Pergamon

Tetrahedron: *Asymmetry* 13 (2002) 211–221

TETRAHEDRON: *ASYMMETRY*

Synthesis of optically active 3-substituted-10-alkyl-10*H*-phenothiazine-5-oxides by enantioselective biotransformations

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Received 23 January 2002; accepted 14 February 2002

Abstract—A series of racemic 10-alkyl-3-formyl-10*H*-phenothiazine-5-oxides (±)-**2a–h** were subjected to biotransformation with baker's yeast resulting in optically active aldehydes (+)-**2a–h** and alcohols (−)-**3a–h** in moderate enantiomeric excess. The racemic 10-alkyl-3-hydroxymethyl-10*H*-phenothiazine-5-oxides (±)-**3a–h** and 3-acetoxymethyl-10-alkyl-10*H*-phenothiazine-5-oxides (±)-**4a–h** obtained from the racemic aldehydes (±)-**2a–h** were also tested in enantioselective lipase-catalyzed acetylations and alcoholysis reactions. The highest enantiomeric purities were achieved by a Novozyme 435-catalyzed acetylation–ethanolysis sequence, leading to optically active alcohols (−)-**3a–h** in 83–92% e.e. A novel NMR method using enantiopure dibenzoyl tartaric acid as chiral additive was developed for determination of the enantiomeric composition of the optically active products.

1. Introduction

Phenothiazines are important psychotropic compounds, but they also have further biological activities.[1–3] For example, phenothiazines have recently been considered as potential drugs in the management of Creutzfeldt-Jacob disease.[4]

Metabolism of phenothiazine-based drugs often results in the formation of 7-hydroxylated derivatives or 5-sulfoxides.[5–7] Because oxidation of asymmetrically substituted phenothiazines at the S(5) position introduces a new stereogenic center, these 5-sulfoxides are chiral. Although chiral 5-sulfoxide metabolites of the phenothiazine drug thioridazine in human plasma were separated by HPLC,[8] to date optically active phenothiazine 5-oxides have not been obtained on preparative scale. Hence, stereoselective methods for the synthesis of optically active phenothiazine-5-oxides would extend the possibilities for investigation of the *S*-oxide metabolites of phenothiazine-based drugs.

Recently, mild methods for *S*-oxidation of the 10-alkylphenothiazines **1a–h** were developed.[9] Vilsmeier–Haack formylation of 10-alkylphenothiazines followed by mild baker's yeast reduction of the 3-formyl-phenothiazine derivatives resulted in the smooth formation of 10-alkyl-3-hydroxy-methylphenthiazines.[10] These results inspired us to prepare racemic 10-alkyl-5-oxo-5,10-dihydro-5λ⁴-phenothiazine-3-carbaldehydes (±)-**2a–h**, (10-alkyl-5-oxo-5,10-dihydro-5λ⁴-phenothiazin-3-yl)-methanols (±)-**3a–h** and their acetates (±)-**4a–h** and study their enantioselective biotransformations (Fig. 1).

2. Results and discussion

Synthesis of the chiral racemic phenothiazine-3-carbaldehydes (±)-**2a–h** was straightforward, starting from the known 10-alkyl-10*H*-phenothiazine-3-carbaldehydes **1a–h**[10] as outlined in Fig. 1.

2.1. Baker's yeast reduction of 10-alkyl-5-oxo-5,10-dihydro-5λ⁴-phenothiazine-3-carbaldehydes (±)-2a–h

Because baker's yeast reduction of (10-alkyl-10*H*-phenothiazine-3-carbaldehydes **1a–h** proceeded easily,[10]

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0957-4166/02/$ - see front matter
PII: S0957-4166(02)00077-0

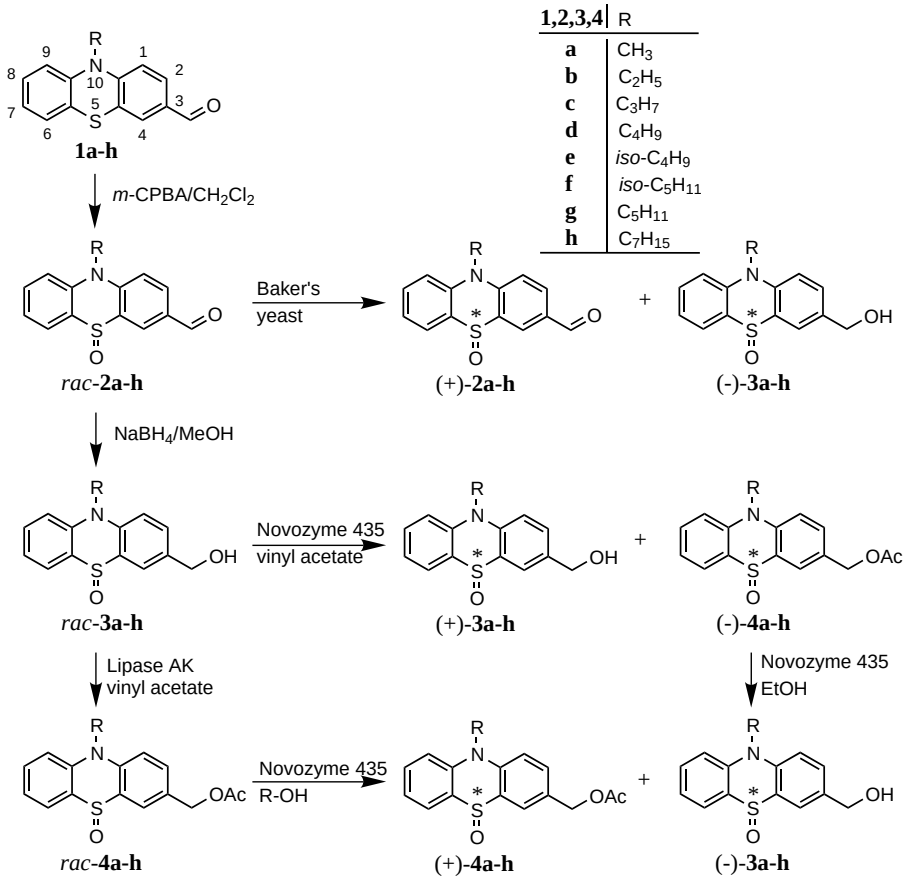

Figure 1. Preparation of optically active 3-substituted-10-alkylphenothiazine-5-oxides.

and enantioselective reduction of racemic aldehydes by fermenting baker's yeast were also reported,[11] the yeast-mediated reduction of the racemic sulfoxides (±)-**2a**–**h** was investigated first (Table 1). According to our expectation, the reduction with fermenting yeast proceeded smoothly. Unfortunately, this fast reduction exhibited only moderate to low enantiomer selectivities. The degree of this selectivity was dependent on the quality of the 10-alkyl moiety having a maxima at around the *N*-ethyl and *N*-propyl substituted aldehydes (±)-**2b** and **2c**. Increasing the bulk of the alkyl substituent gave almost racemic product in the reduction of *N*-(*iso*-butyl) aldehyde (±)-**2f**. Although the unreacted aldehyde fractions exhibited relatively large optical rotation values (e.g. +94.7 for (+)-**2b**; +113.4 for (+)-**2c**), the enantiomeric purities were only moderate (33% and 36% e.e., respectively).

Because it is known that the reduction conditions may strongly influence the outcome of the process,[11] the influence of the reaction conditions on the baker's yeast reduction was investigated using the *N*-propyl deriva-

Table 1. Baker's yeast reduction of 10-alkyl-10*H*-phenothiazine-3-carbaldehyde 5-oxides (±)-**2a**–**h** under fermenting conditions[a]

Starting compound	Time (h)	(+)-**2a**–**h**			(−)-**3a**–**h**			E_p[c]
(±)-**2a**–**h**		Yield (%)	$[\alpha]_D$[b]	E.e. (%)[c]	Yield (%)	$[\alpha]_D$[b]	E.e. (%)[c]	
a	2.0	54	+44	15	39	−6.8	20	1.7
b	1.8	32	+94.7	33	68	−7.1	23	2.4
c	2.0	24	+113.4	36	72	−6.2	17	2.2
d	2.5	37	+56.5	21	63	−3.5	11	1.5
e	2.0	40	+26.2	10	50	−7.0	21	1.9
f	2.0	68	~0	<3	29	~0	<3	1.0
g	2.0	20	+34.2	14	56	−4.0	25	2.2
h	2.0	33	+15.3	5	50	−2.8	10	1.4

[a] For conditions see Section 4.5.1.

