove any remaining solvents, to give the (-)-tartaric salt of (S)-3-amino-5-methoxychroman (22·tartrate (117.8g, 356 mmol, 63%). For analytical purposes a small amount was converted to the hydrochloride salt (22·HCl).

\([\alpha]_D^{23} = -67.1^\circ (c 1, \text{CH}_3\text{OH})\); \(^1\)H NMR (CD₃OD) \(\delta 7.13 (t, J = 8.2, 1H), 6.62-6.49 (m, 2H), 4.24 (ddd, J=11.9, 4.3, 1.9, 1H), 4.15 (dd, J=11.9, 2.0), 1H), 3.87, 3.83(s, 3H), 3.08 (dd, J=18.1, 5.9, 1H), 2.81 (m, 1H); \(^1^3\)C NMR (CD₃OD) \(\delta 159.9, 155.9, 129.3, 110.8, 108.0, 104.4, 66.7, 56.1, 45.5, 24.8\); mp 245-250°C. Anal. C₁₀H₁₃NO₂·HCl C, H, N.

\((S)-3-(Dimethylamino)-5-methoxychroman (23).\) To a solution of the free amine of (22) (1.86g, 10.4 mmol) in 20 mL methanol was added 10 mL 37% aq. Formaldehyde. The resulting mixture was stirred for 15 min and then cooled to 0°C, NaBH₃CN <82.00g, 31.2 mmol) was added. The temperature was allowed to slowly rise to room temperature overnight. The solvent was evaporated under vacuum. To the remaining residue was added 47 mL aq 2M NaOH. The resulting suspension was extracted with 3x30 mL Et₂O. The combined organic phases were dried over K₂CO₃ and concentrated. The obtained oil was further purified by flash chromatography on silica gel, using CHCl₃ as eluent to yield (S)-3-(dimethylamino)-5-methoxychroman (23) (1.86g, 10.4 mmol, 93%) which was converted to the hydrochloride salt.

\([\alpha]_D^{23} = -55.3^\circ (c 1, \text{CH}_3\text{OH})\); \(^1\)H NMR (CD₃OD) \(\delta 7.02 (t, J = 8.2 \text{ Hz}, 1H), 6.51 (d, J = 8.2 \text{ Hz}, 1H), 6.43 (d, J = 8.2 \text{ Hz}, 1H), 4.44 (ddd, J = 12.8 \text{ Hz}, 4.3 \text{ Hz}, 2.0 \text{ Hz}, 1H), 4.16 (dd, J = 12.8 \text{ Hz}, 2.0), 1H), 3.75 (s, 3H), 3.78-3.69,(m, 1H), 3.04-3.00 (m, 2H), 2.88 (s, 6H); \(^1^3\)C NMR (CD₃OD) \(\delta 159.6, 156.0, 129.5, 110.7, 108.0, 104.6, 64.7, 59.9, 56.3, 42.3, 42.0, 22.3\); mp 255-258°C. Anal. C₁₂H₁₇NO₂·HCl C, H, N.

\((S)-3-(Dimethylamino)-5-trifluoromethanesulfonyloxychroman (24).\) The hydrochloride salt of (22) (8.8g, 36.1 mmol) was dissolved in 360 mL 47% HBr under N₂ and the resulting solution refluxed for 2.5h. The HBr and water were evaporated under reduced pressure. 150 mL water was added to the residue and the pH raised to 9 using aqueous NH₃. The aqueous phase was extracted with 3x150 mL CH₂Cl₂. The combined extracts were dried over Na₂SO₄ and concentrated to yield (S)-3-(dimethylamino)-5-hydroxychroman (5.86g, 30.3 mmol, 84%). A small amount was converted to the hydrochloride salt for analytical purposes.

\([\alpha]_D^{23} = -61.0^\circ (c 1, \text{CH}_3\text{OH})\); \(^1\)H NMR (CD₃OD) \(\delta 7.02-6.94 (m, 1H), 6.45 \text{(dd, J = 8.4 Hz, 1.2 Hz, 1H), 6.40 \text{(d, J = 8.2 Hz, 1H), 4.53 \text{(ddd, J = 12.7 Hz, 4.3 Hz, 1.6 Hz, 1H), 4.25 \text{(dd, J = 12.7 Hz, 2.3 Hz, 1H), 3.84-3.78 (m,}}

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1H), 3.16-3.10 (m, 2H), 3.00 (s, 3H), 2.97 (s, 3H); $^{13}$C NMR (CD$_3$OD) δ 157.7, 156.4, 129.2, 109.0, 108.9, 106.8, 64.8, 60.2, 42.2, 22.5. One carbon signal is missing due to overlapping signals; mp 283-285°C. Anal. C$_{11}$H$_{15}$NO$_2$·HCl C, H, N.

To a stirred solution of (S)-3-(dimethylamino)-5-hydroxychroman (3.00 g, 15.5 mmol) and Et$_3$N (3.14 g, 31 mmol), under N$_2$ atmosphere, in 70 mL CH$_2$Cl$_2$ at −40°C, was trifluoromethanesulfonic anhydride (4.82 g, 17.1 mmol) slowly added. The temperature of the mixture was allowed to rise to −10°C over 24 h. The reaction was quenched with water and extracted with 2×50 mL Et$_2$O. The organic phases were washed with brine, dried over K$_2$CO$_3$ and concentrated, to yield (S)-3-(dimethylamino)-5-trifluoromethanesulfonyloxychroman (24), 4.68 g, 14.4 mmol, 93%. A small amount was converted to the hydrochloride salt for analytical purposes.

$[\alpha]_D^{23}= -44.6^\circ$ (c 1, CH$_3$OH); $^1$H NMR (CD$_3$OD) δ 7.42-7.34 (m, 1H), 7.07 (d, $J = 8.2$ Hz, 1H), 7.06 (d, $J = 8.2$ Hz, 1H), 4.69 (ddd, $J = 13.2$ Hz, 4.3 Hz, 2.3 Hz, 1H), 4.43-4.35 (m, 1H), 3.97-3.90 (m, 1H), 3.44, (ddd, $J = 156.9$, 149.4, 130.4, 120.1 (q, $J^{13}$C-$^{19}$F) = 318 Hz), 118.8, 115.9, 113.7, 65.0, 58.8, 42.3, 22.8. One carbon signal is missing due to overlapping signals; mp 200-206°C. Anal. C$_{12}$H$_{14}$F$_3$NO$_4$S·HCl C, H, N.

