Expanding the Synthetic Toolbox through Metal-Enzyme Cascade Reactions

Sergio González-Granda, Jesús Albarrán-Velo, Iván Lavandera and Vicente Gotor-Fernández**

Organic and Inorganic Chemistry Department, Universidad de Oviedo, 33006 Oviedo (Asturias, Spain).

E-mail: lavanderaivan@uniovi.es (I.L.); vicgotfer@uniovi.es (V.G.-F.)

Abstract

Combination of metal-, photo-, enzyme- and/or organocatalysis provides multiple synthetic solutions, especially when the creation of chiral centers is involved. Historically, enzymes and transition metal species have been exploited simultaneously through dynamic kinetic resolutions of racemates. However, more recently, linear cascades have appeared as elegant solutions for the preparation of valuable organic molecules combining multiple bioprocesses and metal-catalyzed transformations. Many advantages are derived from this symbiosis, although there are still bottlenecks to be addressed including the successful coexistence of both catalyst types, the need for compatible reaction media and mild conditions, or the minimization of cross-reactivities. Therefore, solutions are here also provided by means of catalyst co-immobilization, compartmentalization strategies, flow chemistry, etc. A comprehensive review is presented focusing on the period 2015 to early 2022, which has been divided into two main sections that comprise first the use of metals and enzymes as independent catalysts but working in an orchestral or sequential manner, and later their application as bionanohybrid materials through their co-immobilization in adequate supports. Each part has been classified into different subheadings, the first part based on the reaction catalyzed by the metal catalyst, while the development of non-asymmetric or stereoselective processes was considered for the bionanohybrid section.

Table of contents

1. Introduction

Metal catalysis is a mature technology for the study of metal-carbon bond formation reactions, currently finding numerous practical applications in the industrial sector.[1] In fact, organometallic chemistry provides straightforward solutions for a myriad of transformations, including cross-coupling[2] and olefin metathesis reactions among the most outstanding examples.[3] In this field, multiple organometallic complexes and catalysts belong to the synthetic chemist toolbox, finding compounds from copper, iron, palladium, platinum, ruthenium, and zinc as the traditional choices, while others from gold, manganese, and nickel have busted in the last decades as suitable and efficient catalysts. Currently, merging different catalytic processes for increasing molecular complexity is receiving great attention, metal catalysis presenting itself as a proper partner to be combined with photoredox[4] and biocatalytic transformations.[5-7]

On the other hand, Biocatalysis is nowadays considered a consolidated synthetic methodology, especially to obtain optically active products.[8-11] Undoubtedly, the development of biotransformations has exponentially grown in the last decades because of the discovery of new enzymatic reactions and catalyst sources,[12-15] but also due to the possibility of modifying enzyme amino acid sequences by applying molecular biology techniques. Thus, novel enzymatic activities can be found at the same time that biocatalyst preparations display high levels of stability in different solvents at wide pH and temperature ranges, excellent (stereo)selectivity, and even broader substrate specificities.[16-18] Interestingly, advances in directed evolution techniques and bioinformatics have allowed the enzyme modification by generating extensive libraries of engineered enzymes, also resulting in some cases with highly improved catalytic activities.[19,20] Consequently, the potential of enzymes in organic synthesis has exponentially grown up in recent years, especially since the enzyme action can be combined with other catalytic processes (metallo-, photo- and organocatalysis),[5-7,21-27] but also in conjunction with more recent sustainable methodologies including, e.g. microwave irradiation[28,29] or flow chemistry.[30-32]

Nowadays the combination of metal and enzyme catalysis for the development of multi-step and concurrent processes turns out to be a great advance for synthetic chemists, but, at the same time, an enormous challenge (Table 1). On the one hand, a few hurdles must

be overcome, among others: (i) avoid (bio)catalyst inactivation; (ii) achieve the compatibility of all catalysts in the same reaction medium (especially solvent type, pH, and temperature); (iii) minimize cross-reactivity and by-product formation; (iv) drive the equilibrium towards product formation.[33] On the other hand, the main advantages of merging both worlds reside in the possibility to increase molecular complexity in a straightforward manner and under mild reaction conditions. This translates into a series of benefits including the suppression of intermediate isolation(s), which improves the overall process in terms of yield and economy. Therefore, to achieve a robust multicatalytic system, individual reaction optimizations and a deep study of the compatibility of every step will be required, prior to developing downstream processing operations and exploring the scope of a proposed methodology.

Table 1. *Advantages and disadvantages of combining metal and enzyme catalysis in cascade reactions.*

Advantages	Disadvantages
Avoid isolation and purification of reaction intermediates	Metal and enzyme catalysts need to be (partially) compatible under determined reaction conditions
Design high-yielding processes Simplify reaction set-ups and operational steps, reducing costs, wastes, energy, and time	Individual reaction optimization is required Mutual inactivation between (bio)catalysts and auxiliary reagents can exist
Shift the equilibrium towards product formation, avoiding inhibition effects and unstable intermediates	The best scenario is probably not achieved for both catalysts at the same time (solvent, temperature, substrate concentration…)
Water can be used as a predominant environmentally friendly solvent	Necessity of selective processes to avoid catalyst inhibition due to the presence of side-products
Develop novel strategies to make compatible enzyme(s) and metal(s) (co-immobilization, compartmentalization…)	Higher level of sophistication may be necessary depending on the reaction set-up
Tuning enzymes and metal complexes to increase their activity and selectivity	Downstream process can be more difficult due to the different catalyst sources

From an operational point of view, the most elegant and simple manner to achieve these combinations is adding all catalysts and reagents in the same reaction medium from the beginning, achieving a concurrent cascade protocol. In fact, some of the examples described in this contribution will necessarily involve this simultaneous catalysis to find a correct development (e.g., dynamic kinetic resolutions). However, due to the difficulties that these approaches present, as shown in Table 1, in other cases the use of one-pot multi-

step sequential protocols, where the addition of new catalyst(s) and/or reagent(s) occurs at different temporal stages, will be easier for a correct process outcome.

The aim of this contribution is to provide the reader with a description of the main benefits of combining both catalytic approaches toward the development of concurrent processes in a synergistic manner. Obviously, optimization of each individual process is a key item to achieve a global successful method, but our efforts will focus on the identification of the pros and cons of the multicatalytic system rather than deeply analyze their single steps. For that reason, recent examples from the last seven years (2015 onwards) have been collected, analyzed, and discussed. Therefore, this review has been divided into two main sections that comprise first the use of metal and enzyme as independent catalysts but working in an orchestral manner, and later their use as nanohybrid materials through the co-immobilization of both catalyst types in an adequate support. At the same time, each part has been classified with different headings based on the reaction catalyzed by the metal catalyst. Without any doubt, a deep understanding of several metal-enzyme combinations puts on stage the problems associated to their simultaneous use, and at the same time the possible solutions given such as multiple catalyst compartmentalization or the development of sequential processes, among other possibilities.

2. Metal-enzyme cascades using individual catalysts

2.1. Racemization to design chemoenzymatic dynamic kinetic resolutions

Historically, metal-enzyme systems have been used to develop efficient stereoselective dynamic kinetic resolutions (DKRs), in which the enzyme, generally a lipase, carries out the selective transformation while the metal catalyst takes over the racemization of the slow-reacting enantiomer (Figure 1).[34,35] The easiest way to carry out these asymmetric transformations involves the use of lipases in organic and neoteric solvents, thus favoring the solubilization of the metal catalyst. Until 2015, a few interesting reviews appeared in the literature,[36-38] appearing very recently a new one highlighting the possibility to develop multienzymatic and chemoenzymatic DKRs, also including novel approaches through photocatalytic transformations.[39]

Herein, we update this topic with recent advances in the field including examples of DKRs of racemic alcohols and amines where the metal catalyzes the racemization of the unreacted enantiomers. Especial attention will be devoted to the most recent strategies focused on the development of DKR under flow conditions, while in Section 3.2, the potential of bionanohybrids formed by co-immobilization of a lipase and a metal species will be considered to give additional insights in the development of chemoenzymatic DKR processes.

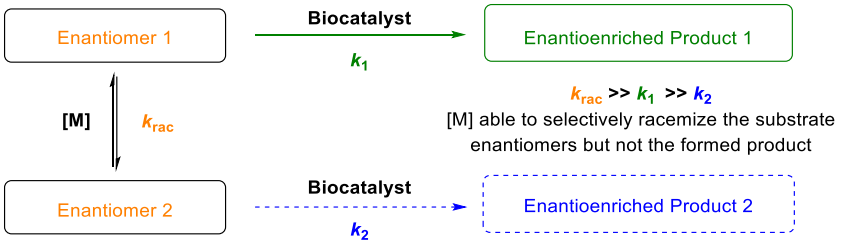
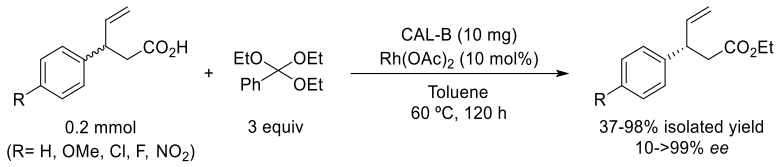


Figure 1. Metal-induced racemization and selective transformation by a biocatalyst.

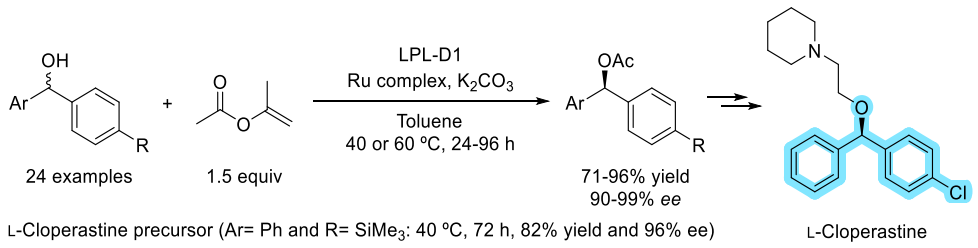
In 2016, Ostaszewski and co-workers reported the first DKR of unsaturated carboxylic acids through the enzymatic esterification of 3-arylpent-4-enoic acids catalyzed by *Candida antarctica* lipase B (CAL-B) under its brand name Novozym 435, combined with a metal-catalyzed racemization employing rhodium(II) acetate ($Rh(OAc)_2$).[40] After optimization of reaction conditions of both individual processes, the lipase-catalyzed KR and the racemization mediated by the metallic species, the DKR of various *para*-substituted unsaturated carboxylic acids was reported on a 0.2 mmol scale using 10 mg of CAL-B (Novozym 435) and 10 mol% of $Rh(OAc)_2$ (Scheme 1). The best results were found employing 3 equiv of the triethyl orthobenzoate, which turned out to be the best ethoxy donor, leading to the corresponding (*S*)-ethyl esters, after a long reaction time (120 h) in toluene at 60 °C, in low to excellent isolated yields after column chromatography purification (37-98%) and variable enantioselectivities (10->99% *ee*). Remarkably, the DKR of a valuable precursor of Femoxetine and Citrocard drugs such as racemic 3-phenylpent-4-enoic acid (R= H) was successfully achieved in a preparative scale (1 mmol), furnishing ethyl enantiopure (*S*)-3-phenylpent-4-enoate in 97% isolated yield.



Scheme 1. DKR of 3-arylpent-4-enoic acids employing a Rh catalyst and CAL-B.

2.1.1. Ruthenium catalysts for the DKR of alcohols

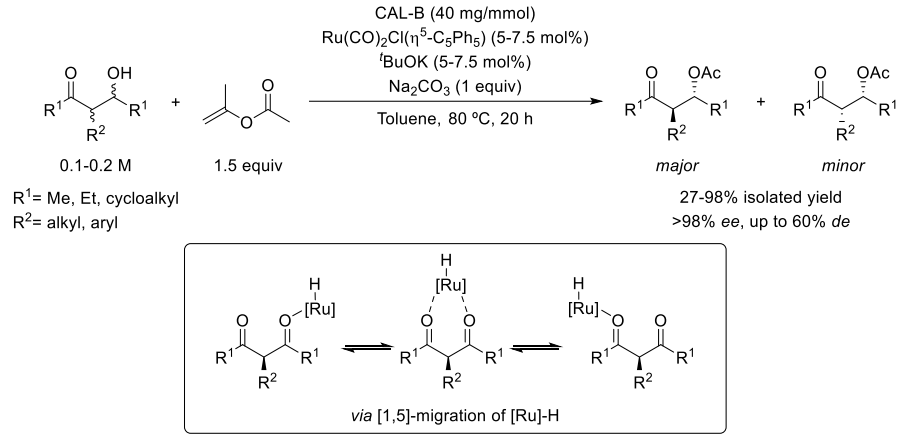
Traditionally, ruthenium complexes have been the racemization agents of choice for the development of DKR protocols involving lipases, including for instance the one described for the chemoenzymatic synthesis of the antitussive drug L-Cloperastine (Scheme 2).[41] The DKR of 24 diarylmethanols was reported using the lipoprotein lipase from *Burkholderia species* (LPL-D1), isopropenyl acetate (1.5 equiv), [{η5-$Ar_4C_4COC(=O)Ar$}$Ru(CO)_2Cl$] with Ar=4-methoxyphenyl (4 mol%), potassium carbonate (K_2CO_3, 1 equiv) and toluene, leading to the diarylmethyl acetates in high to excellent yields (71-96%) and very high to excellent selectivities (90-99% *ee*). Remarkably, the coating of the lipase with a dextrin and an ionic surfactant provided the biocatalyst with a value of about 3000-fold more activity than the native enzyme. Using this methodology, a L-Cloperastine precursor, (*R*)-phenyl[4-(trimethylsilyl)phenyl]methyl acetate was isolated in 82% yield and 96% *ee* after 72 h at 40 ºC, and was later transformed into the final drug in three additional steps.



Scheme 2. DKR of racemic diarylmethanols including the chemoenzymatic synthesis of L-Cloperastine.

Bäckvall and co-workers have recently described the use of $Ru(CO)_2Cl(\eta^5\text{-}C_5Ph_5)$ and lipase B from *Candida antarctica* to achieve the dynamic kinetic asymmetric

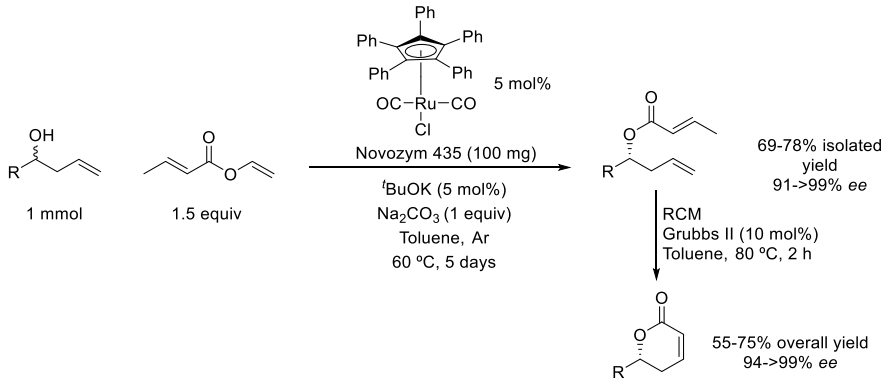
transformation (DYKAT) on α-substituted β-hydroxy ketones (Scheme 3).[42] In this case, due to the presence at the molecule of two stereocenters, up to 4 possible diastereoisomers could be attained. The key to the success of this methodology was the formation of a symmetric diketone intermediate, which in the reaction medium could epimerize in the presence of the Ru catalyst via [1,5]-hydride migration, obtaining the *anti* isomers as the main products (up to 60% *de*). Hence, after optimization, the ruthenium catalyst (5-7.5 mol%), CAL-B (40 mg/mmol), ^{t}BuOK (5-7.5 mol%) and Na_2CO_3 (1 equiv) reacted with the racemic β-hydroxy ketones (0.1-0.2 M) and isopropenyl acetate (1.5 equiv) in toluene under argon atmosphere at 80 ºC. After 20 h, the acetylated products were isolated (27-98%) with excellent enantiomeric excess (>98%) in most cases.



Scheme 3. DYKAT of α-substituted β-hydroxy ketones combining Ru and hydrolase chemistries.

Another interesting example using ruthenium-catalyzed racemization for the synthesis of enantiomerically pure 5,6-dihydropyran-2-ones via a chemoenzymatic sequential DKR-ring-closing metathesis (RCM) reaction is highlighted.[43] The method consists in a two-step approach, the first one based on the DKR of racemic homoallyl alcohols using a ruthenium catalyst and a lipase in the presence of vinyl crotonate as acyl donor, then after filtration of the resulting optically active ester in a silica pad and solvent evaporation, a RCM reaction was carried out using a Grubbs second generation catalyst (Scheme 4). For the first step and after a screening of different lipases, CAL-B (Novozym 435) was found as the most promising enzyme. A ruthenium catalyst [$Ru(CO)_2Cl(\eta^5\text{-}C_5Ph_5)$] was employed to induce the racemization of the homoallylic alcohol, finding that the

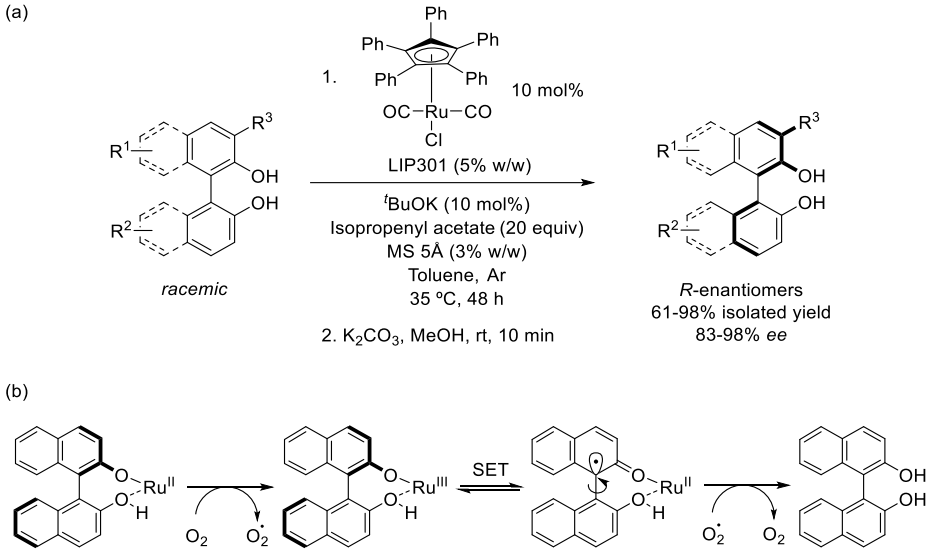
coordination of the catalyst to the double bond of the starting alcohol in the alkoxide intermediate occurs, thus blocking the β-hydride elimination. In this manner, the DKRs of different homoallylic alcohols (1 mmol) were achieved with moderate yields (69-78%) and high enantiomeric excess (91->99%) after prolonged reaction times (3 days) and using Novozym 435 (100 mg), vinyl crotonate (1.5 equiv), the ruthenium catalyst (5 mol%), ^{t}BuOK (5 mol%) and Na_2CO_3 (1 equiv) in dry toluene under inert atmosphere. Next, the stepwise RCM reaction of the obtained (*R*)-esters was described using Grubbs second generation catalyst (10 mol%) for 2 h at 80 ºC. The development of this type of metal-catalyzed metathesis reactions will be fully discussed in Section 2.5.



Scheme 4. Synthesis of enantioenriched 5,6-dihydropyran-2-ones via a stepwise DKR-RCM approach using CAL-B and ruthenium catalysts.

Akai and co-workers showed that axially chiral 2,2'-dihydroxy-1,1'-biaryls could be resolved using a lipase and then, in combination with a ruthenium catalyst, the monoacylated derivatives could be obtained at high extent and enantiomeric excess under dynamic conditions (Scheme 5a).[44] After reaction optimization, commercial immobilized *Pseudomonas* sp. lipase (LIP301, 5% w/w) and $[Ru(CO)_2Cl(\eta^5\text{-}C_5Ph_5)]$ (10 mol%) were selected as the best catalysts to achieve the DKR of 16 substrates in the presence of a base (^{t}BuOK, 10 mol%), molecular sieves (3% w/w), and isopropenyl acetate (20 equiv) as acylating agent in toluene at 35 ºC. After 48 h of reaction and subsequent ester methanolysis, the corresponding (*R*)-diols were obtained with good to excellent isolated yields (61-98%) and *ee* values (83-98%). The mechanism of the racemization in this case did not proceed through the typical transfer hydrogenation pathway, but via single

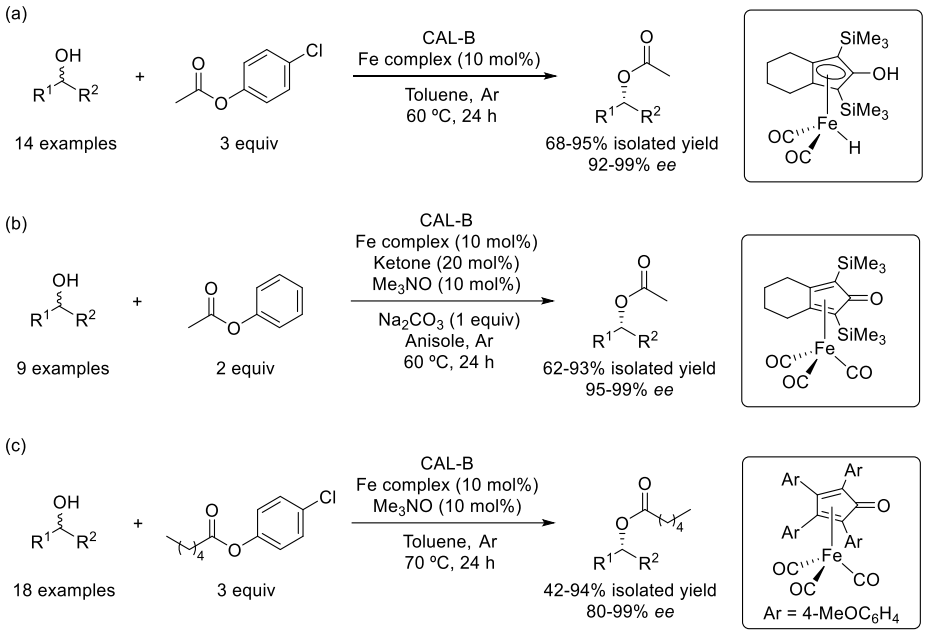
electron transfer (SET) in a Ru(III) complex, forming a radical species bearing an sp^3 carbon at the C1 position, which enabled free rotation around the chiral axis (Scheme 5b).



Scheme 5. (a) Synthesis of axially chiral biaryls through DKR using a lipase and a ruthenium catalyst. (b) Plausible racemization mechanism of the substrates.

2.1.2. Iron catalysts for the DKR of alcohols

The use of iron species has also been studied when developing sustainable lipase-catalyzed DKRs, as described by Rueping and co-workers that studied the behavior of eight different iron complexes, all of them able to racemize aromatic and aliphatic secondary alcohols through a dehydrogenation-hydrogenation process.[45] Merging CAL-B with an air- and moisture-sensitive, but highly active iron complex (10 mol%), it was possible to furnish a series of 14 (hetero)aromatic and aliphatic chiral (*R*)-acetates in 68-95% isolated yield after column chromatography and 92-99% *ee* in toluene at 60 ºC after 24 h under inert atmosphere (Scheme 6a).



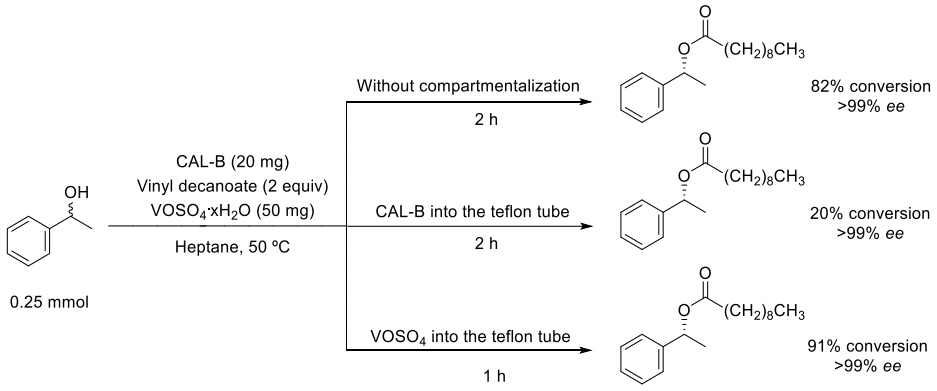
Scheme 6. DKRs of racemic secondary alcohols using iron complexes and CAL-B.

Later, Bäckvall and co-workers developed the DKR of a series of *sec*-alcohols but employing now a precatalyst of the previously described iron complex, that importantly was more stable (Scheme 6b).[46] In this particular system, the addition of a small quantity of the corresponding ketone intermediate (20 mol%), proved to accelerate the racemization rate, while the addition of Me_3NO (10 mol%) as an activator was also convenient. Thus, under these conditions and using CAL-B in anisole at 60 ºC, different O-acetylated (*R*)-alcohols were attained in good to excellent yields (62-93%) and enantiomeric excess (95-99%) after 24 h. Finally, Zhou *et al.* have also described the successful application of another stable iron complex to perform the DKR of different racemic alcohols in the presence of the same lipase at 70 ºC in toluene (Scheme 6c).[47]

2.1.3. Vanadium catalysts for the DKR of alcohols

Exploring additional metal racemization agents, the use of oxovanadium catalysts in combination with lipases has been extensively studied in recent years for the DKRs of racemic secondary alcohols.[48,49] The racemization induced by these types of catalysts follows an addition-elimination mechanism through the formation of a carbocation, thus providing in some cases rearrangement processes simultaneously.

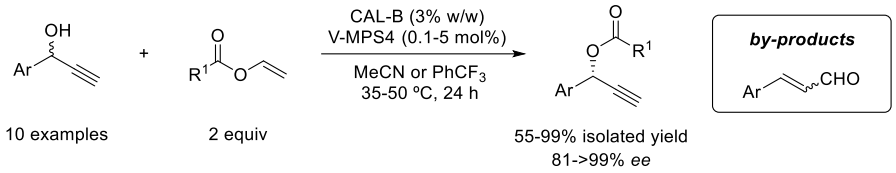
Milagre and co-workers described the combination of CAL-B and $VOSO_4$ for the DKR of 1-phenylethanol and derivatives.[50] Once found heptane and vinyl decanoate as suitable solvent and acyl donor, respectively, up to 82% conversion into the corresponding enantiopure (*R*)-ester was achieved after 2 h at 50 ºC using a 62.5 mM substrate concentration (Scheme 7). The approach resulted to be general for the DKR of ten (het)aromatic secondary alcohols (73-91% isolated yield, 89->99% *ee*, 0.5-2 h). After these results, the authors explored the possibility of compartmentalizing the catalysts in Teflon tubes with micro holes placing only one catalyst inside the tube to prevent physical contact between both, that could lead to their mutual inactivation. Firstly, the compartmentalization of CAL-B led to only a 20% conversion probably caused due to the poor mass transfer of the substrate inside the tube, while with the compartmentalized $VOSO_4$ a 91% conversion into (*R*)-1-phenylethyl decanoate was reached with 98% selectivity and >99% *ee*. These results prove the importance of the physical separation of the heterogeneous catalysts system, allowing the recyclability of the CAL-B-$VOSO_4$ system to achieve good results even after 8 cycles (87% conversion and 99% *ee*).



Scheme 7. DKR of *rac*-1-phenylethanol catalyzed by the CAL-B/$VOSO_4$ system.

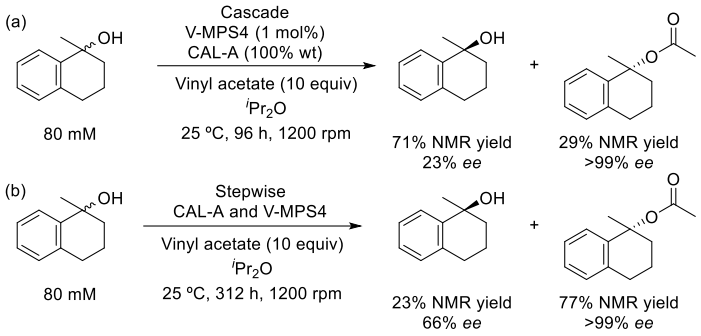
The use of these catalysts has also been successfully applied to achieve the DKR over aromatic propargylic alcohols (Scheme 8).[51] In this particular case, when oxovanadium moieties were covalently immobilized in the inner surface of mesoporous silica with a pore of 4 nm (V-MPS4), it was observed that this material was able to racemize the alcohol substrates. However, it was noticed that the corresponding α,β-unsaturated aldehydes were obtained as by-products via Meyer-Schuster rearrangement, and that the quantities of these by-products were highly dependent on the reaction solvent. Therefore,

after careful optimization and in the presence of CAL-B, it was possible to develop the desired DKRs of 10 aromatic substrates minimizing the formation of the undesired carbonyl compounds (up to 18%). It must be highlighted that the conditions depended greatly on the alcohol substrate, thus being necessary to vary the solvent (MeCN or $PhCF_3$), the temperature (35 or 50 ºC), the acylating agent (vinyl butyrate or vinyl decanoate), and the amount of the metal catalyst (0.1 to 5 mol%). Hence, (*R*)-acylated derivatives were synthesized in moderate to excellent isolated yields (55-99%) and *ee* values (81->99%).



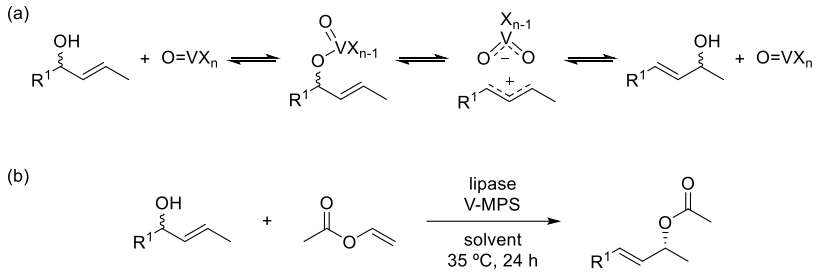
Scheme 8. DKR of different racemic propargylic alcohols catalyzed by CAL-B and immobilized oxovanadium species.

Despite the plethora of examples for the DKRs of secondary alcohols, the application to tertiary alcohols is still in its infancy. Remarkably, Akai, Gröger and co-workers have recently described the use of the oxovanadium catalyst $[VO(OSiPh_3)_3]$ immobilized on mesoporous silica (V-MPS4) and the *Candida antarctica* lipase type A (CAL-A) for the performance of the first example of DKR over these highly hindered substrates.[52] In fact, the reactivity of racemic 1-methyl-1,2,3,4-tetrahydronaphthalen-1-ol with vinyl acetate was exhaustively analyzed considering different parameters such as the catalyst type, temperature and solvent, finding the occurrence of the DKR process, unfortunately, the (*R*)-acetate was isolated only in moderate 29% conversion although in enantiopure form. Overall, the reaction required the addition of fresh CAL-A and acyl donor in a sequential manner because enzyme deactivation was rapidly observed over the time. Under optimized conditions, and after filtering the mixture and continuing with the reaction crude with new addition of the reactants, a 77% of the (*R*)-1-methyl-1,2,3,4-tetrahydronaphthalen-1-yl acetate was found after 312 h in total using 10 equiv of vinyl acetate in diisopropyl ether ($^iPr_2O$) and at low temperature of 25 ºC (Scheme 9).



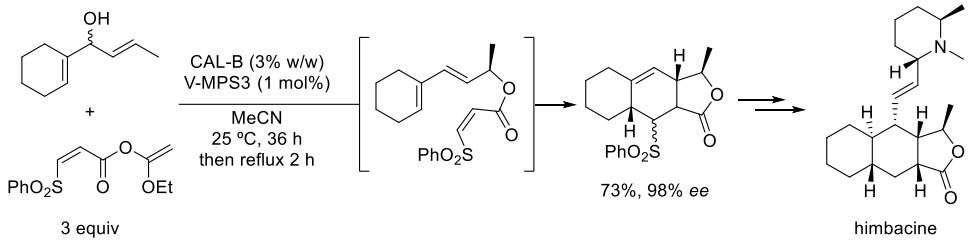
Scheme 9. DKR of a tertiary alcohol by using the V-MPS4/CAL-A system in: (a) cascade mode; (b) stepwise fashion.

As previously mentioned, these catalysts achieve the alcohol racemization through formation of a carbocation intermediate and a vanadate species, which can further evolve via rearrangement processes. For instance, in the case of allylic alcohols, it is well documented the 1,3-transposition of the hydroxy group (Scheme 10a). This particular reactivity can be combined with the action of a lipase to perform DKR processes.[48,49] In this field, Akai's group has been especially active. In a recent contribution, a study of the pore size of the mesoporous silica that acted as support of the oxovanadium catalyst (V-MPS) was accomplished to resolve a series of allylic alcohols combined with CAL-B or lipase from *Burkholderia cepacia* (Scheme 10b).[53] Authors demonstrated that there was a correlation between the pore size of V-MPS and the substrate racemization rate. As higher the polarity of the MPS pores was, more acceleration in the racemization was attained. This fact was attributed to the faster C–O bond cleavage of the vanadate intermediate and stabilization of the carbocation formed. Small pore sizes (2–4 nm) efficiently generated this polar environment inside the pores, while macroporous silica (100–400 nm) did not provide this effect.



Scheme 10. (a) Racemization mechanism of allylic alcohols in the presence of oxovanadium species. (b) Application to the DKR of different racemic secondary allylic alcohols via combination with a lipase.

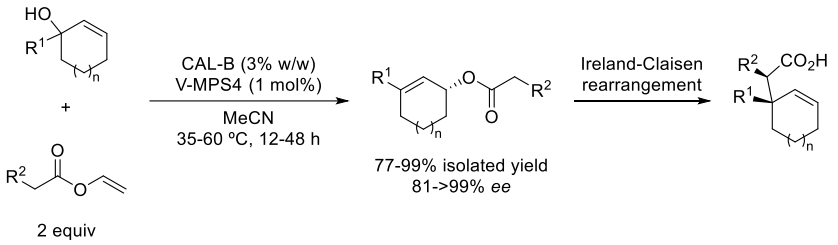
This system has been applied to the synthesis of valuable compounds such as himbacine, an alkaloid with potential applications on Alzheimer's disease. The key step in the proposed synthetic route was the DKR of 1-(1-cyclohexenyl)-2-buten-1-ol (100 mg) in the presence of CAL-B (3% w/w), an immobilized oxovanadium catalyst (V-MPS3, 1 mol%) and an α,β-unsaturated acyl donor (3 equiv), which underwent an spontaneous intramolecular Diels-Alder cycloaddition (Scheme 11).[54] The reaction was performed in acetonitrile for 36 h at 25 ºC, and then refluxed for additional 2 h, providing the desired tricyclic derivative in good isolated yield (73%) and excellent *ee* (98%), although as a diastereomeric mixture of compounds. This intermediate, after few reaction steps, provided himbacine in 6% overall yield.



Scheme 11. Lipase/oxovanadium DKR applied to the synthesis of an intermediate of himbacine.

Due to the particular racemization mechanism using oxovanadium species, it is possible to achieve DKRs on tertiary alcohols, substrates that otherwise are complicated to be selectively modified. In this sense, Akai and co-workers have described the transformation of cyclic tertiary allylic alcohols into derivatives bearing a quaternary

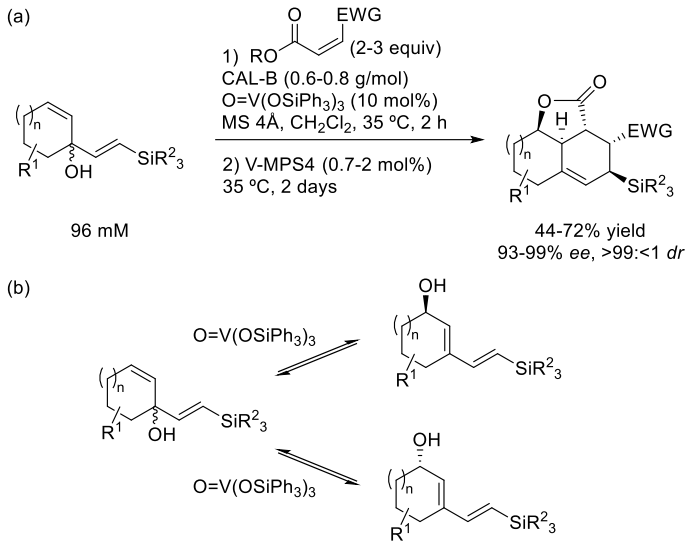
center in enantioenriched form (Scheme 12).[55] The dynamic process was studied with different substrates and acyl donors, and after optimization, it was selected the combination of CAL-B (3% w/w) and immobilized V-MPS4 (1 mol%) in acetonitrile at different temperatures (35-60 ºC) and reaction times (12-48 h) depending on the substrate and acyl donor employed. Thus, (*R*)-O-acylated products were synthesized in good to excellent yields (77-99%) and *ee* values (81->99%). After the DKR-1,3-transposition process, the obtained cyclic esters were submitted to Ireland-Claisen rearrangement conditions (lithium diisopropylamide and hexamethylphosphoramide at –78 ºC, followed by the addition of *tert*-butyldimethylsilyl chloride at –78 ºC and acidic hydrolysis), to afford the corresponding final optically active carboxylic acid compounds containing a quaternary center.



Scheme 12. Synthesis of cycloalkenes containing a quaternary center after DKR of cyclic allylic alcohols combining CAL-B and V-MPS4.

In a subsequent contribution, a lipase-oxovanadium combination was implemented to perform the DKR of a series of cyclic tertiary allylic alcohols followed by an intramolecular Diels-Alder transformation to synthesize tricyclic compounds in a diastereoselective manner (Scheme 13a).[56] The key of this one-pot process was the use of an α,β-unsaturated ester as acylating agent in the lipase-catalyzed reaction, so this intermediate further reacted in order to provide the desired product. Thus, after reaction optimization, CAL-B and $VO(OSiPh_3)_3$ were selected as the most suitable catalysts in dichloromethane at 35 ºC. The addition of molecular sieves improved the reproducibility of the process. After 2 h, V-MPS4 was supplemented to the reaction medium to catalyze the Diels-Alder addition, forming the products in moderate to high yields (44-72%) and usually with excellent enantio- and diastereomeric excess (93-99% *ee* and >99% *de*). As previously described in these systems,[48,49] the vanadium catalyst not only mediated the

substrate racemization but also the alcohol migration, which in fact, after enzymatic acylation, gave the desired intermediate (Scheme 13b).

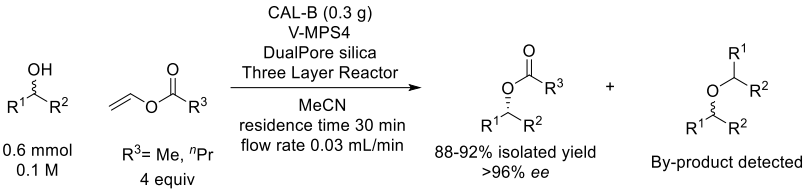


Scheme 13. Vanadium species in DKR processes: (a) DKR of a tertiary allylic alcohol using the $O=V(OSiPh_3)_3$/CAL-B system followed by a Diels-Alder addition catalyzed by V-MPS4. (b) Migration of the alcohol group mediated by the oxovanadium species.

2.1.4. Flow chemistry applied to the DKR of alcohols

The use of a continuous operation mode, also known as flow chemistry, has several advantages over traditional batch transformations, including an increase in the catalyst lifetime and turnover numbers, and also a reduction of the reaction times paving the way to bridge the gap from academic concepts to industrial processes. This has not gone unnoticed for the Biocatalysis area,[30-32] especially when considering the development of DKR processes to circumvent the bio- and chemocatalyst incompatibilities. For instance, Gröger, Akai and co-workers have described the use of the oxovanadium catalyst V-MPS4[52] and CAL-B (Novozym 435) to develop a DKR process of secondary alcohols in flow mode.[57] Firstly, the DKR study of (*E*)-4-phenylbut-3-en-2-ol (0.1 M) was carried out in a packed column with CAL-B and V-MPS4 in the presence of vinyl acetate (4 equiv) at 35 ºC. Unfortunately, a change in the distribution of both catalysts was observed during the process, resulting in a biotransformation efficiency decrease. The authors

justified this loss of activity based on the small particle size of V-MPS4 (0.03-0.05 mm) compared to the immobilized CAL-B particles, that causes its movement along the column when the solution is pumped. To solve this problem, different column packings were tested obtaining the best results when DualPore^{TM} was employed, even maintaining the initial distribution of the catalysts for three days. Another drawback reported by the authors was the formation of significant amounts of a dimeric ether side-product, which was explained due to the reaction of the allyl cation (formed in the racemization process) with one molecule of the starting racemic alcohol. To overcome this problem, a reactor packed with a ratio gradient of V-MPS4 and CAL-B was used and further optimized, obtaining the best results with three-catalyst layers and a 0.03 mL/min flow. Therefore, a series of six (*R*)-esters was recovered with high isolated yields (88-92%) and enantiomeric excess (>96%) after 30 min of residence time with 4 equiv of vinyl acetate or butyrate as acyl donor, acetonitrile as solvent and 35 ºC over three days (Scheme 14).

