**Dynamic Kinetic Resolution using Enzymes**

In the ideal chemical reaction the product obtained would be completely pure in 100% yield with all the materials of the reaction incorporated. However this can be a challenge to accomplish as starting materials can be racemic along with the products produced. Diagram 1 shows an example of a racemic mixture with the enantiomers (denoted as R or S) each with an asymmetric carbon atom (chiral) that are non-superimposable on the other. These compounds are usually hard to separate due to the similarities in physical and chemical properties due to them only differing by the spatial arrangement. Though this may seem innocuous even a slight difference in the spatial arrangement of the compound can have profound effects in regards to its function.



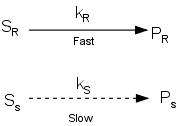
*Diagram 1: Example of a racemic mixture*

*Note R1 ≠ R2*

Enzymes and receptors are also chiral so recognise one enantiomer over the other. Here-in lies the main problem, especially in the pharmaceutical industry, as when a racemic medicine is administered the other enantiomer can cause unwanted side effects elsewhere [ref1]. Since natural products tend to be chiral [ref3] this brings a problem which needs to be overcome as enatiomerically pure compounds are now a standard requirement for drugs [ref3], so a way of separating the enantiomers is required, meaning new pathways to chiral compounds have to be devised.

Resolution is the separating of enantiomers by chemical or physical means which is usually achieved via conversion to diastereomers which then subsequently can be separated due to different chemical properties. From this various techniques have been devised, this essay focuses on dynamic kinetic resolution with enzymes but will briefly cover others.

Kinetic Resolution



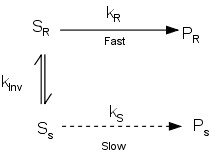
*Diagram 2: Overview of classic kinetic resolution*

Kinetic resolution relies on one enantiomer being kinetically favourable in the asymmetric transformation leading to the desired product being formed while leaving the other enantiomer unreacted. This relies on kR >>> kS to achieve a high enantiomeric excess (ee) and a theoretical yield of 50% though as the conversion reaches near 50% the ee starts to fall (see diagram 3)[ref4]. Another problem is that the unreacted enantiomer and the desired product need to be successfully separated.

*Diagram 3: ee graph/ thermodynamics*

If the Isolated unreacted enantiomer can undergo racemization affording the wanted enantiomer the racemic mixture produced can be subjected to another stage of kinetic resolution and assuming the theoretical yield of 50% is obtained in each stage, by the 5th stage a 97% yield can be achieved [ref5] , however this increases the time and cost so is impractical after the 2nd stage. More practical would to be to combine the two processes into a “dynamic” reaction and thus dynamic kinetic resolution was devised. [ref6]

Dynamic Kinetic Resolution



*Diagram 4: Overview of Dynamic Kinetic Resolution (DKR)*

Dynamic kinetic resolution (DKR) is a kinetic resolution with an additional feature: the enantiomers are being inverted in-situ and thus are in an equilibrium with each other. The fast conversion of one enantiomer into the desired product causes a depletion which shifts the equilibrium in favour of producing more of the faster reacting enantiomer resulting in a skewed reaction making higher yields achievable, theoretically 100%. Also importantly this technique achieves high ee as there is always a racemic mixture present so the asymmetric catalyst isn’t in direct contact with only the undesired enantiomer.

In essence there are two processes going on at once: racemization of the enantiomers and the conversion of one enantiomer into the desired product that need to be fine-tuned as there compatibility is vital. There are ideal conditions that have to be met for a successful DKR including those for a kinetic resolution and avoiding side or spontaneous reactions [ref 5]. The resolution reaction should be irreversible thereby preventing a drop in ee and maintain the shift in equilibrium. The rate of inversion (kInv) needs to be greater than or equal to the rate of the asymmetric conversion (kR) avoiding the supply of the wanted enantiomer depleting thus making the unwanted enantiomer more favourable. High selectivity in the catalyst is desired otherwise the rate of inversion needs to be significantly higher than the rate of conversion.

Racemization

Racemization is arguably the most important aspect of a DKR as its optimization is essential for a DKR to run efficiently. Certain conditions have to be met to induce this which can lead to compatibility problems with the rest of the DKR. Examples of this are specific pH and temperature requirements add in example [ref7]. Therefore this step needs to be tuned affording a balance that maintains an effective inversion but doesn’t hinder the conversion to the desired product and the use of a catalyst in this step keeps the conditions in an acceptable range. Racemization can be accomplished by various methods such as acid or base catalysis but this role can be accomplished by enzymes such as dehydrogenase [ref8].

Biocatalyic Asymmetric conversion- Why Enzymes for DKR?

Chiral asymmetric catalysts based upon transition metals are commonly used to in both the racemization and conversion steps of a DKR but have to be synthesised and usually require a rare metal at its core, most commonly ruthenium is used [ref9]. Enzymes can offer a more attractive alternative.

Enzymes are becoming more viable as tools in organic synthesis especially as the direction of chemistry is moving towards being more responsible [ref10] and enzymes can provide alternative pathways to desired compounds. Enzymes are more environmentally friendly than metal based catalysts as they are biodegradable. In terms of efficiency enzymes can increase reaction rates magnitudes higher [ref11] than there non-bio counterparts and be used in lower quantities.

Enzymes typically act under milder conditions such as low temperature (~ 30⁰C) and near neutral pH. This meansthat reactions can be untaken in ambient conditions avoiding the need for specialized machinery needed to incorporate extreme conditions. Another benefit of this is that it avoids side reactions which would be able to take place at higher temperatures and pressures such as rearrangements, isomerisim and racemization

Enzymes were commonly dismissed as only working in non-aqueous media [ref12] however this is not the case and can even work to temperatures around 70⁰C and beyond in organic solvents [ref13]. Thus allowing for organic substrates that would not dissolve successfully in water to interact with the enzyme. Enzymes can perform in the presence of others [ref14], if the conditions required are similar for both, leading to one pot reactions where multiple enzyme-catalysed reactions are feasible. Enzymes have been shown to be able to carry out reactions on synthesised substances opening their usefulness as they are not limited to natural substrates and can perform a wide variety of transformations.

This is not to say there are not drawbacks to the use of enzymes which need to be considered. The biggest one being that they

Lipases in dkr

One of the most commonly used enzymes in a DKR are lipases which show versatility in being able to catalysed an array of different reactions.

*Synthesis of Esters (esterification)*

*Synthesis of Amides*

*Synthesis of Acids (hydrolysis)*

Other enzymes and reactions carried out under dkr

Bakers yeast

Nitrilase

Baeyer-Villiger Monooxygenase

Etc

Recent developments (2013-2012)

Conclusions and outlook

References