Perkin 1 Abstracts: Biocatalysis in Organic Synthesis

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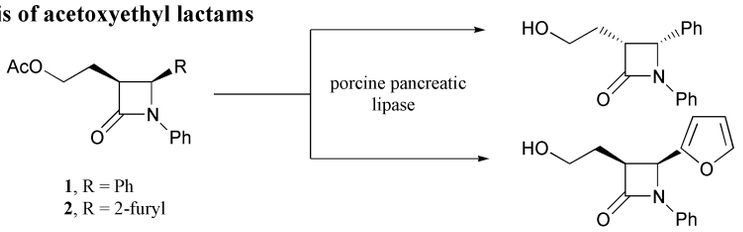
Perkin 1 Abstracts: Biocatalysis in Organic Synthesis aims to cover recent literature concerning the applications of enzymes and micro-organisms as catalysts in organic synthesis. The abstracts will emphasise the key synthetic step(s) that are mediated by the biocatalyst. Emerging technologies for biocatalyst design and optimisation will also be included.

Hydrolysis of acetoxyethyl lactams ***Lipase***

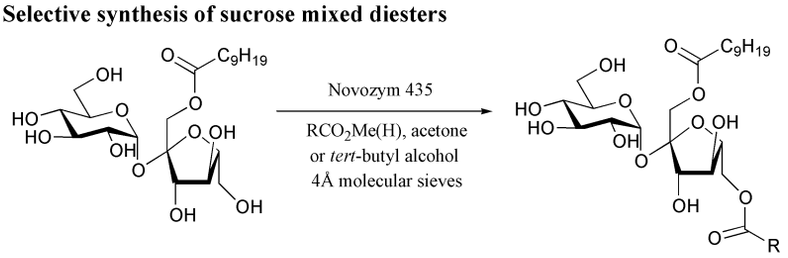


1, R = Ph
2, R = 2-furyl

The hydrolysis of acetoxyethyl lactams **1** and **2** with porcine pancreatic lipase (PPL) proceed with opposite stereoselectivity, the former according to Jones model of PPL, the latter according to the model of Seebach. This divergence was explained with reference to the binding of different substituents in the large hydrophobic pocket of PPL in the case of the two substrates.

A. Basak, K. R. Rudra, H. M. Bdour and J. Dasgupta, *Bioorg. Med. Chem. Lett.*, 2001, **11**, 305.

Selective synthesis of sucrose mixed diesters ***Lipase***

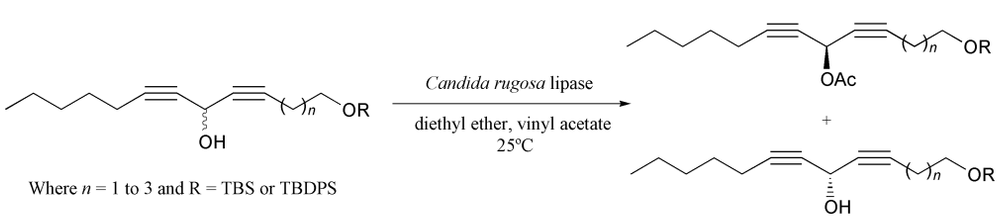


R = (Z)-$(CH_2)_7CH{=}CH$-$(CH_2)_5CH_3$ yield = 89%
R = $(CH_2)_{18}CH_3$ yield = 91%
R = (Z)-$(CH_2)_7CH{=}CH$-$(CH_2)_7CH_3$ yield = 75%

A range of other carbohydrates were acylated in yields ranging from 66 to 94%. The acylation occurs primarily at 6′-OH, on the fructose moiety, even when 1′-OH is available. Long chain fatty acids are better substrates compared to shorter ones and carboxylic acids can be used directly as the acylating agent.