(S)-3-(Dimethylamino)chroman (25). To a solution of (24) (908 mg, 2.8 mmol) in 50 mL DMF under an N$_2$ atmosphere, was Bu$_3$N added (2.07 g, 11.2 mmol) and HCO$_2$H (398 mg, 8.65 mmol). The solution was carefully degassed for 30 min by purging with N$_2$. DPPF (232 mg, 0.42 mmol) and (PPh$_3$)$_2$Pd Cl$_2$ (118 mg, 0.17 mmol) was added. The mixture was kept at 70°C for 4 h and then filtrated through a column with a short plug of Al$_2$O$_3$ with CHCl$_3$ as eluent. The volatiles were evaporated. The obtained crude product was diluted with H$_2$O, the pH raised to 11 with aqueous NH$_3$ and extracted with 3×100 mL Et$_2$O/pentane (1:1). The combined organic phases were dried with K$_2$CO$_3$ and concentrated. The residue was subjected to Kugelrohr distillation (1mm Hg, 65°C) to remove any remaining Bu$_3$N and DMF. The remaining oil was subjected to flash column chromatography with SiO$_2$ (a small amount of Al$_2$O$_3$ on top) using CHCl$_3$ as eluent. The oil was converted to the hydrochloride salt and recrystallized from MeOH/Et$_2$O to give (S)-3-(dimethylamino)chroman (25)·HCl (396 mg, 1.85 mmol, 60%).

$[\alpha]_D^{23}= -44.2^\circ$ (c 1, CH$_3$OH); $^1$H NMR (CD$_3$OD) δ 7.22-7.13 (m, 2H), 6.97 (m, 1H), 6.88 (d, $J = 7.9$ Hz, 1H), 4.58 (ddd, $J = 12.8$ Hz, 4.3 Hz, 2.3 Hz, 1H), 4.31 (dd, $J = 12.8$ Hz, 2.0 Hz, 1H), 3.86-3.77 (m, 1H), 3.40 (dd, $J = 18.1$ Hz, 5.9 Hz, 2H), 3.26-3.16 (m, 1H), 3.00 (s, 6H); $^{13}$C NMR (CD$_3$OD) δ 155.3, 131.2, 129.5, 123.4, 119.1, 118.2, 65.1, 60.3, 42.1, 27.1. One carbon is missing due to overlapping signals; mp 223-225°C. Anal. C$_{12}$H$_{14}$N·HCl C, H, N.
(S)-6-Bromo-3-(dimethylamino)chroman (26). To a well stirred solution of the hydrochloride salt of (S) (361 mg, 1.69 mmol) in 10 mL acetic acid was added bromine (278 mg, 1.74 mmol) dropwise. After 40 min of stirring a precipitate was formed. Et₂O was added to the reaction mixture and the liquors carefully decanted. This was repeated until there was no detectable smell of acetic acid from the solids. The remaining volatiles were evaporated. A slurry was made by mixing the solids with CHCl₃/MeOH (9:1), which was poured on to a column with basic Al₂O₃ and eluted with the above solvent mixture to give the free base of (S)-6-Bromo-3-(dimethylamino)chroman (26) (325 mg, 1.26 mmol, 75%). A small amount was converted to the hydrochloride salt for analytical purposes.

General procedure for the Suzuki Coupling of (S)-6-Bromo-3-(dimethylamino)chroman (6) with the corresponding Boronic acids to give target compounds 27a-27d.

To 1 mL of EtOH/H₂O/DME (37.5:25:62.5:) in a reaction tube were (26) (30 mg, 0.117 mmol), (PPh₃)₄Pd (6.8 mg, 5.9 µmol), Na₂CO₃ (24.8 mg, 0.234 mmol) and the appropriate boronic acid (0.467 mmol) added under a nitrogen atmosphere. The reaction tube was microwave irradiated for 4.3 min with a power of 45W. After cooling, ethanol and DME were evaporated from the mixture under reduced pressure. The above reaction was repeated two times and the reaction mixtures from the three reactions were combined. 15 mL H₂O was added and the pH raised to 12, using aqueous NH₃ and extracted with 3x15 mL CH₂Cl₂. The combined organic phases were dried with K₂CO₃ and evaporated under vacuum. Kugelrohr distillation (1mm Hg, 95°C) was performed to remove any reduced starting material ((S)-3-(Dimethylamino)chroman (5)) formed during the reaction. The crude product was purified by flash column chromatography on silica gel. All compounds were converted to the hydrochloride salt. For eluent system, yield, mp, [α]D₂⁰ and elemental analysis for each compound see below.

(S)-6-(2,6-Dimethoxyphenyl)-3-(dimethylamino)chroman (27a). Eluent system: CHCl₃/MeOH (98:2); Yield 76%; [α]D₂⁰ 71.9° (c 1, CH₃OH); 'H NMR (CD₂OD) δ 7.24 (t, J = 8.2 Hz, 1H), 7.06-6.97 (m, 2H), 6.85 (d, J = 8.2 Hz, 1H), 4.59 (m, 1H), 4.52 (d, J = 12.5 Hz, 1H), 3.83 (m, 1H), 3.65 (s, 6H), 3.46-3.15 (m, 2H), 2.99 (s, 6H); ¹³C NMR (CD₂OD) δ 159.15, 153.9, 133.6, 132.3, 130.0, 129.9, 120.2, 118.0, 117.3,
(S)-6-(2,6-Dimethylphenyl)-3-(dimethylamino)chroman (27b). Eluent system: CHCl₃/MeOH (99:1); Yield 57%; \([\alpha]_D^{23} +65.5^\circ\) (c 1, CH₃OH); \(^1\)H NMR (CD₃OD) δ 7.14-7.03 (m, 3H), 7.00-6.89 (m, 3H), 4.65 (ddd, J = 12.5 Hz, 4.6 Hz, 2.0 Hz, 1H), 4.41 (m, 1H), 3.90 (m, 1H), 3.43, (m, 1H), 3.28-3.21 (m, 1H), 3.25 (s, 6H), 2.01 (s, 3H), 2.00 (s, 3H); \(^13\)C NMR (CD₃OD) δ 154.1, 142.4, 137.2, 137.1, 136.5, 131.7, 130.3, 128.5, 128.3, 119.4, 118.3, 65.2, 42.2, 27.0, 21.2; mp 220-223°C. Anal. for C₁₉H₂₃NO·HCl·1/4 H₂O C, H, N.

(S)-6-Phenyl-3-(dimethylamino)chroman (27c). Eluent system: CHCl₃/MeOH (98:2); Yield 73%; \([\alpha]_D^{23} +93.5^\circ\) (c 1, CH₃OH); \(^1\)H NMR (CD₃OD) δ 7.60-7.54 (m, 2H), 7.48-7.24 (m, 5H), 7.00-6.91 (m, 1H), 4.62 (m, 1H), 4.36 (m, 1H), 3.85 (m, 1H), 3.45 (m, 1H), 3.31, (s, 6H); \(^13\)C NMR (CD₃OD) δ 154.8, 141.9, 137.2, 130.0, 129.6, 128.2, 127.8, 118.6, 65.3, 60.5, 42.1, 27.3. One carbon signal is missing due to overlapping signals; mp 192°C. Anal. for C₁₉H₂₃NO·HCl·1/4 H₂O C, H, N.

