Chemoenzymatic synthesis of (2*S*)-2-arylpropanols through a dynamic kinetic resolution of 2-arylpropanals with alcohol dehydrogenases†

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We applied Horse Liver Alcohol Dehydrogenase (HLADH) to the enantioselective synthesis of six (2*S*)-2-arylpropanols, useful intermediates in the synthesis of Profens. The influence of substrate structure and reaction conditions on yields and enantioselectivity were investigated. The high yields and high enantioselectivity towards the (*S*)-enantiomer obtained in the bioreduction of 2-arylpropionic aldehydes, clearly indicate the achievement of a DKR process through a combination of an enzyme-catalyzed kinetic reduction with a chemical base-catalyzed racemization of the unreacted aldehydes. The racemization step is represented by the keto–enol equilibrium of the aldehyde and can be controlled by modulating pH and reaction conditions.

Introduction

The increasing attention dedicated to biocatalytic processes, which combine selectivity and environmental sustainability, in drug synthesis is not surprising.[1] The main advantages of the use of enzymes as catalysts are represented by the high efficiency in terms of yields, chemo-, regio-, and stereo-selectivity along with their use in mild conditions.[2] Enzymes have been widely employed in the resolution of racemates (kinetic resolution, KR).[3] However, the maximum yield of kinetic resolutions is only 50 per cent, which is economically and ecologically unattractive unless recycling of the undesired enantiomer is easily achievable; the ultimate improvement in this context is represented by dynamic kinetic resolution (DKR).[4] A DKR process combines a kinetic resolution and *in situ* racemization of the unreacted enantiomer and can overcome the 50 per cent yield limitation in classical kinetic resolution whenever the racemization rate successfully competes with that of the resolution reaction.

In DKR processes, enzymes have been successfully employed. Typically, several lipases have been used in esterification or hydrolysis in highly stereoselective DKR, with good yields and under mild conditions.[5] Dehydrogenases have been employed less frequently than lipases in organic synthesis and in DKR because of a number of drawbacks: 1) a limited number of commercially available purified enzymes; 2) limited stability and activity in non-aqueous solvents; and 3) requirement for a co-factor and its associate recycling system that increase the complexity of the reaction. However, recent applications of alcohol dehydrogenases and carbonyl reductases show that the productivity of these systems, particularly in the reduction mode, is constantly increasing, and some seminal examples of DKR by dehydrogenases have been successfully reported.[6] Among dehydrogenases, the most prominent enzymes out of this class are the widely employed yeast alcohol dehydrogenase (YADH, Baker's Yeast) and the horse liver alcohol dehydrogenase (HLADH). In particular, HLADH is a commercially available, NAD(H)-dependent biocatalyst characterized by a very broad substrate tolerance.[7] Recently, a process to produce this enzyme in bacteria, avoiding the use of animal sources, has been established opening the way for large scale applications.

Following our interest in the use of enzymes for the synthesis of bioactive molecules,[8] we recently applied HLADH to the enantioselective synthesis of (2*S*)-2-phenylpropanol and (2*S*)-2-(4-isobutylphenyl)propanol (*S*-Ibuprofenol) *via* an efficient dynamic kinetic resolution (DKR) of the parent racemic aldehydes.[9] (2*S*)-2-Arylpropanols are useful intermediates in flavor manufacture and can be oxidized[10] to (2*S*)-2-arylpropionic acids, active ingredients of the Profen class. Profens are a subclass of the non-steroidal antiinflammatory class of drugs (NSAIDs) and in recent years, have come to dominate this therapeutic area. Ibuprofen, for example, is used in the treatment of a number of inflammatory conditions such as arthritis, muscular strain, and cephalalgia. Profens are chiral drugs and the (*S*)- and (*R*)-enantiomers differ substantially in both their pharmacodynamics and pharmacokinetic properties. Prior to the early nineties, the (*S*)-enantiomer was regarded as the eutomer of the Profens and the (*R*)-enantiomer as the distomer. However, observations in the late eighties made this distinction less clear and it is now recognised that the (*S*)-profens are the enantiomers that inhibit prostaglandin synthetase.[11] It was also evidenced that the (*R*)-enantiomer of Ibuprofen can be interconverted to the (*S*)-enantiomer in the liver and kidney of pigs and rats. However this inversion requires CoA and ATP as cofactors which was validated by showing that (*R*)-Ibuprofen-CoA did not racemize in either buffer solution (pH 7.4) or human plasma.[12] Moreover, the (*R*)-Ibuprofen displays toxicity due to its storage in fatty tissue as a hybrid glycerol ester, whose long-term effects are not known.[13] Effectively, most Profens are moving towards single enantiomer administration as the rule. Enantiopure Profens are usually obtained through a final kinetic resolution of the esters by enzymatic hydrolysis,[14] but performing a dynamic kinetic resolution at an earlier stage may provide an alternative