[b] Optical rotations measurements were obtained at 25°C (*c* 1, $CHCl_3$).

[c] Enantiomeric excess was determined by ^1H NMR (c.f. Section 4.1.4); Measure of selectivity:[10] $E_p = \ln[1-c(1+\text{e.e.}(P))]/\ln[1-c(1-\text{e.e.}(P))]$.

tive (±)-**2c** as test substrate (Table 2). Shortening the reaction time (entry 1) or using Mg^{2+} (a known selectivity modifier for baker's yeast reductions) gave no significant change in selectivity (entry 4). In some cases (entries 2 and 3) both the selectivity and conversion of the reduction decreased.

Although the reduction performed without addition of sugar in a water–hexane two-phase system (entry 5) afforded somewhat higher selectivity than fermenting conditions (Table 1), other biotransformations were investigated in an attempt to enhance the enantiomeric purity of the chiral phenothiazine-5-oxide derivatives (see Section 2.3).

2.2. NMR method using dibenzoyl tartaric acid as chiral additive for determination of the enantiomeric composition of the products

Our first efforts for determination of the enantiomeric composition of the optically active products by chromatographic methods (e.g. HPLC or GLC on cyclodextrin-based chiral phases) were unsuccessful. Because NMR spectroscopy provides a range of powerful methods, which are complementary to chromatographic or electrophoretic approaches,[13] NMR using chiral additives was tried next. Although the use of chiral shift reagents or solvents in NMR are well established methods for determination of the enantiomeric composition of various compounds,[13,14] they require expensive enantiopure additives. Since a large number of enantiomeric excess determinations were needed in our study, we investigated enantiodiscrimination towards our optically active products using easily available enantiopure compounds as additives in ^{1}H NMR. The pure enantiomers of *O*,*O*′-dibenzoyl tartaric acid (DBTA) have been widely used for resolution of racemic amines by diastereomeric salt formation.[14] A report on enantiomer separation of non-basic calix[4]arenes by HPLC with addition of D-(−)-tartaric acid[15] indicated, that not only ionic but hydrogen bonded diastereomeric associates can be formed between solutes and tartaric acid derivatives. ^{1}H NMR was used to determine the equilibrium constants for formation of the diastereomeric complexes of the enantiomers of DBTA with amide type chiral selectors[16,17] or to study chiral recognition of tartaric acid derivatives with chiral receptors.[18] These data prompted us to develop a novel method for the enantiomeric excess determination of our optically active phenothiazine-5-oxides **2**,**3** and **4a**–**h** by (−)-dibenzoyl tartaric acid [(−)-DBTA] as a chiral additive in NMR.

The initial red color which formed on mixing the $CDCl_3$ solutions of the phenothiazine-5-oxides and DBTA (c.f. Section 4.1.4) but disappeared on heating the mixture might be related to formation of charge transfer type complexes. Disappearance of the color can be rationalized by assuming equilibration towards thermodynamically more stable hydrogen bonded/ionic complexes.

The well separated signals (c.f. Section 4.1.4) observed in the ^{1}H NMR spectra allowed not only assessment of the enantiomeric composition of a single optically active compound but simultaneous determination of conversion and enantiomeric compositions of the product and the remaining substrate in our bioconversions (see examples shown in Fig. 2). It is noteworthy that in some cases measurement was repeated with (+)-DBTA resulting in reversal of the signals of the enantiomers in question.

2.3. Lipase-catalyzed acylation of (10-alkyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methanols (±)-3a–h

It is well documented, that lipases are versatile biocatalysts for the enantioselective biotransformations of various racemic alcohols.[11,19] A large number of lipases having a wide substrate specificity range and selectivity are commercially available. Although no report on hydrolase-catalyzed biotransformation of phenothiazine sulfoxides could be found, enzymatic methods are known for kinetic resolution of simpler optically active sulfoxides with shorter distances between sulfoxide and ester moieties.[20] Since $NaBH_4$ reduction of the racemic 10-alkyl-5-oxo-5,10-dihydro-5λ^4-phenothiazine-3-carbaldehydes (±)-**2a**–**h** resulted in smooth formation of the corresponding (phenothiazin-3-yl)methanols (±)-**3a**–**h**, the lipase-mediated enantioselective acylation of these racemic alcohols was also tested. First, the selectivity of acetylations catalyzed by various lipases was tested in anhydrous THF with vinyl acetate using (10-propyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methanol (±)-**3c** as substrate (Table 3.).

All of the lipases in our study accepted the test substrate (±)-**3c** as alcohol in the irreversible transesterifica-

Table 2. Influence of conditions on the baker's yeast-mediated reduction of 10-propyl-10*H*-phenothiazine-3-carbaldehyde 5-oxide (±)-**2c**[a]

Entry	Additive(s) (amount)	Time (h)	Conv. (%)[b]	(+)-**2c** E.e.%[b]	(−)-**3c** E.e.%[b]
1	–	0.5	23	<3	20
2	L-Cysteine (0.5%)	2	35	<3	14
3	Ethyl chloroacetate (0.5%)	2	7	<3	17
4	$MgCl_2$ (2%)	2	64	13	<3
5[c]	Hexane (250 mL)	2	76	26	20

[a] In a system (see Section 4.5.2) containing the additive(s).
[b] Conversion and enantiomeric excess by ^{1}H NMR (c.f. Section 4.1.4).
[c] Without saccharose.

