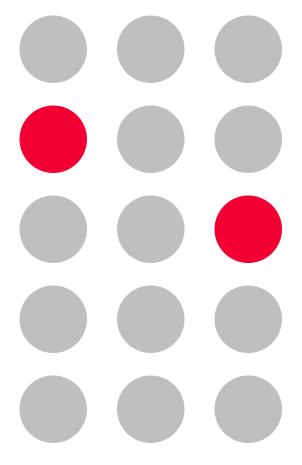


evozymes[®] catalog 10/11





¬ content

catalog issue 01_2010

about evocatal	3
evozymes overview	4
enzyme data sheets	6
orders and inquiries	35
contact information	35
order form	36
terms and conditions	37

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BLZ 300 501 10

BIC DUSSDEDD

IBAN DE38 3005 0110 1004 5299 78



¬ about evocatal

company profile

evocatal was founded in 2006 and has since flourished into a leading partner in the field of enzyme technology. The company name connects the terms 'evolution' and 'catalysis' and thus stands for our core competencies: The tailor-made development of new enzymes and processes for the chemical and pharmaceutical industry. Our goal is to provide new and improved enzymes especially for stereoselective syntheses and to market enzyme-based processes and products.

Furthermore we have strong technology platforms in the fields of enzyme discovery, enzyme expression and secretion by which we support our customers either in contract research projects or by granting licenses. Please contact us for further details about our technologies and services.

evocatal also coordinates various public research projects contributing to the generation of new insights into enzyme technology and the optimization of biocatalytic processes. We're continuously striving to play a leading role as partner for the scientific community.

This catalog gives an overview about our commercial enzymes that we offer from stock. Each enzyme is available in sample quantities as indicated in the price list on pages 4 and 5 as well as in bulk quantities – please contact us any time for further details or information.

We're looking forward to your request,

the evocatal team

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¬ evozymes overview

evozyme		characteristics	catalog-no.	quantity	price	page
Alcohol Dehydrogenases		Overview on page 7	for bulk qu	antities, ple	ase inq	uire!
ADH Screening Kit	NEW!	12 ADH enzymes plus cofactor regeneration system	evo-1.1.100	12 ADHs	1490€	6
ADH 010		(S)-selective, NAD-dependent, recombinant from <i>E. coli</i>	evo-1.1.010	10 kU 100 kU	275 € 550 €	8
ADH 020		(S)-selective, NAD-dependent, recombinant from <i>E. coli</i>	evo-1.1.020	1 kU 5 kU	110€ 275€	9
ADH 030		(S)-selective, NAD-dependent recombinant from <i>E. coli</i>	evo-1.1.030	50 U 200 U	275 € 450 €	10
ADH 040		(S)-selective, NAD-dependent recombinant from <i>E. coli</i>	evo-1.1.040	200 U 1 kU	190€ 450€	11
ADH 130		D-selective, NAD-dependent, recombinant from E. coli	evo-1.1.130	1 kU 10 kU 1 MU	110 € 275 € 550 €	12
ADH 140		L-selective, NAD-dependent, recombinant from E. coli	evo-1.1.140	1 kU 10 kU 100 kU	110 € 275 € 550 €	13
ADH 200	NEW!	(<i>R</i>)-selective , NAD-dependent, recombinant from E. coli	evo-1.1.200	200 U 500 U	275 € 390 €	14
ADH 210 Isoenzyme E from Horse Liver (HLADH)		NAD-dependent, recombinant from <i>E. coli</i>	evo-1.1.210	300 U 1 kU	190€ 520€	15
ADH 250	NEW!	(S)-selective, NADP-dependent, recombinant from <i>E. coli</i>	evo-1.1.250	300 U 1 kU	190€ 450€	16
ADH 260	NEW!	thermostable, NADP-dependent, recombinant from <i>E. coli</i>	evo-1.1.260	500 U 2 kU	190 € 450 €	17
ADH 270 from <i>Lactobacillus kefir</i> (LkADH)	NEW!	(R)-selective, NADP-dependent, recombinant from <i>E. coli</i>	evo-1.1.270	300 U 1 kU	190 € 450 €	18
ADH 190		D-selective, NAD-dependent, recombinant from <i>E. coli</i>	evo-1.1.190	50 U 500 U	110€ 450€	19
Lactate Dehydrogenase 150		L-selective, NAD-dependent, recombinant from <i>E.coli</i>	evo-1.1.150	1 kU 10 kU 100 kU	110 € 275 € 550 €	20

for bulk quantities please inquire!