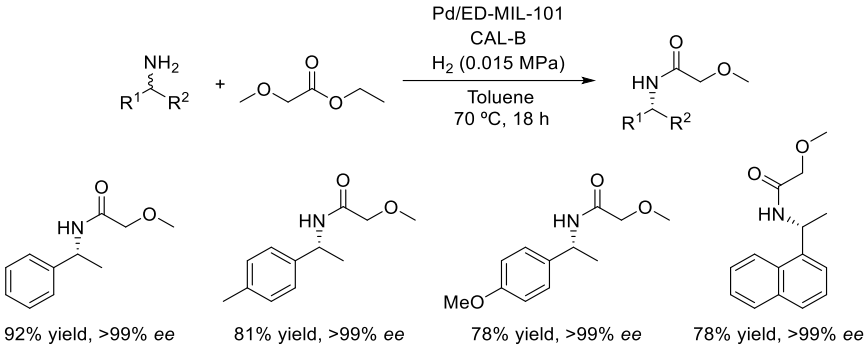


Scheme 14. Continuous-flow DKR of secondary alcohols by lipase-oxovanadium cocatalysis.

Similarly, the DKR of 1-phenylethanol using CAL-B and $VOSO_4$ in a single packed-bed was developed by de Souza and co-workers in continuous flow mode.[58] After optimization of the catalysts loading, temperature, solvent and residence time, the enantiopure (*R*)-1-phenylethyl decanoate and (*R*)-1-phenylethyl acetate were obtained using vinyl decanoate and vinyl acetate as acyl donors, respectively. The same group developed the DKR of 1-(2,6-dichloro-3-fluorophenyl)ethanol (100 mM), an intermediate for the synthesis of the anti-cancer agent Crizotinib, under flow conditions applying different layers of immobilized CAL-B (Novozym 435) and $VOSO_4.xH_2O$ in the presence of isopropenyl acetate (100 mM) as acylating agent in isooctane at 80 ºC. After 5 days with a flow rate of 0.5 mL/min, 57% conversion of the acetylated (*R*)-isomer (96% *ee*) was attained.[59]

2.1.5. DKR of amines

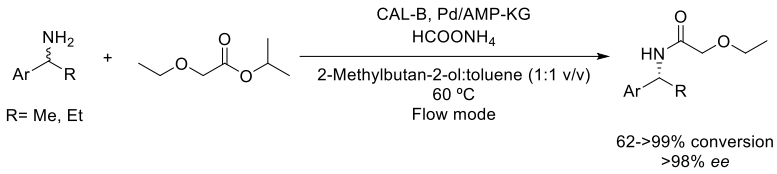
Generally, DKR of amines has been more challenging than those starting from the alcohol counterparts. Li and co-workers described the encapsulation of Pd NPs in cages of ethylenediamine-grafted MIL-101 (Pd-ED-MIL-101), which allowed the metal recycling for racemization purposes.[60] After optimization of the reaction conditions in terms of catalyst loading, temperature, hydrogen pressure, and solvent, four racemic amines were stereoselective acylated under dynamic conditions using ethyl methoxyacetate (1 equiv) as acyl donor in toluene at 70 ºC (Scheme 15). Moreover, both catalysts showed a good recycling capacity (up to 9 cycles), still showing good activity and selectivity. A year later, the same research group proof the compatibility of the Pd-ED-MIL-101 with CAL-B, in this case immobilized as cross-linked enzyme aggregates (CLEAs), for the DKR of both primary and aliphatic amines (81-99% yield, >99% *ee*) including for instance 2-heptylamine and 1-methyl-3-phenylpropylamine.[61]



Scheme 15. DKR of racemic amines using CAL-B and an encapsulated palladium catalyst.

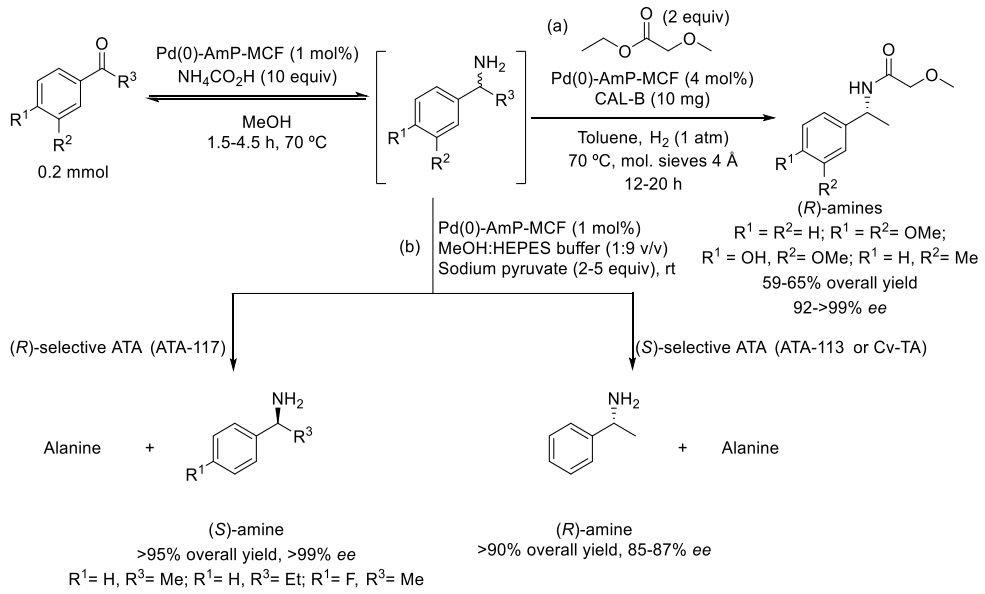
Interestingly, the DKR of amines in continuous mode is also possible as described by Hornyánszky, Poppe and co-workers, which described the resolution of seven benzylic amines using sol-gel immobilized CAL-B as biocatalyst, selecting palladium on 3-aminopropyl-functionalized silica (Pd/AMP-KG) as racemization reagent after testing six different palladium preparations (Scheme 16).[62] The best results were found with isopropyl 2-ethoxyacetate as acyl donor (1 equiv), a substrate concentration of 138 mM

in a mixture 2-methylbutan-2-ol:toluene (1:1 v/v) and ammonium formate (0.6 equiv) as the hydrogen source at 60 ºC.



Scheme 16. DKR of benzylic amines using CAL-B and palladium preparations using flow chemistry.

Córdova and co-workers have described a reductive amination/DKR catalytic relay consisting in the initial reversible transformation of four acetophenones into the corresponding racemic amines using palladium nanoparticles, which were subsequently stereoselectively acylated under DKR conditions using CAL-B and ethyl methoxyacetate in toluene at 70 ºC (Scheme 17a).[63] Interestingly, a sequential methodology is described employing palladium(0)-aminopropyl-mesocellular foam [Pd(0)-AmP-MCF],[64,65] which involves: (a) the reductive amination of the prochiral ketones using ammonium formate under inert conditions in methanol; (b) the DKR of the racemic amine intermediates after methanol evaporation and replacement with toluene in hydrogen atmosphere. Thus, the corresponding (*R*)-methoxyacetamides (92->99% *ee*) were isolated after column chromatography in 59-65% isolated yield. An alternative reductive amination/kinetic oxidative resolution relay sequence is reported in the same contribution, replacing in this case lipases by transaminases. Thus, employing (*S*)- or (*R*)-selective transaminases including commercial ATA-113 and ATA-117 from Codexis Inc. or the one from *Chromobacterium violaceum* (Cv-TA), the KRs were performed with high to excellent selectivity (Scheme 17b).

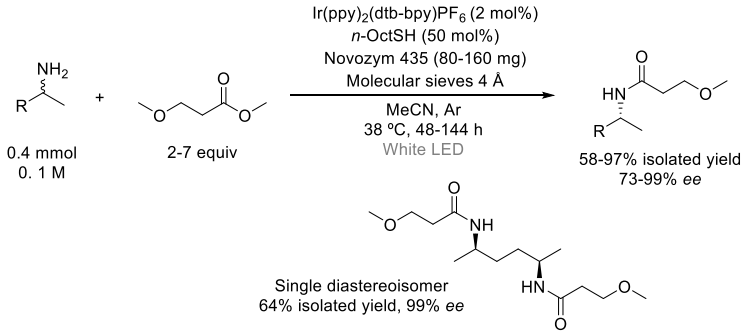


Scheme 17. Heterogeneous metal/enzyme co-catalyzed sequential transformation of ketones into amines via their corresponding racemates using: (a) CAL-B in a DKR approach; (b) amine transaminases for a KR strategy.

2.1.6. DKR using photobiocatalysis

As it has been shown throughout this section, the use of a DKR strategy is an elegant solution to obtain alcohol and amine derivatives with high optical purity and excellent yields. Traditionally, as in the previous examples, transition metals have been applied to induce the racemization process of the unreacted enantiomer, however, in recent years the use of photoredox-mediated processes has emerged as sustainable and innovative alternatives.[66,67] An example of the merging of metal-, photo- and biocatalysis has been described by Zhou and co-workers,[68] describing the racemization of different aliphatic primary amines by the use of an Ir(III) catalyst under white LED in combination with a CAL-B catalyzed stereoselective acylation. To achieve this, the corresponding racemic amine (0.4 mmol) was disposed in the presence of $Ir(ppy)_2(dtb\text{-}bpy)PF_6$ (2 mol%) as photocatalyst and *n*-octanethiol (50 mol%) as hydrogen atom transfer in MeCN, while for the enzymatic process, Novozym 435 (80-200 mg) and methyl 3-methoxypropanoate (2-7 equiv) were employed as enzyme and acyl donor, respectively, in the presence of molecular sieves (4 Å-MS, 400 mg) and under argon atmosphere (Scheme 18). After 2-6

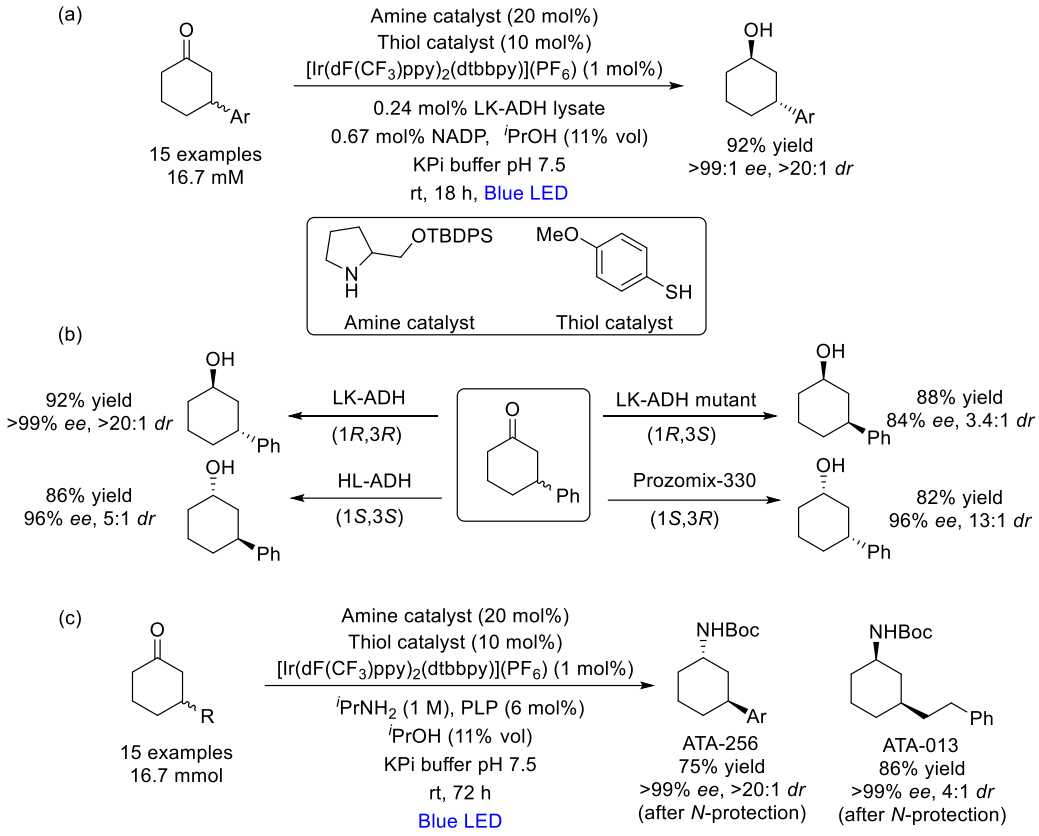
days, at 38 ºC, 19 (*R*)-amides were obtained with moderate to excellent isolated yields (58-97%) and good to excellent *ee* (73-99%). Thiols such as *n*-OctSH were identified as ideal hydrogen atom donors to intercept carbon radicals,[69] and it is interesting to notice that in the absence of light no racemization occurred, while under photocatalytic conditions but without 4 Å-MS, the racemization rate notably decreased. The authors justify this fact by the positive effect of molecular sieves due to a possible coordination of their Al^{3+} cations with amines, that could accelerate the generation of α-amino radicals. Furthermore, this methodology has been extended to hexane-2,5-diamine, furnishing high levels of diastereo- and enantioselectivity, and corresponding to the first report of a DKR of a 1,4-diamine.



Scheme 18. DKR of racemic amines coupling a visible-light photoredox process and a lipase-catalyzed acylation.

More recently, Hyster's group has demonstrated the compatibility of different types of biocatalysts under light conditions, the development of novel photobiocatalytic reactions in which the native repertoire of enzymes is expanded.[70] This research group has overcome one of the key problems related with DKR processes, which relies in the fact that chirality induction only occurs at inducible dynamic stereocenters. However, most of the stereogenic centers in organic molecules are static, which constitutes a serious limitation in asymmetric organic synthesis. Thus, an outstanding photo-, organo-, metal- and biocatalytic approach has been designed to convert β-substituted ketones into chiral γ-substituted amines and alcohols by combining under photocatalytic conditions an iridium catalyst, two organocatalysts and an enzyme such as an alcohol dehydrogenase (ADH) or an amine transaminase (ATA), respectively.[71] Cyclic ketones (16.7 mM) with a distal stereocenter were reacted with 20 mol% of an amine catalyst, 10 mol% of a thiol

catalyst, 1 mol% of [Ir(df(CF_3)ppy)$_2$(dtbbpy)](PF_6) and 0.24 mol% of *Lactobacillus kefir* ADH lysate (including 0.67 mol% NADP and 11% v/v ^{i}PrOH) in a phosphate buffer 50 mM pH 7.5 for 18 h at room temperature and under blue LED light irradiation (Scheme 19a). Thus, the target γ-substituted alcohols were obtained with high yields (68-92%) and enantio-diastereomeric ratio, after the action of 5 different catalytic cycles. It must be highlighted, that all possible 3-phenylcyclohexan-1-ol stereoisomers were prepared starting from racemic 3-phenylcyclohexanone by employing different ketoreductases (*Lactobacillus kefir* alcohol dehydrogenase, a *Lactobacillus kefir* variant, horse liver alcohol dehydrogenase or commercial Prozomix-330 ADH, Scheme 19b). Finally, the authors applied this methodology to the production of chiral amines using different commercial transaminases (ATA-013 and ATA-256 from Codexis) and isopropylamine (^{i}PrNH$_2$) as amine donor, for the biotransamination of two racemic 3-substituted-cyclohexanones (Scheme 19c), which after amine protection as carbamates (Boc, *tert*-butoxycarbonyl group), were isolated in high yields (75-86%) and with excellent optical purities. This example demonstrates the great potential that the combination of different catalytic methodologies offers to develop transformations that in other way would be unthinkable.



Scheme 19. DKR of cyclic ketones substituted at static stereocenters by the combination of photo-, metallo-, organo- and biocatalysis.

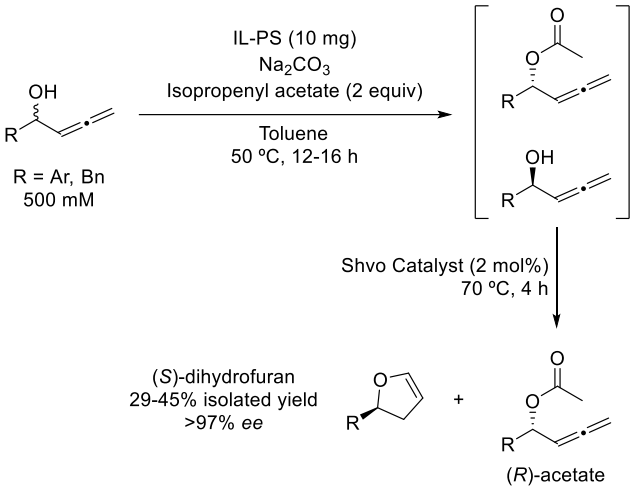
As it has been discussed along this section, the development of one-pot DKR processes combining selective lipase-catalyzed transformations and metal-catalyzed racemizations of the unaltered enantiomer using palladium, rhodium, ruthenium, vanadium and iridium catalysts, still represents the most common strategy for the production of enantiomerically pure esters and amides under dynamic conditions starting from racemic alcohols and amines, respectively. The compatibility of metals and enzymes is usually hampered for the requirement of high temperatures and long reaction times, while inert conditions are necessary to assure a good reactivity of the metal catalyst along the process. However, recent progresses including continuous operational modes, immobilization of the biocatalyst on mesoporous cellular foams (MCFs),[72] or the use of bionanohybrids (see Section 3) open new opportunities including the development of these multicatalytic transformations under safer reaction conditions. Remarkably, the successful combination of metallo-, photo- and biocatalysis provides a promising perspective for DKR processes,

now starting to be developed with several enzyme classes including oxidoreductases and transferases.

2.2. (Cyclo)isomerizations

Isomerization reactions, especially cycloisomerizations, are very attractive transformations from a sustainable point of view,[73] allowing the construction of different molecules with perfect atom economy.[74] The development of these transformations under mild reaction conditions has attracted recent attention, including the use of aqueous media, low temperatures or photocatalytic methods to achieve this aim. In this context, the combination of one-pot metal- and enzyme-catalyzed reactions represents an attractive strategy when starting for instance from allylic, propargylic or allenic alcohols. Mostly, ruthenium complexes have been the metal of choice for isomerization purposes, due to their ability to induce this type of reaction including racemization of secondary alcohols and primary amines,[38,75-77] so next, the potential of chemoenzymatic approaches including isomerizations is deeply discussed.

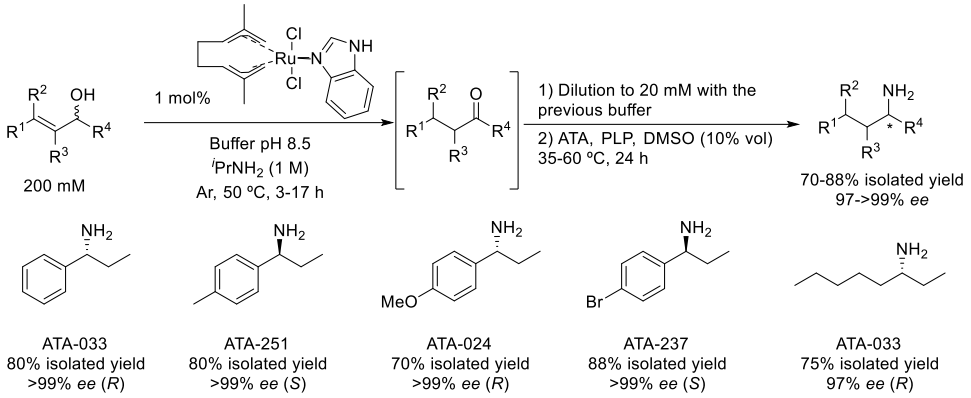
Bäckvall and co-workers reported in 2016 a sequential process involving the lipase-catalyzed KR of racemic α-allenic alcohols followed by the cycloisomerization of the unaltered (*S*)-alcohols to get a series of optically active 2,3-dihydrofurans (Scheme 20).[78] After preliminary enzyme and metal screenings, plus later optimization of the individual reaction conditions, the stereoselective transesterification of the (*R*)-alcohols was successfully developed in a high substrate concentration (500 mM) using an ionic liquid coated *Pseudomonas cepacia* lipase preparation (IL1-PS), and isopropenyl acetate (2 equiv) as acyl donor. Consumption of the preferred alcohol enantiomer was monitored by chiral HPLC (12-16 h), and after this time the Shvo catalyst (2 mol%) was added, stirring the mixture for 4 h at 70 ºC. Thus, eleven (*S*)-2,3-dihydrofurans were produced with excellent selectivity (>97% *ee*) and moderate yields after column chromatography purification (29-45%).



Scheme 20. One-pot sequential lipase-catalyzed kinetic resolution of α-allenic alcohols followed by a ruthenium-catalyzed cycloisomerization.

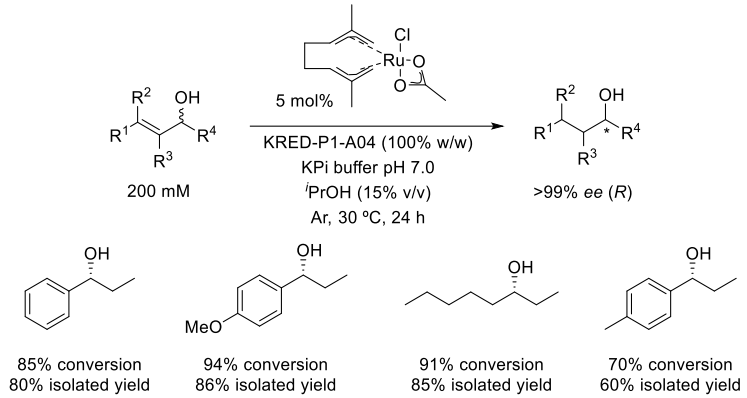
As mentioned before, there is an increasing interest in the design of more sustainable processes including the replacement of organic solvents by aqueous or neoteric media such as deep eutectic solvents (DES).[79-82] The Spanish biotechnological company Entrechem has been particularly active in this area, disclosing the combination of ruthenium catalysts with alcohol dehydrogenases (ADHs) or amine transaminases (ATAs). For instance, in 2015 a chemoenzymatic sequential approach was performed based on a metal-catalyzed isomerization of an allylic alcohol, followed by asymmetric reductive bioamination of the corresponding ketone intermediate.[83] Interestingly, the isomerization of allylic alcohols to the saturated ketones took place with total conversion under argon atmosphere in a phosphate buffer in the presence of a low loading of a ruthenium catalyst (1 mol%) and at high $^{i}PrNH_2$ concentration (1 M, Scheme 21). A key issue when using ATAs is the use of high amine donor concentrations enabling the equilibrium shifting towards the thermodynamically unfavored amine synthesis (normally 50 equiv). The presence of such a high amine donor concentration usually affects the activity of the metal catalysts, so sequential approaches are usually more feasible rather than concurrent strategies. Another issue is the very different concentrations usually employed for each transformation. On one hand, alcohol isomerization efficiently occurs at 200 mM, observing a conversion drop and the appearance of reaction by-products at lower substrate concentrations. On the other hand, biotransaminations are usually developed at 5-25 mM ketone concentration to avoid

enzyme inhibition and favor reaction equilibrium. To tackle this concentration hurdle, the reaction mixture with the ketone intermediate was diluted 10-times in this case, prior adding the corresponding stereoselective ATA, the enzyme cofactor (5'-pyridoxal phosphate, PLP, 1 mM) and an organic cosolvent (DMSO, 10% v/v) commonly employed for a substrate solubility purpose. Finally, the desired amines were obtained in good to high isolated yields (70-88%) and excellent enantiomeric excess (>97%).



Scheme 21. Chemoenzymatic one-pot sequence through ruthenium-catalyzed isomerization of allylic alcohols followed by an enzymatic transamination.

In 2016, the same authors reported a similar approach replacing the biotransamination step by a bioreduction using commercial ketoreductases (KREDs), giving access to the corresponding optically active alcohols (Scheme 22).[84] Remarkably, the redox isomerization of the studied racemic allylic alcohols (200 mM) was carried out under argon atmosphere with a low ruthenium catalyst loading (5 mol%), and successfully coupled with the bioreduction step at 30 ºC in a concurrent manner, rather than developing a sequential approach. Thus, using the commercial KRED-P1-A04 and always that the isomerization took place rapidly, a series of enantiopure alcohols were synthesized in high isolated yields (80-86%) after 24 h, leading to lower yields when the metal-catalyzed process was not so fast as occurred with 1-(*p*-tolyl)propan-1-ol (60% isolated yield).

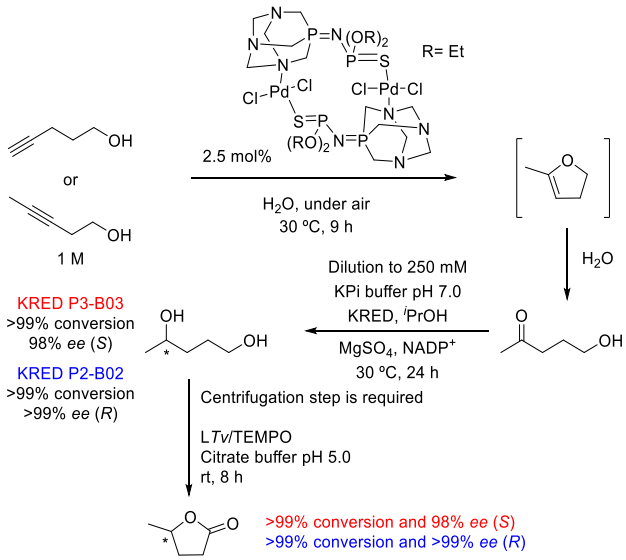


Scheme 22. Chemoenzymatic one-pot concurrent cascade through ruthenium-catalyzed isomerization of allylic alcohols followed by stereoselective bioreduction.

Two years later, the same chemoenzymatic strategy was successfully achieved both in a sequential and concurrent manner by using DES-buffer mixtures as reaction media.[85] This research was the first example of a chemoenzymatic cascade process using this type of solvent mixtures, employing in this case a 1:1 v/v ratio with a DES being composed by choline chloride (ChCl) and glycerol (Gly) in a 1:2 mol/mol ratio. Satisfyingly, the desired alcohols were obtained with moderate to excellent conversions (70-96%) and very high optical purity (93->99% *ee*) in a cascade fashion. Unlike their previous work, both commercial KRED-P2-C11 and *Lactobacillus kefir* ADH were used, requiring an increase in both the metal catalyst loading (5 to 10 mol%) and the temperature (30 to 40 ºC) due to the presence of DES. Without any doubt, the use of ecofriendly solvents such as DES[79-82] provides many opportunities for the recovery and recycling of the catalytic system, also replacing hazardous organic volatile cosolvents. Importantly, these advantages have not gone unnoticed for the development of biocatalyzed redox transformations.[86,87]

A sequential strategy is depicted in Scheme 23 for the cycloisomerization of pent-4-yn-1-ol and pent-3-yn-1-ol followed by spontaneous hydration and ketone bioreduction.[88] This work includes firstly the optimization of the cycloisomerization of these two terminal alkynes using different palladium(II) complexes at 30 ºC in aqueous medium, leading to the formation of 5-methyl-2,3-dihydrofuran as reaction intermediate, which spontaneously hydrolyzed in the reaction medium to form a γ-hydroxy ketone. Next, bioreduction of the resulting carbonyl group using commercial stereoselective KREDs

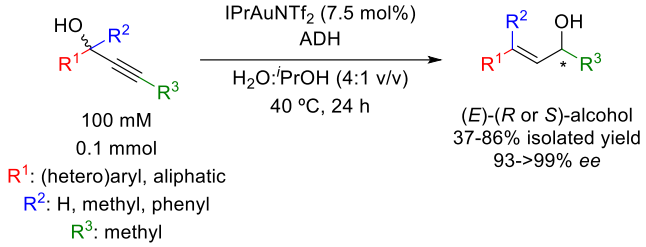
(KRED P2-B02 and KRED P3-B03) gave access to optically active (*R*)- and (*S*)-pentane-1,4-diol, respectively. Furthermore, this process can be combined with an aerobic oxidation using a laccase-mediator system composed by the oxy-radical TEMPO and the laccase from *Trametes versicolor* (L*Tv*) to furnish both γ-valerolactone enantiomers. It must be highlighted that to avoid the detrimental effect of the KRED on the laccase/TEMPO system, a centrifugation step was required to remove the insoluble protein before developing the final oxidative reaction with the resulting supernatant at pH 5.0 and at room temperature.



Scheme 23. Palladium-catalyzed cycloisomerization of terminal alkynes followed by spontaneous hydration, carbonyl bioreduction and regioselective alcohol oxidation to form enantioenriched γ-valerolactone.

Recently, our research group has described the first concurrent cascade involving a N-heterocyclic carbene gold(I) catalyst and an ADH (Scheme 24),[89] demonstrating the great synthetic applicability of the combination of these two types of catalysts to produce enantioenriched β,β-disubstituted allylic alcohols. The approach is based on the Meyer-Schuster rearrangement[90,91] of a broad series of racemic propargylic alcohols to produce the corresponding α,β-unsaturated ketones, and their subsequent *in situ* stereoselective bioreduction. To obtain the enantioenriched alcohols, the starting propargylic alcohol (100 mM) was placed in a water:2-propanol medium (4:1 v/v), in the presence of the IPrAuNTf$_2$ (7.5 mol%) as gold(I) catalyst and an ADH with its corresponding cofactor regeneration system. After 24 h at 40 ºC, a series of fifteen allylic alcohols were obtained

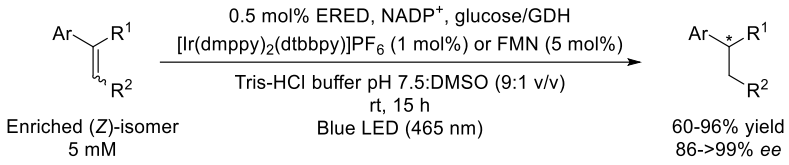
in moderate to high isolated yields (37-86%) and remarkable enantiomeric excess values (93->99%). Depending on the selectivity of the ADH of choice, both allylic alcohol enantiomers were produced under mild reaction conditions, and in a (*E*)-selective manner, since the ADHs employed in this contribution were not capable to transform the (*Z*)-isomers.



Scheme 24. First concurrent cascade of a NHC-gold(I) catalyst and an ADH to produce optically active (*E*)-allylic alcohols.

Finally, Zhao, Hartwig and co-workers have reported the successful combination of a cationic iridium(III) complex and an ene-reductase (ERED) under blue light irradiation for an alkene photoisomerization-bioreduction sequence.[92] Derivatives of 2-arylbut-2-enedioic acid dimethyl ester have been studied as starting products to produce the isomerization between their (*Z*)- and (*E*)-forms, being the last one the isomer preferred for these oxidoreductases. For the development of this cascade process, an alkene enriched in its (*Z*)-form (5 mM) was reacted with a ERED (YersER, XenB, OPR1, TOYE, OYE2 or SYE1, 0.5 mol%) and $[Ir(dmppy)_2(dtbbpy)]PF_6$ (1 mol%) or flavin mononucleotide (FMN, 5 mol%) as photocatalysts, in the presence of $NADP^+$ (0.2 mM), glucose/glucose dehydrogenase (GDH) as cofactor recycling system, DMSO as cosolvent (10% v/v) and Tris-HCl buffer pH 7.5 (Scheme 25). After 15 h at rt under blue light (465 nm), products from alkenes bearing electron-withdrawing groups were obtained in moderate to high conversions (60-96%) and high optical purities (86->99% *ee*). The reactions proceeded in a cooperative fashion (both (bio)catalysts simultaneously catalyzing two reactions in the same medium) since a poor isomerization was achieved when performing the sequential approach by addition of the ERED after 8 h. The synthesized enantioenriched products were subsequently modified leading to highly interesting families of compounds including furans, diols, carboxylic acids, amino acids, amino esters, oxo esters and lactams. Remarkably, the enzymatic reduction of C=C bond

provides a sustainable alternative to the metal-catalyzed traditional hydrogenation approach since the biotransformations can be carried out with excellent stereoselectivity in aqueous medium and under atmospheric pressure and room temperature.



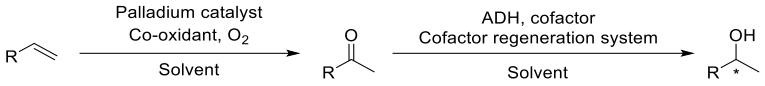
Scheme 25. Cooperative alkene photoisomerization and bioreduction.

Overall, metal-catalyzed isomerization reactions are key transformations in organic chemistry, assuming the simplest way to start building molecular complexity with a perfect atom economy. The reported examples demonstrate the possibility to couple these metal-catalyzed isomerizations with stereoselective ATAs, ADHs and EREDs, providing straightforward chemoenzymatic access towards chiral alcohols, amines and alkenes.

2.3. Oxidation, hydrogenation and reduction reactions

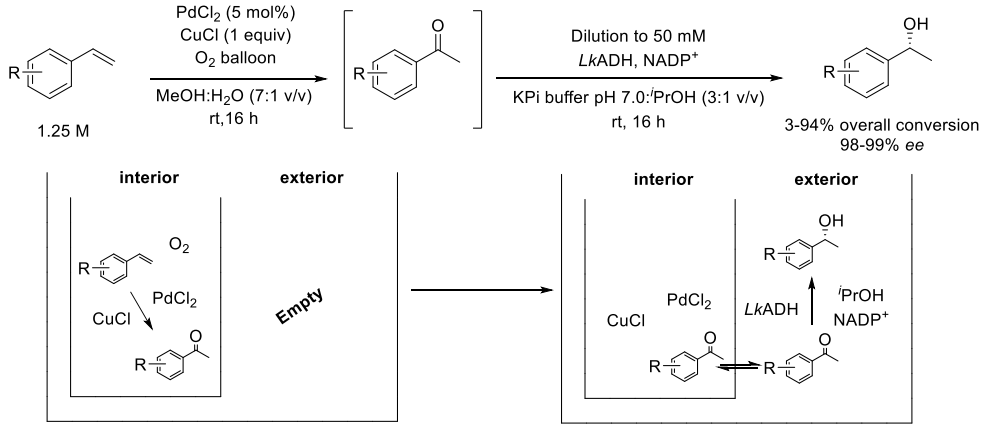
2.3.1. Metal-catalyzed oxidation reactions

The Wacker-Tsuji oxidation,[93,94] consisting of the palladium-catalyzed aerobic transformation of alkenes into carbonyl compounds, is one of the most studied metal-catalyzed oxidative reactions when combining it with a biocatalyzed process. Particularly, Gröger's group has been very active in this field since 2012,[95] demonstrating the compatibility of the Wacker-Tsuji oxidation of styrene derivatives with the use of ADHs and ATAs for the bioreduction or biotransamination of the acetophenone intermediates, respectively. Thus, when considering the use of selective ADHs, a formal asymmetric hydration of alkenes is attained as depicted in Scheme 26. For the oxidative process, a Pd(II) catalyst such as $PdCl_2$ or $Pd(OAc)_2$ is usually employed, coordinating itself with the olefin. To assure the complete reoxidation of Pd(0) to Pd(II), the combination of molecular oxygen as final electron acceptor with a co-catalyst and/or oxidant is necessary, finding Cu(II) salts, benzoquinone or $Fe_2(SO_4)_3$ in stoichiometric amount as adequate reagents to achieve a successful overall oxidation.



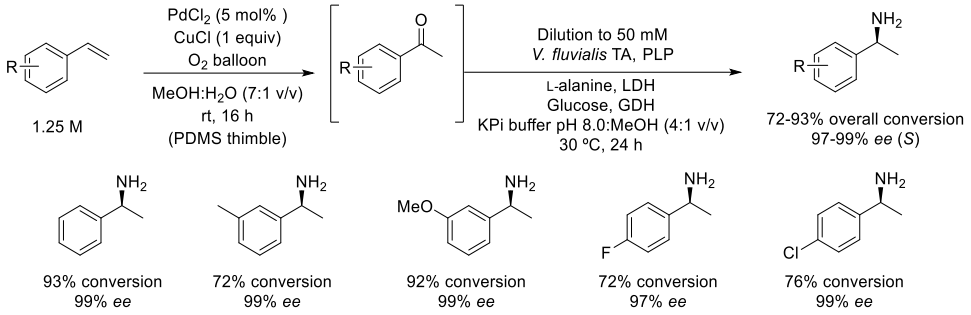
Scheme 26. Chemoenzymatic Wacker-Tsuji oxidation-bioreduction sequence for the formal asymmetric hydration of an alkene.

In 2015, Gröger and co-workers overcame previous limitations[95] in the Wacker-Tsuji oxidation-bioreduction transformation to convert styrene into 1-phenylethanol, via compartmentalization of both reactions to avoid thus the ADH inhibition.[96] Contrary to what many people assume, it was demonstrated that a Pd(II) species like $PdCl_2$ did not influence the ADH activity, concluding that the ADH inactivation was caused due to the presence of Cu(II) species. Thus, a polydimethylsiloxane (PDMS) thimble was used to locate inside both $PdCl_2$ and CuCl, while the enzyme system (ADH and cofactor) was outside to assure that no enzyme inactivation occurred, while the ketone intermediate could go through both compartments. After reaction optimization, a water-methanol mixture was found as the best solvent, but the concurrent cascade resulted not feasible due to a decrease in the methanol content inside the thimble along the process (only 20% alcohol conversion was reached). However, the development of a sequential approach led to the production of various 1-phenylethanol derivatives with high enantiomeric excess (98-99% *ee*) and conversions (up to 94%). $PdCl_2$ (5 mol% loading) and CuCl (1 equiv) were used in a MeOH:H_2O mixture (7:1 v/v) for the first step (Wacker oxidation), and after 16 h at room temperature, *Lactobacillus kefir* ADH (*Lk*ADH), $NADP^+$ and a mixture of buffer:^{i}PrOH (3:1 v/v) were added in the exterior of the thimble to obtain the desired (*R*)-alcohols after additional 16 h (Scheme 27). The recycling of the PDMS thimble was also investigated, observing that the metal-catalytic system could be reused up to 15 cycles with no appreciable activity decrease.



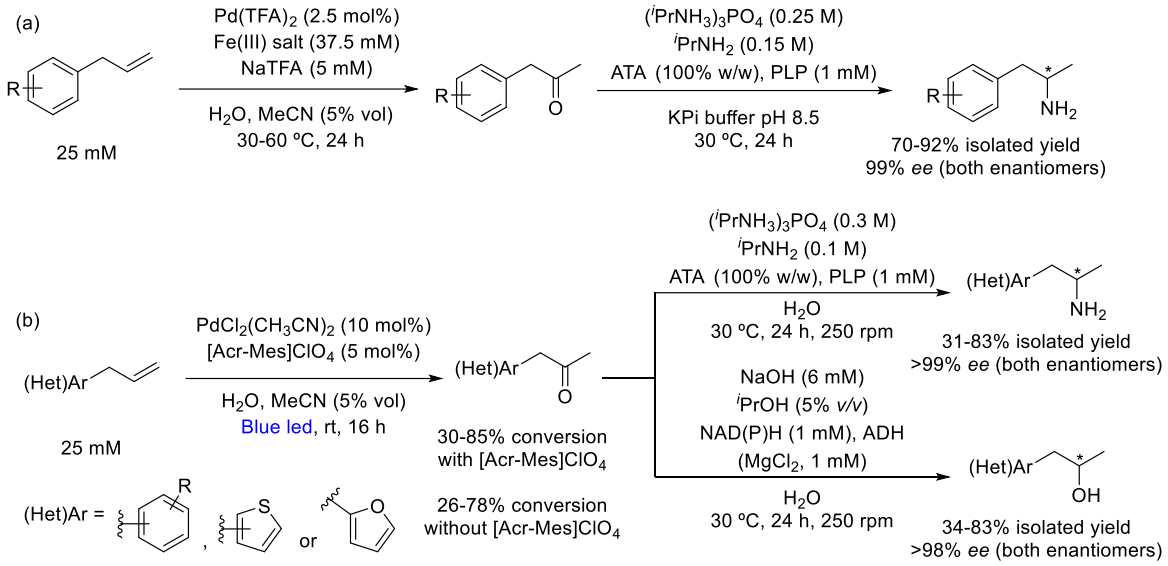
Scheme 27. Sequential Wacker-Tsuji oxidation-stereoselective bioreduction transformation of styrenes into 1-arylethanols through a compartmentalization strategy.

In 2017, the same research group developed a complementary work to stereoselective produce in this case 1-arylethan-1-amines (97-99% *ee*) starting from styrenes (Scheme 28).[97] The Wacker-Tsuji oxidation took place under the same conditions as previously described, this was, 5 mol% $PdCl_2$ and 1 equiv of CuCl in MeOH:H_2O (7:1 v/v) at rt. Since enzyme inhibition was also observed due to the presence of copper ions, the compartmentalized approach was again investigated, the selection of (*S*)-selective transaminase from *Vibrio fluvialis* with L-alanine as amine donor leading to five 1-arylethan-1-amines with good to very high conversions (72-93%) after liquid-liquid extraction. Alternatively and using the same compartmentalization strategy, the synthesis of (*R*)-1-phenylethan-1-amine was also achieved by employing an amine dehydrogenase (AmDH) instead of a transaminase.[98] The selection of a variant from leucine dehydrogenase from *Exigobacterium sibiricum* enabled the replacement of the L-alanine by ammonia as amine donor, thus not requiring the use of lactate dehydrogenase (LDH) in the enzymatic system, only employing glucose/GDH to recycle the nicotinamide cofactor.