(S)-6-(2-Methoxy-1-naphthyl)-3-(dimethylamino)chroman (27d). Eluent system: CHCl₃/MeOH (98:2); Yield 73%; \([\alpha]_D^{23} +93.5^\circ\) (c 1, CH₃OH); \(^1\)H NMR (CD₃OD) δ 8.22-8.16 (m, 1H), 7.90-7.85 (m, 1H), 7.70 (d, J = 8.5 Hz, 1H), 7.59-7.45 (m, 5H), 7.05-7.00 (m, 1H), 4.69-4.60 (m, 1H), 4.45-4.37 (m, 1H), 3.92-3.85 (m, 1H), 3.58,(s, 3H), 3.57-3.49 (m, 1H), 3.33-3.28 (m, 1H), 3.06 (s, 3H), 3.03 (s, 3H); \(^13\)C NMR (CD₃OD) δ 154.6, 154.3, 135.9, 134.1, 132.1, 130.7, 130.2, 129.9, 129.4, 127.4, 125.3, 123.4, 119.1, 118.2, 65.3, 61.6, 60.3, 42.3, 27.2; mp 192°C. Anal. for C₁₉H₂₃NO·HCl·1/4 H₂O C, H, N.

(S)-6-(2,6-Dihydroxyphenyl)-3-(dimethylamino)chroman (27e). The hydrochloride salt of (S)-6-(2,6-dimethoxyphenyl)-3-(dimethylamino)chroman (27a) (94 mg, 0.269 mmol) was dissolved in 10 mL 47% HBr, under N₂ atmosphere, and refluxed for 2.5 h. The HBr was evaporated under reduced pressure. 20 mL water was added to the residue and the pH raised to 9 using aqueous NH₃. The aqueous phase was extracted with 3x10 mL CH₂Cl₂. The combined organic phases were dried over Na₂SO₄ and evaporated under vacuum. The obtained crude product was further purified by flash column chromatography on silica gel using CHCl₃/MeOH (95:5) as eluent to yield (S)-6-(2,6-Dihydroxyphenyl)-3-(dimethylamino)chroman (27e) (60 mg, 0.186 mmol, 69%), which was converted to the hydrochloride salt.

\([\alpha]_D^{23} +88.3^\circ\) (c 1, CH₃OH); \(^1\)H NMR (CD₃OD) δ 7.20-7.15 (m, 2H), 6.97-6.87 (m, 2H), 6.39 (m, 1H), 4.63 (ddd, J = 12.1 Hz, 5.4 Hz, 2.7 Hz, 1H),...
4.33 (m, 1H), 3.82 (m, 1H), 3.43, (m, 1H), 3.23 (m, 1H), 3.02 (s, 6H); $^{13}$C NMR (CD$_3$OD) δ 156.7, 153.9, 133.7, 132.5, 130.1, 129.4, 118.0, 117.4, 117.3, 65.2, 60.6, 48.2, 27.3. One carbon signal is missing due to overlapping signals; mp = (260-267)°C. Anal. C$_{17}$H$_{19}$NO$_3$·HCl·1/4 H$_2$O C, H, N

10.3. Supplementary Information Paper III

Pär Holmberg and Adolf Gogoll.

Experimental

$^1$H and $^{13}$C NMR spectra were recorded at ambient temperature on a JEOL JNM-EX 400 or a JEOL JNM-EX 270 spectrometer at 270.2 and 67.8 or 399.8 and 100.5 MHz, respectively. Chemical shifts are given in ppm from internal standards. For $^1$H NMR and $^{13}$C NMR spectra the internal reference was residual CHCl$_3$ and CDCl$_3$ (δ 7.26 or δ 77.0 ppm), or CD$_3$SOCD$_2$H and DMSO-d$_6$ (δ 2.49 or δ 39.5 ppm), respectively. Multiplet patterns are designated as follows: s, singlet, d, doublet, dd, doublet of doublets, t, triplet, q, quartet and m, multiplet. GC-MS was performed with a Varian Saturn 2100 massspectrometer connected to a Varian 3900 gas chromatograph equipped with a CP-Sil 8CB lowbleed/MS column (30m X 0.25mm) using a 70-300°C temperature gradient. Analysis of enantiomerial purity of the 1-(3-furyl)-3-buten-1-ol was performed with HPLC on a Chiracel OD-H (0.46cm X 25cm, 5µm) column with a Chiracel OD (0.46cm x 5cm, 10µm) gardcolumn using a Merck Hitachi L-6200 intelligent pump, a Merck Hitachi L-4000 UV-detector operated at 214 nm and a Spectra Physics SP4270 integrator. The mobile phase consisted of isohexane and isopropanol (99:1) and the flow rate was 0.83 mL/min. Analysis of enantiomeric purity of 1-(3-furyl)-3-buten-1-yl acetate were performed with GC on a Varian 3800 gas chromatograph equipped with a Supelco β-dex 120 column (30m x 0.25mm) and a flame-ionisation detector using isothermal conditions at 90°C, He as carrier gas and the flow rate was 1.1ml/min. The enzymatic kinetic resolution was monitored with $^1$H NMR (DMSO-d$_6$ solution) with 1,4-dimethoxybenzene as internal standard by comparison between the peaks at 7.52, 7.47 (ester) 7.40, 7.43 (alcohol) and the peaks at 6.82 and 3.6 (1,4-dimethoxybenzene). The peaks at 6.43 (ester) and 6.39 (alcohol) were also used when they appeared without overlap by other signals. Preparative chromatographic separations were achieved by using silica gel and monitored with thin layer chromatography, performed on silica gel (Merck) mounted on aluminium cards, monitored with UV (254 nm), and with GC-MS. An Orion 420A benchtop pH/ISE meter was used for pH measurement. The buffer solutions used in the enzymatic kinetic resolutions were prepared by mixing 100mL of
0.1M Na₂HPO₄ with 100 mL of 0.00064M NaH₂PO₄ and thereafter adjusting the pH to 7.0 by addition of 1.0M NaOH drop wise. The buffer solutions were stored under nitrogen atmosphere.

1-(3-Furyl)-3-buten-1-ol (37). Zink powder (1.1 g, 17.4 mmol) was added to a mixture of 1.4 g (14.5 mmol) 3-furaldehyd and 2.1 g (17.4 mmol) 3-bromo-propene (1.7 g, 12.5 mmol) in 20mL saturated aqueous NH₄Cl and 4mL THF. A slightly exothermic reaction followed and the mixture was stirred for an additional hour or until the reaction had cooled to r.t. The reaction mixture was extracted with ether (3x25 mL). The combined organic phases were dried over MgSO₄ and concentrated to give 2 g (99%) of 37 as an oil. Spectroscopic data were in accordance with the literature.¹

¹H-NMR (CDCl₃) δ 7.50-7.39 (m, 2H), 6.40 (s, 1H), 5.82-5.76 (m, 2H), 5.20-5.13 (m, 1H), 4.71 (m, 1H), 2.52-2.47 (m, 2H), 2.05 (s, 1H); ¹³C-NMR (CDCl₃) δ 143.5, 139.3, 134.3, 128.6, 118.8, 108.7, 66.3, 42.6. 37

