Journal of Chromatography A, 666 (1994) 557–563
Elsevier Science B.V., Amsterdam

CHROM. 25 879

CHIRBASE, a molecular database for the separation of enantiomers by chromatography

Bernhard Koppenhoefer*, Rudolph Graf, Helga Holzschuh, Andreas Nothdurft and Ulrich Trettin

Institut für Organische Chemie der Universität, Auf der Morgenstelle 18, D-72076 Tübingen (Germany)

Patrick Piras and Christian Roussel

ENSSPICAM, URA 1410, F-13397 Marseille Cedex 13 (France)

ABSTRACT

A selected review on the separation of the stereoisomers of menthol by gas chromatography serves to demonstrate a new approach to tackle the increasing number of publications on enantiomer separation. The graphical molecular database CHIRBASE covers information (structural, bibliographic and chromatographic data) on liquid chromatography, supercritical fluid chromatography and gas chromatography. CHIRBASE is based on standard database software, thus meeting the requirements of "state of the art" information management in the chemical and pharamaceutical industries and academia. The scrutiny required to create and maintain a factual database revealed that the information content of the contemporary literature with respect to chromatographic data requires improvement.

INTRODUCTION

Enantiomers often show huge differences in biological activities. Given a racemic drug, the inactive enantiomer must be considered like any other impurity. As a consequence, single enantiomer drug development has become the common standard in Japan, and other countries are likely to follow this trend after the recent policy statement by the US Food and Drug Administration [1]. It may be expected that synthetic food additives, fragrances, pesticides and herbicides will be treated along the same lines. These restrictions on the sale of racemic compounds lead to a strong demand for information about how to obtain and test enantiomeric samples in every stage of research.

The separation of enantiomers on a chiral stationary phase (CSP) is performed for two major reasons: (i) on a preparative scale to obtain enantiomerically pure compounds and (ii) on an analytical scale to determine the enantiomeric purity of a chiral component in a sample of chemical or biological origin. Until January 1994, more than 230 chiral stationary phases for GC have been described in the literature, 40 of which are commercially available. In LC and supercritical fluid chromatography (SFC) more than 150 of 530 known CSPs have been commercialized.

PURPOSE OF CHIRBASE

In the initial state of a particular separation task, the analyst is confronted with the crucial question of which CSP he or she should select and what the optimum operating conditions are. To give an appropriate answer, we have created the database CHIRBASE, designed to store and retrieve the user's own experiments along with

* Corresponding author.

0021-9673/94/$07.00
SSDI 0021-9673(94)00016-3

separations taken from the literature. As the chromatographic separation of enantiomers on a CSP is based on a molecular recognition mechanism, a graphical molecular database, allowing direct access to the molecular structure, is certainly the method of choice.

Present status

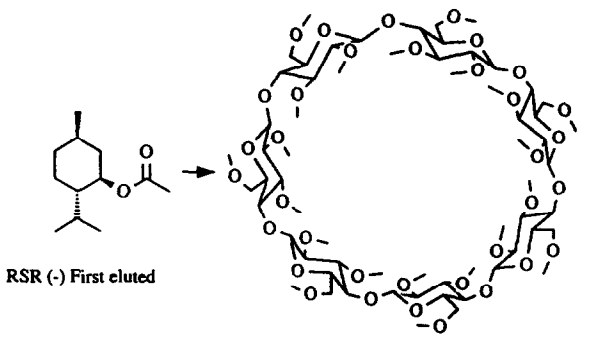
CHIRBASE currently (January 1994) covers more than 31 400 chromatographic separations of approximately 12 000 enantiomeric pairs by LC, SFC and GC; additional methods are in preparation. Without resorting to a time-consuming literature search, the chromatographic conditions may be retrieved and sorted within a few seconds. Even for a new separation problem, a sub-structure search (see below) will provide the user with a list of the most promising candidates out of more than 760 CSPs contained in CHIRBASE.

Software and hardware requirements

Originally, CHIRBASE was built on the database software ChemBase (MDL), a single-user solution running under DOS on an IBM PC and compatibles, preferably 386/33 MHz or higher; the whole database package including ChemBase requires a total of 50 MB of free disk space.