Scheme 28. One-pot Wacker-Tsuji oxidation-biotransamination sequence to convert styrene derivatives into optically active 1-arylethan-1-amines.

The synthesis of amphetamine derivatives has traditionally attracted great synthetic attention due to their powerful biological effects, and our research group has recently described their stereoselective synthesis from allylarenes through a sequential Wacker-Tsuji oxidation-biotransamination sequence.[99] After optimization of the initial step, the system composed by palladium(II) trifluoroacetate [$Pd(TFA)_2$] (2.5 mol%), iron(III) sulfate (1.5 equiv) and sodium trifluoroacetate (NaTFA, 20 mol%) at 30-60 ºC for 24 h was selected as an efficient oxidative combination, working appropriately in aqueous medium (H_2O:MeCN mixture 95:5 v/v). The one-pot sequential process, developed without compartmentalization requirements, was performed at a 25 mM substrate concentration, adjusting the pH of the reaction medium with a phosphate buffer pH 8.5 previous to the addition of isopropylammonium phosphate (0.25 M), isopropylamine (0.15 M), PLP and the corresponding transaminase (100% w/w) for the biotransamination step at 30 ºC for 24 h. Depending on the ATA of choice, both amine enantiomers could be obtained with high isolated yields (70-92%) and enantiomeric excess (99%, Scheme 29a).

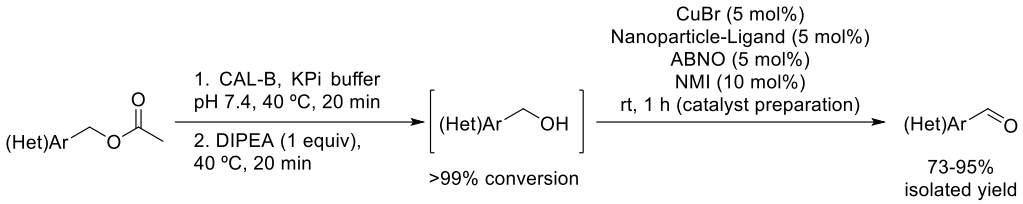


Scheme 29. One-pot (photo)metal-Wacker-Tsuji oxidation of allyl(hetero)arenes-biotransformation sequences: (a) Involving a biotransamination to furnish optically active 1-arylpropan-2-amines; (b) using light irradiation with a biotransamination or a bioreduction process.

Our research group has also recently described the possibility to develop a light-driven Wacker-Tsuji oxidation of different allyl(hetero)arenes to provide with high selectivity the formation of the ketone intermediates. After an intensive optimization, the best conditions were found in a H_2O:MeCN mixture (95:5 v/v) at rt for 16 h, with bis(acetonitrile)palladium(II) dichloride (10 mol%) acting as the oxidative catalyst and 9-mesityl-10-methylacridinium perchlorate ([Acr-Mes]ClO_4, 5 mol%) as the photocatalyst. Nevertheless, it was confirmed that the photosensitizer was not strictly necessary in this process, as it was noticed that even in its absence, high conversions into the ketones were obtained, although a higher extent of several by-products were detected. Furthermore, this methodology was applied in two sequential photo-metal-biocatalytic approaches to furnish the corresponding enantioenriched amines and alcohols by using ATAs or ADHs, respectively. Thus, depending on the enzyme selectivity, both enantiomers of the different 1-(hetero)arylpropan-2-amines or 1-(hetero)arylpropan-2-ols were usually isolated with high yields (31-83% for amines and 34-83% for alcohols) and excellent enantiomeric excess (>99% for amines and >98% for alcohols, Scheme 29b).[100]

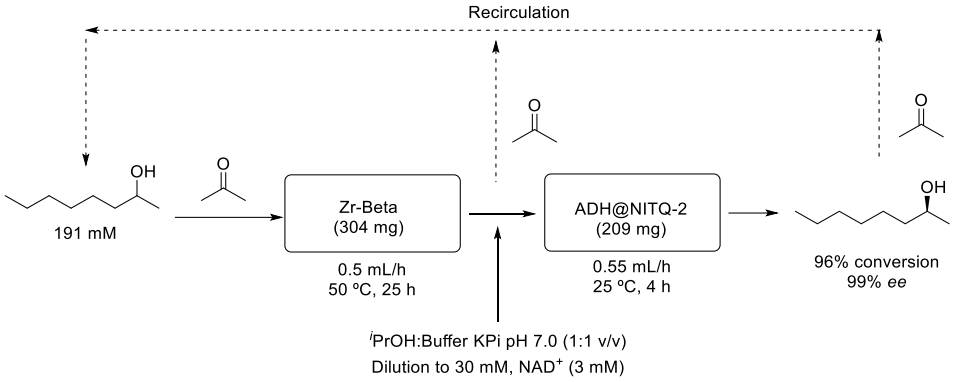
These examples showed that the combination of the Wacker-Tsuji oxidation and a second biocatalytic step has become a powerful tool to obtain optically active alcohols or amines from alkenes. However, these approaches require a compartmentalization or a sequential strategy as a smart solution to avoid mutual metal and enzyme inhibition. Nonetheless, the discovery of mild and suitable conditions for this oxidative step, for instance without requiring stoichiometric amounts of the co-oxidizing agent would minimize the impact in the biocatalytic step, maybe facilitating the design of a concurrent cascade approach.

As it has been previously described, when both bio- and metal-catalyzed steps are not compatible, this limitation can be surpassed by employing a compartmentalized system. In this context, Weberskirch and Sand developed a one-pot multistep chemoenzymatic approach to covert acetyl esters into the corresponding aldehydes in aqueous medium (Scheme 30).[101] The process combined the enzymatic ester hydrolysis catalyzed by CAL-B with the alcohol oxidation using a Cu(I)/bipyridine catalyst in the presence of a *N*-oxyl radical oxidant. The ester (160 μmol, 1 equiv) and the immobilized CAL-B were incubated for 20 min at 40 ºC in the phosphate buffer, then, *N*,*N*-diisopropylethylamine (DIPEA, 1 equiv) was added to neutralize the acetic acid formed, and the mixture stirred for additional 20 min at 40 ºC. Finally, *N*-methylimidazole (NMI, 10 mol%), employed as base, 9-azabicyclo[3.3.1]nonane *N*-oxyl (ABNO, 5 mol%), and previously prepared nanoparticles containing the copper ions were added, and the reaction stirred for additional 4 h. The resulting aldehydes were obtained with good to high yields after liquid-liquid extraction (73-95%), the oxidative process being highly dependent on the substrate structure, finding the poorest results with the heteroaromatic compounds (2-thiophene and 2-furan derivatives, 73-80% yield). Upscale experiments with benzyl acetate and 4-methoxyphenyl acetate (3.2 mmol) allowed the formation of the corresponding aldehydes in 93 and 95% isolated yield, respectively, after liquid-liquid extraction and column chromatography.



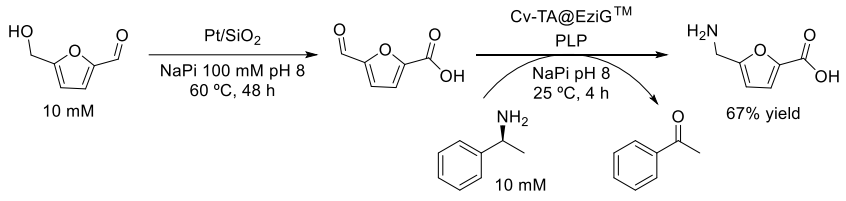
Scheme 30. Conversion of acetyl esters into aldehydes through enzymatic ester hydrolysis and Cu-mediated alcohol oxidation.

Very recently, Iborra, Corma and co-workers have described the production of enantioenriched alcohols from racemic mixtures by the combination of a chemical oxidation and a bioreduction step operating both in continuous mode (Scheme 31).[102] The deracemization strategy consisted of the Oppenauer oxidation of racemic aliphatic and aromatic alcohols employing a Lewis acid zeolite (Zr-Beta) and a second asymmetric reduction step using an ADH immobilized on a two-dimensional zeolite (ITQ-2). Various examples have been published in which Al- and Ti-Beta zeolites were used to carry out this oxidation step,[103] however Zr-Beta zeolites are more adequate Lewis acid promoters.[104] The first oxidative step implied the use of acetone as auxiliary ketone, which was reduced to 2-propanol, its formation serving for substrate solubilization and for enzyme cofactor recycling purposes in the second step. The deracemization process of secondary racemic alcohols involved two fixed-bed continuous reactors. Depending on the selectivity of the immobilized ADH (Prelog-selective ADH030 or anti-Prelog ADH270, both commercial enzymes from Evocatal, now called Evoxx Technologies), the (*S*)- or the (*R*)-enantiomers, respectively, could be attained. Coupling both steps in a flow reactor, 2-octanol (191 mM) in acetone was fed in the Zr-Beta zeolite bed and maintained for 25 h with a flow of 0.5 mL/h at 50 ºC. Then, after acetone elimination, the bioreduction was set-up by the addition of the cofactor (NADH for ADH030 or NADPH for ADH270) and ^{i}PrOH/phosphate buffer pH 7.0 (1:1 v/v) to the second bed at a 0.55 mL/h flow at 25 ºC.



Scheme 31. Sequential continuous approach for the deracemization of 2-octanol employing a Zr-Beta zeolite and an immobilized ADH.

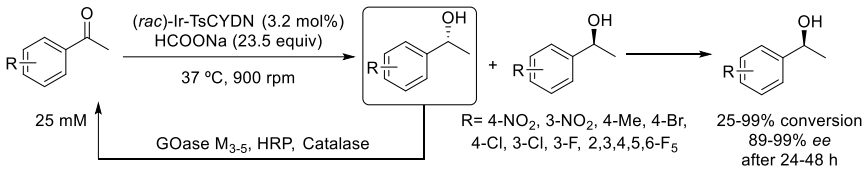
The combination of metal catalysis and enzymes has also been described to synthesize aminated furan derivatives. Thus, Froidevaux, Heuson and co-workers described the synthesis of 5-aminomethyl-2-furancarboxylic acid (AMFC) from HMF, through the combined use of immobilized Pt nanoparticles and transaminases (Scheme 32).[105] After metal and enzymatic screenings, the best catalysts were Pt NPs impregnated on silica and *Chromobacterium violaceum* TA (Cv-TA) immobilized by affinity on a EziG^{TM} support from the company EnginZyme AB. Due to the different reaction conditions, this process was developed in a sequential manner. The first oxidative step was performed treating HMF (10 mM) in phosphate buffer 100 mM pH 8 at 60 ºC with the platinum catalyst (20 mg), attaining 5-aldehyde-2-furancarboxylic acid (AFCA) as intermediate. After 48 h, the reaction was cooled down at 25 ºC, and the enzyme (10 mg) and buffer containing PLP and the amine donor [(*S*)-α-methylbenzylamine (10 mM)] were added and the reaction was stirred for 4 h. After purification, 67% yield of AMFC was obtained.



Scheme 32. Synthesis of AMFC from HMF using Pt nanoparticles followed by transaminase-catalyzed amination.

2.3.2. Metal-catalyzed hydrogenation and reduction reactions

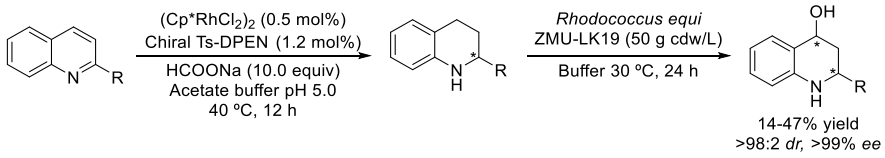
Conversely, the chemoenzymatic deracemization of secondary alcohols can be accomplished following other approaches employing an enzyme for oxidation purposes and a metal catalyst for the reduction step. In this context, an example has been recently reported by Fei, Turner and co-workers combining a non-selective iridium-catalyzed transfer hydrogenation process of a series of acetophenones, while a selective galactose oxidase was employed to selectively oxidize one of the resulting alcohol enantiomers (Scheme 33), rendering the enantioenriched (*S*)-1-arylethanols in a cyclic cascade.[106] Therefore, 4-nitroacetophenone (25 mM) was reacted with *rac*-Ir-*N*-(*p*-toluenesulfonyl)-1,2-diaminocyclohexane (TsCYDN, 3.2 mol%) as metal catalyst and HCOONa (24 equivalents) as hydrogen donor for the reduction step. After 1.5 h at 900 rpm and 37 ºC, the desired racemic alcohol was obtained with 94% conversion. Then, the (*R*)-selective galactose oxidase GOase $M_{3\text{-}5}$ (29.2 µM), horseradish peroxidase (HRP, 22 µM) and a catalase (0.4 µM) were added to produce the enantioselective oxidation, achieving the (*S*)-alcohol in 99% yield and 98% *ee* after 48 h of running this cascade reaction. This protocol was performed with other acetophenone derivatives, attaining variable results depending on the pattern aromatic substitution (25-99% conversion and 91-99% *ee*) after 24-48 h. Remarkably and as expected, better results can be achieved when attempting the asymmetric hydrogenation with a chiral catalyst such as the (*S*)-selective Rh-TSDPEN, for instance the enantiopure (*S*)-1-(4-nitrophenyl)ethan-1-ol was prepared in 96% yield in comparison with only a 70% obtained when employing the racemic metal complex under identical conditions (37 ºC, 48 h, 900 rpm).



Scheme 33. Production of optically active 1-arylethanols using a transfer hydrogenation-enzymatic oxidation cyclic cascade.

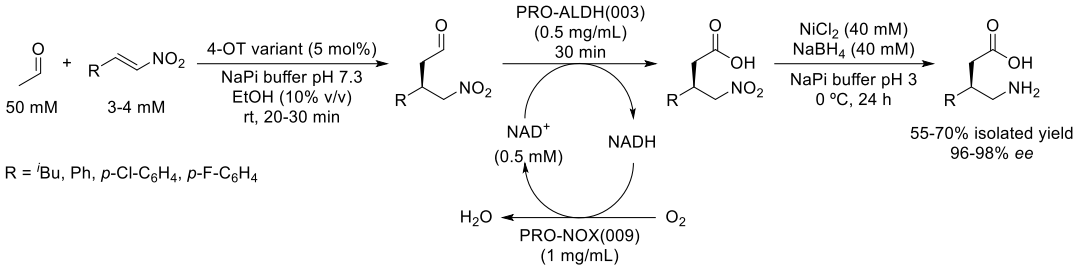
In another example described by Chen and co-workers, a rhodium complex was used as catalyst applied to an alkene reduction process.[107] The approach consisted of the sequential combination of a chiral Rh catalyst *in situ* generated, to produce the

stereoselective hydrogenation of 2-substituted-quinolines, which were later enzymatically hydroxylated at the C-4 position (Scheme 34). To achieve this aim, four 2-substituted quinolines (100 mM) were dissolved in acetate buffer pH 5.0 in the presence of a chiral Rh catalyst (10 mol%) and HCOONa (10 equiv) as hydrogen source, and after 12 h at 40 ºC, the reaction mixture was diluted to 6 mM using the same or phosphate buffer and divided in 17 portions. *Rhodococcus equi* ZMU-LK19 cells were suspended in each portion up to an optical density (OD) of 50 g cdw/L, and the mixture was incubated at 30 ºC for 24 h, obtaining 4 tetrahydroquinolin-4-ols containing different alkyl substitutions at the C-2 position with high *dr* (>98:2, in some cases after recrystallization) and *ee* (>99%), although with low yields (14-47%).



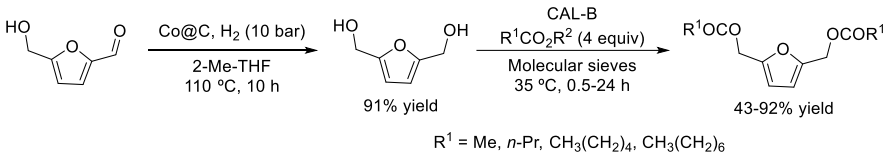
Scheme 34. Rh-catalyzed ATH and enzymatic hydroxylation to produce chiral 2-substituted tetrahydroquinolin-4-ols.

Drug synthesis is a challenging task, especially when enantioselective and greener approaches are pursued. In this context, chiral analogues of γ -aminobutyric acid (GABA) have been reported as relevant targets due to their application as anticonvulsants, antidepressants or to relieve neuropathic pain. Poelarends and co-workers have recently reported a one-pot three-step procedure consisting of the use of an evolved oxalocrotonate tautomerase for the Michael addition of acetaldehyde to a series of nitroalkenes (3-4 mM), followed by the enzymatic aldehyde oxidation using an aldehyde dehydrogenase (ALDH) into the γ-nitrobutyric acid intermediate, and nickel boride-catalyzed reduction of the nitro group to obtain four GABA derivatives (pregabalin, phenibut, baclofen and fluorophenibut) in a straightforward manner (Scheme 35).[108] The oxidized cofactor needed by ALDH was recycled using a nicotinamide oxidase (NOX). Due to the different pH and temperature windows where enzymes and metal catalysts appropriately worked, this protocol was performed in a stepwise manner.



Scheme 35. One-pot three-step chemoenzymatic approach to produce chiral GABA derivatives from nitroalkenes and acetaldehyde.

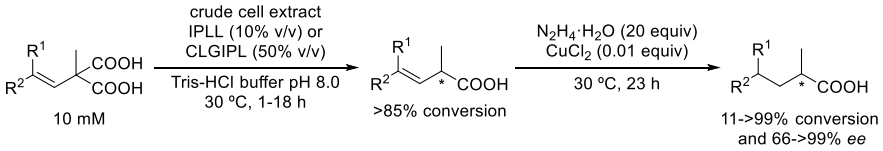
The production of valuable feedstocks from biomass is attracting great attention in recent years due to the chemical versatility of carbohydrates[109,110] or furan molecules such as 5-hydroxymethylfurfural (HMF) or 2,5-bis(hydroxymethyl)furan (BHMF).[111] For instance, Arias *et al.* have reported a chemoenzymatic cascade to produce diesters that can be used as biobased plasticizers. The approach involved the chemical hydrogenation of HMF (100 mM) using cobalt nanoparticles (Co@C) to produce BHMF at 110 ºC in a green solvent such as 2-methyltetrahydrofuran (2-Me-THF), which was later enzymatically esterified using CAL-B in combination with aliphatic carboxylic acids or vinyl esters as acyl donors at 35 ºC in the presence of molecular sieves to trap the water released (Scheme 36).[112] The process was firstly developed in batch, and after optimization of the reaction conditions, the system was developed in two fixed-bed reactors in a continuous mode, demonstrating high stability after 60 h. The results attained with vinyl esters such as vinyl hexanoate can be highlighted, improving the conversions obtained with carboxylic acids as acyl donors that led to partial enzyme deactivation.



Scheme 36. Hydrogenation of HMF using cobalt nanoparticles followed by lipase-catalyzed esterification with carboxylic acids or vinyl esters.

Another interesting example has been shown by Kourist's group, who developed a cofactor-free asymmetric decarboxylation of prochiral aliphatic arylmalonate derivatives using a series of stereocomplementary mutants of arylmalonate decarboxylase from

Bordetella bronchiseptica (AMDases IPLL and CLGIPL), followed by alkene reduction with diimide as reductant (Scheme 37).[113] The use of this hydrogen donor avoided a loss in the optical purity of the corresponding carboxylic acids, which was observed when using heterogeneous Pd(0) catalyst in hydrogenation reactions, the catalyst being formed *in situ* by reaction between hydrazine and copper(II) chloride. Therefore, both (*R*)- and (*S*)-carboxylic acids were obtained depending on the enzyme selectivity under mild reaction conditions, and a semipreparative scale reaction (110 mg of substrate) was carried out in a sequential manner to produce (*R*)-2-methylbutanoic acid with 98% *ee* and 83% yield after liquid-liquid extraction and column chromatography.



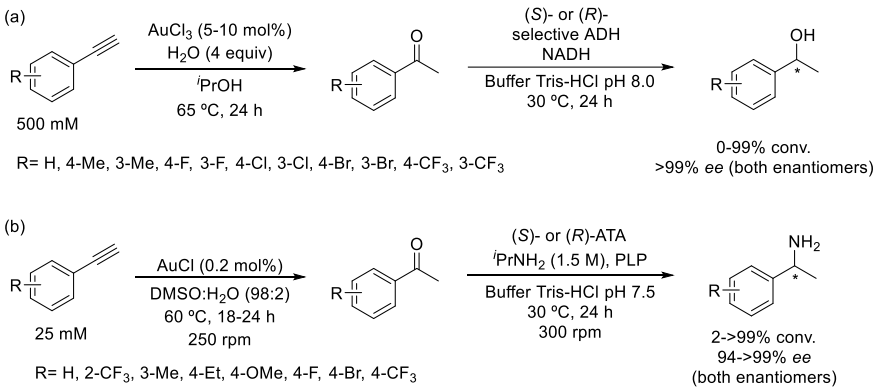
Scheme 37. Enzymatic decarboxylation of prochiral aryl malonates and chemical C=C reduction sequence to obtain enantiopure α-substituted carboxylic acids.

2.4. Hydration reactions

The functionalization of olefins and alkynes provides multiple possibilities to the organic chemists by developing fascinating regioselective reactions including carbonylations, hydroaminations, hydrocyanations and hydrations, among other types of transformations.[114] Remarkably, alkynes are suitable carbonyl surrogates through hydration reactions, transformations that occur with a perfect atom economy.[115] However, traditionally the hydration of alkynes has been developed under drastic reaction conditions, for instance through highly toxic oxymercuration reactions that also present low compatibility with many functional groups,[116] or in the presence of strongly acidic media.[117] Nowadays, the search for more efficient procedures combined with the use of less hazardous catalysts and reagents is actively on-going, appearing gold catalysts as suitable candidates for alkyne hydration reactions, which present the advantage to be compatible with different enzyme classes.[6]

A significant number of examples have recently appeared in the literature dealing with the development of chemoenzymatic concurrent and sequential processes involving a

metal-catalyzed alkyne hydration step. For instance, Mihovilovic and co-workers developed the stereoselective preparation of 1-arylethanols following a sequence based on the hydration of arylacetylenes and subsequent ketone enzymatic reduction.[118] Once the alkyne hydration step was optimized using the $AuCl_3$ (5-10 mol%), H_2O (4 equiv) and ^{i}PrOH as solvent, a series of ketones were obtained in moderate to excellent extent (49-99%). Later, the sequential process was studied using an ADH for the second step, furnishing the (*S*)-and (*R*)-alcohols in a full conversion range (0-99%) but in all cases in enantiomerically pure form (Scheme 38a). The overall process proceeded by developing first the alkyne hydration for 24 h at 65 ºC, and then cooling down the mixture to 30 ºC before the addition of the (*R*)-selective *Lactobacillus kefir* ADH or the (*S*)-selective ADH-A from *Rhodococcus ruber*, Tris-HCl buffer 350 mM pH 8.0 and NAD^+ to the reaction media. This sequential strategy resulted to be compulsory because of the requirements of high temperatures for this gold-catalyzed reaction, while the enzyme was not active at this temperature. Incubation for additional 24 h at 30 ºC allowed the synthesis of a set of eleven alcohols, even at semi-preparative scale: (*S*)-1-(4-fluorophenyl)ethanol (50 mg, 71% yield) and (*S*)-1-(3-chlorophenyl)ethanol (50 mg, 64% yield).

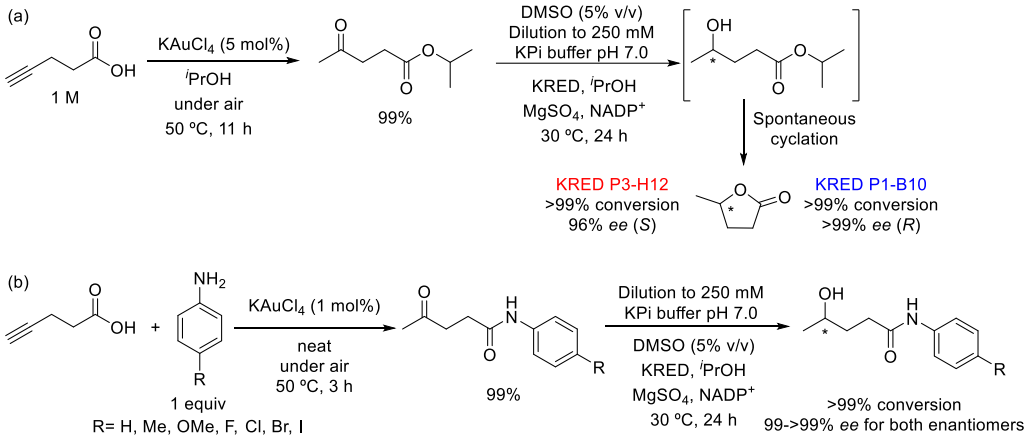


Scheme 38. Combination of Au-catalyzed arylacetylene hydration and stereoselective: (a) ketone bioreduction to produce enantiopure (*R*)- and (*S*)-1-arylethanols; and (b) transamination to synthesize (*R*)- and (*S*)-1-arylethanamines.

Rueping and co-workers have recently expanded the possibilities of gold chemistry, in this case using gold(I) chloride, by producing chiral amines from alkynes in a two-step sequential approach (Scheme 38b).[119] This strategy is based on the hydration of arylacetylenes (25 mM) at 60 ºC for 18-24 h, followed by addition of isopropylamine (1.5

M), PLP, a stereoselective ATA and Tris·HCl buffer (100 mM, pH 7.5) to develop the biotransamination of the acetophenone intermediates for additional 24 h at 30 ºC. Thus, the corresponding enantioenriched 1-arylethanamines were obtained in variable conversions (2->99%) and very high to excellent selectivities (94->99% *ee*). After optimization of the hydration reaction conditions and screening of commercial ATAs, it was found that very low loadings of AuCl (0.2 mol%) were required for the hydration step, while the access to both enantiomers for the eight synthesized 1-arylethanamines was feasible by the proper selection of the biocatalyst. The reactions were performed in more diluted conditions than the previous report from Mihovilovic in the case of the alcohol analogues, since the transamination was done after diluting to 5 mM the intermediate ketone. Finally, preparative transformations were successfully achieved to produce 1-phenylethanamine (92% yield, ATA-237) and 1-(4-methoxyphenyl)ethanamine (59% yield, ATA-254) both after column chromatography.

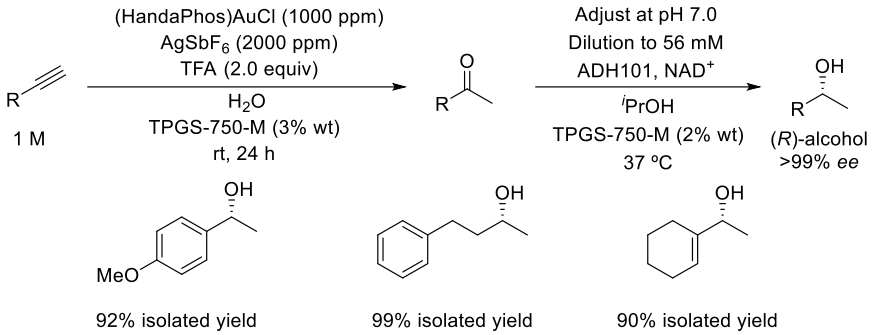
García-Álvarez, González-Sabín and co-workers reported the conversion of 4-pentynoic acid into enantiopure γ-valerolactone using potassium gold(III) chloride in alcohol media (2-propanol, ^{i}PrOH) followed by a bioreduction step (Scheme 39a).[88] Compared with previous observations in which a negligible effect by the action of palladium species was detected over enzyme catalysts, in this case the Au(III) catalyst caused a partial KRED inactivation (around 50% conversion was achieved). To solve this problem, the addition of DMSO (5% v/v) at the end of the first hydration/esterification step was performed. Thus, DMSO acting as a ligand, irreversibly bound to the gold catalyst, and consequently, did not have a negative effect on the bioreduction step. The first stage was performed at 50 ºC for 11 h, while the enzymatic reaction was achieved at 30 ºC for 24 h. Remarkably, the scope of this reaction was extended to the synthesis of chiral γ-hydroxy amides through a hydration-amidation-bioreduction sequence using only 1 mol% of the same gold(III) catalyst and various selective ADHs (KRED P1-A04, KRED P1-B02 or KRED P3-B03, Scheme 39b).



Scheme 39. Gold-catalyzed alkyne hydration and bioreduction sequential processes to obtain enantioenriched: (a) γ-valerolactone; and (b) γ-hydroxy amides.

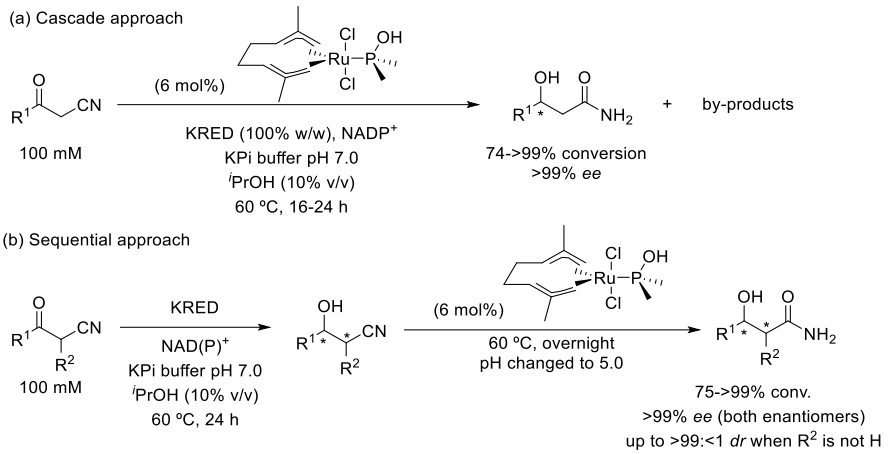
Moving again to gold(I) catalysis, Lipshutz and co-workers have reported the use of a micellar aqueous system to combine metal and enzyme catalysis.[120] Thus, micelles containing a hydrophobic core can accommodate the organic reactions, which are developed without the need of an additional organic solvent, and therefore can be performed in aqueous media. Interestingly, apart from alkyne hydration, Heck and Sonogashira couplings and 1,4-additions can be combined with the use of ADHs in a 100% aqueous media as will be commented in Section 2.6. Focusing on the hydration of arylacetylenes, DL-α-tocopherol methoxypolyethylene glycol succinate (TPGS-750-M) was employed as benign surfactant, its hydrophobic core composed of vitamin E, housing lipophilic substrates and transition metal in a compartmentalized nanoparticle that contains several micelles.[121] The surfactant resulted highly compatible with a commercial ADH (ADH 101 and ADH 112) in phosphate buffer, leading to faster reactions than in the absence TPGS-750-M and maintaining a perfect stereoselection (Scheme 40). This benefit is explained based on the difficulties caused due to the presence of water-insoluble substrates and products, while the micelles act as reservoir of these components, housing and releasing them along the time. Regarding the overall process, in a first step, the carbon-carbon triple bond (1 M) was hydrated in the presence of (HandaPhos)AuCl (0.5 mol%) as catalyst, silver hexafluoroantimonate ($AgSbF_6$, 1 mol%), trifluoroacetic acid (TFA, 2 equiv), TPGS-750-M (3% w/v) and water at rt for 24 h. Once the hydration was completed, and the ketone was generated, the reaction was diluted and the pH was adjusted to a value of 7 with NaOH 1 M, then ADH101, TPGS-750-M (2% w/v) and

phosphate buffer were added to obtain the desired enantiopure alcohols with high isolated yields (90-99%) after extraction and flash chromatography. Overall, the use of surfactants provides a great advance to traditional pure aqueous systems or even the use of cosolvents in terms of conversion values, enabling the use of low catalyst loadings.



Scheme 40. Use of a surfactant to compartmentalize a gold-catalyzed alkyne hydration-bioreduction sequence.

The hydration process is not only limited to the use of alkynes as substrates, but also to nitriles for the formation of amides, ruthenium-catalysts serving as valuable tools for the development of cascade reactions, for instance when combined with asymmetric ketone bioreduction.[122] The compatibility between Ru catalysts and KREDs was explored by means of concurrent cascades (Scheme 41a), or alternatively via sequential approaches in those cases with low KRED stereoselectivity or incomplete conversions (Scheme 41b). After deep optimization, six (hetero)aromatic β-ketonitriles at 100 mM concentration were converted into the desired β-hydroxy amides after 16-24 h at 60 ºC using a ruthenium complex (6 mol%) and a commercial KRED (KRED P2-C11, KRED P2-G03 or KRED P1-B10, 100% w/w) with the presence of ^{i}PrOH (10%, *v/v*) for cofactor recycling purposes in a phosphate buffer pH 7.0. Alternatively, for the sequential approach, firstly the bioreduction and later the hydration both were achieved at 60 ºC, furnishing the desired products with high conversions (75->99%) and excellent selectivities (>99%). Interestingly, DKR processes for cyclic and acyclic β-ketonitriles bearing a stereogenic carbon at α-position were successfully carried out, the change of the pH for the second step to a value of 5.0 being crucial for a more efficient process (>99% *ee* and 85:15 to >99:<1 for diastereomeric ratios).



Scheme 41. Chemoenzymatic synthesis of optically active β-hydroxy amides from β-ketonitriles using a cascade (a) or sequential (b) strategy combining a Ru-mediated hydration and a bioreduction.

The hydration of carbon-carbon and carbon-heteroatom triple bonds is an area worthy of research due to its perfect atom efficiency in the production of versatile intermediates. In this field, especially gold complexes are capable to produce valuable ketones, and while sequential approaches have been described, it is expected that their reaction conditions can be suitable for using enzymes such as ADHs in aqueous media with the presence of an organic cosolvent or a surfactant in a cascade manner. Without any doubt, this area is attracting high attention, the enzyme diversity providing a plethora of possibilities to produce complex and high-added value chiral organic molecules.

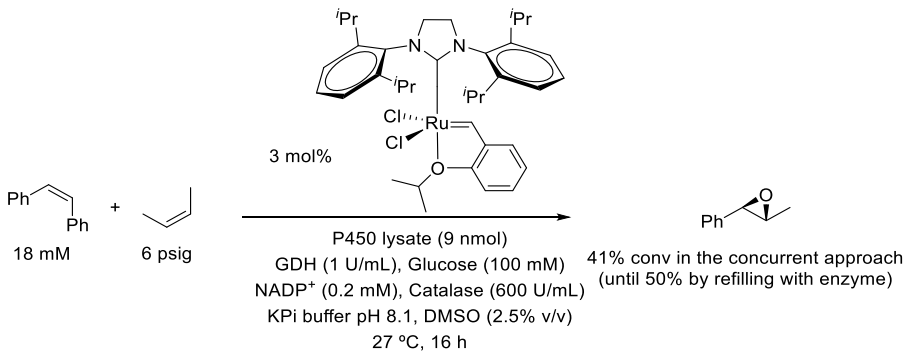
2.5. Metathesis reactions

Since olefin metathesis was discovered, it has become the most powerful tool to the formation of double bonds, allowing the creation of structural complexity from simple raw molecules.[3,123] Therefore, the use of different metal complexes has been found as a valuable tool for olefin metathesis reactions with importance in various fields such as petrochemistry, materials and pharmaceutical industries.[124] Before 2015, few research groups studied the combination of metal-catalyzed olefin metathesis and a biocatalytic step including esterases[125] or cytochromes P450.[126] Next, recent examples of concurrent

cross metathesis (CM) and ring-closing metathesis (RCM) processes with enzymatic transformations are deeply discussed.

2.5.1. Cross Metathesis (CM)

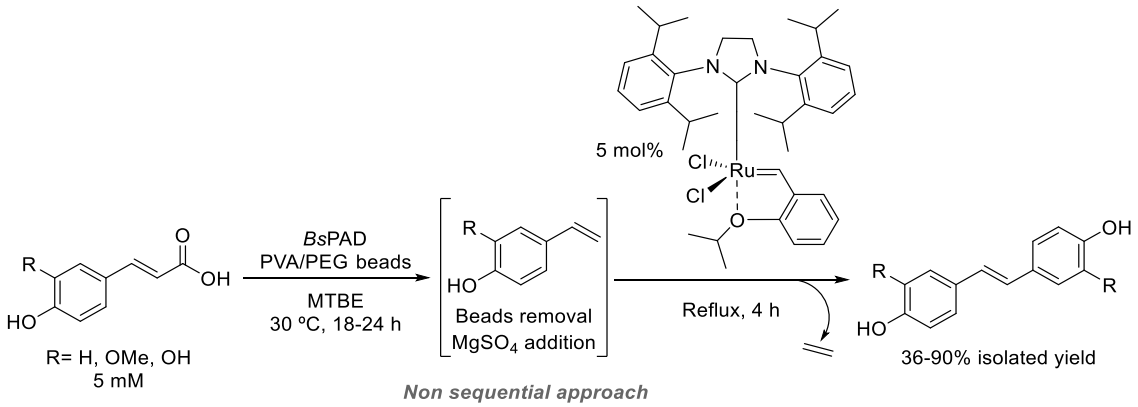
Hartwig, Zhao and co-workers expanded the potential of ruthenium catalysts and P450 enzymes,[126] developing in 2015 a cascade approach that combined a ruthenium-catalyzed alkene metathesis with an enzymatic epoxidation using a cytochrome P450 mutant.[127] After optimization of the reaction conditions to accommodate both catalytic reactions for a series of substrates, the tandem reaction starting from (*Z*)-stilbene was performed taking advantage that the enzyme did not recognize this substrate, so its epoxidation was not detected. The first step required a ruthenium complex (3 mol%) for the cross-metathesis reaction between (*Z*)-stilbene and gaseous (*Z*)-2-butene in phosphate buffer, adding later the corresponding P450-BM3 variant (RLYF, KT2, or RH47) from *Bacillus megaterium* and the cofactor recycling system (glucose/GDH). After 16 h at 27 ºC, the *trans*-epoxide was obtained with moderate conversion (41%), but with excellent (*E*)-selectivity using P450 KT2 (Scheme 42). Furthermore, the addition of a second batch of enzyme after 16 h increased the yield until 50% after 24 h. Unfortunately, tandem reactions involving (*Z*)-stilbene and (*E*)-3-hexene as substrates led to the corresponding *trans*-epoxide in low conversions (up to 25%), as part of a mixture of products such as β-ethylstyrene, highlighting the lower activity of the tested P450 variants for the epoxidation of this alkene intermediate.



Scheme 42. Concurrent alkene-metathesis and enantioselective bioepoxidation.

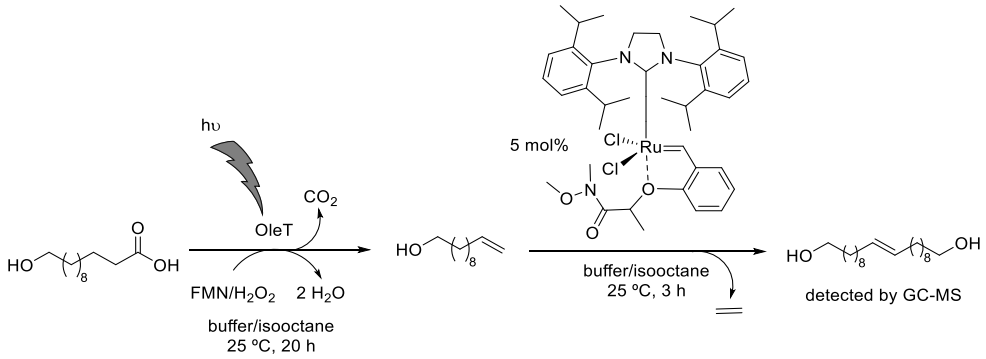
Kourist and co-workers have combined the activity of a ruthenium catalyst and an enzyme in an organic solvent and water, respectively, through the encapsulation of a phenolic acid

decarboxylase from *Bacillus subtilis* (*Bs*PAD). This biocatalyst was encapsulated in poly(vinyl alcohol)/poly(ethylene glycol) (PVA/PEG) cryogel, allowing to entrap the enzyme in an internal aqueous environment. Thus, the design of a one-pot sequential decarboxylation of coumaric acid derivatives using *Bs*PAD and the ruthenium-catalyzed homodimerization of the resulting olefins was developed as depicted in Scheme 43.[128] The coumaric acid derivative (5 mM) was dissolved in MTBE and the encapsulated *Bs*PAD was added, reacting the mixture until full conversion was achieved at 30 ºC (18-24 h). After removing the beads, the solvent was dried with $MgSO_4$, and the ruthenium catalyst (5 mol%) was added, to obtain three homodimerization products with low to very high isolated yields (36-90%) after refluxing the mixture for 4 h and column chromatography purification. From the three strategies tested: (i) Sequential two-pot reaction with intermediate isolation; (ii) combination of both steps in a one-pot biphasic system; or (iii) sequential one-pot reaction with encapsulated *Bs*PAD in pure organic system, then bead separation and intermediate drying, followed by the metathesis step; the latest resulted to be the best choice for synthetic purposes. Okuda, Schwaneberg, and co-workers designed a similar approach in aqueous medium. In this case, the key to success of this protocol was the immobilization of the Ru catalyst on an outer membrane protein from *E. coli* (Ferric hydroxamate uptake protein component A). Hence, after performing the decarboxylation step employing ferulic acid decarboxylase from *Saccharomyces cerevisiae* (FDC1) in a phosphate buffer:THF (98:2 v/v) mixture (1 h, 35 ºC), the supported ruthenium catalyst was added, obtaining after additional 4 h at 35 ºC the coupled products in good conversions (64-74%).[129]



Scheme 43. One-pot biocatalyzed decarboxylation of coumaric acids followed by ruthenium-catalyzed metathesis.