1-(3-furyl)-3-buten-1-yl acetate (53). To a cooled solution (0°C) of 37 (2 g, 14.5 mmol) in 30ml dry dichloromethane was added Dimethylaminopyridin (0.2 g, 1.45 mmol) and acetic anhydride (2.9 mg, 28.9 mmol), followed by the dropwise addition of Et₃N (5.9 g, 57.9 mmol). The resulting mixture was stirred for 1.5h in r.t. The reaction was quenched with the addition of 40 mL 2M HCl and the phases were partitioned. The organic phase was washe with 40 mL NaHCO₃, followed by 40mL Brine. The volatiles were dried over MgSO₄ and concentrated. The resulting brown oil was eluted with diethylether through a pad of silica and concentrated to give 53 (2.4 g, 85%). Spectroscopic data were in accordance with literature.²

²H-NMR (CDCl₃) δ 7.42 (s, 1H), 7.37 (s, 1H), 6.39 (s, 1H), 5.66-5.84 (m, 2H), 5.03-5.13 (m, 2H), 2.51-2.66 (m, 2H), 2.04 (s, 3H); ¹³C-NMR (CDCl₃) δ 170.1, 143.1, 140.2, 133.1, 124.4, 118.0, 108.8, 67.6, 39.0, 20.9.

General method for the enzyme catalyzed hydrolysis of 37(analytical scale).To a mixture of 10mL of a phosphate buffer at pH 7.0 and 1mL DMSO was added 53 (100 mg, 0.55 mmol). Lipase from the appropriate enzyme (10 mg) was added and the mixture was stirred at 40°C in a heating block. Samples (500 µL) were taken twice a day and mixed with DMSO-d₆ (500 µL) and a known amount of 1,4-dimethoxybenzen (5-10 mg), then analysed with ¹H NMR.
Resolution of 37 in a 0.8 g scale.
To 66 mL phosphate buffer (pH7.0) and 6,6mL DMSO was added 53 (793 mg, 4.4 mmol). Lipase from *Pseudomonas cepacia* (73 mg) was added and the mixture was stirred at 40°C. Samples (500 µL) were taken twice a day and mixed with DMSO-d6 (500µl) and 1,4-dimethoxybenzen as the internal standard then analysed with ¹H NMR. The hydrolysis mixture was extracted with diethylether-pentane (7:3) (3x70mL)when the degree of conversion was 67%. The ester was separated from the alcohol by flash column chromatography (pentane-diethyl ether 9:1 v/v) to yield 245 mg (31%) of 53 (ee >98%) and 270 mg (44%) of 37 (ee = 75%,) [α]D²⁰ 20 (c 1.0, CH₂Cl₂), corresponding to (R)-37 according to ref 1.

The absolute configuration of 53 was determined hydrolysing the ester in 1M NaOH, then measuring the optical rotation on the corresponding alcohol; [α]D²⁰ -25 (c 1.0, CH₂Cl₂), corresponding to (S)-37 according to ref 1.

References


10.4. Supplementary Information Paper IV

Pär Holmberg and Adolf Gogoll.

Experimental
Melting points were determined on a Büchi SMP-20 apparatus. ¹H and ¹³C NMR spectra were recorded at ambient temperature on a Varian Unity 400, Varian Gemini 300 or Varian Unity 500 instrument. Chemical shifts are given in ppm from internal standards. For ¹H NMR and ¹³C NMR spectra the internal reference was residual CHCl₃ and CDCl₃ (δ 7.26 or δ 77.0 ppm). Coupling constants are given in Hertz, and the splitting patterns are designated as follows: s, singlet, d, doublet, dd, doublet of doublets, t, triplet, q, quartet, quint, quintett, hept, heptet, sext, sextet, oct, octett and app for apparent. NMR signals were assigned from gHMBC,¹ gHSQC,² gNOESY,³ P.E-COSY⁴ and TOCSY.⁵ When characterizing crude reaction mixture the WET pulse sequence was used.⁶ Infrared spectra (IR) were obtained on a Perkin-Elmer 1605 FT-IR spectrophotometer. Thin layer chromatography was performed on silica gel (Merck) mounted on aluminium cards or glass
plates and with a fluorescent indicator (254 nm). Preparative chromatographic separations were achieved by using silica gel (Merck) or alumina (Merck). Elemental analyses (C,H,N) were performed by MIKRO KEMI AB, Uppsala, Sweden or by Analytische Laboratorien, Gummersbach, Germany. The values were within ±0.4% of theoretical if not otherwise indicated. DMDO was synthesized by the method of Adam et al.7

Representative procedure for the synthesis of 1-silyloxy-1,3-dienes, exemplified with compound 57b. To a cooled solution (0°C) of 80 mg (0.7 mmol) 56 in 4 mL dry CH2Cl2 was added 162 μL (0.9 mmol) of DIEA, followed by the dropwise addition of 213 μL (0.9 mmol) triethylsilyl triflate. The reaction was stirred for ½ h under nitrogen atmosphere. The characterization of 57b was performed by taking a 1mL sample from the reaction mixture. The sample was diluted with 0.5 mL CDCl3 then characterized by NMR. For quantification studies 1,4-dimethoxybenzene was used as internal standard in the quantification studies by NMR. The NMR yield was determined to 72%.

1H NMR (CDCl3) δ 5.45 (s, 1H), 4.51 (d, 2H), 2.48-2.41 (m, 2H), 2.26-2.18 (m, 2H), 2.64-2.58 (m, 2H); 13C NMR (CDCl3) δ 155.8, 144.1, 109.2, 105.3, 32.1, 31.9, 25.1.

54b. 54b was synthesized and characterized according to the general procedure described for compound 57b. NMR spectra were run without any lock-signal in the reaction medium using WET pulse sequence. The NMR yield was determined to 80%.

1H NMR δ 6.22 (bs, 1H), 5.38 (bs, 1H), 5.27 (bs, 1H), 3.43 (m, 1H, obscured by solvent signal), 2.25 (m, 2H), 1.72 (m, 2H); 13C NMR δ 108.9, 69.2, 63.8, 57.9, 42.1, 23.1. Three carbon signals are missing due obscuring solvent peaks. Further NMR experiments are needed to fully characterize 57b.

Compound 58. To a cooled solution (0°C) of hexamethyldisilazane (150 μL, 0.7 mmol) in 10 mL dry THF was added dropwise n-butyl lithium (445 μL, 0.7 mmol) The reaction was stirred under nitrogen atmosphere for ½ h. The reaction temperature was lowered to −78°C and a precooled (-78°C) mixture of Hagemann’s ester 31 (100 mg, 0.6 mmol) and TBDMSCl (110 mg, 0.7 mmol) in 2 mL dry THF was added dropwise. The reaction was stirred for 20 min then the volatiles were evaporated. The residue was eluted through SiO2 with hexanes/Et2O (1:1) to yield 94 mg (60%) crude 54a. To a cooled solution of crude 54a (-50°C) (94 mg, 0.3 mmol) in 10mL dry CH2Cl2 was added a precooled (-20°C) 0.06M acetone solution of DMDO (12 mL 0.8 mmol). The reaction mixture was stirred at −50°C for 1 ½ h, then was added dropwise Et3N·3HF (135 μL, 0.8 mmol) and the reaction was stirred o.n. and the temperature of the reaction allowed to rise to 15°C. The volatiles
were evaporated and the residue flash column chromatographed [hexanes/Et2O, 1:1] to yield 26.5 mg (45%), calculated on the starting material 31, of 58 as an oil.

