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Stereoselective amination of racemic *sec*-alcohols through sequential application of laccases and transaminases

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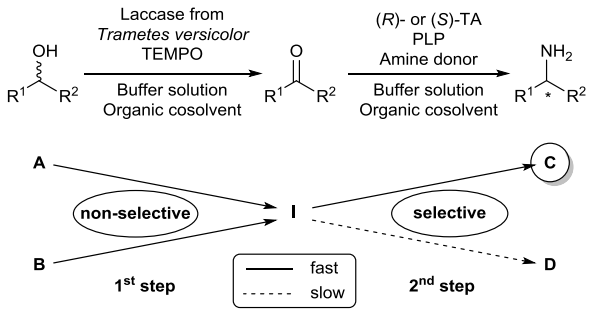
A one-pot/two-step bienzymatic asymmetric amination of secondary alcohols is disclosed. The approach is based on a sequential strategy involving the use of a laccase/TEMPO catalytic system for the oxidation of alcohols into ketone intermediates, and their following transformation into optically enriched amines by using transaminases. Individual optimizations of the oxidation and biotransamination reactions have been carried out, studying later their applicability in a concurrent process. Therefore, 17 racemic (hetero)aromatic *sec*-alcohols with different substitutions in the aromatic ring have been converted into enantioenriched amines with good to excellent selectivities (90-99% *ee*) and conversion values (67-99%). The scalability of the process was also demonstrated when two different amine donors were used in the transamination step, such as isopropylamine and *cis*-2-buten-1,4-diamine. Satisfyingly, both sacrificial amine donors can shift the equilibrium toward the amine formation, leading to the corresponding isolated enantioenriched amines with good to excellent results.

Introduction

Optically active amines are valuable building blocks in the synthesis of pharmaceuticals and agrochemicals, also possessing multiple applications as resolving agents, chiral auxiliaries, and organocatalytic reagents.[1] Nowadays, biocatalytic methods provide versatile strategies for the stereoselective synthesis of chiral amines by using a wide number of enzymes such as lipases, amino oxidases, imine reductases and transaminases (TAs), among others.[2-5] In this context, the use of enzymes for industrial applications has been boosted due to the requirements established by national regulatory agencies, and the continuous efforts for the development of chemical transformations under safer and milder reaction conditions.[6,7] As an excellent example, it can be mentioned a TA-catalyzed stereoselective amination that constitutes the key step in the synthesis of the antidiabetic drug Sitagliptin.[8]

A current trend in biotransformations is the development of concurrent catalytic strategies, which can be performed in a cascade or sequential mode.[9-14] These strategies allow to minimize the number of operational steps and reaction vessels used, improving the 'pot-economy'[15] of the processes. These systems have recently provided access to enantiopure amines by the proper employment of chemoenzymatic or multienzymatic systems.[16] Remarkably, the selective amination of alcohols[17] has been described through elegant cascades by combining an alcohol dehydrogenase (ADH)-catalyzed oxidation of racemic alcohols into ketones, and a subsequent biotransamination using TAs[18,19] or amine dehydrogenases (AmDHs).[20,21] However, in the previously cited strategies, due to the excellent selectivities commonly shown by ADHs when a racemic alcohol is used as starting material, two enzymes with opposite stereopreferences are usually compulsory. Herein, we propose a one-pot chemoenzymatic approach involving the use of an alternative redox system, the laccase from *Trametes versicolor*/TEMPO, for the non-selective oxidation of racemic alcohols. Its action will be later combined with a stereoselective TA-catalyzed transformation over the corresponding ketone intermediates (Scheme 1).



Scheme 1 Synthesis of optically active amines from racemic alcohols through a one-pot sequential bienzymatic system using the *Trametes versicolor* laccase and a transaminase. A and B are the substrate enantiomers; I is the ketone intermediate; C and D are the product enantiomers.

Laccases are blue multicopper oxidases able to catalyze the oxidation of low molecular weight phenols as natural substrates at the expense of the environmentally friendly four-electron reduction of oxygen into water.[22-25] Multiple applications have been found for these redox enzymes in industrial sectors,[26,27] possessing attractive possibilities applied to synthetic chemistry.[28-30] In order to carry out effective oxidations of primary and secondary alcohols, the enzyme requires the use of a chemical mediator. Particularly, the 2,2,6,6-tetramethylpiperidinoxyl radical (TEMPO) is one of the most recurrent reagents for efficient protocols.[31-33]

On the other hand, transaminases belonging to the transferase class, are pyridoxal 5'-phosphate (PLP)-dependent enzymes. These biocatalysts have received increasing attention in the last decade due to their possibilities in the synthesis of optically active amines.[34-39] A key issue for this transformation is the use of sacrificial amines as donors (i.e. isopropylamine), normally employed in a high molar excess for shifting the equilibrium toward the amine synthesis.[40]

Results and Discussion

The synthesis of enantiopure amines has been envisaged from inexpensive racemic alcohols using the *Trametes versicolor* laccase/TEMPO system and different commercially available transaminases. The possibility to develop a concurrent one-pot process is here discussed searching for compatible enzymatic reaction conditions. A panel of 17 racemic (hetero)aromatic alcohols was selected in order to design a general and stereoselective strategy toward the synthesis of interesting amines (Fig. 1).

