

Enantiomerenanalytik

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Outline

Introduction

- ◆ Chirality, physiological activity of enantiomers
- ◆ Definitions
- ◆ Databases, Literature, useful links

Analytical (Separation) Methods

- ◆ TLC, GC, HPLC, SFC, CE, NMR, Polarimetry, Crystallization
- ◆ Enantioselective Interactions

Method Selection

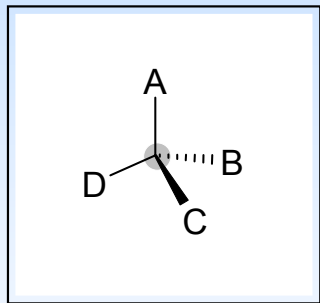
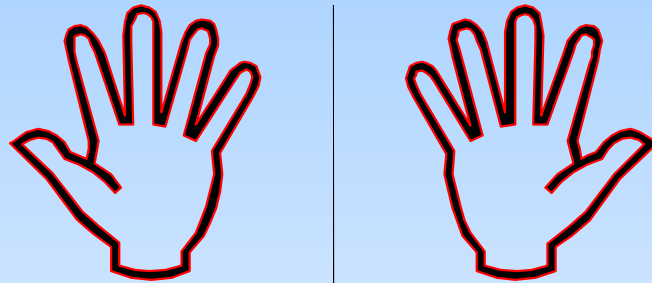
- ◆ Where to start
- ◆ Comparative separations

Not covered:

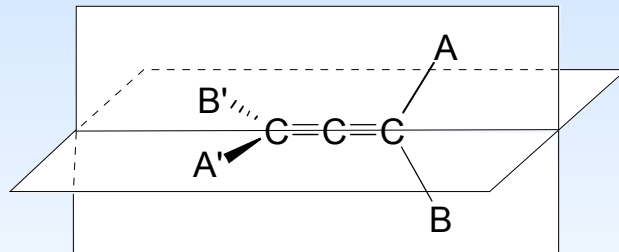
Preparative separations

Chirality

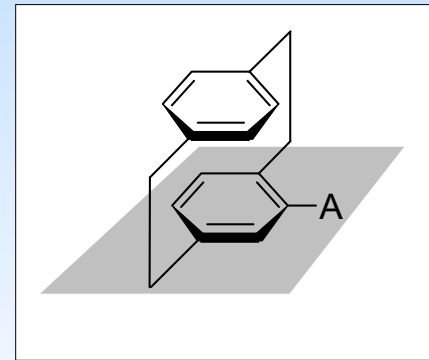
Chirality: Description for the property of any object to be different from its own mirror image



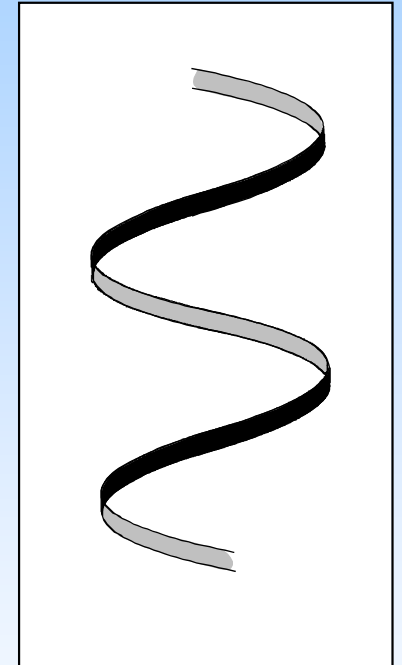
central



axial



planar



helical

Enantiomers: Mirror images of any object

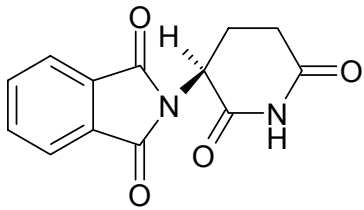
Racemate: equimolar mixture of two enantiomers

Diastereomers: Stereoisomers, which do not behave as image and mirror image (possible for molecules with more than one chirality elements).

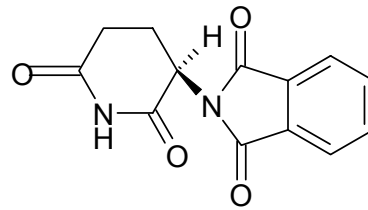
Physiological Activity of Enantiomers

Therapeutics

Thalidomide



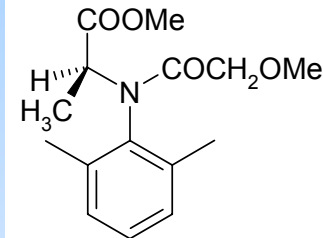
(S)
teratogenic



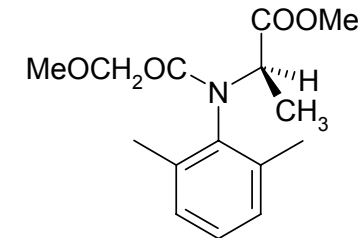
(R)
sedative

Environment

Metalaxyl



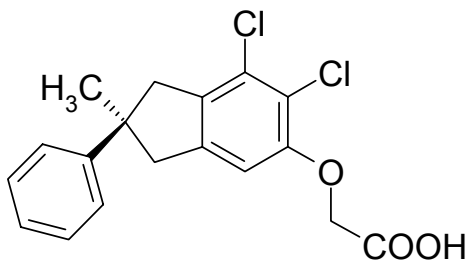
(S)
inactive



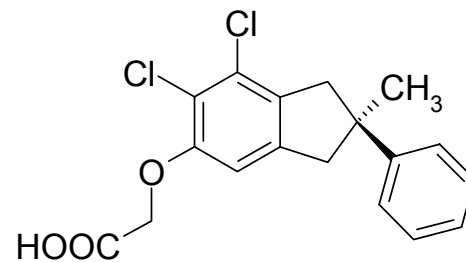
(R)
active fungicide

Food / Feed

Indacrinone

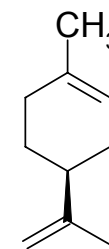


(S)
promotes
uric acid
secretion

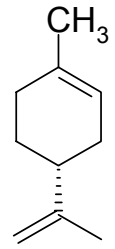


(R)
diuretisch

Limonen

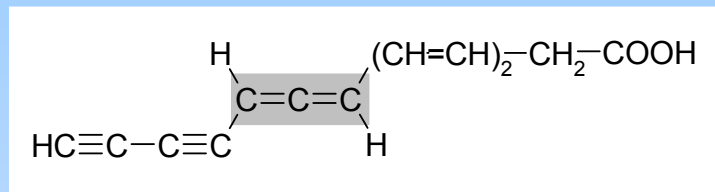


(S)
Citron Flavour

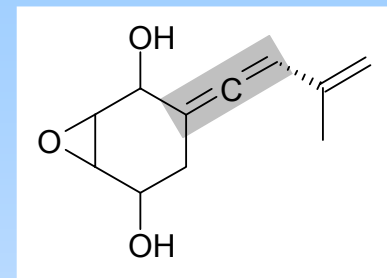


(R)
Orange Flavour

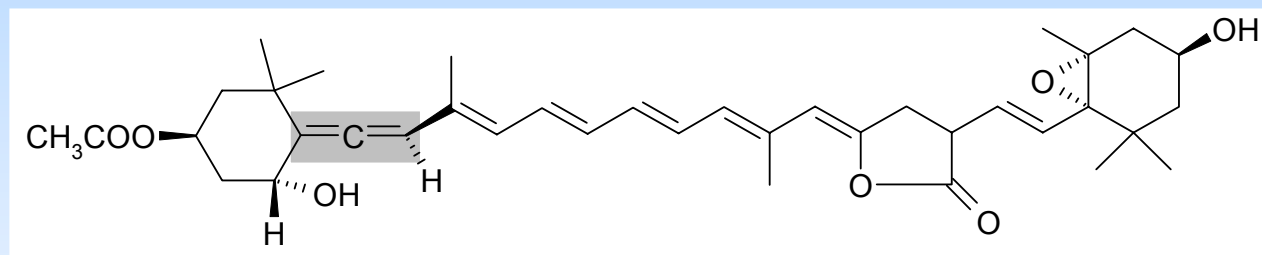
Axial Chirality



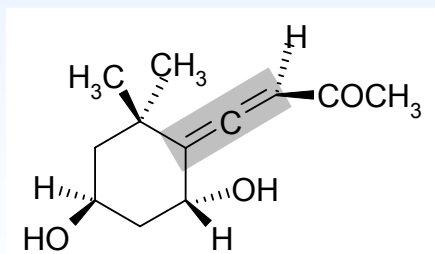
Mycomycin (*Nocardia acidophilus*)



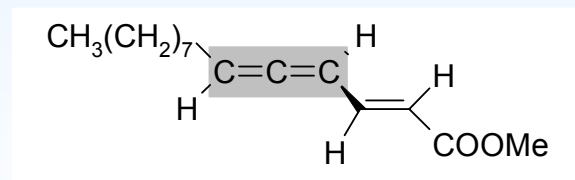
Plant pathogen (*Eutypa lata*)



Peridinin
(e .g. from dinoflagellates)



Secret for ant defense
(*Romalea microptera*)



Sexual pheromone
(*Acanthoscelides obtectus*)

Access to optically pure compounds

1. Isolation from natural sources

2. Fermentation (*de novo* synthesis)

3. Resolution of racemates

- Separation of Diastereoisomers (crystallization)
- Chromatography using chiral stationary phases
- **Biocatalysis**