P. Potier, A. Bouchu, G. Descotes and Y. Queneau, *Synthesis*, 2001, **3**, 458.

Enzymatic resolution of intermediates for lipoxygenase substrates ***Lipase***



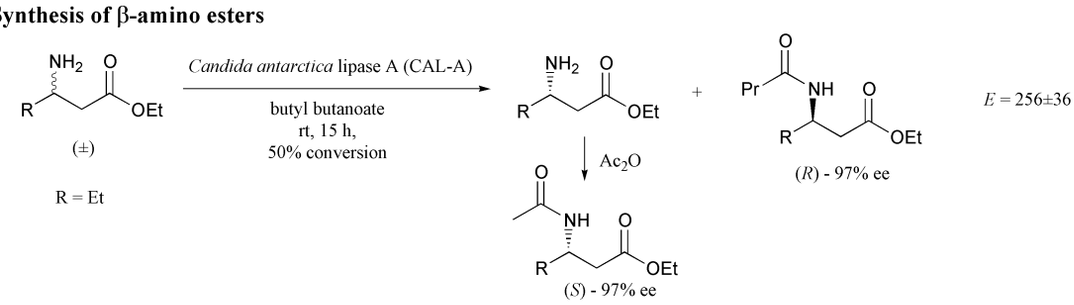
Where n = 1 to 3 and R = TBS or TBDPS

Where n =3 and R = TBDPS ee = 95% and yield = 39%

Where n =3 and R = TBDPS ee = 77% and yield = 37%

J. S. Yadav, S. Nanda and A. B. Rao, *Tetrahedron: Asymmetry*, 2001, **12**, 53.

Synthesis of β-amino esters ***Lipase***

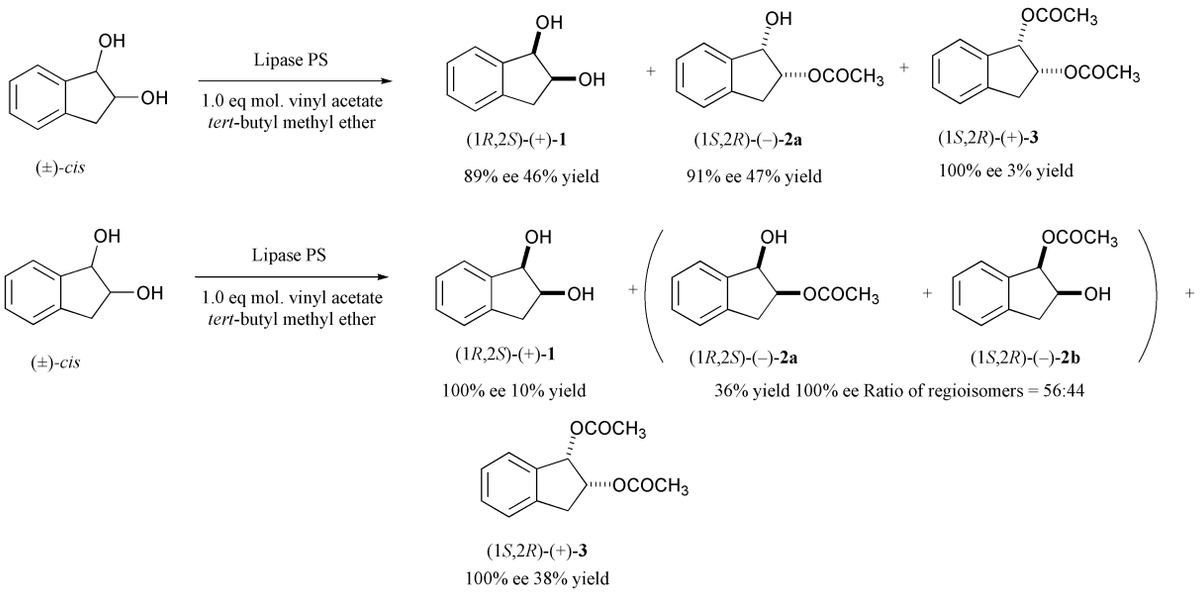


S. Gedey, A. Liljeblad, L. Lázár, F. Fülöp and L. T. Kanerva, *Tetrahedron: Asymmetry*, 2001, **12**, 105.

Various R substituents were screened. When R = branched chain aliphatic substituent, 2,2,2-trifluoroethyl butanoate was used as the acyl donor.

Kinetic resolution of *cis*-indane-1,2-diol

Lipase

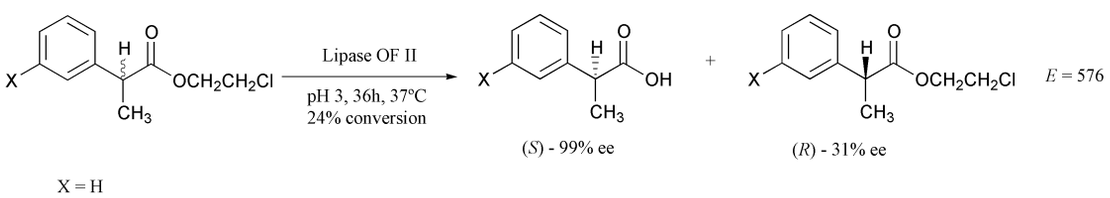


S. Nakano, Y. Igarashi and H. Nohira , *Tetrahedron: Asymmetry*, 2001, **12**, 59.

(1*S*,2*R*)-**2a** was subjected to the Ritter reaction to provide (1*S*,2*R*) *cis*-1-aminoindan-2-ol in 78% yield with 96% ee and under the same conditions (1*R*,2*S*) **2a** & **2b** gave (1*R*,2*S*) *cis*-1-aminoindan-2-ol in 70% yield and 100% ee.