MOL NAME	Menthyl acetate					
	Fst	RSR	Rt1	k'1	k2/k1	CHIRALITY
ID 1392	Sec	SRS	Rt2	k'2	RES 1.00	3 Centers

Structure



RSR (-) First eluted

METHOD GC	AMOUNT	Analytical	
DETECTION FID	COLUMN TREATMENT		
CARRIER GAS	Helium	TEMP(°C)	
TYPE OF COLUMN	Fused-silica 25 m * 0.25 mm		
FLOW-RATE(ml/min):	INLET PRESSURE(bar):	1.00	
CSP NAME	2,3,6-Tri-O-methyl-beta-cyclodextrin		
TRADE NAME	FS-CYCLODEX beta-I/P		
SUPPLIER	CS-Chromatographie Service, Langerwehe, FRG	CSP NO 54	
AUTHOR	Werkhoff, P.; Brennecke, S.; Bretschneider, W.		
JOURNAL	Chem. Mikrobiol. Technol. Lebensm.		
REF NO 20363	YEAR 1991	VOLUME 13	PAGE 129-152

Multicolumn. Chromatogram reported under temperature programming: 40°C, 2°C/min, 200°C. Baseline separation. Underestimated resolution. Data kindly checked by the authors.

Fig. 1. Reaction database of CHIRBASE/GC (example: menthyl acetate).

More recently, major changes in the database structure were made to adapt CHIRBASE to MDL's new software ISIS/Base that can be used on both IBM PC (under Windows) and Macintosh, and also to MDL's mainframe software REACCS and MACCS (for VAX computers), as well as ISIS/Host (for VAX and UNIX workstations).

Database organization and search example

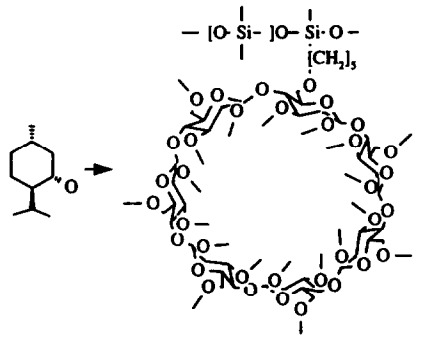
CHIRBASE is divided into different sections, each representing a particular chromatographic method. One of these sections is CHIRBASE/GC, again divided into three parts: a reaction database for full information (Figs. 1 and 2), a molecular database for fast retrieval of analytes (Fig. 3) and another molecular database for supplementary information on the CSPs (Fig. 4).

After drawing the molecular structure of the analyte (*i.e.*, Fig. 3) with a few mouse clicks, the user will find eleven entries for menthol. Separate searches for neomenthol and for isomenthol are possible, each yielding another five entries. The graphical visualization of the molecular structures of both analyte and CSP in the reaction database (Figs. 1 and 2) gives the chemist a better idea of the intermolecular interactions possibly involved.

In order to compare and sort the individual separation conditions on the various CSPs re-

MOL NAME			Menthol				
	Fst	Rt1	12.5	k'1	18.83	k2/k1 1.040	CHIRALITY
ID 380	Sec	Rt2	13.1	k'2	19.59	RES 1.71	3 Centers

Structure



METHOD GC	AMOUNT	Analytical (split 1:100)	
DETECTION FID	COLUMN TREATMENT	None	
CARRIER GAS Hydrogen	TEMP(°C)	110	
TYPE OF COLUMN Fused-silica 25 m * 0.25 mm			
FLOW-RATE(ml/min):	INLET PRESSURE(bar):	1.00	
CSP NAME 2,3,6-Tri-O-methyl-beta-CD-pentamethylen-polysiloxane			
TRADE NAME Chirasil-Dex 2			
SUPPLIER Chrompack, Middelburg, NL	CSP NO 11		
AUTHOR Schurig, V.; Schmalzing, D.; Mühleck, U.; Jung, M.; Schleimer, M.; Mussche, P.; Duvekot, C.; Buyten, J.C.			
JOURNAL J. High Res. Chromatogr.			
REF NO 20093	YEAR 1990	VOLUME 13	PAGE 713-717

Chromatogram reported. Baseline separation. No order of elution. Data expected from the authors.

Fig. 2. Reaction database of CHIRBASE/GC (example: menthol).

