Combinatorial Chemistry

Synthesis, Analysis, Screening

Edited by
Günther Jung

WILEY-VCH

Weinheim · New York · Chichester · Toronto · Brisbane · Singapore

This Page Intentionally Left Blank

Combinatorial Chemistry

Edited by Günther Jung

WILEY-VCH

This Page Intentionally Left Blank

Combinatorial Chemistry

Synthesis, Analysis, Screening

Edited by
Günther Jung

WILEY-VCH

Weinheim · New York · Chichester · Toronto · Brisbane · Singapore

Prof. Dr. Günther Jung
Institut für Organische Chemie
Universität Tübingen
Auf der Morgenstelle 18
D-72076 Tübingen
Germany

Reprint 2001

Library of Congress Card No: applied for

British Library Cataloguing-in-Publication Data: A catalogue record for this book is available from the British Library

Die Deutsche Bibliothek – CIP Einheitsaufnahme
Ein Titeldatensatz für diese Publikation ist bei der Deutschen Bibliothek erhältlich.

Composition: Datascan GmbH, D-64295 Darmstadt
Printing: Strauss Offsetdruck, D-69509 Mörlenbach
Bookbinding: Osswald & Co. D-67433 Neustadt (Weinstraße)

Preface

Since I edited *Combinatorial Peptide and Nonpeptide Libraries* – the first comprehensive book on the subject – in 1996, progress in the field of combinatorial chemistry has been truly dramatic. However, before I began to write this preface, I looked back and found the early work by no means outdated – as evidenced by the number of complimentary letters received from satisfied readers. A number of excellent books on combinatorial chemistry have appeared since then, covering different aspects of the subject. However, despite this earlier 'boom', I was encouraged to edit a second volume, and recognized that soon there will be very little overlap of *Combinatorial Chemistry* with the contents of other such books.

The past five years have seen the development and introduction of new and improved approaches to the combinatorial sciences, their practical applications having resulted in an almost 'explosive' increase in the number of experimental publications and new journals devoted exclusively to these areas. Thus, a comprehensive book which emphasizes the state of the art of most of the methods and instrumentation of combinatorial chemistry may be of value to those who may consider entering this exciting field. It is possible also that experts in combinatorial organic chemistry may find it interesting to examine more closely exciting new fields such as catalyst research, chemosensor development, new analytical tools, and the creation of biodiversity.

As a logical consequence of the demand for combinatorial chemists in industry, teaching the subject and related key technologies should be a major topic at our universities. It is hoped that some of the chapters of this book are sufficiently convincing as to encourage future scientists not only to use combinatorial methods in their research, but also to offer training for their students or co-workers. Indeed, for some years, I have offered public courses in multiple methods in peptide chemistry, and presently my group teaches theoretical and practical methods in combinatorial organic chemistry.

As we approach the end of the 20th century, we observe two major trends in the drug discovery process: first, the production of large numbers of single compounds and complex libraries in small scale (microgram) quantities; and second, the production of preparative-scale amounts (mg) of single compounds with complete analytical characterization and defined purity criteria, and less complex mixtures. At present, the nanoscale synthesis and on-line screening of 100000 compounds per day remains a dream for organic chemists – except perhaps for primitive compounds and rather simple biological targets. Nonetheless, such wishes may be realized for oligomers accessible by repetitive synthetic steps.

Combinatorial Chemistry begins with a general overview on recent advances in the subject, with expert surveys provided on selected solid-phase organic reactions and – for the first time – also on solution-phase combinatorial chemistry. These chapters are followed by a detailed survey of the broad research into multicomponent reactions.

The chapter on solid supports in the first book has now been supplemented by a complete listing of the various solid-phase anchors and linkers used in organic chemistry – information which may be of particular value to newcomers in the field.