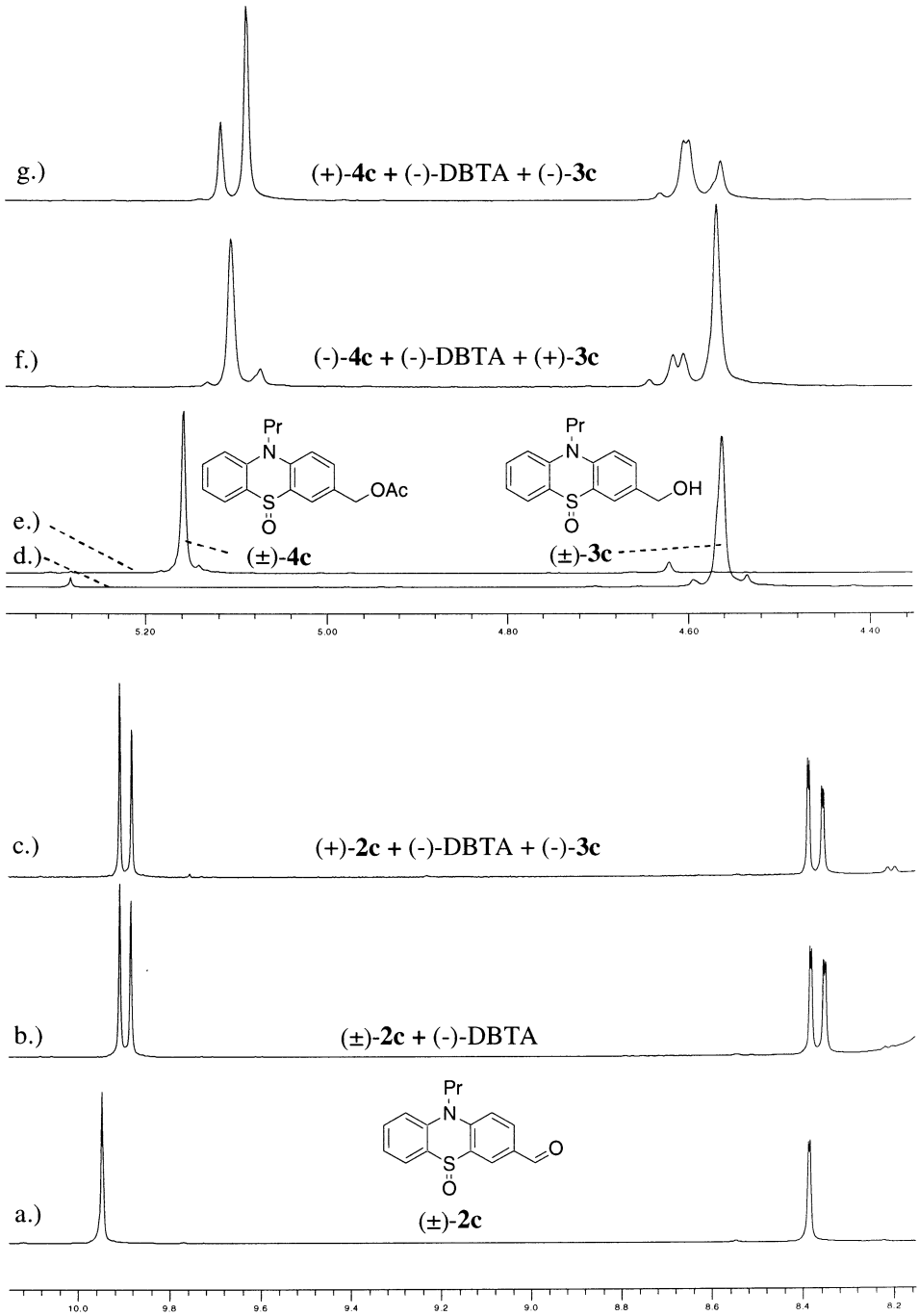

Figure 2. Determination of enantiomeric composition of the optically active 3-substituted-phenothiazine-5-oxides by ^{1}H NMR (a) (±)-**2c**; (b) (±)-**2c** in presence of (−)-DBTA; (c) (Baker's yeast reduction of (±)-**2c** in the presence of L-cysteine)+(−)-DBTA; (d) (±)-**3c**; (e) (±)-**4c**; (f) (Novozyme 435-mediated acetylation of (±)-**3c** in acetone)+(−)-DBTA; (g) (Novozyme 435 mediated ethanolysis of (±)-**4c**)+(−)-DBTA.

tion of vinyl acetate under anhydrous conditions (entries 1–12, Table 3). The tested lipases, however, showed a wide variety of reactivity and enantiomer selectivity. While Lipase AK exhibited excellent reactivity without apparent selectivity (entry 2, Table 3) and thus resulted in a fast and mild formation of the racemic acetate (±)-**4c**, most of the lipases showed low to moderate enantioselectivity and reactivity (entries 1, 3–11, Table 3).

Since the best selectivity was observed with Novozyme 435™ (entry 10, Table 3), this lipase was selected for further studies with the whole series of the racemic (10-alkyl-phenothiazin-3-yl)methanols (±)-**3a**–**h** (Table 4).

The Novozyme 435-catalyzed acetylation reaction with vinyl acetate proceeded fast, almost independently from the size of the alkyl substituents in the racemic (10-alkyl-phenothiazin-3-yl)methanols (±)-**3a**–**h**.

Table 3. Enantioselective acetylation of (10-propyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methanol (±)-**3c** mediated by lipases[a]

Entry	Enzyme (amount mg)	Time (h)	Conv. (%)[b]	(+)-**3c** E.e. (%)[b]	(−)-**4c** (%)[b]
1	Lipase A (100)	72	17	<3	<3
2	Lipase AK (100)	0.08	100	–	<3
3	Lipase AY (100)	90	12	<3	<3
4	Lipase G (100)	72	18	<3	<3
5	Lipase M (100)	144	34	4	7
6	Lipase N (100)	72	15	<3	<3
7	Lipase PS (100)	0.25	12	3	4
8	Lipase R (100)	72	21	<3	<3
9	Lipozyme IM 20 (100)	0.25	65	8	12
10	Novozyme 435 (100)	0.42	47	36	43
11	PPL (100)	92	27	4	3

[a] For conditions see Section 4.6.1.
[b] Conversion and enantiomeric excess by ^{1}H NMR (c.f. Section 4.1.4)

Table 4. Enantioselective acetylation of (10-alkyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methanols (±)-**3a**–**h** mediated by Novozyme 435[a]

Starting compound[a] (±)-**3a**–**h**	Time (min)	Yield (%)	(+)-**3a**–**h** $[\alpha]_D$[b]	E.e. (%) [c]	Yield (%)	(−)-**4a**–**h** $[\alpha]_D$[b]	E.e. (%) [c]	E_p[c]
a	13	47	+17.8	40	49	−24.8	43	3.8
b	18	22	+18.8	42	57	−24.6	33	4.9
c	15	21	+16.7	39	70	−15.1	23	3.3
d	20	42	+15.3	38	57	−28.0	44	4.5
e	10	43	+10.5	25	54	−14.8	25	2.2
f	10	51	+13.5	43	49	−22.3	44	3.8
g	10	73	+6.50	15	22	−28.2	47	3.2
h	10	49	+19.1	47	49	−17.5	46	4.2

[a] For conditions see Section 4.6.2.
[b] Optical rotations were measured at 25°C (*c* 1, $CHCl_3$).
[c] Enantiomeric excess was determined by ^{1}H NMR (c.f. Section 4.1.4); measure of selectivity:[12] $E_p = \ln[1-c(1+\text{e.e.}(P))]/\ln[1-c(1-\text{e.e.}(P))]$.

The selectivity varied only slightly depending on the steric properties of the *N*-alkyl moiety. Although the degree of enantioselectivity increased slightly compared to the baker's yeast-mediated reduction (from $E=1.0$–2.4 to 2.2–4.9), it was still insufficient for obtaining products with e.e. of over 80%.

Because significant dependence on the solvent enhancement of the enantioselectivity was observed in lipase-catalyzed acylations in many cases,[11,19] the Novozyme 435-mediated acetylation was carried out in different solvents using the *N*-propyl compound (±)-**3c** as a test substrate (Table 5).

The cost of the slight increase in selectivity (from $E=3.3$ to 4.3–4.5) in pyridine or acetone (entries 2 and 4, Table 5, respectively), however, was a significant drop in the rate of acetylation.

2.4. Lipase-catalyzed alcoholysis of (10-alkyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methyl acetates 4a–h

It is usual that enantiopreference in the acylation and

Table 5. Solvent effect on Novozyme 435-mediated acetylation of (10-propyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)-methanol (±)-**3c**[a]

Entry	Solvent	Time (min)	Conv. (%)[b]	(+)-**3c** E.e. (%)[b]	(−)-**4c** E.e. (%)[b]
1	Dichloromethane	25	48	29	33
2	Pyridine	240	63	49	36
3	Chloroform	30	46	16	23
4	*t*-Butanol	120	31	13	33
5	Acetone	240	54	43	46
6	Tetrahydrofuran	15	73	39	23

[a] For conditions see Section 4.6.3.
[b] Conversion and enantiomeric excess by ^{1}H NMR (c.f. Section 4.1.4).

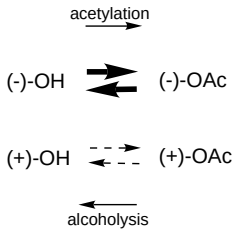

Figure 3. Enantiopreferences in enzymic acylation versus alcoholysis.

alcoholysis/hydrolysis reactions of the same enzyme remain unaltered[11,19] (Fig. 3.)

Because the selectivity towards the same enantiomer might be similar or even higher in alcoholysis than in acetylation, these reactions were also studied using the *N*-propyl (phenothiazin-3-yl)methyl acetate (±)-**4c** as test substrate (Table 6.).

According to the expectations, the enantiopreference in alcoholysis remained unaltered compared to that found in acetylation, and thus the alcoholysis/hydrolysis of the (−)-acetate [(−)-**4c**] was always preferred. A strong maxima—exhibiting the highest enantioselectivity ($E=$ 10.5) so far—was found using ethanol as nucleophile in alcoholysis (entry 2, Table 6).

Because the (−)-enantiomer was the faster reacting in both the acetylation and ethanolysis reactions ((−)-**3c** and (−)-**4c**, respectively) their selectivities can be combined in a sequential acetylation–ethanolysis process. Accordingly, the Novozyme 435-catalyzed ethanolysis of the partially resolved acetates (−)-**4a**–**h** (prepared by Novozyme 435-mediated acetylation, c.f. Section 4.6.1 and Table 4) yielded the optically active (−)-(10-alkyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methanols (−)-**3a**–**h** in enhanced enantiomeric excess (Table 7.).