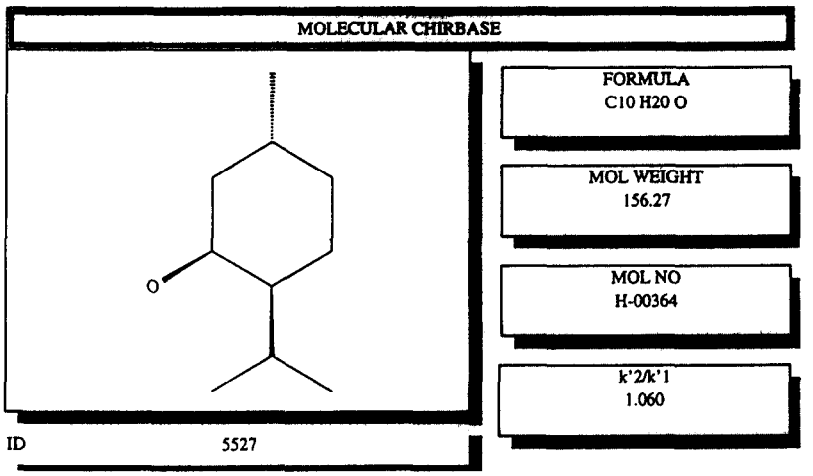


Fig. 3. Molecular database of analytes (example: menthol).

ported, the data are compiled in a table (Table I), making trends easy to recognize. Thus, CHIRBASE allows the user to evaluate the most suitable separation conditions, including possible derivatization methods, operating conditions and commercial availability of chiral stationary phases. Derivatives are found by a sub-structure search for a given analyte in the same run. For example, on CSPs of the cyclodextrin type the acetyl derivative of menthol appears to be the most suitable (Fig. 1) [7,10,11], whereas the separation of the isopropylurethane derivative on an amide phase provides an alternative solution [12]. If the separation of the underivatized analyte is given a high priority, a metal complex phase [2] is still first choice for this particular molecule (Table I), although a user may find it easier to access the polysiloxane-bound cyclodextrin depicted in Fig. 2.

INFORMATION CONTENT IN THE LITERATURE

The information content of papers referring to the separation of enantiomers by chromatography is not very standardized, and is generally fairly low. Publications with a main focus on the synthesis of enantiomers, on the one hand, seldomly provide the analytical data of interest. This is unfortunate, as these publications today constitute a major proportion (several thousand)

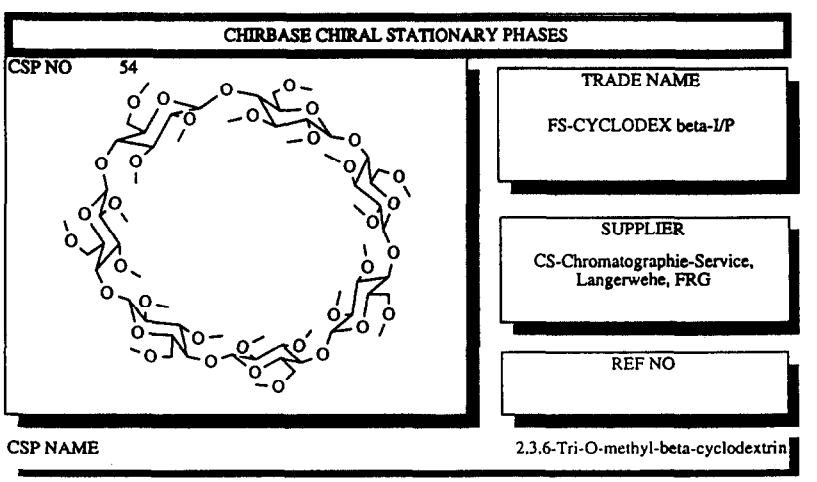


Fig. 4. Molecular database of stationary phases (example: 2,3,6-tri-O-methyl-β-cyclodextrin)

