Enantioselective preparation of ω-functionalized $O$-acylated cyanohydrins

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Stockholm, 14th March to 17th August 2011
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# List of Abbreviations

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Meaning</th>
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<tbody>
<tr>
<td>AcCN</td>
<td>acetyl cyanide</td>
</tr>
<tr>
<td>Brine</td>
<td>concentrated sodium chloride solution</td>
</tr>
<tr>
<td>CAL-A</td>
<td>Candida Antartica Lipase A</td>
</tr>
<tr>
<td>CAL-B</td>
<td>Candida Antartica Lipase B</td>
</tr>
<tr>
<td>CRL</td>
<td>Candida Rugosa Lipase</td>
</tr>
<tr>
<td>DCM</td>
<td>dichloromethane</td>
</tr>
<tr>
<td>DIBAL-H</td>
<td>diisobutylaluminium hydride</td>
</tr>
<tr>
<td>DIPEA</td>
<td>N-Diisopropylethylamine</td>
</tr>
<tr>
<td>DMF</td>
<td>dimethylformamide</td>
</tr>
<tr>
<td>DMP</td>
<td>Dess-Martin-Periodiane</td>
</tr>
<tr>
<td>EtOAc</td>
<td>ethyl acetate</td>
</tr>
<tr>
<td>equiv.</td>
<td>equivalents</td>
</tr>
<tr>
<td>ee</td>
<td>enantiomeric excess</td>
</tr>
<tr>
<td>GC</td>
<td>gas chromatography</td>
</tr>
<tr>
<td>HPLC</td>
<td>high pressure liquid chromatography</td>
</tr>
<tr>
<td>MER</td>
<td>minor enantiomer recycling</td>
</tr>
<tr>
<td>NMR</td>
<td>nuclear magnetic resonance</td>
</tr>
<tr>
<td>PCC</td>
<td>pyridinium chlorochromate</td>
</tr>
<tr>
<td>rt</td>
<td>room temperature</td>
</tr>
<tr>
<td>TEA</td>
<td>triethylamine</td>
</tr>
<tr>
<td>Abbreviation</td>
<td>Full Form</td>
</tr>
<tr>
<td>--------------</td>
<td>-----------</td>
</tr>
<tr>
<td>THF</td>
<td>tetrahydrofuran</td>
</tr>
<tr>
<td>TLC</td>
<td>thin layer chromatography</td>
</tr>
<tr>
<td>TMS-Cl</td>
<td>trimethylsilyl chloride</td>
</tr>
<tr>
<td>UV</td>
<td>ultra violet</td>
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</table>
Abstract

A minor enantiomer recycling one-pot process using ω-functionalized prochiral aldehydes as starting materials and two reinforcing catalysts has been reported. The desired aldehyde for these process studies was 5-bromo-1-pentanal.

In a two-phase solvent system, enzyme-catalyzed hydrolysis of the minor enantiomer regenerates continuously the prochiral starting material and Lewis acid catalysed addition of acetyl cyanide provides the $O$-acetylated cyanohydrins. The minor enantiomer recycling process has been studied and improved for 5-bromo-1-pentanal to receive high enantiomeric excess and yield of the expected $O$-acetylated cyanohydrin.
1. Introduction

1.1 Asymmetric Synthesis

Asymmetric synthesis is defined as the production of enantiomers from prochiral molecules, where one of the two enantiomers is produced in excess. The synthesis of such enantiomERICALLY enriched compounds, which are of interest for example in pharmacology, is often associated with difficulties.

Enantiomers have, besides rotating plane-polarised light in opposite direction, the same physical properties. They can therefore not be purified by conventional methods, as for example column chromatography and recrystallization. For that reason several other synthetic processes have been developed to produce one of the two enantiomers in excess. Such processes use for example chiral catalysts, enzymes or auxiliaries or the enantiomers can be converted to diastereomers which can be separated easier. The success of asymmetric synthesis can be seen in the enantiomeric excess, ee, which can be calculated as described in formula (1).

\[
ee = \frac{|(R - S)|}{R + S} \cdot 100 \tag{1}
\]

The enantiomeric purity is of interest for example in pharmaceuticals. One example for this purpose is the Contergan scandal in Germany in the 1960´s, were many cases of deformities at unborn babies appeared. This circumstance was taken in context with the medicament Contergan, which many pregnant women swallowed as a sedative. The active substance in that medicament is Thalidomide, which consists of two different enantiomers. One enantiomer has a calming effect and the other one is toxic to reproduction. Until today it is not exactly proved which enantiomer is the toxic one.\[¹\] Even if the medicament consists of enantioenriched substrate both enantiomers will be formed because of an acid catalyzed transformation. If the enantioenriched substance gets to the stomach, where hydrochloric acid is present, the enantiomers will be hydrolyzed and form the racemic mixture, why both enantiomers will be present.
1.2 Minor Enantiomer Recycling

Often asymmetric syntheses are resulting in products with insufficient enantiomeric purity. The minor enantiomer recycling (MER), where two reinforcing catalysts are used, presents a way to improve the enantiomeric excess of the products. The studies of MER have been made on the synthesis of O-acylated enantioenriched cyanohydrins because these molecules serve as versatile synthetic building blocks.