The high enantiomeric excess of the producing (−)-alcohols [(−)-**3a**–**h**, 83–92% e.e.] indicated that consecutive utilization of reactions having moderate enantioselectivities towards the same enantiomeric forms can provide the desired products in high enantiomeric purity.

3. Conclusion

Stereoselective synthetic methods have been elaborated for preparation of the optically active 3-substituted-10-alkyl-10*H*-phenothiazine-5-oxides **2**,**3** and **4a**–**h** for the first time. Among the biotransformations studied, the Novozyme 435-catalyzed acylation with vinyl acetate combined with Novozyme 435-catalyzed alcoholysis in ethanol resulted in the highest enantiomeric purities.

A convenient new NMR method using (−)-dibenzoyl tartaric acid as chiral additive was developed for determination of the enantiomeric composition of the optically active products **2**,**3** and **4a**–**h** even from unseparated reaction mixtures.

Table 7. Novozyme 435-catalyzed ethanolysis of partially resolved (10-alkyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methyl acetates, (−)-**4a**–**h**

Acetate[a] (−)-**4a**–**h**	Time (min)	Conversion (%)[b]	(−)-**3a**–**h** E.e. (%)[b]
a	120	30	87
b	120	33	89
c	120	26	83
d	120	25	86
e	120	39	85
f	120	24	89
g	120	25	92
h	120	22	89

[a] The partially resolved acetates (−)-**4a**–**h** were prepared according to Section 4.6.1. For ethanolysis conditions see Section 4.7.2.
[b] Conversion and enantiomeric excess by ^{1}H NMR (c.f. Section 4.1.4)

4. Experimental

4.1. Materials and methods

4.1.1. Reagents and solvents. *m*-CPBA, $NaBH_4$, vinyl acetate and solvents were Aldrich or Fluka products. All solvents were purified and dried by standard methods as required.

4.1.2. Biocatalysts. Baker's yeast produced as wet cakes by Budafok Ltd., Hungary was from a local store. Lipase A, Lipase AK, Lipase AY, Lipase G, Lipase AK, Lipase M, Lipase N, Lipase PS and Lipase R were gifts from Amano Europe, England. Novozyme 435™ and Lipozyme IM 20™ were gifts from Novo Nordisk, Denmark. Lipase from porcine pancreas (PPL) was obtained from Sigma.

Table 6. Novozyme 435-catalyzed alcoholyses of (10-propyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methyl acetate (±)-**4c** in different media[a]

Entry	Nucleophile	Time (h)	Conv. (%)[b]	(−)-**3c** E.e. (%)[b]	(+)-**4c** E.e. (%)[b]	E_P[b]
1	MeOH	18	12	63	5	1.1
2	EtOH	18	45	75	71	10.5
3	PrOH	18	40	30	22	1.8
4	BuOH	18	30	40	11	1.3
5	H_2O/acetone (1:4, v:v)	18	15	39	5	1.1

[a] For conditions see Section 4.7.1.
[b] By ^{1}H NMR (c.f. Section 4.1.4); $E_p=\ln[1-c(1+ee(P))]/\ln[1-c(1-ee(P))]$[12]

4.1.3. Analytical methods. The ^1H and ^{13}C NMR spectra were recorded in $CDCl_3$ solution on a Brucker DRX-500 spectrometer operating at 500 and 125 MHz, respectively. Chemical shifts are expressed in ppm values from TMS as internal standard. In ^1H NMR measurements, long relaxation time ($D_1 = 20$ s) was applied for enhancing the accuracy of the integration. IR spectra were recorded in KBr on a Specord 2000 spectrometer. Mass spectra were taken on a VG QUATTROO mass spectrometer in M+H+ES and ES modes. TLC was carried out using Merck Kieselgel $60F_{254}$ sheets. Spots were visualized by treatment with 5% ethanolic phosphomolybdic acid solution and heating. Preparative chromatographic separations were performed using vacuum chromatography[21] on Merck Kieselgel 60 (0.063–0.200 μm). Melting points were determined by hot plate method and are uncorrected. Optical rotations were determined on a Perkin–Elmer 241 polarimeter.

4.1.4. Determination of enantiomeric excess of the products by NMR using (+)- or (−)-dibenzoyl tartaric acid as chiral additive. Enantiomeric composition of the optically active products was determined by ^1H NMR in $CDCl_3$ using (−)-dibenzoyl tartaric acid as chiral additive. For the measurements the sample (~50 mg, compounds **2**,**3** and **4a**–**h** either as separated compounds or even as unseparated reaction mixtures) and 1 molar equivalent (−)-dibenzoyl tartaric acid (~55–65 mg) were separately dissolved in $CDCl_3$ (~0.5 mL, each). The combined solutions were mixed and heated (~55°C) in a closed NMR tube until the red color—which usually appeared on mixing—disappeared (15–30 min) and then the ^1H NMR spectra was recorded.

Significant splitting in the presence of (−)-DBTA was seen for several signals [phenothiazine-3-carbaldehydes, **2a**–**h**: aldehyde singlet at 9.95–10.24 ppm and aromatic doublet at around 8.39–8.46 ppm; (phenothiazin-3-yl)methanols **3a**–**h**: the Ar-C$\underline{H}_2$-O singlet at 4.56–4.66 ppm; (phenothiazin-3-yl)methyl acetates **4a**–**h**: the Ar-C$\underline{H}_2$-OAc singlet at 5.13–5.29 ppm]. Precise integration of the signals showing significant enantiomer splitting in the presence of (−)-DBTA allowed the calculation of the enantiomeric compositions (and the chemical conversion) for the samples. In the case of ambiguous integration results, measurement was repeated with the same amount of (+)-DBTA resulting in reversal of the signals of the enantiomers in question.

Illustrative examples of simultaneous determination of enantiomeric compositions/conversion for the optically active phenothiazine derivatives **2**,**3** and **4a**–**h** are shown in Fig. 2.

4.2. *S*-Oxidation of 10-alkyl-10*H*-phenothiazine-3-carbaldehydes 1a–h with *m*-CPBA

To a stirred solution of 10-alkyl-10*H*-phenothiazine-3-carbaldehyde (**1a**–**h**, 5 mmol) in dichloromethane (25 mL), a solution of *meta*-chloroperoxybenzoic acid (5 mmol, ~1.23 g, ~70%, freshly titrated iodometrically) dissolved in anhydrous dichloromethane (20 mL) was added dropwise over 30 minutes at 0–5°C. The reaction was completed within 2–4 h (by TLC). The reaction mixture was washed with 10% potassium hydroxide solution (15 mL, twice), 5% HCl solution (10 mL), and saturated $NaHCO_3$ solution (10 mL) and dried over $MgSO_4$. After the solvent was distilled off, the product was isolated from the residue by column chromatography on silica gel with dichloromethane:acetone 9:1. Finally the resulting sulfoxides (±)-**2a**–**h** were recrystallized from hexane.

4.2.1. 10-Methyl-5-oxo-5,10-dihydro-5λ^4-phenothiazine-3-carbaldehyde (±)-2a. Yield: 82%. Mp: 207–208°C; ^1H NMR: 7.38 (1H, t), 7.50 (2H, m), 7.71 (1H, t), 7.99 (1H, t), 8.14 (1H, dd), 8.44 (1H, d), 10.02 (1H, s); ^{13}C NMR: 36.20, 116.34, 116.42, 123.58, 124.96, 125.27, 130.39, 131.28, 132.86, 133.47, 134.73, 139.16, 143.95, 189.61; IR: 1680, 1624, 1600, 1584, 1464, 1384, 1352, 1200, 1160, 1128, 1048, 1024, 752; MS: 259, 258, 243, 242, 241, 227, 226, 214, 213, 212, 200, 199, 198. Anal. calcd for $C_{14}H_{11}NO_2S$: C, 65.35; H, 4.31; N, 5.44; S, 12.46. Found: C, 65.28; H, 4.21; S, 12.43%.

