Biocatalysis & Multicomponent Reactions: The Ideal Synergy

Asymmetric Synthesis of Substituted Proline Derivatives

The demand for fast, clean, atom and step efficient chemical processes is increasing by the day. Sustainable synthesis is not only a key feature for the chemical industry but also for the pharmaceutical world. The key objective of the research in this thesis is to combine two methodologies that have proven to be efficient and environmentally benign: (i) multicomponent reactions (MCRs) and (ii) biocatalysis. These methods offer significant advantages by reducing time, saving money, energy and raw materials, thus resulting in both economical and environmental benefits. The structurally complex products formed in MCRs often contain new stereocenters which are difficult to control. For most MCRs, catalytic asymmetric methods to control the stereochemical outcome of the reaction are so far not available. The broad repertoire of stereospecific conversions by biocatalysts presents a unique opportunity to address the stereoselectivity issue of certain MCRs.

We have developed several methods (Scheme 1) making use of the enzymatic desymmetrization of meso-pyrrolidines by means of a monoamine oxidase N (MAO-N) from *Aspergillus niger* optimized by directed evolution and its combination with multicomponent chemistry.

**Scheme 1**: Access to different scaffolds and follow-up chemistry using MAO-N and MCRs.
Multicomponent reactions (MCRs) combine essentially all the atoms of at least three components in one pot for atom and process efficiency with reduced waste. MCRs are significantly more efficient than classical multistep synthesis in terms of time and resources. Biocatalysis employs enzymes and microorganisms in the chemical or food industry, making manufacturing processes more environmentally friendly. They can produce high yields of specific products with low energy use and minimal waste generation. Despite the great potential of biocatalysis and MCRs, the combination of these two types of methodologies to generate optically pure complex compounds has hardly been described in literature (Chapter 1).

The main biocatalyst described in this thesis is monoamine oxidase N (MAO-N) from Aspergillus niger. This enzyme has proven to be particularly suitable for directed evolution. Mutant MAO-N tolerates primary, secondary, and tertiary amines as substrates. In addition, Aspergillus niger. This enzyme has proven to be particularly suitable for directed evolution. The main biocatalyst described in this thesis is monoamine oxidase N (MAO-N) from Aspergillus niger. This enzyme has proven to be particularly suitable for directed evolution. Moreover, the stereoselectivity of the biocatalyst is very high (ee > 94%) and no additional cofactor is required. This makes MAO-N one of the leading biocatalysts for amine oxidation (Chapter 2).

Since 3,4-substituted meso-pyrrolidines 1 proved excellent substrates for MAO-N, different optically pure 3,4-disubstituted 1-pyrrolines 2 were generated (Scheme 2). These 3,4-disubstituted 1-pyrrolines offer the possibility to do a subsequent MCR since imines are intermediates for several of these MCRs. 3,4-Disubstituted 1-pyrrolines were reacted with carboxylic acids and isocyanides in a highly diastereoselective Ugi-type multicomponent reaction (Joullié-Ugi 3CR, Scheme 2) to give pharmaceutically relevant substituted prolyl carboxylic acids and isocyanides in a highly diastereoselective Ugi-type multicomponent reaction (Joullié-Ugi 3CR, Scheme 2) to give pharmaceutically relevant substituted prolyl peptides 3 that would require lengthy reaction sequences using other methods. The resulting prolyl peptides can also be used as highly active organocatalysts (4) for stereoselective conjugate addition of aldehydes to nitroolefins (Chapter 3).

![Scheme 2: Desymmetrization and diastereoselective Ugi-type 3CR towards optically pure 3,4-substituted prolyl peptides](image)
We envisioned that combining the diastereoselective Joullié-Ugi 3CR approach with cyclization reactions could further increase the resulting molecular complexity and diversity considerably. Reacting our 3,4-disubstituted 1-pyrrolines with α-ketocarboxylic acids and homoveratryl isocyanide afforded the ketoamide intermediate 5 which subsequently undergoes a Pictet–Spengler cyclization to afford highly complex alkaloid-like polycyclic compounds (2,5-diketopiperazines; DKPs) 6 with high diversity (Scheme 3). In addition, the experimental procedure is simple and very efficient. To the best of our knowledge, it also constitutes the first example of MCR chemistry to synthesize 5-membered ring-fused DKPs (Chapter 4).

**Scheme 3**: Diastereoselective Ugi-type 3CR followed by a Pictet-Spengler reaction resulting in DKPs 6.

The efficiency and generality of the approach and the resulting molecular diversity and complexity makes our methodology highly interesting for medicinal chemistry. Most notably, a very short and efficient synthesis of important hepatitis C drug Telaprevir (Incivek™), featuring a biocatalytic desymmetrization and two multicomponent reactions (Passerini and Joullié-Ugi 3CR) as the key steps was developed. The classical issue of lack of stereoselectivity in Ugi- and Passerini-type reactions was circumvented. The atom economic and convergent nature of the synthetic strategy requires only very limited use of protective groups. The use of protective groups is limited to two intermediate methyl esters and an acetate. The use of carbamate protective groups is avoided altogether. The synthesis comprises only eleven steps in total (seven steps in the longest linear sequence) compared to twenty-four in the originally reported procedure. The total yield (over the longest linear sequence) is 45% starting from L-cyclohexylglycine methyl ester. Our approach is general and will be applicable to many other HCV NS3 protease inhibitors that can be derived from meso-pyrrolidines, such as e.g. Boceprevir (Victrelis™) and Narlaprevir. The combination of synthetic efficiency and convergence in our approach allows both faster development of second generation inhibitors and a more economical production of Telaprevir (Chapter 5).
We then decided to investigate the possibility of reacting our 3,4-disubstituted 1-pyrrolines in other MCRs. The Ugi-Smiles reaction, a variation of the Ugi reaction in which the carboxylic acid component is substituted by an electron-deficient phenol derivative (or a heteroaromatic analog) is a promising candidate for combination with our biocatalytic oxidation due to its mechanistic similarity with the Ugi reaction. Reacting our 3,4-disubstituted 1-pyrrolines with electron-deficient phenols and different isocyanides afforded the N-aryl proline (thio) amides 7 in high diastereoselectivity (Scheme 4). These 3,4-substituted N-aryl proline (thio) amides are pharmaceutically relevant since similar compounds have been reported to be potent VLA-4 antagonists. Therefore, these compounds may be useful in the treatment of diseases like asthma, atherosclerosis, rheumatoid arthritis, inflammatory bowel disease, and multiple sclerosis. This method represents the first report of a fully asymmetric Ugi-Smiles process (Chapter 6).

In summary, the results described in this thesis demonstrate that biocatalysis and MCRs is a powerful combination for the production of optically pure complex compounds. We were able to synthesize two distinct scaffolds; N-aryl proline amides and 3,4-disubstituted prolyl peptides. The latter showed to be a very versatile scaffold providing access to organocatalysts, synthetic alkaloids and even to the important Hepatitis C drug Telaprevir (Incivek™).