¬ evozymes overview

evozyme	characteristics	catalog-no.	quantity	price p	age
Cofactor Regenerating Enzymes	Overview on page 21				
Glucose Dehydrogenase (GDH)	NAD(P)-dependent, recombinant from <i>E. coli</i>	evo-1.1.060	1 kU 10 kU 100 kU	110 € 275 € 550 €	22
Formate Dehydrogenase (FDH) NEW!	NAD-dependent, recombinant from <i>E. coli</i>	evo-1.1.230	50 U 100 U	275 € 450 €	23
Isocitrate Dehydrogenase	NAD-dependent, recombinant from <i>E. coli</i>	evo-1.1.070	50 U 100 U	275 € 450 €	24
Malate Dehydrogenase 080 (decarboxylating)	NADP-dependent, bacterial	evo-1.1.080	200 U 1 kU	110 € 275 €	25
Malate Dehydrogenase 090 (decarboxylating)	NAD-dependent, bacterial	evo-1.1.090	200 U 1 kU	110 € 275 €	26
Dehydrogenases					
Leucine Dehydrogenase 110	NAD-dependent, bacterial L-selective	evo-1.1.110	200 U 1 kU	110 € 275 €	27
Alanine Dehydrogenase 160	NAD-dependent, bacterial L-selective	evo-1.1.160	100 U 1 kU	110€ 250€	28
Hydroxynitrilases		for bulk quant	tities please	inquire!	
Hydroxynitrile Lyase 4.010 (AtHNL)	(R)-selective eukaryotic (A. thaliana) recombinant from E. coli	evo-1.4.010	200 U 1 kU	110 € 275 €	29
Hydroxynitrile Lyase 4.020 NEW! (MeHNL)	(S)-selective eukaryotic (<i>Manihot exculenta</i>) recombinant from <i>E. coli</i>	evo-1.4.020	200 U 1 kU	110€ 275€	31
Lipases					
Lipase Screening Kit	35 Lipases for screening applications recombinant from <i>E. coli</i>	evo-1.3.100	please	inquire	32
soon available: Transaminase Screening Kit	6 novel Transaminases for screening applications recombinant from <i>E. coli</i>	evo-1.2.100	please	inquire	34



¬ Alcohol Dehydrogenase Kit

evo-1.1.100

11 Alcohol Dehydrogenases – Just add substrate!

Kit Description

The evocatal Alcohol Dehydrogenase Kit comprises 12 lyophilized preparations of different alcohol dehydrogenases (ADH) of prokaryotic and eukaryotic origin. It is intended for quick screening for a desired enzyme activity to aid in the selection of the right biocatalyst for the synthesis of your target **chiral alcohol**.

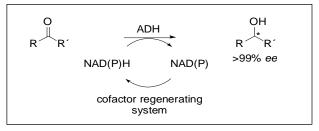


Fig. 1: General reaction catalyzed by alcohol dehydrogenases.

Enzyme Specificities

Table 1 provides a general overview of the substrate specificities and relative activities of the enzymes included in this kit (following page). The indicated data give a general overview — activities may vary according to the specific substrates, assays and reaction conditions.

Formulation and Storage Conditions

All enzymes in this kit are provided as lyophilized powder in quantities of about 100 mg per sample. The lyophilisates are prepared from buffered solutions. Store at -20°C. Avoid repeated freeze/thaw cycles, storage in "frost-free" freezers is not recommended.

Screening Service and Enzyme Requests

evocatal offers this screening as a service:

Send us your compound - we will test all our ADHs (kit-enzymes plus a number of additional ones) for best performance on your substrate of interest. For further information please contact us!

Any evozyme of this kit can be requested from evocatal in larger quantities for further investigation or upscaling purposes. evocatal is open for licensing requests. Please contact us for further details.

Cofactor Regeneration

Cofactors NAD and NADP can be recycled throughout the reaction process (Fig. 1) either by using 2-propanol as co-solvent and co-substrate or by coupling the reaction to a second enzyme, the Glucose Dehydrogenase (evo-1.1.060) which is part of the kit as well. The enzyme re-reduces NAD or NADP using glucose as a hydride donor. Cofactor regeneration reduces the costs of a process significantly. evocatal offers a wide range of solutions in this field – for further information please contact us!





¬ Alcohol Dehydrogenase Kit

evo-1.1.100

Tab. 1: Overview of the evocatal alcohol dehydrogenase activities

(+++ very high activity, ++ high activity, + fair activity, - no activity)

evozyme cat. no:	1.1.010	1.1.020	1.1.030	1.1.040	1.1.130	1.1.140	1.1.190²	1.1.200	1.1.210	1.1.250	1.1.260	1.1.270
selectivity	(R/S) ¹	(R/S) ¹	(S)	(R/S) ¹	(R)	(S)	(R)	(R)	(S)	(S)	(R/S) ¹	(R)
cofactor	NAD	NAD	NAD	NAD	NAD	NAD	NADP	NAD	NAD	NADP	NADP	NADP
$ \begin{array}{ccc} O & x = 0 - 3 \\ X & y & y = 0, 1 \end{array} $ aliphatic ketones	++	++	++	+	-	-	-	+++	-	+	++	++
acetophenone and derivatives	-	-	++	-	-	-	-	++	-	-	-	++
α-ketoesters and derivatives	+++	++	+	-	+++	+++	-	++	+	+	-	-
O O R' β-ketoesters and derivatives	+++	++	+	-	-	-	-	+++	+	++	-	+++
R-CHO R = aliphatic side chain aldehydes	-	++	-	+++	-	-	+++	-	++	+	-	+
x = 0, 1 cyclic ketones	+	+	-	-	-	-	-	++	+	-	++	++

¹substrate dependent (R)- or (S)-selective

²evo-1.1.190 prefers (*R*)-glyceraldehyde over (*S*)-glyceraldehyde.