4.2.2. 10-Ethyl-5-oxo-5,10-dihydro-5λ^4-phenothiazine-3-carbaldehyde (±)-2b. Yield: 84%. Mp: 204°C; ^1H NMR: 1.63 (3H, t), 4.42 (2H, q), 7.37 (1H, t), 7.55 (2H, m), 7.71 (1H, t), 7.99 (1H, t), 8.14 (1H, dd), 8.45 (1H, d), 10.01 (1H, s); ^{13}C NMR: 12.61, 43.86, 116.47, 116.57, 123.72, 124.31, 124.82, 130.42, 132.36, 133.00, 133.87, 136.02, 137.68, 142.48, 189.75; IR: 1676, 1628, 1600, 1584, 1468, 1392, 1368, 1200, 1164, 1136, 1048, 1024, 760; MS: 273, 272, 257, 256, 255, 240, 229, 228, 227, 226, 200, 199, 198. Anal. calcd for $C_{15}H_{13}NO_2S$: C, 66.40; H, 4.83; N, 5.16; S, 11.82. Found: C, 66.35; H, 4.79; N, 5.18; S, 11.76%.

4.2.3. 10-Propyl-5-oxo-5,10-dihydro-5λ^4-phenothiazine-3-carbaldehyde (±)-2c. Yield: 92%. Mp: 158°C; ^1H NMR: 1.10 (3H, t), 2.00 (2H, m), 4.19 (2H, t), 7.25 (1H, t), 7.46 (2H, m), 7.64 (1H, t), 7.93 (1H, t), 8.07 (1H, dd), 8.39 (1H, s), 9.95 (1H, s); ^{13}C NMR: 11.42, 20.26, 50.63, 116.73, 116.84, 123.73, 124.53, 125.04, 130.42, 132.18, 132.91, 133.76, 135.83, 137.95, 142.79, 189.74; IR: 1684, 1628, 1608, 1588, 1472, 1440, 1392, 1360, 1224, 1164, 1108, 1048, 1016, 752; MS: 287, 286, 271, 270, 269, 243, 241, 240, 227, 226. Anal. calcd for $C_{16}H_{15}NO_2S$: C, 67.34; H, 5.30; N, 4.91; S, 11.23. Found: C, 67.39; H, 5.36; N, 4.98; S, 11.23%.

4.2.4. 10-Butyl-5-oxo-5,10-dihydro-5λ^4-phenothiazine-3-carbaldehyde (±)-2d. Yield: 89%. Mp: 134–135°C; ^1H NMR: 1.09 (3H, t), 1.60 (2H, m), 1.98 (2H, m), 4.30 (2H, t), 7.37 (1H, t), 7.53 (2H, t), 7.71 (1H, t), 8.00 (1H, d), 8.14 (1H, d), 8.45 (1H, d), 10.01 (1H, s); ^{13}C NMR: 14.17, 28.86, 48.89, 116.68, 116.79, 123.72, 124.53, 125.04, 130.40, 132.21, 132.92, 133.78, 135.87, 137.94, 142.77, 189.73; IR: 1688, 1636, 1600, 1584, 1464, 1368, 1336, 1228, 1176, 1072, 1048, 1020, 764; MS: 301, 300, 285, 284, 283, 282, 242, 241, 240, 227, 226. Anal. calcd for $C_{17}H_{17}NO_2S$: C, 68.20; H, 5.7; N, 4.68; S, 10.71. Found: C; 68.28; H, 5.68; N, 4.65; S, 10.78%.

4.2.5. 10-(3-Methylpropyl)-5-oxo-5,10-dihydro-5λ^4-phenothiazine-3-carbaldehyde (±)-2e. Yield: 95%. Mp: 125–127°C; ^1H NMR: 0.99 (6H, d), 2.34 (1H, m), 4.20 (2H,

t), 7.35 (1H, t), 7.57 (2H, m), 7.66 (1H, t), 7.96 (1H, t), 8.10 (1H, dd), 8.41 (1H, d), 10.24 (1H, s); ^{13}C NMR: 20.49, 27.64, 54.90, 117.59, 117.74, 123.84, 126.27, 126.71, 130.58, 131.26, 132.45, 133.22, 134.97, 138.88, 143.97, 189.77; IR: 1680, 1628, 1604, 1584, 1464, 1384, 1352, 1204, 1144, 1104, 1072, 1032, 756; MS: 301, 300, 285, 284, 283, 241, 240. Anal. calcd for $C_{17}H_{17}NO_2S$: C, 68.20; H, 5.72; N, 4.68; S, 10.71. Found: C, 68.24; H, 5.68; N, 4.59; S, 10.65%.

4.2.6. 10-(3-Methylbutyl)-5-oxo-5,10-dihydro-5λ^4-phenothiazine-3-carbaldehyde (±)-2f. Yield: 92%. Mp: 138–139°C; ^{1}H NMR: 1.12 (6H, d), 1.89 (3H, m), 4.33 (2H, t), 7.37 (1H, t), 7.53 (2H, m), 7.71 (1H, t), 8.00 (1H, d), 8.15 (1H, dd), 8.45 (1H, d), 10.01 (1H, s); ^{13}C NMR: 22.88, 26.99, 27.01, 35.22, 47.75, 116.57, 116.67, 123.70, 124.60, 125.11, 130.40, 132.24, 132.93, 133.77, 135.88, 137.88, 142.68, 189.73; IR: 1688, 1636, 1608, 1600, 1584, 1464, 1384, 1316, 1208, 1168, 1156, 1108, 1052, 1020, 752; MS: 315, 314, 299, 298, 297, 241, 240, 227, 226. Anal. calcd for $C_{18}H_{19}NO_2S$: C, 68.98; H, 6.11; N, 4.47; S, 10.23. Found: C, 68.95; H, 6.09; N, 4.42; S, 10.31%.

4.2.7. 10-Pentyl-5-oxo-5,10-dihydro-5λ^4-phenothiazine-3-carbaldehyde (±)-2g. Yield: 91%. Mp: 146–147°C; ^{1}H NMR: 1.10 (3H, t), 1.57–1.47 (4H, m), 2.00 (2H, m), 4.29 (2H, t), 7.37 (1H, t), 7.52 (2H, m), 7.71 (1H, t), 8.00 (1H, d), 8.14 (1H, dd), 8.45 (1H, d), 10.01 (1H, s); ^{13}C NMR: 14.44, 22.77, 26.55, 29.28, 49.14, 116.64, 116.74, 123.70, 124.57, 125.09, 130.41, 132.21, 132.90, 133.76, 135.88, 137.90, 142.72, 189.73; IR: 1688, 1648, 1608, 1600, 1584, 1464, 1368, 1328, 1228, 1196, 1156, 1144, 1048, 1032, 756; MS: 315, 314, 299, 298, 297, 241, 240, 227, 226. Anal. calcd for $C_{18}H_{19}NO_2S$: C, 68.98; H, 6.11; N, 4.47; S, 10.23. Found: C, 68.88; H, 6.12; N, 4.40; S, 10.28%.

4.2.8. 10-Heptyl-5-oxo-5,10-dihydro-5λ^4-phenothiazine-3-carbaldehyde (±)-2h. Yield: 93%. Mp: 144–145°C; ^{1}H NMR: 0.95 (3H, t), 1.46– 1.28 (4H, m), 1.57 (2H, m), 1.60 (2H, m), 2.01 (2H, m), 4.30 (2H, t), 7.38 (1H, t), 7.53 (2H, m), 7.72 (1H, d), 8.01 (1H, d), 8.15 (1H, dd), 8.46 (1H, d), 10.02 (1H, s); ^{13}C NMR: 14.45, 22.96, 26.85, 27.18, 29.31, 32.15, 49.21, 116.65, 116.74, 123.70, 124.53, 125.05, 130.42, 132.24, 132.89, 133.77, 135.94, 137. 92, 142.73, 189.72; IR: 1688, 1636, 1608, 1600, 1588, 1464, 1368, 1328, 1272, 1200, 1144, 1073, 1032, 756; MS: 343, 342, 327, 326, 325, 240, 227, 226. Anal. calcd for $C_{20}H_{23}NO_2S$: C, 70.35; H, 6.79; N, 4.10; S, 9.39. Found: C, 70.25; H, 6.71; N, 4.18; S, 9.42%.