4. Asymmetric Synthesis

- Chemo-Catalysis
- **Biocatalysis**

Definitions

Resolution, R_s

$$R_s = 2 \cdot \frac{t_s'' - t_s'}{W_b'' + W_b'} = 1.177 \cdot \frac{t_s'' - t_s'}{b_{0.5}'' + b_{0.5}'}$$

$R_s > 1.5$ results usually in base line separation

Enantiomeric excess, ee

$$ee [\%] = \frac{[R] - [S]}{[R] + [S]} \times 100$$

Efficiency of separation

Retention factor, k

$$k = \frac{(t_s - t_0)}{t_0}$$

t_0 = "dead volume"

t_s = retention time

Separation factor, α

$$\alpha = \frac{k''}{k'}$$

k	$\alpha = 2.00$	$\alpha = 1.10$	$\alpha = 1.01$
0.1	1936	193 600	1 936 000
1	64	6 400	640 000
10	19.4	1 936	193 600
- $\Delta(\Delta G)$ (kcal/mol)	410	56	6

Databases

Chemical abstracts – SciFinder

The logo for SciFinder Scholar 2002 is displayed on a dark blue and purple background with diagonal light streaks. The text 'SciFinder' is in a large, white, serif font with a registered trademark symbol. Below it, 'SCHOLAR' is written in a smaller, white, sans-serif font, and '2002' is in a large, white, sans-serif font. Two horizontal white lines separate the three text elements.

SciFinder®
SCHOLAR
2002

Recent Advances in Chiral Separations

A Bibliometric Study Using SciFinder Scholar

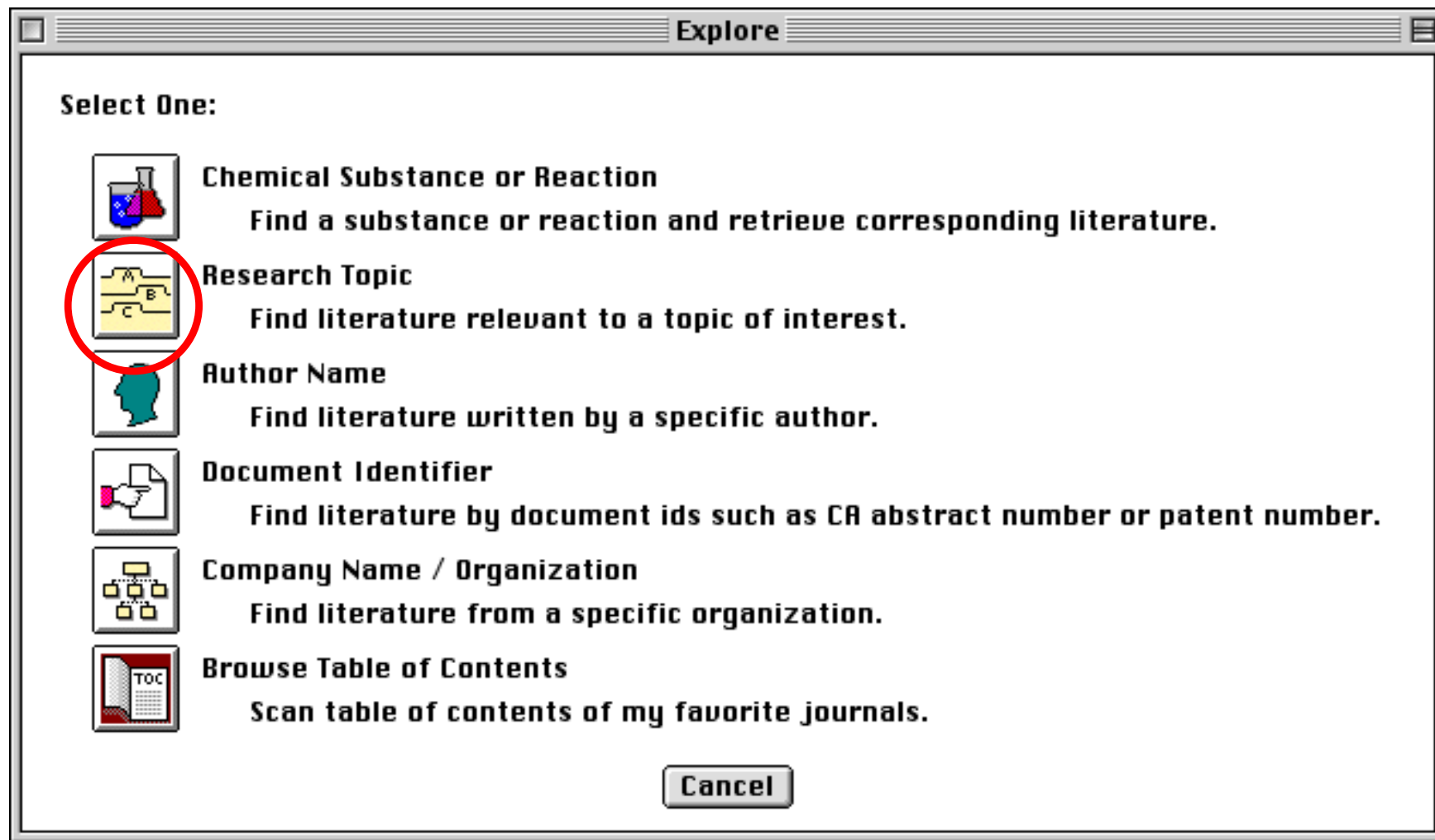
Prof. Pietzsch,
MLU Halle-Wittenberg, FB IW

SciFinder Scholar: a powerful search tool for locating scientific information

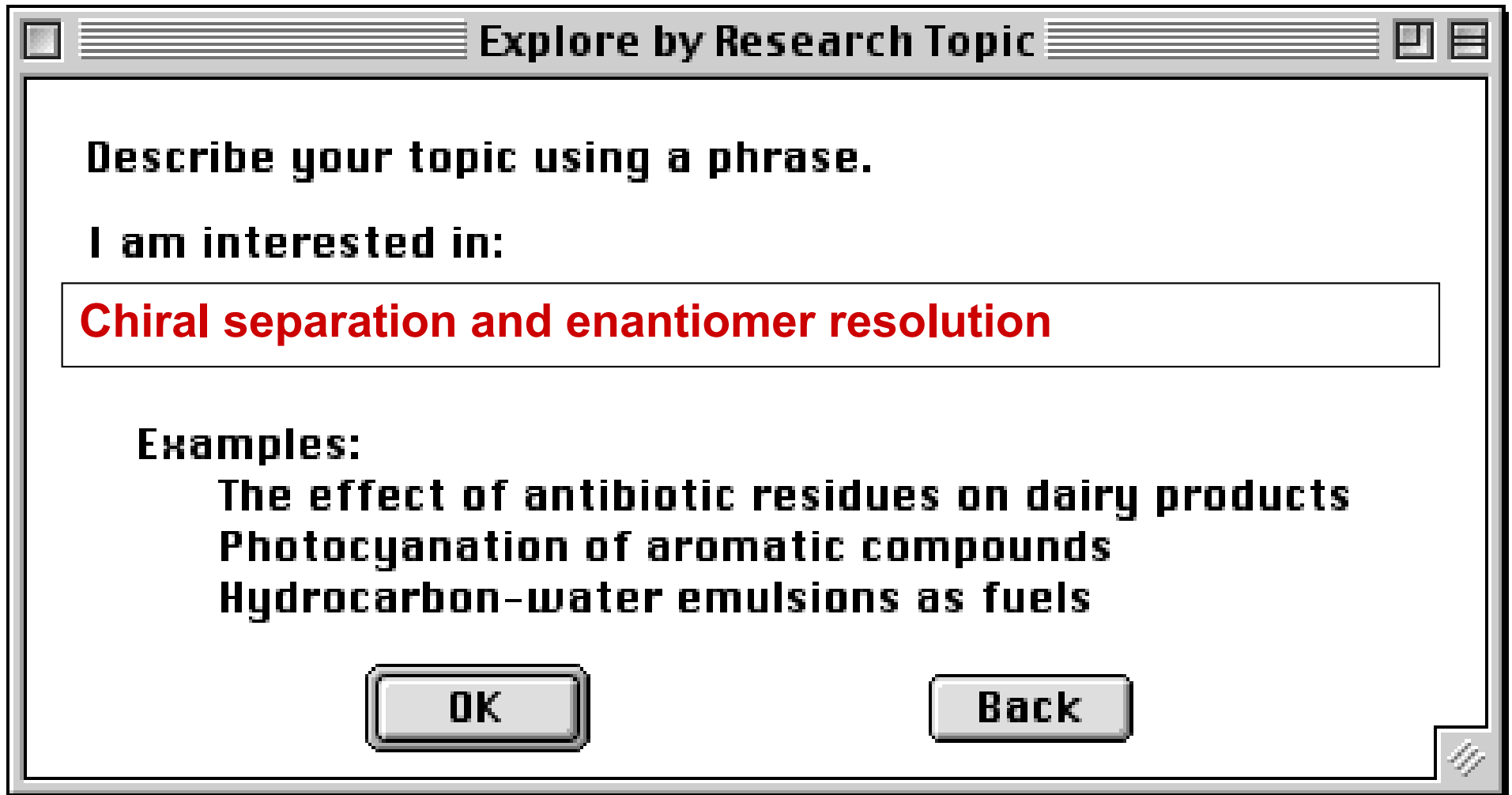
- Scholar provides many access points to retrieve information:
 - chemical name
 - chemical structure
 - keywords
 - author names...
- Use Scholar to analyze your search results

Explore by Research Topic

Begin your search by selecting “Research Topic”



Input Your Search Terms



Explore by Research Topic

Describe your topic using a phrase.