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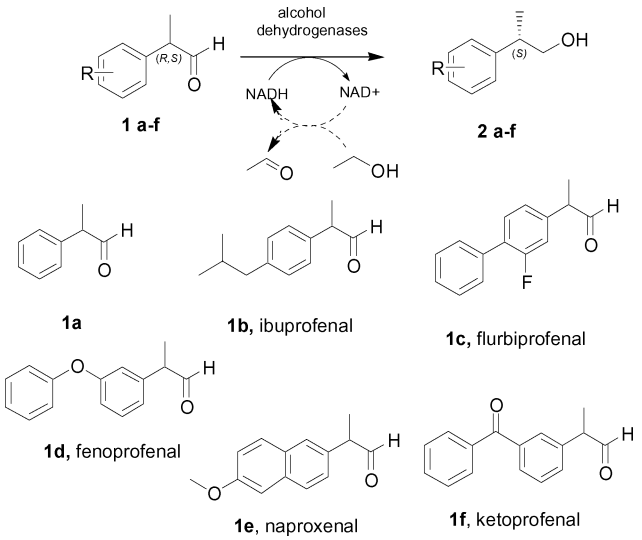
route to their asymmetric synthesis thus making the process simpler and more economically advantageous.

Here we present a detailed study of the enzymatic reduction by dehydrogenases of arylpropionic aldehydes to obtain (2*S*)-2-arylpropanols. In particular we investigated the racemization step and the influence of pH on both the racemization of the starting aldehyde and the enzymatic reduction. We then applied the reaction to a wider series of racemic 2-arylpropanals (Scheme 1) and even on a semi-preparative scale, and the influence of substrate structures and reaction conditions on yields and enantioselectivities was discussed in depth. As a further development, we demonstrated the maintenance of the optical purities of (2*S*)-flurbiprofenol and (2*S*)-fenoprofenol in a subsequent oxidation reaction to the corresponding acids, thus realizing the enantioselective synthesis of Flurbiprofen and Fenoprofen.



Scheme 1 Enzymatic reduction of racemic 2-arylpropanals **1a–f** to the corresponding (2*S*)-2-arylpropanols **2a–f**.

Results and discussion

Preliminary experiments were performed on racemic 2-phenylpropanal. The enzymatic reduction was explored using the two commercial alcohol dehydrogenases, YADH and HLADH in phosphate buffer with an excess of NADH. Data are reported in Table 1. With both enzymes (entries 3 and 5) the enantioselectivity was good and the yield exceeded 50%, thus indicating that a DKR process was in action but with YADH reaction times were longer, reflecting the broader substrate specificity of HLADH. The enantiomeric excess was always in favour of the (*S*)-enantiomer and noticeably improved by lowering the enzyme amount (entries 6 and 7). To increase aldehyde solubility in the reaction mixture, the reduction with HLADH was tested also in the presence of an organic co-solvent (CH_3CN, THF, and hexane). The reaction proceeded in all cases but yields were lower and in hexane the stereoselectivity was poor, probably because of the excess of enzyme needed to catalyze the reaction.

The same process was tested in the presence of ethanol as a co-substrate to realize the *in situ* co-factor recycling (Table 2). With YADH the reduction did not proceed whereas with HLADH the

Table 1 Enzymatic reduction of 2-phenylpropanal **1a** to 2-phenylpropanol **2a** with excess NADH.[a]

Ent.	Enzyme, amount (U/mmol), time	Co-solvent (%)	**2a** (yield %)[b]	*S*/*R*
1	YADH 500, 5 h	—	18	99/1
2	YADH 500, 24 h	—	53	93/7
3	YADH 500, 96 h	—	79	92/8
4	HLADH 25, 5 h	—	81	82/18
5	HLADH 25, 24 h	—	89	81/19
6	HLADH 5, 5 h	—	75	99/1
7	HLADH 5, 24 h	—	89	95/5
8	HLADH 25, 5 h	CH_3CN (10)	72	89/11
9	HLADH 25, 5 h	THF (10)	55	96/4
10	HLADH 500, 5 h	Hexane (99)	77	63/37
11	HLADH 500, 5 h	Hexane (98)	80	62/38

[a] Reaction conditions: method A, 0.5 mM 2-phenylpropanal **1a**, 1 mM NADH, 0.1 M phosphate buffer pH 7.5, 27 °C, $V = 5$ mL, enzyme amount expressed as U/mmol of aldehyde **1a**; U refers to ethanol/acetaldehyde activity as reported by suppliers. [b] Yields obtained by HPLC through calibration plot.

Table 2 Enzymatic reduction of 2-phenylpropanal **1a** to 2-phenylpropanol **2a** with NADH recycling.[a]