\(^1\)H NMR (CDCl\(_3\)) \(\delta\) 6.22 (bs, 1H), 4.35 (q, 2H), 3.79 (s, OH), 2.68-2.61 (m, 2H), 2.41 (ddd, 1H), 2.23 (ddd, 1H), 1.35 (s, 3H); \(^{13}\)C NMR (CDCl\(_3\)) \(\delta\) 200.0, 176.2, 158.5, 129.1, 75.2, 62.7, 34.0, 32.3, 19.1, 15.2.

**Compound 59.** To a cooled solution (0°C) of 31 (50 mg, 0.3 mmol) in 4 mL dry CH\(_2\)Cl\(_2\) was added DIEA (62 \(\mu\)L, 0.4mmol), followed by the dropwise addition of triethylsilyl triflate (81 \(\mu\)L, 0.4 mmol). The reaction was stirred for \(\frac{1}{2}\) h under nitrogen atmosphere. The temperature was lowered to –78°C and was added a precooled (-20°C) 0.09 M acetone solution of DMDO (5 mL, 0.4 mmol). The reaction mixture was stirred for \(\frac{1}{2}\) h, then the volatiles were evaporated. The residue was taken up in CDCl\(_3\) and immediately analyzed by NMR. Selected chemical shifts for 59 (omitting the triethyl silyl group).

\(^1\)H NMR (CDCl\(_3\)) \(\delta\) 6.22 (bs, 1H), 4.38 (dd, 1H), 4.29 (dd, 1H), 4.21 (q, 2H), 3.38 (m, 1H) 2.58 (m, 1H), 2.42-2.36 (m, 2H), 2.22 (m, 1H); \(^{13}\)C NMR (CDCl\(_3\)) \(\delta\) 200.0, 172.3, 160.1, 124.2, 64.9, 45.1, 42.1, 37.7, 24.1, 15.1.

**2\(\alpha/\beta\)-Hydroxycholest-4-en-3-one (61).** To a cooled solution (0°C) of (+)-4-cholesten-3-one (100 mg, 0.3 mmol) in 10 mL dry CH\(_2\)Cl\(_2\) was added Et\(_3\)N (75 \(\mu\)L, 0.5 mmol), followed by the dropwise addition of triethylsilyl triflate (120 \(\mu\)L, 0.5 mmol). The reaction was stirred for \(\frac{1}{2}\) h under nitrogen atmosphere. The temperature was lowered to –78°C and was added a precooled (-20°C) 0.09 M acetone solution of DMDO (4.5 mL, 0.4 mmol). The reaction was stirred for \(\frac{1}{2}\) h then was added Et\(_3\)N·3HF (65 \(\mu\)L, 0.4 mmol). The reaction was stirred over night and the temperature allowed reach r.t. The reaction was quenched by the addition of 10 mL 5% aq. NaHCO\(_3\) and the phases were partitioned. The organic phase was washed with additional portions (2x10mL) 5% aq. NaHCO\(_3\). The volatiles were dried over MgSO\(_4\) and concentrated. The residue was subjected to flash column chromatography [SiO\(_2\) hexanes/ether, 1:1] to yield 55 mg (53%) of 61 as a 3:1 mixture of 2\(\alpha/\beta\)-isomers. Selected chemical shifts for the isomers of 61:

- 2\(\alpha\)-Hydroxycholest-4-en-3-one. \(^1\)H NMR (CDCl\(_3\)) \(\delta\) 5.77 (d, \(J=1.5\) Hz, 1H, H-4), 4.24 (dd, \(J= 5.7, 13.7\) Hz 1H, H-2), 3.60 (br s, OH), 2.34 (dd, \(J=5.7, 12.6\) Hz, 1H, H-1\(_{eq}\)), 1.53 (dd, \(J=12.6, 13.7\) Hz, 1H, H-1\(_{ax}\)), 1.27 (s, 3H, Me-19); \(^{13}\)C NMR (CDCl\(_3\)) \(\delta\) 200.0 (C=O), 173.7 (C-5), 120.3 (C-4) 69.6 (C-2), 40.9 (C-6), 44.3 (C-1), 33.2 (C-7).

- 2\(\beta\)-Hydroxycholest-4-en-3-one. \(^1\)H NMR (CDCl\(_3\)) \(\delta\) 5.78 (d, \(J=1.3\) Hz, 1H, H-4), 4.17 (dd, \(J= 5.6, 13.9\) Hz 1H, H-2), 3.60 (brs, OH), 2.46 (dd, \(J=5.6, 13.6\) Hz, 1H, H-1\(_{eq}\)), 1.53 (obscured m, 1H, H-1\(_{ax}\)), 1.16 (s, 3H, Me-19); \(^{13}\)C NMR (CDCl\(_3\)) \(\delta\) 200.0 (C=O), 175.7 (C-5), 118.7 (C-4) 68.7 (C-2), 41.8 (C-6), 39.8 (C-1), 33.7 (C-7).
References


10.5. Supplementary Information for Unpublished Compounds Described in the Thesis

Pär Holmberg.

**Experimental**

Melting points were determined on a Büchi SMP-20 apparatus. $^1$H and $^{13}$C NMR spectra were recorded on a Varian Unity 400 MHz, a Varian MercuryPlus 300 MHz, or a Varian Unity Inova 500 MHz spectrometer. Chemical shifts are referenced indirectly to tetramethylsilane (0.0 ppm) via the (residual) solvent signals. Coupling constants are given in Hertz, and the splitting patterns are designated as follows: s, singlet, d, doublet, dd, doublet of doublets, t, triplet, q, quartet, quint, quintett, hept, heptet, sext, sextet, oct, octett and app for apparent. NMR signals were assigned from gHMBC,$^1$ gHSQC,$^2$ gNOESY,$^3$ P.E-COSY$^4$ and TOCSY$^5$ spectra. When characterizing crude reaction mixtures in non-deuterated solvents, the WET pulse sequence element was used.$^6$ Thin layer chromatography was performed on silica gel (Merck) mounted on aluminium cards or glass plates and with a fluorescent indicator (254 nm). Preparative chromatographic separations were achieved by using silica gel (Merck) or alumina (Merck). Elemental analyses (C,H,N) were performed by Mikro Kemi AB, Uppsala, Sweden or by Analytische Laboratorien, Gummersbach, Germany. The values were within ±0.4% of theoretical if not otherwise indicated.
10.5.1. Chromans

3-Nitro-8-methoxy-2-chromen (10). 5 g (32.9 mmol) of 2-hydroxy-3-methoxybenzaldehyde, 5 g (16.4 mmol) n-Bu₂NH₂Cl, 4.8 g nitroethanol (52.6 mmol) and 120 mL iso-pentyl acetate were mixed in a one necked 200 mL round bottom flask, fitted with a Soxhlet extractor filled with 4Å molecular sieves. The mixture was vigorously refluxed for 24 h. Then the reaction mixture was cooled to r.t. and the black solids filtered off. The black solids were rinsed with several portions of EtOAc and the organic phases concentrated. The residue was subjected to flash column chromatography [SiO₂, CH₂Cl₂] to yield 4 g crystals. Further analysis of the obtained crystals by GC-MS showed three peaks with the correct mass and one peak corresponding to N-nitrosodibutylamine. Recrystallization from hexanes/benzene provided 2.8 g (13.5 mmol, 41%) of 10.