4.3. Reduction of 10-alkyl-5-oxo-5,10-dihydro-5λ^4-phenothiazine-3-carbaldehydes (±)-2a–h with sodium borohydride

The substrate (±)-**2a**–**h**, 0.2 g) was dissolved in dry methanol (5 mL) and $NaBH_4$ (0.1 g) was added portionwise to the stirred solution at room temperature and stirring was continued for 1 h. The reaction was quenched by dropwise addition of 2N HCl and the resulting mixture was evaporated to dryness. The residue was extracted with water–dichloromethane (1:2). The organic layer was separated and dried over $MgSO_4$. After removing the solvent the residual crude (10-alkyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methanols (±)-**3a**–**h** were purified by column chromatography on silica gel using toluene–acetone (9:1) and finally recrystallized from toluene.

4.3.1. (10-Methyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methanol (±)-3a. Yield: 85%. Mp: 187°C; ^{1}H NMR: 2.73 (1H, OH), 3.75 (3H, s), 4.66 (2H, s), 7.26 (1H, m), 7.36 (2H, m), 7.57 (1H, m), 7.63 (1H, m), 7.85 (1H, d), 7.91 (1H, m); ^{13}C NMR: 35.44, 63.81, 115.59, 115.70, 121.88, 124.01, 124.15, 129.24, 131.11, 131.72, 132.95, 135.28, 130.10, 139.91; IR: 3296, 1592, 1468, 1344, 1264, 1192, 1128, 1048, 992, 760; MS: 261, 260, 255, 246, 245, 244, 243, 242, 230, 229, 228, 226, 214, 213, 212, 200, 199, 198. Anal. calcd for $C_{14}H_{13}NO_2S$: C, 64.84; H, 5.05; N, 5.40; S, 12.34. Found: C, 64.82; H, 5.42; S, 12.43%.

4.3.2. (10-Ethyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methanol (±)-3b. Yield: 89%. Mp: 147–149°C; ^{1}H NMR: 1.56 (3H, t), 3.02 (1H, OH), 4.33 (2H, q), 4.64 (2H, s), 7.25 (1H, m), 7.43 (2H, m), 7.62 (2H, m), 7.87 (1H, s), 8.00 (1H, d); ^{13}C NMR: 12.46, 30.09, 43.19, 64.08, 115.91, 116.05, 122.09, 123.45, 123.62, 130.28, 130.41, 132.35, 133.50, 135.54, 137.65, 138.49; IR: 3248, 1592, 1468, 1368, 1256, 1208, 1136, 1048, 996, 752; MS: 275, 274, 259, 258, 257, 256, 242, 229, 228, 226. Anal. calcd for $C_{15}H_{15}NO_2S$: C, 65.91; H, 5.53; N, 5.12; S, 11.73. Found: C, 65.88; H, 5.61; S, 11.82%.

4.3.3. (10-Propyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methanol (±)-3c. Yield: 94%. Mp: 118–120°C; ^{1}H NMR: 1.09 (3H, t), 1.93 (2H, m), 3.41 (1H, OH), 4.13 (2H, t), 4.56 (2H, s), 7.21 (1H, t), 7.2–7.38 (2H, m), 7.55 (1H, d), 7.60 (1H, t), 7.82 (1H, t), 7.89 (1H, d); ^{13}C NMR: 10.99, 19.56, 46.64, 63.37, 115.68, 115.77, 121.59, 123.00, 123.22, 129.71, 131.72, 131.82, 132.97, 135.32, 137.35, 138.31; IR: 3320, 1592, 1464, 1368, 1256, 1208, 1144, 1048, 1000, 752; MS: 289, 288, 273, 272, 271, 270, 243, 242, 240, 229, 228. Anal. calcd for $C_{16}H_{17}NO_2S$: C, 66.87; H, 5.96; N, 4.87; S, 11.13. Found: C, 68.85; H, 5.90; N, 4.88; S, 11.23%.

4.3.4. (10-Butyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methanol (±)-3d. Yield: 95%. Mp: 114–117°C; ^{1}H NMR: 1.04 (3H, t), 1.55 (2H, m), 2.16 (2H, m), 3.16 (1H, OH), 4.18 (2H, t), 4.58 (2H, s), 7.22 (1H, t), 7.34–7.40 (2H, m), 7.55/7.63 (2H, m), 7.82 (1H, d), 7.91 (1H, d); ^{13}C NMR: 13.77, 20.01, 28.21, 47.86, 63.44, 115.74, 121.59, 123.12, 123.33, 129.76, 131.75, 131.82, 132.97, 135.25, 137.34, 138.28; IR: 3376, 1592, 1464, 1376, 1248, 1184, 1144, 1048, 996, 744; MS: 303, 302, 287, 286, 285, 284, 243, 242, 240, 229, 228. Anal. calcd for $C_{17}H_{19}NO_2S$: C, 67.75; H, 6.35; N, 4.65; S, 10.64. Found: C, 67.81; H, 6.32; N, 4.68; S, 10.56%.

4.3.5. [10-(3-Methylpropyl)-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl]methanol (±)-3e. Yield: 92%. Mp: 65–66°C; ^{1}H NMR: 0.93 (6H, d), 2.30 (1H, m), 2.89 (1H, OH), 4.07 (2H, t), 4.60 (2H, s), 7.21 (1H, t), 7.38–7.45 (2H, m), 7.51–7.58 (2H, m), 7.81 (1H, d), 7.87 (1H, d); ^{13}C NMR: 20.14, 26.99, 53.94, 63.65, 116.65, 116.77,

121.84, 125.07, 125.25, 129.02, 130.92, 131.32, 132.48, 135.33, 138.49, 139.38; IR: 3360, 1592, 1464, 1360, 1248, 1176, 1144, 1096, 1008, 756; MS: 303, 302, 300, 286, 285, 284. Anal. calcd for $C_{17}H_{19}NO_2S$: C, 67.75; H, 6.35; N, 4.65; S, 10.64. Found: C, 67.79; H, 6.38; N, 4.60; S, 10.58%.

4.3.6. [10-(3-Methylbutyl)-5-oxo-5,10-dihydro-5λ⁴-phenothiazin-3-yl]methanol (±)-3f. Yield: 92%. Mp: 146–147°C; ^{1}H NMR: 1.08 (6H, d), 1.84–2.05 (3H, m), 3.18 (1H, OH), 4.31 (2H, t), 4.60 (2H, s), 7.23 (1H, d), 7.38–7.41 (2H, m), 7.58–7.62 (2H, m), 7.84 (1H, d), 7.91 (1H, d); ^{13}C NMR: 22.54, 26.58, 34.57, 46.73, 63.50, 115.63, 115.65, 121.61, 123.41, 123.89, 129.83, 131.81, 131.87, 133.01, 135.25, 137.28, 138.20; IR: 3336, 1592, 1464, 1368, 1264, 1208, 1096, 1048, 1008, 752; MS: 317, 316, 302, 301, 300, 299, 298, 243, 242, 230, 229, 228. Anal. calcd for $C_{18}H_{21}NO_2S$: C, 68.54; H, 6.71; N, 4.44; S, 10.16. Found: C, 68.62; H, 6.68; N, 4.48; S, 10.25.

4.3.7. (10-Pentyl-5-oxo-5,10-dihydro-5λ⁴-phenothiazin-3-yl)methanol (±)-3g. Yield: 90%, semisolid; ^{1}H NMR: 0.99 (3H, t), 1.28 (2H, m), 1.47 (4H, m), 1.95 (2H, m), 3.16 (1H, OH), 4.19 (2H, t), 4.63 (2H, s), 7.23 (1H, t), 7.40 (2H, m), 7.62 (2H, m), 7.92 (1H, d), 7.99 (1H, d); ^{13}C NMR: 14.08, 22.41, 25.93, 28.97, 48.26, 63.66, 115.66, 115.83, 121.66, 123.34, 123.66, 129.90, 131.84, 131.95, 133.05, 135.17, 137.40, 138.27; IR: 3392, 1592, 1464, 1372, 1260, 1188, 1144, 1048, 1008, 760; MS: 317, 316, 301, 300, 299, 298, 243, 242, 229, 228. Anal. calcd for $C_{18}H_{21}NO_2S$: C, 68.54; H, 6.71; N, 4.44; S, 10.16. Found: C, 68.58; H, 6.78; N, 4.42; S, 10.20%.