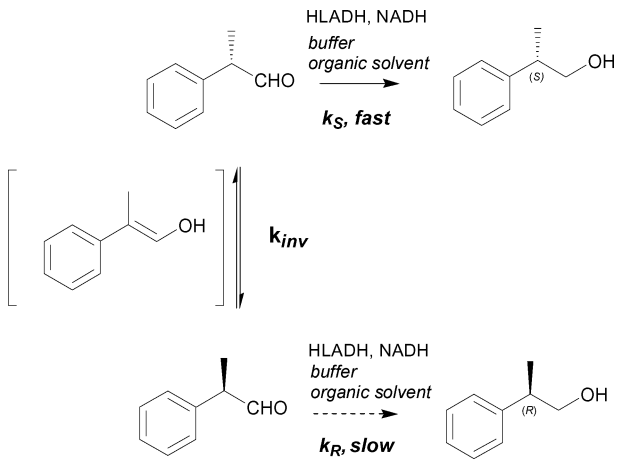
Ent.	Enzyme, amount (U/mmol), time	Co-solvent (%)	pH	**2a** (yield %)[b]	*S*/*R*
1	YADH 500, 5 h	—	7.5	—	—
2	YADH 500, 24 h	—	7.5	traces	—
3	YADH 50, 96 h	—	7.5	5	50/50
4	HLADH 25, 5 h	—	7.5	89	84/16
5	HLADH 25, 24 h	—	7.5	99	83/17
6	HLADH 5, 24 h	—	8.0	85	95/5
7	HLADH 25, 5 h	CH_3CN (10)	7.5	72	97/3
8	HLADH 5, 96 h[c]	CH_3CN (16)	7.5	90	94/6
9	HLADH 25, 24 h	CH_3CN (10)	8.0	85	98/2
10	HLADH 25, 5 h	THF (10)	7.5	54	95/5
11	HLADH 25, 24 h	THF (10)	8.0	80	99/1
12	HLADH 25, 24 h	THF (5)	8.0	88	95/5
13	HLADH 500, 5 h	Hexane (99)	7.5	55	90/10
14	HLADH 500, 5 h	Hexane (95)	7.5	68	72/28

[a] Reaction conditions: method B, 0.5 mM 2-phenylpropanal, 0.01 mM NADH, 0.5 M EtOH, 0.1 M phosphate buffer, 27 °C, $V = 15$ mL, enzyme amount expressed as U/mmol of aldehyde **1a**. [b] Yields obtained by HPLC through calibration curve. [c] 1 mmol **1a** (45 mM), 96 h, $V = 24$ mL, isolated yield.

results obtained at pH 7.5, in terms of yields and enantioselectivity, were at least comparable with those obtained with NADH in excess (*cf.* entry 4 Table 1 and entry 4 Table 2). On increasing the pH or adding an organic co-solvent, the enantiomeric ratio increased, even if the co-solvent addition lowered the enzyme activity and extended the reaction time to obtain a complete conversion.

The high yields obtained in the bioreduction of 2-phenylpropionic aldehyde, clearly indicate the achievement of a DKR process through a combination of an enzyme-catalyzed kinetic resolution with a chemical base-catalyzed racemization. In our case, the racemization step is represented by the keto–enol equilibrium of the starting aldehyde. The stereoinversion of arylpropionic aldehydes in aqueous medium is a feasible process and supported by the reported racemization of *R*- and *S*-Ibuprofen

(2-(4-isobutyl)phenyl-propionic acid)[15] and 2-phenyl ketones.[16] In the reaction medium the unreacted (*R*)-aldehyde undergoes a racemization process through the achiral enolic form (Scheme 2). The stereoinversion was definitively proved by the reduction with HLADH of the enantiopure (2*R*)-2-phenylpropanal; the (2*S*)-2-phenylpropanol was obtained in 95% yield and an enantiomeric ratio *S*/*R* of 95 : 5.[9]

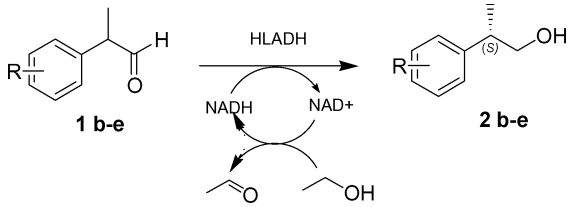


Scheme 2 Resolution and racemization steps in reductive DKR of 2-phenylpropanal.

To extend the application scope, the enzymatic reduction with HLADH was applied to 2-arylpropanals **1b–f** suitable for use as intermediates in the synthesis of Profens (Scheme 1, Table 3 and 4). The racemic aldehydes are not commercially available and, as a shortcut for the purpose of this work, have been obtained from the corresponding commercial 2-arylpropionic acids (see ESI† for details). The bioreductions were performed using NADH in catalytic amount, and in the presence of EtOH for the co-factor recycling.

On changing substrates and reaction conditions the interplay between the resolution and the racemization step becomes fundamental to get satisfactory yields and enantioselectivities (Table 3). In the case of ibuprofenal **1b** and flurbiprofenal **1c** very good yields and complete enantioselelectivity were achieved using a 10% mixture of CH_3CN in phosphate buffer (pH 7.5) (Table 3, entries 2 and 5). In the same conditions with fenoprofenal **1d** the *S*/*R* ratio was not satisfactory (entry 9) but it was improved by increasing pH and lowering the enzyme amount (entry 14). To keep the enzyme concentration low and obtain quantitative yields, the enzyme was added in two aliquots at different reaction times (Table 3, *cf.* entries 13 and 14). In the case of naproxenal **1e**, the enantiomeric ratio was good (entry 20) but high yields could not be obtained even if in some cases more than 50% was obtained thus indicating the presence of a DKR (entry 21). In all cases we obtained the (2*S*)-arylpropanol as the major enantiomer. For **2b** and **2e** the configuration was established by comparison with samples of enantiopure (*S*)-alcohols obtained by reduction with $BH_3{\cdot}Me_2S$ of the commercial (*S*)-acids. For **2c** and **2d** (flurbiprofenol and fenoprofenol), the enzymatic reduction was performed on a semi-preparative scale (Table 3, entries 7 and 15), the (*S*)-alcohols were oxidized to the corresponding (2*S*)-2-arylpropionic acid (flurbiprofen and fenoprofen) using $KMnO_4$ in acetone and the