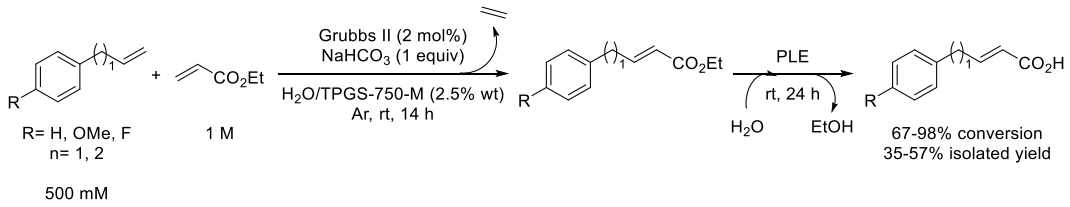
The fatty acid decarboxylase cytochrome P450 OleT from *Jeotgalicoccus* sp. is able to convert aliphatic long-chain fatty acids into the corresponding terminal alkenes. This enzyme has been successfully coupled with other (bio)catalysts to transform ω-functionalized fatty acids into other valuable compounds. Among the different reactions tested, a Ru-catalyzed metathesis transformation was studied starting from ω-hydroxylauric acid (Scheme 44).[130] Furthermore, a light-driven *in situ* formation of hydrogen peroxide was used for the enzymatic step, adding flavin mononucleotide (FMN) as photosensitizer under LED light. Then, different reaction set-ups were studied to perform the metal-catalyzed transformation, furnishing the dimer compound in a one-pot sequential manner utilizing isooctane as the second phase.



Scheme 44. One-pot sequential synthesis of a diol from ω-hydroxylauric acid through light-driven biocatalyzed decarboxylation followed by ruthenium-catalyzed metathesis.

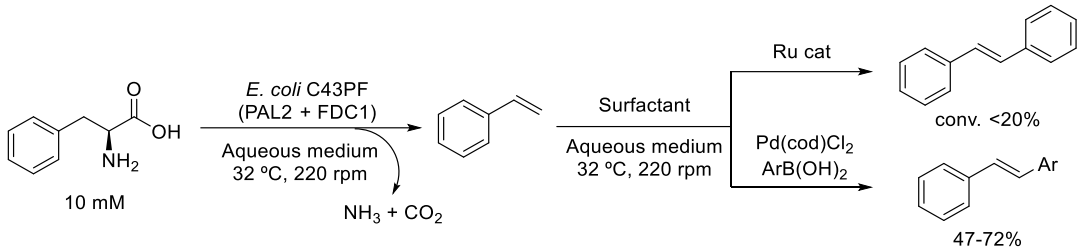
The addition of surfactants to form micelles in the reaction medium has been proved as an excellent approximation to perform metathesis-enzymatic transformations in a one-pot fashion. Hence, Hastings and co-workers recently described the coupling of the Ru-catalyzed metathesis of a series of (homo)allylarenes with ethyl acrylate, to obtain the corresponding α,β-unsaturated ester intermediates, which were subsequently hydrolyzed into the carboxylic acids by means of an esterase (Scheme 45).[131] The corresponding aromatic substrate (500 mM) reacted with ethyl acrylate (2 equiv) in the presence of sodium bicarbonate (1 equiv) and Grubbs second generation catalyst (2 mol%) in water containing TPGS-750-M (2.5% w/v) under inert atmosphere of argon. After 14 h at rt, pig liver esterase (PLE) was added to the reaction medium, and the mixture was left stirring additional 24 h. The products were isolated after column chromatography purification in moderate yields (35-57%). Remarkably, the concurrent one-pot reaction

between allylbenzene and ethyl acrylate was also successfully tested, providing the corresponding carboxylic acid (n= 1 and R= H) in 91% conversion. Again, this is a nice example of the potential that surfactants can have to develop chemoenzymatic cascade protocols under mild reaction conditions.



Scheme 45. One-pot sequential synthesis of α,β-unsaturated carboxylic acids combining a ruthenium-catalyzed metathesis with an esterase-catalyzed hydrolysis.

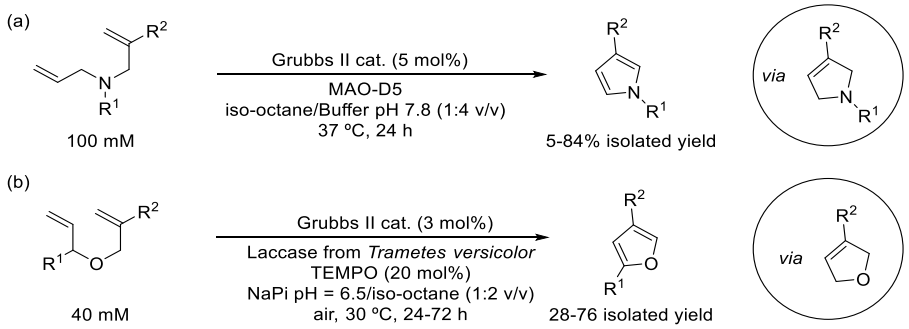
Starting from an α-amino acid, the synthetic possibilities of styrene were further explored by merging whole-cell biocatalysis with metal transition C–C coupling through two independent approaches (Scheme 46).[132] In both cases, styrene was produced by ammonia lysis of L-phenylalanine (10 mM) catalyzed by a phenylalanine ammonia lyase from *Arabidopsis thaliana* (PAL2) to furnish cinnamic acid, which was next decarboxylated by the action of a ferulic acid decarboxylase from *Saccharomyces cerevisae* (FDC1) in aqueous medium at 32 ºC. The expression of both proteins together was possible in *E. coli* C43PF, the use of a surfactant (TPGS-750-M, 2.5% w/v) improving the reaction outcome. Satisfyingly, these one-pot reaction conditions were compatible with the action of ruthenium Grubbs-type metathesis catalysts, and also with palladium(0) and palladium(II) species with a series of arylboronic acids, that allowed the metathesis and cross-coupling reactions, respectively, to produce a series of stilbene compounds. While the ruthenium complexes afforded low conversions (<20%), the best results were achieved with dichloro(1,5-cyclooctadiene)palladium(II) ($Pd(cod)Cl_2$) as catalyst (47-72%).



Scheme 46. Enzymatic production of styrene for one-pot transformations into stilbene derivatives using ruthenium or palladium catalysts.

2.5.2. Ring Closing Metathesis (RCM)

Olefin Ring-Closing Metathesis (RCM) is nowadays considered an effective strategy to obtain heterocycles in a straightforward manner,[133,134] its combination with biotransformations allowing the development of several concurrent methods towards functionalized heterocycles. For instance, Castagnolo and co-workers developed a chemoenzymatic cascade for the synthesis of pyrroles, combining the Grubbs second generation catalyst and a monoamine oxidase either from *Aspergillus niger* (MAO-N) or 6-hydroxy-D-nicotine-oxidase (6-HDNO) from *Arthrobacter nicotinovorans*.[135] In this work, the authors carried out a RCM approach to prepare different 3-pyrrolines from diallylamines and diallylanilines, and the subsequent MAO-catalyzed aromatization allowed the generation of different pyrroles. The RCM process was carried out in aqueous medium, finding *iso*-octane as the best cosolvent, which has also later a crucial role to prevent the interaction between the metal and enzyme catalysts mimicking the compartmentalization of cellular processes. Once this parameter was optimized, the cascade reaction was developed by dissolving the diallyl compound (100 mM) in a mixture of *iso*-octane and phosphate buffer pH 7.8 using Grubbs second generation catalyst (5 mol%) and mutant MAO-D5 in whole cell form, obtaining different pyrroles with low to high isolated yields (5-84%, Scheme 47a) after 24 h at 37 ºC. Otherwise, a sequential methodology was developed in which an additional amount of MAO-D5 was re-added after 6 h, slightly improving the results for those low-yielding substrates (*e.g.*, R^1= 4-^{i}Pr-C_6H_4 and R^2= H from 22 to 70% isolated yield).



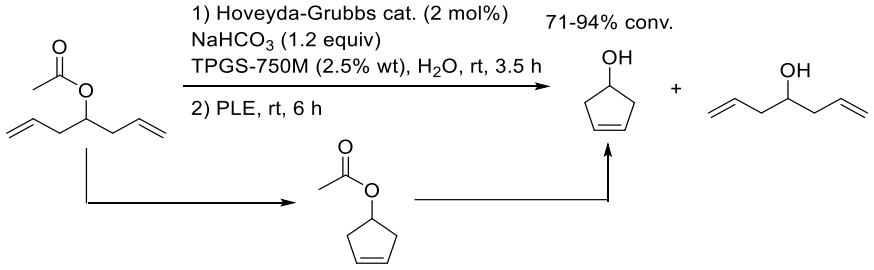
Scheme 47. One-pot cascades to synthesize (a) pyrroles, or (b) furans through a RCM-aromatization approach combined with the use of a MAO or a LMS, respectively.

The same research group later reported a chemoenzymatic metathesis/aromatization cascade for the synthesis of oxygen-containing heterocycles,[136] using a laccase mediator system as a biocatalytic system (Scheme 47b). The one-pot combination of Grubbs second generation catalyst for RCM of diolefins to generate 2,5-dihydrofuran derivatives, with the system composed by the laccase from *Trametes versicolor* and TEMPO catalyzing the subsequent aromatization, allowed the synthesis of the corresponding furan derivatives. Ambitioning the development of the cascade reaction, diallyl ethers (40 mM) were dissolved under aerobic conditions in a phosphate buffer pH 6.5/*iso*-octane (1:2, v/v) mixture, then Grubbs second generation catalyst (3 mol%), laccase from *Trametes versicolor* (67 U) and TEMPO (20 mol%) were added. After 24-72 h at 30 ºC, the desired compounds were isolated with low to moderate yields after liquid-liquid extraction and flash chromatography (28-76%).

Both RCM-aromatization processes represent a smart proof of concept, since simple and mild experimental conditions have been found for the cascade reaction of Grubbs catalysts and oxidative enzymes to obtain oxygenated and nitrogenated heterocycles. Perhaps, future research in this field will focus on increasing the number of chiral centers replacing aromatization processes with the enzymatic stereoselective modification of selected functionalities.

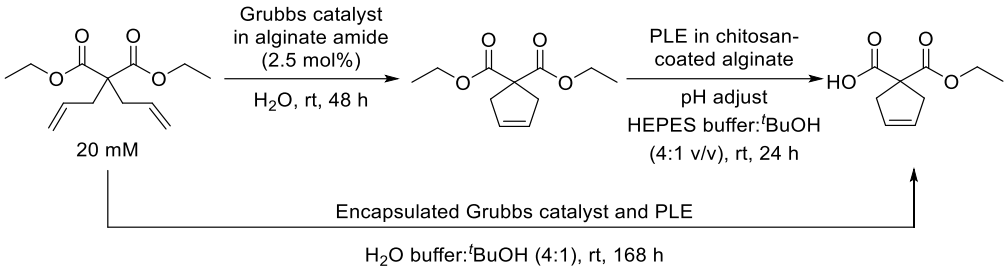
Hastings and co-workers reported the use of aqueous systems for the micellar ruthenium-catalyzed ring-closing of hepta-1,6-dien-4-yl acetate followed by PLE-catalyzed hydrolysis of the cyclopent-3-en-1-yl acetate (Scheme 48),[131] studying the possibility to develop the process in both sequential or concurrent fashion. The main limitation of the

concurrent approach was the occurrence of the enzyme-catalyzed hydrolysis the starting material in some extent, detecting the formation of the undesired hepta-1,6-dien-4-ol in 29%. For that reason, the sequential strategy was developed leading to 80% of the desired product in pure water, while the presence of the TPGS-750M surfactant (2.5% wt) increased the conversion up to a remarkable 94%.



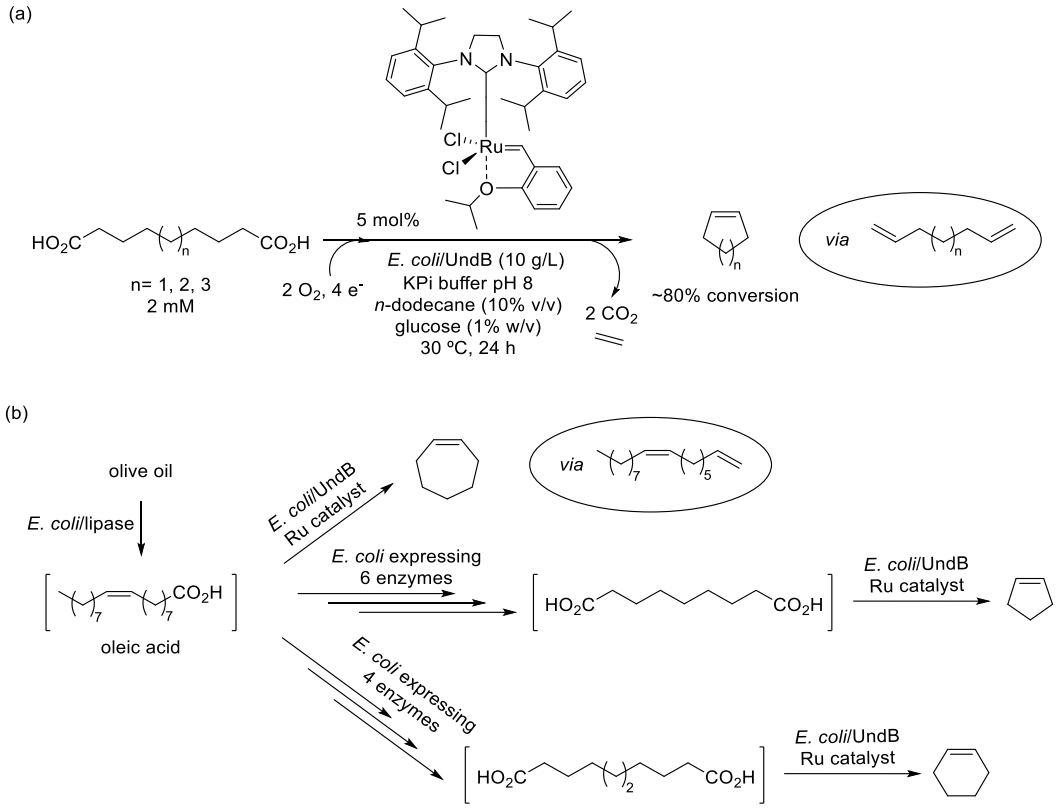
Scheme 48. RCM of hepta-1,6-dien-4-yl acetate followed by enzymatic ester hydrolysis.

Catalyst encapsulation is an interesting strategy to preserve its activity and develop one-pot procedures as previously shown in the cross-metathesis section.[128] For instance, it must be highlighted the work from Patel and co-workers, which have encapsulated Grubbs second generation catalyst in alginate amide and pig liver esterase (PLE) in chitosan-coated alginate beads, to develop a sequential RCM-ester hydrolysis in aqueous media (Scheme 49).[137] In this manner, after diethyl diallyl malonate (20 mM) ring-closing metathesis, the resulting cyclopent-3-ene-1,1-dicarboxylate was selectively hydrolyzed, affording the monocarboxylic acid. The one-pot process was run in a sequential manner to provide the adequate media for both catalysts, and improving previous results in the synthesis of malonic acid derivatives after 72 h.[125] The system could be recycled up to 7 cycles with excellent results. To minimize the side-product formation in the tandem process, the use of different types of encapsulations, co-encapsulation of both catalysts and alternatively their compartmentalization were investigated. The design of a specific large-sized bead (13.5 nm size) increased the diffusion facility of the Grubbs catalyst-containing shell, the substrate diffusing later into the PLE-containing core to attain a 84% total conversion and producing the desired carboxylic acid with a selectivity up to 75% after 168 h at room temperature (Scheme 49).[138]



Scheme 49. One-pot sequential RCM-ester hydrolysis using individual or encapsulated catalysts.

As an excellent recent contribution in this field, it must be highlighted the work developed by Wu, Ward and co-workers, who designed several enzymatic cascades expressing different biocatalysts in a host microorganism (*E. coli*) and coupled them with a final RCM step mediated by a Ru catalyst to obtain cycloalkenes from fatty acids.[139] After selecting the proper bio- and metal-catalysts, the first cascades studied were related with the transformation of several diacids into the corresponding cycloalkenes combining *E. coli* cells expressing a membrane-bound desaturase-like enzyme from *Pseudomonas* (UndB) and a (Hoveyda)-Grubbs ruthenium(II) catalyst (Scheme 50a). The outcome of the concurrent reactions highly improved in the presence of *n*-dodecane (10% v/v) as second phase. Thus, starting with the diacid (2 mM) and using *E. coli*/UndB (10 g/L) and the Ru catalyst (5 mol%) in a phosphate buffer pH 8 and *n*-dodecane mixture, it was possible to furnish the cycloalkenes in conversions around 80% after 24 h at 30 ºC. As a further extension of this methodology, the use of bio-based compounds as starting material such as oleic acid or olive oil was also attempted expressing a series of biocatalysts necessary to obtain the desired dialkene derivatives that reacted lastly in the RCM process. Especially challenging was the transformation of olive oil into cycloalkenes, since up to eight enzymes overexpressed in up to three different *E. coli* hosts were needed involving ester hydrolysis, alkene hydrations, decarboxylations, and alcohol, aldehyde and Baeyer-Villiger oxidations (Scheme 50b). Using olive oil (1 g/L) and under similar conditions than the ones shown before, up to 0.7 mM concentrations of cyclopentene, cyclohexene and cycloheptene could be detected. While still a proof-of-concept contribution, it is undeniable the potential that this work demonstrates by combining genetically modified microorganisms and metal catalysts in a concurrent manner to generate value to bio-based compounds.



Scheme 50. Cascade transformations to obtain cycloalkenes involving a RCM transformation from: (a) diacids and (b) olive oil.

2.6. C–C and C–X Cross-Couplings

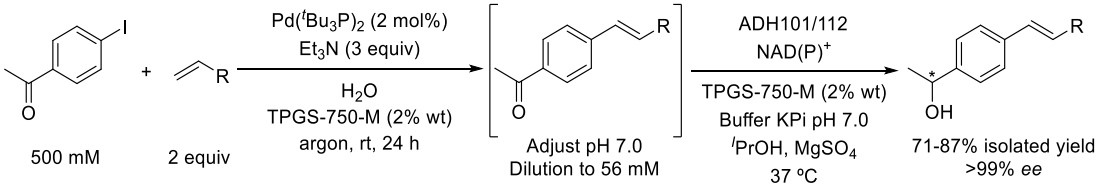
Since the discovery of cross-coupling reactions, their use in organic chemistry has exponentially increased becoming nowadays pivotal reactions in the synthesis of several (natural) product families.[2,140-145] This successful history found recognition in 2010 with the Chemistry Nobel award, personalized in the works from Heck, Negishi and Suzuki.[146] This section attempts to summarize the combination of metal-catalyzed cross-coupling reactions with the development of biocatalytic processes, therefore a classification based on the coupling transformation type has been made.

2.6.1. Heck reactions

The Heck-reaction, also called as Mizoroki-Heck reaction, is a C–C bond formation reaction known now for almost 50 years, which consists of the reaction between an

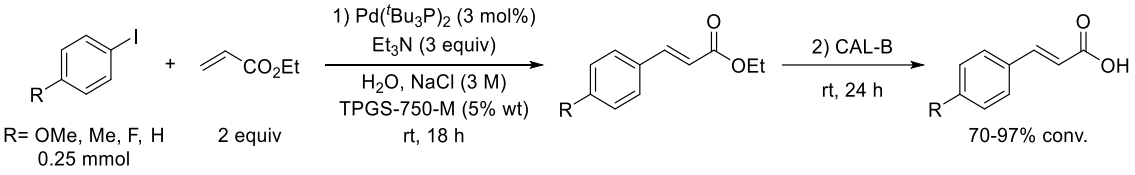
aryl/alkenyl halide or triflate with an alkene in the presence of a palladium catalyst and a base.[147] Herein, we will discuss the examples reported for the combination of this metal-catalyzed process with enzymatic transformations, which have involved the participation of ADHs or phenolic acid decarboxylases.

As previously mentioned in Section 2.4, the use of micelles has allowed the performance of different metal-catalyzed processes in aqueous media, providing an appropriate environment inside the micelle for the desired metal-catalyzed reaction. The achievements made in this field by Lipshutz's group were already discussed regarding the design of an alkyne hydration-bioreduction sequence.[120] In the same contribution, a sequential approach is also described consisting of a Heck reaction and a subsequent stereoselective bioreduction of the carbonyl group. The Heck coupling involves the reaction of an aryl iodide (500 mM), also containing a ketone moiety, with an alkene (2 equiv) in the presence of Et_3N (3 equiv) and a palladium catalyst (2 mol%). The process was performed in water using a TPGS-750-M surfactant (2% wt) under argon for 24 h at rt (Scheme 51). After that time, the pH and the substrate concentration were adjusted (pH 7.0, 56 mM) by addition of HCl and additional TPGS-750-M/phosphate buffer mixture, developing next the bioreduction of the ketone intermediate. The desired enantiopure 1-(4-substituted-phenyl)ethanols were obtained in good isolated yields (71-87%). Again, the use of a small percentage of surfactant allowed the development of a typical organometallic process in a purely aqueous medium, making it compatible with a biocatalytic transformation. Disclosing sequential strategies has opened many synthetic possibilities in an open field with perspectives towards the design of concurrent processes by minimizing interferences between both metal and enzyme catalysts.



Scheme 51. Heck cross-coupling reactions followed by stereoselective bioreduction of the ketone intermediate using a surfactant-aqueous mixture.

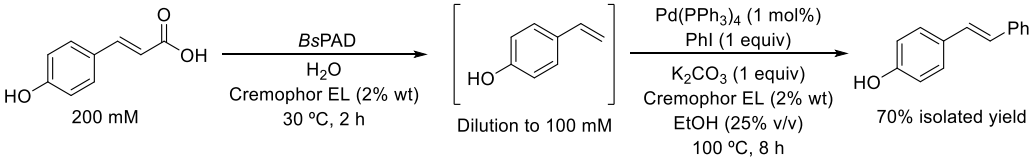
More recently, Hastings and co-workers have described a similar system for the Heck reaction between 4-iodoanisole and ethyl acrylate in the presence of ionic surfactants, which was followed in this case by enzyme-catalyzed hydrolysis of the cinnamate ester intermediate using either CAL-B or PLE.[131] Optimization of the sequence design was undertaken, finding the best results by diluting the reaction mixture after the Pd-catalyzed Heck coupling prior addition of CAL-B (Scheme 52). Thus, 4-methoxycinnamic acid was recovered in 97% yield through the sequential one-pot approach, while the concurrent reaction stopped with a 76% of ethyl cinnamate and 24% of cinnamic acid, clearly suggesting the inactivation of the hydrolase due to the metal presence. Finally, the chemoenzymatic strategy was successfully extended to the synthesis of other three cinnamic acids with variable substituents at the *p*-position of the aromatic ring (R= Me, F, H, 70-97% acid formation).



Scheme 52. Heck cross-coupling reaction followed by hydrolysis of the ester intermediate using a surfactant-aqueous mixture.

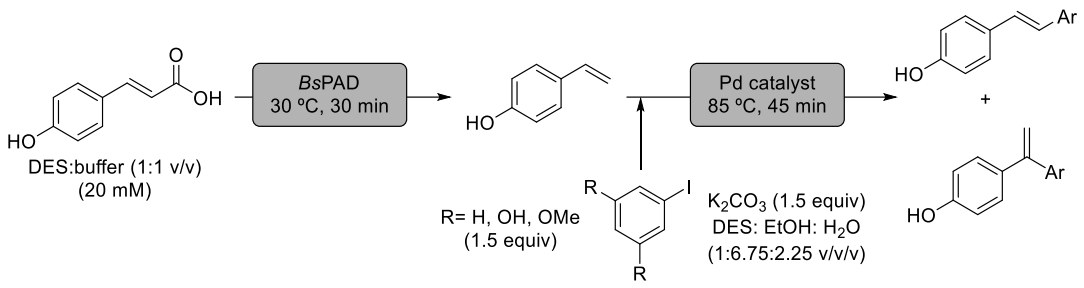
González-Sabín, García-Álvarez and co-workers have disclosed another sequential strategy, this time combining an enzymatic decarboxylation-Heck reaction sequence in a benign reaction medium.[148] In this approach, phenolic acid decarboxylase from *Bacillus subtilis* (*Bs*PAD) catalyzed the initial decarboxylation step of coumaric acid to obtain *p*-hydroxystyrene, which next reacted with tetrakis(triphenylphosphine)palladium [$Pd(PPh_3)_4$] and phenyl iodide for the Heck cross-coupling. Similarly, to the work reported by Lipshutz and co-workers,[120] the reaction was carried out employing a surfactant agent to generate micelles, Cremophor EL in this case. Water-deep eutectic solvent mixtures seemed to be adequate for the individual transformations, but after optimization of the enzymatic decarboxylation of coumaric acid (200 mM), best results were achieved in H_2O containing Cremophor EL (2% wt) for 2 h at 30 ºC. In this intermediate stage, the medium containing the resulting styrene was diluted to 100 mM prior developing the Heck reaction adding then the palladium catalyst (1 mol%) with

phenyl iodide (1 equiv) in the presence of potassium carbonate (1 equiv) and EtOH as cosolvent (25% v/v). After 8 h at 100 ºC, liquid-liquid extraction and flash chromatography on silica gel, (*E*)-4-hydroxystilbene was prepared with 70% isolated yield (Scheme 53). This strategy overcomes the limitation of similar decarboxylation-Heck coupling approaches, that involve the development of the decarboxylation in flow, although prior to the Heck coupling it is necessary to perform an extraction and column chromatography purification of the styrene intermediate.[149]



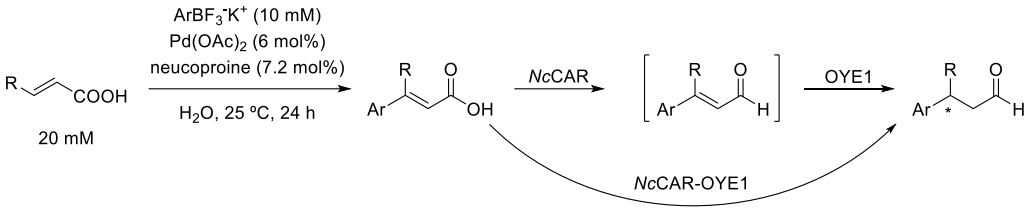
Scheme 53. Sequential decarboxylation-Heck coupling of *p*-hydroxycinnamic acid.

Following with the progress of decarboxylation-Heck coupling protocols in continuous mode using *Bs*PAD and a heterogeneous Pd catalyst, Grabner *et al.* developed a fully integrated two-step system to produce (*E*)-4-hydroxystilbene, the use of a DES [ChCl:Gly (1:2 mol/mol)] and phosphate buffer pH 6.0 (1:1 v/v) being crucial to overcome the solvent compatibility and enabling an increase of the substrate concentration from 5 mM in pure buffer to 20 mM in this system (Scheme 54).[150] The study involved the optimization of both individual reactions under batch and flow conditions, to later develop the cascade first in batch and stepwise mode, which required 30 ºC for the *p*-coumaric acid decarboxylation, filtration of the PAD alginate beads and development of the Heck coupling with iodobenzene (R= H) at 85 ºC. The cascade in flow was successfully operated for more than 16 h, achieving full conversion for the first step to produce 4-vinylphenol, although the second step was limited by the formation of 4-(1-phenylvinyl)phenol as major product, only observing the formation of (*E*)-4-hydroxy-stilbene in around 20% conversion. Some months later, the same authors reported the use of a 3D printed continuous stirred tank reactor that allowed an increase in the substrate concentration, and therefore of the productivity since the overall conversion was around 15% for the same reaction.[151] Fine-tuning of the reaction conditions of the original report,[150] and extension to other iodoaryl substrates allowed the synthesis of different stilbenes in 32-54% conversion under similar conditions.[152]



Scheme 54. Decarboxylation-Heck coupling cascade using *p*-coumaric acid and derivatives with iodoarenes in a DES-buffer system under flow conditions.

The production of fragrance aldehydes has been recently described by combining Heck coupling and subsequent carboxylate and alkene bioreduction steps with these two enzymatic activities being co-expressed (Scheme 55).[153] Firstly, the oxidative Heck coupling between a series of aryl trifluoroborates (10 mM) and substituted acrylic acids (20 mM) led to the corresponding cinnamic acid derivatives in low to moderate yields (8-76%) using $Pd(OAc)_2$ (6 mol%) and neocuproine (7.2 mol%) as ligand under aerobic conditions in H_2O at room temperature. Next, various strains were constructed expressing a carboxylic acid reductase from *Neurospora crassa* (CAR, *Nc*CAR) and a ERED from *Saccharomyces pastorianus* (OYE1), to perform two consecutive reduction steps. On the one hand, the carboxylate functionality was reduced to produce the corresponding cinnamaldehyde intermediates using the *Nc*CAR, and on the other hand, the alkene conjugated to the carbonyl group was converted into the corresponding aliphatic aldehyde by using the OYE1. Starting directly from cinnamic acid, the bienzymatic process occurred at high extent using *E. coli* cells co-expressing both enzymes to obtain 3-phenylpropanal, however, when trying to couple both processes, just a small quantity of cinnamaldehyde was detected, probably due to the molar excess of the acrylic acid present after the metal-catalyzed reaction. Therefore, authors envisaged CAR protein engineering or modification of the Heck conditions to avoid the use of an excess of the acrylic acid as possible ways to improve this cascade protocol.



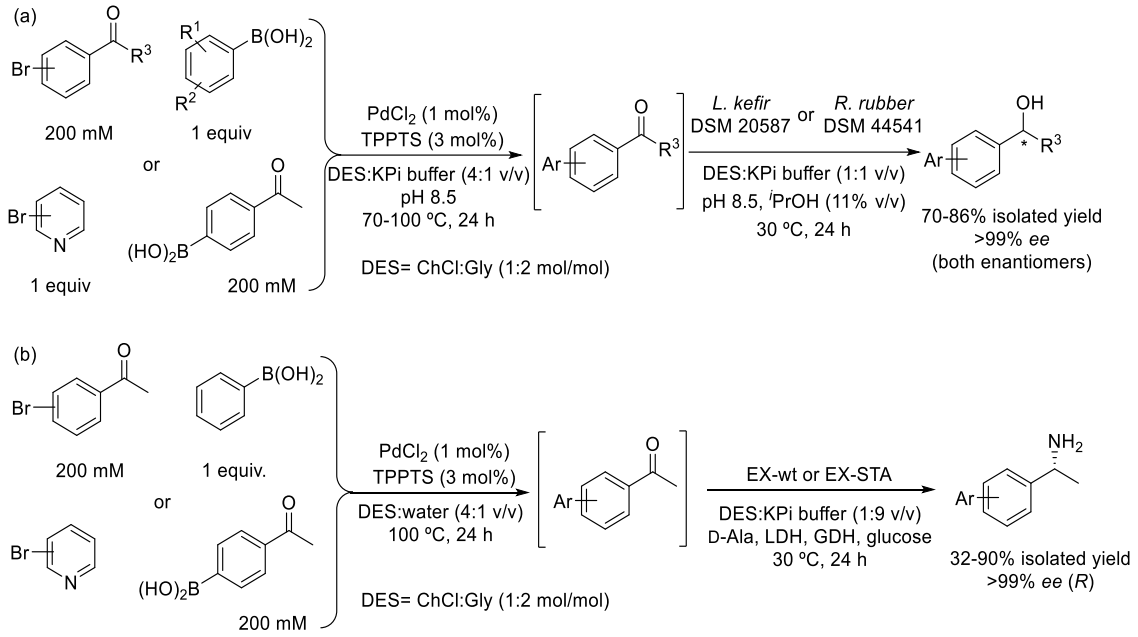
Scheme 55. Oxidative Heck coupling followed by CAR and ERED reductions to produce chiral aldehydes.

Overall, the combination of palladium-catalyzed Heck coupling with different biotransformations (bioreduction, decarboxylation or hydrolysis, among others) is limited by key factors including different optimal individual reaction temperatures and mutual catalyst inactivation and cross-reactivity when concurrent approaches are considered. Therefore, sequential processes are usually accomplished, which also include various strategies such as enzyme encapsulation, design of continuous mode sequences in separated modules, but in the less-favored scenarios also the isolation of reaction intermediates including purification steps or enzyme centrifugation need to be addressed.[149,154] Interestingly, the use of cheaper low-cost transition metals such as Co, Cu, Fe and Ni has been recently found as an alternative to conventional Pd-catalyzed Heck couplings,[155] therefore these discoveries open new possibilities to overcome current limitations in this area, and an important impact is expected in the near future.

2.6.2. Suzuki reaction

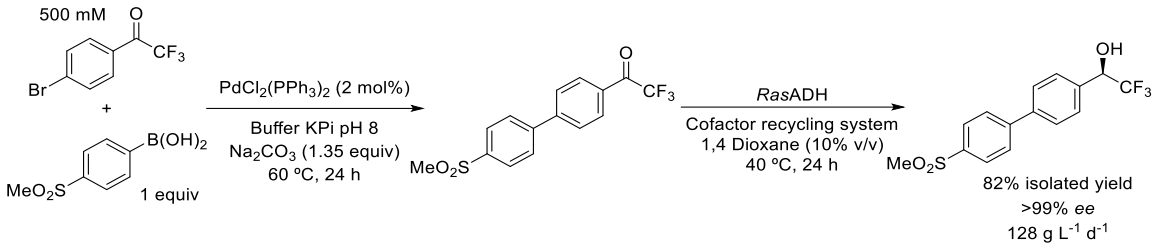
Suzuki reaction is perhaps the most popular cross-coupling process, for which palladium species are commonly employed as catalysts when combining boronic acids (or derivatives) and organohalides.[156,157] Next, recent combinations of Suzuki cross-coupling with biotransformations are reviewed, most of the examples corresponding to the use of palladium- and ADH-catalyzed transformations, which are known for more than a decade.[158-160] For instance, Gröger, González-Sabín and co-workers have developed the sequential synthesis of enantiopure biaryl alcohols using deep eutectic solvents-water as reaction media.[161] After optimization of both individual steps in these neoteric solvents, the sequential approach was envisaged by firstly dissolving equimolar amounts of the

organic bromide and the boronic acid containing a ketone moiety (200 mM) in a ChCl:Gly 1:2 (mol/mol) DES:phosphate buffer pH 8.5 mixture (4:1 v/v), in the presence of palladium(II) chloride (1 mol%) and tris(3-sulfonatophenyl)phosphine hydrate sodium salt (TPPTS, 3 mol%, Scheme 56a). After 24 h at 70 ºC or 100 ºC, the reaction was cooled down to room temperature and diluted (~110 mM) adding the buffer and 2-propanol as hydrogen donor to perform the stereoselective bioreduction using either ADH from *Lactobacillus kefir* (*Lk*ADH) or from *Rhodococcus ruber* (ADH-A). Additional 24 h at 30 ºC led to a series of enantiopure biaryl alcohols with high isolated yields (70-86%) after liquid-liquid extraction and column chromatography. The practical applicability of this approach was demonstrated by scaling-up the synthesis of (*S*)-1-(4-(pyridin-3-yl)phenyl)ethanol (364 mg, 85%) and (*R*)-1-[4-(pyridin-4-yl)phenyl]ethanol (368 mg, 80%). A similar procedure was later described by the same authors but developing an asymmetric biotransamination of the ketone intermediate using a transaminase.[162] The same DES was employed (ChCl:Gly 1:2 mol/mol) using two transaminases from *Exophiala xenobiotica* (wild type EX or T273S mutant EX-STA) and D-alanine as amine donor, furnishing enantiopure (*R*)-biarylamines with low to high isolated yields (32-90%, Scheme 56b).



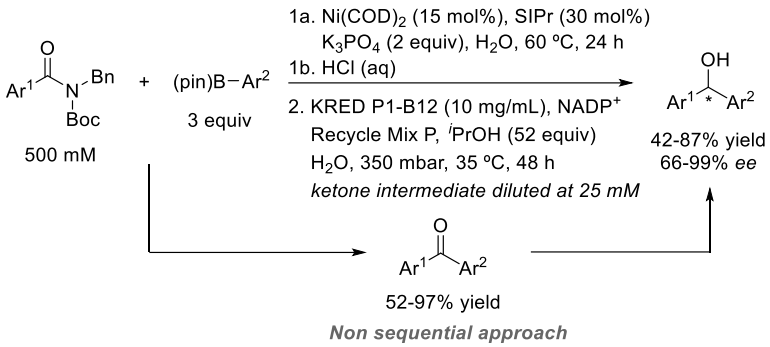
Scheme 56. Sequential chemoenzymatic synthesis involving Suzuki cross-coupling and stereoselective biotransformations: (a) Bioreduction to produce biaryl alcohols; (b) Biotransamination towards biarylamines.

Our research group has taken advantage of this methodology for the synthesis of a precursor of Odanacatib, a Cathepsin K inhibitor (cysteine protease expressed in osteoclasts that are the cells responsible for bone resorption), using a Suzuki cross-coupling/asymmetric bioreduction sequence.[163] Due to the fact that the cascade approach was not possible because the enzymatic crude inactivated the palladium catalyst, while the boronic acid inhibited the ADH, a sequential approach was developed at 500 mM ketone concentration employing 1 equiv of the corresponding boronic acid, 2 mol% of bis(triphenylphosphine)palladium dichloride, 1.35 equiv of sodium carbonate in a phosphate buffer pH 8.0 (Scheme 57). After 24 h at 60 ºC, the ketone intermediate was formed and the pH slightly decreased until 7.5-8.0, resulting optimum for the bioreduction with the ADH from *Ralstonia* sp. (*Ras*ADH). Then, the cofactor recycling system and 1,4-dioxane (10% v/v) were added and the reaction kept for additional 24 h at 40 ºC, obtaining the desired enantiopure alcohol in 82% yield and with high productivity (128 g L^{-1} d^{-1}).



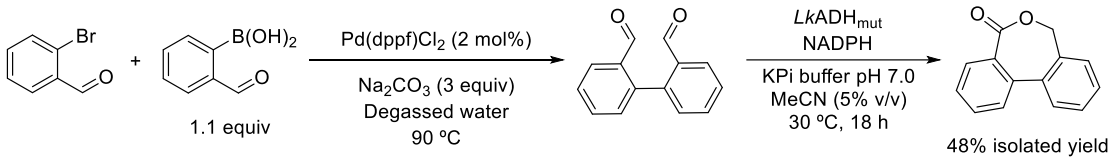
Scheme 57. Sequential Suzuki cross-coupling/*Ras*ADH-catalyzed bioreduction for the stereoselective synthesis of an Odanacatib alcohol precursor.

In addition to palladium species, nickel catalysts are also able to catalyze Suzuki-Miyaura couplings. In 2019, Garg and co-workers developed a sequential one-pot chemoenzymatic approach to yield enantioenriched alcohols starting from amides in aqueous media.[164] This is one of the few reports showing Ni-catalyzed Suzuki-Miyaura reactions in aqueous medium, which is necessary for the bioreduction process, just requiring toluene as additive in some cases. The coupling between an aromatic amide (500 mM, 1.0 equiv) and an arylboronate (3.0 equiv) was accomplished by $Ni(COD)_2$ (15 mol%) in water in the presence of 1,3-bis(2,6-diisopropylphenyl)-4,5-dihydroimidazol-2-ylidine (SIPr, 30 mol%) as ligand and K_3PO_4 (2.0 equiv) as base at 60 ºC during 24 h. In few selected examples, toluene (25% v/v) was added to favor the substrate solubility. Once the C–C coupling was finished the solution was quenched with an aqueous HCl 1 M solution, and the bioreduction of the formed diaryl ketones was performed adding commercially available KRED-P1-B12 (40 mg) under suitable conditions for cofactor regeneration purposes [^{i}PrOH (52.0 equiv) and commercial recycle Mix P]. Then, the reaction was diluted with water to have a substrate concentration of 25 mM and heated up to 35 ºC under reduced pressure (350 mbar) to remove the acetone formed. Thus, the equilibrium was shifted towards enantioenriched alcohol formation, stirring the reaction at 900 rpm during 24 h. Finally, additional $NADP^+$ (0.044 equiv) was added and stirred under the same conditions for additional 24 h (Scheme 58). The practical utility of this approach was demonstrated through the synthesis of both orphenadrine enantiomers, although this drug employed for the treatment of muscle pain is administrated as a racemate.