![Energy profile of a typical asymmetric synthesis](image)

**Figure 1:** Energy profile of a typical asymmetric synthesis

Forward and backward reaction of the recycling process are following the microscopic reversibility law, formulated 1924 by R. C. Tolman [2, 3], which is according to IUPAC defined as follows:

"The principle of microscopic reversibility at equilibrium states that, in a system at equilibrium, any molecular process and the reverse of that process occur, on the average, at the same rate." [4]

Thus forward and backward reaction, which are in equilibrium, have to overcome the same transition state barrier with the identical energy profile (Figure 1). According to this law a recycling process (Figure 2) would not be possible because the backward reaction would not be supported for the reason of the unfavourable transition state barrier.
Thus for the generation of an unidirectional cycle a thermodynamic driving force is required.\cite{5} Therefore first the continuous addition of acetyl cyanide to the prochiral aldehyde is needed. This pushes, following the le Chatelier principles, the equilibrium of the forward reaction to the product side because of the mass flow, resulting from the formation of hydrogen cyanide and carboxylate ion, which will go to the aqueous phase and thus out of the reaction. Also the addition of an enzyme is needed to generate a recycling process. The addition of the enzyme, which is enantioselective, creates an additional reaction, a hydrolysis, which is also following the microscopic reversibility law. It generates the backward reaction of the recycling process, where the driving force is also a mass balance. The acetic acid, which is formed during the hydrolysis, is transformed to the acetate with the help of a pH 8 buffer and thus it is irreversible going out of the reaction. This shifts the equilibrium, which results in prochiral aldehyde, to the product side. The reaction follows again the principles from le Chatelier.
With a look at the kinetics of the reaction it can be easily understood how the recycling process is working (Figure 3). For the forward reaction (black line), the energy barrier for the $S$ enantiomer is higher than that for the $R$ enantiomer, means that, with a continuous addition of $B$ (= acetyl cyanide) and the use of a chiral Lewis acid (Ti-salen-complex) as enantioselective catalyst, the $R$ enantiomer is formed easier and in a higher amount than $S$ ($k_R > k_S$). If it is also assumed that for the backward reaction (blue line), the enzyme is enantioselective for the $S$ enantiomer ($k_s' \gg k_{r'}$), the major amount of $S$ is recycled back to prochiral aldehyde, while the hydrolysis of the $R$ enantiomer is non quantitative and slower. This explanation shows that the recycling process consists of two systems which are both equivalent to the microscopic reversibility law and the energy profile shown in figure 1.[5, 6]

In the recycling process two different catalysts, a titanium-salen-complex and an enzyme, are working together in a two-phase system. They effect in that way that the minor enantiomer changes back to starting material via enzymatic hydrolysis to get the chance to form new product with the help of the titanium-salen-complex.

The titanium-salen-complex activates the aldehyde and the acetyl cyanide at the same time since the oxygen atoms of both compounds are coordinating each to one of the two titanium-metal-centers in the complex (Figure 4). Thus the activated reactants are close to each other, thereby favoring the reaction. In earlier studies Moberg et.al. suggested this mechanism for the enantioselective addition of $\alpha$-ketonitriles to aldehydes at -40 °C with the use of a Lewis acid and a Lewis base as catalysts. This double activation is needed to start the reaction at this temperature.[7] The addition of Lewis base is not needed at room temperature or higher temperature because the reaction barrier is low enough for the

![Figure 3: Recycling process](image-url)
substances react without an additional Lewis base. The second catalyst, the enzyme, is chosen in that way that it hydrolyses only one of the two enantiomers. Thus the desired enantiomer is built in excess while the minor enantiomer is formed back to prochiral starting material to get the chance to react again. The configuration of the formed product is finally depending on the combination of Ti-salen-complex and used enzyme. Moberg et al. showed that the (S, S)-Ti-salen-complex in combination with Candida antarctica Lipase B (CAL-B) usually provides the products with the absolute configuration R whereas the (R, R)-Ti-salen-complex in combination with Candida rugosa lipase (CRL) usually provides product with the opposite configuration, S.[5]

![Diagram](image)

**Figure 4:** Proposed mechanism for addition of ketonitriles to aldehydes (left)[7]; Structure of the Ti-salen-complex (right)[5]

The main advantage of MER processes is the positive effect on enantiomeric excess. Due to the enzymatic hydrolysis of the minor enantiomer back to starting material and the possibility for the new formed starting material to react again the ee is able to increase. This circumstance can be explained as follows. The minor enantiomer is hydrolyzed selectively by the enzyme back to staring material and therefore the ee is increasing, what can be easily understood with formula (1). If \( S \) gets smaller numerator and denominator are approximating to the same value, what results in a higher ee.

The yield can be affected, namely by the variation of the amounts of the two different catalysts. If the amount of the Ti-salen-complex (cat. 1) is higher than that for the enzyme (cat. 2) the yield will increase because more product is formed while less product is hydrolyzed. With this combination of amounts of catalysts also the ee is affected in that way that it needs longer time to reach steady state. When the amount of enzyme is higher than the amount of Ti-salen-complex the yield will be lower because less product is
formed, what moreover hydrolyzes faster back to starting material. The ee will in this case be affected in that way that it reaches steady state faster.

1.3 Aims of this Project

The main part of this project includes studies of the minor enantiomer recycling process using ω-functionalized prochiral aldehydes as starting material. In that case 5-bromo-1-pentanal was used as prochiral aldehyde. The products which are formed in the cyanation reaction are of interest because they undergo a variety of useful transformations. In earlier studies γ-functionalized cyanohydrins, which were obtained for example from 4-bromo-1-butanal, have been transformed to a piperidin-3-ol (Figure 5) which is found in many different bioactive compounds.[8]

![Reaction scheme for the synthesis of (R)-benzyl-3-hydroxypiperidine-1-carboxylate][8]

Figure 5: Reaction scheme for the synthesis of (R)-benzyl-3-hydroxypiperidine-1-carboxylate[8]

Also the cyanohydrins from the higher homologue have been used for the preparation of 2-cyanopiperidines (Figure 6).[19]
For that reason the MER process using 5-bromo-1-pentanal as prochiral ω-functionalized starting material was studied and improved to finally get the wanted enantiomer in excess for further enantiomerically enriched synthesis. Furthermore, attempts were made to substitute the bromo atom by different secondary amines because the products of these reactions are useful for further syntheses to substrates which are found in the nature.