1H NMR (CDCl₃) δ 6.99 (t, 1H), 6.84 (m, 1H), 6.71 (m, 1H), 4.44 (s, 2H), 3.88 (s, 3H), 3.59 (s, 1H); 13C NMR (CDCl₃) δ 148.1, 143.8, 139.1, 129.2, 122.6, 122.2, 118.9, 116.3, 63.1, 56.2.

8-Methoxy-3-chromanamine (11) A mixture of 20 mL (20 mmol) 1 M BH₃·THF and 0.1 g (2.4 mmol) NaBH₄ in 100 mL dry THF, under nitrogen atmosphere, was cooled to 0°C and 10 (1.0 g, 4.8 mmol) added dropwise. The external ice bath was removed and the reaction mixture heated to 65°C for 24 h. The reaction mixture was then cooled to 0°C and 40 mL H₂O added cautiously. The pH of the reaction mixture was lowered to 1 with 1.5 M H₂SO₄. Then the reaction mixture was heated to 60°C for 1 h. The THF was evaporated and the water phase washed with two portions of diethyl ether and two portions of CH₂Cl₂. The water phase was made basic with solid NaOH (pH>12) and continuously extracted (Soxhlet extractor) for 72 h. The organic phase was dried over K₂CO₃ and concentrated to yield 0.78 g (4.40 mmol, 90%) of 11.

1H NMR (CDCl₃) δ 6.89-6.6.62 (m, 3H), 4.23 (m, 1H), 4.0-3.80 (m, 4H), 3.44 (m, 1H), 3.1 (m, 1H), 2.65 (m, 1H); 13C NMR (CDCl₃) δ 147.9, 142.9, 121.8, 120.8, 120.0, 108.89, 71.5, 55.5, 43.7, 34.1.

3-Isocyanato-8-methoxychroman (17). To a solution of 100 mg (0.56 mmol) 11 and 0.83 mg (0.3 mmol) triphosgene in CH₂Cl₂ at 0°C was added 113 mg (1.1 mmol) Et₃N. The reaction mixture was allowed to warm to r.t and stirred for an additional 3 ½ h. The volatiles were concentrated and anhydrous EtOAc was added until a white precipitate was formed. The supernatant was pipetted off. The procedure was repeated three times, then the volatiles were evaporated. The residue was further purified by Kugelrohr distillation (5 mm Hg) to yield 80 mg (0.36 mmol, 64%) of 17.
1\textsuperscript{H} NMR (CDCl\textsubscript{3}) \(\delta\) 6.86 (m, 1H), 6.77 (m, 1H), 6.65 (m, 1H), 4.29-4.11 (m, 2H), 3.98 (m, 1H), 3.11 (m, 1H), 2.90 (m, 1H); \(\textsuperscript{13}\text{C}\) NMR (CDCl\textsubscript{3}) \(\delta\) 148.4, 142.9, 123.1, 121.9, 121.2, 119.3, 110.2, 69.2, 56.1, 47.6, 33.4.

10.5.2. Salvinorin A and Analogues

\textbf{Isolation of Salvinorin A.} 50 g dried \textit{Salvia divinorum} leaves obtained from Botanic-Art, Maassluis, Netherlands were steeped in 400 mL acetone for 15 min. The leaves were filtered off and the organic phase collected. The leaves were extracted two more times with the same procedure. The pooled organic phases were evaporated. The residue was column chromatographed [activated charcoal/celite (1:1), acetone] to yield an oil. The oil was triturated with a small amount of MeOH followed by 20 mL diethyl ether. The supernatant was carefully pipetted off. Two additional volumes of diethyl ether were added, every time pipetting off the supernatant. The crystals were dried under vacuum over night to yield 70 mg of pure salvinorin A. The collected mother liquors were evaporated and subjected to the same treatment as above to provide an additional 20 mg of salvinorin A. The total yield was 90 mg from 50 g of dried leaves. Spectroscopic data were in accordance with the literature.\textsuperscript{7}

\(1\textsuperscript{H} \) NMR (CDCl\textsubscript{3}) \(\delta\) 7.41 (dd, \(J=0.9, 1.6\) Hz, 1H), 7.38 (dd, \(J=1.7, x\) Hz, 1H), 6.37 (dd, \(J=1.7, 0.6\) Hz, 1H), 5.52 (m, 1H), 5.15 (m, 1H), 3.75 (s, 3H), 2.75 (dd, \(J=7.2, 9.9\) Hz, 1H), 2.50 (dd, \(J=5.2, 13.4\) Hz, 1H), 2.31 (m, 2H), 2.17 (m, 5H), 2.04 (m, 1H), 1.73-1.54 (m, 4H), 1.45 (s, 3H), 1.12 (s, 3H); \(\textsuperscript{13}\text{C}\) NMR (CDCl\textsubscript{3}) \(\delta\) 200.1, 172.3, 170.1, 169.1, 144.1, 140.1 124.3, 108.2, 76.2, 64.0, 54.2, 51.9, 51.1, 43.3, 42.1, 38.2, 35.5, 30.9, 20.5, 16.4, 15.2, 18.3.

1-(3-Furyl)-3-methyl-but-3-en-1-ol (34). Zink powder (0.8 g, 12.2 mmol) was added to a mixture of 1 g (10.4 mmol) 3-furaldehyd and 1.7 g (12.6 mmol) 3-bromo-2-metylpropene in 10mL saturated aqueous NH\textsubscript{4}Cl and 2mL THF. A slightly exothermic reaction followed and the mixture was stirred for an additional hour or until the mixture had cooled to r.t. The reaction mixture was extracted with diethyl ether (3x25 mL). The combined organic phases were dried over MgSO\textsubscript{4} and concentrated to give 2 g (9.6 mmol, 78\%) of 34 as an oil.