I am interested in:

Chiral separation and enantiomer resolution

Examples:

- The effect of antibiotic residues on dairy products**
- Photocyanation of aromatic compounds**
- Hydrocarbon-water emulsions as fuels**

OK **Back**

Topic Candidates

SciFinder Scholar presents candidates of interest based on the research topic input. Over 15,015 answers are retrieved.

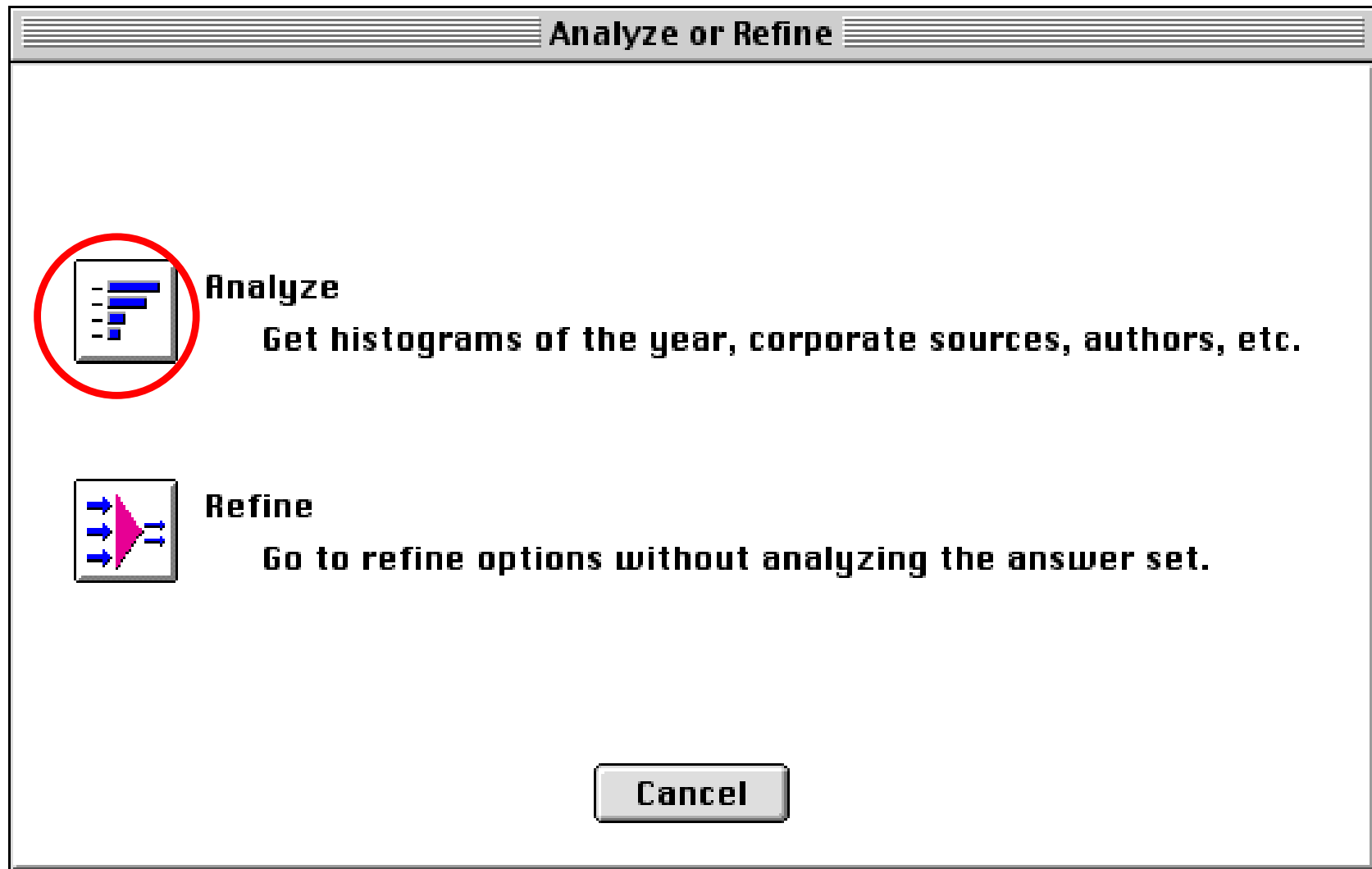
The screenshot displays the SciFinder Scholar application window. The title bar reads "SciFinder Scholar". The menu bar includes "File", "Edit", "View", "Task", "Tools", and "Help". The toolbar contains icons for "NewTask", "Back", "Forward", "Print", "Save As", "Full Text", "Prefs", "Database", "History", "Internet", "Help", and "Exit".

The main content area shows a list of search results, each with a checkbox on the left and a magnifying glass icon on the right. The results are as follows:

- Wang, Xing; Liu, Na; Cao, Hui; Zhao, Xiaogang; Zhang, Wanjin. **Chiral separation of racemic BN-DIF trimer.** Polymer Preprints (American Chemical Society, Division of Polymer Chemistry) (2003), 44(2), 812. CODEN: ACPPAY ISSN:0032-3934. AN 2003:658260 CAPLUS
- Yang, Guo-Sheng; Vazquez, Piedad Parrilla; Frenich, Antonia Garrido; Vidal, Jose L. Martinez; Aboul-Enein, Hassan Y. **Chiral separation of organic phosphonate compounds on cellulose CSP (chiral stationary phase) under reversed phase mode.** Analytical Sciences (2003), 19(8), 1157-1161. CODEN: ANSCEN ISSN:0910-6340. AN 2003:657671 CAPLUS
- Usachev, Sergey V.; Nikiforov, Grygorii A.; Strelenko, Yurii A.; Chervin, Ivan I.; Lyssenko, Konstantin A.; Kostyanovsky, Remir G. **Nitrogen chirality via the sterical veto of N inversion.** Mendeleev Communications (2003), (3), 136-139. CODEN: MENCEX ISSN:0959-9436. AN 2003:655712 CAPLUS
- Vinogradov, Maxim G.; Kurilov, Dmitry V.; Chel'tsova, Galina V.; Ferapontov, Vladimir A.; Heise, Glenn L. **Efficient resolution of (\pm)- α -cyclopropylethanol by crystallization of its inclusion complex with chiral diols.** Mendeleev Communications (2003), (3), 124. CODEN: MENCEX ISSN:0959-9436. AN 2003:655704 CAPLUS
- Kostyanovsky, Remir G.; Kostyanovsky, Vasilii R.; Kadorkina, Gul'nara K.; Lyssenko, Konstantin A. **Asymmetric three-coordinated nitrogen compounds: Spontaneous resolution and absolute asymmetric synthesis.** Mendeleev Communications (2003), (3), 111-113. CODEN: MENCEX ISSN:0959-9436. AN 2003:655699 CAPLUS
- Bredikhin, Alexander A.; Bredikhina, Zemfira A.; Lazarev, Sergey N.; Savell'ev, Dmitry V. **Systematic search for conglomerates among glycerol aromatic monoethers: Guaifenesin and mephenesin are the cases.** Mendeleev Communications (2003), (3), 104-105. CODEN: MENCEX ISSN:0959-9436. AN 2003:655696 CAPLUS
- Lukin, Oleg; Mueller, Walter M.; Mueller, Ute; Kaufmann, Astrid; Schmidt, Claus; Leszczynski, Jerzy; Voegtle, Fritz. **Covalent chemistry and conformational dynamics of topologically chiral amide-based molecular knots.** Chemistry--A European Journal (2003), 9(15), 3507-3517. CODEN: CEUJED ISSN:0947-6539. AN 2003:651618 CAPLUS