Another part of this project was the synthesis of 6-hydroxyhexanal starting from ε-caprolactone and the study of the minor enantiomer recycling process with this substrate.

Also the synthesis and the recycling process of 3-bromo-1-propanal were part of the research project.

**Figure 6**: Reaction schema for the synthesis of (S)-1-benzyl-2-cyanopiperidine[^9^]
2. Results and Discussion

2.1 Substrate synthesis

For the MER studies, different aldehydes (3, 8 and 13) had to be synthesized as starting materials.

Aldehyde 3 was synthesized from 1,5-pentanediol (1). First a nucleophilic substitution with aqueous hydrobromic acid HBr(aq) was used to synthesize 5-bromo-1-pentanol (2). The bromo alcohol was then oxidized with PCC to produce the wanted aldehyde, 5-bromo-pentanal (3) (Figure 8). These two steps have previously been reported in the literature.\[10\]

For further studies, 3 was converted to the racemic mixture of O-acylcyanohydrin, by treatment with acetyl cyanide in the presence of triethylamine as base to receive racemic 5-bromo-1-cyanopentyl acetate (rac-4) (Figure 9).
Racemate rac-4 was then used for some substitution reactions where the bromo atom was replaced by secondary amines.

The substitution reactions were carried out in acetonitrile in the presence of DIPEA as base. For the synthesis of rac-5 (Figure 10), the reaction mixture was heated up to 40 °C. Out of the results of the NMR spectra, which were taken after finishing the reaction, the success of the reaction could not be proven because the peaks in the NMR spectra could not be assigned to either starting material or expected product.

For the synthesis of rac-6 (Figure 10), the reaction mixture was refluxed in the presence of a catalytic amount of potassium iodide, which was added to generate a better leaving group instead of the bromo atom. The measured ^1H- and ^13C-NMR spectra showed that the correct product was formed. The yield for rac-6 was 47%.

Since it was obtained that the substitution reaction of the bromo atom of the racemic mixture was successful with the use of morpholine as nucleophile further substitution reactions were considered. For these substitution reactions the enantioenriched product (R)-4 was used as starting material in combination with different amines as nucleophiles. The enantioenriched product was used to control how the ee would be affected by substitution of the bromo atom from an amine. For these reactions morpholine could not be used because it has no properties which would allow it to do analysis of the substitution product using for example an HPLC with an UV-detector. The first
substitution reaction was carried out with the chiral amine \((S)-(\cdot)-\text{phenylethylamine}\). This reaction would result in a diastereomeric mixture which would be visible on NMR. The reaction product could also be analysed using HPLC with the UV-detector. The aromatic amines benzylamine and N-benzylmethylamine were chosen because they are also UV-visible and thus the products of the substitution reactions using these amines would be also able to analyse with HPLC (Figure 11).

![Reaction Diagram](image)

**Figure 11:** Reactions of enantioenriched 5-bromocyanohydrin with amines

These reactions were also carried out acetonitrile in the presence of DIPEA as base. For the synthesis of \((R)-7\) the reaction mixture was stirred for 20 h at rt in the presence of a catalytic amount of potassium iodide. The crude \(^1\text{H}-\text{NMR}\) spectrum of the reaction mixture did not show characteristic product peaks.
For the synthesis of (R)-8 the reaction mixture was also stirred at rt for 23 h in the presence of a catalytic amount of potassium iodide. The results of this reaction would be interesting because such enantiomeric enriched amine products could be used as starting material for further natural product syntheses. Out of the $^1$H-NMR spectra, which were made before and after the purification column, the success of the reaction could not be proven because the assignment of the obtained peaks to the supposed product peaks was not possible.

Substitution product (R)-9 was also attempted to be synthesized by stirring the reaction mixture at rt for 44 h. In this reaction after 24 h another 0.5 equiv. of N-benzylmethylamine were added because the crude $^1$H-NMR showed a large amount of starting material. After evaporation of the solvent the residue was purified by column chromatography. Unfortunately the $^1$H-NMR spectrum of the purified substance showed a mixture of the starting materials, N-benzylmethylamine and 5-bromo-1-cyanopentyl acetate.

Another aldehyde that was used in the studies was 6-hydroxy-1-hexanal (11). ε-Caprolactone (10) was used as staring material. With DIBAL-H 10 was reduced to the free hydroxyaldehyde 11 in 25% yield (Figure 12). A low yield was obtained because there was no possibility to purify the whole amount of aldehyde neither with column chromatography nor with recrystallization. Only a small amount of 25% yield could be purified using these methods. This procedure has been reported in the literature.[11]

![Figure 12: Attempt synthesis of 6-hydroxy-1-hexanal (11) from ε-caprolactone](image)

For further study, attempts were made to convert aldehyde 11 to the corresponding racemate rac-12. In this reaction, the base catalyzed equilibrium reaction from 11 to the hemiacetal 13 had to be considered (Figure 13).
The crude $^{13}$C-NMR spectrum of the reaction mixture showed that the diacylated product rac-12a was formed. For that reason a new procedure was considered where 5-hydroxy-pentanal was synthesized from 1,5-pentanediol, to examine if the preparation of the racemate is better with this substrate (Figure 14). First the mono-protected diol 14 had to be synthesized to guarantee that at the following bromination and oxidation reactions only one hydroxyl group would react. This protection was previously made with the use of a tert-butyldimethylsilyl- protecting group (TBS)$^{[12]}$, but in this case TMSCl was used instead because this protecting group is not as bulky. This is important for a good working enzyme in the MER process because a too big protecting group will prevent that the molecule could get into the active part of the enzyme where it should react.