Enhancement of the enantioselectivity of lipase OF catalysed hydrolysis

Lipase



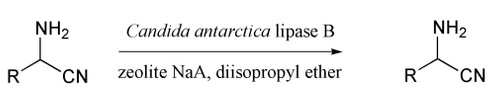
X = H

Y.-F. Chang and D.-F. Tai, *Tetrahedron: Asymmetry*, 2001, **12**, 177.

Three lipase OF fractions (*i.e.* I, II and III) were isolated from crude lipase OF using an agarose mercurial affinity column. These fractions were screened with three distinct ester substrates (*i.e.* X = H, isobutyl and benzoyl) with varying enantioselectivities.

Enantioselective acylation of α-aminonitriles

Lipase



1 R=Ph
2 R=CH_2CH_2Ph
3 R=$CH(CH_3)_2$

a R'=CH_3
b R'=CH_2Ph

4 R=Ph
5 R=CH_2CH_2Ph
6 R=$CH(CH_3)_2$

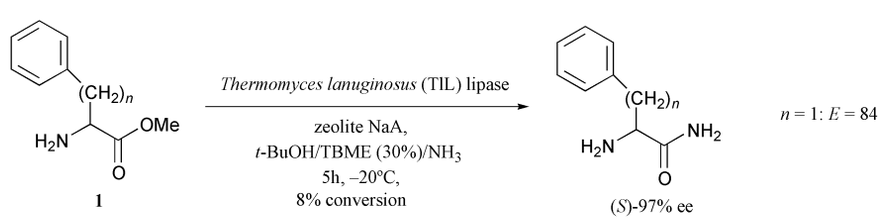
(*S*)-**4a**: 51% conversion, 10% ee
(*S*)-**4b**: 47% conversion, >99% ee
(*S*)-**5a**: 56% conversion, 5% ee
5b: no conversion
(*S*)-**6a**: 76% conversion, 0% ee
6b: no conversion

P. López-Serrano, J. A. Jongejan, F. van Rantwijk and R. A. Sheldon, *Tetrahedron: Asymmetry*, 2001, **12**, 219.

The lipase catalysed acylation of **1** by ethyl phenylacetate occurs with >99% preference for the *S*-enantiomer. The acylation of **1**, **2** and **3** by ethyl acetate is also *S*-selective, however a turnover-related racemisation of the product occurred affecting the *S*-enantiomer more than the *R*-enantiomer. A mechanism is proposed to account for the observed racemisation.

Ammoniolysis of amino acid derivatives

Lipase



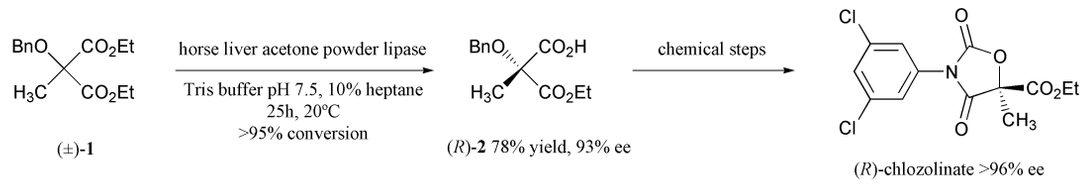
n = 1: *E* = 84

P. López-Serrano, M. A. Wegman, F. van Rantwijk and R. A. Sheldon, *Tetrahedron: Asymmetry*, 2001, **12**, 235.

Several lipases were screened for substrates with *n* = 1, 2 and 3. In the ammoniolysis of **1** by TIL a decrease in temperature to –20°C enhanced the enantioselectivity.

Chemoenzymatic asymmetric synthesis of (*R*)-chlozolinate

Lipase

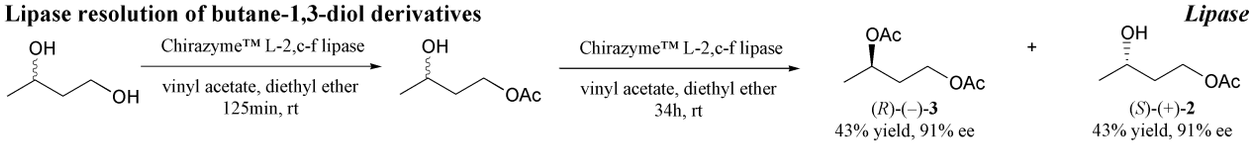


G. Guanti, L. Banfi, K. Powles, M. Rasparini, C. Scolastico and N. Fossati, *Tetrahedron: Asymmetry*, 2001, **12**, 271.

(*RS*)-Chlozolinate is used as an antifungal agent against *Botrytis cinerea* strains. The asymmetric synthesis was developed to ascertain the relative biological activities of its two enantiomers.