SciFinder Scholar Analyze Tool

Sort and visualize your search results

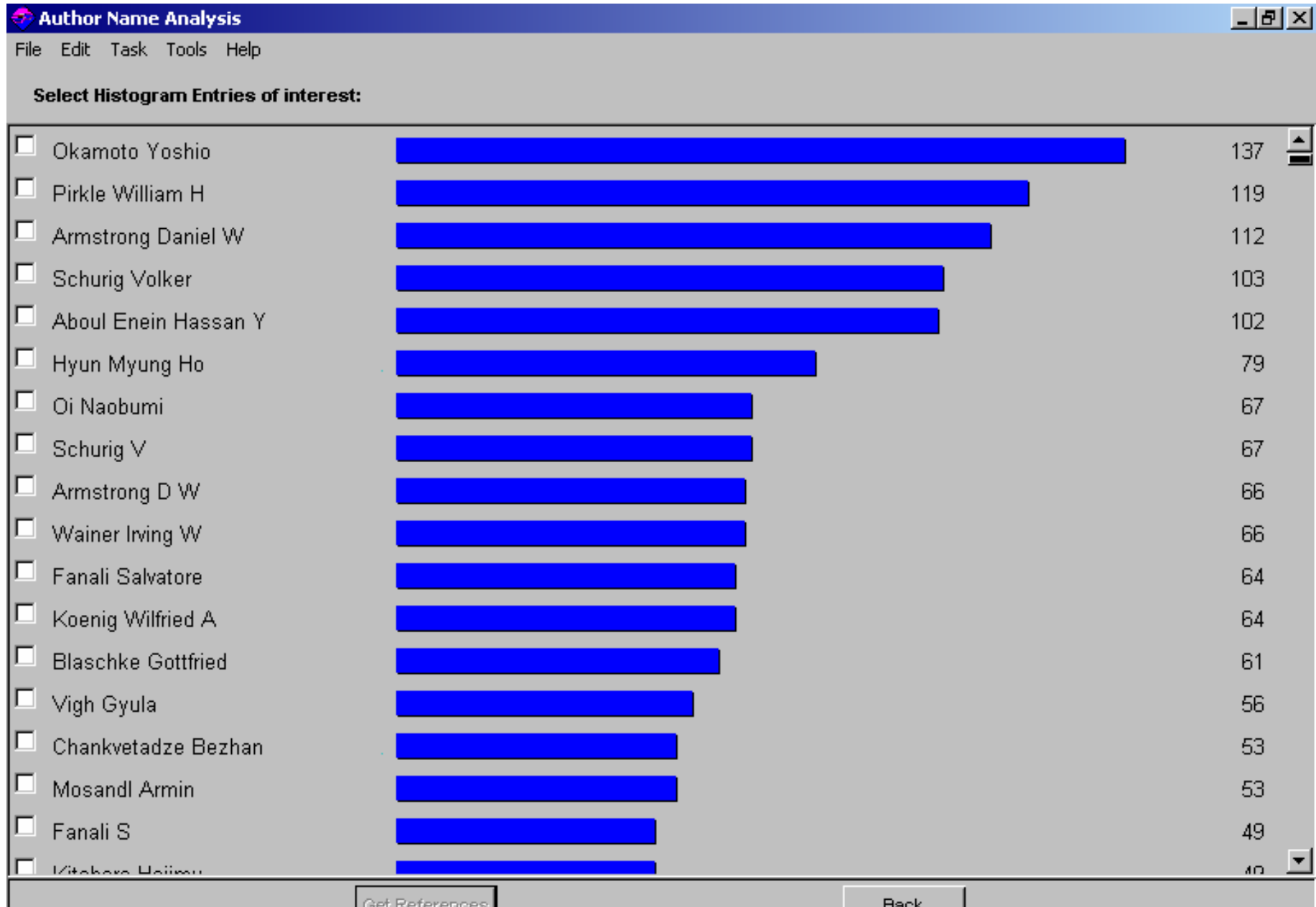


Analyze Choices

- **Author Name** **Index Term**
- **CAS Registry Number** **Journal Name**
- **CA Section Title** **Language**
- **Company/Organization** **Publication Year**
- **Supplementary Term** **Database**
- **Document Type**

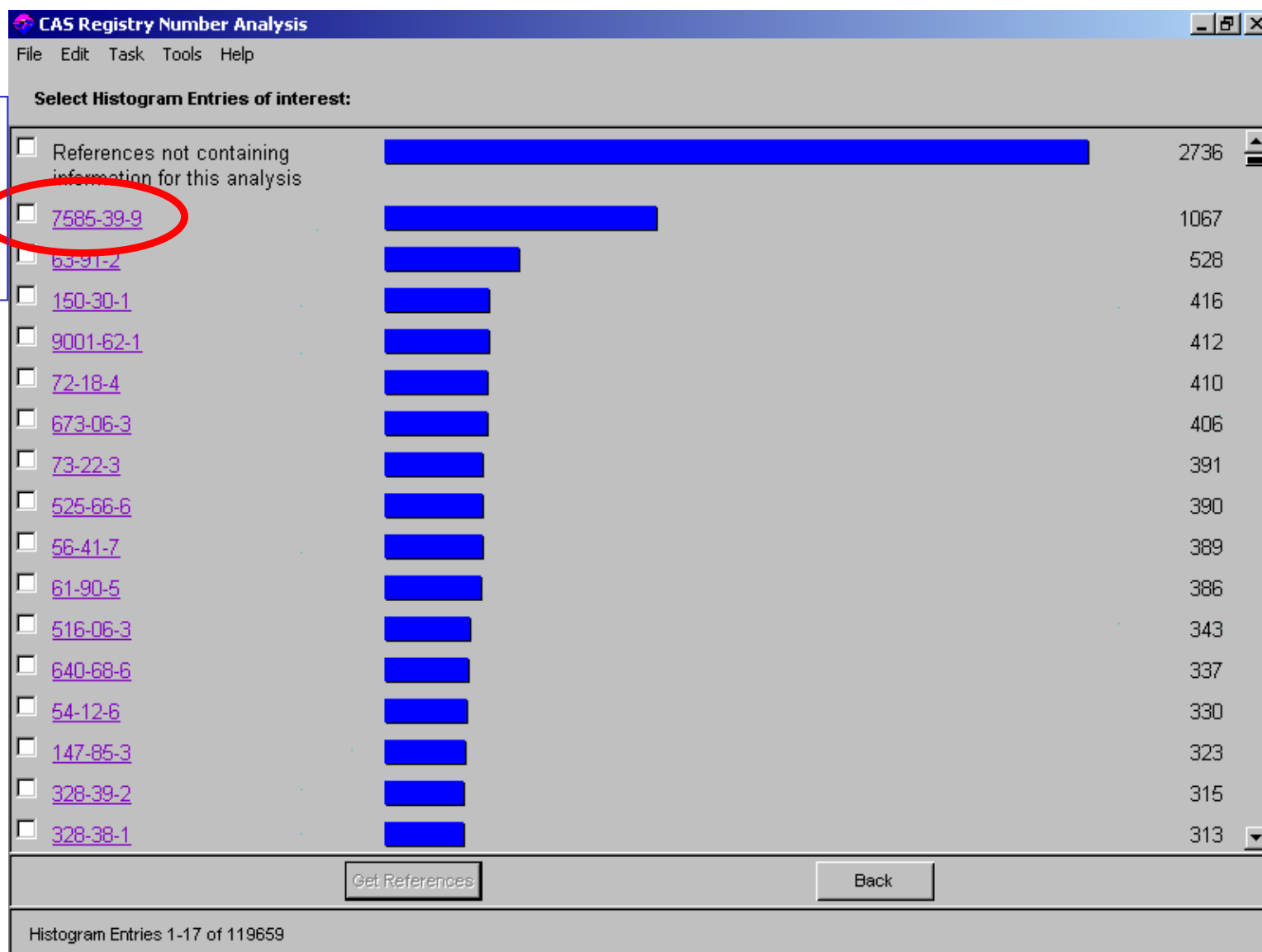
SciFinder Scholar Analyze Tool

Answer set sorted by **author**



CAS Registry Number Analysis

- There are more than 119,659 different substances indexed
- The most frequently indexed compound is β -cyclodextrin

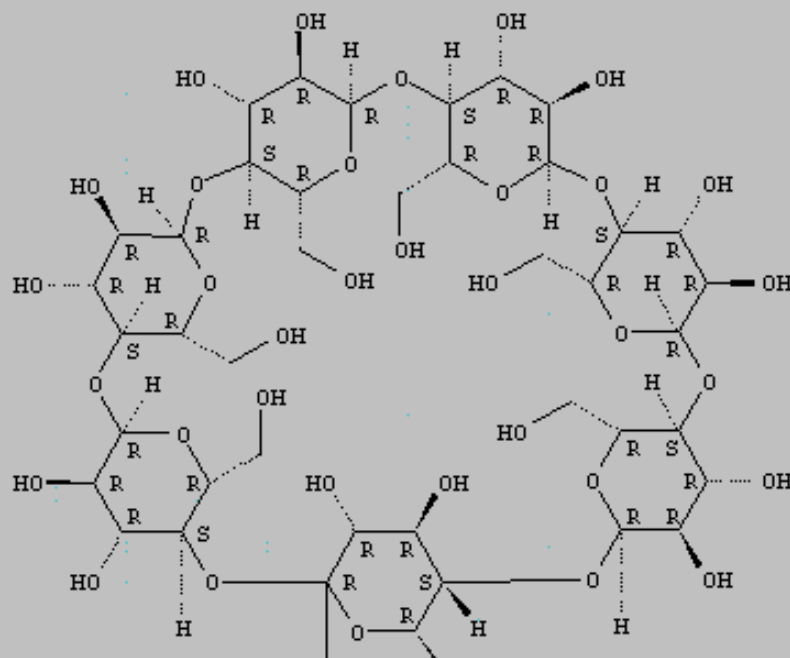


Click to see structure

Registry Number: 7585-39-9

Absolute stereochemistry.