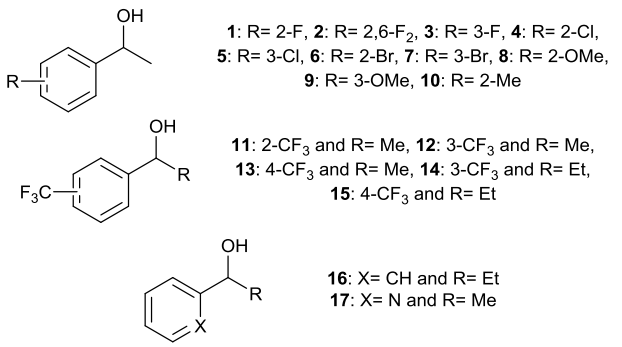
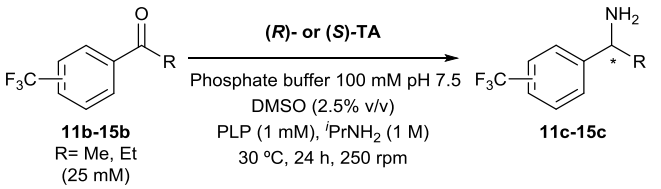


Fig. 1 Panel of substrates under study.

Preliminary optimizations of the reaction conditions were performed for both laccase- and transaminase-catalyzed reactions. The catalytic system composed by the laccase from *Trametes versicolor* and TEMPO (33 mol%) as chemical mediator was studied in citrate buffer 50 mM at pH 5 and 30 °C, adapting previously optimized conditions for the oxidation of benzylic alcohols.[33,41] The stirring mode and the use of different organic cosolvents were analyzed (see Tables S1 and S2 in the ESI), finding a quantitative conversion for the oxidation of 1-(2-fluorophenyl)ethanol (**1a**) after 16 h using magnetic stirring and 50% v/v of methyl *tert*-butyl ether (MTBE). Furthermore, attempting to scale the process under more sustainable conditions, lower amounts of TEMPO were studied. Gladly, when TEMPO was used in 20 and 33 mol%, complete oxidation of 1-(2-bromophenyl)ethanol (**6a**) was achieved in both cases after 16 h. On the contrary, a decrease in the conversion was observed for other substrates when employing 20 mol% of TEMPO. For instance, the oxidation of the racemic alcohols 1-(2-chlorophenyl)ethanol (**4a**) and 1-(3-methoxyphenyl)ethanol (**9a**), two substrates that had been quantitatively transformed into the corresponding ketones when a 33 mol% had been used, led to lower conversion values (66% for **4b** and 83% for **9b**, respectively) when the loading of TEMPO was reduced to 20 mol%.

For the biotransamination step, while there were already reports in the literature for some of the ketones tested,[8,42-47] additional transaminase activity screenings were performed for the methoxy derivatives **8b** and **9b** (Tables S3 and S4 in the ESI, respectively) and for the trifluoromethylated ketones **11b-15b** (Table 1 and Tables S5-S9 in the ESI). In this study commercially available TAs were used, although TAs with well-known sequences could also be employed for this reaction. These substrates were selected due to the relevance of the synthesized amines. Hence, amine (*S*)-**9c** is a precursor of Rivastigmine, a drug used in the treatment of dementia diseases,[44,48] and organofluorinated compounds present a broad applicability in medicinal chemistry.[49-51]

Table 1 Biotransamination of selected trifluoromethylated ketones.[a]



Entry	Substrate	Transaminase	*c* (%)[b]	*ee* (%)[c]
1	2-CF_3 and R= Me (**11b**)	ATA-024	91	>99 (*R*)
2		ATA-113	95	>99 (*S*)
3	3-CF_3 and R= Me (**12b**)	ATA-033	77	96 (*R*)
4		ATA-251	71	>99 (*S*)
5	4-CF_3 and R= Me (**13b**)	ATA-025	86	>99 (*R*)
6		ATA-251	86	>99 (*S*)
7	3-CF_3 and R= Et (**14b**)	ATA-415	71	>99 (*R*)
8		ATA-237	67	>99 (*S*)
9	4-CF_3 and R= Et (**15b**)	ATA-025	78	>99 (*R*)
10		ATA-237	74	>99 (*S*)

[a] For reaction conditions, see the ESI. [b] Conversion values were determined by GC analysis. [c] Enantiomeric excess values were determined by chiral GC or HPLC analyses from acetamide derivatives of amines **11c-15c**, otherwise indicated in the ESI.