TABLE I

SELECTED DATA FROM CHIRBASE/GC FOR MENTHOL, NEOMENTHOL AND ISOMENTHOL

Compound	Chiral stationary phase[a]	α[b]	First eluted	T (°C)[c]	Ref.
Menthol	Ni(II) bis[3-heptafluorobutanoyl-(1*R*)-camphorate][d]	1.060	*RSR*	110	2
	2,3,6-Tri-O-methyl-β-cyclodextrin pentamethylenepolysiloxane[d]	1.040	–[e]	110	3
	2,3,6-Tri-O-methyl-β-CD[d]	1.039	–	85	3
	2,3,6-Tri-O-methyl-β-CD trimethylenepolysiloxane[d]	1.037	–	110	3
	N-(1R,3*R*)-*trans*-Chrysanthemoyl-(*R*)-1-(α-naphthyl)ethylamine	1.018	–	80	4
	2,3,6-Tri-O-methyl-β-CD (DB-1701)[d]	1.017	*SRS*	105	5
	2,6-Di-O-methyl-3-O-pentyl-β-CD (OV-1701)	1.004	–	TP	6
	2,3,6-Tri-O-methyl-γ-CD (OV-1701-OH)	–[e]	–	TP	7
	2,3,6-Tri-O-methyl-α-CD (OV-1701-OH)	–	–	TP	7
	2,6-Di-O-pentyl-3-O-methyl-β-CD (OV-1701)	1.000	–	–[e]	6
	2,3,6-Tri-O-pentyl-β-CD (OV-1701)	1.000	–	–	6
Neomenthol	2,3,6-Tri-O-methyl-β-CD[d]	1.059	–	85	3,8
	2,3,6-Tri-O-methyl-β-cyclodextrin pentamethylenepolysiloxane[d]	1.047	–	110	3
	2,6-Di-O-methyl-3-O-TFA-β-CD	1.030	–	100	8
	2,3,6-Tri-O-methyl-β-cyclodextrin trimethylenepolysiloxane[d]	1.038	–	110	3
	2,3,6-Tri-O-methyl-β-CD (DB-1701)[d]	1.018	*SRS*	105	5
Isomenthol	2,3,6-Tri-O-methyl-β-cyclodextrin pentamethylenepolysiloxane[d]	1.058	–	110	3
	2,3,6-Tri-O-methyl-β-cyclodextrin trimethylenepolysiloxane[d]	1.055	–	110	3
	2,6-Di-O-methyl-3-O-TFA-β-CD	1.040	–	100	8
	2,3,6-Tri-O-methyl-β-CD[d]	1.034	–	85	3,8
	2,3,6-Tri-O-methyl-β-cyclodextrin 5-octen-1-ylsiloxane[d]	–	–	TP	9
O-Acetyl derivative	2,3,6-Tri-O-methyl-β-CD (OV-1701-OH)[d]	1.110	–	–	7
	2,3,6-Tri-O-methyl-β-CD (OV-1701)[d]	1.043	–	140	10
	2,3,6-Tri-O-methyl-β-CD[d]	–	*RSR*	TP	11
O-Isopropylurethane derivative	XE-60–(*S*)-Valine-(*S*)-α-phenylethylamide	–	–	–	12

[a] CD = Cyclodextrin; TFA = trifluoroacetyl.
[b] $\alpha = k_2'/k_1'$,
[c] TP = Temperature programming.
[d] Commercial phase.
[e] Not published.

of the different analytes stored in the database. On the other hand, most papers introducing new stationary phases, typically published in journals specializing in analytical chemistry, tend to deal with a restricted set (only a few hundred) of test compounds. Even those papers often do not contain all data necessary for a complete description of the separation experiments.

Given that the molecular structures of both analyte and stationary phase can be seen from the article itself or other information sources, the authors should at least provide the following

data, under isothermal operation conditions: temperature (T); column dimensions; dead time (t_0); net retention times (t_R) and/or reduced data derived therefrom, *i.e.*, resolution factor (α) and net capacity factors (k'); peak resolution (R); flow-rate and/or pressure applied at the column head; and carrier gas. Further data are optional, such as detection method, amount injected, possible pretreatment of the column wall and origin of the column. Bearing in mind that an unambiguous peak assignment is the first step in the characterization of any chromatographic experiment, knowledge of the elution order is essential. This shows in any application of the separation, such as preparative chromatography, determination of the enantiomeric excess and modelling the intermolecular interaction.