PAGE 1-A



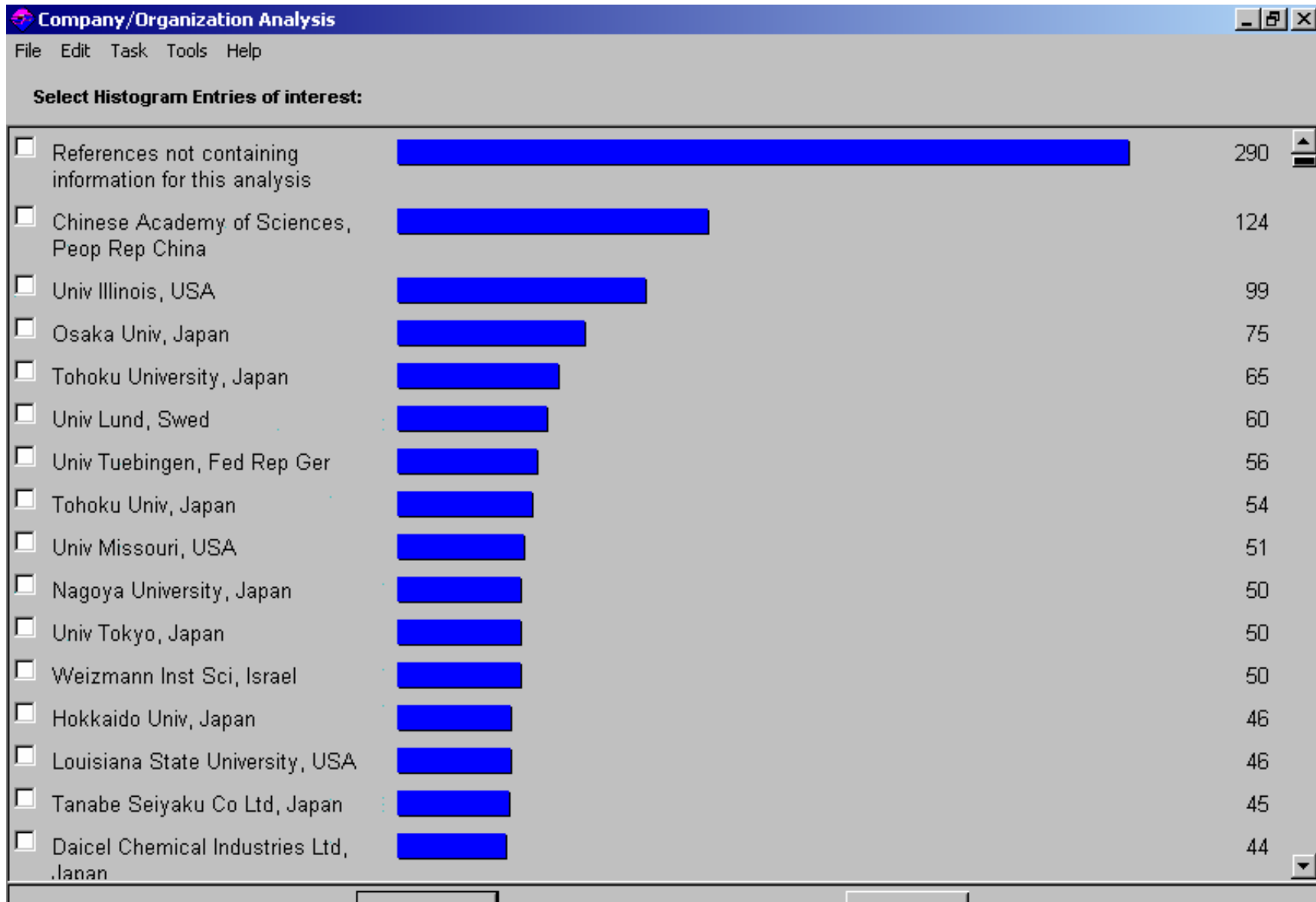
PAGE 2-A



Close

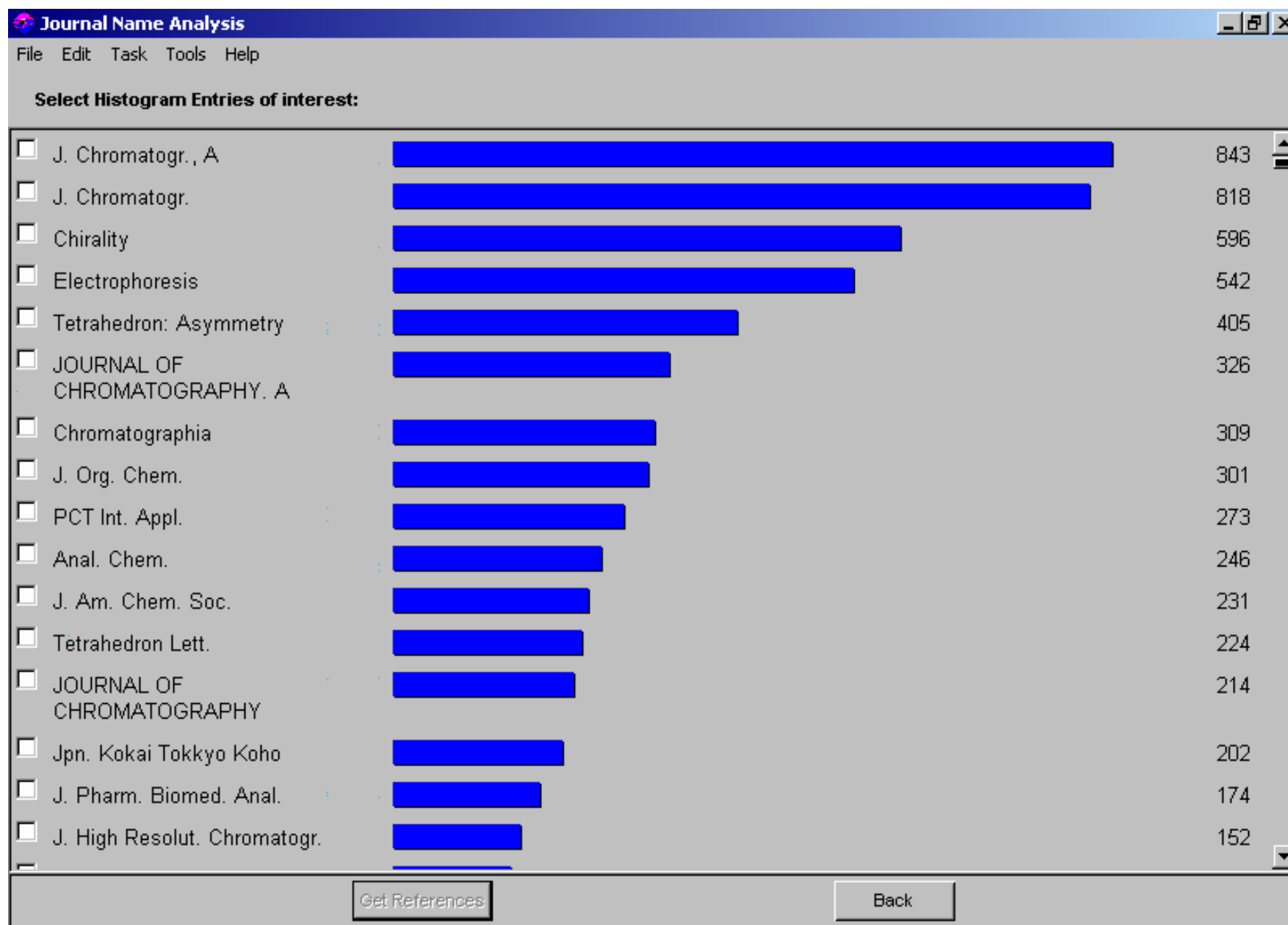
Company/Organization Analysis

- There are over 7,000 companies and organizations represented.
- Top organizations can be cross correlated with the top authors.



Journal Name Analysis

- There are 1619 Journal titles listed



Databases - cont.

Chemical abstracts – SciFinder

Chirbase <http://chirbase.u-3mrs.fr>

Chirbase

A powerful Information System for Chiral Separations of Enantiomers by Chromatography

CHIRBASE is a molecular database which contains :

- 98,000 chiral separations
- 25,000 molecular structures
- 4,000 new separations updated every four months
- A multitude of unpublished Data
- Three types of format : ISIS/Base/Local, ISIS/Host and REACCS

Price

Companies	5000 \$	single PC version	1000 \$ / year for 3 updates
Academia	on request and project		

Columns for Chiral Separation

Chirbase : W3 DataBase - Microsoft Internet Explorer

Datei Bearbeiten Ansicht Favoriten Extras ?

Zurück Suchen Favoriten Medien

Adresse <http://chirbase.u-3mrs.fr/chirbase/database.html> Wechseln zu Links

Google Web-Suche Site-Suche Seiten-Info Aufwärts Hervorheben

Chirbase *W3 DataBase*

W3 DataBase **Chirbase Project**
Commercially Available Chiral Stationary Phases

This database is the first stage of chirbase project : Internet / Intranet.
You can try the Search Engine, but the administration part is reserved to the Chirbase team.

Search Engine Administration

Powered By **Mini SQL**

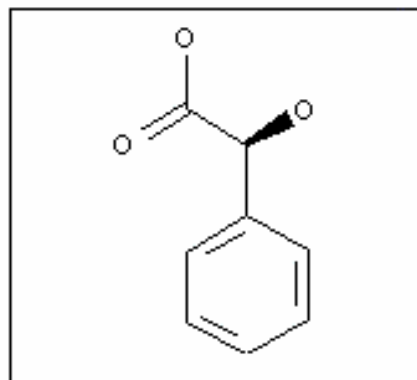
CHEMSCAPE
Now!

For more information
CHIRBASE Office

[[Overview](#) | [Searching Examples](#) | [W3 DataBase](#) | [Email Discussion List](#) | [Laboratory Publication](#) | [Courses & Training](#) | [Chemistry Links](#)]

Internet

<http://chirbase.u-3mrs.fr/chirbase/database.html>

CHIRBASE Summary

Comment:

Sample: Mandelic acid

Chiral Stationary Phases

	<i>name</i>	<i>trade name</i>
1	beta-Cyclodextrin	Cyclobond I
2	Beta-Cyclodextrin	Cyclobond I
3	Beta-cyclodextrin	Cyclobond I

Conditions Litref 2 of 3

	<i>flow_rate</i>	<i>temp</i>	<i>mobile_phase</i>
1	1.00e+000	21.0	4:96 MeOH / Triethylammonium acetate, pH 7.1
2	1.00e+000	25.0	60:40 CH3CN / Phosphate buffer (pH 4.3, 0.1M)
3	1.00e+000	22.0	50:50 CH3OH / H2O

Separation data

RXN		1st	2nd	k'1	k'2	k'1/k'2	Res	Rt1	Rt2
9	1	(?)	(?)	2.04	2.18	1.07	1.40		
	2	(?)	(?)			1.00			
	3	(?)	(?)	0.67	0.67	1.00			
501	1	(D)	(L)	0.83	0.83	1.00			
	2					1.10		4.60	4.80
923	1	S+	R-			1.47	1.91		
	2	(?)	(?)	3.67	6.56	1.78	1.70		

Databases - cont.