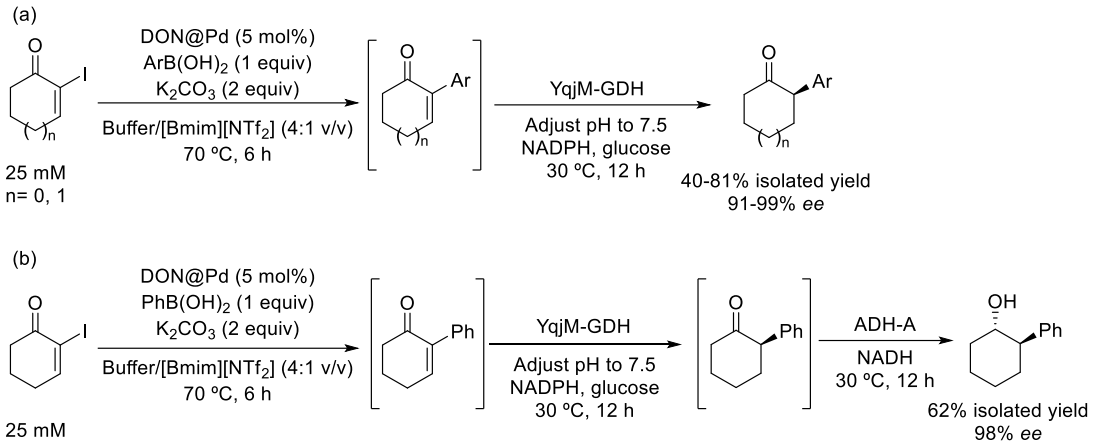
Scheme 58. One-pot sequential Ni-catalyzed Suzuki-Miyaura coupling and stereoselective ketone bioreduction.

Another example combining a Suzuki cross-coupling reaction and a biocatalytic step has been recently published by Hall and co-workers (Scheme 59).[165] In this case, a regioselective self-sufficient Tishchenko-type reaction, a disproportionation reaction that leads to the formation of lactones through a dimerization step of dialdehydes,[166] has been achieved through formal intramolecular hydride transfer using an ADH. The focus has been put in the enzymatic process, although the sequential approach has been demonstrated by developing the Suzuki cross-coupling reaction between 2-bromobenzaldehyde and 2-formylphenylboronic acid (1.1 equiv) in the presence of Na_2CO_3 (3 equiv) and [1,1'-bis(diphenylphosphino)ferrocene]palladium(II) dichloride ($Pd(dppf)Cl_2$, 2 mol%) at 90 ºC in degassed water, followed by the Tishchenko-type reaction after cooling the mixture to room temperature, dilution and pH adjustment with phosphate buffer pH 7.0 until 15 mM intermediate concentration, and addition of a cosolvent such as MeCN (5% v/v), a *Lactobacillus kefir* ADH mutant (*Lk*ADH_{mut}) and NADPH (0.5 mM), gratifyingly dibenzo[c,e]oxepin-5(7H)-one was isolated with 48% isolated yield after 18 h at 30 ºC after liquid-liquid extraction and chromatographic purification.



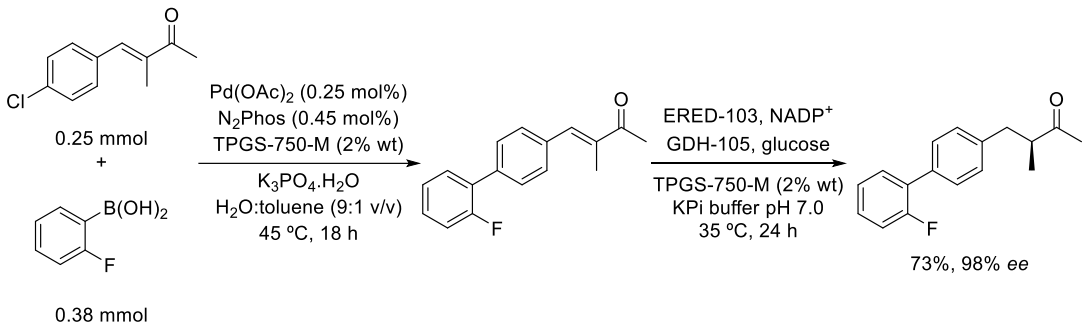
Scheme 59. Sequential chemoenzymatic approach combining a Suzuki-Miyaura coupling and an ADH-catalyzed lactonization.

Furthermore, Suzuki cross-coupling reaction has been combined not only with a carbonyl bioreduction step but also with a biocatalytic C=C asymmetric reduction. For instance, Liu, Jiang and co-workers described the sequential chemoenzymatic synthesis of tertiary α-aryl cyclic ketones through Pd-catalyzed Suzuki coupling and ene-reductase alkene asymmetric reduction step (Scheme 60a).[167] To develop the integration of both transformations in one-pot, the Pd catalyst was immobilized into dendritic organosilica nanoparticles (DON@Pd) with high specific surface area. Thus, the starting 2-iodocycloenone (25 mM) was placed in the presence of DON@Pd (5 mol%), the corresponding arylboronic acid (1 equiv), K_2CO_3 (2 equiv) and a mixture of buffer Tris-HCl (50 mM, pH 7.5) and the water-immiscible ionic liquid 1-butyl-3-methylimidazolium bis(trifluoromethylsulfonyl)imide ([Bmim][NTf_2]) in a 4:1 v/v ratio. After 6 h at 70 ºC, the mixture was cooled to 30 ºC, the pH adjusted to 7.5, and then whole cells co-expressing YqjM ERED from *Bacillus subtilis* and GDH from *Bacillus megaterium*, NADPH (0.2 mM) and glucose (200 mM) were added, furnishing the desired α-aryl cycloketones with moderate to high yields (40-81%) and high enantiomeric excess (>91%). To complement this research, a variant from YqjM displaying the opposite stereoselectivity was also successfully applied to obtain the corresponding ketone enantiomer. Interestingly, even a second biotransformation could be combined such as the carbonyl reduction of the resulting α-aryl cyclic ketone, so after the sequential chemoenzymatic Suzuki C=C reduction protocol was finished, ADH-A from *Rhodococcus ruber* and NADH were added leading to (1*S*,2*R*)-2-phenylcyclohexan-1-ol in 62% isolated yield and 98% *ee* after liquid-liquid extraction and chromatographic purification (Scheme 60b). Recycling studies of the Pd catalyst alone or co-immobilizing both metal- and biocatalysts (YqjM and GDH) were performed, observing that after 5 cycles these preparations were still active.



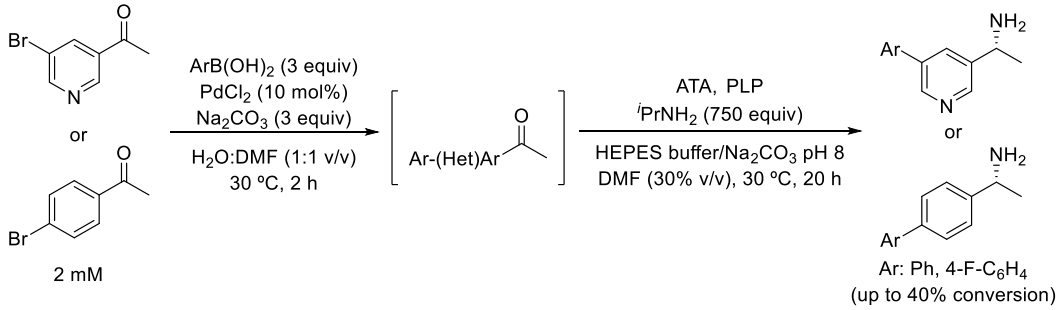
Scheme 60. Sequential Suzuki cross-coupling and alkene bioreduction (strategy a), including a final ADH-catalyzed bioreduction step of the α-aryl cyclic ketone (strategy b).

More recently, Lipshutz and co-workers have reported a one-pot sequence consisting of the palladium-catalyzed Suzuki-Miyaura coupling of 2-fluoroboronic acid (1.52 equiv) with (*E*)-4-(4-chlorophenyl)-3-methylbut-3-en-2-one, followed by the ERED-catalyzed reduction of the resulting olefin (Scheme 61).[168] The cascade was developed in sequential manner and in aqueous medium with the presence of the surfactant TPGS-750-M (2% wt) to assure the correct action of the enzyme in the second step. Once that the C–C coupling was achieved with $Pd(OAc)_2$ (0.25 mol%) and adding a phosphine ligand (0.45 mol%) in a water:toluene mixture (9:1 v/v), after 18 h at 45 ºC, the pH was adjusted to 7.0 to perform the reduction process using a commercial enzyme in a selective manner (98% *ee*) and a good overall yield (73%). Also in this contribution, other multi-step sequential protocols were successfully achieved in one-pot: (i) cyanation of (*E*)-4-(4-bromophenyl)-3-methylbut-3-en-2-one with $Zn(CN)_2$ (0.55 equiv) catalyzed by Xantphos palladacycle, followed by ERED-103-catalyzed reduction of the olefin, and subsequent carbonyl bioreduction using the commercial ADH-101; (ii) ERED reduction of (*E*)-4-(2-nitrophenyl)-3-methylbut-3-en-2-one, palladium-catalyzed hydrogenation of the nitro moiety under hydrogen atmosphere with concomitant intramolecular cyclization and imine reduction, and final chemical acetylation with acetic anhydride to afford a N-protected tetrahydroquinoline.



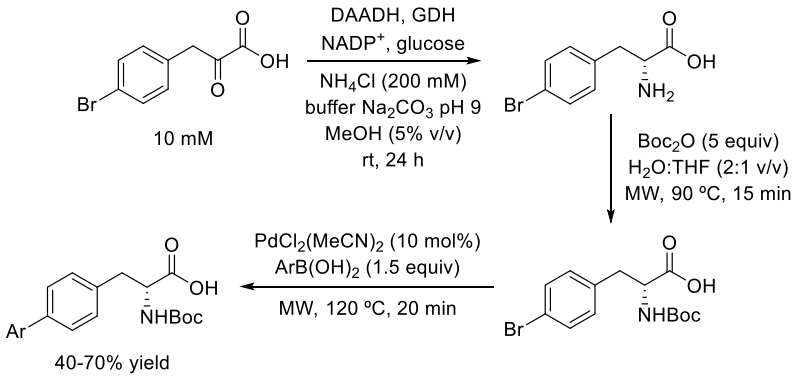
Scheme 61. Sequential Suzuki-Miyaura cross-coupling and alkene bioreduction.

As previously mentioned,[162] chemoenzymatic sequential approaches combining Suzuki cross-couplings and transaminases to synthesize chiral amines have also been described. For instance, Bornscheuer and co-workers developed an efficient system employing engineered transaminases to produced chiral biaryl amines (Scheme 62).[169] Thus, a palladium phosphine ligand free ($PdCl_2$, 10 mol%) was used as catalyst for the coupling between a brominated ketone (2 mM) and a phenyl boronic acid (1.5 equiv) in a water:DMF mixture (1:1 v/v) with the presence of sodium carbonate (3 equiv). The coupling reaction was performed for 2 h at 30 ºC, later after dilution with buffer, an engineered transaminase from *Aspergillus fumigatus* (4CHI-TA) and isopropylamine as amine donor (750 mM) were added, obtaining various biaryl (*R*)-amines (>99% *ee* and up to 88% conversion) after reacting for additional 20 h at 30 ºC. Furthermore, its application in flow system was successfully developed to furnish 1-(5-phenylpyridin-3-yl)ethan-1-amine with moderate conversion (43%) after 3.5 h of residence time in a carrier in which the variant 4CHI-I146A was immobilized via metal affinity resin.



Scheme 62. Sequential Suzuki cross-coupling between brominated ketones and aryl boronic acids, followed by ketone biotransamination.

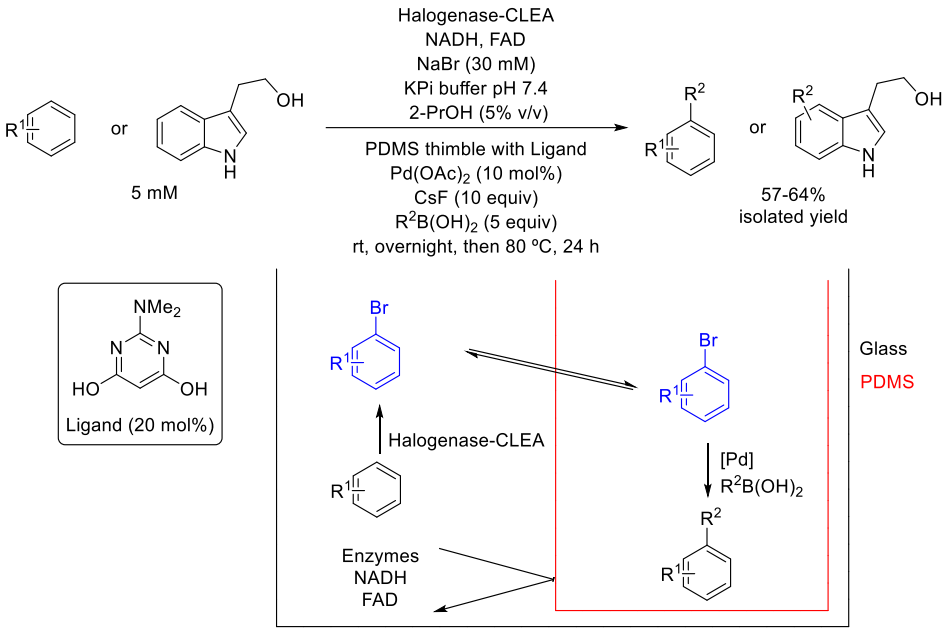
Asymmetric chemoenzymatic multistep synthesis of biaryl amino acids such as L- and D-biarylalanines is also possible as reported by Turner and co-workers through a sequence consisting of an asymmetric reductive amination of the starting keto acid, nitrogen protection and subsequent Suzuki cross-coupling reaction, involving the last two steps microwave irradiation (Scheme 63).[170] Hence, 3-(4-bromophenyl)-2-oxopropanoic acid (10 mM in aqueous medium) was transformed after 24 h into (*R*)-2-amino-3-(4-bromophenyl)propanoic acid employing a D-amino acid dehydrogenase (DAADH) from *Corynebacterium glutamicum* and a glucose dehydrogenase to recycle the nicotinamide cofactor. Then THF, deionized water and Boc_2O (5 equiv) were added for *N*-Boc protection under microwave irradiation at 90 ºC for 15 min. After cooling the mixture, the corresponding arylboronic acid (1.5 equiv) and $PdCl_2(MeCN)_2$ (10 mol%) were added to the recipient, and the mixture was heated at 120 ºC for additional 20 min under microwave conditions. The desired D-biarylalanines were obtained with moderate to high yields (40-70%). Alternatively, L-derivatives were synthesized in 33-65% yield, using a variant from phenylalanine ammonia lyase from *Anabaena variabilis* (*Av*PAL), although this chemoenzymatic approach was not a cascade reaction due to the need of removing ammonium salts from the buffer and unreacted starting material by adsorption on an ion-exchange resin, before the last two steps (*N*-Boc protection and Suzuki-cross coupling).



Scheme 63. Chemoenzymatic one-pot sequential synthesis of D-biarylalanines employing DAADH, *N*-Boc protection and Suzuki cross-coupling reaction.

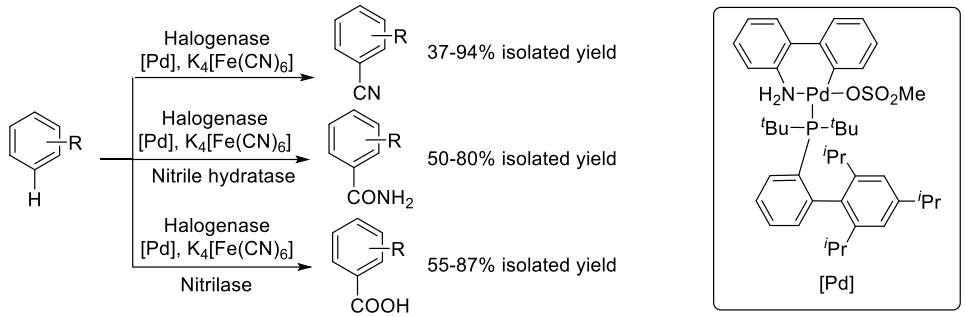
Selective C–H activation has transformed organic chemistry due to its multiple synthetic applications, metal catalysis being traditionally used for this type of activation. However, several (evolved) halogenases have recently appeared as suitable biocatalysts for this

transformation under mild reaction conditions and able to be part of cascades involving the Suzuki reaction.[171] However, after the selective halogenation of tryptophan, biocatalyst filtration is required to perform the metal-catalyzed cross-coupling.[172] Greaney, Micklefield and co-workers have overcome this limitation proposing a compartmentalized cascade approach for the regioselective halogenation of (het)arenes (2-5 mM) and their subsequent palladium-catalyzed Suzuki coupling with boronic acids.[173] In order to develop the cascade approach, the enzymatic halogenation was carried out using CLEAs containing different halogenases, namely, tryptophan 5-halogenase from *Streptomyces rugosporus* (PyrH), tryptophan 7-halogenase from *Lechevalieria aerocolonigenes* (RebH) and tryptophan 6-halogenase from *Streptomyces toxytricini* (SttH),[174] which allowed to greatly reduce the necessary cross-coupling palladium catalyst loading (10 mol%) in comparison to the use of pure protein (Scheme 64). Cesium fluoride (10 equiv) was added to improve the reaction outcome and the boronic acid derivative was used in molar excess (5 equiv). As halogenating agent, NaBr (30-100 mM) was employed. After studying different set-ups, a compartmentalization strategy using polydimethylsiloxane thimbles was chosen as the simplest one as it allowed to separate the enzyme from the palladium catalyst, solving the inhibitions and incompatibilities between catalysts and additives, and yielding four aromatic compounds in moderate isolated yields (57-64%) and exquisite regioselectivity.



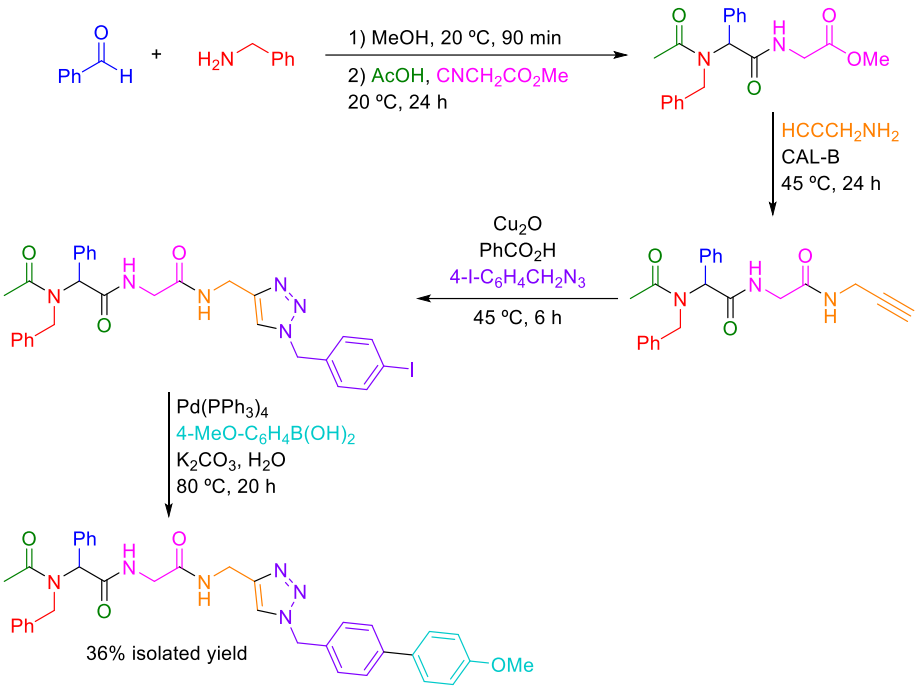
Scheme 64. PDMS compartmentalization for the regioselective cascade biohalogenation-Suzuki cross-coupling of arenes.

The same research group has found another interesting application of halogenases, displaying the possibility to prepare several nitrile aromatic compounds by the combination of a FAD-dependent halogenase (Fl-Hal) with Pd catalysis using a non-toxic cyanide source such as potassium ferrocyanide ($K_4[Fe(CN)_6]$).[175] Thus, a series of aromatic substrates (1.5-3 mM) reacted with different halogenases (wild-type or mutants) and NaBr (30 mM) in phosphate buffer and MeOH or ^{i}PrOH (95:5 v/v) overnight at rt (Scheme 65). Once the enzymatic halogenation finished, the cyanation reagents, ^{t}BuXPhos-Pd-G3 (10 mol%) and $K_4[Fe(CN)_6]$ (50 mol%), and THF as cosolvent (16.7% v/v) were added, heating the mixture under nitrogen overnight at 80 ºC. While different setups were studied, the best results were attained using enzymatic CLEA preparations. The corresponding nitriles were synthesized with 37-94% isolated yield depending on the aromatic structure. Additional functionalization was feasible by converting the cyano group into amides (50-80%) or carboxylic acids (55-87%) using nitrile hydratases or nitrilases, respectively, after THF evaporation.



Scheme 65. Two- and three-component chemo-biocatalytic systems involving cyanation of aromatic compounds using a halogenase and palladium catalysis.

To end this section, a consecutive Ugi-four component reaction, CAL-B-catalyzed aminolysis of the resulting ester with a propargylic amine, copper-catalyzed alkyne-azide cycloaddition (CuAAC) and final Suzuki coupling with 4-methoxyphenylboronic acid is presented, representing a one-pot seven-component reaction developed in a sequential manner (Scheme 66).[176] Optimization of individual and consecutive transformations were the key to provide a straightforward protocol to synthesize a 1-biaryl-4-triamide disubstituted triazole in 36% isolated yield after two purification protocols, a column chromatography followed by recrystallization.

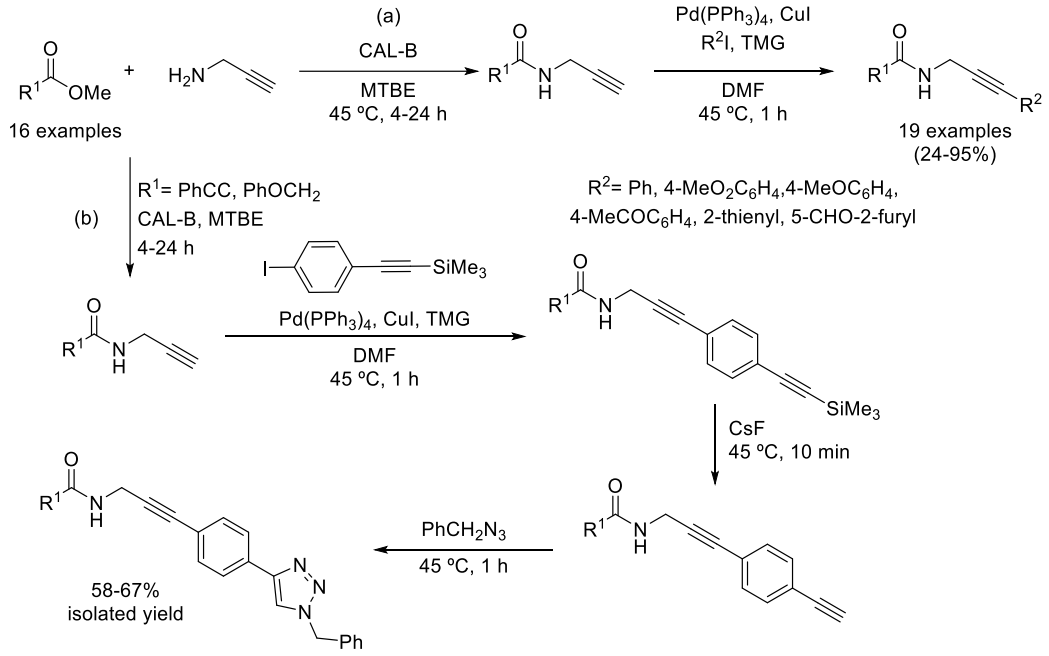


Scheme 66. One-pot consecutive seven-component reaction including Ugi-4 component reaction, lipase-catalyzed aminolysis, CuAAC, and Suzuki coupling using tetrakis(triphenylphosphine)palladium(0).

2.6.3. Other Cross-Coupling Reactions

In addition to the well-known Heck and Suzuki cross-coupling reactions, other similar metal-catalyzed transformations have resulted to be compatible with selected enzymatic activities in cascade processes. The Sonogashira coupling is one of these pivotal examples, allowing carbon-carbon bond formation reactions between a terminal alkyne and an aryl or vinyl halide employing a palladium catalyst and a co-catalyst, usually a copper species, to accelerate the reaction rate.[141,177,178] Following with the combination of lipases and cross-coupling reactions already seen in the previous section, Müller and co-workers reported a one-pot consecutive three-component synthesis of (hetero)arylated propargyl amides consisting of a CAL-B-catalyzed aminolysis of methyl carboxylates followed by Sonogashira coupling with (hetero)aryliodides (Scheme 67).[179] After optimization of the Sonogashira conditions in terms of catalyst, base and solvent, $Pd(PPh_3)_4$ was found as the superior Pd(0) catalyst precursor, and adequately combined

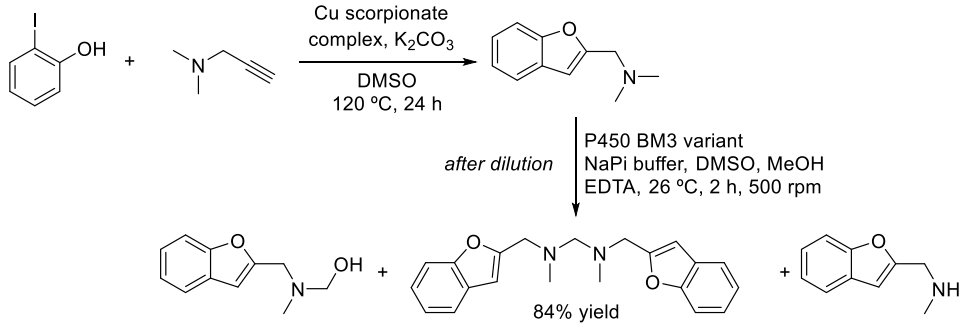
with 1,1,3,3-tetramethyl guanidine (TMG) and DMF. Then, the sequence was performed by conducting first the aminolysis of the starting ester (1.2 equiv) with propargyl amine (500 mM, 1 equiv) in MTBE at 45 ºC, and after 4-24 h, the $Pd(PPh_3)_4$ (2 mol%), CuI (4 mol%), the corresponding aryl iodide (R^2I, 1 equiv), TMG (1 equiv), and DMF were added to provide after 1 h at the same temperature the propargyl amides (Scheme 67a). Interestingly, the sequence can be further extended when selecting *p*-iodo[(trimethylsilyl)ethynyl]benzene as aryl iodide to concatenate additional chemical desilylation and CuAAC, furnishing two 1,4-disubstituted 1,2,3-triazole derivatives containing an arylated propargyl amide (58-67%, Scheme 67b).



Scheme 67. One-pot consecutive three-component synthesis of (hetero)arylated propargyl amides through aminolysis and Sonogashira coupling sequence (strategy a), and possibility to further concatenate a desilylation and CuAAC reactions (strategy b).

Continuing with Sonogashira-biotransformation cascades, a sequential one-pot two-step catalytic reaction has been reported for the synthesis of a methylene-bridged bis(2-substituted benzofuran).[180] The concurrent cascade approach was precluded due to the high temperatures required for the Sonogashira transformation (120 ºC) as previously noticed in other described cross-coupling reactions. Hence, the reaction between 2-iodophenol (1 equiv) and 3-dimethylamino-1-propyne (1 equiv) was performed under

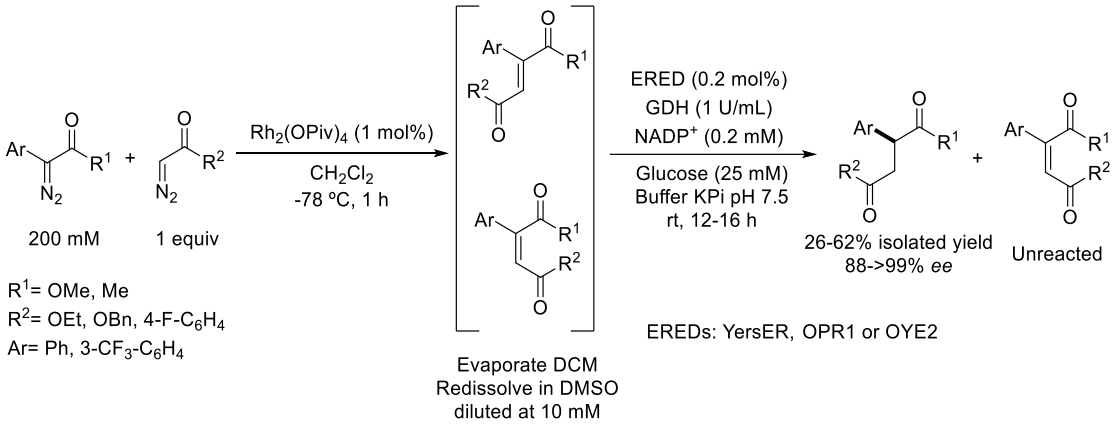
palladium-free conditions using a copper scorpionate complex (10 mol%) in the presence of K_2CO_3 (2 equiv). After 24 h, the hydroxylation was developed of the benzofuran intermediate (after dilution, 0.5 mM) using a monooxygenase P450 BM3 variant (A74G-F87V-L188Q) with the occurrence of concomitant elimination reactions providing a dimer as the main product (Scheme 68). Remarkably, the addition of EDTA as chelating agent allowed the capture/complex of metal ions, increasing the yield up to 84%, which gave a similar value than the sequential reaction including benzofuran intermediate isolation after the first step (88%). The immobilization of the enzyme as microgel was also studied, which presents the benefit of a possible reuse of the enzyme, although the attained yields were significantly lower.



Scheme 68. One-pot two-step Cu-mediated Sonogashira coupling and P450-catalyzed biohydroxylation performed in a sequential manner.

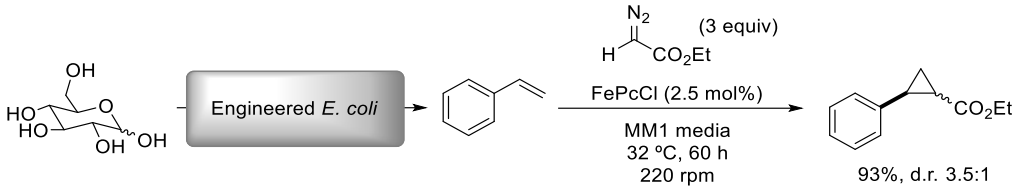
Diazo compounds are the most important and versatile precursors of organic carbenes, so its chemistry has been extensively studied,[181] while the application of carbene transferases in C–C bond formation reactions results highly attractive due to the advances in enzyme evolution.[182,183] Metal-enzyme combinations to take advantage of the diazo-compound reactivity has been explored by Hartwig, Zhao and co-workers for the sequential rhodium-catalyzed C–C bond formation and asymmetric C=C reduction using an ERED.[184] Dirhodium(II) tetrapivalate [$Rh_2(Opiv)_4$] displayed an excellent selectivity towards the formation of (*E*)-alkenes (>9:1) in the diazocoupling reaction between two α-diazocarbonyl compounds, which allowed the development of the chemoenzymatic cascade because these isomers were accepted by the tested EREDs, while the (*Z*)-alkenes obtained as minor products remained unaltered. To carry out the sequential process, firstly, the diazo compounds (200 mM) were dissolved in dichloromethane, and

$Rh_2(Opiv)_4$ (1 mol%) was added. After 1 h at –78 ºC, the solvent was evaporated and the crude redissolved in DMSO, adding next a phosphate buffer pH 7.5 to have a final substrate concentration of 10 mM, and the corresponding ERED (0.2 mol%, YersER from *Yersinia bercovieri*, OPR1 from *Lycopersicum esculentum* or OYE2 from *Saccharomyces cerevisiae*). Glucose (25 mM) and GDH were also added to recycle the nicotinamide cofactor needed by the ERED. Consequently, 2-aryl-substituted succinate derivatives were obtained with low to moderate isolated yield (26-62%) and high enantiomeric excess (88->99%) after additional 12-16 h at room temperature (Scheme 69).



Scheme 69. Sequential Rh-ERED approach to synthesize enantioenriched 2-aryl-substituted succinate derivatives.

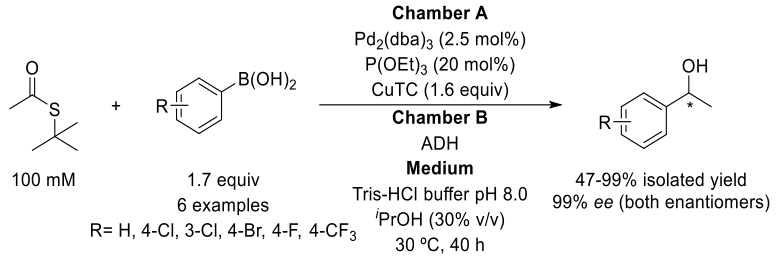
Wallace and Balskus have reported the transformation of D-glucose into styrene using a living system such as *E. coli* followed by the action of a biocompatible iron(III) phthalocyanine catalyst (FePcCl) able to catalyze olefin cyclopropanation reactions (Scheme 70).[185] In this context, after individual reaction optimization, medium and iron catalyst screening, the cyclopropanation of styrene was performed with ethyl diazoacetate (3 equiv, added portionwise to avoid its dimerization) using FePcCl (2.5 mol%), producing ethyl 2-phenylcyclopropane-1-carboxylate (*cis*:*trans* 3.5:1 ratio) in 93% isolated yield after 60 h at 32 ºC.



Scheme 70. Conversion of D-glucose into styrene by an engineered *E. coli* and iron-catalyzed cyclopropanation reaction with ethyl diazoacetate.

The Liebeskind-Srogl cross-coupling reaction is much less known than those previously discussed, typically occurring between a thioester and a boronic acid to generate ketones in the presence of a palladium(0) catalyst, a phosphine as an external ligand, and a stoichiometric additive, which is usually a Cu(I) species.[186] The proposed mechanism consists of the coordination of the Cu(I) species, as Lewis acid, to the electron-rich sulfur atom, then the oxidative insertion of palladium(0) catalyst into the C-S bond is produced, next transmetalation between the palladium species and the boronic acid occurs and, finally, a reductive elimination releases the formed ketone regenerating the Pd(0) catalyst. Liebeskind-Srogl cross-coupling reaction differs from the Suzuki counterpart because it takes place at a neutral pH in the absence of a base. However, as a disadvantage, it requires an additive in stoichiometric amount. This reaction together with an asymmetric bioreduction were described in a compartmentalized cascade methodology to obtain enantiopure alcohols,[187] employing a PDMS membrane since the presence of boronic acids, Cu(I) salts and high phosphine ligand loadings provoked the inactivation of the tested ADHs. The opposite selectivity of ADH-A and *Lk*ADH allowed the production of both desired alcohol enantiomers. Thus, the thioester (100 mM) was suspended in H_2O inside the "Liebeskind-Srogl chamber" with 1.7 equiv of boronic acid, 1.6 equiv of copper(I) thiophene-2-carboxylate (CuTC) and catalytic amounts of tris(dibenzylideneacetone)-dipalladium(0) [$Pd_2(dba)_3$, 2.5 mol%] and triethyl phosphite [$P(OEt)_3$] (20 mol%) as catalyst precursor and ligand, respectively. On the "enzymatic chamber" the corresponding ADH was added together with Tris-HCl buffer pH 8.0 and ^{i}PrOH (30% v/v) as the cofactor regeneration system. After 40 h at 30 ºC, the desired enantiopure alcohols were obtained with moderate to very good isolated yields after liquid-liquid extraction and column chromatography (47-99%, Scheme 71). This protocol was also tried with transaminases to furnish the corresponding chiral amines, but it failed,

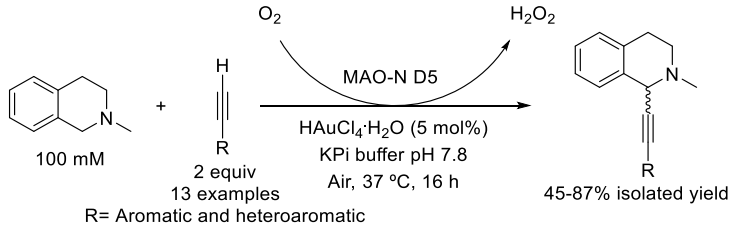
therefore, another sequential approach performing first the metal-catalyzed process and then the transamination in a biphasic system involving heptane as organic solvent and sodium polyacrylate/H_2O for trapping the components of the biocatalytic reaction, afforded enantiopure (*R*)-1-phenylethan-1-amine in 51% isolated yield.



Scheme 71. Compartmentalized concurrent Liebeskind-Srogl cross-coupling and stereoselective bioreduction approach to obtain enantiopure 1-arylethanol enantiomers.

In spite of the multiple applications of gold complexes for C–C bond formation reactions,[188,189] the combination of gold and enzymes is still in its infancy especially if we compare with previously described palladium-biotransformation cascades. One of the most interesting features of gold catalysts is that the oxidation state of the metal often remains unchanged during the catalytic cycle, unlike others such as palladium or nickel catalysts that experiment oxidative-addition processes and change their oxidation state from 0 to +2 during the catalytic cycle.[190] Bergman, Toste and co-workers described in 2013, a tandem ester hydrolysis-hydroalkoxylation using an esterase or a lipase as biocatalyst and Me_3PAuCl as metal catalyst,[191] which depending on the starting acetate, could be developed via kinetic resolution or in a non-selective fashion. More recently, Greaney, Turner and co-workers have developed the chemoenzymatic alkynylation of *N*-methyl-tetrahydroisoquinolines (*N*-Me-THIQs, 100 mM) with acetylenes (2 equiv) in aqueous medium without catalyst compartmentalization requirements.[192] The cross-dehydrogenative coupling reaction consisted of the oxidation of *N*-Me-THIQs using the mutant monoamine oxidase MAO-N D5 to regioselectively generate a cyclic imine, gold(III) chloride trihydrate (5 mol%) catalyzing the cross-coupling reaction between the imine and terminal alkynes through gold-acetylide formation. This work represents the first example of a cascade approach employing Au(III) catalyst and an enzyme to produce a C–C coupling, isolating the racemic 1-substituted-*N*-Me-THIQs with moderate to high isolated yields (25-87%, Scheme 72) after 16 h at 37 ºC. The scope of the reaction resulted

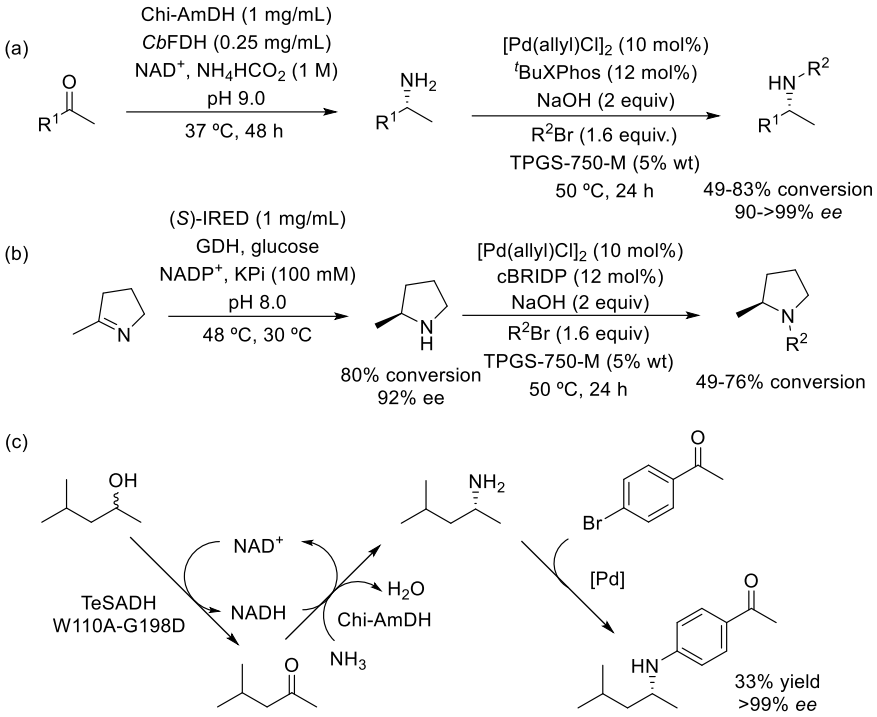
wide in terms of the used alkyne, although more limited in terms of the amine accepting *N*-ethyl-THIQ (88% with phenylacetylene), although the reaction did not proceed in any extension with *N*-acetyl, *N*-benzyl or deprotected THIQs. On the other hand, following a sequential approach (first the enzymatic step and then the gold(III)-catalyzed coupling), aliphatic alkynes and *N*-allyl- and *N*-homoallyl-THIQs could also be applied as substrates (41-46%).