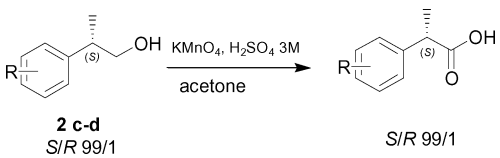
Table 3 Enzymatic reduction of 2-arylpropanals **1b–e** to 2-arylpropanols **2b–e** with NADH recycling.[a]



Ent.	Sub.	Enzyme amount (U/mmol)	pH	10% Co-solvent	Yield (%)[b]	*S*/*R*
1	**1b**	25	7.5	—	69	97/3
2	**1b**	25	7.5	CH_3CN	93	>99/1
3	**1b**	25	7.5	THF	46	>99/1
4	**1c**	25	7.5	—	92	96/4
5	**1c**	25	7.5	CH_3CN	98	>99/1
6	**1c**	25	7.5	THF	49	>99/1
7	**1c**[d]	25	8	CH_3CN	63	>99/1
8	**1d**	25	7.5	—	93	66/34
9	**1d**	25	7.5	CH_3CN	92	82/18
10	**1d**	25	7.5	THF	68	>99/1
11	**1d**	10[c]	8	—	99	81/19
12	**1d**	25	8	THF	82	93/7
13	**1d**	25	8	CH_3CN	97	82/18
14	**1d**	10[c]	8	CH_3CN	98	93/7
15	**1d**[d]	25	8	CH_3CN	82	>99/1
16	**1e**	25	7.5	—	44	92/8
17	**1e**	25	7.5	THF	42	88/12
18	**1e**	25	7.5	CH_3CN	38	90/10
19	**1e**	25	8.0	—	63	86/14
20	**1e**	25	8.0	THF	58	98/2
21	**1e**	25	8.0	CH_3CN	67	92/8

[a] Reaction conditions: method B, 0.1 M phosphate buffer, 0.5 mM 2-arylpropanals **1b–e**, 0.01 mM NADH, 0.5 M EtOH, 24 h, enzyme amount expressed as U/mmol of aldehyde. [b] Yields obtained by HPLC through calibration curve. [c] Total amount, added in two aliquots at $t = 0$ and $t = 5$ h. [d] Reaction conditions: method C, 0.5 mmol aldehyde (5 mM), 96 h, isolated yield.

S-enantiopreference was definitively confirmed by comparison with literature optical rotation data and HPLC analyses on chiral columns (Scheme 3, see ESI† for details).

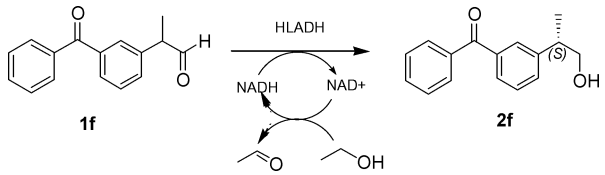


Scheme 3

In both cases no racemization occurred in the oxidation process and the enantiomeric ratio of the alcohols was conserved in acids. With all substrates, when the reaction yields were low, the unreacted aldehyde was not recovered but we detected the corresponding 2-arylmethylketones as by-products. The formation of a by-product is particularly evident in the case of naproxenal **1e** which never gives high yields and forms a significant amount of arylmethylketone **3e** (Scheme 4).

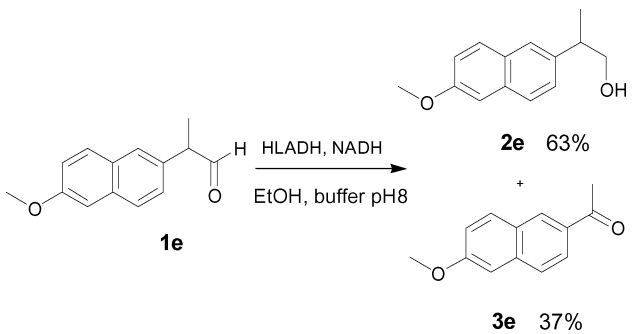
The conversion of 2-phenylpropanal into acetophenone is reported in the literature through a metal catalysis,[17] and it is known also in the case of other 2-arylpropanals.[18] In our case we identified arylmethylketones in blank reactions with aldehydes in the same

Table 4 Enzymatic reduction of ketoprofenal **1f** to ketoprofenol **2f** with NADH recycling.[a]



Entry	Enzyme amount (U/mmol)	pH	10% Co-solvent	**2f** (yield %)[b]	*S*/*R*
1	25	7.5	—	95	75/25
2	10[c]	7.5	—	99	93/7
3	25	7.5	CH_3CN	80	94/6
4	10[c]	7.5	CH_3CN	54	99/1
5	25	7.5	THF	88	95/5
6	10 [c]	7.5	THF	36	99/1
7	25	8.0	—	37	99/1
8	25	8.0	CH_3CN	9	99/1

[a] Reaction conditions: method B, phosphate buffer, 0.5 mM **1f**, 0.01 mM NADH, 0.5 M EtOH, 24 h, enzyme amount expressed as U/mmol of aldehyde (**1f**). [b] Yields obtained by HPLC through calibration curve. [c] Total amount, added in two aliquots at $t = 0$ and $t = 5$ h.