Chemical abstracts – SciFinder

Chirbase <http://chirbase.u-3mrs.fr>

Several suppliers provide information about separations carried out using their equipment

e. g. Advanced Separation Technologies
http://www.astecusa.com/applications/index_mod.asp

Books

G. Subramanian, "*Chiral Separation Techniques*", 2nd ed., Wiley VCH, **2001**.

S. Ahuja, "*Chiral Separations by chromatography*", Oxford University Press, **2000**.

Thomas E. Beesley, Raymond P. W. Scott, "*Chiral Chromatography*", John Wiley & Sons, **1998**.

B. Chankvetadze, "*Capillary Electrophoresis in Chiral Analysis*", John Wiley & Sons, **1997**.

W. A. König, "*Gas Chromatographic Enantiomer Separations with Modified Cyclodextrins*", Hüthig Buch Verlag, **1992**.

In press

G. Gubitz, "*Chiral Separations: Methods and Protocols* (Methods in Molecular Biology, 243)", Humana Press, ISBN: 1588291502, in prep, **Nov. 2003**

Other useful links

http://family.swu.ac.kr/~cat/links_chromatography.htm

lists information on

- ACADEMY & INSTITUTE
- LINKS & RESOURCES
- CHROMATOGRAPHY DISCUSSION GROUP & SOCIETY
- MANUFACTURERS & SUPPLIERS
- CORE JOURNALS for CHROMATOGRAPHY & SEPARATION
- Research

Conferences

ISCD-14 / 14th International Symposium on Chirality (2002) / Hamburg, Germany

The *14th International Symposium on Chirality* (ISCD-14) was held at the University of Hamburg, Hamburg, Germany, from September 8 - 12, 2002.

Separation methods

- **Thin layer chromatography, TLC**
- **Gas chromatography, GC**
- **High pressure liquid chromatography, HPLC**
- **Supercritical fluid chromatography, SFC**
- **Capillary electrophoresis, CE**

Efficiency of separation

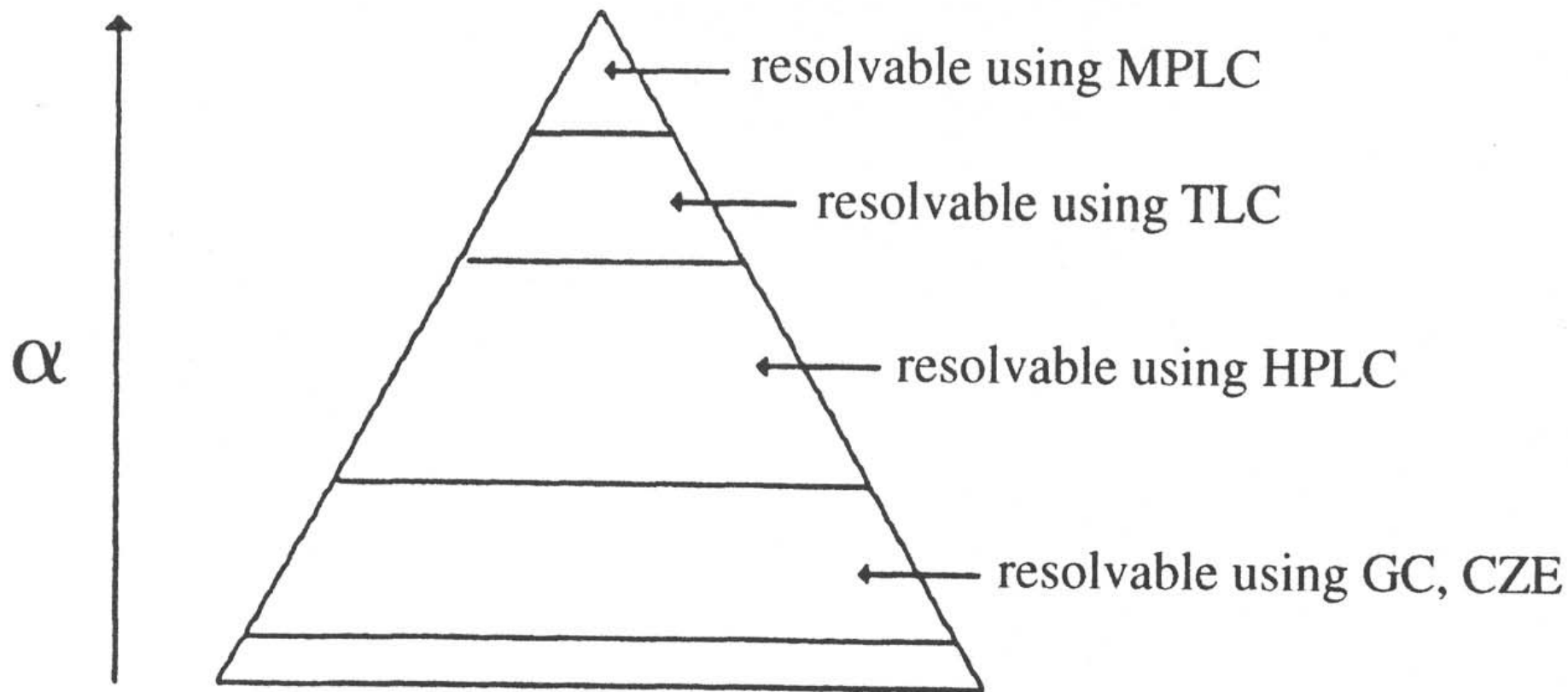
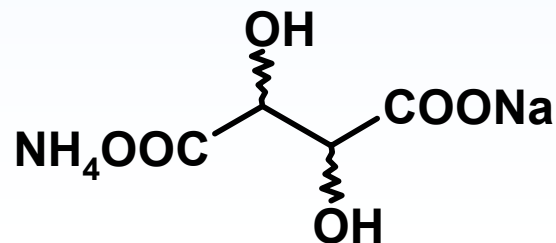
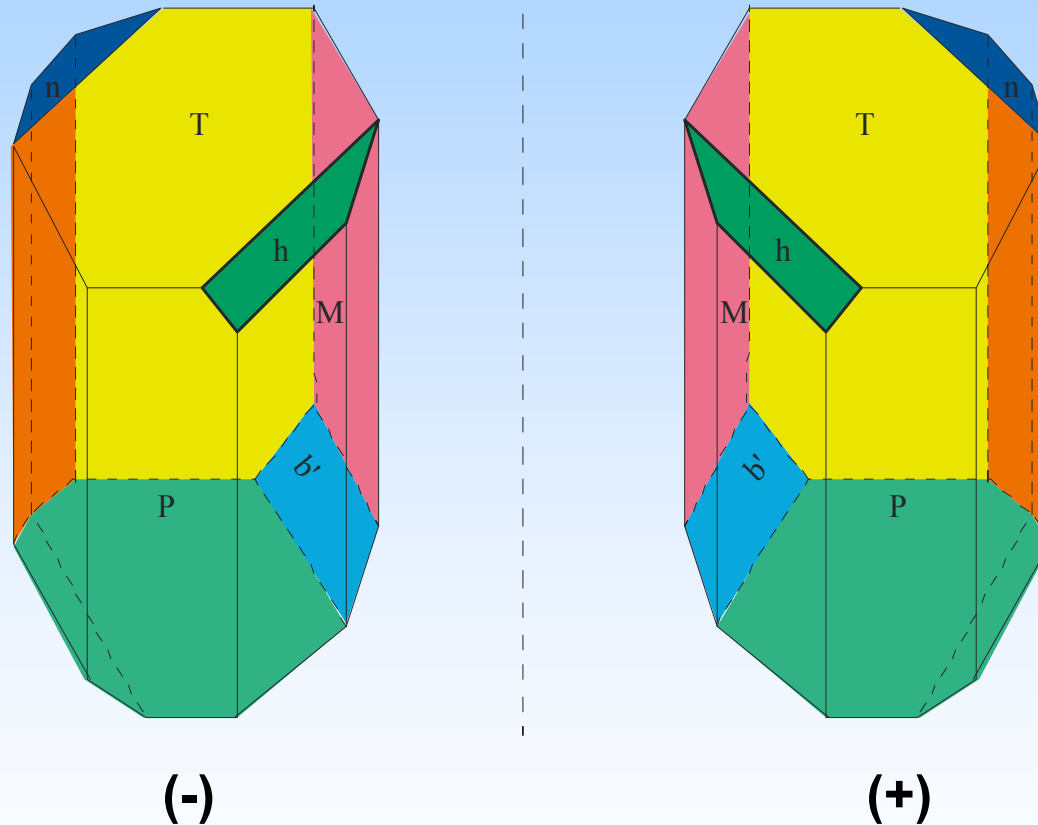


Figure 6.1 Resolution with various chromatographic techniques. Reproduced from S. Ahuja, W. Pirkle, and C. Welch, *Chiral Separations by Chromatography*, American Chemical Society Short Course, 1994. Copyright 1994 American Chemical Society.