butyric acid ethyl ester

Alcohol Dehydrogenase 010

evo-1.1.010

Application: selective reduction of ketones

Educts: ketoesters,

ketones with two large side chains

 $\begin{array}{c} O \\ O \\ O \\ O \\ \end{array} \begin{array}{c} + \text{NADH} + \text{H}^+ \\ \end{array} \begin{array}{c} ADH \ 010 \\ O \\ OH \\ \end{array} \begin{array}{c} O \\ OH \\ \end{array} \begin{array}{c} + \text{NAD}^+ \\ \end{array}$ 2-Oxo-4-phenyl-butyric (S)-2-Hydroxy-4-phenyl-

2-Oxo-4-phenyl-butyric acid ethyl ester

> See page 7 for a detailed activity spectrum and further educts!

enzyme properties:

stereoselectivity: mostly (S)-selective, substrate dependent

cofactor dependence: NADH

cofactor regeneration: cofactor can be regenerated enzymatically, e.g. evo-1.1.060

> see page 21 for details!

source: recombinant from E. coli

activity and formulations: > 10000 U/mL* technical grade, liquid

> 100 U/mg* lyoph. lyophilized powder

ref. substrate: 2-Oxo-4-phenyl butyric acid ethyl ester

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.





butyric acid ethyl ester

Alcohol Dehydrogenase 020

evo-1.1.020

Application: selective reduction of ketones

Educts: ketoesters,

ketones with two large side chains

2-Oxo-4-phenyl-butyric ADH 020

ADH 020

(S)-2-Hydroxy-4-phenyl-

2-Oxo-4-phenyl-butyric acid ethyl ester

> See page 7 for a detailed activity spectrum and further educts!

enzyme properties:

stereoselectivity: mostly (S)-selective, substrate dependent

cofactor dependence: NADH

cofactor regeneration: cofactor can be regenerated enzymatically

> see page 21 for details!

source: recombinant from E. coli

activity and formulations: > 1500 U/mL* technical grade, liquid (50% Glycerol)

> 40 U/mg* lyoph. lyophilized powder

ref. substrate: 2-Oxo-4-phenyl butyric acid ethyl ester

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.





evo-1.1.030

Application: selective reduction of ketones and ketoesters

Educts: aliphatic and aromatic ketones, ketoesters

> See page 7 for a detailed activity spectrum and further educts!

enzyme properties:

stereoselectivity: (S)-selective

cofactor dependence: NADH

cofactor regeneration: 2-Propanol as co-substrate

> see page 21 for details!

source: recombinant from E. coli

activity and formulations: > 10-50 U/mL* technical grade, liquid (50 % Glycerol)

> 0.25 U/mg* lyoph. lyophilized powder

ref. substrate: 4'-Chloroacetophenone

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.





Alcohol Dehydrogenase 040

evo-1.1.040

Application: reduction of aldehydes

Educts: aromatic and aliphatic aldehydes

> See page 7 for a detailed activity spectrum and further educts!

enzyme properties:

cofactor dependence: NADH

cofactor regeneration: cofactor can be regenerated enzymatically, e.g. evo-1.1.060

> see page 21 for details!

source: recombinant from E. coli

activity and formulations: > 50 U/mL* technical grade, liquid (50 % Glycerol)

> 1.7 U/mg* lyoph. lyophilized powder

ref. substrate: trans-2-Hexenal

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.





evo-1.1.130

Application: D-selective reduction of aromatic α -ketoacids

Co-substrate: Phenylglyoxylate

> See page 7 for a detailed activity spectrum and further educts!

enzyme properties:

stereoselectivity: (R)-selective, substrate dependent

cofactor dependence: NADH

cofactor regeneration: cofactor can be regenerated enzymatically, e.g. evo-1.1.060

> see page 21 for details!

source: recombinant from *E. coli*

activity and formulations: > 5000 U/mL* technical grade, liquid

> 65 U/mg* lyoph. lyophilized powder

ref. substrate: Phenylglyoxylate

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.





evo-1.1.140

Application: L-selective reduction of aliphatic α -ketoacids

Co-substrate: 2-Ketoisocaproate

> See page 7 for a detailed activity spectrum and further educts!

enzyme properties:

stereoselectivity: (S)-selective, substrate dependent

cofactor dependence: NADH

cofactor regeneration: cofactor can be regenerated enzymatically, e.g. evo-1.1.060

> see page 21 for details!

source: recombinant from *E. coli*

activity and formulations: > 1500 U/mL* technical grade, liquid

> 15 U/mg* lyoph. lyophilized powder

ref. substrate: 2-Ketoisocaproate

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.





evo-1.1.200

<u>Application</u>: (R)-selective reduction of ketones

Educts: aromatic and aliphatic ketones and

ketoesters

> See page 7 for a detailed activity spectrum and further educts!

enzyme properties:

stereoselectivity: (R)-selective

cofactor dependence: NADH

cofactor regeneration: 2-Propanol as co-substrate

> see page 21 for details!

source: recombinant from E. coli

activity and formulations: > 200 U/mL* technical grade, liquid

> **1.0 U/mg* lyoph.** lyophilized powder ref. substrate: Ethyl acetoacetate

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.





Alcohol Dehydrogenase 210 - HLADH-E

evo-1.1.210

<u>Application</u>: selective reduction of aldehydes and cyclic ketones

Educts: cyclic and bicyclic ketones, aldehydes

> See page 7 for a detailed activity spectrum and further educts!