As a consequence of the progress and success of diverse oligomer libraries, experts in combinatorial synthetic oligomers, glycopeptide and oligosaccharide libraries, RNA- and DNA-aptamers have each contributed their news in these fields, while novel experimental examples of the use of templates in combinatorial chemistry for the solid-phase synthesis of multiple core structure libraries provide some insight to praxis-relevant work.

A complete chapter is devoted to novel combinatorial approaches to highly selective chemoreceptors designed for parallel analysis by sensor devices. An assay-oriented chapter on peptide libraries in T-cell-mediated immune response illustrates how libraries of extreme complexity (10^{13}) can lead directly to single molecules with activities that may be up to 100000-fold higher than those of natural epitopes.

Results of combinatorial biosynthesis using gene clusters responsible for metabolites of potential pharmaceutical interest illustrate the state of the art in this field. Library design and diversity analysis are of increasing importance, and the present programs and approaches are discussed, together with expert views and opinions. In particular, the chapter on a miniaturized ultra-high throughput screening system highlights the immediate future in assay systems of this type.

Combinatorial approaches in materials sciences and solid-state catalysts are relatively new, and these are reviewed, together with expert contributions in the field.

Among the last five chapters, four are devoted to the important instrumentation used for analytical on- and off-bead controls. Whereas tagging concepts appear increasingly less interesting due to excessive time constraints and inherent problems introduced in the drug discovery process, a number of methods have proved suitable for routine single-bead analysis, including infra-red microscopy, separation techniques coupled to electrospray-mass spectrometry, and ion-cyclotron resonance-FT-mass spectrometry to determine the elemental composition of each component in a library. In addition, MAS-NMR has become a popular means of examining resin-bound intermediates during feasibility and optimization studies.

Finally, *Combinatorial Chemistry* closes with a critical discussion of the various attempts to automate combinatorial synthesis, with a presentation of a newly developed workstation used to perform fully automated parallel synthesis, including all steps through work-up to sample preparation for screening modules.

I would like to close this Preface by expressing my gratitude to all authors and co-authors who spent their precious time in contributing to this work. I also acknowledge the present and former co-workers of my group who contributed novel experimental results.

I am very grateful to Mrs. Ursula Sanzenbacher for her help during the editorial work, and to Dr. Gudrun Walter and Dr. Peter Gölitz from Wiley-VCH for their continuous interest on this edition.

During the past few years – and in parallel with combinatorial chemistry – communication via the Internet and the availability of published material from databases have emerged exponentially. However, I am confident that a broad readership may still – from time to time – enjoy reading a scientific book such as Combinatorial Chemistry.

Tübingen, September 1999 Günther Jung

This Page Intentionally Left Blank

Contents

2. Survey of Solid-Phase Organic Reactions

Susan E. Booth, C. Marijke Dreef-Tromp, Petro H.H. Hermkens, Jos A.P.A. de Man and H.C.J. Ottenheijm