Lipase resolution of butane-1,3-diol derivatives

Lipase

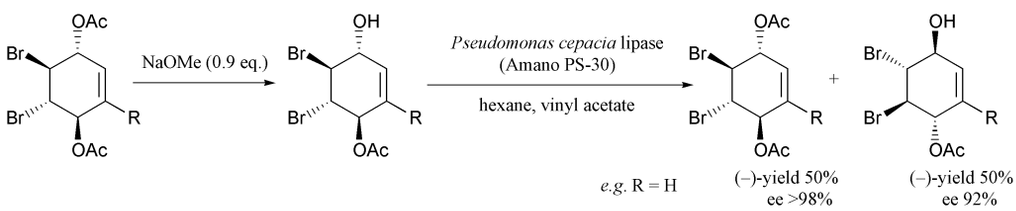


I. Izquierdo, M. T. Plaza, M. Rodríguez, J. A. Tamayo and A. Martos, *Tetrahedron: Asymmetry*, 2001, **12**, 293.

A new and simple procedure for the preparation of enantiopure samples of the major component of the pheromone bouquet of the common wasp.

Resolution of some *p*-benzoquinone derivatives

Lipase

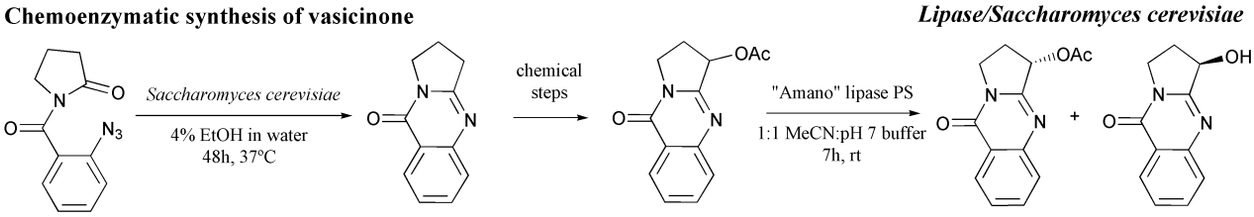


J. Gu, M. J. Heeg and C. R. Johnson, *Tetrahedron Lett.*, 2001, **42**, 1213.

Four substrates were subjected to esterification: R = H, Br, Me and CH_2OTBS. Highest yields and ee's were observed for R = H. For R = CH_2OTBS, *Candida antarctica* lipase B was found to be most effective, with product absolute configuration suspected to be (+). In each case, recrystallisation improved the ee.

Chemoenzymatic synthesis of vasicinone

Lipase/Saccharomyces cerevisiae

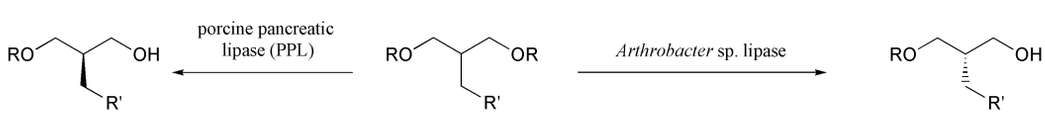


A. Kamal, K. V. Ramana and M. V. Rao, *J. Org. Chem.*, 2001, **66**, 997.

An enzymatic reductive cyclisation provides a method for the synthesis of the desired pyrrolo[2,1-*b*]quinazoline alkaloid skeleton, albeit in lower yields than the conventional chemical method. Lipase hydrolysis of ester **1** yields the target compound **2**. Chemical hydrolysis of **1** to compound **2** followed by lipase esterification was also studied.

A new esterase from *Arthrobacter* sp.

Esterase

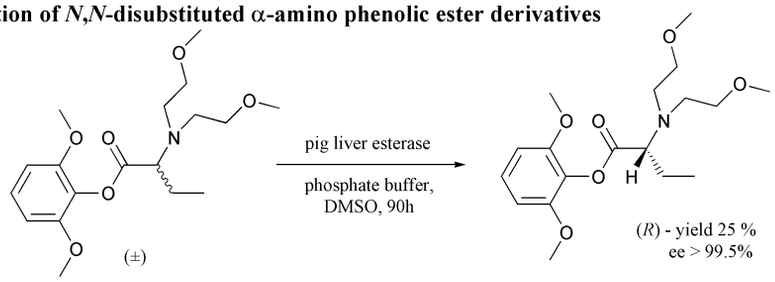


S. Johri, V. Verma, R. Parshad, S. Koul, S. C. Taneja and G. N. Qazi, *Bioorg. Med. Chem.*, 2001, **9**, 269.

The purification and characterisation of a novel esterase from *Arthrobacter* sp. RRLJ-1/95 was reported. The enzyme had a molecular weight of 32 kDa and a pH optimum of 8.5. A series of asymmetrisation reactions were performed with lypholised preparations of the enzyme which was shown to catalyse predominantly (*S*)-specific hydrolyses, enantiocomplementary to those catalysed by PPL.