Scheme 4

reaction conditions of the DKR process but in the absence of the enzyme. The reduced products of the arylmethylketones, *i.e* 1-arylethanols, were never detected in the enzymatic reactions.

Ketoprofenal is a particularly interesting substrate for the enzymatic reduction (Table 4) since **1f** bears two carbonyl groups, a diaryl ketone and an aldehyde. HLADH demonstrated a high degree of chemoselectivity producing as a unique compound the ketoalcohol **2f** in high yield. No traces of the possible ketone reduction by-products (the diol or the hydroxy-aldehyde) were detected. With this substrate both better yields and enantioselectivity were obtained in the absence of a co-solvent (entry 2); by using 10% of CH_3CN and THF, *S*/*R* values were always good but yields were significantly lower.

The high (*S*)-enantioselectivity of the enzymatic reduction, in terms of the kinetic constants, means that the rate constant for (*S*)-aldehydes is higher than that for (*R*)-aldehydes ($k_S \gg k_R$), but the efficiency of the entire DKR process is also strictly related to the rate of the racemization k_{inv}. Noyori *et al.*[19] developed a quantitative treatment of a DKR process generated by a reduction coupled with a stereoinversion and showed that quantitative conversions and good enantioselectivity can be still obtained when the racemization rate constant (k_{inv}) and the rate constant for reduction of the faster enantiomer are the same (Scheme 2). If the k_{inv} becomes slower than k_{fast} a decrease in the ee of the preferred enantiomer at high conversions can occur but a good DKR process can still be obtained if k_{inv} is higher than the rate constant for reduction of the slower reacting enantiomer ($k_{inv} > k_{slow}$). We observed with time a decrease of the *S*/*R* ratio on increasing the reaction yields (entries 4–7, Table 1 and entries 4–5, Table 2) and this could be due to a k_{inv} slower than k_S. Moreover, both resolution and racemization steps are expected to be pH dependent, particularly the keto–enol tautomerism that occurs *via* proton transfer.[20] Therefore we studied the bioreduction of 2-phenylpropanal at different pH values in the range of pH 6–8.5 in consideration of the pH sensitivity of HLADH (Fig. 1). As expected, yields are strongly influenced by pH value (Fig. 1a). At pH 7.5 the yield was particularly good, but at pH 8.5 it was low. This result could be due to a decreased enzyme activity or co-factor binding[21] at higher pH values. However, at the higher pH the best enantioselectivity was obtained (Fig. 1b). Moreover a decreased (*S*)-enantiomer % on increasing the reaction time and conversion confirms that the racemization rate k_{inv} is very close to or even lower than k_S, according to the kinetic analysis by Noyori.[19]

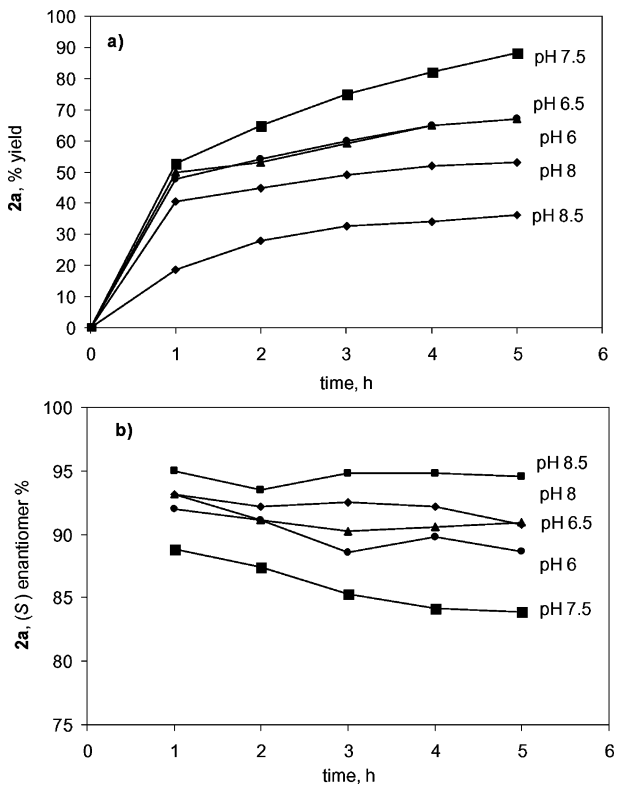


Fig. 1 Time course for pH effects on a) yields and b) enantioselectivity. Reaction conditions: 0.5 mM **1a**, 0.1 M phosphate buffers, 0.01 mM NADH, 0.5 M EtOH.