All reactions were carried out at 30 °C in phosphate buffer 100 mM pH 7.5, using 2.5% v/v of DMSO as cosolvent to enhance the ketone solubility. Satisfyingly, both (*R*)- and (*S*)-amine enantiomers were isolated in optically pure form excepting 1-[(3-trifluoromethyl)phenyl]ethanamine (**12c**), although the (*R*)-enantiomer was obtained in an also remarkable 96% *ee* (entry 3). Methyl ketones **11b**-**13b** displayed a higher reactivity than the ethyl ketones **14b** and **15b**. Particularly the *ortho*-substituted **11b** was transformed with the best conversions (91 and 95%). The highest reactivity shown by the *ortho*-substituted derivatives was

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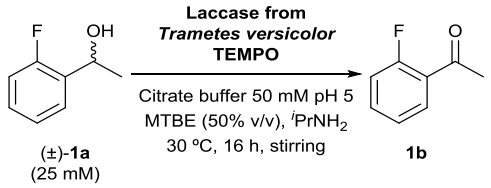
in agreement with a previous study which uses this transaminase kit and structurally similar substrates.[46]

Aiming to develop a concurrent one-pot methodology, once both steps of the bienzymatic approach were individually explored, the oxidative transformation was attempted in the presence of the amine donor (isopropylamine, $^{i}PrNH_2$). As a positive feedback would allow the possibility of carrying out the amination of alcohols in a cascade manner, the substrate **1a** was oxidized in the presence of different amounts of $^{i}PrNH_2$. Its concentration varied from a huge excess (1 M), which is usually required for shifting the equilibrium toward the amine synthesis, to a 50 mM concentration (Table 2). As a consequence of the loss of the activity shown by the laccase/TEMPO system in the presence of the amine donor, we deduced that $^{i}PrNH_2$ should be added once the oxidation step had concluded. Therefore, we envisioned the development of a one-pot/two-step sequential approach to achieve the stereoselective synthesis of amines.

Table 2 Oxidation of substrate **1a** catalyzed by the laccase/TEMPO system in the presence of isopropylamine.[a]



Entry	[$^{i}PrNH_2$] (mM)	**alcohol** (%)[b]	**ketone** (%)[b]
1	1000	>99	<1
2	500	>99	<1
3	200	98	2
4	100	98	2
5	75	98	2
6	50	94	6

[a] For reaction conditions, see the ESI. [b] Percentages of products measured by GC.

Hence, the asymmetric amination of racemic alcohols **1a**-**17a** was studied. Firstly, the catalytic system composed by the laccase from *Trametes versicolor* and TEMPO (33 mol%) was used for the oxidation step, and subsequently, the transaminases that showed better activity and selectivity values for the formation of the amines were utilized for the asymmetric biotransaminations. It is noteworthy that during the oxidation step at 50 mM substrate concentration,[52] complete evaporation of MTBE was observed.

One of the main drawbacks of the combined use of these enzymes is the different pHs in which these biocatalysts are active. Meanwhile laccases reach their best values at acidic pHs (between 4.5 and 5.5), TAs display their best activities at neutral or basic pH values (7-10). This limitation was overcome by pH adjustment through addition of phosphate buffer 200 mM pH 9 to the reaction medium. This addition was made to obtain a final 25 mM substrate concentration, optimal conditions for the transaminase-catalyzed reactions.

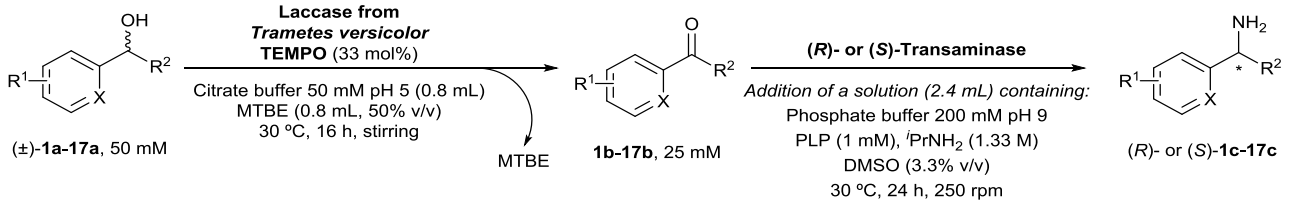
The best results for the sequential strategy are shown in Table 3. To our delight, in all cases the oxidation step proceeded with complete conversion, yielding the ketones **1b**-**17b** that were later subjected to amination experiments in a sequential mode. Thus, for 9 out of 17 substrates, transaminases were found to selectively produce both enantiomers of the corresponding amines in more than 90% *ee* (entries 1-8, 10, 11, 13, 14, 19, 20, 22, 23, 25 and 26). For the other 8 alcohols, at least one amine enantiomer was produced in optically pure form and more than 70% conversion (entries 9, 12, 15-18, 21 and 24).