Resolution of *N*,*N*-disubstituted α-amino phenolic ester derivatives

Esterase

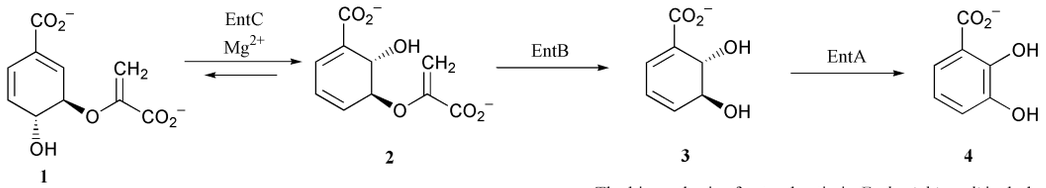


D. J. Bennett, K. I. Buchanan, A. Cooke, O. Epemolu, N. M. Hamilton, E. J. Hutchinson and A. Mitchell, *J. Chem. Soc., Perkin Trans. 1*, 2001, 362.

Two other substrates were also tested - one with a *p*-methyl group and one with ^{n}Pr in place of Et at the stereogenic carbon. The enantiomerically pure (*R*) products were of interest as anaesthetics.

Synthesis of cyclohexadiene-*trans*-diols

Recombinant Escherichia coli



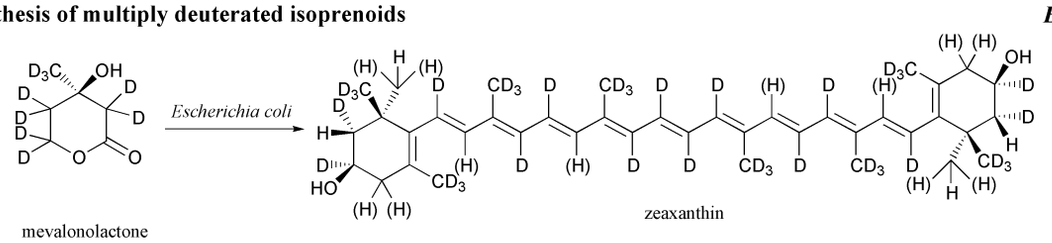
The biosynthesis of enterobactin in *Escherichia coli* includes the three step conversion of chorismate **1** to 2,3-dihydroxybenzoate **4** catalysed by the enzymes, EntA, EntB and EntC. Strains of *E. coli* have been engineered to over produce cyclohexadiene-*trans*-diols such as **3**, by transforming *E. coli* strains deficient in the aromatase EntA with plasmids containing EntB and EntC.

D. Franke, G. A. Sprenger and M. Muller, *Angew. Chem., Int. Ed.*, 2001, **40**, 555.

These strains produced 27 mg h^{-1} per g dry cell mass of **3.**

Synthesis of multiply deuterated isoprenoids

Escherichia coli

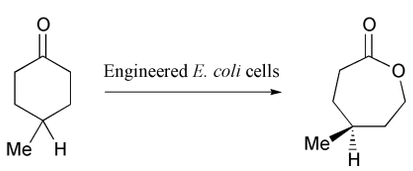


Using a strain of *E. coli* which had been triply engineered for a mevalonate biosynthetic pathway, fully deuterated mevalonolactone-d_9 was converted to zeaxanthin. No trace of nonlabelled zeaxanthin was produced. ^{1}H NMR studies revealed details of the biosynthetic pathway.

K. Kakinuma, Y. Dekishima, Y. Matsushima, T. Eguchi, N. Misawa, M. Takagi, T. Kuzuyama and H. Seto, *J. Am. Chem. Soc.*, 2001, **123**, 1238.

Asymmetric Baeyer–Villiger oxidations

Escherichia coli

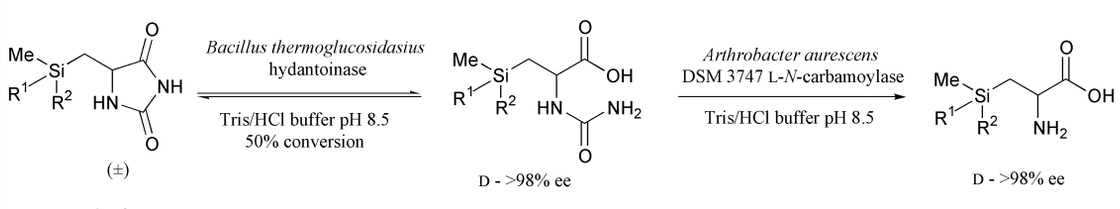


E. coli was engineered to overexpress cyclohexanone monooxygenase. The system was tested for its effectiveness in oxidising cyclohexanone substrates to the corresponding Baeyer–Villiger products. Eleven *meso* cyclohexanones with variation in the substituents at the 4 position were used, including some 4,4-dialkyl-substituted examples. Where applicable, the cyclohexanones which were known to be substrates for cyclohexanone monooxygenase from *S. cerevisiae* gave comparable yields and ee's using the *E. coli* cells. Yields varied from 54 to 91%, and ee's from 75% to ≥ 98%.