The first enantio-separation

Mechanical Separation of the Enantiomers of tartaric acid

Pasteur, L. (1848), *Comptes Rendus, Acad. Sci. (Paris)*, **26**, 535-538



Enantioselective Interactions

Three-point interaction theory (Dalglish, 1952; Pirkle, 1986)

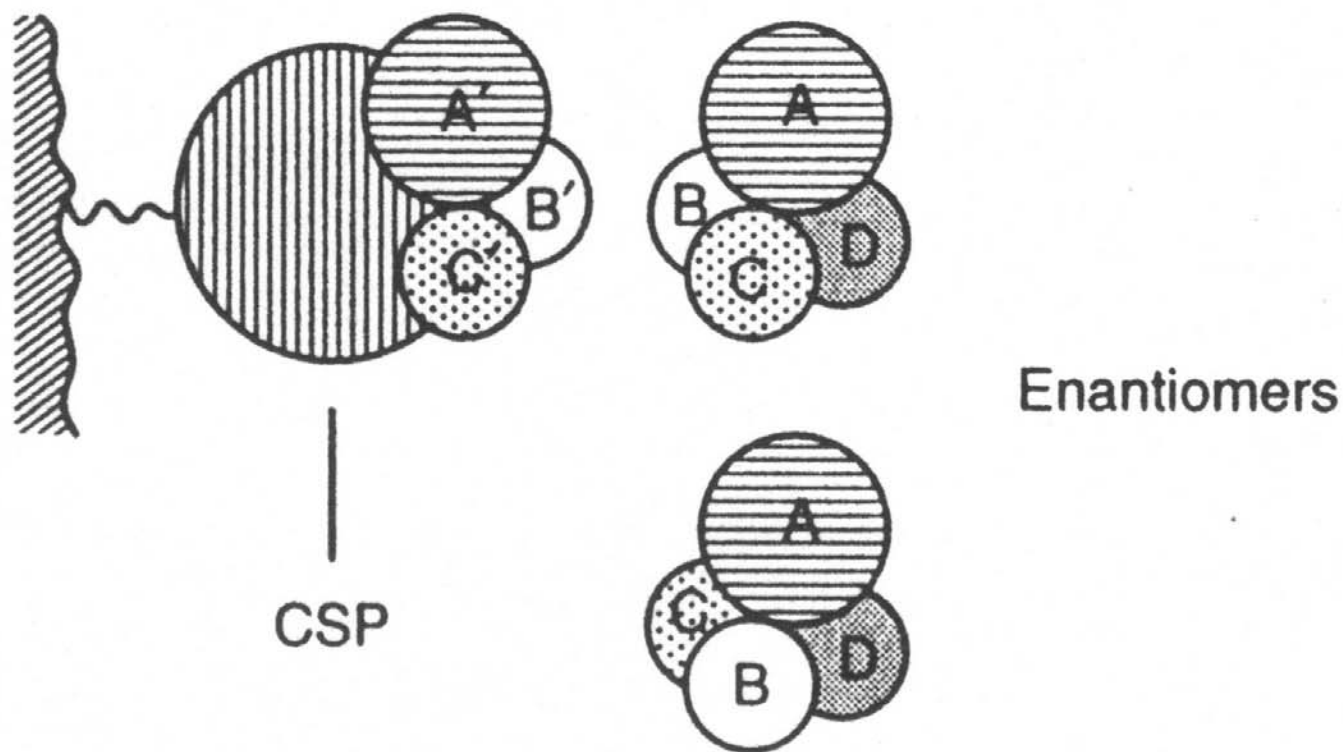


Figure 7.1 A cartoon of three-point interaction.

Enantioselective Interactions

Three-point interaction theory (Dalglish, 1952; Pirkle, 1986)

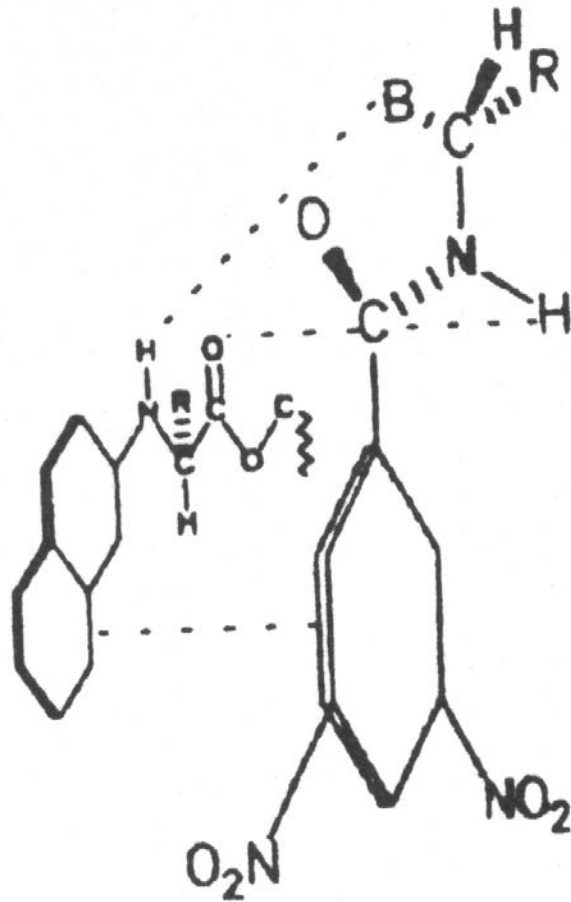


Figure 7.2 A chiral recognition model.

Where to start

1. For enantiomer resolutions: Is my molecule chiral?
2ⁿ stereoisomers (n = number of stereogenic centers)
2. Of course: Literature and Database search
3. Equipment available, Costs for new equipment
4. Method selection (personal opinion)

Method	Con	Pro
TLC	qualitative semi-quantitative	fast for method develop.
GC	Only for volatile compounds Sometimes derivatization required	Long columns possible
HPLC		Often applicable without derivatization

GC vs HPLC

	GC	HPLC
Column efficiency	Very high	Moderate
Separation factors, α	1.05 – 2	Up to 30
Temperature influence	Important for medium to high $\Delta(\Delta H)$	Little known

Temperature influence

HPLC

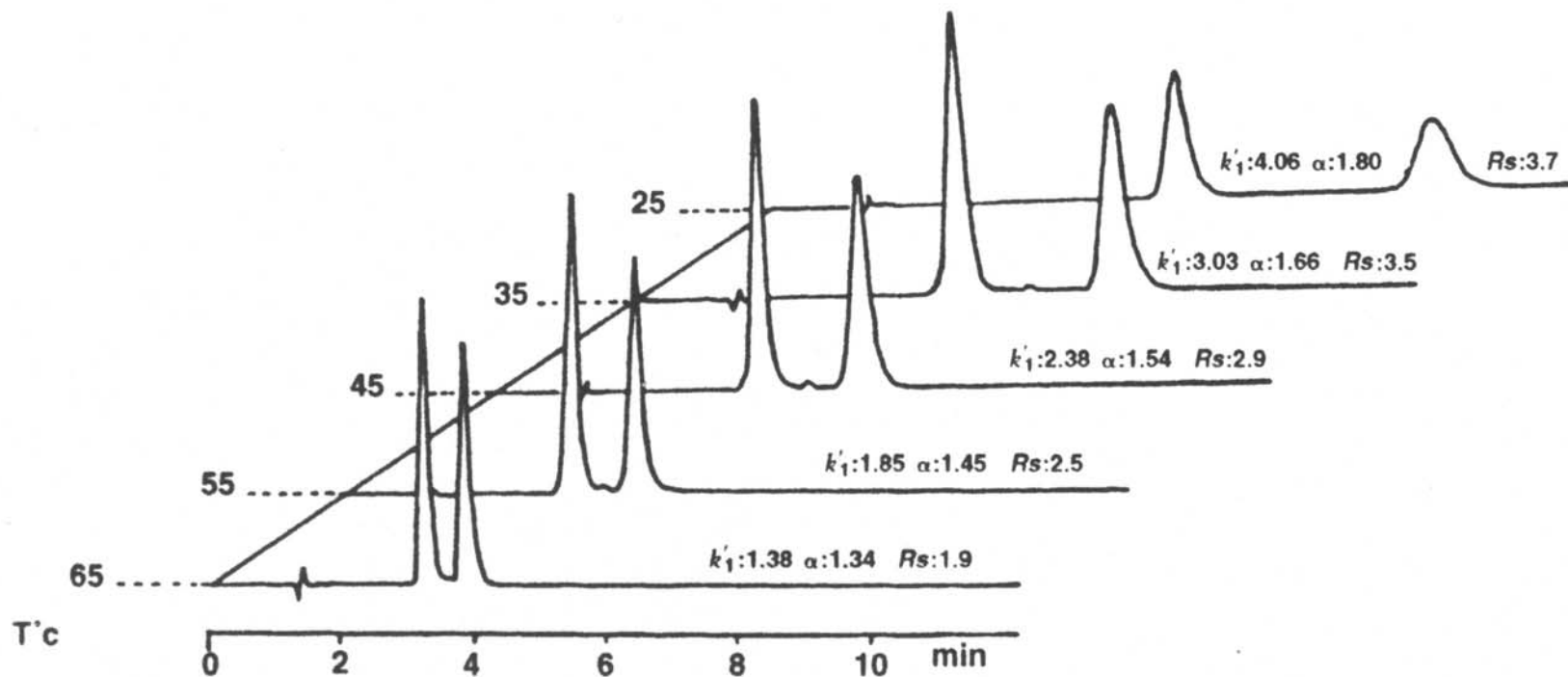


Figure 7.7 Temperature influence on k' , α and resolution of an amido compound. Reproduced with permission from F. Gasparrini, D. Misiti, and G. Villani, in *Recent Advances in Chiral Separations*, D. Stevenson and I. D. Wilson, Eds., Plenum, New York, 1990. Copyright 1990 Plenum Publishers.