4.3.8. (10-Heptyl-5-oxo-5,10-dihydro-5λ⁴-phenothiazin-3-yl)methanol (±)-3h. Yield: 93%. Mp: 103–104°C; ^{1}H NMR: 0.93 (3H, t), 1.35 (4H, m), 1.40 (2H, m), 1.51 (2H, m), 1.94 (2H, m), 3.14 (1H, OH), 4.18 (2H, t), 4.61 (2H, s); ^{13}C NMR: 14.06, 22.56, 26.20, 26.79, 28.95, 31.78, 48.21, 63.54, 115.61, 115.71, 121.60, 123.19, 123.39, 129.83, 131.81, 132.96, 135.21, 135.27, 137.36, 138.27; IR: 3328, 1592, 1472, 1376, 1248, 1196, 1144, 1036, 760; MS: 387, 386, 370, 369, 368, 328, 327, 326. Anal. calcd for $C_{20}H_{25}NO_2S$: C, 69.94; H, 7.34; N, 4.08; S, 9.33. Found: C, 69.85; H, 7.31; N, 4.08; S, 9.43%.

4.4. Acylation of (10-alkyl-5-oxo-5,10-dihydro-5λ⁴-phenothiazin-3-yl)methanols (±)-3a–h with vinyl acetate by a non-stereoselective lipase

To a solution of racemic (10-alkyl-5-oxo-5,10-dihydro-5λ⁴-phenothiazin-3-yl)-methanol (±)-**3a**–**h**, 200 mg, each) in anhydrous THF (6 mL) Lipase AK (200 mg) and vinyl acetate (2 mL) were added and the mixture was stirred at room temperature for 1 h. Then the enzyme was filtered off and washed with acetone (10 mL). Solvents were distilled off from the combined filtrate, the residue was purified by column chromatography on silica gel with dichloromethane and finally recrystallized from hexane.

4.4.1. (10-Methyl-5-oxo-5,10-dihydro-5λ⁴-phenothiazin-3-yl)methyl acetate (±)-4a. Yield: 92%. Mp: 148°C; ^{1}H NMR: 2.01 (3H, s), 3.83 (3H, s), 5.29 (2H, s), 7.26 (1H, t), 7.31 (2H, d), 7.49 (2H, m), 7.86 (2H, m); ^{13}C NMR: 20.95, 35.43, 65.13, 115.53, 115.60, 115.72, 122.05, 124.38, 129.61, 130.98, 131.05, 131.12, 132.93, 133.13, 139.67, 170.78; IR: 1740, 1592, 1468, 1356, 1232, 1048, 1024, 756; MS: 303, 302, 287, 286, 285, 271, 270, 245, 244, 243, 242, 241, 228, 226, 225, 214, 213, 195, 194. Anal. calcd for $C_{16}H_{15}NO_3S$: C, 63.77; H, 5.02; N, 4.65; S, 10.64. Found: C, 63.75; H, 5.11; N, 4.68; S, 10.62%.

4.4.2. (10-Ethyl-5-oxo-5,10-dihydro-5λ⁴-phenothiazin-3-yl)methyl acetate (±)-4b. Yield: 95%. Mp: 121°C; ^{1}H NMR: 1.58 (3H, t), 2.11 (3H, s), 4.37 (2H, q), 5.18 (2H, s), 7.29 (1H, t), 7.49 (2H, m), 7.65 (2H, m), 7.97 (2H, d); ^{13}C NMR:11.96, 20.94, 42.74, 65.09, 115.50, 115.64, 121.62, 121.85, 123.73, 123.77, 129.36, 131.80, 131.92, 133.01, 133.22, 137.84, 170.75; IR: 1736, 1592, 1448, 1368, 1224, 1048, 1024, 752; MS: 318, 316, 300, 299, 284, 272, 271, 270, 258, 257, 256, 240, 239, 228, 227, 211, 208. Anal. calcd for $C_{17}H_{17}NO_3S$: C, 64.74; H, 5.43; N, 4.44; S, 10.17. Found: C, 64.75; H, 5.41; N, 4.48; S, 10.13%.

4.4.3. (10-Propyl-5-oxo-5,10-dihydro-5λ⁴-phenothiazin-3-yl)methyl acetate (±)-4c. Yield: 95%. Mp: 133°C; ^{1}H NMR: 1.11 (3H, t), 1.95 (2H, m), 2.08 (3H, s), 4.32 (2H, t), 5.16 (2H, s), 7.25 (1H, t), 7.42 (2H, m), 7.63 (2H, d), 7.95 (2H, m); ^{13}C NMR: 10.99, 19.57, 20.94, 49.68, 65.09, 115.76, 115.91, 121.60, 121.89, 123.77, 129.40, 129.85, 131.65, 131.78, 132.97, 133.18, 138.16, 170.78; IR: 1736, 1592, 1464, 1456, 1368, 1228, 1044, 1020, 744; MS: 331, 330, 314, 313, 312, 272, 271, 270. Anal. calcd for $C_{18}H_{19}NO_3S$: C, 65.63; H, 5.81; N, 4.25; S, 9.73; Found: C, 65.58; H, 6.79; N, 4.28; S, 9.81%.

4.4.4. (10-Butyl-5-oxo-5,10-dihydro-5λ⁴-phenothiazin-3-yl)methyl acetate (±)-4d. Yield: 91%. Mp: 98°C; ^{1}H NMR: 1.04 (3H, t), 1.54 (2H, m), 1.91 (2H, m), 2.09 (3H, s), 4.21 (2H, t), 5.13 (2H, s), 7.23 (1H, t), 7.61 (2H, m), 7.63 (2H, m), 7.94 (2H, m); ^{13}C NMR: 13.72, 19.96, 20.91, 28.19, 47.81, 65.07, 115.68, 115.83, 121.83, 123.92, 123.96, 129.34, 129.82, 131.62, 131.75, 132.88, 133.12, 138.10, 170.71; IR: 1736, 1592, 1464, 1372, 1240, 1048, 1024, 764; MS: 345, 344, 329, 328, 327, 326, 286, 285, 284, 271, 270, 230, 228, 212, 211. Anal. calcd for $C_{19}H_{21}NO_3S$: C, 66.45; H, 6.16; N, 4.08; S, 9.33; Found: C, 66.38; H, 6.11; N, 4.18; S, 9.43%.

4.4.5. [10-(3-Methylpropyl)-5-oxo-5,10-dihydro-5λ⁴-phenothiazin-3-yl]methyl acetate (±)-4e. Yield: 92%. Mp: 78°C; ^{1}H NMR: 0.94 (6H, d), 2.09 (3H, s), 2.29 (1H, m), 4.09 (2H, d), 5.15 (2H, s), 7.24 (1H, t), 7.56 (2H, t), 7.59 (2H, m), 7.91 (2H, m); ^{13}C NMR: 20.12, 20.99, 26.99, 54.02, 65.17, 116.70, 116.83, 122.10, 125.67, 129.59, 130.78, 130.98, 132.45, 132.72, 139.24, 145.53, 145.97, 170.83; IR: 1736, 1592, 1464, 1360, 1228, 1028, 756; MS: 345, 344, 286, 285, 284, 229, 228, 212, 211. Anal. calcd for $C_{19}H_{21}NO_3S$: C, 66.45; H, 6.16; N, 4.08 S, 9.33; Found. C, 66.38; H, 6.21; N, 4.11; S, 9.38%.

4.4.6. [10-(3-Methylbutyl)-5-oxo-5,10-dihydro-5λ⁴-phenothiazin-3-yl]methyl acetate (±)-4f. Yield: 93%, semisolid; ^{1}H NMR: 1.10 (3H, t), 1.29–1.45 (4H, m), 1.76–1.83 (2H, m), 2.10 (3H, s), 4.31 (2H, t), 5.18 (2H, s), 7.26

(1H, t), 7.42 (2H, t), 7.65 (2H, m), 7.96 (2H, m); ^{13}C NMR: 20.96, 22.50, 26.57, 34.59, 46.74, 65.12, 115.61, 115.76, 121.88, 123.95, 124.00, 125.83, 129.39, 131.72, 131.86, 132.96, 133.19, 138.09, 170.77; IR: 1732, 1592, 1472, 1360, 1248, 1052, 1020, 760; MS: 359, 358, 342, 341, 340, 300, 299, 298, 284, 229, 228. Anal. calcd for $C_{20}H_{23}NO_3S$: C, 67.20; H, 6.49; N, 3.92 S, 8.97. Found: C, 67.25; H, 6.41; N, 3.98; S, 8.93%.