M. D. Mihovilovic, G. Chen, S. Wang, B. Kyte, F. Rochon, M. M. Kayser and J. D. Stewart , *J. Org. Chem.*, 2001, **66**, 733.

Deracemization of DL-5-silylmethylated hydantoins

Hydantoinase/carbamoylase

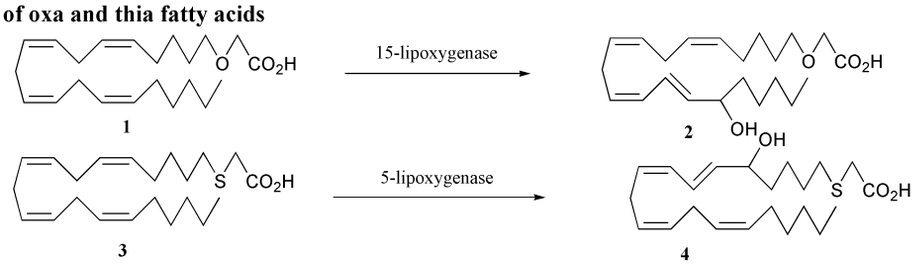


R. J. Smith, M. Pietzsch, T. Waniek, C. Syldatk and S. Bienz, *Tetrahedron: Asymmetry*, 2001, **12**, 157.

The use of hydantoinase from *A. aurescens* DSM 3745 gave similar enantioselectivities, however the extent of selectivity was substrate dependent.

Oxidation of oxa and thia fatty acids

Lipoxygenase

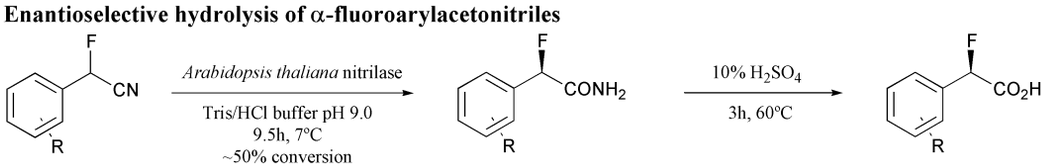


Analogues of arachidonic acid such as **1** and **3** have been oxidised using both 15- and 5-lipoxygenase. Substrates for 15-lipoxygenase were oxidised regioselectively at the ω-6 position (*e.g.* **2**). Some S oxidation was also observed. Oxidations by 5-lipoxygenase occurred at the carboxy end of the polyene moiety in most cases to yield products such as **4.**

C. J. Easton, T. A. Robertson, M. J. Pitt, D. A. Rathjen, A. Ferrante and A. Poulos, *Bioorg. Med. Chem.*, 2001, **9,** 317.

Enantioselective hydrolysis of α-fluoroarylacetonitriles

Nitrilase



(±)-**1**

a: R=H
b: R=*m*-Me
c: R=*m*-OMe

(*R*)-**2**

a: 72% yield, 92% ee
b: 61% yield, 88% ee
c: 67% yield, 91% ee

(*R*)-**3**

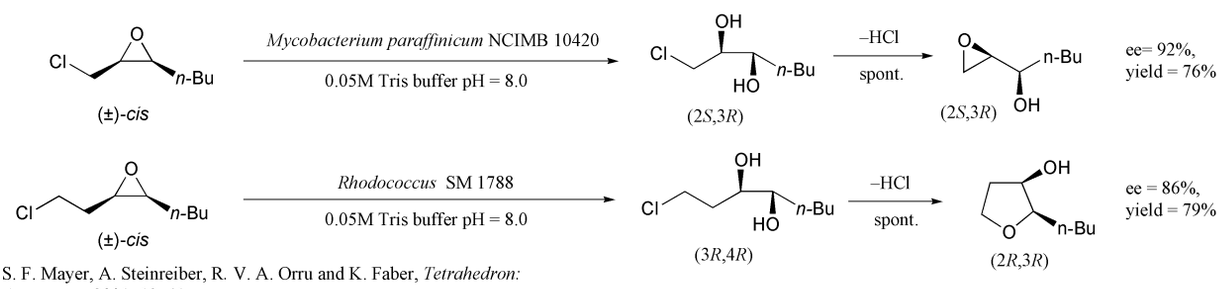
a: 91% yield, >99% ee
b: 90% yield, 98% ee
c: 88% yield, >99% ee

F. Effenberger and S. Oβwald, *Tetrahedron: Asymmetry*, 2001, **12**, 279.

Nitrilase catalysed hydrolysis shows advantage over chemical hydrolysis in that no decomposition products are observed. Virtually no enzymatic hydrolysis to the acid is observed under these conditions. Recrystallisation after enzymatic hydrolysis increases ee to >98%.