\(1\textsuperscript{H} \) NMR (CDCl\textsubscript{3}) \(\delta\) 1.77 (s, 3H) 2.18 (s, 1H) 2.42 (m, 2H), 4.83 (m, 2H) 4.91 (m, 1H) 6.41 (m, 1H), 7.29-7.46 (m, 2H). \(\textsuperscript{13}\text{C}\)-NMR (CDCl\textsubscript{3}) \(\delta\) 22.4, 46.8, 64.3, 108.6, 114.2, 128.6, 139.1, 142.1, 143.3.

2-(3-Furyl)-4-methyl-3,6-dihydro-2H-pyran (36). 1.2 g (7.9 mmol) of 34 was dissolved in ice cold dry tetrahydrofuran under a nitrogen atmosphere. To this solution was added 0.4 g (16.7 mmol) NaH was added followed by the addition of 2.9 g (24.0 mmol) 3-bromopropene.The solution was stirred at r.t. over night. The reaction was quenched with water and extracted with
diethyl ether (3x50 mL). The combined organic phases were dried over MgSO₄ and concentrated to give 1.3 g (6.76 mmol, 86%) of 35 as an oil, MS: m/z 192, which immediately was used without any further characterization in the ring closing metathesis.

To a heated (40°C) solution of 700 mg (3.65 mmol) 35 in dry dichloromethane under nitrogen atmosphere was added 163 mg (0.2 mmol) of catalyst ii and the mixture kept at reflux for 2h. The volatiles were concentrated and the residue eluted with ether through a pad of silica. The volatiles were evaporated and the residue further purified by flash column chromatography [SiO₂, hexanes/ether (1:1)] to yield 483 mg (69%) of pure 36. ¹H NMR (CDCl₃) δ 77.46-7.41 (m, 1H), 6.48 (s, 1H), 5.45 (m, 1H), 4.61 (m, 1H) 4.35-4.26 (m, 2H), 2.33 (m, 1H), 2.15 (m, 1H), 1.8 (s, 3H); ¹³C NMR (CDCl₃) δ 143.7, 139.5, 127.0, 119.6, 109.0, 78.7, 64.2, 38.1, 32.0, 22.1.

6-(3-Furyl)-4-methyl-5,6-dihydro-pyran-2-one (A). A solution of 36 (0.3 g, 1.8 mmol) in CH₂Cl₂ (2 mL), cooled to -50°C, was added dropwise to a solution of 1.8 g (18.2 mmol) of CrO₃ and 1.7 g (18.2 mmol) 3,5-dimethylpyrazine at –50°C in 30 mL CH₂Cl₂. The reaction was kept under N₂ atmosphere and stirred for 2.5h. The reaction was quenched by the addition of 20 mL 5M NaOH cooled to 0°C. The reaction was stirred for 1h at 0°C. The phases were allowed to separate and the organic phase was washed with 30 mL brine followed by additional washings with 1M HCl (3x20 mL). The organic phase was dried over Na₂SO₄ and concentrated. The residue was purified by flash column chromatography [SiO₂, hexanes/ether (1:1)] to yield 73.8 mg (0.41 mmol, 23%) of A.

¹H NMR (CDCl₃) δ 7.51 (s, 1H), 7.43 (s, 1H), 6.47 (s, 1H), 5.89 (s, 1H), 5.41 (m, 1H), 2.73 (m, 1H) 2.45 (m, 1H), 2.05 (s, 3H); ¹³C NMR (CDCl₃) δ 165.3, 157.2, 144.3, 144.0, 125.1, 117.5, 109.4, 72.9 36.0, 24.1

1-(3-Furyl)-3-buten-1-ol (37). 1.1 g (17.4 mmol) Zink powder was added to a mixture of 1.4 g (14.5 mmol) 3-furaldehyd and 2.1 g (17.4 mmol) 3-bromo-propene in 20 mL saturated aqueous NH₄Cl and 4mL THF. A slightly exothermic reaction followed and the mixture was stirred for an additional hour or until the reaction had cooled to r.t. The reaction mixture was extracted with ether (3x25 mL). The combined organic phases were dried over MgSO₄ and concentrated to give 2 g (14.5 mmol, 99%) of 37 as an oil.

¹H NMR (CDCl₃) δ 7.39 (m, 2H), 6.40 (s, 1H), 5.80 (m, 1H), 5.17 (m, 2H), 4.71 (m, 1H), 2.51-2.46 (m, 2H), 2.05 (s, 1H); ¹³C NMR (CDCl₃) δ 143.5, 139.3, 134.3, 128.6, 118.8, 108.7, 66.3, 42.6.

2-(3-Furyl)-3,6-dihydro-2H-pyrany (39). 2 g (14.5 mmol) of 37 was dissolved in ice cold dry tetrahydrofuran under nitrogen atmosphere. 1.2 g (50 mmol) NaH was added followed by the addition of 3.8 g (31.4 mmol) 3-bromopropene. The solution was stirred at r.t. over night. The reaction was
quenched with water and extracted with diethyl ether (3x50 mL). The combined organic phases were dried over MgSO₄ and concentrated to give 1.3 g (7.3 mmol, 50%) of 38 as an oil, which immediately was used in the ring closing metathesis.

To a heated (40°C) solution of 38 (200 mg, 1.1 mmol) in dry dichloromethane under nitrogen atmosphere, was added 48 mg (0.06 mmol) of catalyst ii and the solution was kept at reflux for 2h. The volatiles were concentrated and the residue eluted with ether through a pad of silica. The volatiles were evaporated and the residue further purified by flash column chromatography [SiO₂, hexanes/ether (1:1)] to yield 131 mg (0.87 mmol, 80%) of 41.

1H NMR (CDCl₃) δ 7.45 (s, 1H), 7.43 (s 1H ), 6.48 (s, 1H), 5.92 (m, 1H), 5.82 (m, 1H), 4.61 (m, 1H), 4.40-4.28 (m, 2H), 2.42 (m, 1H), 2.28 (m, 1H) ; 13C NMR (CDCl₃) δ 143.0, 139.7, 126.1, 125.2, 121.7, 109.5, 71.9, 69.5, 35.0.

1-(3-Furyl)-but-3-enyl acrylate (40) To a cooled solution(0oC) of 37 (0.7 g, 5.1 mmol) and acryloyl chloride (0.9 g, 9.9 mmol) in 10 mL dry dichloromethane, under an nitrogen atmosphere, was Et₃N (1.5 g, 15 mmol) added dropwise. The solution was stirred at room temperature for 1h. 50 mL diethyl ether was added to the solution and white crystals precipitated. The crystals were filtered off and the volatiles concentrated. The obtained residue was dissolved in diethyl ether, filtered and concentrated to give 0.7 g (3.6 mmol, 71%) of 40 as an oil.

1H NMR (CDCl₃) δ 7.43 (m, 1H), 7.36 (m, 1H), 6.45-6.35 (m 2H), 6.11 (m, 1H), 5.59-5.95 (m, 3H) 5.08 (m, 2H), 2.59 (m, 2H); 13C NMR (CDCl₃) δ 165.3, 143.1, 140.3, 133.0, 130.8, 128.4, 124.3, 118.2, 108.9, 67.8, 39.1.