The well-known Horse Liver Enzyme!

enzyme properties:

stereoselectivity: mostly (S)-selective, substrate dependent

cofactor dependence: NADH

cofactor regeneration: cofactor can be regenerated enzymatically, e.g. evo-1.1.060

> see page 21 for details!

source: recombinant from E. coli

activity and formulations: > 30 U/mL* technical grade, liquid

> 0.5 U/mg* lyoph. lyophilized powder

ref. substrate: Benzaldehyde

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.





evo-1.1.250

<u>Application</u>: (S)-selective reduction of ketones and diketones

Educts: aliphatic ketones, diketones, α - and β -

ketoesters

> See page 7 for a detailed activity spectrum and further educts!

enzyme properties:

stereoselectivity: (S)-selective

cofactor dependence: NADPH

cofactor regeneration: cofactor can be regenerated enzymatically, e.g. evo-1.1.060

> see page 21 for details!

source: recombinant from E. coli

activity and formulations: > 150 U/mL* technical grade, liquid

> 4.0 U/mg* lyoph. lyophilized powder

ref. substrate: 2,5-Hexanedione

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.





evo-1.1.260

<u>Application</u>: (R)-selective reduction of small aliphatic ketones

Educts: aliphatic aldehydes, aliphatic and cyclic ketones

> See page 7 for a detailed activity spectrum and further educts!

<u>enzyme properties:</u> This enzyme is thermostable!

stereoselectivity: (R)-selective for small ketones, (S)-selective for large ketones

cofactor dependence: NADPH

cofactor regeneration: 2-Propanol as co-substrate

> see page 21 for details!

source: recombinant from E. coli

activity and formulations: > 500 U/mL* technical grade, liquid

> 7.0 U/mg* lyoph. lyophilized powder

ref. substrate: 2-Propanol

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.





evo-1.1.270

<u>Application</u>: (R)-selective reduction of ketones and aldehydes

Educts: aromatic, cyclic and aliphatic ketones, aldehydes

> See page 7 for a detailed activity spectrum and further educts!

enzyme properties:

stereoselectivity: (*R*)-selective

cofactor dependence: NADPH

cofactor regeneration: 2-Propanol as co-substrate

> see page 21 for details!

source: recombinant from E. coli

activity and formulations: > 450 U/mL* technical grade, liquid

> 3.5 U/mg* lyoph. lyophilized powder

ref. substrate: Acetophenone

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.





Glycerol Dehydrogenase

evo-1.1.190

Application: reduction of aldehydes

Educts: D-Glyceraldehyde

OH HO CHO + NADPH + H
$$^+$$
 ADH 190 OH + NADP $^+$

D-Glyceraldehyde Glycerine

> See page 7 for a detailed activity spectrum and further educts!

enzyme properties:

stereoselectivity: (*R*)-selective with respect to Glyceraldehyde

cofactor dependence: NADPH

cofactor regeneration: cofactor can be regenerated enzymatically, e.g. evo-1.1.060

> see page 21 for details!

source: recombinant from E. coli

activity and formulations: > 40 U/mL* technical grade, liquid

> **0.75 U/mg* lyoph.** lyophilized powder

ref. substrate: D-Glyceraldehyde

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.





L-Lactate Dehydrogenase

evo-1.1.150

Application: Reduction of pyruvate

Substrate: Pyruvate

> L-LDH reduces pyruvate to L-lactate and oxidizes NADH to NAD+.

enzyme properties:

stereoselectivity: L-selective

cofactor dependence: NADH

cofactor regeneration: cofactor can be regenerated enzymatically, e.g. evo-1.1.060

> see page 21 for details!

source: recombinant from E. coli

activity and formulations: $> 500 \text{ U/mL}^*$ technical grade, liquid (2M (NH₄)₂SO₄)

ref. substrate: Pyruvate

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.





Overview

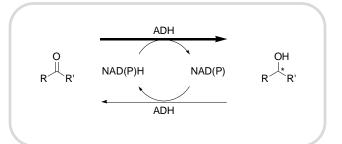
Cofactors

Alcohol Dehydrogenases depend on either NAD (nicotinamide adenine dinucleotide) or NADP (nicotinamide adenine dinucleotide phosphate) as a cofactor, which is an essential part of the enzymatic reaction. The reduced form of these cofactors (NAD(P)H) delivers the hydride-equivalent needed for the reduction of carbonyl groups and is oxidized during this reaction (Scheme 1). The back reaction, namely the oxidation of alcohols to ketones and reduction of the cofactor, is feasible as well, but generally not favored.

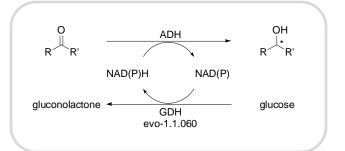
Cofactor Regeneration

Cofactors like NADH and NADPH are usually prohibitively expensive. Thus using them in stoichiometric amounts is hardly ever applicable, except in small scale screening applications. In commercial processes, the cofactors are recycled and used in catalytic amounts, thus reducing production costs significantly. Cofactor regeneration can be performed either by the same enzyme using a co-substrate (Scheme 1) or by a second enzyme in a coupled reaction (Scheme 2).

Scheme 1: Regeneration of the cofactor NAD(P)H during a reduction by the same enzyme using 2-Propanol as a cosubstrate.



Scheme 2: Regeneration of the cofactor NAD(P)H during a reduction coupled to a parallel reaction using a second enzyme like Glucose Dehydrogenase (evo-1.1.060).