Enzymatic hydrolysis of (±)-2,3-disubstituted *cis*-chloroalkyl epoxides

Rhodococcus sp./Mycobacterium paraffinicum



(±)-*cis*

(2*S*,3*R*)

ee= 92%, yield = 76%

(±)-*cis*

(3*R*,4*R*)

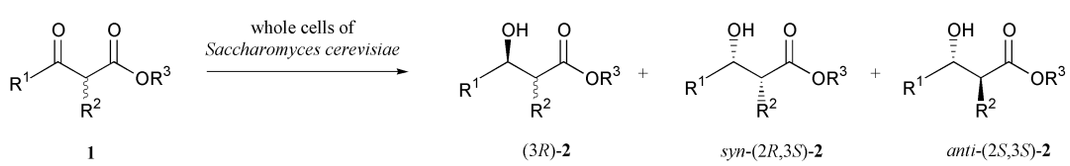
(2*R*,3*R*)

ee = 86%, yield = 79%

S. F. Mayer, A. Steinreiber, R. V. A. Orru and K. Faber, *Tetrahedron: Asymmetry*, 2001, **12**, 41.

Reductions of β-keto esters with genetically engineered yeast

Saccharomyces cerevisiae



1

(3*R*)-**2**

syn-(2*R*,3*S*)-**2**

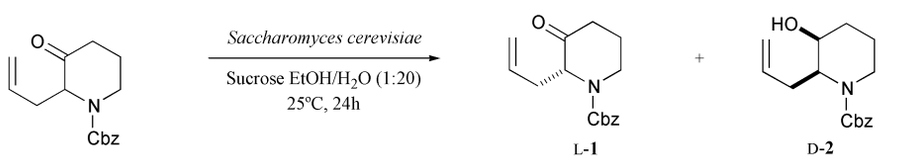
anti-(2*S*,3*S*)-**2**

Nine β-keto esters were tested, including 5 α-unsubstituted substrates. Initial modifications to the whole cell system (overexpression, or knockout of fatty acid synthase (Fasp), aldo-keto reductase (Ypr1p) or *α*-acetoxy ketone reductase (Gre2p)) resulted in changes to the stereoselectivity. These changes gave inconsistent results, but by combining gene alterations to target specific enantiomers resulted in high ee's (>98%), though in some instances the selectivity was opposite to expectations.

S. Rodríguez, M. M. Kayser and J. D. Stewart, *J. Am. Chem. Soc.*, 2001, **123**, 1547.

Yeast reduction of benzyl 2-allyl-3-oxopiperidinecarboxylate

Saccharomyces cerevisiae



DL -**1**

L-**1**
31% yield 93% ee

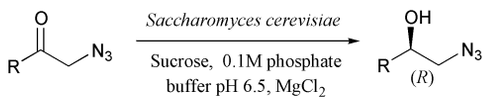
D-**2**
41% yield 97% ee

Y. Takeuchi, K. Azuma, K. Takakura, H. Abe, H.-S. Kim, Y. Wataya and T. Harayama, *Tetrahedron*, 2001, **57**, 1213.

The addition of K_2CO_3, to epimerize L-**1**, gave D-**2** in 62**%** yield (97% ee) and L-**1** in 14% yield (14% ee). (D)-Febrifugine was synthesized in 5 steps from D- **2**.

Stereoselective reduction of 2-azido-1-aryl ketones

Saccharomyces cerevisiae



Where R = *p*-ClC_6H_4 yield = 93%, ee =100%

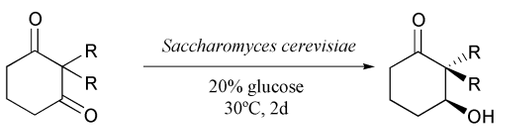
R = aryl, naphthyl or thienyl

J. S. Yadav, P. T. Reddy, S. Nanda and A. B. Rao, *Tetrahedron: Asymmetry*, 2001, **12**, 63.

A range of 2-azido-1-aryl ketones were reduced to the (*R*)-azido alcohols with yields in the range 88–94% and ee in the range 92–100%. The reduction of thiopenyl azidoketone gave the (*S*)-azido alcohol. The apparent enzyme kinetics were also reported.