Scheme 72. Chemoenzymatic cascade to achieve the alkynylation of *N*-methyl tetrahydroisoquinolines.

Parmeggiani, Turner and co-workers have described the sequential synthesis of *N*-arylamines by combining different bioamination processes and a Buchwald-Hartwig *N*-arylation reaction (Scheme 73).[193] Remarkably, TPGS-750-M was employed as surfactant to improve the solubility of the palladium and phosphine species in the aqueous medium, thus increasing the reaction rate of different chemoenzymatic approaches. Firstly, an asymmetric reductive amination catalyzed by a chimeric amine dehydrogenase (ChiAmDH) was combined with a palladium-catalyzed Buchwald-Hartwig reaction in a sequential manner. Better results were found with the purified AmDH instead of the cell-free extract form avoiding a negative impact on the metallic step, so the starting ketone (50 mM) was incubated in an ammonium formate buffer 1 M pH 9.0 containing NAD^+, formate dehydrogenase from *Candida boidini* (*Cb*FDH, 0.25 mg/mL) and the purified ChiAmDH (1 mg/mL). After 48 h at 37 ºC, the reaction mixture was added to a degassed aqueous solution of TPGS-750-M (5% w/v), NaOH (2 equiv), ^{t}BuXPhos (12 mol%), $[Pd(allyl)Cl]_2$ (10 mol%) and the corresponding aryl bromide (1.6 equiv), to furnish the (*R*)-N-substituted amines with moderate to good conversions (49-83%) and high enantiomeric excess values (>90%) after additional 24 h at 50 ºC (Scheme 73a). An alternative ATA-catalyzed ketone biotransamination followed by Buchwald-Hartwig arylation sequence was also attempted, however, the presence of a strong nucleophilic

amine as donor (D-alanine) was deleterious to the overall process. To overcome the metallic catalyst inactivation, the reaction crude was extracted with toluene after the biotransamination step, developing then a stepwise approach instead of a chemoenzymatic cascade. The use of an imine reductase (IRED) was also studied, looking for the synthesis of enantioenriched *N*-protected cyclic amines from the corresponding imines. For instance, 5-methyl-3,4-dihydro-2*H*-pyrrole was reacted with a (*S*)-IRED from *Streptomyces* sp. in phosphate buffer pH 8.0 in the presence of DMSO (5% v/v) as cosolvent, glucose and GDH to recycle the NADP cofactor for 48 h at 30 ºC. This mixture was added to a degassed aqueous solution of TPGS-750-M (5% w/v), NaOH (2 equiv), di-*tert*-butyl(2,2,-diphenyl-1-methyl-1-cyclopropyl)phosphine (cBRIDP, 12 mol%), $[Pd(allyl)Cl]_2$ (10 mol%), bubbling nitrogen for 10 minutes before the addition of the corresponding aryl bromide (1.6 equiv), obtaining a series of pyrrolidines after additional 24 h at 50 ºC with moderate to good conversions (49-76%, Scheme 73b). Finally, the same methodology was compatible with a sequential biocatalytic hydrogen-borrowing amination of a *sec*-alcohol, starting from a racemate that was oxidized by the action of a non-enantioselective variant of an ADH from *Thermoanaerobacter ethanolicus*, coupled with ChiAmDH to produce the enantioenriched amine, which finally underwent the Buchwald-Hartwing arylation reaction, yielding enantiopure (*R*)-1-[4-((4-methylpentan-2-yl)amino)phenyl]ethan-1-one in 33% yield (Scheme 73c).

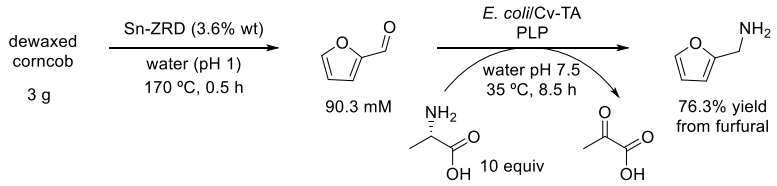


Scheme 73. Different sequential syntheses of chiral *N*-arylamines involving various biocatalytic processes and Buchwald-Hartwig arylations.

Recently, Heckmann and Paradisi have combined the Pd-mediated Buchwald-Hartwig amination of (chiral) amines obtained by the ATA-catalyzed biotransamination of a series of aldehyde and ketone precursors.[194] Firstly, the (*R*)-selective transaminase from *Thermomyces stellatus* (*Ts*RTA) was used in combination with D-alanine (5 equiv), glucose (1.2 equiv), and GDH in a buffer-DMSO (9:1 v/v) system, requiring enzyme centrifugation and protein elimination, before the addition of the amine intermediate solution to the vessel containing the metal catalyst for the second step. The final addition of $[Pd(allyl)Cl]_2$ (5 mol%), sodium *tert*-butoxide (7 equiv), a GPhos ligand (6 mol%), and an aryl halide (1.2 equiv) in a biphasic buffer-toluene mixture made possible this two-step transformation, which afforded the (enantioenriched) amines in low to high conversions.

2.7. Miscellaneous

Furfurylamine is a valuable biomass-product that is widely used to produce food additives, polymers and pharmaceuticals, among others. He and co-workers recently described the treatment of dewaxed corncob with acidified solid acid based on Sn-zirconium dioxide (Sn-ZRD) to furnish furfural by hydrolysis and dehydration, which subsequently was enzymatically aminated using a transaminase in a one-pot sequential approach, giving access to furfurylamine (Scheme 74).[195] Thus, corncob obtained after alkali pretreatment (3 g) was reacted with water in the presence of Sn-ZRD (3.6% wt) at 170 ºC during 30 min, synthesizing furfural (90.3 mM), and after that time pH was adjusted at 7.5, and *E. coli* cells overexpressing Cv-TA and L-alanine (10 equiv) as amine donor were added. The reaction was maintained at 35 ºC for 8.5 h, isolating the desired amine in 76.3% yield (from quantified furfural). Later, *E. coli* whole cells were immobilized on carrageenan, and the beads formed were cross-linked with glutaraldehyde. This enzymatic preparation was studied with the Sn-ZRD particles to study the recyclability of the system, being active after 6 reaction cycles. As a further extension of this methodology, sulfonated Sn on perlite and *E. coli*/Cv-TA cells were utilized to obtain furfurylamine from bamboo shoot shell, corncob and rice straw in a one-pot sequential manner. In this case, the metal-catalyzed step was performed in a γ-valerolactone/water (1:4 v/v) mixture, and the transamination was achieved using isopropylamine (3 equiv) as amine donor, forming furfurylamine at high yield (approx. 0.4 g/g xylan in biomass).[196] The same group has also developed a similar one-pot system to prepare furfuryl alcohol from a xylose-rich hydrolysate using sequentially a Sn-based solid acid as catalyst and an alcohol dehydrogenase from *Sporidiobolus salmonicolor*.[197]



Scheme 74. Synthesis of furfurylamine from corncob combining a metal-dehydration step with an enzymatic transamination.

3. Artificial metal-enzyme combinations for cascade reactions

In the previous section, the role of (organo)metallic complexes and enzymes for the development of cascade reactions has been described, focusing on the individual characteristics of both types of catalysts. Thus, their combined use has allowed the design of efficient one-pot multicatalytic transformations, searching for compatible conditions (concurrent processes) or changing some reaction parameters at an intermediate stage, for example pH, temperature, reaction medium, substrate concentration, or additional reagents/catalysts, among others (sequential approaches). Herein, the design of artificial metal-enzyme combinations will be presented as an emerging source of active and selective catalysts,[198,199] which for the development of cascade processes mainly can be divided in two types: metalloenzymes and bionanohybrids (BNHs).

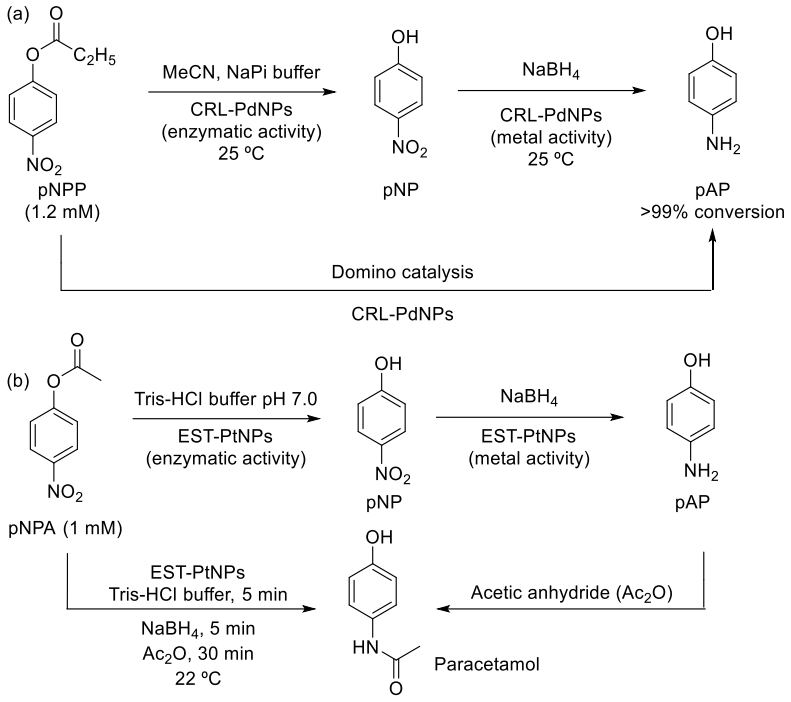
Artificial metalloenzymes can be obtained either by the incorporation of a non-native organometallic complex in a protein matrix or replacing the native transition metal present in natural metalloproteins, combinations that have allowed the appearance of an exciting research field with outstanding contributions excellently reviewed in various reviews in recent years.[200-203] The reconstruction of proteins brings new interesting catalytic properties, even *in vivo*,[204] which in some cases has allowed the design of cascade transformations,[7,205] ranging from the design of two consecutive reactions[206] to more complicated systems.[139] Based on the extensive number of reviews in the field, we have next exclusively focused on the possibilities of BNHs for the development of cascade reactions.

Metal nanoparticles (NPs) have attracted increasing attention in the last decade due to their unique physicochemical properties compared to their traditional bulk counterparts. Many different applications have been disclosed in blockbuster scientific areas such as catalysis[207] and biomedicine,[208,209] NP size and shape being crucial parameters for their latest use. Interestingly, enzymes provide a three-dimensional structure to coordinate metal atoms such as gold, palladium, platinum, silver, etc., which represents an elegant form to catalyze single and multiple reactions. Based on its metallic and enzymatic composition, the main advantage of BNHs resides in their double reactivity, therefore its application in concurrent processes has received exponential attention in recent years.[210] Therefore, examples related to the use of BNHs for single catalytic transformations

(acylations, C–C couplings, oxidations…)[211-214] are omitted since they are out of the scope of this review focus on multicatalytic cascades. Next, a division of BNH-catalyzed multistep transformations has been made depending on their development in a non-selective or stereoselective fashion, paying special attention in the design of overall one-pot cascade transformations rather than describing the catalyst preparation and characterization.[215]

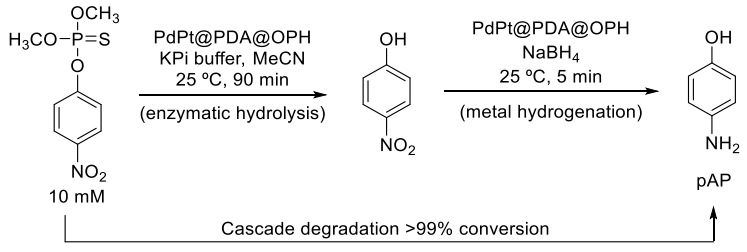
3.1. Bionanohybrids in non-stereoselective cascade processes

Palomo and co-workers have developed novel BNHs composed by the lipase from *Candida rugosa* (CRL) and palladium nanoparticles (CRL-PdNPs) to catalyze the single oxidation of aromatic alcohols such as 4-methoxybenzyl alcohol, 4-nitrobenzyl alcohol and 1-phenylethanol as racemates or single enantiomers, but also the domino transformation of 4-nitrophenyl propionate (pNPP) to 4-aminophenol (pAP).[216] This two-step cascade consisted of the lipase-catalyzed hydrolysis of pNPP (1.2 mM) to 4-nitrophenol (pNP) that smoothly occurred in a phosphate buffer after 50 minutes at 25 ºC (Scheme 75a), followed by the reductive action of the palladium component after addition of sodium borohydride ($NaBH_4$) at the same temperature. Similarly, the combination of an oligomeric esterase from *Mycobacterium smegmatis* and platinum nanoparticles (EST-PtNPs) has been described to produce pAP in this case from 4-nitrophenyl acetate (pNPA, 1 mM).[217] The advantage of this BNH was demonstrated in the multistep synthesis of the analgesic and antipyretic drug acetaminophen (paracetamol) by just adding acetic anhydride to the reaction medium after pAP formation (Scheme 75b). All these transformations were performed under very mild conditions (22 ºC) and very short reaction times.



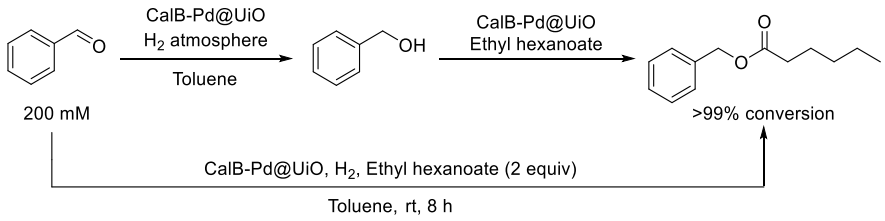
Scheme 75. Conversion of 4-nitrophenyl propionate (pNPP) and 4-nitrophenyl acetate (pNPA) to 4-aminophenol (pAP) using hydrolase-metal NPs.

Gao, Jiang and co-workers reported the synthesis of a mesoporous core-shell nanostructure PdPt@PDA with a bimetallic core using K_2PtCl_4, H_2PtCl_6, Na_2PdCl_4, a surfactant (Pluronic F127) and polydopamine (PDA), immobilizing later an organophosphorus hydrolase (OPH) on the outer surface of the PDA shell though a bioadhesion-inspired strategy.[218] The so-obtained BNH (PdPt@PDA@OPH) catalyzed the enzymatic hydrolysis of the organophosphate nerve agent, parathion-methyl (10 mM), and subsequently the reduction of the nitro group occurred to obtain the desired pAP (Scheme 76).



Scheme 76. One-pot degradation of parathion-methyl into pAP using PdPt@PDA@OPH.

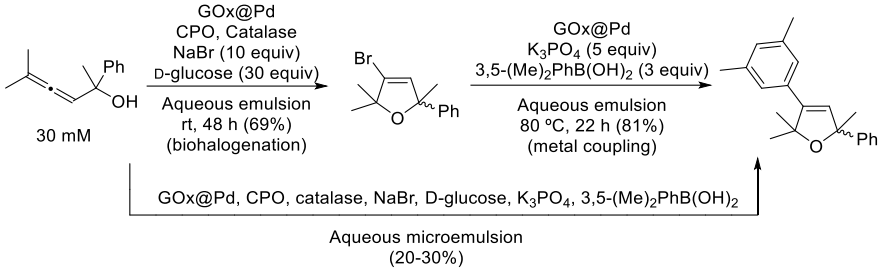
The combination of Pd NPs and CAL-B in a metal organic framework (UiO-66-NH_2) has been applied to the expedient synthesis of benzyl hexanoate from benzaldehyde using an organic cosolvent such as toluene.[219] The best results were obtained when the metal scaffold was treated with lauric acid. This one-pot concurrent cascade consisted of the hydrogenation of the aldehyde (200 mM) to produce benzyl alcohol, that in the same reaction medium was esterified using 2 equiv of ethyl hexanoate (Scheme 77). The same reaction has been described using BNHs obtained after co-immobilization of Pd NPs and CAL-B into a functionalized mesoporous silica material. Thus, their mutual inactivation was avoided, running the reaction with benzaldehyde at 250 mM concentration and benzyl hexanoate (2 equiv) at 25 ºC in toluene.[220] The same research group has demonstrated the ability of carbon nitride (*g*-C_3N_4) as an excellent platform to co-immobilize Pd nanoparticles and CAL-B using glutaraldehyde as crosslinker.[221] This BNH was able to transform benzaldehyde (400 mM) into benzyl hexanoate (82%) after hydrogenation and transesterification reactions at rt after 12 h, although the reusability of the co-catalyst needed to be improved for synthetic purposes due to a serious loss of the enzyme activity after four cycles.



Scheme 77. Hydrogenation-esterification concurrent cascade in organic solvent using a BNH composed by Pd NPs and CAL-B.

Recently, Deska and co-workers have disclosed a combination of enzymatic halocyclization and subsequent Suzuki-type cross coupling.[222] A glucose oxidase-chloroperoxidase-catalase system in the presence of D-glucose and sodium bromide was reported to be efficient for enzymatic allene cyclizations at rt, using later Na_2PdCl_4 to catalyze the cross-coupling step in the presence of potassium phosphate and a boronic acid at 80 ºC in a heptane:buffer mixture (1:1 v/v). After demonstrating the versatility of this one-pot sequential approach with different allenols and boronic acids, a direct arylative allenol cyclization of 5-methyl-2-phenylhexa-3,4-dien-2-ol with 3,5-

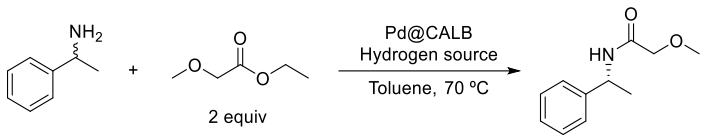
dimethylphenylboronic acid was envisaged using a GOx@Pd BNH in an aqueous emulsion (Scheme 78). Although the use of the BNH in a cascade manner only afforded 20-30% yield, lower than when the hybrid catalyzed the individual steps (69% and 81% for the first and second steps, respectively), it represents an exciting finding for future improvements.



Scheme 78. Enzymatic halogenation and subsequent Suzuki-type cross coupling in an aqueous emulsion using a GOx@Pd BNH.

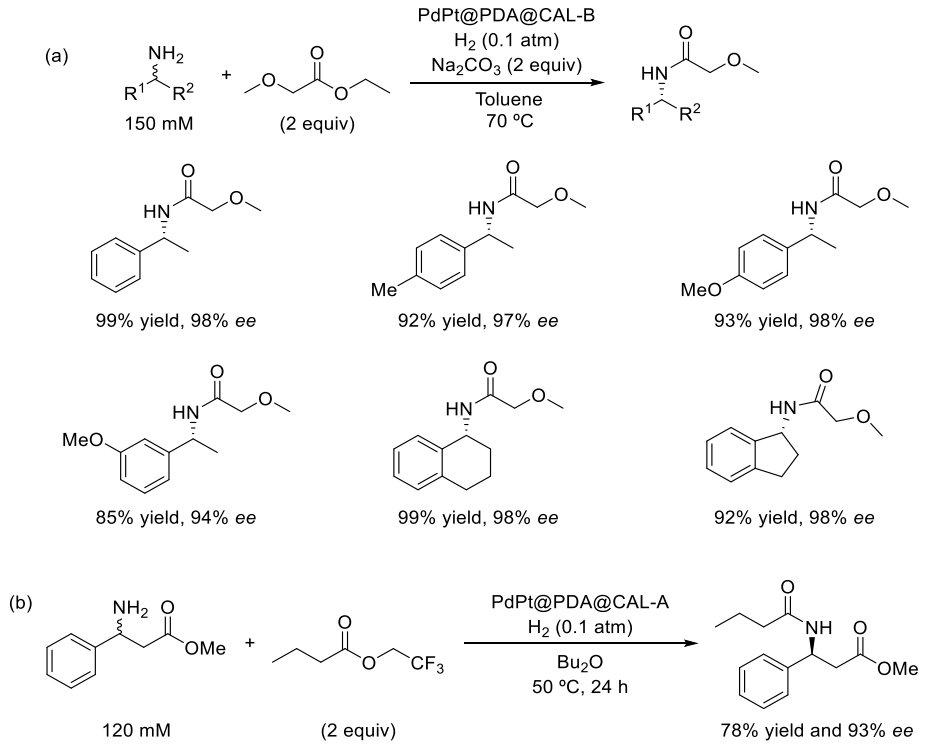
3.2. Bionanohybrids in stereoselective cascade processes

Since the seminal work from Bäckvall and co-workers dealing with the DKR of 1-phenylethylamine using Pd NPs and CAL-B co-immobilized in siliceous mesocellular foams,[223] many different research groups have performed the DKR of racemic amines using BNHs in organic solvents (Scheme 79). For instance, other Pd@CAL-B combinations have been efficiently employed for the DKR of the same amine, using in these cases toluene and 2 equiv of ethyl methoxyacetate as acyl donor at 70 ºC under hydrogen atmosphere or employing formate as hydrogen donor.[224,225] The possibility to recycle and reuse the biocatalyst was studied in successive DKRs of 1-phenylethylamine, finding a notable decrease of the activity after five reaction cycles (55-84%), while the selectivity was mostly maintained (98->99% *ee*).[224] Itabaiana Jr and co-workers applied this combination not only in batch for around 12 h, but also in continuous mode allowing the performance of faster processes (up to 9 h), although with significant by-product formation in both cases.[226]



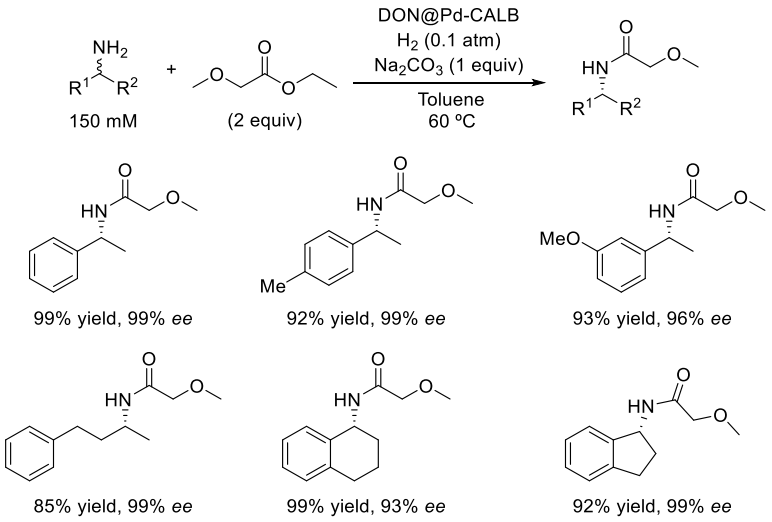
Scheme 79. DKR of 1-phenylethan-1-amine using Pd@CAL-B BNHs with ethyl methoxyacetate in toluene.

The possibilities of these types of hybrid catalysts have been fully demonstrated in recent years, by disclosing highly stereoselective DKRs of a wide set of racemic amines apart from 1-phenylethylamine, which has always been used as the model substrate for reaction conditions optimization. These reports have employed different co-immobilized palladium species and lipases as BNHs, broadening the possibilities for chiral amines syntheses.[218,227-231] A mesoporous core-shell structured nanocatalyst with a PdPt bimetallic core and CAL-B co-immobilized on polydopamine (PdPt@PDAt@CAL-B) has been applied for the DKR of six primary amines using ethyl methoxyacetate (2 equiv) as acyl donor (Scheme 80a).[218] The recycling of the catalyst was possible after a centrifugation process, maintaining the stereoselectivity after five cycles (98% *ee*, for 1-phenylethylamine), while the activity just moderately decreased, as 75% yield of the product was still attained. To expand the possibilities of these types of materials, a new BNH, in this case with CAL-A instead of CAL-B was synthesized and applied in the DKR of a β-amino ester (methyl 3-amino-3-phenylpropanoate), obtaining after reaction the corresponding (*S*)-amide in 78% yield and 93% *ee* (Scheme 80b).



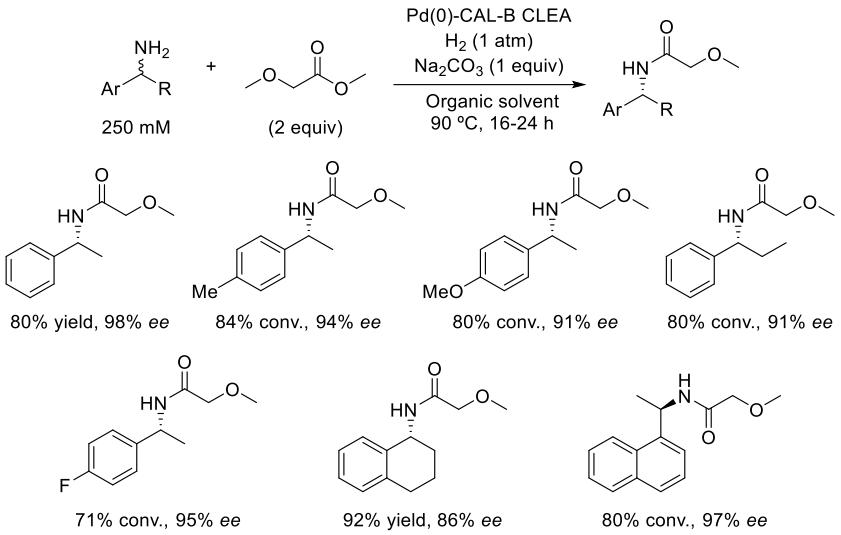
Scheme 80. DKR of racemic amines and methyl 3-aminopropanoate using a Pd-CAL-B (a) or Pd-CAL-A BNH (b), respectively.

In a similar manner, other porous materials have been recently applied by the same research group for identical purposes and similar reaction conditions. For instance, the co-immobilization of Pd NPs and CAL-B as dendritic organosilica nanoparticles (DONs) has been satisfactorily demonstrated in the DKR of a series of primary amines in high to excellent yields (85-99%) and selectivities (93-99% *ee*) using ethyl methoxyacetate as acyl donor and toluene as solvent at 60 ºC (Scheme 81).[229] More recently, the use of porous imine molecule cages has served to entrap palladium nanoparticles and CAL-B,[230] which were successfully applied in the DKR of similar primary amines to those described in the previous two examples (toluene, 60 ºC, 4-8 h).[218,229]



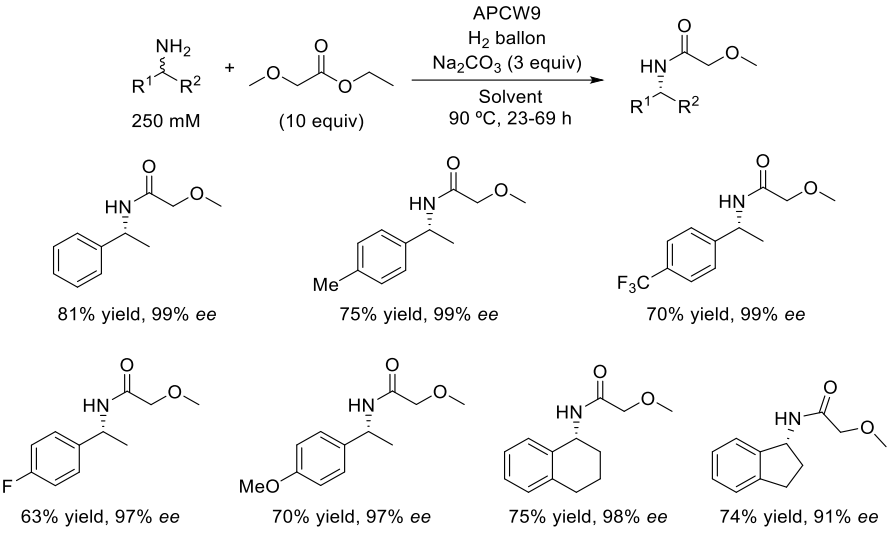
Scheme 81. DKR of racemic amines using palladium and CAL-B co-immobilized into dendritic organosilica particles.

Protein-polymer nanoconjugates have been utilized as confined nanoreactors for the BNH generation, making possible the action of both lipase and metal catalysts at an ideal temperature of 55 ºC.[227] In this case ethyl acetate (3 equiv) was used as acyl donor and toluene as solvent under an argon atmosphere (0.4 MPa) for the successful DKR of 1-phenylethylamine, 1-aminoindane and 1,2,3,4-tetrahydro-1-naphthylamine at 100 mM substrate concentration. Bäckvall and co-workers showed the efficient resolution of a series of benzylic amines (Scheme 82), by using a BNH catalyst consisting of palladium nanoparticles immobilized on CLEAs from CAL-B, that apart of the achievement of good yields (80-92%) and selectivities (86-98% *ee*), the hybrid immobilized system was easily recovered and reused, the activity remaining almost unaltered after 5 cycles when toluene was used as solvent in the DKR of 1-phenylethylamine (72-78% amide formation). Compared to the previous examples, the temperature needed to be raised up to 90 ºC, however it is also remarkable that under these conditions the lipase remained stable, demonstrating the potential of this material.[228]



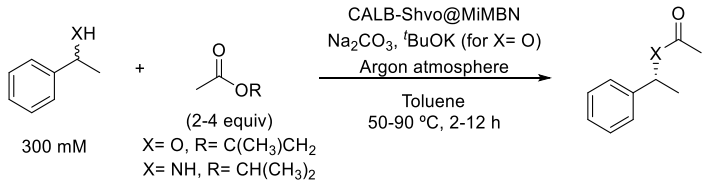
Scheme 82. DKR of racemic amines using a Pd(0)-CAL-B CLEA in 1,4-dioxane or toluene.

Recently, Bäckvall, Córdova and co-workers have developed the DKR of different primary amines assembling cellulose-based artificial plant cell wall (APCW) containing palladium nanoparticles and CAL-B as (bio)catalysts.[231] Hence, modified cellulose fibers were used as support of metal particles first, and then, in the presence of a surfactant, the lipase was co-immobilized. A series of racemic substrates (250 mM) were reacted in the presence of this material (APCW9) containing 1.1 mol% of Pd, sodium carbonate (3 equiv) as base and ethyl methoxyacetate (10 equiv) as acyl donor. Reactions were carried out in organic solvents such as toluene, 1,4-dioxane or 3-methylpentan-3-ol under hydrogen atmosphere (1 atm) at 90 ºC, furnishing the corresponding (*R*)-amides in high isolated yields (63-81%) and *ee* values (>91%) after 23-69 h (Scheme 83).



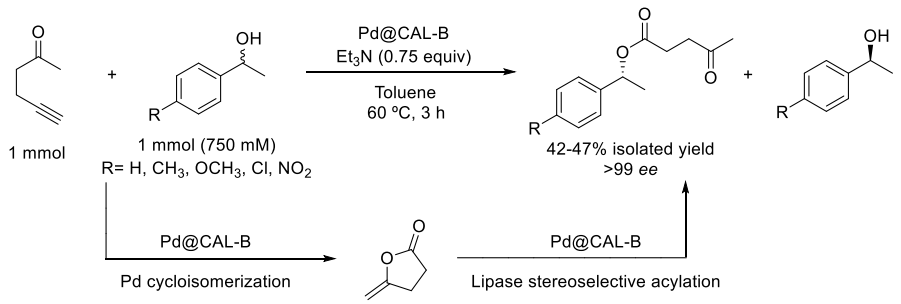
Scheme 83. DKR of racemic amines using assembling cellulose-based artificial plant cell wall containing palladium nanoparticles and CAL-B.

The kinetic resolution of racemic secondary alcohols using BNHs has been less studied than in the case of the amine racemates, however, Chen and co-workers described the co-encapsulation of CAL-B and the Shvo catalyst into a 2-methylimidazole-metal-biosurfactant nanocomposite (CALB-Shvo@MiMBN),[232] which was later applied in the DKR of 1-phenylethanol and 1-phenylethylamine (Scheme 84). Higher reaction temperatures were required (80-90 ºC) and isopropyl acetate (4 equiv) for the resolution of the amine derivative in comparison with the process for the secondary alcohol (50-70 ºC) and isopropenyl acetate (2 equiv) as acyl donor. The reaction times were shortened in comparison with the use of the free Shvo catalyst together with commercial Novozym 435 under the same reaction conditions.



Scheme 84. DKR of 1-phenylethanol and 1-phenylethan-1-amine using a BNH composed by Shvo and CAL-B catalysts.

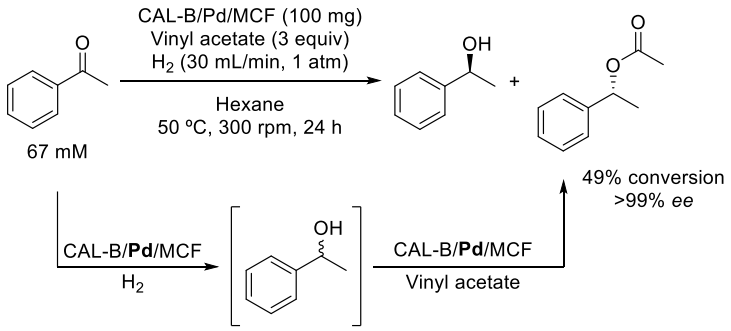
There is an interesting work reported by Johnston, Bäckvall and co-workers showing the design and application of a Pd@CAL-B BNH using lipase cross-linked enzyme aggregates to obtain optically active O-levulynated 1-arylethanols through a metal-catalyzed lactonization of the acyl donor and lipase-mediated kinetic resolution sequence.[233] Hence, the metal component catalyzed the cycloisomerization of 4-pentyonic acid into 5-methylene-dihydrofuran-2(3*H*)-one, lactone that served as acyl donor in the CAL-B-catalyzed kinetic resolution of a series of 1-arylethanols to obtain the corresponding (*R*)-esters and remaining (*S*)-alcohols with good selectivity after 3 h at 60 ºC in toluene (Scheme 85). The reaction was also successfully extended to a bulkier substrate such as 1-(naphthalen-2-yl)ethanol (45% yield and 99% *ee*), and the recycling of the BNH was demonstrated in the reaction with racemic 1-phenylethanol, observing after 5 uses a minimum decrease in the activity (42-45%) and stereoselectivity (98-99% *ee*) for the formation of the (*R*)-ester. Recycling studies demonstrated that after 6 cycles, the BNHs was still active (42-46% yield, 98-99% *ee*), SEM images demonstrated that the morphology of the recycled catalyst was identical to that of the fresh catalyst Pd(0)-CAL-B CLEA catalyst.



Scheme 85. Cycloisomerization and kinetic resolution cascade to obtain enantioenriched secondary alcohols using a Pd@CAL-B CLEA preparation.

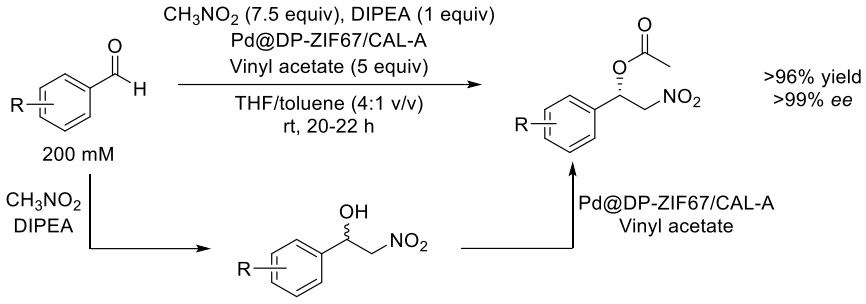
Magadum and Yadav reported the chemoenzymatic synthesis of (*R*)-1-phenylethyl acetate through a tandem process involving the hydrogenation of acetophenone (67 mM) and subsequent stereoselective acylation of the formed racemic 1-phenylethanol (Scheme 86).[234] This strategy was based on the co-immobilization of palladium and CAL-B on a mesoporous foam, affording enantiopure (*R*)-1-phenylethyl acetate with 49% conversion

when using vinyl acetate as acyl donor. In this case, it must be noted that the catalytic hydrogenation was possible at atmospheric hydrogen pressure, obtaining a complete conversion into the racemic alcohol after 10 h, while after optimization of the enzymatic transformation, 3 equiv of vinyl acetate, *n*-hexane as solvent, 50 ºC and 300 rpm resulted the best tested conditions for the sequential combination of metal and enzyme catalysis under flow conditions.



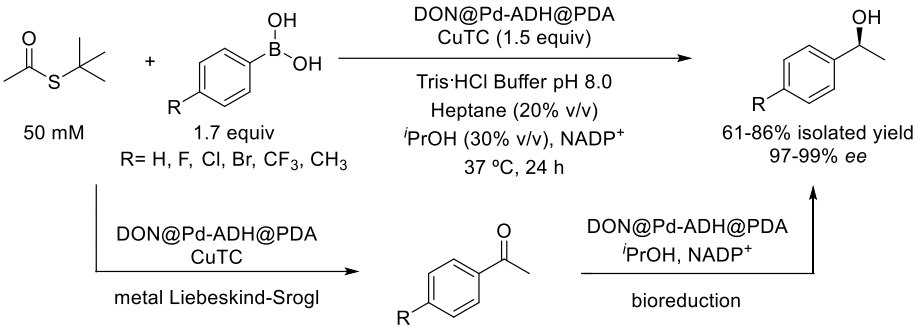
Scheme 86. Acetophenone hydrogenation followed by stereoselective acetylation using a co-immobilized Pd-CAL-B BNH on a mesoporous foam in a flow system.

In another study involving secondary alcohols, a DKR approach was recently demonstrated by combining in this case palladium and CAL-A in a Co-based material (Scheme 87).[235] After design and synthesis of multimodal catalytic nanoreactors (MCNRs) based on a mesoporous metal-organic framework, which allowed a synergistic action of metal and enzyme, producing β-nitroalcohol derivatives in excellent yields and optical purities. The reaction consisted of the one-pot concurrent reaction between a series of substituted benzaldehydes (0.2 mmol) and nitromethane (1.5 mmol) in the presence of a base (DIPEA), forming the racemic alcohols, which afterwards were resolved with CAL-A encapsulated in Pd@DP-ZIF67, that was also the responsible of the DKR of these intermediates with vinyl acetate (1 mmol) in a mixture of THF:toluene (4:1 v/v) at room temperature after 20-22 h.



Scheme 87. One-pot nitro aldol formation-DKR cascade process using a co-immobilized palladium-CAL-A catalyst at room temperature.

Finally, hydrophobic nanopores, fabricated through co-immobilization of palladium nanoparticles and the ADH from *Rhodococcus ruber* into dendritic organosilica nanoparticles (DON@Pd-ADH@PDA), have been used in a one-pot cascade consisting of a Liebeskind-Srogl reaction followed by asymmetric carbonyl bioreduction to produce chiral *sec*-alcohols (Scheme 88).[229] Therefore, the drawbacks associated to the enzymatic inhibition caused by the presence of Cu^{+} ions from copper(I) thiophene-2-carboxylate required for the first step was avoided, since the ion bound to the PDA shell, circumventing the use of compartmentalization strategies or biphasic systems as alternative solutions. In this manner, *S*-(*tert*-butyl) ethanethioate was reacted with a series of arylboronic acids in the presence of ^{i}PrOH as hydrogen donor, obtaining six (*S*)-1-arylethanols with excellent selectivity (97-99% *ee*) and good to very high yields (61-86%) after column chromatography purification. The use of *n*-heptane resulted highly beneficious, probably because the overenrichment of organic substrates and products near the catalyst active sites which inhibited the enzyme in the absence of the cosolvent. The reusability of the BNH was studied finding a continuous loss of the catalyst activity, around 20% of the conversion after four uses, but mostly maintaining the stereoselectivity, suggesting palladium leaching.



Scheme 88. C–C coupling and carbonyl bioreduction cascade to obtain enantiopure *sec*-alcohols using Pd-ADH co-immobilized in dendritic organosilica nanoparticles.

Nowadays, BNHs have consolidated their potential in organic synthesis due to their efficient action with a double catalytic activity, which allow them to promote different sequential and concurrent processes both in aqueous and organic media depending on the immobilization strategy. One of their main advantages compared to traditional multicatalyst approaches is the possibility to recycle the hybrid material in a simple way, acting with great and cooperative efficiency in successive catalytic cycles and avoiding possible inadequate interactions between catalysts and reagents. Since the application of BNHs is still in its infancy, many chemo- and stereoselective cascades are envisaged to be proximately disclosed.

4. Challenges, perspectives and conclusions

The combination of metal and enzymes in cascade process has become a very useful and versatile methodology for the synthesis of a broad number of (chiral) molecules, minimizing reaction steps and tedious intermediate isolations, especially advantageous when they are unstable. Herein, the synergy between both types of catalysts has been deeply revised, paying special attention not only to current existing possibilities and perspectives, but also showing the ways to avoid undesired catalyst deactivations, which in many cases led to the design of stepwise strategies due to the need of the removal of the first catalyst in the cascade before developing the second step, intermediate isolation or even purification.[236-241] Probably, this limitation has hampered the implementation of scalable metalloenzymatic cascade in the industrial sector in order to replace current

existing synthetic methods.[10,242] Some of these challenges have been solved through different approaches, for instance, by developing compartmentalization strategies including the use of thimbles, encapsulations, adding surfactants or creating biphasic systems by addition of organic solvents or deep eutectic solvents, or exploiting continuous flow reactions. Interestingly, emerging bionanohybrids currently receive great attention since they allow the cooperative work of the metal and enzyme units.