The methyl ketones bearing substitutions at *ortho*-position led to the best conversion values (87-99%), while the *meta*-substituted ones provided moderate to high conversions (71-82%). Comparing the reactivity of *meta*-trifluoromethylated **12b** and *para*-substituted **13b**, a better conversion was observed for the latter, although a slight decrease of selectivity was also detected (entries 18-20). Otherwise, excellent *ee* values were observed when considering the homologue ethyl ketones **14b** and **15b**, or the non-substituted aromatic compound **16b** (entries 21-24). Remarkably, both enantiomers of 1-(pyridin-2-yl)ethanamine (**17c**) were obtained in almost optically pure form and with very high conversions (entries 25 and 26).

To demonstrate the applicability of this sequential strategy, some selected examples were scaled-up using 100 mg of the corresponding alcohol. To this purpose, we selected two racemic alcohols based on different reasons. On the one hand, chlorinated alcohol **4a** was considered due to the excellent results obtained at smaller scale (entry 7, Table 3). Satisfyingly, the reaction under identical oxidative conditions and using ATA-025 in the second step led to 99% conversion. The enantiopure amine (*R*)-**4c** was recovered in 93% isolated yield after a simple extraction protocol. On the other hand, we focused on **9a** (entry 15, Table 3) since amine **9c** can serve as a valuable precursor for (*S*)-Rivastigmine.[44,48] Under similar conditions, the reaction scaled-up with ATA-254 led to the desired (*S*)-**9c** in 98% *ee* and 67% conversion. The corresponding enantioenriched amine was isolated in 62% yield. It must be mentioned that due to its high boiling point, the organic cosolvent DMSO was replaced by acetonitrile (MeCN) in the scale-up biotransaminations in order to facilitate the isolation of the final products.

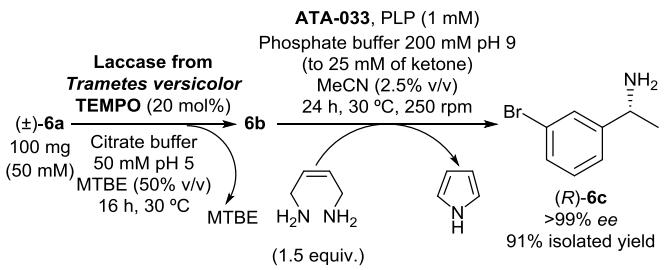
Recently, we have demonstrated the versatility of sacrificial diamines for driving the equilibrium toward amine synthesis when using them at almost stoichiometric amounts.[53] This is the case of *cis*-but-2-ene-1,4-diamine which presents additional advantages. For instance, the compounds of interest are easily isolated through simple extraction protocols since, on the one side, the diamine is completely soluble in water media, and on the other side, the insoluble polypyrrole is formed as by-product. Therefore, 100 mg of brominated alcohol **6a** reacted with the laccase from *Trametes versicolor* and only 20 mol% of TEMPO, obtaining quantitatively the ketone intermediate **6b**. Afterwards, it was converted into the enantiopure amine (*R*)-**6c** using ATA-033 and *cis*-but-2-ene-1,4-diamine (1.5 equiv.) in 97% conversion and 91% isolated yield after extraction (Scheme 2).

Table 3 Sequential bienzymatic transformation of racemic alcohols into optically active amines using a laccase and a transaminase.[a]