4.4.7. (10-Pentyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methyl acetate (±)-4g. Yield: 90%. Mp: 105–106°C; ^{1}H NMR: 1.01 (3H, t), 1.36–1.50 (4H, m), 1.76–1.83 (2H, m), 2.08 (3H, s), 4.22 (2H, t), 5.18 (2H, s), 7.26 (1H, t), 7.42 (2H, t), 7.65 (2H, d), 7.95 (2H, d); ^{13}C NMR: 14.04, 20.98, 22.38, 25.90, 28.92, 30.89, 48.20, 65.14, 115.69, 115.84, 121.90, 123.95, 125.48, 129.12, 129.40, 131.72, 131.85, 132.97, 133.20, 138.13, 170.80; IR: 1740, 1592, 1464, 1364, 1228, 1032, 1020, 752; MS: 359, 358, 342, 341, 340, 300, 299, 298, 284, 229, 228. Anal. calcd for $C_{20}H_{23}NO_3S$: C, 67.20; H, 6.49; N, 3.92; S, 8.97. Found: C, 67.19; H, 6.44; N, 3.88; S, 8.89.

4.4.8. (10-Heptyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methyl acetate (±)-4h. Yield: 89%; semisolid; ^{1}H NMR: 0.93 (3H, t), 1.26–1.37 (4H, m), 1.44 (2H, m), 1.95 (2H, m), 2.08 (3H, s), 2.12 (2H, m), 4.22 (2H, t), 5.18 (2H, s), 7.27 (1H, t), 7.63 (2H, t), 7.65 (2H, m), 7.96 (2H, m); ^{13}C NMR: 14.02, 20.94, 22.52, 26.19, 26.75, 28.90, 31.73, 48.21, 65.11, 115.68, 115.82, 121.87, 123.83, 123.88, 125.42, 129.38, 131.69, 131.81, 132.95, 133.17, 138.12, 170.76; IR: 1740, 1592, 1468, 1364, 1228, 1032, 756; MS: 387, 386, 370, 369, 368, 328, 327, 326. Anal. calcd for $C_{22}H_{27}NO_3S$: C, 68.54; H, 7.06; N, 3.63; S, 9.35. Found: C, 68.44; H, 7.13; N, 3.73; S, 9.39%.

4.5. Enantioselective reduction of 10-alkyl-5-oxo-5,10-dihydro-5λ^4-phenothiazine-3-carbaldehydes (±)-2a–h by baker's yeast

4.5.1. Reduction of 10-alkyl-5-oxo-5,10-dihydro-5λ^4-phenothiazine-3-carbaldehydes (±)-2a–h with fermenting baker's yeast. Ethanolic solution of 10-alkyl-5-oxo-5,10-dihydro-5λ^4-phenothiazine-3-carbaldehyde (±)-**2a**–**h**, (250 mg/25 mL) was added to a suspension of fresh baker's yeast (25 g) and saccharose (25 g) in water (150 mL). The mixture was stirred (250 rpm) at room temperature and the reaction was monitored by TLC. At 30–70% conversion level, the mixture was extracted with dichloromethane (300 mL) and the separated organic layer was dried over $MgSO_4$. After distilling off the solvent in vacuum, the residue was separated by column chromatography on silica gel (dichloromethane:acetone 9:1) resulting optically active (+)-**2a**–**h** and (−)-**3a**–**h** products. Reaction time, yield, enantiomeric excess and specific rotation data for the products are given in Table 3.

4.5.2. Reduction of the 10-propyl-5-oxo-5,10-dihydro-5λ^4-phenothiazine-3-carbaldehyde (±)-2c with baker's yeast under different conditions. A suspension of fresh baker's yeast (25 g), saccharose (25 g) and water (250 mL) was stirred at room temperature (at 250 rpm) for 15 min. and then additive(s) (as indicated in Table 2) followed by 10-propyl-5-oxo-5,10-dihydro-5λ^4-phenothiazine-3-carbaldehyde (±)-**2c** (250 mg) in ethanol (25 mL) (after further 15 min) were added and the reaction was monitored by TLC. After the time indicated in Table 2, the mixture was extracted with dichloromethane (300 mL). The further work up was performed as described in Section 4.5.1. Reaction conditions, conversion and enantiomeric excess data for the resulting (+)-**2c** and (−)-**3c** products are given in Table 2.

4.6. Lipase-mediated enantioselective acylation of (10-alkyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)-methanols (±)-3a–h with vinyl acetate

4.6.1. Acylation of (10-propyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methanol (±)-3c mediated by different lipases. To a solution of racemic (10-propyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)-methanol (±)-**3c**, 100 mg) in anhydrous THF (3 mL) vinyl acetate (1 mL, 12.45 mmol) and lipase (see Table 3) were added and the reaction mixture was stirred at room temperature. The enzyme was filtered off and washed with acetone (10 mL). Solvents were distilled off from the filtrate and the residue was analyzed with ^{1}H NMR in the presence of (−)-DBTA. Reaction conditions, conversion and enantiomeric excess data for the products [(+)-**3c** and (−)-**4c**] are given in Table 3.

4.6.2. Enantioselective acylation (10-alkyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methanols (±)-3a–h mediated by Novozyme 435. To a solution of racemic (10-alkyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)-methanol (±)-**3a**–**h** (100 mg) in anhydrous THF (3 mL) vinyl acetate (1 mL, 12.45 mmol) and Novozyme 435™ (100 mg) were added and the reaction mixture was stirred at room temperature. Then the enzyme was filtered off and washed with acetone (10 mL). Solvents were distilled off from the filtrate and the residue was separated by column chromatography on silica gel with dichloromethane. Reaction conditions, yield, specific rotation and enantiomeric excess data for the resulting products [(+)-**3a**–**h** and (−)-**4a**–**h**] are given in Table 4.

4.6.3. Acylation of racemic (10-propyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methanol (±)-3c mediated by Novozyme 435 in different solvents. To a solution of racemic (10-propyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)-methanol (±)-**3c** (100 mg) in the corresponding solvent (3 mL) vinyl acetate (1 mL, 12.45 mmol) and Novozyme 435™ (100 mg) were added and the reaction mixture was stirred at room temperature. Work up and analysis of the products were performed according to Section 4.6.1. Conditions, conversion and enantiomeric excess data for the products [(+)-**3c** and (−)-**4c**] are given in Table 5.

4.7. Novozyme 435-mediated enantioselective alcoholysis of (10-alkyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)-methyl acetates (4a–h)

4.7.1. Novozyme 435-catalyzed alcoholysis of (10-propyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methyl acetate (±)-4c in different media. To a solution of racemic

(10-propyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)-methyl acetate (±)-**4c** (40 mg) in the corresponding alcohol or in water–acetone mixture (2 mL), was added Novozyme 435 (20 mg) and the reaction mixture was stirred at room temperature overnight. The enzyme was filtered off and washed with acetone (5 mL). The solvents were distilled off from the filtrate and the residue was analyzed with ^{1}H NMR in the presence of (-)-DBTA. Conversion and enantiomeric excess data for the products [(−)-**3c** and (+)-**4c**] are given in Table 6.

4.7.2. Novozyme 435-catalyzed ethanolysis of (−)-(10-alkyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methyl acetates [(−)-4a–h]. Novozyme 435 (20 mg) was added to an ethanolic solution (10 mL) of (−)-(10-alkyl-5-oxo-5,10-dihydro-5λ^4-phenothiazin-3-yl)methyl acetate [(−)-**4a**–**h**, 40 mg, obtained as described in Section 4.6.3] and the resulting mixture was stirred at room temperature for 2 h. The enzyme was filtered off and washed with acetone (5 mL). Work up was performed according to Section 4.7.1. Conversion and enantiomeric excess data for the resulting optically active products [(−)-**3a**–**h**] are given in Table 7.

Acknowledgements

Financial support for the Hungarian OTKA Foundation (T-033112) is gratefully acknowledged. Cs.P. and C.M. thank Domus Hungarica Foundation for short term fellowships.

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