The reaction was not successful under these conditions, because of the main difficulty to get the solvent DMF evaporated. Attempts to completely remove the DMF from the reaction mixture resulted in too much loss of product, so that the obtained yield was only 3%. The $^1$H-NMR spectrum showed also that not only the mono-protected diol 14 was formed, also the di-protected reaction product was obtained. For this reason the reaction
conditions were changed. The reaction was then carried out in THF with the use of TEA as base instead of DMF and imidazole (Figure 15).

![Figure 15: Attempt synthesis of mono protected diol 14 with TMSCl](image)

After this experiment it had to be noticed that the mono-protection with the TMS group was not successful. NMR spectra showed that only the di-protected diol 14a was formed with the new chosen conditions. These results can be explained with the size of the protecting group. The bigger the protecting group, the higher is the sterical hindrance and the more the mono-protected product is formed. For the reaction in this synthesis a smaller TMS group was used, while in the literature [12] a bulky TBS group was chosen, for the synthesis of the mono-protected diol.

Another aldehyde, which was also of interest for the studies, was 3-bromo-1-propanal 16. Attempts were made to synthesize 16 from 3-bromo-1-propanol (15) (Figure 16). For this purpose two different methods were used. First the alcohol was oxidized with PCC, following the same procedure as used for the synthesis of 3, to obtain the expected aldehyde.

![Figure 16: Synthesis of 3-bromo-1-propanal from 3-bromo-1-propanol with PCC](image)

The evaluated NMR spectra showed that the PCC- method was not successful for this synthesis because after 6 h reaction time the amount of aldehyde 16 was low and too much starting material was left. For that reason the Dess-Martin-oxidation (Figure 17) was considered for the synthesis of 16.
Figure 17: Synthesis of 3-bromo-1-propanal from 3-bromo-1-propanol via Dess-Martin-oxidation\cite{13}

First the reaction was carried out at room temperature and the solvent was evaporated after 1.5 h when the reaction was finished. The crude $^1$H-NMR showed nearly no aldehyde $^{16}$. It was concluded that the formed 3-bromo-propanal was highly volatile, so that it was also evaporated together with the solvent. For that reason the same reaction was made again but that time the solvent was removed by airstream. The crude $^1$H-NMR showed a large aldehyde peak but also a large amount of starting material. To try to minimize the amount of starting material the reaction conditions were changed, according to another procedure\cite{14}, so that the reaction started at 0 °C and warmed up slowly to room temperature and stirred for 62 h. The reaction was not successful since in the $^1$H-NMR no aldehyde peak was obtained. Since neither the PCC-method nor the Dess-Martin-oxidation with 3-bromo-propanol as starting material were successful for the synthesis of $^{16}$ another starting material and another synthesis pathway should be found to synthesize the desired aldehyde $^{16}$.

2.2 Minor enantiomer recycling

The minor enantiomer recycling process was studied and attempted to improve only for 5-bromo-1-pentanal ($^3$) as starting material. The aldehydes $^{11}$ and $^{16}$ could not be used because for $^{11}$ the diacylated product was obtained and aldehyde $^{16}$ was too difficult to synthesize and purify in good yield.

The asymmetric synthesis and the hydrolysis of the racemic mixture of 5-bromo-pentanal ($^3$) with different enzymes were first studied. The acetyl cyanation was carried out at -40 °C in DCM as solvent. $^3$ reacted with AcCN in the presence of the (S,S)-Ti-salen-complex and NEt$_3$ as Lewis base to produce $^4$ in 67% ee (Figure 18).
Figure 18: Asymmetric synthesis of 5-bromo-1-cyanopentyl acetate

For the hydrolysis tests the enzymes *Candida Antartica* Lipase A (CAL-A), *Candida Antartica* Lipase B (CAL-B) and *Candida Rugosa* Lipase (CRL) were chosen (Figure 19). The hydrolysis of racemate rac-4 was performed in a two-phase-solvent system consisting of toluene and a 1M phosphate buffer pH 8.

Figure 19: Scheme for hydrolysis of 4 with the different enzymes

For CAL-A and CRL the hydrolysis was performed at room temperature. The evaluated GC-results for these enzymes are represented in the Figures 20 and 21.
The hydrolysis with CAL-B was performed at 40 °C and was also followed with GC measurements. The results are represented in Figure 22.
CAL-B and CRL were the most selective. In the case of CRL the \( R \) enantiomer was hydrolysed more rapidly than the \( S \) enantiomer why in the MER process the \((R,R)\)-Ti-salen complex had to be used to guarantee that enantiomer \( S \) becomes the major enantiomer. For CAL-B the hydrolysis worked much better compared to CRL because enantiomer \( S \) was completely hydrolysed after 30 h whereas the relative amount of the desired enantiomer \( R \) decreased only around 10%. In the later MER process the \((S,S)\)-Ti-salen complex, which was synthesized following the procedure described in the experimental part at 4.2, had to be used in combination with CAL-B to get the desired enantiomer \( R \) in excess. Because of the selective hydrolysis these two enzymes were chosen for the MER studies with 5-bromo-1-pentanal (3) as starting material.
The cycle for that substrate works as shown in the following scheme.