6-(3-Furyl)-5,6-dihydro-pyran-2-one (41). To a heated (40 oC) solution of 40 (100 mg, 0.5 mmol) in dry dichloromethane under nitrogen atmosphere, was added catalyst ii (22 mg, 0.03mmol) and kept at reflux for 2h. The volatiles were concentrated and the residue eluted with ether through a pad of silica. The volatiles were evaporated and the residue further purified by flash column chromatography [SiO₂, hexanes/ether (1:1)] to yield 60 mg (0.37 mmol, 73%) of 41.

1H NMR (CDCl₃) δ 7.45 (s, 1H), 7.43 (s 1H ), 6.46 (s, 1H), 5.46 (dd, 1H), 5.29 (m, 1H), 2.58-2.85 (m, 2H), 2.58-2.85 (m, 2H); 13C NMR (CDCl₃) δ 165.1, 159.3, 144.6, 143.7, 139.9, 121.7, 108.5, 71.9, 30.1.

49. A mixture of ethyl-4-oxocyclohexanecarboxylate (1 g, 5.9 mmol) and pyrrolidine (540 µL, 6.5) mmol in 50 mL benzene was kept at reflux under azeotropic conditions for 14 h. The volatiles were evaporated and the residue
kept under vacuum in a 40°C water bath for ½ h, to assure that excess pyrrolidine was evaporated. To a solution of the crude enamine in 30 mL anhydrous ethanol was added freshly distilled methyl acrylate (480 µL, 5.3 mmol) added. The reaction was heated at 60°C under nitrogen atmosphere for 1h. 0.4 mL water was added and the enamine was hydrolyzed for ½ h at 50°C. The volatiles were evaporated and kugelrohr distillation at 150°C (2 mm Hg) afforded 1 g (3.9 mmol, 68%) of 49 as a 1:1 mixture of cis/trans isomers.

\[ ^{1}H \text{NMR (CDCl}_{3} \] \( \delta \) 4.22-4.06 (m, 4H), 3.61 (s, 6H ), 2.84-2.72 (m, 2H), 2.45-2.25 (m, 13H), 5.29 (m, 1H), 2.10-1.44 (m, 9H), 1.27-1.70 (m, 6H); \[ ^{13}C \text{NMR (CDCl}_{3} \] \( \delta \) 211.8, 210.6, 174.2, 174.1, 174.0, 173.9, 61.0, 60.9, 51.7, 47.9, 46.8, 42.4, 40.8, 38.8, 38.5, 36.0, 34.6, 31.8, 31.5, 30.0, 28.8, 25.1, 24.6, 14.4, 14.3. One carbon signal is missing, probably due to overlapping signals.

Ethyl-[3-(tert-butyldimethylsilyloxy)-5-(methoxycarbonyl)ethyl]-4-oxocyclohexanoat 51. To a cooled (0°C) solution of 49 (150 mg, 0.58 mmol) in 10 ML CH\textsubscript{2}Cl\textsubscript{2} under a nitrogen atmosphere, was Et\textsubscript{3}N (165 µL, 1.16 mmol) added dropwise, followed by the dropwise addition of triethylsilyl triflate (265 µL, 1.17 mmol). After ½ h the temperature was lowered to –78°C and was added a cooled (–20°C) 0.1M acetone solution of DMDO (1l mL,1.2 mmol). After 20 min was Et\textsubscript{3}N·3HF (200 µL 1.2 mmol) added. The temperature was allowed to rise to r.t over night. The reaction was quenched with aqueous 10 mL NaHCO\textsubscript{3} and the phases were partitioned. The organic phase was washed with additional portions of aqueous NaHCO\textsubscript{3} (2x10 mL), followed by washing with 10 mL brine. The organic phase was dried over MgSO\textsubscript{4} and concentrated to provide a crude yield of 126 mg (0.46 mmol, 80%) of 50a and 50b as an oil which were used in the following reaction without any further purification.

To a solution of crude 50a and 50b (126 mg, 0.0.46 mmol) in 5 mL CH\textsubscript{2}Cl\textsubscript{2} was imidazole (63 mg, 0.92 mmol), DMAP (5 mg, 0.05 mmol) and TBDMOSCI (140 mg, 0.93 mmol) added. The reaction was kept under nitrogen and stirred over night at r.t. The volatiles were evaporated and the residue subjected to flash column chromatography [SiO\textsubscript{2}, hexanes/ether, 1:1] to give 27mg (0.07 mmol, 15%) 51 as an oil. The ratio between 51a and 51b was determined by NMR to be 3:2.
Compound 51a. 1H NMR (CDCl₃) δ 4.21 (m, 1H, H-2a), 4.15 (q, J=7.0, 2H, H-11), 2.86 (m, 1H, H-4a), 2.48 (m, 1H, H-3e), 2.47 (m, 1H, H-6a), 2.37 (m, 2H, C-8) 2.30 (m, 1H, H-5a), 2.09 (m, 1H, H-7), 1.88 (m, 1H, H-3e), 1.55 (m, 1H, H-5e), 1.26 (J=7.0, 3H, H-12), 0.89 (s, 9H, tert-butyl-Si), 0.12 (s, 3H, Me-Si), 0.01 (s, 3H, Me-Si); 13C NMR (CDCl₃) δ 208.5 (C=O), 174.0 (C-9), 172.1 (C-13), 75.9 (C-2) 61.1 (C-11), 51.8 (C-10), 45.4 (C-6), 40.7 (C-4), 39.8 (C-3), 36.0 (C-5), 31.7 (C-8), 24.4 (C-7), 18.6 (Me₃C-Si), 14.2 (C-12), -5.2 (Me-Si), -4.5 (Me-Si).

Compound 51b. 1H NMR (CDCl₃) δ 4.41 (m, 1H, H-2a), 4.23 (q, J=7.2, 2H, H-11), 2.90 (m, 1H, H-4a), 2.61 (m, 1H, H-3e), 2.42 (m, 1H, H-5e), 2.41 (m, 1H, H-6a) 2.37 (m, 1H, H-8), 2.09 (m, 1H, H-7), 1.84 (m, 1H, H-3e), 1.57 (m, 1H, H-5e), 1.60 (m, 1H, H-7), 1.32 (J=7.2, 3H, H-12), 0.90 (s, 9H, tert-butyl-Si), 0.12 (s, 3H, Me-Si), 0.02 (s, 3H, Me-Si); 13C NMR (CDCl₃) δ 209.2 (C=O), 174.2 (C-9), 172.9 (C-13), 74.5 (C-2) 61.2 (C-11), 51.8 (C-10), 46.4 (C-6), 38.5 (C-4), 38.4 (C-3), 35.0 (C-5), 31.7 (C-8), 26.2 (t-butyl-Si), 24.4 (C-7), 18.6 (Me₃C-Si), 14.4 (C-12), -5.2 (Me-Si), -4.5 (Me-Si).

References

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