The evocatal ADHs that do accept 2-propanol as cosubstrate are labelled respectively on the particular datasheets. For any further questions please contact us.

Please note: evocatal offers process development as service! We find the most suitable enzyme system for your chiral product by screening our enzymes as well as other available ones. We develop the most efficient process for you! We're looking forward to your success!





Glucose Dehydrogenase

evo-1.1.060

Application: Regeneration of NADH / NADPH

Co-substrate: D-Glucose

SDH is an universal enzyme for cofactor regeneration with high activity, that accepts NAD and NADP and works well in aqueous solutions.

enzyme properties:

regenerated cofactor: NAD(P)H

source: recombinant from E. coli

activity and formulations: > 1500 U/mL* technical grade, liquid

> 35 U/mg* lyoph. lyophilized powder

ref. substrate: D-Glucose

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.



¬ Cofactor Regeneration



Formate Dehydrogenase

evo-1.1.230

Application: Regeneration of NADH

Co-substrate: Formate

$$\begin{array}{c} \text{NAD}^+ + \begin{array}{c} \text{O} \\ \text{H} \end{array} \begin{array}{c} \text{FDH} \\ \end{array} \end{array} \begin{array}{c} \text{NADH + CO}_2 \\ \end{array}$$

> FDH is able to reduce NAD⁺ to NADH. Advantage of this enzyme is is the formation of the volatile carbon dioxide. This allows to shift reaction equilibrium to complete conversion and simplifies product isolation and purification.

enzyme properties:

regenerated cofactor: NADH

source: recombinant from E. coli

activity and formulations: > 15 U/mL* technical grade, liquid

ref. substrate: Formate

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.





Isocitrate Dehydrogenase

evo-1.1.070

Application: Regeneration of NADPH

Co-substrate: D-threo-Isocitrate

> IDH is able to reduce NADP+ to NADPH. Advantage of this enzyme is the formation of the volatile carbon dioxide. This allows to shift reaction equilibrium to complete conversion.

enzyme properties:

regenerated cofactor: NADPH

source: recombinant from *E. coli*

activity and formulations: > 100 U/mL* technical grade, liquid

ref. substrate: D-threo-Isocitrate

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.



¬ Cofactor Regeneration



Malate Dehydrogenase

evo-1.1.080

Application: Regeneration of NADPH

Co-substrate: L-Malate

> MAE-80 is able to reduce NADP+ to NADPH. Advantage of this enzyme is the formation of the volatile carbon dioxide. This allows to shift reaction equilibrium to complete conversion.

enzyme properties:

regenerated cofactor: NADPH

source: recombinant from E. coli

activity and formulations: > 200 U/mL* technical grade, liquid

ref. substrate: L-Malate

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.



¬ Cofactor Regeneration



Malate Dehydrogenase

evo-1.1.090

Application: Regeneration of NADH

Co-substrate: L-Malate

> MAE-90 is able to reduce NAD+ to NADH. Advantage of this enzyme is the formation of the volatile carbon dioxide. This allows to shift reaction equilibrium to complete conversion.

enzyme properties:

regenerated cofactor: NADH

source: recombinant from E. coli

activity and formulations: > 150 U/mL* technical grade, liquid (5% BSA)

ref. substrate: L-Malate

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.





L-Leucine Dehydrogenase

evo-1.1.110

Application: L-selective formation of amino acids from α -ketoacids

Substrate: Pyruvate and aliphatic α -ketoacids

> LeuDH converts aliphatic α -ketoacids to L-amino acids. Consumed cofactor is NADH.

enzyme properties:

stereoselectivity: L-selective

cofactor dependence: NADH

cofactor regeneration: cofactor can be regenerated enzymatically, e.g. evo-1.1.060

> see page 21 for details!

source: recombinant from *E. coli*

activity and formulations: > 150 U/mL* technical grade, liquid

> 5 U/mg* lyoph. lyophilized powder

ref. substrate: L-Leucine

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.





L-Alanine Dehydrogenase

evo-1.1.160

Application: L-selective formation of α -amino acids

Substrate: Pyruvate and aliphatic α -ketoacids

O + NADH +
$$NH_4^+$$
 AlaDH NH_2 O + NAD^+

Pyruvate

L-Alanine

> L-AlaDH converts Pyruvate to L-Alanine and oxidizes NADH to NAD+.

enzyme properties:

stereoselectivity: L-selective

cofactor dependence: NADH

cofactor regeneration: cofactor can be regenerated enzymatically, e.g. evo-1.1.060

> see page 21 for details!

source: recombinant from E. coli

activity and formulations: > 350 U/mL* technical grade, liquid

ref. substrate: L-Alanine

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.





Hydroxynitrilase 010

evo-1.4.010

<u>Application</u>: (R)-selective formation of hydroxynitriles

Educts: aromatic and aliphatic aldehydes and ketones

> for bulk quantities, please inquire!

enzyme properties:

stereoselectivity: (R)-selective

source: recombinant from E. coli

activity and formulations: > 500 U/mL* technical grade, liquid

> 10 U/mg* lyoph. lyophilized powder

ref. substrate: racemic Mandelonitrile

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.