![Scheme](image)

**Figure 23:** MER scheme for substrate 3

The MER process with CRL was carried out at room temperature with an addition of 3 equivalents of acetyl cyanide. The used acetyl cyanide was therefore synthesized according to the procedure described in the experimental part at 4.2, over 6.25 h (experimental data s. 4.3). For this experiment the maximum ee was 49%, which was obtained after 1.5 h. But while continuing the experiment the ee decreased to a value of 46% (Figure 24). In contrast, the obtained yield increased with time to a maximum value of 65% (Figure 25).
In comparison to the results obtained with CRL, the use of CAL-B supplied better results. The MER process performed also at room temperature with the use of 20 mg of CAL-B for 0.2 mmol of the aldehyde and an addition of 3 equivalents of acetyl cyanide over 10 h (experimental data s. 4.3) a maximum ee of 59% (Figure 26) and a maximum yield of 66% (Figure 27) were obtained.

**Figure 24:** Obtained enantiomeric excess ee as a function of the time

**Figure 25:** Obtained yield as a function of the time
Out of this comparison it could be concluded that the enzyme CAL-B gave the best results for the MER process, using aldehyde 3 as starting material. For that reason further experiments were made using CAL-B as enzyme to improve the results for the enantiomeric excess and the yield. In these experiments several parameters were changed to see how they affect ee and yield. The change of the temperature for example would affect both ee and yield, while the variation of the amount of enzyme could affect only the yield. The amount of used aldehyde (33 mg, 0.2 mmol) was kept constant during the studies. For a MER process carried out at 40 °C for 6.25 h the maximal obtained ee at
steady state reached a value of 79% and a yield of 45%. Different addition times of the three equivalents of acetyl cyanide affected the values for the enantiomeric excess and yield; for a slower addition of acetyl cyanide value for the yield was increasing while the ee value needed longer time to reach the steady state. A recycling process carried out at room temperature over 10 h with 20 mg CAL-B supplied a maximum ee of 58% and a yield of 66% whereas for a cycle performed also at room temperature over 24 h a maximum ee of 75% and a yield of 70% were obtained. Doubling the amount of CAL-B under previously described conditions with 10 h addition of acetyl cyanide a maximum ee of 67% and a yield of 67% and for the 24 h addition maximal values of 85% ee and 58% yield were obtained. The results described above is visualized in Figure 28 and 29 and summarized in table 1.

**Figure 28:** MER process over 10 h
As expected, with a slow addition of acetyl cyanide and a larger amount of enzyme the steady state value of the ee was reached faster than with half the amount of enzyme after the same addition time. Also a lower yield was expected using more enzyme, but found for the 10 h addition was a slightly higher yield than with less enzyme at the same addition time. These results are due to the amount of CAL-B. Since more enzyme hydrolyses more $S$ in a shorter time, more $R$ can be produced what affects that the ee reached the steady state value faster while the yield decreased because of the faster hydrolysis of the formed product.
3. Conclusion and Outlook

A minor enantiomer recycling process using \( \omega \)-functionalized aldehydes as prochiral starting material and two reinforcing catalysts, an enzyme and a Ti-salen-complex, has been reported.

The synthesis of 5-bromo-1-pentanal as starting material was successful, and it was therefore used for further MER studies. The aldehydes 6-hydroxyhexanal and 3-bromo-propanal were not able to synthesize as pure compounds. The different attempted syntheses pathways did not fit for the formation of the desired products.

The MER process was carried out with 5-bromo-pentanal as prochiral starting material. The optimization of this process was studied by variation of different parameters, as temperature, amount of enzyme and type of enzyme. The best results for the ee were obtained using CAL-B and carrying out the MER process at room temperature with an addition of three equivalents acetyl cyanide. The obtained ee value at steady state was 85%. The maximum yield, with a value of 70%, was obtained by carrying out the MER process at room temperature with an addition of three equivalents acetyl cyanide over 24 h and a small amount of enzyme (20 mg CALB).

For further optimization of this MER process the change of the amount of added acetyl cyanide up to four equivalents would be interesting to see how it affects ee and yield. Also the change of the relation between the two catalysts, Ti-salen-complex and enzyme, could be another way to optimize the values of ee and yield.
4. Experimental Part

4.1 General Experimental Procedure

The $^1$H-NMR were recorded at 400 MHz or 500 MHz and the $^{13}$C-NMR were recorded at 100 MHz or 125 MHz on a Bruker Avance 400 or a Bruker Avance DMX 500 instrument in CDCl$_3$. The solvent peak was used as internal standard (CDCl$_3$, $^1$H: $\delta = 7.26$ ppm, $^{13}$C: $\delta = 77.36$ ppm) and the chemical shifts were observed in the $\delta$-scale with multiplicity (s = singlet, d = doublet, t = triplet, q = quartet, quint = quintet, m = multiplet), coupling constants in Hz.

The TLC analyses were performed on Merck silica gel 60 F$_{254}$ plates. For visualization anisaldehyde and ninhydrine were used in combination with a heat gun. For column chromatography the silica gel named YMC*Gel Silica 120A S – 50 $\mu$m SL 12S50 was used.

To determine the ee, the GC from Aglient Technologies 6850 was used, containing a column (30m x 0,250 mm x 0,25$\mu$m) with Cyclosil-B as stationary phase and hydrogen and helium in the feed. For the analysis a flow of 2.0 ml/min and the following temperature program (table 2) was used.