While at first sight one could expect that these combinations cannot be straightforward designed due to the different solvent media preference for enzymes (water as natural medium) and metals (organic solvents as the preferred environment), in the last decade a tremendous growth of one-pot chemoenzymatic methods has been noticed, and it is expected that these methodologies will be further expanded. In this sense, palladium and ruthenium catalysts have received great attention for the development of sequential protocols and concurrent cascades due to their unparalleled reactivity with respect to the biocatalysts, but apart from these main protagonists, other metal complexes are gaining relevance in the field, especially highlighting the versatility of gold for various synthetic transformations. In fact, many organometallic transformations such rearrangements and C–C or C–X coupling reactions have been historically described in organic solvents. Remarkably, the development of novel complexes has demonstrated that selected transformations can be performed in an aqueous environment under mild conditions, opening-up the possibility to perform novel one-pot (concurrent) processes. In this context, the other side of the coin, the enzyme counterpart, has also a key role, and due to the tremendous progresses in strategies such as biocatalyst immobilization and enzyme evolution, improved and highly selective biocatalysts that can accept non-natural substrates and catalyze non-conventional reactions, can likewise add more structural molecular complexity to target products in a very selective and robust manner.

Until now, lipases and keto-converting enzymes such as ADHs and ATAs have attracted major attention in the field, which is likely related to their accessibility from commercial sources. In addition, the easiness of use of lipases, their ability to work in organic media as immobilized forms and the lack of external cofactor addition has made possible their broad application in the preparation of a wide number of organic molecules such as esters, amides, carbonates, carbamates, etc. However, an increasing number of metalloenzymatic

approaches using, e.g. cytochrome P450 enzymes, amine oxidases, EREDs, IREDs or decarboxylases are expected in the next few years, opening new insights that will provide elegant, robust, and scalable chemoenzymatic approaches towards valuable targets. It is obvious that this will affect to the expertise that young scientists working in Biotechnology, mainly in the Biocatalysis field, would acquire, as they will need to be trained in organometallic chemistry and *vice versa*, since many enzymes are themselves metalloproteins containing a metal ion cofactor, such as cobalt, copper, iron, manganese or zinc, being essential for the enzyme action. However, the combination of both worlds must be employed in a reasonable way, taking advantage of both catalysis but also being aware of existing limitations to advance, and therefore considering solutions in the reaction set-ups.[243-246] There is no more powerful technique than organometallic chemistry for C–C or C–X bond formation, while enzyme can provide unique possibilities for chiral induction, so their cooperative work in one-pot multistep transformations undoubtedly constitutes one of the most straightforward manners to design controlled complexity. Gladly, this is an area in continuous evolution during 2022, and remarkable examples have already appeared in the literature including valuable chemoenzymatic cascades.[247-258]

References

(1) Chiusoli, G. P.; Maitlis, P. M. Metal-Catalysis in Industrial Organic Processes. RSC Publishing, Cambridge (United Kingdom). 2007; pp 1-290.

(2) Johansson Seechurn, C. C. C.; Kitching, M. O.; Colacot, T. J.; Snieckus, V. Palladium-Catalyzed Cross-Coupling: A Historical Contextual Perspective to the 2010 Nobel Prize. *Angew. Chem. Int. Ed.* **2012**, *51*, 5062–5085.

(3) Ogba, O. M.; Warner, N. C.; O'Leary, D. J.; Grubbs, R. H. Recent Advances in Ruthenium-Based Olefin Metathesis. *Chem. Soc. Rev.* **2018**, *47*, 4510–4544.

(4) Fernandes, A. A. G.; Jurberg, I. D. Synergisms between Metal and Photoredox Catalysis: Deconvoluting Complex Systems. *Quim. Nova* **2020**, *43*, 928–950.

(5) Li, X.; Cao, X.; Xiong, J.; Ge, J. Enzyme–Metal Hybrid Catalysts for Chemoenzymatic Reactions. *Small* **2020**, *16*, 1902751.

(6) Ríos-Lombardía, N.; García-Álvarez, J.; González-Sabín, J. One-Pot Combination of Metal- and Bio-Catalysis in Water for the Synthesis of Chiral Molecules. *Catalysts* **2018**, *8*, 75.

(7) Wang, Y.; Zhao, H. Tandem Reactions Combining Biocatalysts and Chemical Catalysts for Asymmetric Synthesis. *Catalysts* **2016**, *6*, 194.

(8) Albarrán-Velo, J.; González-Martínez, D.; Gotor-Fernández, V. Stereoselective Biocatalysis: A Mature Technology for the Asymmetric Synthesis of Pharmaceutical Building Blocks. *Biocatal. Biotransform.* **2018**, *36*, 102–130.

(9) Adams, J. P.; Brown, M. J. B.; Diaz-Rodriguez, A.; Lloyd, R. C.; Roiban, G.-D. Biocatalysis: A Pharma Perspective. *Adv. Synth. Catal.* **2019**, *361*, 2421–2432.

(10) Wu, S.; Snajdrova, R.; Moore, J. C.; Baldenius, K.; Bornscheuer, U. T. Biocatalysis: Enzymatic Synthesis for Industrial Applications. *Angew. Chem. Int. Ed.* **2021**, *60*, 88–119.

(11) Benítez-Mateos, A. I.; Roura Padrosa, D.; Paradisi, F. Multistep Enzyme Cascades as a Route towards Green and Sustainable Pharmaceutical Syntheses. *Nat. Chem.* **2022**, *14*, 489–499.

(12) O'Brien, P. J.; Herschlag, D. Catalytic Promiscuity and the Evolution of New Enzymatic Activities. *Chem. Biol.* **1999**, *6*, R91–R105.

(13) Sheldon, R. A.; Brady, D. Broadening the Scope of Biocatalysis in Sustainable Organic Synthesis. *ChemSusChem* **2019**, *12*, 2859–2881.

(14) Sheldon, R. A.; Brady, D.; Bode, M. L. The Hitchhiker's Guide to Biocatalysis: Recent Advances in the Use of Enzymes in Organic Synthesis. *Chem. Sci.* **2020**, *11*, 2587–2605.

(15) Hauer, B. Embracing Nature's Catalysts: A Viewpoint on the Future of Biocatalysis. *ACS Catal.* **2020**, *10*, 8418-8427.

(16) Sheldon, R. A.; Pereira, P. C. Biocatalysis Engineering: The Big Picture. *Chem. Soc. Rev.* **2017**, *46*, 2678–2691.

(17) Arnold, F. H. Directed Evolution: Bringing New Chemistry to Life. *Angew. Chem. Int. Ed.* **2018**, *57*, 4143–4148.

(18) Salvi, H. M.; Yadav, G. D. Process Intensification Using Immobilized Enzymes for the Development of White Biotechnology. *Catal. Sci. Technol.* **2021**, *11*, 1994–2020.

(19) Bornscheuer, U. T.; Huisman, G. W.; Kazlauskas, R. J.; Lutz, S.; Moore, J. C.; Robins, K. Engineering the Third Wave of Biocatalysis. *Nature* **2012**, *485*, 185–194.

(20) Reetz, M. T. What Are the Limitations of Enzymes in Synthetic Organic Chemistry? *Chem. Rec.* **2016**, *16*, 2449–2459.

(21) Rudroff, F.; Mihovilovic, M. D.; Gröger, H.; Snajdrova, R.; Iding, H.; Bornscheuer, U. T. Opportunities and Challenges for Combining Chemo- and Biocatalysis. *Nat. Catal.* **2018**, *1*, 12–22.

(22) Dumeignil, F.; Guehl, M.; Gimbernat, A.; Capron, M.; Ferreira, N. L.; Froidevaux, R.; Girardon, J.-S.; Wojcieszak, R.; Dhulster, P.; Delcroix, D. From Sequential Chemoenzymatic Synthesis to Integrated Hybrid Catalysis: Taking the Best of Both Worlds to Open up the Scope of Possibilities for a Sustainable Future. *Catal. Sci. Technol.* **2018**, *8*, 5708–5734.

(23) Huang, X.; Cao, M.; Zhao, H. Integrating Biocatalysis with Chemocatalysis for Selective Transformations. *Curr. Opin. Chem. Biol.* **2020**, *55*, 161–170.

(24) Heuson, E.; Dumeignil, F. The Various Levels of Integration of Chemo- and Bio-Catalysis towards Hybrid Catalysis. *Catal. Sci. Technol.* **2020**, *10*, 7082–7100.

(25) Liu, Y.; Liu, P.; Gao, S.; Wang, Z.; Luan, P.; González-Sabín, J.; Jiang, Y. Construction of Chemoenzymatic Cascade Reactions for Bridging Chemocatalysis and

Biocatalysis: Principles, Strategies and Prospective. *Chem. Eng. Technol.* **2021**, *420*, 127659.

(26) An, C.; Maloney, K. M. Designing for Sustainability with Biocatalytic and Chemoenzymatic Cascade Processes. *Curr. Opin. Green Sustain. Chem*. **2022**, *34*, 100591.

(27) Bering, L.; Thompson, J.; Micklefield, J. New Reaction Pathways by Integrating Chemo- and Biocatalysis. *Trends Chem.* **2022**, *4*, 392–408.

(28) Khan, N. R.; Rathod, V. K. Microwave Assisted Enzymatic Synthesis of Speciality Esters: A Mini - Review. *Process Biochem.* **2018**, *75*, 89–98.

(29) Horikoshi, S.; Serpone, N. Microwave Flow Chemistry as a Methodology in Organic Syntheses, Enzymatic Reactions, and Nanoparticle Syntheses. *Chem. Rec.* **2019**, *19*, 118–139.

(30) Romero-Fernández, M.; Paradisi, F. Protein Immobilization Technology for Flow Biocatalysis. *Curr. Opin. Chem. Biol.* **2020**, *55*, 1–8.

(31) Britton, J.; Majumdar, S.; Weiss, G. A. Continuous Flow Biocatalysis. *Chem. Soc. Rev*. **2018**, *47*, 5891–5918.

(32) Thompson, M. P.; Peñafiel, I.; Cosgrove, S. C.; Turner, N. J. Biocatalysis Using Immobilized Enzymes in Continuous Flow for the Synthesis of Fine Chemicals. *Org. Process Res. Dev.* **2019**, *23*, 9–18.

(33) Muschiol, J.; Peters, C.; Oberleitner, N.; Mihovilovic, M. D.; Bornscheuer, U. T.; Rudroff, F. Cascade Catalysis – Strategies and Challenges En Route to Preparative Synthetic Biology. *Chem. Commun.* **2015**, *51*, 5798–5811.

(34) Allen, J. V.; Williams, J. M. J. Dynamic Kinetic Resolution with Enzyme and Palladium Combinations. *Tetrahedron Lett.* **1996**, *37*, 1859–1862.

(35) Dinh, P. M.; Howarth, J. A.; Hudnott, A. R.; Williams, J. M. J.; Harris, W. Catalytic Racemisation of Alcohols: Applications to Enzymatic Resolution Reactions. *Tetrahedron Lett.* **1996**, *37*, 7623–7626.

(36) de Miranda, A. S.; Miranda, L. S. M.; de Souza, R. O. M. A. Lipases: Valuable Catalysts for Dynamic Kinetic Resolutions. *Biotechnol. Adv*. **2015**, *33*, 372–393.

(37) Långvik, O.; Saloranta, T.; Murzin, D. Y.; Leino, R. Heterogeneous Chemoenzymatic Catalyst Combinations for One-Pot Dynamic Kinetic Resolution Applications. *ChemCatChem* **2015**, *7*, 4004–4015.

(38) Verho, O.; Bäckvall, J.-E.; Chemoenzymatic Dynamic Kinetic Resolution: A Powerful Tool for the Preparation of Enantiomerically Pure Alcohols and Amines. *J. Am Chem. Soc.* **2015**, *137*, 3996–4009.

(39) Yang, L.-C.; Deng, H.; Renata, H. Recent Progress and Developments in Chemoenzymatic and Biocatalytic Dynamic Kinetic Resolution. *Org. Process Res. Dev.* **2022**, *26*, 1925–1943.

(40) Koszelewski, D.; Brodzka, A.; Żądło, A.; Paprocki, D.; Trzepizur, D.; Zysk, M.; Ostaszewski, R. Dynamic Kinetic Resolution of 3-Aryl-4-pentenoic Acids. *ACS Catal*. **2016**, *6*, 3287–3292.

(41) Lee, J.; Oh, Y.; Choi, Y. K.; Choi, E.; Kim, K.; Park, J.; Kim, M.-J. Dynamic Kinetic Resolution of Diarylmethanols with an Activated Lipoprotein Lipase. *ACS Catal*, **2015**, *5*, 683–689.

(42) Hilker, S.; Posevins, D.; Unelius, C. R.; Bäckvall, J.-E.; Chemoenzymatic Dynamic Kinetic Asymmetric Transformations of β-Hydroxyketones. *Chem. Eur. J.* **2021**, *27*, 15623–15627.

(43) Koszelewski, D.; Borys, F.; Brodzka, A.; Ostaszewski, R. Synthesis of Enantiomerically Pure 5,6-Dihydropyran-2-ones via Chemoenzymatic Sequential DKR-RCM Reaction. *Eur. J. Org. Chem.* **2019**, 1653–1658.

(44) Moustafa, G. A. I.; Oki, Y.; Akai, S. Lipase-Catalyzed Dynamic Kinetic Resolution of C_1- and C_2-Aymmetric Racemic Axially Chiral 2,2'-Dihydroxy-1,1'-biaryls. *Angew. Chem. Int. Ed.* **2018**, *57*, 10278–10282.

(45) El-Sepelgy, O.; Alandini, N.; Rueping. M. Merging Iron Catalysis and Biocatalysis–Iron Carbonyl Complexes as Efficient Hydrogen Autotransfer Catalysts in Dynamic Kinetic Resolutions. *Angew. Chem. Int. Ed.* **2016**, *55*, 13602–13605.

(46) Gustafson, K. P. J.; Guðmundsson, A.; Lewis, K.; Bäckvall, J.-E. Chemoenzymatic Dynamic Kinetic Resolution of Secondary Alcohols Using an Air- and Moisture-Stable Iron Racemization Catalyst. *Chem. Eur. J.* **2017**, *23*, 1048–1051.

(47) Yang, Q.; Zhang, N.; Liu, M.; Zhou, S. New Air-Stable Iron Catalyst for Efficient Dynamic Kinetic Resolution of Secondary Benzylic and Aliphatic Alcohols. *Tetrahedron Lett.* **2017**, *58*, 2487–2489.

(48) Akai, S.; Tanimoto, K.; Kanao, Y.; Egi, M.; Yamamoto, T.; Kita, Y. A Dynamic Kinetic Resolution of Allyl Alcohols by the Combined Use of Lipases and $[VO(OSiPh_3)_3]$. *Angew. Chem. Int. Ed*. **2006**, *45*, 2592–2595.

(49) Egi, M.; Sugiyama, K.; Saneto, M.; Hanada, R.; Kato, K.; Akai, S. A Mesoporous-Silica-Immobilized Oxovanadium Cocatalyst for the Lipase-Catalyzed Dynamic Kinetic Resolution of Racemic Alcohols. *Angew. Chem. Int. Ed*. **2013**, *52*, 3654–3658.

(50) De Almeida, L. A.; Marcondes, T. H.; Milagre, C. D. F.; Milagre, H. M. S. Lipase-Oxovanadium Heterogeneous Catalysis System: A Robust Protocol for the Dynamic Kinetic Resolution of *sec*-Alcohols. *ChemCatChem* **2020**, *12*, 2849–2858.

(51) Kawanishi, S.; Oki, S.; Kundu, D.; Akai, S. Lipase/Oxovanadium Co-Catalyzed Dynamic Kinetic Resolution of Propargyl Alcohols: Competition between Racemization and Rearrangement. *Org. Lett.* **2019**, *21*, 2978–2982.

(52) Kühn, F.; Katsuragi, S.; Oki, Y.; Scholz, C.; Akai, S.; Gröger, H. Dynamic Kinetic Resolution of a Tertiary Alcohol. *Chem. Commun.* **2020**, *56*, 2885–2888.

(53) Sugiyama, K.; Oki, Y.; Kawanishi, S.; Kato, K.; Ikawa, T.; Egi, M.; Akai, S. Spatial Effects of Oxovanadium-Immobilized Mesoporous Silica on Racemization of Alcohols and Application in Lipase-Catalyzed Dynamic Kinetic Resolution. *Catal. Sci. Technol.* **2016**, *6*, 5023–5030.

(54) Sugiyama, K.; Kawanishi, S.; Oki, Y.; Kamiya, M.; Hanada, R.; Egi, M.; Akai, S. Lipase-Catalyzed Asymmetric Synthesis of Naphtho[2,3-*c*]furan-1(3*H*)-one Derivatives by a One-Pot Dynamic Kinetic Resolution/Intramolecular Diels–Alder Reaction: Total Synthesis of (–)-Himbacine. *Bioorg. Med. Chem.* **2018**, *26*, 1378–1386.

(55) Kawanishi, S.; Sugiyama, K.; Oki, Y.; Ikawa, T.; Akai, S. Preparation of Optically Active Cycloalkenes Bearing All-Carbon Quaternary Stereogenic Centres via Lipase–Oxovanadium Combo-Catalysed Dynamic Kinetic Resolution. *Green Chem.* **2017**, *19*, 411–417.

(56) Tsuchimochi, I.; Hori, S.; Takeuchi, Y.; Egi, M.; Satoh, T.; Kanomata, K.; Ikawa, T.; Akai, S. Four-Step One-Pot Catalytic Asymmetric Synthesis of Polysubstituted Tricyclic Compounds: Lipase-Catalyzed Dynamic Kinetic Resolution Followed by an Intramolecular Diels–Alder Reaction. *Synlett* **2021**, *32*, 822–828.

(57) Higashio, K.; Katsuragi, S.; Kundu, D.; Adebar, N.; Plass, C.; Kühn, F.; Gröger, H.; Akai, S. Continuous-Flow Dynamic Kinetic Resolution of Racemic Alcohols by Lipase–Oxovanadium Cocatalysis. *Eur. J. Org. Chem.* **2020**, 1961–1967.

(58) de Miranda, A. S.; Silva, M. V. M.; Dias, F. C.; de Souza, S. P.; Leão, R. A. C.; de Souza, R. O. M. A. Continuous Flow Dynamic Kinetic Resolution of *rac*-1-Phenylethanol Using a Single Packed-Bed Containing Immobilized CAL-B Lipase and $VOSO_4$ as Racemization Catalysts. *React. Chem. Eng.* **2017**, *2*, 375–381.

(59) de França, A. S.; Silva, M. V. M.; Neves, R. V.; de Souza, S. P.; Leão, R. A. C.; Monteiro, C. M.; Rocha, Â.; Afonso, C. A. M.; de Souza, R. O. M. A. Studies on the Dynamic Resolution of Crizotinib Intermediate. *Bioorg. Med. Chem.* **2018**, *26*, 1333–1337.

(60) Xu, S.; Wang, M.; Feng, B.; Han, X.; Lan, Z.; Gu, H.; Li, H.; Li, H. Dynamic Kinetic Resolution of Amines by Using Palladium Nanoparticles Confined inside the Cages of Amine-Modified MIL-101 and Lipase. *J. Catal.* **2018**, *363*, 9–17.

(61) Wang, M.; Wang, X.; Feng, B.; Li, Y.; Han, X.; Lan, Z.; Gu, H.; Sun, H.; Shi, M.; Li, H. *et al.* Combining Pd Nanoparticles on MOFs with Cross-Linked Enzyme Aggregates of Lipase as Powerful Chemoenzymatic Platform for One-Pot Dynamic Kinetic Resolution of Amines. *J. Catal.* **2019**, *378*, 153–163.

(62) Farkas, E.; Oláh, M.; Földi, A.; Kóti, J.; Éles, J.; Nagy, J.; Gal, C. A.; Paizs, C.; Hornyánszky, G.; Poppe, L. Chemoenzymatic Dynamic Kinetic Resolution of Amines in Fully Continuous-Flow Mode. *Org. Lett.* **2018**, *20*, 8052–8056.

(63) Palo-Nieto, C.; Afewerki, S.; Anderson, M.; Tai, C.-W.; Berglund, P.; Córdova, A. Integrated Heterogeneous Metal/Enzymatic Multiple Relay Catalysis for Eco-Friendly and Asymmetric Synthesis. *ACS Catal*. **2016**, *6*, 3932–3940.

(64) Parvulescu, A.; De Vos, D.; Jacobs, P. Efficient Dynamic Kinetic Resolution of Secondary Amines with Pd on Alkaline Earth Salts and a Lipase. *Chem. Commun.* **2005**, 5307–5309.

(65) Gustafson, K. P. J.; Lihammar, R.; Verho, O.; Engström, K.; Bäckvall, J.-E. Chemoenzymatic Dynamic Kinetic Resolution of Primary Amines Using a Recyclable Palladium Nanoparticle Catalyst Together with Lipases. *J. Org. Chem*. **2014**, *79*, 3747–3751.

(66) Zitler, K. Photoredox Catalysis with Visible Light. *Angew. Chem. Int. Ed*. **2009**, *48*, 9785–9789.

(67) Silvi, M.; Melchiorre, P. Enhancing the Potential of Enantioselective Organocatalysis with Light. *Nature* **2018**, *554*, 41–49.

(68) Yang, Q.; Zhao, F.; Zhang, N.; Liu, M.; Hu, H.; Zhang, J.; Zhou, S. Mild Dynamic Kinetic Resolution of Amines by Coupled Visible-Light Photoredox and Enzyme Catalysis. *Chem. Commun.* **2018**, *54*, 14065–14068.

(69) Gastaldi, S.; Escoubet, S.; Vanthuyne, N.; Gil, G.; Bertrand, M. P. Dynamic Kinetic Resolution of Amines Involving Biocatalysis and in situ Free Radical Mediated Racemization. *Org. Lett.* **2007**, *9*, 837–839.

(70) Sandoval, B. A.; Hyster, T. K. Emerging Strategies for Expanding the Toolbox of Enzymes in Biocatalysis. *Curr. Opin. Chem. Biol.* **2020**, *55*, 45–51.

(71) DeHovitz, J. S.; Loh, Y. Y.; Kautzky, J. A.; Nagao, K.; Meichan, A. J.; Yamauchi, M.; MacMillan, D. W. C.; Hyster, T. K. Static to Inducibly Dynamic Stereocontrol: The Convergent Use of Racemic β-Substituted Ketones. *Science* **2020**, *369*, 1113–1118.

(72) Stradomska, D.; Heba, M.; Czernek, A.; Kuźnik, N.; Gillner, D.; Maresz, K.; Pudło, W.; Jarzębski, A.; Szymańska, K. Lipase Immobilized on MCFs as Biocatalysts for Kinetic and Dynamic Kinetic Resolution of *sec*-Alcohols. *Catalysts* **2021**, *11*, 518.

(73) Liu, X.; Li, B.; Liu, Q. Base-Metal-Catalyzed Olefin Isomerization Reactions. *Synthesis* **2019**, *51*, 1293–1310.

(74) Sheldon, R. A. The E Factor: Fifteen Years on. *Green Chem.* **2007**, *9*, 1273–1283.

(75) Martín-Matute, B.; Edin, M.; Bogár, K.; Kaynak, F. B.; Bäckvall, J.-E. Combined Ruthenium(II) and Lipase Catalysis for Efficient Dynamic Kinetic Resolution of Secondary Alcohols. Insight into the Racemization Mechanism. *J. Am. Chem. Soc.* **2005**, *127*, 8817–8825.

(76) Bogár, K.; Martín-Matute, B.; Bäckvall, J.-E. Large-Scale Ruthenium- and Enzyme-Catalyzed Dynamic Kinetic Resolution of (*rac*)-1-Phenylethanol. *Beilstein J. Org. Chem.* **2007**, *3*, 50.

(77) Karvembu, R.; Prabhakaran, R.; Tamizh, M. M.; Natarajan, K. Ruthenium and Enzyme-Catalyzed Dynamic Kinetic Resolution of Alcohols. *C. R. Chimie* **2009**, *12*, 951–962.

(78) Yang, B.; Zhu, C.; Qiu, Y.; Bäckvall, J.-E. Enzyme- and Ruthenium-Catalyzed Enantioselective Transformation of α-Allenic Alcohols into 2,3-Dihydrofurans. *Angew. Chem. Int. Ed.* **2016**, *55*, 5568–5572.

(79) Smith, E. L.; Abbott, A. P.; Ryder, K. S. Deep Eutectic Solvents (DESs) and Their Applications. *Chem. Rev.* **2014**, *114*, 11060–11082.

(80) Kourist, R.; González-Sabín, J. Non-Conventional Media as Strategy to Overcome the Solvent Dilemma in Chemoenzymatic Tandem Catalysis. *ChemCatChem* **2020**, *12*, 1903–1912.

(81) Hansen, B. B.; Spittle, S.; Chen, B.; Poe, D.; Zhang, Y.; Klein, J. M.; Horton, A.; Adhikari, L.; Zelovich, T.; Doherty, B. W. *et al.* Deep Eutectic Solvents: A Review of Fundamentals and Applications. *Chem. Rev.* **2021**, *121*, 1232–1285.

(82) Cicco, L.; Dilauro, G.; Perna, F. M.; Vitale, P.; Capriati, V. Advances in Deep Eutectic Solvents and Water: Applications in Metal- and Biocatalyzed Processes, in the Synthesis of APIs, and other Biologically Active Compounds. *Org. Biomol. Chem.* **2021**, *19*, 2558–2577.

(83) Ríos-Lombardía, N.; Vidal, C.; Cocina, M.; Morís, F.; García-Álvarez, J.; González-Sabín, J. Chemoenzymatic One-Pot Synthesis in Aqueous Medium: Combination of Metal-Catalysed Allylic Alcohol Isomerisation–Asymmetric Bioamination. *Chem. Commun.* **2015**, *51*, 10937–10940.

(84) Ríos-Lombardía, N.; Vidal, C.; Liardo, E.; Morís, F.; García-Álvarez, J.; González-Sabín, J. From a Sequential to a Concurrent Reaction in Aqueous Medium: Ruthenium-Catalyzed Allylic Alcohol Isomerization and Asymmetric Bioreduction. *Angew. Chem. Int. Ed*. **2016**, *55*, 8691–8695.

(85) Cicco, L.; Ríos-Lombardía, N.; Rodríguez-Álvarez, M. J.; Morís, F.; Perna, F. M.; Capriati, V.; García-Álvarez, J.; González-Sabín, J. Programming Cascade Reactions Interfacing Biocatalysis with Transition-Metal Catalysis in *Deep Eutectic Solvents* as Biorenewable Reaction Media. *Green Chem.* **2018**, *20*, 3468–3475.

(86) Gotor-Fernández, V.; Paul, C. E. Deep Eutectic Solvents for Redox Biocatalysis. *J. Biotechnol.* **2019**, *293*, 24–35.

(87) Ibn Majdoub Hassani, F. Z.; Amzazi, S.; Lavandera, I. The Versatile Applications of DES and Their Influence on Oxidoreductase-Mediated Transformations. *Molecules* **2019**, *24*, 2190.

(88) Rodríguez-Álvarez, M. J.; Ríos-Lombardía, N.; Schumacher, S.; Pérez-Iglesias, D.; Morís, F.; Cadierno, V.; García-Álvarez, J.; González-Sabín, J. Combination of Metal-Catalyzed Cycloisomerizations and Biocatalysis in Aqueous Media: Asymmetric Construction of Chiral Alcohols, Lactones, and γ-Hydroxy-Carbonyl Compounds. *ACS Catal*. **2017**, *7*, 7753–7759.

(89) González-Granda, S.; Lavandera, I.; Gotor-Fernández, V. Alcohol Dehydrogenases and N-Heterocyclic Carbene Gold(I) Catalysts: Design of a Chemoenzymatic Cascade towards Optically Active β,β-Disubstituted Allylic Alcohols. *Angew. Chem. Int. Ed.* **2021**, *60*, 13945–13951.

(90) Engel, D. A.; Dudley, G. B. The Meyer–Schuster Rearrangement for the Synthesis of α,β-Unsaturated Carbonyl Compounds. *Org. Biomol. Chem.* **2009**, *7*, 4149–4158.

(91) Roy, D.; Tharra, P.; Baire, B. Intercepted Meyer–Schuster Rearrangements in Organic Synthesis. *Asian J. Org. Chem.* **2018**, *7*, 1015–1032.

(92) Litman, Z. C.; Wang, Y.; Zhao, H.; Hartwig, J. F. Cooperative Asymmetric Reactions Combining Photocatalysis and Enzymatic Catalysis. *Nature* **2018**, *560*, 355–359.

(93) Tsuji, J. Synthetic Applications of the Palladium-Catalyzed Oxidation of Olefins to Ketones. *Synthesis* **1984**, 369–384.

(94) Fernandes, R. A.; Jha, A. K.; Kumar, P. Recent Advances in Wacker Oxidation: From Conventional to Modern Variants and Applications. *Catal. Sci. Technol.* **2020**, *10*, 7448–7470.

(95) Schnapperelle, I.; Hummel, W.; Gröger, H. Formal Asymmetric Hydration of Non-Activated Alkenes in Aqueous Medium through a "Chemoenzymatic Catalytic System". *Chem. Eur. J*. **2012**, *18*, 1073–1076.

(96) Sato, H.; Hummel, W.; Gröger, H. Cooperative Catalysis of Noncompatible Catalysts through Compartmentalization: Wacker Oxidation and Enzymatic Reduction in a One-Pot Process in Aqueous Media. *Angew. Chem. Int. Ed*. **2015**, *54*, 4488–4492.

(97) Uthoff, F.; Sato, H.; Gröger, H. Formal Enantioselective Hydroamination of Non-Activated Alkenes: Transformation of Styrenes into Enantiomerically Pure 1-Phenylethylamines in Chemoenzymatic One-Pot Synthesis. *ChemCatChem* **2017**, *9*, 555–558.

(98) Uthoff, F.; Gröger, H. Asymmetric Synthesis of 1-Phenylethylamine from Styrene via Combined Wacker Oxidation and Enzymatic Reductive Amination. *J. Org. Chem*. **2018**, *83*, 9517–9521.

(99) González-Martínez, D.; Gotor, V.; Gotor-Fernández, V. Stereoselective Synthesis of 1-Arylpropan-2-amines from Allylbenzenes through a Wacker-Tsuji Oxidation-Biotransamination Sequential Process. *Adv. Synth. Catal.* **2019**, *361*, 2582–2593.

(100) Albarrán-Velo, J.; Gotor-Fernández, V.; Lavandera, I. Markovnikov Wacker-Tsuji Oxidation of Allyl(hetero)arenes and Application in a One-Pot Photo-Metal-Biocatalytic Approach to Enantioenriched Amines and Alcohols. *Adv. Synth. Catal.* **2021**, *363*, 4096–4108.

(101) Sand, H.; Weberskirch, R. Chemoenzymatic One-Pot Reaction of Noncompatible Catalysts: Combining Enzymatic Ester Hydrolysis with Cu(I)/Bipyridine Catalyzed Oxidation in Aqueous Medium. *RSC Adv.* **2017**, *7*, 33614–33626.

(102) Carceller, J. M.; Mifsud, M.; Climent, M. J.; Iborra, S.; Corma, A. Production of Chiral Alcohols from Racemic Mixtures by Integrated Heterogeneous Chemoenzymatic Catalysis in Fixed Bed Continuous Operation. *Green Chem.* **2020**, *22*, 2767–2777.

(103) van der Waal, J. C.; Tan, K.; van Bekkum, H. Zeolite Titanium Beta: A Selective and Water Resistant Catalyst in Meerwein-Ponndorf-Verley-Oppenauer Reactions. *Catal. Lett.* **1996**, *41*, 63–67.

(104) Boronat, M.; Corma, A.; Renz, M.; Viruela, P. M. Predicting the Activity of Single Isolated Lewis Acid Sites in Solid Catalysts. *Chem. Eur. J.* **2006**, *12*, 7067–7077.

(105) Lancien, A.; Wojcieszak, R.; Cuvelier, E.; Duban, M.; Dhulster, P.; Paul, S.; Dumeignil, F.; Froidevaux, R.; Heuson, E. Hybrid Conversion of 5-Hydroxymethylfurfural to 5-Aminomethyl-2-furancarboxylic Acid: Toward New Biosourced Polymers. *ChemCatChem* **2021**, *13*, 247–259.

(106) Yuan, B.; Debecker, D. P.; Wu, X.; Xiao, J.; Fei, Q.; Turner, N. J. One-Pot Chemoenzymatic Deracemisation of Secondary Alcohols Employing Variants of Galactose Oxidase and Transfer Hydrogenation. *ChemCatChem* **2020**, *12*, 6191–6195.

(107) Wang, J.; Li, K.; Zhou, X.; Han, W.; Wan, N.; Cui, B.; Wang, H.; Yuan, W.; Chen, Y. Asymmetric Combinational ‘‘Metal-Biocatalytic System”: One Approach to Chiral 2-Substituted-tetrahydroquinoline-4-ols towards Two-Step One-Pot Processes in Aqueous Media. *Tetrahedron Lett.* **2017**, *58*, 2252–2254.

(108) Biewenga, L.; Saravanan, T.; Kunzendorf, A.; van der Meer, J.-Y.; Pijning, T.; Tepper, P. G.; van Merkerk, R.; Charnock, S. J.; Thunnissen, A.-M. W. H.; Poelarends, G. J. Enantioselective Synthesis of Pharmaceutically Active γ-Aminobutyric Acids Using a Tailor-Made Artificial Michaelase in One-Pot Cascade Reactions. *ACS Catal.* **2019**, *9*, 1503–1513.

(109) Sperl, J. M.; Carsten, J. M.; Guterl, J.-K.; Lommes, P.; Sieber, V. Reaction Design for the Compartmented Combination of Heterogeneous and Enzyme Catalysis. *ACS Catal.* **2016**, *6*, 6329–6334.

(110) Sun, J.; Li, H.; Huang, H.; Wang, B.; Xiao, L.-P.; Song, G. Integration of Enzymatic and Heterogeneous Catalysis for One-Pot Production of Fructose from Glucose. *ChemSusChem* **2018**, *11*, 1157–1162.

(111) Zhou, Y.; Wu, S.; Bornscheuer, U. T. Recent Advances in (Chemo)enzymatic Cascades for Upgrading Bio-Based Resources. *Chem. Commun.* **2021**, *57*, 10661–10674.

(112) Arias, K. S.; Carceller, J. M.; Climent, M. J.; Corma, A.; Iborra, S. Chemoenzymatic Synthesis of 5-Hydroxymethylfurfural (HMF)-Derived Plasticizers by Coupling HMF Reduction with Enzymatic Esterification. *ChemSusChem* **2020**, *13*, 1864–1875.

(113) Enoki, J.; Mügge, C.; Tischler, D.; Miyamoto, K.; Kourist, R. Chemoenzymatic Cascade Synthesis of Optically Pure Alkanoic Acids by Using Engineered Arylmalonate Decarboxylase Variants. *Chem. Eur. J.* **2019**, *25*, 5071–5076.

(114) Beller, M.; Seayad, J.; Tillack, A.; Jiao, H. Catalytic Markovnikov and anti-Markovnikov Functionalization of Alkenes and Alkynes: Recent Developments and Trends. *Angew. Chem. Int. Ed.* **2004**, *43*, 3368–3398.

(115) Hintermann, L.; Labonne. A. Catalytic Hydration of Alkynes and Its Application in Synthesis. *Synthesis* **2007**, 1121–1150.

(116) Olah, G. A.; Meidar. D. Heterogeneous Catalysis by Solid Superacids; 10. Mercury Impregnated Nafion-H Perfluorinated Resinsulfonic Acid Catalyzed Hydration of Alkynes. *Synthesis* **1978**, 671–672.

(117) Hou, S.; Yang, H.; Cheng, B.; Zhai, H.; Li. Y. Cobaloxime-Catalyzed Hydration of Terminal Alkynes without Acidic Promoters. *Chem. Commun.* **2017**, *53*, 6926–6929.

(118) Schaaf, P.; Gojic, V.; Bayer, T.; Rudroff, F.; Schnürch, M.; Mihovilovic. M. D. Easy Access to Enantiopure (*S*)- and (*R*)-Aryl Alkyl Alcohols by a Combination of Gold(III)-Catalyzed Alkyne Hydration and Enzymatic Reduction. *ChemCatChem* **2018**, *10*, 920–924.

(119) Mathew, S.; Sagadevan, A.; Renn, D.; Rueping. M. One-Pot Chemoenzymatic Conversion of Alkynes to Chiral Amines. *ACS Catal.* **2021**, *11*, 12565–12569.

(120) Cortes-Clerget, M.; Akporji, N.; Zhou, J.; Gao, F.; Guo, P.; Parmentier, M.; Gallou, F.; Berthon, J.-Y.; Lipshutz, B. H. Bridging the Gap between Transition Metal- and Bio-Catalysis via Aqueous Micellar Catalysis. *Nat. Commun.* **2019**, *10*, 2169.

(121) Andersson, M. P.; Gallou, F.; Klumphu, P.; Takale, B. S.; Lipshutz, B. H. Structure of nanoparticles derived from designer surfactant TPGS-750-M in water, as used in organic synthesis. *Chem. Eur. J.* **2018**. *24*, 6778–6786.

(122) Liardo, E.; González-Fernández, R.; Ríos-Lombardía, N.; Morís, F.; García-Álvarez, J.; Cadierno, V.; Crochet, P.; Rebolledo, F.; González-Sabín, J. Strengthening the Combination between Enzymes and Metals in Aqueous Medium: Concurrent Ruthenium-Catalyzed Nitrile Hydration - Asymmetric Ketone Bioreduction. *ChemCatChem* **2018**, *10*, 4676–4682.

(123) Grubbs, R. H. Olefin Metathesis. *Tetrahedron* **2004**, *60*, 7117–7140.

(124) Grubbs, R. H.; Wenzel, A. G. Handbook of Metathesis, Wiley-VCH, Weinheim (Germany), 2nd edn, vol. 2, 2015; pp 1-744.

(125) Tenbrink, K.; Seßler, M.; Schatz, J.; Gröger. H. Combination of Olefin Metathesis and Enzymatic Ester Hydrolysis in Aqueous Media in a One-Pot Synthesis. *Adv. Synth. Catal*. **2011**, *353*, 2363–2367.

(126) Denard, C. A.; Huang, H.; Bartlett, M. J.; Lu, L.; Tan, Y.; Zhao, H.; Hartwig. J. F. Cooperative Tandem Catalysis by an Organometallic Complex and a Metalloenzyme. *Angew. Chem. Int. Ed*. **2014**, *53*, 465–469.

(127) Denard, C. A.; Bartlett, M. J.; Wang, Y.; Lu, L.; Hartwig, J. F.; Zhao, H. Development of a One-Pot Tandem Reaction Combining Ruthenium-Catalyzed Alkene Metathesis and Enantioselective Enzymatic Oxidation to Produce Aryl Epoxides. *ACS Catal*. **2015**, *5*, 3817–3822.

(128) Gómez-Baraibar, Á.; Reichert, D.; Mügge, C.; Seger, S.; Gröger, H.; Kourist, R. A One-Pot Cascade Reaction Combining an Encapsulated Decarboxylase with a Metathesis Catalyst for the Synthesis of Bio-Based Antioxidants. *Angew. Chem. Int. Ed*. **2016**, *55*, 14823–14827.

(129) Mertens, M. A. S.; Sauer, D. F.; Markel, U.; Schiffels, J.; Okuda, J.; Schwaneberg, U. Chemoenzymatic Cascade for Stilbene Production from Cinnamic Acid Catalyzed by

Ferulic Acid Decarboxylase and an Artificial Metathease. *Catal. Sci. Technol.* **2019**, *9*, 5572–5576.

(130) Bojarra, S.; Reichert, D.; Grote, M.; Gómez Baraibar, Á.; Dennig, A.; Nidetzky, B.; Mügge, C.; Kourist, R. Bio-based α,ω-Functionalized Hydrocarbons from Multistep Reaction Sequences with Bio- and Metallo-catalysts Based on the Fatty Acid Decarboxylase $OleT_{JE}$. *ChemCatChem* **2018**, *10*, 1192–1201.

(131) Hastings, C. J.; Adams, N. P.; Bushi, J.; Kolb, S. J. One-pot Chemoenzymatic Reactions in Water Enabled by Micellar Encapsulation. *Green Chem.* **2020**, *22*, 6187–6193.

(132) Maaskant, R. V.; Chordia, S.; Roelfes, G. Merging Whole-cell Biosynthesis of Styrene and Transition-metal Catalyzed Derivatization Reactions. *ChemCatChem* **2021**, *13*, 1607–1613.

(133) Deiters, A.; Martin, S. F. Synthesis of Oxygen- and Nitrogen-Containing Heterocycles by Ring-Closing Metathesis. *Chem. Rev.* **2004**, *104*, 2199–2238.