Entry	Substrate	R^1	R^2	X	TA	ratio (%)[b] Alcohol **1a-17a**	Ketone **1b-17b**	Amine **1c-17c**	*ee* (%)[c] **1c-17c**
1	**1a**	2-F	Me	CH	ATA-033	<1	<1	>99	>99 (*R*)
2					ATA-P1-A06	<1	<1	>99	>99 (*S*)
3	**2a**	2,6-F_2	Me	CH	ATA-013	<1	<1	>99	>99 (*R*)
4					ATA-P1-F03	<1	<1	>99	>99 (*S*)
5	**3a**	3-F	Me	CH	ATA-033	<1	21	79	>99 (*R*)
6					ATA-P1-G06	<1	23	77	91 (*S*)
7	**4a**	2-Cl	Me	CH	ATA-025	<1	<1	>99	>99 (*R*)
8					ATA-P1-A06	<1	<1	>99	>99 (*S*)
9	**5a**	3-Cl	Me	CH	ATA-033	<1	18	82	>99 (*R*)
10	**6a**	2-Br	Me	CH	ATA-025	<1	<1	>99	>99 (*R*)
11					ATA-P1-A06	<1	<1	>99	>99 (*S*)
12	**7a**	3-Br	Me	CH	ATA-025	<1	22	78	>99 (*R*)
13	**8a**	2-OMe	Me	CH	ATA-033	<1	5	95	97 (*R*)
14					ATA-256	<1	13	87	>99 (*S*)
15	**9a**	3-OMe	Me	CH	ATA-254	<1	26	74	>99 (*S*)
16	**10a**	2-Me	Me	CH	ATA-024	<1	10	90	>99 (*R*)
17	**11a**	2-CF_3	Me	CH	ATA-024	<1	4	96	>99 (*R*)
18	**12a**	3-CF_3	Me	CH	ATA-251	<1	29	71	>99 (*S*)
19	**13a**	4-CF_3	Me	CH	ATA-025	<1	27	73	96 (*R*)
20					ATA-251	<1	14	86	90 (*S*)
21	**14a**	3-CF_3	Et	CH	ATA-237	<1	24	76	>99 (*S*)
22	**15a**	4-CF_3	Et	CH	ATA-025	<1	25	75	>99 (*R*)
23					ATA-237	<1	33	67	>99 (*S*)
24	**16a**	H	Et	CH	ATA-025	<1	28	72	>99 (*R*)
25	**17a**	H	Me	N	ATA-033	<1	4	96	>99 (*R*)
26					ATA-P1-A06	<1	2	98	98 (*S*)

[a] For reaction conditions, see the ESI. [b] Ratios of products were determined by GC analysis of an aliquot after the two enzymatic steps.
[c] Enantiomeric excess values were determined by chiral GC or HPLC analyses from acetamide derivatives of amines **1c-17c**, otherwise indicated in the ESI.



Scheme 2 Preparative synthesis of (*R*)-**6c** using the *Trametes versicolor* laccase, TEMPO (20 mol%), ATA-033 and 1.5 equiv. of *cis*-but-2-ene-1,4-diamine as sacrificial amine donor.

At this point, a simple quantification of the environmental impact of this system was calculated using the *E*-factor concept.[54,55] Thus, this parameter was calculated employing the EATOS tool[56] and it was compared to the values obtained for two similar previous strategies, which combined the use of an ADH and a TA,[18] and of an ADH and an AmDH,[20] respectively (see ESI for more details). Particularly, we focused on the impact of the reaction conditions regarding the reagents, catalysts and media employed, as the work-up for the three protocols (a liquid-liquid extraction) is identical for all of them. For the reaction shown in Scheme 2, an *E*-factor of 8.3 was obtained (excluding solvents),[57] while values of 21 and 57 were respectively attained for the others. These results can be ascribed to the lower conversions achieved with the ADH/TA method, and to the high concentration of ammonium formate required for the ADH/AmDH system. As a result, these data demonstrate the favorable ecological impact of the laccase/TEMPO-TA amination protocol.

Conclusions

A practical one-pot/two-step sequential strategy has been disclosed for the synthesis of enantioenriched amines starting from racemic alcohols under very mild reaction conditions. A laccase-transaminase bienzymatic system has been used through the formation of stable prochiral ketone intermediates, which are later stereoselectively transformed into both amine enantiomers

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depending on the transaminase used. After studying different reaction conditions, the addition of isopropylamine after the oxidative process was found to be the key to success in the effective synthesis of enantioenriched amines. Additionally, scalable processes at 100 mg substrate scale were performed for different derivatives. Satisfyingly, the applicability of the sequential methodology has been proved when a high molar excess of isopropylamine, or alternatively, only 1.5 equiv of *cis*-but-2-ene-1,4-diamine were used as sacrificial amine donors. Particularly, this last example significantly improves the atom efficiency of the process. Overall, this sequential strategy is a robust example of a multienzymatic process as synthetic tool. Thus, transformations that can be challenging under mild and safe reaction conditions by other chemical means, have been carried out in an efficient and environmentally friendly manner.

Experimental

General

Laccase from *Trametes versicolor* (0.9 U/mg) was purchased from Sigma Aldrich. Codex Transaminase ATA Screening Kit (ATASK-000250) and PLP were purchased from Codexis. All other reagents were obtained from commercial sources and used as received.