**Table 2:** Temperature program for the GC

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<th>Hold [min]</th>
<th>Run [min]</th>
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</tr>
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<td></td>
<td>50</td>
<td>0</td>
<td>48</td>
</tr>
</tbody>
</table>

Reactions, which were air and moisture sensitive, were performed in oven dried, septum-covered flasks under atmospheric pressure of nitrogen and with dry solvents from a dispersing system.
4.2 Compounds

5-Bromo-1-pentanol (2)

Hydrobromic acid (0.5 g, 4.8 mmol, 48% (aq), 1 equiv.) was added to a stirred solution of 1,5-pentanediol (0.81 g, 4.8 mmol, 1 equiv.) in toluene (10 ml). The reaction mixture was heated for 6 h at 80 °C and then stirred for 16 h at room temperature. The reaction mixture was transferred into a separating funnel and washed with brine. After drying the organic phase over MgSO₄ and evaporating the solvent, the product was obtained as transparent oil (0.6 g, 70%).

¹H-NMR (400 MHz, CDCl₃): δ = 3.66 (t, J = 6.2 Hz, 2H); 3.42 (t, J = 6.8 Hz, 2H); 1.89 (quint, J = 7.3 Hz, 2H); 1.65 – 1.48 (m, 4H); 1.33 (s, 1H).

5-Bromo-1-pentanal (3)

To a stirred solution of PCC (2.34 g, 10.84 mmol, 1.2 equiv.) in DCM (46 ml) 5-bromo-1-pentanol (1.51 g, 9.03 mmol, 1 equiv.) was added. The reaction mixture was stirred for 3 h at room temperature. 46 ml diethyl ether was added to the mixture in one portion resulting in a black gum. The black gum was extracted with dry ether until it turns to a black granulate. The combined ether phases were filtered through a short silica gel column with first diethyl ether and then ethyl acetate as eluants. After purification by column chromatography on silica gel (2% EtOAc in n-pentane) and evaporation of the solvent the product was obtained (0.79 g, 53%).

¹H-NMR (400 MHz, CDCl₃): δ = 9.79 (s, 1H); 3.42 (t, J = 6.6 Hz, 2H); 2.50 (t, J = 6.8 Hz, 2H); 1.95 - 1.85 (m, 2H); 1.84 - 1.75 (m, 2H).
**5- Bromo-1-cyanopentyl acetate (rac-4)**

![Structure of 5- Bromo-1-cyanopentyl acetate (rac-4)](image)

Rac-5-Bromo-1-pentanal (0.41 g, 2.49 mmol, 1 equiv.) and AcCN (0.21 g, 2.99 mmol, 1.2 equiv.) were dissolved in DCM (6.5 ml). Triethylamine (0.05 g, 0.49 mmol, 0.2 eq.) was added to the solution and it turned yellow with smoke development. After a total of 6 h the solution was washed with 0.1 M HCl, the organic phase was dried over MgSO₄ and the solvent was evaporated. The product was purified by column chromatography on silica gel (3% EtOAc in n-pentane) (0.29 g, 51%).

$$^1$$H-NMR (400 MHz, CDCl₃): $\delta = 5.34$ (t, $J = 6.4$ Hz, 1H); 3.42 (t, $J = 6.4$ Hz, 2H), 2.10 (s, 3H); 1.93 (quint, $J = 7.5$ Hz, 4H); 1.73 - 1.63 (m, 2H).

**Attempted preparation of 1-Cyano-5-(diethylamino)pentyl acetate (rac-5)**

![Structure of 1-Cyano-5-(diethylamino)pentyl acetate (rac-5)](image)

Rac-5-Bromo-1-cyanopentyl acetate (0.030 g, 0.13 mmol, 1.1 equiv.) was dissolved in acetonitrile (1 ml) under nitrogen atmosphere and diethylamine (0.0121 ml, 0.116 mmol, 1 equiv.) and N-ethyldiisopropylamine (0.031 ml, 0.18 mmol, 1.5 equiv.) were added. The reaction mixture was stirred for 22 h at 40 °C. The solvent was evaporated and the residue was purified by column chromatography (50% EtOAc in n-pentane).
1-Cyano-5-morpholinopentyl acetate (rac-6)

Rac-5-Bromo-1-cyanopentyl acetate (0.10 g, 0.43 mmol, 1 equiv.) was dissolved in acetonitrile (1 ml) under nitrogen atmosphere and two crystals of potassium iodide, morpholine (0.045 ml, 0.52 mmol, 1.2 equiv.) and N-ethyldiisopropylamine (0.11 ml, 0.65 mmol, 1.5 equiv.) were added. The reaction mixture was refluxed for 26 h at 82 °C and stirred for 16 h at rt. The solvent was evaporated and the product was purified by column chromatography (1. 20% EtOAc in n-pentane; 2. 20% EtOAc in n-pentane with 3% TEA; 3. 50% EtOAc in n-pentane with 3% TEA) on silica gel (0.048 g, 0.20 mmol, 47%).

$^1$H-NMR (400 MHz, CDCl$_3$): $\delta$ = 5.34 (t, 1H); 3.71 (m, 4H); 2.42 (s, 4H); 2.34 (s, 2H); 2.15 (s, 3H); 1.93 (s, 2H); 1.55 (s, 4H overlapping with H$_2$O).

$^{13}$C-NMR (400 MHz, CDCl$_3$): $\delta$ = 169.5 (C$_q$, C=O); 117.2 (C$_q$, -CN); 67 – 67.3 (2C, CH$_2$-O-CH$_2$); 61.4 (-CH); 58.7 (N-CH$_2$); 54.1 (2C, CH$_2$-N-CH$_2$); 32.5 (-CH-CH$_2$-); 26.1 (N-CH$_2$-CH$_2$-); 22.8 (N-CH$_2$-CH$_2$-CH$_2$-); 20.8 (-CH$_3$)

Attempt synthesis of (R)-5-(benzylamino)-1-cyanopentyl acetate ((R)-7)

(R)-5-Bromo-1-cyanopentyl acetate (31.79 mg, 0.14 mmol, 1 equiv.) was dissolved in acetonitrile (0.3 ml) under nitrogen atmosphere and two crystals of potassium iodide,
benzylamine (0.015 ml, 0.14 mmol, 1 equiv.) and N-ethyldiisopropylamine (0.035 ml, 0.20 mmol, 1.5 equiv.) were added. The reaction mixture was stirred for 10 h at rt.