Mono-reduction of cyclohexane-1,3-diones bearing two identical C(2) substituents

Saccharomyces cerevisiae



R = Me

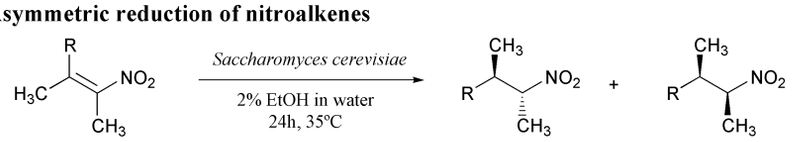
(*S*) - >96% ee, 48% yield

Z.-L. Wei, Z.-Y. Li and G.-Q. Lin, *Tetrahedron: Asymmetry*, 2001, **12**, 229.

Various R substituents were tested for reactivity. Reactivity increases along the series R = $CH_2CH_2CH_3$, $CH_2CH{=}CH_2$, $CH_2C{\equiv}CH$ and CH_3.

Asymmetric reduction of nitroalkenes

Saccharomyces cerevisiae



1a-e

a: R=Ph
b: R=Ph (*E*-isomer)
c: R=*o*-Cl-Ph
d: R=*m*-Cl-Ph
e: R=*p*-Cl-Ph

(2*R*,3*R*)-**2a-e** + (2*S*,3*R*)-**3a-e**

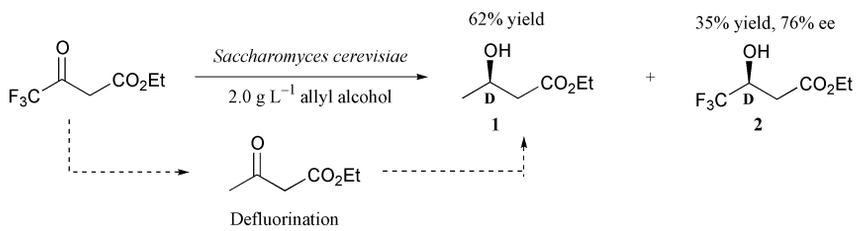
	Yield	de 2	ee 2	ee 3
a	72	20	98	97
b	54	19	87	83
c	n.r.	-	-	-
d	52	36	82	81
e	67	43	94	92

Y. Kawai, Y. Inaba and N. Tokitoh, *Tetrahedron: Asymmetry*, 2001, **12**, 309.

Enzymatic reduction of nitroalkenes by *Saccharomyces cerevisiae* is chemoselective, however the stereoselectivity is strongly influenced by the substitution pattern of the alkene. α,β-Disubstituted alkenes show very low selectivity whereas trisubstituted nitroalkenes show satisfactory stereoselectivity and modest diastereoselectivity.

Dehalogenation and enantioselective reduction of ethyl 4,4,4-trifluoroacetoacetate

Saccharomyces cerevisiae

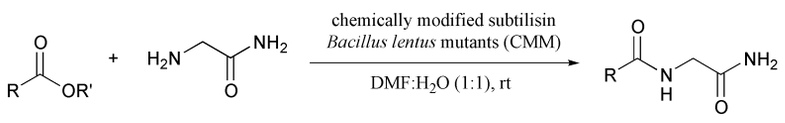


M. Bertau, *Tetrahedron Lett.*, 2001, **42**, 1267.

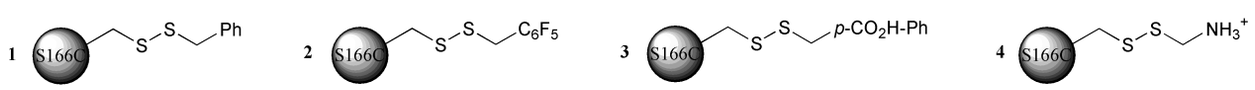
With 1.0 g l^{-1} allyl alcohol, **2** is formed in 39% yield and 28% ee with no defluorination detected. The defluorination occurs before keto reduction.

Expanding the utility of proteases in synthesis

Subtilisin



R= phenyllactic benzyl esters
mandelic acid benzyl esters
β-amino acid esters



K. Khumtaveeporn, A. Ullmann, K. Matsumoto, B. G. Davis and J. B. Jones, *Tetrahedron: Asymmetry*, 2001, **12**, 249.

Four CMM's (**1-4**) were identified as possible catalysts for amide formation reactions. The scope of the reaction was studied using 13 acyl donors and glycinamide and alaninamide as acyl acceptors. The stereospecificity of the catalysts was also examined. CMM's showed general improvement over subtilisin *Bacillus lentus* wild type enzyme.