The frequency of three crucial figures, *i.e.*, separation factor (α), capacity factors (k') and peak resolution (R), in a sample of 488 papers is depicted in Fig. 5; thus, only 17% of the papers contain all three figures. According to Postma and Kateman [13], a similar deficiency in information content is a problem inherent to analytical chemistry in general. In order to improve this situation, the CHIRBASE project attempts to update all relevant information through correspondence with authors. The central collection of a defined set of chromatographic data prior to publication will be one of the future challenges in this field. The shining example is X-ray structural analysis, where there is general agreement on the pathway and content of information on the molecular structure to be delivered to the Cambridge Crystallographic Data File, prior to publication. Today, information on compound properties and chemical reactions is spread worldwide by a few authorized institutions, *e.g.*, the Scientific and Technical Network (STN), maintained jointly by *Chemical Abstracts* (CA) in the USA, the *Fachinformationszentrum* (FIZ) in Germany and the *Japan Information Centre of Science and Technology*. It may be hoped for the future that CHIRBASE will have an equally rationalizing effect on the chromatographic community.

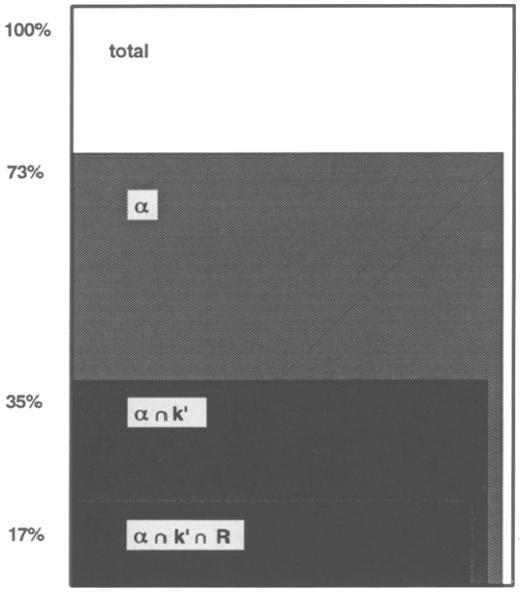


Fig. 5. Information content of 488 publications on the resolution of enantiomers by gas chromatography on a chiral stationary phase. Frequency of separation factor (α), capacity factors (k') and peak resolution (R).

CONCLUSIONS

The database package CHIRBASE does not replace the chemist's mind in evaluating possible solutions for a given separation task; however, it provides him or her with virtually all the information necessary for a rational decision. A substantial increase in efficiency of the chromatographic experiment can be expected from this approach.

REFERENCES

1 Department of Health and Human Service of the USA, Food and Drug Administration's Policy Statement for the Development of New Stereoisomeric Drugs, *Fed. Regist.*, 57, 2 May (1992) 102; *Science*, 256 (1992) 965.
2 C. Bicchi and A. Pisciotta, *J. Chromatogr.*, 508 (1990) 341–348.
3 V. Schurig, D. Schmalzing, U. Mühleck, M. Jung, M. Schleimer, P. Mussche, C. Duvekot and J.C. Buyten, *J. High Resolut. Chromatogr.*, 13 (1990) 713–717.
4 N. Oi, H. Kitahara, Y. Inda and T. Doi, *J. Chromatogr.*, 213 (1981) 137–141.
5 F & W Scientific, *J. Chromatogr. Sci.*, 29 (1991) 89.
6 C. Bicchi, G. Artuffo, A. D'Amato, V. Manzin, A. Galli and M. Galli, *J. High Resolut. Chromatogr.*, 15 (1992) 710–714.

7 C. Bicchi, G. Artuffo, A. D'Amato, G.M. Nano, A. Galli and M. Galli, *J. High Resolut. Chromatogr.*, 14 (1991) 301–305.
8 H.-P. Nowotny, D. Schmalzing, D. Wistuba and V. Schurig, *J. High Resolut. Chromatogr.*, 12 (1989) 383–393.
9 V. Schurig, D. Schmalzing and M. Schleimer, *Angew. Chem.*, 103 (1991) 994–996.
10 *Gas-Chromatographie Katalog*, Macherey–Nagel, Düren, 1992, pp. A52–A101.
11 P. Werkhoff, S. Brennecke and W. Bretschneider, *Chem. Mikrobiol. Technol. Lebensm.*, 13 (1991) 129–152.
12 G. Langrand, M. Secchi, G. Buono, J. Baratti and C. Triantaphylides, *Tetrahedron Lett.*, 26 (1985) 1857–1860.
13 G.J. Postma and G. Kateman, *Anal. Chim. Acta*, 265 (1992) 133–155.