As above mentioned, the racemization step is very important for the efficiency of a DKR process, we thus studied in more detail the racemization of 2-phenylpropanal, ibuprofenal and naproxenal (**1a,1b** and **1e**) in the absence of the enzymatic reduction. We measured the decrease of optical rotation [22] with time of solutions of enantiopure (2*S*)-2-phenylpropanal, (*S*)-ibuprofenal and (*S*)-naproxenal (**1a,1b** and **1e**) in phosphate buffer 0.1 M at pH 7.5 and 8, CH_3CN was used as co-solvent to ensure complete substrate

dissolution. Data are plotted in Fig. 2. Considering racemization as the formation of a racemate from a pure enantiomer in an irreversible first-order reaction it follows that: [23]

$$-\ln(\alpha_t/\alpha_o) = k_{inv}t \quad (1)$$

and

$$t_{1/2} = \ln 2/k_{inv} \quad (2)$$

α_t being the observed optical rotation at time t, α_0 the initial observed optical rotation, $t_{1/2}$ the racemization half-life and k_{inv} the racemization constant.

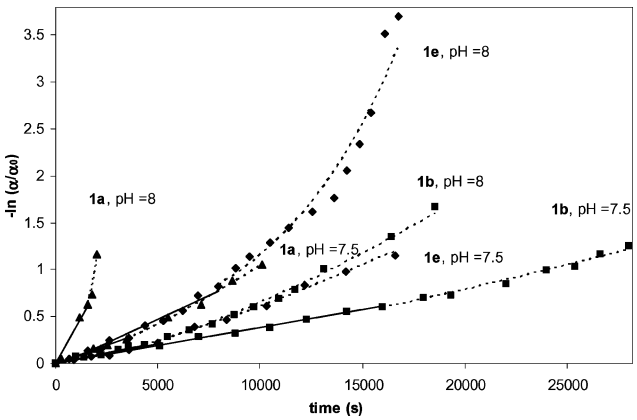


Fig. 2 Time course of optical rotation of aldehydes **1a** (▲), **1b** (■) and **1e** (♦) at pH 7.5 and 8. Conditions: aldehyde concentration 5 mM, phosphate buffer 0.1 M, CH_3CN 30%.

According to eqn (1) a linear relationship with slope corresponding to k_{inv} values should be obtained. In our case, as can be seen in Fig. 2, linearity was not observed during the entire experimental course because the aldehydes form chiral hydrates and by-products with time (HPLC monitoring). However over the initial time range a linear behaviour (solid lines in Fig. 2) was observed for all plots allowing calculation of the k_{inv} values reported in Table 5. Half-lives $t_{1/2}$calc have been calculated from k_{inv} through eqn (2) and have been compared with the observed $t_{1/2}$ ($t_{1/2obs}$), the time corresponding to $\alpha_o/2$.

As expected, the racemization rate is highly controlled by pH, for all aldehydes k_{inv}s increase and half-lives decrease with increasing pH (Table 5, *cf.* entries 1 and 2, 3 and 4, 5 and 6). 2-Phenylpropanal is the aldehyde with the fastest racemization rate and this is also confirmed by the experimental observation that enantiopure 2-phenylpropanal has a very short shelf-life. Ibuprofenal **1b** and **1e** have slower k_{inv} values but whereas for naproxenal **1e** the whole process is limited by the aldehyde decomposition into the 2-arylmethylketone, in the case of **1b** a good DKR is obtained with longer reaction times.

Table 5 Racemization constants k_{inv}, half-lives $t_{1/2}$calc and $t_{1/2}$obs obtained from data in Fig. 2

Entry	Aldehyde	pH	$k_{inv}/10^{-6}s^{-1}$	$t_{1/2}$ obs/min	$t_{1/2}$ calc/min
1	**1a**	7.5	75 ± 6	120	154
2	**1a**	8	419 ± 30	30	28
3	**1b**	7.5	29 ± 1	320	397
4	**1b**	8	52 ± 2	180	224
5	**1e**	7.5	41 ± 2	180	279
6	**1e**	8	95 ± 3	115	113

Conclusions

The enzymatic reduction with HLADH was successfully applied to six racemic 2-arylpropanals affording (2*S*)-2-arylpropanols, suitable for use as intermediates in Profen syntheses. In most cases both excellent enantioselectivity and yields could be obtained by modulating the reaction conditions, thus influencing the resolution and the racemization step of the DKR process. In the case of ketoprofenal complete chemoselectivity was also achieved.

The starting racemic 2-arylpropanals are in some cases already used as intermediates in the synthesis of Profen[24] or can be obtained by hydroformylation[25] or hydrovinylation[26] of vinylarenes. The enantiopure products, (2*S*)-2-arylpropanols, can in turn be oxidized to (2*S*)-2-arylpropanoic acids, so that the process here described could represent an interesting alternative for the synthesis of enantiomerically pure Profens. Work is in progress to study alternative more sustainable procedures for the final oxidation step.