Product Information

evo-1.4.010

Enzyme Description

This Hydroxynitrile Lyase from *Arabidopsis thaliana* catalyzes the formation of (*R*)-hydroxynitriles highly selectively. It shows highest activity against a wide variety of substrates, especially in non-aqueous media. The enzyme is recombinantly expressed from *E. coli* and available in all quantities on demand

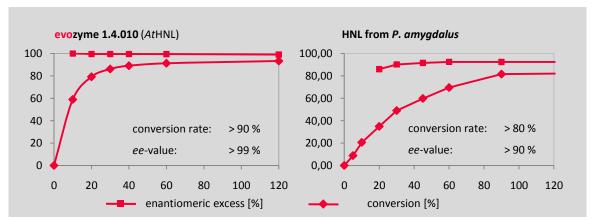
Process Example: Synthesis of enantiopure (*R*)-Mandelonitrile

Examples* for well accepted educts CHO R = -H, -F, -CI CHO O O O

* see J. Andexer, J. von Langermann, A. Mell, M. Bocola, U. Kragl, T. Eggert, M. Pohl. An *R*-selective Hydroxynitrile Lyase from *Arabidopsis thaliana* with an a/b-Hydrolase Fold. *Angew. Chem. Int. Ed.* **46**: 8679 – 8681.

Performance of evozyme 1.4.010

The graph shows HNL from *A. thaliana* compared to HNL from *P. amygdalus* (almonds) in the conversion of Benzaldehyde to (*R*)-Mandelonitrile







Hydroxynitrilase 020

evo-1.4.020

Application: (S)-selective formation of hydroxynitriles

Educts: aromatic and aliphatic aldehydes and ketones

> for bulk quantities, please inquire!

enzyme properties:

stereoselectivity: (S)-selective

properties: pH_{opt}: 5.5, T_{opt}: 40°C

source: recombinant from E. coli

Examples* for well accepted educts

CHO CHO

* see S. Förster, J. Roos, F. Effenberger, H. Wajant, A. Sprauer. The First Recombinant Hydroxynitrile Lyase and its Application in the Synthesis of (S)-Cyanohydrins. *Angew. Chem. Int. Ed.* **35**: 437 – 439.

activity and formulations: > 170 U/mL* technical grade, liquid

ref. substrate: racemic Mandelonitrile

availability: standard quantities (lyophilisates) are available from stock

^{*}U = Enzymatic Unit. 1 U is defined as the amount of enzyme that converts 1 μmol of a defined substrate in one minute. It is strictly dependent on the substrate referred to. The reference substrates in this catalog is always given for each enzyme individually.





¬ Lipase Screening Kit

evo-1.3.100

evozymes 1.3.101 - 1.3.135

Kit Description

The evocatal Lipase Screening Kit comprises 35 preparations of different lipases and esterases of microbial origin. It is intended for quick screening for a desired enzyme activity to aid in the selection of the right biocatalyst for your specific application.

The preparations originate from bacterial cultures whereas evozymes 1.3.101 – 1.3.116 are overexpressed from recombinant bacterial strains and evozymes 1.3.117-1.3.135 stem from a metagenomic screening. The latter are expressed from cosmids under control of their wildtype promoters.

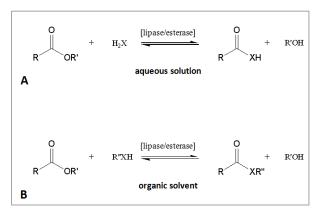


Fig. 1: General reactions catalyzed by lipases and esterases including hydrolysis (A) and transesterification (B).

Activities and Specificities

Table 1 provides a general overview of the substrate specificities and activities of the enzymes included in this kit (following page). The indicated data give only a general overview about the activities which may vary according to the specific substrates, assays and reaction conditions.

Formulation and Storage Conditions

All enzymes in this kit are provided as lyophilized powder in quantities of about 100 mg per sample. The Lyophilisates are prepared from solutions in 150 mM sodium phosphate buffer pH 8.2. Store at -20°C. Avoid repeated freeze/thaw cycles, storage in "frost-free" freezers is not recommended.

Enzyme Requests

Any evozyme of this kit that reveals interesting activities can be requested from evocatal in larger quantities for further investigation or upscaling purposes. evocatal is open for licensing requests. In case of the metagenomic clones evocatal offers the subcloning and overexpression of the specific enzyme on a short time-scale. For further information please inquire.





¬ Lipase Screening Kit

evo-1.3.100

Lipase Screening Kit continued

Tab. 1: Overview of lipase and esterase activities of the kit enzymes.

The activities are given in mU / mg lyophilisate referring to the hydrolysis reactions. One unit is defined as the amount of enzyme that converts one μ mol of substrate per minute.