Sequential reactions were performed in a single sealed tube [(19 x 130 x 3) mm], otherwise indicated. Oxidation step mediated by laccase/TEMPO catalytic system was performed open-to-air using magnetic stirring; while for the transamination step, the sealed tube was closed and orbital shaking (250 rpm) was used.

NMR spectra were recorded on a Bruker AV300 MHz spectrometer. All chemical shifts (δ) are given in parts per million (ppm) and referenced to the residual solvent signal as internal standard. Gas chromatography (GC) analyses were performed on an Agilent HP7820 GC chromatograph equipped with a FID detector. High performance liquid chromatography (HPLC) analyses were carried out in a Hewlett Packard 1100 chromatograph UV detector at 210 nm. Thin-layer chromatography (TLC) was conducted with Merck Silica Gel 60 F254 precoated plates and visualized with UV and potassium permanganate stain. Column chromatography was performed using Merck Silica Gel 60 (230-400 mesh).

General procedure for the sequential one-pot/two-step synthesis of enantiopure amines from racemic alcohols

The enantiopure amines **1c**-**17c** were obtained in a one-pot process according to the following procedure after two sequential reactions involving a laccase/TEMPO oxidation in a first step, and a transaminase-catalyzed reaction in a second one, leading to the desired optically active amines (*S*)- or (*R*)-**1c**-**17c** depending on the transaminase selectivity. In an open-to-air sealed tube, TEMPO (4.1 mg, 33 mol%) was added to a solution of the racemic alcohol **1a**-**17a** (0.08 mmol, 50 mM) in a biphasic mixture of oxygen-saturated citrate buffer 50 mM pH 5 and MTBE (50% v/v, for a total volume of 1.6 mL). The reaction mixture was stirred for a few minutes to dissolve all the reagents, and then the laccase from *Trametes versicolor* (5 U) was added. The reaction was stirred for 16 h at 30 °C, observing the complete evaporation of MTBE along this time. This fact led to a volume reduction from the initial 1.6 mL to 0.8 mL, and in consequence, the substrate concentration was increased from the initial 50 mM to approximately 100 mM. To the resulting reaction crude, phosphate buffer 200 mM pH 9 (2.4 mL) containing isopropylamine (1.33 M), PLP (1 mM) and DMSO (3.3% v/v) was added, leading to approximately 25 mM, 1 M and 2.5% v/v as substrate, isopropylamine and DMSO final concentrations, respectively. At the same time, the addition of this concentrated buffer to the reaction media, caused an increase in the pH from an initial value of 5 to approximately 7.5, therefore, further pH adjustment was not required. Finally, the corresponding commercially available transaminase (12 mg) was added. The sealed tube was closed and the reaction was shaken at 30 °C and 250 rpm for 24 h. After this time, the reaction was stopped by addition of an aqueous NaOH 10 M solution (3 mL). Then, the mixture was extracted with EtOAc (5 mL) and the organic layer was separated by centrifugation (3 min, 4,900 rpm). This centrifugation protocol was performed twice and, finally, the organic layers were combined and dried over Na_2SO_4. Conversion values into the corresponding amines **1c**-**17c** and their enantiomeric excess measurements were determined by GC analysis (Table 3, see the ESI for further details).

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Notes and references

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Electronic Supplementary Information (ESI) available: [experimental procedures, extensive enzymatic screenings for selected substrates, characterization for new compounds, analytical data, and NMR spectra are described]. See DOI: 10.1039/c000000x/

1 *Chiral Amine Synthesis: Methods, Developments and Applications*, Ed. T. C. Nugent, Wiley-VCH: Weinheim, 2010.
2 M. Höhne and U. T. Bornscheuer, *ChemCatChem*, 2009, **1**, 42-51.
3 W. Kroutil, E.-M. Fischereder, C. S. Fuchs, H. Lechner, F. G. Mutti, D. Pressnitz, A. Rajagopalan, J. H. Sattler, R. C. Simon and E. Siirola, *Org. Process Res. Dev.*, 2013, **17**, 751-759.
4 D. Ghislieri and N. J. Turner, *Top. Catal.*, 2014, **57**, 284-300.
5 H. Kohls, F. Steffen-Munsberg and M. Höhne, *Curr. Opin. Chem. Biol.*, 2014, **19**, 180-192.
6. J.-M. Choi, S.-S. Han and H.-S. Kim, *Biotechnol. Adv.*, 2015, **33**, 1443-1454.
7. G. Torrelo, U. Hanefeld and F. Hollmann, *Catal. Lett.*, 2015, **145**, 309-345.
8 C. K. Savile, J. M. Janey, E. C. Mundorff, J. C. Moore, S. Tam, W. R. Jarvis, J. C. Colbeck, A. Krebber, F. J. Fleitz, J. Brands, P. N. Devine, G. W. Huisman and G. J. Hughes, *Science*, 2010, **329**, 305-309.
9 C. A. Denard, J. F. Hartwig and H. Zhao, *ACS Catal.*, 2013, **3**, 2856-2864.