(134) Groso, E. J.; Schindler; C. S. Recent Advances in the Application of Ring-Closing Metathesis for the Synthesis of Unsaturated Nitrogen Heterocycles. *Synthesis* **2019**, *51*, 1100–1114.

(135) Scalacci, N.; Black, G. W.; Mattedi, G.; Brown, N. L.; Turner, N. J. Castagnolo, D. Unveiling the Biocatalytic Aromatizing Activity of Monoamine Oxidases MAO-N and 6-HDNO: Development of Chemoenzymatic Cascades for the Synthesis of Pyrroles. *ACS Catal*. **2017**, *7*, 1295–1300.

(136) Risi, C.; Zhao, F.; Castagnolo, D. Chemo-Enzymatic Metathesis/Aromatization Cascades for the Synthesis of Furans: Disclosing the Aromatizing Activity of Laccase/TEMPO in Oxygen-Containing Heterocycles. *ACS Catal*. **2019**, *9*, 7264–7269.

(137) Pauly, J.; Gröger, H.; Patel, A. V. Catalysts Encapsulated in Biopolymer Hydrogels for Chemoenzymatic One-Pot Processes in Aqueous Media. *ChemCatChem* **2019**, *11*, 1503–1509.

(138) Pauly, J.; Gröger, H.; Patel, A. V. Developing Multicompartment Biopolymer Hydrogel Beads for Tandem Chemoenzymatic One-Pot Process. *Catalysts* **2019**, *9*, 547.

(139) Wu, S.; Zhou, Y.; Gerngross, D.; Jeschek, M.; Ward, T. R. Chemo-Enzymatic Cascades to Produce Cycloalkenes from Bio-Based Resources. *Nat. Commun.* **2019**, *10*, 5060.

(140) Geist, E.; Kirschning, A.; Schmidt, T. sp^3-sp^3 Coupling Reactions in the Synthesis of Natural Products and Biologically Active Molecules. *Nat. Prod. Rep.* **2014**, *31*, 441–448.

(141) Chinchilla, R.; Nájera, C. The Sonogashira Reaction: A Booming Methodology in Synthetic Organic Chemistry. *Chem. Rev.* **2007**, *107*, 874–922.

(142) Haas, D.; Hammann, J. M.; Greiner, R.; Knochel, P. Recent Developments in Negishi Cross-Coupling Reactions. *ACS Catal*. **2016**, *6*, 1540–1552.

(143) Brittain, W. D.; G. Cobb; S. L. Negishi Cross-Couplings in the Synthesis of Amino Acids. *Org. Biomol. Chem*. **2018**, *16*, 10–20.

(144) Koshvandi, A. T. K.; Heravi, M. M.; Momeni, T. Current Applications of Suzuki-Miyaura Coupling Reaction in The Total Synthesis of Natural Products: An update. *Appl. Organometal. Chem*. **2018**, *32*, e4210.

(145) Ghosh, T. Reductive Heck Reaction: An Emerging Alternative in Natural Product Synthesis. *ChemistrySelect* **2019**, *4*, 4747–4755.

(146) Wu, X.-F.; Anbarasan, P.; Neumann, H.; Beller, M. From Noble Metal to Nobel Prize: Palladium-Catalyzed Coupling Reactions as Key Methods in Organic Synthesis. *Angew. Chem. Int. Ed.* **2010**, *49*, 9047–9050.

(147) Paul, D.; Das, S.; Saha, S.; Sharma, H.; Goswami, R. K. Intramolecular Heck Reaction in Total Synthesis of Natural Products: An Update. *Eur. J. Org. Chem.* **2021**, 2057–2076.

(148) Ríos-Lombardía, N.; Rodríguez-Álvarez, M. J.; Morís, F.; Kourist, R.; Comino, N.; López-Gallego, F.; González-Sabín, J.; García-Álvarez, J. DESign of Sustainable One-

Pot Chemoenzymatic Organic Transformations in Deep Eutectic Solvents for the Synthesis of 1,2-Disubstituted Aromatic Olefins. *Front. Chem.* **2020**, *8*, 139.

(149) Peng, M.; Mittmann, E.; Wenger, L.; Hubbuch, J.; Engqvist, M. K. M.; Niemeyer, C. M.; Rabe. K. S. 3D-Printed Phenacrylate Decarboxylase Flow Reactors for the Chemoenzymatic Synthesis of 4-Hydroxystilbene. *Chem. Eur. J.* **2019**, *25*, 15998–16001.

(150) Grabner, B.; Schweiger, A. K.; Gavric, K.; Kourist, R.; Gruber-Woelfler, H. A Chemo-Enzymatic Tandem Reaction in a Mixture of Deep Eutectic Solvent and Water in Continuous Flow. *React. Chem. Eng.* **2020**, *5*, 263–269.

(151) Maier, M. C.; Valotta, A.; Hiebler, K.; Soritz, S.; Gavric, K.; Grabner, B.; Gruber-Woelfler, H. 3D Printed Reactors for Synthesis of Active Pharmaceutical Ingredients in Continuous Flow. *Org. Process Res. Dev.* **2020**, *24*, 2197–2207.

(152) Lackner, F.; Hiebler, K.; Grabner, B.; Gruber-Woelfler; H. Optimization of a Catalytic Chemoenzymatic Tandem Reaction for the Synthesis of Natural Stilbenes in Continuous Flow. *Catalysts* **2020**, *10*, 1404.

(153) Schwendenwein, D.; Ressmann, A. K.; Entner, M.; Savic, V.; Winkler, M.; Rudroff, F. Chemo-Enzymatic Cascade for the Generation of Fragrance Aldehydes. *Catalysts* **2021**, *11*, 932.

(154) Gruß, H.; Belu, C.; Bernhard, L. M.; Merschel, A.; Sewald, N. Fluorogenic Diversification of Unprotected Bromotryptophan by Aqueous Mizoroki–Heck Cross-Coupling. *Chem. Eur. J.* **2019**, *25*, 5880–5883.

(155) Alisha, M.; Philip, R. M.; Anilkumar, G. Low-Cost Transition Metal-Catalyzed Heck-Type Reactions: An Overview. *Eur. J. Org. Chem.* **2022**, e202101384.

(156) Hooshmand, S. E.; Heidari, B.; Sedghi, R.; Varma, R. S. Recent Advances in the Suzuki–Miyaura Cross-Coupling Reaction Using Efficient Catalysts in Eco-Friendly Media. *Green Chem.* **2019**, *21*, 381–405.

(157) Kadu, B. S. Suzuki–Miyaura Cross Coupling Reaction: Recent Advancements in Catalysis and Organic Synthesis. *Catal. Sci. Technol.* **2021**, *11*, 1186–1221.

(158) Burda, E.; Hummel, W.; Gröger, H. Modular Chemoenzymatic One-Pot Syntheses in Aqueous Media: Combination of a Palladium-Catalyzed Cross-Coupling with an Asymmetric Biotransformation. *Angew. Chem. Int. Ed.* **2008**, *47*, 9551–9554.

(159) Gauchot, V.; Kroutil, W.; Schmitzer, A. R. Highly Recyclable Chemo-/Biocatalyzed Cascade Reactions with Ionic Liquids: One-Pot Synthesis of Chiral Biaryl Alcohols. *Chem. Eur. J.* **2010**, *16*, 6748–6751.

(160) Borchert, S.; Burda, E.; Schatz, J.; Hummel, W.; Gröger, H. Combination of a Suzuki Cross-Coupling Reaction Using a Water-Soluble Palladium Catalyst with an Asymmetric Enzymatic Reduction towards a One-Pot Process in Aqueous Medium at Room Temperature. *J. Mol. Catal. B: Enzym.* **2012**, *84*, 89–93.

(161) Paris, J.; Ríos-Lombardía, N.; Morís, F.; Gröger, H.; González-Sabín J. Novel Insights into the Combination of Metal- and Biocatalysis: Cascade One-Pot Synthesis of Enantiomerically Pure Biaryl Alcohols in Deep Eutectic Solvents. *ChemCatChem* **2018**, *10*, 4417–4423.

(162) Paris, J.; Telzerow, A.; Ríos-Lombardía, N.; Steiner, K.; Schwab, H.; Morís, F.; Gröger, H.; González-Sabín, J. Enantioselective One-Pot Synthesis of Biaryl-Substituted Amines by Combining Palladium and Enzyme Catalysis in Deep Eutectic Solvents. *ACS Sustain. Chem. Eng*. **2019**, *7*, 5486–5493.

(163) González-Martínez, D.; Gotor, V.; Gotor-Fernández, V. Chemoenzymatic Synthesis of an Odanacatib Precursor through a Suzuki-Miyaura Cross-Coupling and Bioreduction Sequence. *ChemCatChem* **2019**, *11*, 5800–5807.

(164) Dander, J. E.; Giroud, M.; Racine, S.; Darzi, E. R.; Alvizo, O.; Entwistle, D.; Garg, N. K. Chemoenzymatic Conversion of Amides to Enantioenriched Alcohols in Aqueous Medium. *Commun. Chem.* **2019**, *2*, 82.

(165) Tassano, E.; Merusic, K.; Buljubasic, I.; Laggner, O.; Reiter, T.; Vogel, A.; Hall, M. Regioselective Biocatalytic Self-Sufficient Tishchenko-type Reaction via Formal Intramolecular Hydride Transfer. *Chem. Commun.* **2020**, *56*, 6340–6343.

(166) Morris, S. A.; Gusev, D. G. Rethinking the Claisen–Tishchenko Reaction. *Angew. Chem. Int. Ed.* **2017**, *56*, 6228–6231.

(167) Luan, P.; Liu, Y.; Li, Y.; Chen, R.; Huang, C.; Gao, J.; Hollmann, F.; Jiang, Y. Aqueous Chemoenzymatic One-Pot Enantioselective Synthesis of Tertiary α-Aryl Cycloketones *via* Pd-Catalyzed C–C Formation and Enzymatic C=C Asymmetric Hydrogenation. *Green Chem.* **2021**, *23*, 1960–1964.

(168) Akporji, N.; Singhania, V.; Dussart-Gautheret, J.; Gallou, F.; Lipshutz, B. H. Nanomicelle-Enhanced, Asymmetric ERED-Catalyzed Reductions of Activated Olefins. Applications of 1-pot Chemo- and Bio-Catalysis Sequences in Water. *Chem. Commun.* **2021**, *57*, 11847–11850.

(169) Dawood, A. W. H.; Bassut, J.; de Souza, R. O. M. A.; Bornscheuer, U. T. Combination of the Suzuki–Miyaura Cross-Coupling Reaction with Engineered Transaminases. *Chem. Eur. J.* **2018**, *24*, 16009–16013.

(170) Ahmed, S. T.; Parmeggiani, F.; Weise, N. J.; Flitsch, S. L.; Turner, N. J. Chemoenzymatic Synthesis of Optically Pure L- and D-Biarylalanines through Biocatalytic Asymmetric Amination and Palladium-Catalyzed Arylation. *ACS Catal.* **2015**, *5*, 5410–5413.

(171) Schnepel, C.; Minges, H.; Frese, M.; Sewald, N. A High-Throughput Fluorescence Assay to Determine the Activity of Tryptophan Halogenases. *Angew. Chem. Int. Ed.* **2016**, *55*, 14159–14163.

(172) Frese, M.; Schnepel, C.; Minges, H.; Voß, H.; Feiner, R.; Sewald, N. Modular Combination of Enzymatic Halogenation of Tryptophan with Suzuki–Miyaura Cross-Coupling Reactions. *ChemCatChem* **2016**, *8*, 1799–1803.

(173) Latham, J.; Henry, J.-M.; Sharif, H. H.; Menon, B. R. K.; Shepherd, S. A.; Greaney, M. F.; Micklefield, J. Integrated Catalysis Opens New Arylation Pathways via Regiodivergent Enzymatic C-H Activation. *Nat. Commun.* **2016**, *7*, 11873.

(174) Frese, M.; Sewald, N. Enzymatic Halogenation of Tryptophan on a Gram Scale. *Angew. Chem. Int. Ed*. **2015**, *54*, 298–301.

(175) Craven, E. J.; Latham, J.; Shepherd, S. A.; Khan, I.; Diaz-Rodriguez, A.; Greaney, M. F.; Micklefield, J. Programmable Late-Stage C–H Bond Functionalization Enabled by Integration of Enzymes with Chemocatalysis. *Nat. Catal.* **2021**, *4*, 385–394.

(176) Gesse, P.; Müller, T. J. J. Consecutive Five-Component Ugi-4CR-CAL B-Catalyzed Aminolysis Sequence and Concatenation with Transition Metal Catalysis in a One-Pot Fashion to Substituted Triamides. *Eur. J. Org. Chem.* **2019**, 2150–2157.

(177) Chincilla, R.; Nájera C. Recent Advances in Sonogashira Reactions. *Chem. Soc. Rev.* **2011**, *40*, 5084–5121.

(178) Wang, D.; Gao, S. Sonogashira Coupling in Natural Product Synthesis. *Org. Chem. Front.* **2014**, *1*, 556–566.

(179) Hassan, S.; Ullrich, A.; Müller, T. J. J. Consecutive Three-Component Synthesis of (Hetero)arylated Propargyl Amides by Chemoenzymatic Aminolysis–Sonogashira Coupling Sequence. *Org. Biomol. Chem.* **2015**, *13*, 1571–1576.

(180) Mertens, M. A. S.; Thomas, F.; Nöth, M.; Moegling, J.; El-Awaad, I.; Sauer, D. F.; Dhoke, G. V.; Xu, W.; Pich, A.; Herres-Pawlis, S.; Schwaneberg, U. One-Pot Two-Step Chemoenzymatic Cascade for the Synthesis of a Bis-benzofuran Derivative. *Eur. J. Org. Chem.* **2019**, 6341–6346.

(181) For diazo compounds reactivity: Ford, A.; Miel, H.; Ring, A.; Slattery, C. N.; Maguire, A. R.; McKervey, M. A. Modern Organic Synthesis with α-Diazocarbonyl Compounds. *Chem. Rev*. **2015**, *115*, 9981–10080.

(182) Zhang, R. K.; Chen, K.; Huang, X.; Wohlschlager, L.; Renata, H.; Arnold, F. H. Enzymatic Assembly of Carbon–Carbon Bonds via Iron-Catalysed sp^3 C–H functionalization. *Nature* **2019**, *565*, 67–72.

(183) Carminati, D. M.; Fasan, R. Stereoselective Cyclopropanation of Electron-Deficient Olefins with a Cofactor Redesigned Carbene Transferase Featuring Radical Reactivity. *ACS Catal*. **2019**, *9*, 9683–9697.

(184) Wang, Y.; Bartlett, M. J.; Denard, C. A.; Hartwig, J. F.; Zhao, H. Combining Rh-Catalyzed Diazocoupling and Enzymatic Reduction to Efficiently Synthesize Enantioenriched 2-Substituted Succinate Derivatives. *ACS Catal*. **2017**, *7*, 2548–2552.

(185) Wallace, S.; Balskus, E. P. Interfacing Microbial Styrene Production with a Biocompatible Cyclopropanation Reaction. *Angew. Chem. Int. Ed.* **2015**, *54*, 7106–7109.

(186) Liebeskind, L. S.; Srogl, J. Thiol Ester-Boronic Acid Coupling. A Mechanistically Unprecedented and General Ketone Synthesis. *J. Am. Chem. Soc*. **2000**, *122*, 11260–11261.

(187) Schaaf, P.; Bayer, T.; Koley, M.; Schnürch, M.; Bornscheuer, U. T.; Rudroff, F.; Mihovilovic, M. D. Biocompatible Metal-Assisted C–C Cross-Coupling Combined with Biocatalytic Chiral Reductions in a Concurrent Tandem Cascade. *Chem. Commun.* **2018**, *54*, 12978–12981.

(188) Hashmi, A. S. K. Gold-Catalyzed Organic Reactions. *Top. Organomet. Chem.* **2013**, *44*, 143–164.

(188) Dorel, R.; Echavarren, A. M. Gold(I)-Catalyzed Activation of Alkynes for the Construction of Molecular Complexity. *Chem. Rev.* **2015**, *115*, 9028–9072.

(190) Boorman, T. C.; Larrosa, I. Gold-Mediated C–H Bond Functionalization. *Chem. Soc. Rev.* **2011**, *40*, 1910–1925.

(191) Wang, Z. J.; Clary, K. N.; Bergman, R. G.; Raymond K. N.; Toste, F. D. A Supramolecular Approach to Combining Enzymatic and Transition Metal Catalysis. *Nat. Chem*. **2013**, *5*, 100–103.

(192) Odachowski, M.; Greaney, M. F.; Turner, N. J. Concurrent Biocatalytic Oxidation and C−C Bond Formation via Gold Catalysis: One-Pot Alkynylation of *N*-Alkyl Tetrahydroisoquinolines. *ACS Catal.* **2018**, *8*, 10032–10035.

(193) Cosgrove, S. C.; Thompson, M. P.; Ahmed, S. T.; Parmeggiani, F.; Turner, N. J. One-Pot Synthesis of Chiral *N*-Arylamines by Combining Biocatalytic Aminations with Buchwald–Hartwig *N*-Arylation. *Angew. Chem. Int. Ed.* **2020**, *59*, 18156–18160.

(194) Heckmann, C. M.; Paradisi, F. GPhos Ligand Enables Production of Chiral *N*-Arylamines in a Telescoped Transaminase-Buchwald-Hartwig Amination Cascade in the Presence of Excess Amine Donor. *Chem. Eur. J.* **2021**, *27*, 16616–16620.

(195) Zhang, P.; Liao, X.; Ma, C.; Li, Q.; Li, A.; He, Y. Chemoenzymatic Conversion of Corncob to Furfurylamine via Tandem Catalysis with Tin-Based Solid Acid and Transaminase Biocatalyst. *ACS Sustain. Chem. Eng.* **2019**, *7*, 17636–17642.

(196) Di, J.-H.; Gong, L.; Yang, D.; He, Y.-C.; Tang, Z. Y.; Ma, C.-L. Enhanced Conversion of Biomass to Furfurylamine with High Productivity by Tandem Catalysis with Sulfonated Perlite and ω-Transaminase Whole-Cell Biocatalyst. *J. Biotechnol.* **2021**, *334*, 26–34.

(197) He, Y.; Ding, Y.; Ma, C.; Di, J.; Jiang, C.; Li, A. One-Pot Conversion of Biomass-Derived Xylose to Furfuralcohol by a Chemo-Enzymatic Sequential Acid-Catalyzed Dehydration and Bioreduction. *Green Chem.* **2017**, *19*, 3844–3850.

(198) Palomo, J. M.; Filice, M. New Emerging Bio-Catalysts Design in Biotransformations. *Biotechnol. Adv.* **2015**, *33*, 605–613.

(199) Heuson, E.; Froidevaux, R.; Itabaiana, Jr., I.; Wojcieszak, R.; Capron, M.; Dumeignil, F. Optimisation of Catalysts Coupling in Multi-Catalytic Hybrid Materials: Perspectives for the Next Revolution in Catalysis. *Green Chem.* **2021**, *23*, 1942–1954.

(200) Davis, H. J.; Ward, T. R. Artificial Metalloenzymes: Challenges and Opportunities. *ACS Cent. Sci.* **2019**, *5*, 1120–1136.

(201) Reetz, M. T. Directed Evolution of Artificial Metalloenzymes: A Universal Means to Tune the Selectivity of Transition Metal Catalysts? *Acc. Chem. Res.* **2019**, *52*, 336–344.

(202) Matsuo, T.; Miyake, T.; Hirota, S. Recent Developments on Creation of Artificial Metalloenzymes. *Tetrahedron Lett.* **2019**, *60*, 151226.

(203) Alonso-Cotchico, L.; Rodríguez-Guerra, J.; Lledós, A.; Maréchal, J.-D. Molecular Modeling for Artificial Metalloenzyme Design and Optimization. *Acc. Chem. Res.* **2020**, *53*, 896–905.

(204) Jeschek, M.; Reuter, R.; Heinisch, T.; Trindler, C.; Klehr, J.; Panke, S.; Ward, T. R. Directed Evolution of Artificial Metalloenzymes for *in vivo* Metathesis. *Nature* **2016**, *537*, 661–665.

(205) Liang, A. D.; Serrano-Plana, J.; Peterson, R. L.; Ward, T. R. Artificial Metalloenzymes Based on the Biotin−Streptavidin Technology: Enzymatic Cascades and Directed Evolution. *Acc. Chem. Res.* **2019**, *52*, 585–595.

(206) Köhler, V.; Wilson, Y. M.; Dürrenberger, M.; Ghislieri, D.; Churakova, E.; Quinto, T.; Knörr, L.; Häussinger, D.; Hollmann, F.; Turner, N. J. *et al.* Synthetic Cascades Are Enabled by Combining Biocatalysts with Artificial Metalloenzymes. *Nat. Chem.* **2013**, *5*, 93–99.

(207) Xu, Y.; Chen, L.; Wang, X.; Yao, W.; Zhang, Q. Recent Advances in Noble Metal Based Composite Nanocatalysts: Colloidal Synthesis, Properties, and Catalytic Applications. *Nanoscale* **2015**, *7*, 10559–10583.

(208) Mashwani, Z.-u.-R.; Khan, T.; Khan, M. A.; Nadhman, A. Synthesis in Plants and Plant Extracts of Silver Nanoparticles with Potent Antimicrobial Properties: Current Status and Future Prospects. *Appl. Microbiol. Biotechnol.* **2015**, *99*, 9923–9934.

(209) Gonzalez-Moragas, L.; Maurer, L. L.; Harms, V. M.; Meyer, J. N.; Laromaine, A.; Roig, A. Materials and Toxicological Approaches to Study Metal and Metal-Oxide Nanoparticles in the Model Organism *Caenorhabditis elegans*. *Mater. Horiz.* **2017**, *4*, 719.

(210) Palomo, J. M. Nanobiohybrids: A New Concept for Metal Nanoparticles Synthesis. *Chem. Commun.* **2019**, *55*, 9583–9589.

(211) Filice, M.; Marciello, M.; Morales, M. P.; Palomo, J. M. Synthesis of Heterogeneous Enzyme–Metal Nanoparticle Biohybrids in Aqueous Media and Their Applications in C–C Bond Formation and Tandem Catalysis. *Chem. Commun*. **2013**, *49*, 6876–6878.

(212) Kisukuri, C. M.; Palmeira, D. J.; Rodrigues, T. S.; Camargo, P. H. C.; Andrade, L. H. Bimetallic Nanoshells as Platforms for Metallo- and Biometallo-Catalytic Applications. *ChemCatChem* **2016**, *8*, 171–179.

(213) Cuenca, T.; Filice, M.; Palomo, J. M. Palladium Nanoparticles Enzyme Aggregate (PANEA) as Efficient Catalyst for Suzuki–Miyaura Reaction in Aqueous Media. *Enzyme Microb. Technol.* **2016**, *95*, 242–247.

(214) Lopez-Tejedor, D.; de las Rivas, B.; Palomo, J. M. Ultra-Small Pd(0) Nanoparticles into a Designed Semisynthetic Lipase: An efficient and Recyclable Heterogeneous Biohybrid Catalyst for the Heck Reaction under Mild Conditions. *Molecules* **2018**, *23*, 2358.

(215) Palomo, J. M.; Filice, M. Biosynthesis of Metal Nanoparticles: Novel Efficient Heterogeneous Nanocatalysts. *Nanomat.* **2016**, *6*, 84.

(216) O'Brien, P.; Lopez-Tejedor, D.; Benavente, R.; Palomo, J. M. Pd Nanoparticles-Polyethylenemine-Lipase Bionanohybrids as Heterogeneous Catalysts for Selective Oxidation of Aromatic Alcohols. *ChemCatChem* **2018**, *10*, 4992–4999.

(217) San, B. H.; Ravichandran, S.; Park, K.-s.; Subramani, V. K.; Kim, K. K. Bioinorganic Nanohybrid Catalyst for Multistep Synthesis of Acetaminophen, an Analgesic. *ACS Appl. Mater. Interfaces* **2016**, *8*, 30058–30065.

(218) Gao, S.; Wang, Z.; Ma, L.; Liu, Y.; Gao, J.; Jiang, Y. Mesoporous Core–Shell Nanostructures Bridging Metal and Biocatalyst for Highly Efficient Cascade Reactions. *ACS Catal.* **2020**, *10*, 1375–1380.

(219) Wang, Y.; Zhang, N.; Zhang, E.; Han, Y.; Qi, Z.; Ansorge-Schumacher, M. B.; Ge, Y.; Wu, C. Heterogeneous Metal–Organic-Framework-Based Biohybrid Catalysts for Cascade Reactions in Organic Solvent. *Chem. Eur. J.* **2019**, *25*, 1716–1721.

(220) Zhang, N.; Hübner, R.; Wang, Y.; Zhang, E.; Zhou, Y.; Dong, S.; Wu, C. Surface-Functionalized Mesoporous Nanoparticles as Heterogeneous Supports to Transfer Bifunctional Catalysts into Organic Solvents for Tandem Catalysis. *ACS Appl. Nano Mater.* **2018**, *1*, 6378–6386.

(221) Wang, Y.; Zhang, N.; Hübner, R.; Tan, D.; Löffler, M.; Facsko, S.; Zhang, E.; Ge, Y.; Qi, Z.; Wu, C. Enzymes Immobilized on Carbon Nitride (C_3N_4) Cooperating with Metal Nanoparticles for Cascade Catalysis. *Adv. Mater. Interfaces* **2019**, *6*, 1801664.

(222) Naapuri, J. M.; Åberg, G. A.; Palomo, J. M.; Deska, J. Arylative Allenol Cyclization via Sequential One-pot Enzyme & Palladium Catalysis. *ChemCatChem* **2021**, *13*, 763–769.

(223) Engström, K.; Johnston, E. V.; Verho, O.; Gustafson, K. P. J.; Shakeri, M.; Tai, C.-W.; Bäckvall, J.-E. Co-Immobilization of an Enzyme and a Metal into the Compartments of Mesoporous Silica for Cooperative Tandem Catalysis: An Artificial Metalloenzyme. *Angew. Chem. Int. Ed.* **2013**, *52*, 14006–14010.

(224) de Souza, S. P.; Leão, R. A. C.; Bassut, J. F.; Leal, I. C. R.; Wang, S.; Ding, Q.; Li, Y.; Lam, F. L.-Y.; de Souza, R. O. M. A.; Itabaiana Jr, I. New Biosilified Pd-Lipase Hybrid Biocatalysts for Dynamic Resolution of Amines. *Tetrahedron Lett.* **2017**, *58*, 4849–4854.

(225) Zhang, X.; Jing, L.; Chang, F.; Chen, S.; Yang, H.; Yang, Q. Positional Immobilization of Pd Nanoparticles and Enzymes in Hierarchical Yolk–Shell@Shell Nanoreactors for Tandem Catalysis. *Chem. Commun.* **2017**, *53*, 7780–7783.

(226) Ferraz, C. A.; do Nascimento, M. A.; Almeida, R. F. O.; Sergio, G. G.; Junior, A. A. T.; Dalmônico, G.; Caraballo, R.; Finotelli, P. V.; Leão, R. A. C.; Wojcieszak, R. *et al.* Synthesis and Characterization of a Magnetic Hybrid Catalyst Containing Lipase and

Palladium and its Application on the Dynamic Kinetic Resolution of Amines. *Mol. Catal.* **2020**, *493*, 111106.

(227) Li, X.; Cao, Y.; Luo, K.; Sun, Y.; Xiong, J.; Wang, L.; Liu, Z.; Li, J.; Ma, J.; Ge, J. *et al.* Highly Active Enzyme–Metal Nanohybrids Synthesized in Protein–Polymer Conjugates. *Nat. Catal.* **2019**, *2*, 718–725.

(228) Gustafson, K. P. J.; Görbe, T.; de Gonzalo-Calvo, G.; Yuan, N.; Schreiber, C. L.; Shchukarev, A.; Tai, C.-W.; Persson, I.; Zou, X.; Bäckvall, J.-E. Chemoenzymatic Dynamic Kinetic Resolution of Primary Benzylic Amines using Pd^0-CalB CLEA as a Biohybrid Catalyst. *Chem. Eur. J.* **2019**, *25*, 9174–9179.

(229) Gao, L.; Wang, Z.; Liu, Y.; Liu, P.; Gao, S.; Gao, J.; Jiang, Y. Co-Immobilization of Metal and Enzyme into Hydrophobic Nanopores for Highly Improved Chemoenzymatic Asymmetric Synthesis. *Chem. Commun.* **2020**, *56*, 13547–13550.

(230) Gao, S.; Liu, Y.; Wang, L.; Wang, Z.; Liu, P.; Gao, J.; Jiang, Y. Incorporation of Metals and Enzymes with Porous Imine Molecule Cages for Highly Efficient Semiheterogeneous Chemoenzymatic Catalysis. *ACS Catal.* **2021**, *11*, 5544–5553.

(231) Deiana, L.; Rafi, A. A.; Naidu, V. R.; Tai, C.-W.; Bäckvall, J.-E.; Córdova, A. Artificial Plant Cell Walls as Multi-Catalyst Systems for Enzymatic Cooperative Asymmetric Catalysis in Non-Aqueous Media. *Chem. Commun.* **2021**, *57*, 8814–8817.

(232) Li, H.; Qiu, C.; Cao, X.; Lu, Y.; Li, G.; He, X.; Lu, Q.; Chen, K.; Ouyang, P.; Tan, W. Artificial Nanometalloenzymes for Cooperative Tandem Catalysis. *ACS Appl. Mater. Interfaces* **2019**, *11*, 15718–15726.

(233) Görbe, T.; Gustafson, K. P. J.; Verho, O.; Kervefors, G.; Zheng, H.; Zou, X.; Johnston, E. V.; Bäckvall, J.-E. Design of a Pd(0)-CalB CLEA Biohybrid Catalyst and Its Application in a One-Pot Cascade Reaction. *ACS Catal.* **2017**, *7*, 1601–1605.

(234) Magadum, D. B.; Yadav, G. D. Design of Tandem Catalyst by Co-Immobilization of Metal and Enzyme on Mesoporous Foam for Cascaded Synthesis of (*R*)-Phenyl Ethyl Acetate. *Biochem. Eng. J.* **2018**, *129*, 96–105.

(235) Dutta, S.; Kumari, N.; Dubbu, S.; Jang, S. W.; Kumar, A.; Ohtsu, H.; Kim, J.; Cho, S. H.; Kawano, M.; Lee, I. S. Highly Mesoporous Metal-Organic Frameworks as Synergistic Multimodal Catalytic Platforms for Divergent Cascade Reactions. *Angew. Chem. Int. Ed.* **2020**, *59*, 3416–3422.

(236) Gaßmeyer, S. K.; Wetzig, J.; Mügge, C.; Assmann, M.; Enoki, J.; Hilterhaus, L.; Zuhse, R.; Miyamoto, K.; Liese, A.; Kourist. R. Arylmalonate Decarboxylase-Catalyzed Asymmetric Synthesis of Both Enantiomers of Optically Pure Flurbiprofen. *ChemCatChem* **2016**, *8*, 916–921.

(237) Wedde, S.; Rommelmann, P.; Scherkus, C.; Schmidt, S.; Bornscheuer, U. T.; Liese, A.; Gröger, H. An Alternative Approach towards Poly-ε-caprolactone through a Chemoenzymatic Synthesis: Combined Hydrogenation, Bio-Oxidations and Polymerization without the Isolation of Intermediates. *Green Chem.* **2017**, *19*, 1286–1290.

(238) Cui, B.; Yang, M.; Shan, J.; Qin, L.; Liu, Z.; Wan, N.; Han, W.; Chen, Y. Chemoenzymatic Synthesis of β-Hydroxyl-sulfoxides by a Two-Step Reaction of Enzymatic Reduction using *Pseudomonas monteilii* Species and Sulfoxidation with Chiral Titanium Complexe. *Tetrahedron* **2017**, *73*, 5200–5206.

(239) Plass, C.; Hinzmann, A.; Terhorst, M.; Brauer, W.; Oike, K.; Yavuzer, H.; Asano, Y.; Vorholt, A. J.; Betke, T.; Gröger, H. Approaching Bulk Chemical Nitriles from Alkenes: A Hydrogen Cyanide-Free Approach through a Combination of Hydroformylation and Biocatalysis. *ACS Catal.* **2019**, *9*, 5198–5203.

(240) Enoki, J.; Linhorst, M.; Busch, F.; Gomez Baraibar, A.; Miyamoto, K.; Kourist, R.; Mügge, C. Preparation of Optically Pure Flurbiprofen via an Integrated Chemoenzymatic Synthesis Pathway. *Mol. Catal.* **2019**, *467*, 135–142.

(241) Blakemore, C. A.; France, S. P.; Samp, L.; Nason, D. M.; Yang, E.; Howard, R. M.; Coffman, K. J.; Yang, Q.; Smith, A. C.; Evrard, E. *et al.* Scalable, Telescoped Hydrogenolysis−Enzymatic Decarboxylation Process for the Asymmetric Synthesis of (*R*)-α-Heteroaryl Propionic Acids. *Org. Process Res. Dev.* **2021**, *25*, 421−426.

(242) Domínguez de María, P.; de Gonzalo, G.; Alcántara, A. R. Biocatalysis as Useful Tool in Asymmetric Synthesis: An Assessment of Recently Granted Patents (2014–2019). *Catalysts* **2019**, *9*, 802.

(243) Schmidt, S.; Castiglione, K.; Kourist, R. Overcoming the Incompatibility Challenge in Chemoenzymatic and Multi-Catalytic Cascade Reactions. *Chem. Eur. J.* **2018**, *24*, 1755–1768.

(244) Winkler, C. K.; Schrittwieser, J. H.; Kroutil, W. Power of Biocatalysis for Organic Synthesis. *ACS Cent. Sci.* **2021**, *7*, 55–71.

(245) Sheldon, R. A.; Brady, D. Streamlining Design, Engineering, and Applications of Enzymes for Sustainable Biocatalysis. *ACS Sustain. Chem. Eng.* **2021**, *9*, 8032–8052.

(246) Ascaso-Alegre, C.; Mangas-Sánchez, J. Construction of Chemoenzymatic Linear Cascades for the Synthesis of Chiral Compounds. *Eur. J. Org. Chem.* **2022**, e202200093.

(247) González-Granda, S.; Escot, L.; Lavandera, I.; Gotor-Fernández, V. Unmasking the Hidden Carbonyl Group Using Gold(I) Catalysts and Alcohol Dehydrogenases: Design of a Thermodynamically-Driven Cascade toward Optically Active Halohydrins. *ACS Catal.* **2022**, *12*, 2552–2560.

(248) Chang, F.; Wang, C.; Chen, Q.; Zhang, Y.; Liu, G. A Chemoenzymatic Cascade Combining a Hydration Catalyst with an Amine Dehydrogenase: Synthesis of Chiral Amines. *Angew. Chem. Int. Ed.* **2022**, *61*, e202114809.

(249) Adriaensen, K.; Vercammen, J.; Van Goethem, C.; Agrawal, K. V.; De Vos, D. In Depth Analysis of Heterogeneous Catalysts for the Chemoenzymatic Dynamic Kinetic Resolution of β-Amino Esters. *Catal. Sci. Technol.* **2022**, *12*, 2894–2902.

(250) Seide, S.; Arnold, L.; Wetzels, S.; Bregu, M.; Gätgens, J.; Pohl, M. From Enzyme to Preparative Cascade Reactions with Immobilized Enzymes: Tuning Fe(II)/α-Ketoglutarate-Dependent Lysine Hydroxylases for Application in Biotransformations. *Catalysts* **2022**, *12*, 354.

(251) López-Agudo, M.; Ríos-Lombardía, N.; González-Sabín, J.; Lavandera, I.; Gotor-Fernández, V. Chemoenzymatic Oxosulfonylation-Bioreduction Sequence for the Stereoselective Synthesis of β-Hydroxy Sulfones. *ChemSusChem* **2022**, *15*, e202101313.

(252) Gastaldi, C.; Mekhloufi, G.; Forano, C.; Gautier, A.; Guérard-Hélaine, C. Mixing Chemo- and Biocatalysis for Rare Monosaccharide Production by Combining Aldolase and N-Heterocyclic Carbene Gold Catalysts. *Green Chem.* **2022**, *24*, 3634–3639.

(253) Wood, A. B.; Kincaid, J. R. A.; Lipshutz, B. H. Dehydration of Primary Amides to Nitriles in Water. Late-Stage Functionalization and 1-Pot Multistep Chemoenzymatic Processes under Micellar Catalysis Conditions. *Green Chem.* **2022**, *24*, 2853–2858.

(254) Bering, L.; Craven, E. J.; Thomas, S. A. S.; Shepherd, S. A.; Micklefield, J. Merging Enzymes with Chemocatalysis for Amide Bond Synthesis. *Nat. Commun.* **2022**, *13*, 380.

(255) Horino, S.; Nishio, T.; Kawanishi, S.; Oki, S.; Nishihara, K.; Ikawa, T.; Kanomata, K.; Wagner, K.; Gröger, H.; Akai, S. Enantiodivergent Chemoenzymatic Dynamic Kinetic Resolution: Conversion of Racemic Propargyl Alcohols into Both Enantiomers. *Chem. Eur. J.* **2022**, *28*, e202202437.

(256) González-Granda, S.; Tzouras, N. V.; Nolan, S. P.; Lavandera, I.; Gotor-Fernández, V. Merging Gold(I) Catalysis with Amine Transaminases in Cascade Catalysis: Chemoenzymatic Transformation of Propargylic Alcohols into Enantioenriched Allylic Amines. *Adv. Synth. Catal.* **2022,** doi.org/10.1002/adsc.202200777.

(257) Naapuri, J. M.; Losada-Garcia, N.; Rothemann, R. A.; Pichardo, M. C.; Prechtl, M. H. G.; Palomo, J. M.; Deska J. Cascade Catalysis through Bifunctional Lipase Metal Biohybrids for the Synthesis of Enantioenriched O-Heterocycles from Allenes. *ChemCatChem* **2022**, *14*, e202200362.

Acknowledgments

Financial support from the Spanish Ministry of Science and Innovation (MCI, PID2019-109253RB-I00) and Asturian Regional Government (AYUD/2021/51542) are gratefully

acknowledged. S.G.-G. thanks University of Oviedo for a predoctoral fellowship (PAPI-21-PF-14) inside the “Programa de Apoyo y Promoción de la Investigación”.

Table of contents

Biographical sketches

Sergio González-Granda

Dr. Sergio González-Granda (1993) studied chemistry at the University of Oviedo where he graduated on July 2017. He recently completed his PhD studies under the supervision of Iván Lavandera and Vicente Gotor-Fernández working into the design of new cascade approaches combining gold(I) species and enzymes to synthesize high-added value compounds under mild reaction conditions. His main research interests include asymmetric catalysis, and merging biocatalysis with other methodologies such as metal- or photocatalysis to develop new synthetic routes and activation modes.

Jesús Albarrán Velo

Dr. Jesús Albarrán-Velo (1992) studied chemistry at the University of Extremadura, obtaining first his degree in 2014 and later a Green Chemistry master degree in 2015. He completed his PhD studies under the supervision of Prof. Iván Lavandera and Vicente Gotor-Fernández on 2021, studying the development of oxidative processes and their application in multicatalytic systems using oxidoreductases and transaminases, including the design of photobiocatalytic processes. Currently he is working as a research associate

at the company Eurofins Villapharma on the production of high added value organic compounds.

Iván Lavandera

Dr. Iván Lavandera graduated in Chemistry in 1998 at the University of Oviedo (Spain), where he completed his PhD studies in 2003 with Prof. Vicente Gotor and Prof. Miguel Ferrero. In 2005, he moved to the University of Graz as a postdoctoral researcher under the supervision of Prof. Wolfgang Kroutil. He returned to Oviedo in 2008, where he became first post-doctoral researcher, and then, since 2015, he is Associate Professor at the Organic and Inorganic Chemistry Department at the University of Oviedo. He has been co-author of two patents and more than 110 publications. In 2021 he edited with Prof. Gonzalo de Gonzalo the book "Biocatalysis for Practitioners". His main research interests are focused on Biocatalysis, especially the use of oxidoreductases and transferases to develop green processes, and also new synthetic routes combining bio- and chemocatalysis in a concurrent manner.

Vicente Gotor-Fernández

Dr. Vicente Gotor-Fernández studied his Degree in Chemistry at the University of Oviedo, specializing in Organic Chemistry (1997). He completed his PhD studies working in the chemoenzymatic synthesis of vitamin D_3 analogues at the Bioorganic Chemistry research group (2001). Then, he moved to the University of Edinburgh, joining the research group of Prof. Nicholas Turner with a Marie Curie postdoctoral contract, and after two years, he returned to Oviedo as postdoctoral researcher. In June 2012, he obtained a permanent position as Associate Professor. He is co-author of around 170 scientific contributions including research articles, reviews, chapter books and patents, mostly in the field of Biocatalysis and organic synthesis. His main current research interest involves the development of chemoenzymatic routes for the synthesis of organic compounds using hydrolases, oxidoreductases and transferases, especially focusing on the design of chemo- and multienzymatic concurrent processes in different media.