evozyme	general a	general activities		
cat. no.	short chain esters	long chain esters		
1.3.101	> 50	> 1		
1.3.102	> 150	> 100		
1.3.103	> 120	> 150		
1.3.104	> 150	> 50		
1.3.105	> 150	> 40		
1.3.106	> 150	> 50		
1.3.107	> 45	> 10	recombinant	
1.3.108	> 0.7	~ 0.1	overexpression	
1.3.109	> 7	> 0.5	in E. coli	
1.3.110	> 150	~ 0.1	III E. COII	
1.3.111	> 100	> 70		
1.3.112	> 0.5	-		
1.3.113	> 120	> 100		
1.3.114	> 25	> 15		
1.3.115	> 1	> 0.1		
1.3.116	> 15	> 0.8		
1.3.117	> 25	> 10		
1.3.118	> 1	> 0.1		
1.3.119	> 7	> 1		
1.3.120	> 1	> 0.1		
1.3.121	> 1	> 0.1		
1.3.122	> 1	> 0.5		
1.3.123	> 1	> 0.1		
1.3.124	> 1	> 0.1		
1.3.125	> 1	> 0.1	metagenomic clones	
1.3.126	> 7	> 0.3	expressed from	
1.3.127	> 4	> 0.2	cosmids in <i>E. coli</i>	
1.3.128	> 4	~ 0.1		
1.3.129	> 2	> 0.1		
1.3.130	> 5	-		
1.3.131	> 3	> 0.1		
1.3.132	> 1	> 0.1		
1.3.133	> 1	> 0.1		
1.3.134	> 1	> 0.1		
1.3.135	> 1	> 0.1		





¬ Transaminase Screening Kit

evo-1.2.100

seven bacterial transaminases (evozymes 1.2.101 - 1.2.106)

- > bacterial transaminases for screening applications
- > recombinantly expressed in *E. coli soon available!*

reaction schemes:

tested substrates: Acetophenone

Propiophenone

Ethyl acetoacetate

soon available!

2-Pentanone

Cyclohexanone

stereoselectivity: predominantly (S)-selective

amine donors: L-Alanine is a good amine donor for all enzymes,

Isopropylamine works fine in most cases

technical information:

activity: 3.0 -7.0 mU/mg (Ethylacetoacetate)

formulation: lyophilized powder

properties: pH-Optima range between 6.0 and 8.0

Temp.-Optima range between 30° C and 37°C (amination)

storage: -20°C



¬ ordering information

evozymes

contact

For ordering sample quantities please refer to the pricing information on page 3. To place an order please contact us by phone or email or fill in the fax form on the next page and fax it to +49 211 1576 0959.

For any further information or ordering of bulk quantities, please contact us by phone or email.

We're looking forward to your request!

evocatal GmbH Merowingerplatz 1a 40225 Düsseldorf

Fon: +49 (0) 211 15760950 Fax: +49 (0) 211 15760959

web: www.evocatal.com email: sales@evocatal.com



order form



To place an order please fill in this form and send it by fax to +49 211-15760959 or mail it to:

Firma evocatal GmbH Merowingerplatz 1a 40225 Düsseldorf Germany

ORDER:

catalog. no.:	product name:	quantity:	unit price (€):	total price (€):
			total (€):	
				,
delivery add	<u>Iress:</u>	billing	g address	
name:		name:		
institute/compa	ny:	institu	te/company:	
address:		addre	SS:	
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phone number:			number:	
fax number:		for nu		
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credit card (Visa	a or Master):	expira	tion date (MM/YYYY):	
credit card no.:		card v	validation code (CVC):	
date		sign	ature (company st	(amp)

¬ General Terms and Conditions

evozymes

Art. 1

Application of these General Terms and Conditions of Sale, Quotation and Conclusion of Contract,

Subject Matter of Contract

1. All quotations and all supplies and services from evocatal GmbH shall be provided exclusively on the basis of these General Terms and Conditions of Sale. They shall be an integral part of all of the contracts that evocatal GmbH concludes with the customer concerning the supplies and services offered by evocatal GmbH. They shall also apply to all future quotations, supplies and services, even if their application is not once again separately agreed upon.

The customer's general terms and conditions of business or those of a third party shall not be applied, even if evocatal GmbH does not separately object to their application in individual cases.

- 2. All of evocatal GmbH's quotations shall be subject to confirmation and without obligation, unless they are expressly designated as binding or a certain time limit for acceptance is contained therein. The contract concluded between evocatal GmbH and the customer in writing, by e-mail or by fax completely reproduces all of the agreements between the parties to the contract regarding the subject matter of the contract. Verbal agreements between the parties to the contract shall only apply if this is expressly determined therein.
- 3. The subject matter of the contract shall be exclusively the product as declared in the contract. At the same time details of the subject matter of the contract such as weights, dimensions or data on physical and technical characteristics as well as technical data are only approximately authoritative, unless the usability for the purpose provided for in the contract necessitates exact congruence. The details are not quality statements but solely a description or characterisation of the supply or service.
- 4. The supply of a product or the provision of a service by evocatal GmbH shall only comprise consent to the use of evocatal GmbH's intellectual property embodied in them or related to them (in particular of evocatal GmbH's patents), if this is expressly agreed upon with the customer.

Art. 2

Prices, Payment

- 1. The prices are to be understood as being ex works (place of the registered head office of evocatal GmbH in 40225 Düsseldorf). Added to the prices shall be unless expressly stated otherwise in the contract the cost of packaging, transport and, if necessary, if agreed the transport insurance. The agreed prices shall be net prices, on which statutory VAT is additionally payable.
- 2. Invoice amounts are to be paid within 14 days without deduction. If the customer does not pay by the due date, default interest in the amount of 10 percentage points above the base interest rate per annum shall be incurred; the claiming of higher interest and further losses in the event of default shall remain unaffected.
- 3. Offsetting with any of the customer's counter-claims and the withholding of payments because of such counter-claims shall only

be admissible if the customer's counter-claims are undisputed or have been established by a final and non-appealable legally effective court decision.