**Attempt synthesis of (R)-1-cyano-5(((S)-1-phenylethyl)amino)pentyl acetate ((R)-8)**

(R)-5-Bromo-1-cyanopentyl acetate (34.79 mg, 0.15 mmol, 1 equiv.) was dissolved in acetonitrile (0.3 ml) under nitrogen atmosphere and two crystals of potassium iodide, (S)-(-)-phenylethylamine (0.019 ml, 0.15 mmol, 1 equiv.) and N-ethyldiisopropylamine (0.039 ml, 0.22 mmol, 1.5 equiv.) were added. The reaction mixture was stirred for 23 h at rt. The solvent was evaporated and the residue was purified by column chromatography (1. 10% EtOAc in n-pentane with 3% TEA; 2. 20% EtOAc in n-pentane with 3% TEA; 3. 50% EtOAc in n-pentane with 3% TEA).

**(R)-5-(benzylmethylamino)-1-cyanopentyl acetate ((R)-9)**

(R)-5-Bromo-1-cyanopentyl acetate (30.51 mg, 0.13 mmol, 1 equiv.) was dissolved in acetonitrile (0.3 ml) under nitrogen atmosphere and two crystals of potassium iodide, N-benzylmethylamine (0.024 ml, 0.20 mmol, 1.5 equiv.) and N-ethyldiisopropylamine (0.034 ml, 0.19 mmol, 1.5 equiv.) were added. The reaction mixture was stirred for 44 h at rt. Then the solvent was evaporated and the residue was purified by column chromatography (1. 10% EtOAc in n-pentane with 3% TEA; 2. 20% EtOAc in n-pentane with 3% TEA; 3. 50% EtOAc in n-pentane with 3% TEA).
6-Hydroxy-1-hexanal (11)

ε-Caprolactone (0.5 g, 4.4 mmol, 1 equiv.) was dissolved in THF (4 ml) in a dried, evacuated and with nitrogen refilled round-bottom flask. Then DIBAL-H (3.5 ml, 20.6 mmol, 4.7 equiv.) was added dropwise to the -78 °C cold solution, which turned opaque and more viscous. After 6 h 20 min the reaction was quenched with 10 ml H₂O, warmed up to room temperature and 0.5 ml 0.5 M HCl were added. The reaction mixture was then extracted with DCM. The solvent was evaporated and the product was purified by recrystallization out of ethyl acetate as solvent (0.1 g, 25%).

¹H-NMR (400 MHz, CDCl₃): δ = 9.73 (s, 1H); 3.61 (t, J = 6.4 Hz, 2H); 2.42 (t, J = 7.2 Hz, 2H); 1.67 - 1.43 (m, 6H); 1.22 (t, J = 7 Hz, 1H).

1-Cyanohexane-1,6-diyl diacetate (rac-12a)

6-Hydroxyhexanal (0.12 g, 1.04 mmol, 1 equiv.) was dissolved in DCM (2.6 ml). Then AcCN (0.13 ml, 1.76 mmol, 1.7 equiv.) was added to the cooled solution. Finally NEt₃ (0.03 ml, 0.21 mmol, 0.2 equiv.) was added slowly to the solution. After a total of 74 h 45 min stirring at rt, the reaction mixture was washed with 50 ml 0.1 M HCl. The organic phase was dried over MgSO₄ and the solvent was evaporated. The product was purified by column chromatography (1. 10% EtOAc in n-pentane; 2. 50% EtOAc in n-pentane) (0.06 g, 0.33 mmol, 31%).

¹³C-NMR (500 MHz, CDCl₃): δ = 171.1 (C₉, H₃C-COO-CH₂-); 169.1 (C₉, H₃C-COO-CH-CN); 116.8 (C₉, -CN); 64.2 (O-CH₂-); 60.9 (CN-CH); 32.1 (-CH-CH₂-); 28.4 (O-CH₂-CH₂-); 25.4 (O-CH₂-CH₂-CH₂-); 24.3 (-CH-CH₂-CH₂-); 21 (-CH₃); 20.9 (-CH₃).
5-((Trimethylsilyl)oxy)-pentane-1-ol (14)

1,5-Pentanediol (0.30 g, 2.89 mmol, 1 equiv.) and imidazole (0.39 g, 5.77 mmol, 2 equiv.) were dissolved in DMF (4 ml). The mixture was cooled to 0 °C and TMSCl (0.37 ml, 2.89 mmol, 1 equiv.) was added under nitrogen atmosphere. The reaction mixture was warmed up to room temperature and stirred for 24 h. Then the reaction was quenched with 8 ml of water. The mixture was extracted with ethyl acetate (~12 ml) and the combined organic phases were then washed with brine, dried over MgSO₄ and evaporated.

1,5-((Bistrimethylsilyl)oxy)-pentane-1-ol (14a)

1,5-Pentanediol (0.3 g, 2.89 mmol, 1 equiv.) and NEt₃ (0.80 ml, 5.77 mmol, 2 equiv.) were dissolved in THF (10 ml). The mixture was cooled to 0 °C and TMSCl (0.37 ml, 2.89 mmol, 1 equiv.) was added slowly under nitrogen atmosphere. The reaction mixture was slowly warmed up to room temperature. After 48 h stirring the solvent was evaporated. The white crystals were dissolved in cold water and the liquid was extracted with 20 ml of diethyl ether. The combined organic phases were dried over MgSO₄ and evaporated.