10 I. Oroz-Guinea and E. García-Junceda, *Curr. Opin. Chem. Biol.*, 2013, **17**, 236-249.
11 E. García-Junceda, I. Lavandera, D. Rother and J. H. Schrittwieser, *J. Mol. Catal B: Enzym.*, 2015, **114**, 1-6.
12 K. B. Otte and B. Hauer, *Curr. Opin. Biotechnol.*, 2015, **35**, 16-22.
13 V. Köhler and N. J. Turner, *Chem. Commun.*, 2015, **51**, 450-464.
14 J. Muschiol, C. Peters, N. Oberleitner, M. D. Mihovilovic, U. T. Bornscheuer and F. Rudroff, *Chem. Commun.*, 2015, **51**, 5798-5811.
15 Y. Hayashi, *Chem. Sci.*, 2016, **7**, 866-880.
16 R. C. Simon, N. Richter, E. Busto and W. Kroutil, *ACS Catal.*, 2014, **4**, 129-143.
17 There are other examples of non-selective biocatalytic amination of primary alcohols combining alcohol dehydrogenases and TAs: J. H. Sattler, M. Fuchs, K. Tauber, F. G. Mutti, K. Faber, J. Pfeffer, T. Haas and W. Kroutil, *Angew. Chem. Int. Ed.*, 2012, **51**, 9156-9159; or alcohol oxidases and TAs: M. Fuchs, K. Tauber, J. Sattler, H. Lechner, J. Pfeffer, W. Kroutil and K. Faber, *RSC Adv.*, 2012, **2**, 6262-6265; M. Pickl, M. Fuchs, S. M. Glueck and K. Faber, *ChemCatChem*, 2015, **7**, 3121-3124.
18 K. Tauber, M. Fuchs, J. H. Sattler, J. Pitzer, D. Pressnitz, D. Koszelewski, K. Faber, J. Pfeffer, T. Haas and W. Kroutil, *Chem. Eur. J.*, 2013, **19**, 4030-4035.
19 A. Lerchner, S. Achatz, C. Rausch, T. Haas and A. Skerra, *ChemCatChem*, 2013, **5**, 3374-3383.
20 F. G. Mutti, T. Knaus, N. S. Scrutton, M. Breuer and N. J. Turner, *Science*, 2015, **349**, 1525-1529.
21 F.-F. Chen, Y.-Y. Liu, G.-W. Zheng and J.-H. Xu, *ChemCatChem*, 2015, **7**, 3838-3841.
22 E. I. Solomon, U. M. Sundaram and T. E. Machonkin, *Chem. Rev.*, 1996, **96**, 2563-2605.
23 H. Claus, *Arch. Microbiol.*, 2003, **179**, 145-150.
24 S. G. Burton, *Curr. Org. Chem.*, 2003, **7**, 1317-1331.
25 S. Riva, *Trends Biotechnol.*, 2006, **24**, 219-226.
26 J.-R. Jeon and Y.-S. Chang, *Trends Biotechnol.*, 2013, **31**, 335-341.
27 C. Pezzella, L. Guarino and A. Piscitelli, *Cell. Mol. Life Sci.*, 2015, **72**, 923-940.
28 S. Witayakran and A. J. Ragauskas, *Adv. Synth. Catal.*, 2009, **351**, 1187-1209.
29 M. Mogharabi and M. A. Faramarzi, *Adv. Synth. Catal.*, 2014, **356**, 897-927.
30 I. Gonçalves, C. Silva and A. Cavaco-Paulo, *Green Chem.*, 2015, **17**, 1362-1374.
31 O. G. Mancheño and T. Stopka, *Synthesis*, 2013, **45**, 1602-1611.
32 N. E. Leadbeater and E. J. M. Bobbitt, *Aldrichim. Acta*, 2014, **47**, 65-74.
33 K. Kędziora, A. Díaz-Rodríguez, I. Lavandera, V. Gotor-Fernández and V. Gotor, *Green Chem.* 2014, **16**, 2448-2453.
34 D. Koszelewski, K. Tauber, K. Faber and W. Kroutil, *Trends Biotechnol.*, 2010, **28**, 324-332.
35 H. C. Hailes, P. A. Dalby, G. J. Lye, F. Baganz, M. Micheletti, N. Szita and J. M. Ward, *Curr. Org. Chem.*, 2010, **14**, 1883-1893.
36 S. Mathew and H. Yun, *ACS Catal.*, 2012, **2**, 993-1001.
37 M. S. Malik, E.-S. Park and J.-S. Shin, *Appl. Microbiol. Biotechnol.*, 2012, **94**, 1163-1171.