4. evocatal GmbH shall be entitled to execute or to perform any still outstanding supplies or services only on the terms of cash in advance or the provision of security, if after the conclusion of a contract circumstances become known which are liable to significantly reduce the customer's creditworthiness and by means of which the payment of the outstanding accounts due to evocatal GmbH regarding the outstanding supply or service is endangered.

Art.

Delivery, Delivery Time

- 1. All deliveries shall be made ex works (place of the registered head office of GmbH in 40225 Düsseldorf), unless agreed otherwise in the contract.
- 2. Delivery dates or dates for the provision of services proposed by evocatal GmbH shall be without engagement unless a fixed time or a fixed date has been expressly agreed upon in the contract. If forwarding was agreed upon in the contract, the delivery periods and the delivery dates shall refer to the point in time of handover to the third party entrusted with transportation.
- 3. evocatal GmbH shall be entitled to make part deliveries, if the part delivery is useable for the customer in accordance with the contractually assumed purpose, delivery of the remaining goods has been ensured and the customer does not have any significant additional work as a result of the part delivery.
- 4. If evocatal GmbH defaults on supply or service or if supply or service no matter for what reason becomes impossible for it, its liability for damages shall be restricted in accordance with Art. 6 of these General Terms and Conditions of Sale.

Art. 5

Warranty

- 1. The warranty period shall be one year from delivery or as far as an acceptance inspection is necessary in the individual case from acceptance. If attention is drawn in the contract to the fact that the subject matter of the contract shall show stability or a minimum durability over a period of less than one year, a warranty shall only be assumed for this stability period stated or minimum durability period.
- 2. The product supplied is to be examined carefully for any defects. The product shall be deemed to have been approved when evocatal GmbH has, with regard to obvious defects or defects which were recognisable during careful examination without undue delay, no notice of defect has been received in writing or by fax or e-mail within seven days of the delivery and/or acceptance or otherwise within seven days after the point in time, at which the defect was recognisable for the customer during normal use of the subject matter of the contract without any closer examination.
- 3. The customer must give evocatal GmbH the opportunity to examine the subject matter of the contract at the place where it is located, therefore, if necessary, on the customers' premises, if the customer claims that there is a material defect.





¬ General Terms and Conditions

evozymes

4. If a product supplied shows a defect of title in the form of a thirdparty right, the customer shall give evocatal GmbH, at its option, the opportunity to either deliver an item free from defects or to remove the defects within a reasonable period of time by obtaining approval of the holder of the right. If the form of subsequent performance chosen by evocatal proves abortive, the customer shall, at its option, have the right to reduce the price or withdraw from the contract.

5. If a defect is based on a fault on the part of evocatal GmbH, the customer can demand damages on the conditions defined in Art. 6.

Art. 6

Liability for Damages due to Culpability

1. The liability of evocatal GmbH for damages, no matter for what legal reason, in particular due to default, impossibility, defective or incorrect supply, breach of contract, breach of duties at contract negotiations or from a tortious act, is – to the extent that in each case it is dependent upon fault – restricted in accordance with this provision in Art. 6:

The company evocatal GmbH shall not be liable in the case of slight negligence of its organs, statutory representatives, employees or other vicarious agents;

b) gross negligence of its non-executive employees or other vicarious agents, unless the breach of duties essential to the contract is involved.

Essential to the contract are the obligations relating to timely delivery free of defects as well as consulting, protection and order duties, which are meant to make contractual use of the subject matter of the contract possible for the customer, or the purpose of which is the protection of life and limb of the customer or third party or of the customer's property from considerable damage/loss.

To the extent that evocatal GmbH shall be liable for damages on the merits in accordance with this provision, this liability shall be limited to that damage/loss which evocatal GmbH, at the time of the conclusion of the contract and with the exercising of ordinary care, foresaw as the possible consequence of a breach of contract or, taking into account the circumstances of which it was aware or should have been aware, it would have had to foresee. Indirect damage/loss and consequential damage which is the consequence of defects of the subject matter of the contract, shall, moreover, only be recoverable if such damage/loss is typically to be expected in the case of use of the subject matter of the contract for the intended purpose.

2. In the case of liability for slight negligence, the liability for damages of evocatal GmbH for damage/loss of every kind shall be restricted to an amount of € 2,000,000.00 per case of damage even if a breach of duties not in conformity with the contract is involved.

The above mentioned exclusions from liability and restrictions shall apply to the full extent also in favour of the organs, statutory representatives, employees and other vicarious agents of evocatal GmbH.

To the extent that evocatal GmbH provides technical or physical information or becomes active in an advisory capacity and this information or these advisory services are not part of the scope of services owed by evocatal and contractually agreed upon, this shall be done free of charge and to the exclusion of all liability. The restrictions in accordance with this Art. 6 shall not apply to the liability on the part of evocatal GmbH for intentional behaviour, for statements of warranted nature and quality, for harm to life and limb or health impairment or in accordance with the Product Liability Act.

Art. 7

Choice of Law, Place of Jurisdiction

- 1. The exclusive place of jurisdiction shall be Düsseldorf.
- 2. The contractual relations between evocatal GmbH and the customer shall be subject to the law of the Federal Republic of Germany to the exclusion of the provisions of private international law. The provisions of the UNCITRAL law on the sale of goods shall not apply, either.