¹H-NMR (400 MHz, CDCl₃): δ = 3.57 (t, J = 6.6 Hz, 4H); 1.53 (m, 2H overlapped with H₂O); 1.44 - 1.19 (m, 4H); 0.11 (s, 18H).

Attemped synthesis of 3-bromo-1-propanal (16)
Synthesis path 1

To a stirred solution of PCC (0.095 g, 0.44 mmol, 1.2 equiv.) in dry DCM (1.5 ml) 3-bromo-1-propanol (0.032 ml, 0.37 mmol, 1 equiv.) was added. The reaction mixture was stirred for 6 h at room temperature. The reaction was followed by TLC and NMR (after 3 h and 6 h). The NMR data showed that no product was formed.

Synthesis path 2

To a stirred solution of DMP (0.116 g, 0.273 mmol, 1.25 equiv.) in dry DCM (2 ml), under nitrogen atmosphere, 3-bromo-1-propanol (0.019 ml, 0.219 mmol, 1 equiv.) was added. The reaction mixture was stirred for 1 h 45 min at room temperature. Then 2 ml saturated Na₂S₂O₃ solution was added to the mixture which was stirred until two phases appeared. The two phases were separated and the organic phase was extracted with DCM. The combined organic phases were washed with saturated NaHCO₃ solution, dried over MgSO₄ and the solvent was evaporated.

Acetyl cyanide

Copper cyanide (7.28 g, 0.081 mol, 1.2 equiv.) was weighed into a round-bottom flask, acetyl bromide (5 ml, 0.068 mol, 1 equiv.) was added and the stirred mixture was refluxed for 4 h at 75 °C. The product was purified by bulb to bulb distillation (3.02 g, 54%).

¹H-NMR (400 MHz, CDCl₃): δ = 2.55 ppm (s, 3H).
The ligand \((R,R)\)-\((-\)N,N\(^{\prime}\)-Bis(3,5-di-tert-butylsalicylidene)-1,2-cyclohexanediamine (0.51 g, 0.93 mmol) was weighed into a round-bottom flask, which was closed with a septum and taken into the glove box. DCM and titanium (IV) chloride (1.3 ml, 11.86 mmol) were added. The reaction mixture was stirred outside the glove box at rt for 4 h. The solvent was evaporated until a brown residue was left, which was then taken up with diethyl ether (~50 ml) and filtered through cotton. The cotton was then washed with DCM (~100 ml) and the resulting red solution was stirred with pH 7 phosphate buffer (~100 ml) at room temperature until it turned yellow. The organic phase was separated, evaporated and dried overnight under vacuum to get the desired product (0.50 g, 45%).

4.3 Minor enantiomer recycling

**Procedure for Lewis – acid – CRL- catalyzed synthesis of O-acetylated cyanohydrins**

Activated CRL (15 mg) and a 1M phosphate buffer pH8 (1 ml) were added to a solution of \((R, R)\)-[(salen)Ti-(\(\mu\)-O)]\(_2\) (0.012 g, 0.010 mmol, 0.05 equiv.), aldehyde 3 (0.033 g, 0.20 mmol, 1 equiv.) and internal standard undecane C\(_{11}\)H\(_{24}\) (10 \(\mu\)l, 0.005 mmol) in toluene (1 ml). Acetyl cyanide (42.7 \(\mu\)l, 0.60 mmol, 3 equiv.) diluted to 0.25 ml with toluene was then added at rt over 6.25 h to the vigorously stirred reaction mixture using a syringe pump. The reaction was followed by taking aliquots from the organic phase (ca. 30 \(\mu\)l, which were filtered through a plug of silica and eluted with dry diethyl ether), which were analyzed with GC.
General procedure for Lewis – acid – CAL-B- catalyzed synthesis of O-acetylated cyanohydrins

Immobilized CAL-B (20 mg) and a 1M phosphate buffer pH8 (1ml) were added to a solution of (S, S) -[(salen)Ti-(μ-O)]2 (0.012 g, 0.011 mmol, 0.05 equiv.), aldehyde 3 (0.033 g, 0.20 mmol, 1 equiv.) and internal standard undecane C11H24 (10 μl, 0.005 mmol) in toluene (1 ml). Acetyl cyanide (3 equiv.) diluted to 0.25 ml with toluene was added at room temperature (or 40 °C) over 6 - 24 h to the vigorously stirred reaction mixture using a syringe pump. The reaction was followed by taking aliquots from the organic phase (ca. 30 μl, which were filtered through a plug of silica and eluted with dry diethyl ether), which were analyzed with GC.

5. References


6. Acknowledgement

First I want to thank Professor Christina Moberg for giving me the chance to be a member of her research group at KTH. It was a pleasure for me to work in that group on a very interesting field of research. At the discussions with the group I learned a lot for my future as chemist.

I’d like to thank also Khalid, Robin and Anna for helping me with any problems and answering every question I had. It was always nice atmosphere in the group, what I enjoyed very much.

Further I want to thank Prof. Dr. Sabine Laschat for making it possible for me to participate at the ERASMUS-program and also I’d like to thank Ursula Henn for the organization.

I want to thank Anna, Lara and Philipp for the funny breaks at university and the great time in Stockholm. Thanks also to the whole “Mariatorget-group”, whose members made the time in Stockholm unforgettable for me.

And last but not least I want to thank my parents who made it possible for me to study abroad and who supported me in everything I did.