Experimental

General

Chemicals were purchased from Sigma-Aldrich or TCI and used without further purification. YADH: Sigma A3263. HLADH: Sigma A9589 from Horse Liver, or evo-1.1.211 cloned isoenzyme E from Evocatal, or cloned enzyme prepared according to existing protocols[27] in Dr Paradisi's laboratory (UCD, Ireland) and kindly given to us for the purpose of this research. TLC: Merck 60 F254. Column chromatography: Merck silica gel 20–300 mesh. FT-IR: Nicolet 308 measured as films between NaCl plates, wave numbers reported in cm^{-1}. ^1H and ^{13}C NMR spectra: obtained on a Varian GEMINI 200 and Varian INOVA 300 spectrometers with a 5 mm probe. All chemical shifts have been quoted relative to deuterated solvent signals, δ in parts per million, J in hertz. Elemental analysis: Perkin-Elmer 2400 Series II CHNS/O analyzer. Reverse phase HPLC: Agilent Technology HP1100, column ZORBAX-Eclipse XDB-C8 Agilent Technologies. The compounds were eluted with CH_3CN–H_2O, gradient: from 30% to 100% of CH_3CN in 15 min, then 100% of CH_3CN for 10 min, $T = 30$ °C. Direct phase HPLC: Hewlett-Packard HP1090 Series II, columns Daicel's Chiralpack (0.46 cm Ø x 25 cm) Chiralcel OD or OF. The compounds were eluted with Hexane/iPrOH, flux 0.5 mL min^{-1}, $T = 40$ °C (see Table 5). 2-Phenylpropanal **1a** and 2-phenylpropanol **2a** are commercial and re-distilled before use. Non-commercial racemic 2-arylpropanals **1b–f** can be prepared starting from the corresponding commercial carboxylic acids using two alternative procedures: method 1) preliminary esterification and then iBu_2AlH reduction,[9] or method 2) over-reduction to primary alcohols with $BH_3{\cdot}Me_2S$[28] and then Swern oxidation to aldehydes.[29] In both procedures 2-arylpropanol alcohols **2b–f** can be obtained as intermediates or by-products and can be used for HPLC calibration plot. In the case of ketoprofenal **1f**, to avoid ketone protection/deprotection steps, method B was used. Full details and data in ESI.†

General procedure for enzymatic reduction.

Method A with excess NADH: into a vial equipped with a magnetic stirrer (double spinfin magnetic stirring bar) all reagents were added in the following order: 0.5 mL of a 5 mM solution of the starting aldehyde in CH_3CN or THF, 0.5 mL of a 10 mM solution of NADH freshly prepared in 0.1M phosphate buffer (pH 7.5), 0.1M phosphate buffer (pH 7.5) to reach a total final volume of 5 mL and the chosen amount of enzyme (indicated in Table 1–4) from a freshly prepared solution in phosphate buffer. In the case of co-solvent absence, the aldehyde was directly added to the reaction mixture. In the case of enzymatic reductions in hexane, data were obtained by adding solid lyophilized cofactor together with lyophilized enzyme into the reaction vessel before adding the solvent mixture and the substrate.

Method B with NADH recycling: into a vial equipped with a magnetic stirrer (double spinfin magnetic stirring bar) all reagents were added in the following order: 1.5 mL of a 5 mM solution of the starting aldehyde in CH_3CN or THF, 0.45 mL of EtOH, 1.5 mL of a 0.1 mM solution of NADH freshly prepared in the appropriate 0.1 M buffer, 0.1M buffer to reach a total final volume of 15 mL and the chosen amount of enzyme from a freshly prepared solution in buffer. In the case of co-solvent absence, 2-arylpropanals were diluted in EtOH.

Method C, semi-preparative scale: into a flask equipped with a magnetic stirrer were added in the following order: 0.5 mmol of the starting aldehyde **1a,c–d** in 10 mL of CH_3CN, 3.4 mL of EtOH and phosphate buffer 0.1 M (pH = 8) to reach a final total volume of 100 mL. After 15 min NADH (0.01 mmol, 7.1 mg) and the enzyme (1 mg, 0.46 U/mg) were added to the solution. After 96 h the reactions were worked up by adding 1 g of solid NaCl to the aqueous phase and extracting 3 times with EtOAc (3 × 75 mL). The organic phases were dried over Na_2SO_4 and concentrated *in vacuo*. Yields after flash chromatography (80/20 : cyclohexane/ethylacetate). Oxidation to 2-arypropionic acids is reported in the ESI.†

Formation of arylpropanols was monitored by inverse phase HPLC analysis: at different reaction times, aliquot samples were filtered, diluted and directly injected. Calibration curves obtained with pure arylpropanols (five dilutions, each in triplicate) were used for quantitative analysis. Enantiomeric ratios were determined by HPLC analysis on chiral columns.

2-(4-Isobutylphenyl)-propanol (Ibuprofenol) 2b. ^{1}H NMR (300 MHz, $CDCl_3$): δ 0.92 (d, J = 6.6, 6H, $(CH_3)_2CH$), 1.28 (d, J = 7.2, 3H, CH_3CH), 1.4 (bs, 1H, OH), 1.86 (m, 1H, $(CH_3)_2CHCH_2$), 2.46 (d, J = 7.2, 2H, CH_2), 2.93 (sextet, J = 7.2, 1H, CH), 3.70 (d, J = 7.2, 2H, CH_2OH), 7.10–7.17 (m, 4H, arom); ^{13}C NMR (75 MHz, $CDCl_3$): δ 17.5, 22.2, 30.0, 41.8, 44.8, 68.4, 127.0, 129.0, 139.6, 140.9; IR: ν = 3372, 1684, 1513, 1465; elemental analysis calcd (%) for $C_{13}H_{13}O$: C 81.20, H 10.48; found: C 81.15, H 10.52.