38 M. Fuchs, J. E. Farnberger and W. Kroutil, *Eur. J. Org. Chem.*, 2015, 6965-6982.
39 R. C. Simon, E. Busto, E.-M. Fischereder, C. S. Fuchs, D. Pressnitz, N. Richter and W. Kroutil, *Science of Synthesis, Biocatalysis in Organic Synthesis*, Eds. K. Faber, W.-D. Fessner and N. J. Turner, Georg Thieme Verlag, Stuttgart, 2015, pp. 383-420.
40 K. E. Cassimjee, C. Branneby, V. Abedi, A. Wells and P. Berglund, *Chem. Commun.*, 2010, **46**, 5569-5571.
41 A. Díaz-Rodríguez, L. Martínez-Montero, I. Lavandera, V. Gotor and V. Gotor-Fernández, *Adv. Synth. Catal.*, 2014, **356**, 2321-2329.
42 A. Iwasaki, Y. Yamada, N. Kizaki, Y. Ikenaka and J. Hasegawa, *Appl. Microbiol. Biotechnol.*, 2006, **69**, 499-505. (amine **9c**)
43 D. Koszelewski, I. Lavandera, D. Clay, D. Rozzell and W. Kroutil, *Adv. Synth. Catal.*, 2008, **350**, 2761-2766. (amine **16c**)
44 M. Fuchs, D. Koszelewski, K. Tauber, W. Kroutil and K. Faber, *Chem. Commun.*, 2010, **46**, 5500-5502. (amines **8c** and **9c**)
45 M. D. Truppo, H. Strotman and G. Hughes, *ChemCatChem*, 2012, **4**, 1071-1074. (amine **16c**)
46 C. E. Paul, M. Rodríguez-Mata, E. Busto, I. Lavandera, V. Gotor-Fernández, V. Gotor, S. García-Cerrada, J. Mendiola, O. de Frutos and I. Collado, *Org. Process Res. Dev.*, 2014, **18**, 788-792. (amines **1c**-**7c** and **17c**)
47 N. Ríos-Lombardía, C. Vidal, M. Cocina, F. Morís, J. García-Álvarez and J. González-Sabín, *Chem. Commun.*, 2015, **51**, 10937-10940. (amine **16c**)
48 J. Mangas-Sánchez, M. Rodríguez-Mata, E. Busto, V. Gotor-Fernández and V. Gotor, *J. Org. Chem.*, 2009, **74**, 5304-5310.
49 V. M. Muzalevskiy, A. V. Shastin, E. S. Balenkova, G. Haufe and V. G. Nenajdenko, *Synthesis*, 2009, 3905-3929.
50 F.-L. Qing and F. Zheng, *Synlett*, 2011, 1052-1072.
51 F. Meyer, *Chem. Commun.*, 2016, **52**, 3077-3094.
52 From previous studies in our research group, the laccase/TEMPO system can perfectly oxidize secondary alcohols at 25-50 mM concentrations.
53 L. Martínez-Montero, V. Gotor, V. Gotor-Fernández and I. Lavandera, *Adv. Synth. Catal.*, 2016, **358**, 1618-1624.
54 R. A. Sheldon, *Chem. Ind.*, 1992, 903-906.
55 R. A. Sheldon, *Green Chem.*, 2007, **9**, 1273-1283.
56 EATOS: Environmental Assessment Tool for Organic Syntheses, http://www.metzger.chemie.uni-oldenburg.de/eatos/english.htm. See: M. Eissen and J. O. Metzger, *Chem.–Eur. J.*, 2002, **8**, 3580-3585.
57 *E*-factor calculations including the solvents used in the reaction can be found in the ESI. Values vary between 274 (laccase/TEMPO-TA methodology) and 495 (ADH-AmDH method).

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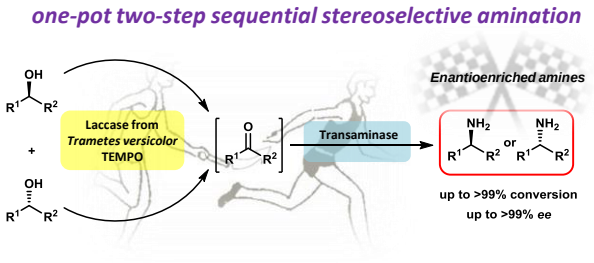

A one-pot/two-step chemoenzymatic sequential methodology has been developed for the selective amination of secondary alcohols by combining the laccase from *Trametes versicolor*/TEMPO catalytic system with the stereoselective action of transaminases