- In general:
- α increased with *decreasing* temperature
 - 1 - 2% per °C

Choice of the stationary phase

Table 8.1 Classification of Chiral Stationary Phases (CSP)

Type	Description	Examples	Usual Mode
1	Brush-type (donor acceptors)	DNB-glycine, DNB-leucine, naphthylalanine	Normal phase (polar modifier)
2	Cellulose triesters or carbamates on silica	Chiralcel OA, OB, OF, OJ, etc.	Normal phase (polar modifier)
3	Inclusion CSPs, e. g. cyclodextrins, crown ethers, polyacrylates, polyacrylamides	Cyclobond 1-3, Chiralcel CR, Chiralpak OP, OT, Merck grafted	Reversed phase, e. g. aqueous MeCN or MeOH
4	Ligand exchangers	Proline, OH-proline	Reversed phase (aqueous buffer)
5	Proteins	Albumin, glycoprotein	Reversed phase (aqueous buffer)

Choice of the stationary phase

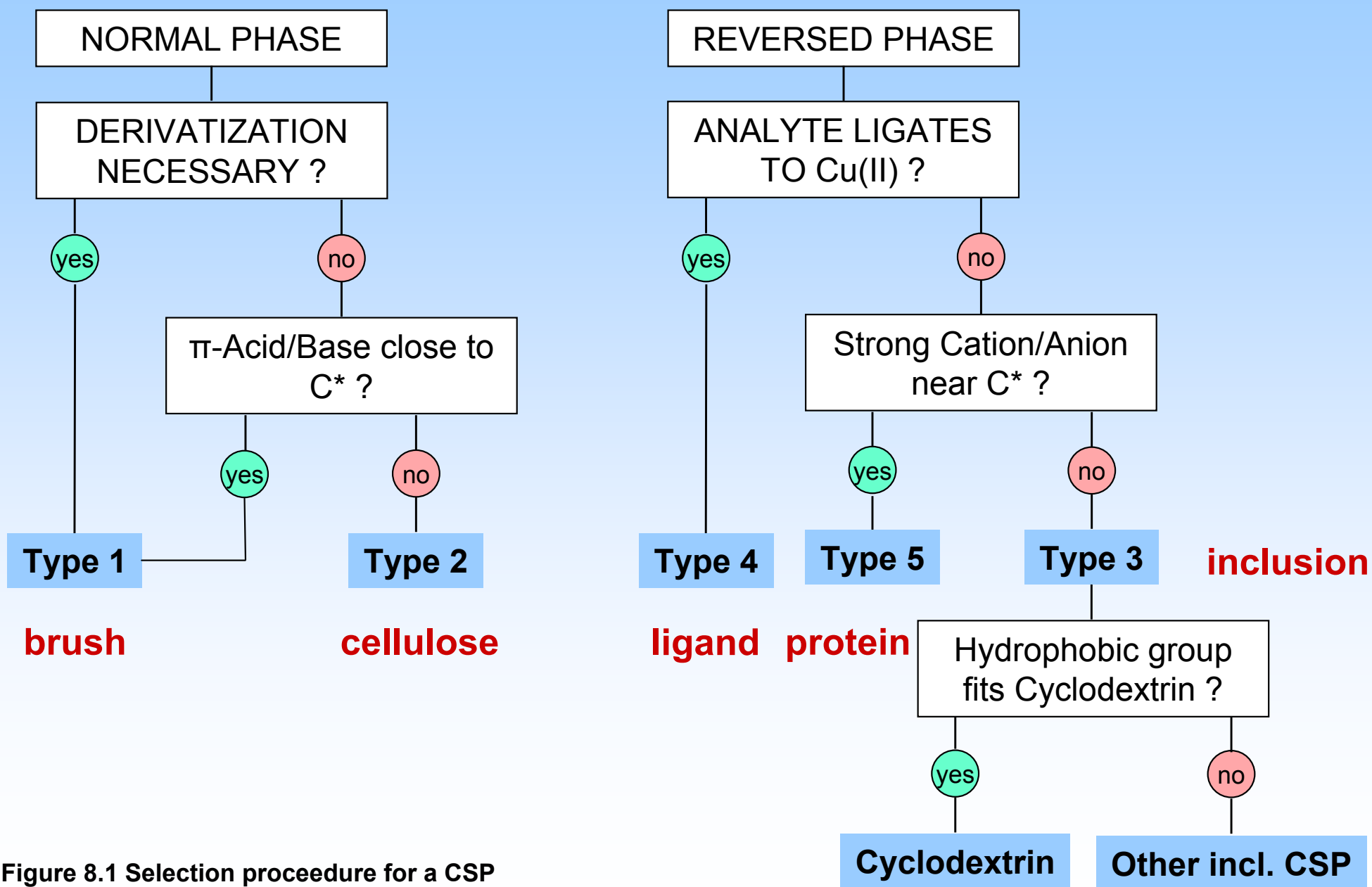
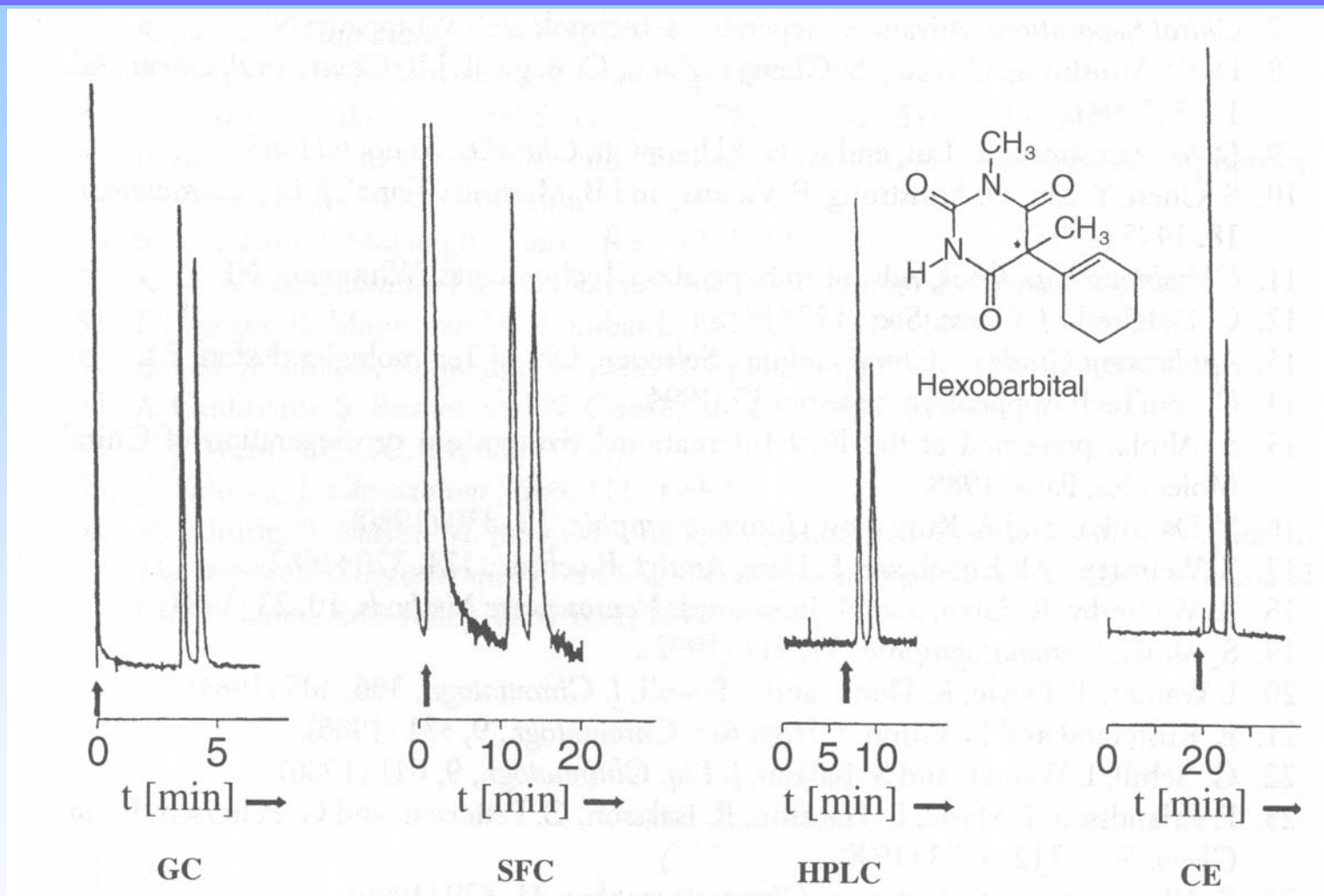


Figure 8.1 Selection procedure for a CSP

Comparative Separations



Results: α for CE = HPLC > SFC > GC
 R_s for CE > HPLC > GC > SFC
 N (first peak) for CE > HPLC = GC > SFC
 CE slowest method

The End