2-(2-Fluoro-biphenyl-4-yl)-propan-1-ol (Flurbiprofenol) 2c. Obtained in 12% yield as a by-product in **1c** preparation following method A or in 95% yield following method B. ^{1}H NMR (200 MHz, $CDCl_3$): δ 1.34 (d, J = 7.0, 3H, CH_3), 1.57 (bs, 1H, OH), 3.02 (sextet, J = 7.0, 1H, CH), 3.77 (d, J = 7.0, 2H, CH_2), 7.05–7.16 (m, 2H, arom), 7.28–7.61 (m, 6H, arom); ^{13}C NMR (50 MHz, $CDCl_3$): δ 17.4, 41.9, 68.4, 114.9 (d, $J_{(C,F)}$ = 23), 123.5 (d, $J_{C,F}$ = 3), 127.2 (d, 1C, $J_{C,F}$ = 18), 127.5, 128.4, 128.9 (d, $J_{C,F}$ = 3), 130.7 (d, J(C,F) = 4), 135.7, 145.5 (d, 1C, $J_{C,F}$ = 7), 159.9 (d, 1C, $J_{C,F}$ = 247); IR: ν = 3373, 1023; elemental analysis calcd (%) for $C_{15}H_{15}FO$: C 78.24, H 6.57; found: C 78.19, H 6.56. (*S*)-**2c** obtained from semi-preparative enzymatic reaction: $[\alpha]_D^{20} = -11.5$, (c = 2.6, $CHCl_3$).

2-(3-Phenoxy-phenyl)-propan-1ol (Fenoprofenol) 2d. Obtained in 15% yield as a by-product in **1c** preparation. ^{1}H NMR (300 MHz, $CDCl_3$): δ 1.30 (d, J = 7.2, 3H, CH_3), 1.4 (bs, 1H, OH), 2.97 (sextet, J = 7.2, 1H, CH), 3.72 (d, J = 7.2, 2H, CH_2), 6.89–7.19 (m, 6H, arom), 7.30–7.42 (m, 3H, arom); ^{13}C NMR (75 MHz, $CDCl_3$): δ 17.5, 42.2, 68.4, 116.7, 117.9, 118.8, 122.3, 123.2, 129.7, 145.9, 157.0, 157.4. IR: ν = 3350, 1584, 1489, 1250; elemental analysis calcd (%) for $C_{15}H_{16}O_2$: C 78.92, H 7.06; found: C 79.02, H 7.05.

(*S*)-**2d** obtained from semi-preparative enzymatic reaction: $[\alpha]_D^{20} = -10.0$ (c = 2.4, $CHCl_3$).

2-(6-Methoxy-naphthalen-2-yl)-propan-1-ol (Naproxol) 2e. Obtained in 10% yield as a by-product in **1e** preparation. ^{1}H NMR (300 MHz, $CDCl_3$): δ 1.38 (d, J = 7.0, 3H, CH_3CH), 1.4 (bs, 1H, OH), 3.11 (sextet, J = 7.0, 1H, CH), 3.70 (d, J = 7.0, 2H, CH_2), 3.94 (s, 3H, OCH_3), 7.14–7.28 (m, 2H, arom), 7.34–7.40 (m, 1H, arom), 7.63–7.76 (m, 3H, arom); ^{13}C NMR (50 MHz, $CDCl_3$): δ 17.6, 42.4, 55.3, 68.6, 105.6, 118.9, 125.9, 126.3, 126.2, 127.2, 129.1, 133.5, 138.7, 157.5; IR: ν = 3300, 1604, 1028; elemental analysis calcd (%) for $C_{14}H_{16}O_2$: C 77.75, H 7.46; found: C 77.91, H 7.51.

[3-(2-Hydroxy-1-methyl-ethyl)-phenyl]-phenyl-methanone (Ketoprofenol) 2f. Obtained in 30% yield as a by-product in the obtaining of **1f**. ^{1}H NMR (300 MHz, $CDCl_3$): δ 1.35 (d, J = 7.0, 3H, CH_3CH), 1.4 (bs, 1H, OH), 3.07 (m, 1H, CH), 3.79 (d, J = 6.6, 2H, CH_2), 7.44–7.86 (m, 9H, arom); ^{13}C NMR (75 MHz, $CDCl_3$): δ 17.6, 42.3, 68.5, 128.3, 128.4, 128.6, 128.9, 130.0, 131.7, 132.4, 137.6, 137.8, 144.2, 196.8; elemental analysis calcd (%) for $C_{16}H_{16}O_2$: C 79.97, H 6.71; found: C 80.05, H 6.69.

Determination of enantiomeric ratio and configurational assignment of major stereoisomers.

Enantiomeric ratios were determined by HPLC analysis on chiral columns (see Table 6 in ESI† for full details). In the case of alcohols **2a,2b,2e** and **2f** the (*S*)-configuration of the major isomer was established by direct comparison with commercial (*S*)-2-phenylpropanol or (*S*)-alcohols, obtained by reduction with $BH_3{\cdot}Me_2S$ of the commercial (*S*)-acids. For **2c** and **2d** the (*S*)-configuration of the major isomer was established by converting the alcohols obtained in the semi-preparative procedure into acids by oxidation with $KMnO_4$ and comparing the optical rotation with reported data (see ESI†).

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