3. Solution-Phase Combinatorial Chemistry
Volkhard Austel

4. Combinatorial Chemistry of Multicomponent Reactions
Ivar Ugi, Alexander Dömling and Birgit Ebert

9. RNA and DNA Aptamers

Michael Famulok

10. Combinatorial Approaches to Molecular Receptors for Chemosensors

Dietmar Leipert, Jürgen Mack, Rolf Tünnemann and Günther Jung

11. Peptide Libraries in T-Cell-Mediated Immune Response

Burkhard Fleckenstein, Günther Jung and Karl-Heinz Wiesmüller

List of Abbreviations

A-3CR	Asinger – three component reaction
Aba	2-aminobenzoic acid
Ac	acetyl
ACN	acetonitrile
ACP	acyl carrier protein
ADME	absorption, distribution, metabolism, excretion
ADPV	5-(4-aminomethyl-3,5-dimethoxy-phenoxy)-valeric acid
Alloc	allyloxycarbonyl
AM	aminomethyl
AMCA	7-amino-4-methyl-coumarin-3-acetic acid
ANP	3-amino-3-(2-nitrophenyl) propionic acid
APCI	atmospheric pressure chemical ionization
ATP	adenosine triphosphate
ATR-IR	attenuated total reflection infrared spectroscopy
BAL	backbone amide linker
BAP	borane/pyridine complex
BB-4CR	Berg-Bucherer – four component reaction
BBTO	bis-(tributyl)tinoxide
BEMP	2-tert-butylimino-2-diethylamino-1,3-dimethylperhydro-1,3,2-diazaphosphorine
BHA	benzyhydrylamine
BINAP	2,2′-bis(diphenylphosphino)-1,1′-binaphthyl
Bn, Benz	benzyl
Boc	tert.-butyloxycarbonyl
BOP	benzotriazolyloxy-tris(dimethylamino)phosphonium hexafluorophosphate
Bz	benzoyl
CAD	collision activated dissociation
CALD	computer assisted library design
CAN	ceric ammonium nitrate
CBP	carbohydrate binding proteins
Cbz	benzyloxycarbonyl
CD	circular dichroism
CDI	N,N-carbonyldiimidazol
CE	capillary electrophoresis

CHC	a-cyano-4-hydroxy-cinnamic acid
CHO	chinese hamster ovary (cells)
Chx	cyclohexyl
CID	collision induced decay
CID	collision induced decomposition
CLEAR	cross-linked ethoxylate acrylate resin
CLF	chain length factor protein
CLIP	class II-associated invariant chain peptide
CMPP	chloromethyl phenyl pentyl polystyrene
CoA	coenzyme A
Cod	cycloactadienyl
CPG	controlled pore glass
CRD	carbohydrate recognition domain
CTL	cytotoxic T lymphocytes
cHex	cyclohexyl
Cy	cyclohexyl
CZE	capillary zone electrophoresis
Da	dalton
DABCO	1,4-diazabicyclo[2.2.2]octane
DBE	double bond equivalent
DBU	1,8-diazabicyclo[5.4.0]undec-7-ene
DCI	desorption chemical ionization
DCM	dichloromethane
Dde	1-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene)ethyl
DDQ	dichlorodicyano-p-benzochinon
DEAD	diethylazodicarboxylate
DEBS	6-deoxyerythronolide B synthase
DH	dehydratase
DIAD	diisopropyl azodicarboxylate
DIBAH	diisobutylaluminium hydride
DIBAL	diisobutylaluminium hydride
DIC	diisopropylcarbodiimide
DIEA	diisopropylethylamine
DMAE	dimethylaminoethanol
DMAP	4-N,N-dimethylaminopyridine
DMF	N,N-dimethylformamide
DMSO	dimethylsulfoxide
DMTST	dimethyl(methylthio)sulfonium-trifluoromethylsulfonate
DNA	desoxyribonucleic acid
DRIFTS	diffuse reflectance infrared Fourier transform spectroscopy
DSB	4-(2,5-dimethyl-4-methylsulfinylphenyl)-4-hydroxybutyric acid
DVB	divinylbenzene
EA	enzyme acceptor
EBV	Epstein-Barr virus
ED	enzyme donor

EDC	1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride
EI	electron impact
ELISA	enzyme-linked immunosorbent assay
ER	endoplasmatic reticulum
ER	enoyl reductase
ESI	electrospray ionization
ES-MS	electrospray-mass spectrometry
Et	ethyl
FAB	fast atom bombardment
FCS	fluorescence correlation spectroscopy
FITC	fluoresceinisothiocyanate
FMN	flavinemononucleotide
Fmoc	fluorenylmethyloxycarbonyl
FP	fluorescence polarization
FRET	fluorescence resonance energy transfer
FT	Fourier transform
FT-IR	Fourier transform infrared spectroscopy
Fuc	L-fucose
GalNAc	N-acetyl-D-galactosamine
GC	gas chromatography
GlcNAc	N-acetyl-D-glucosamine
HFIP	hexafluoroisopropanol
HHR	hammer head ribozyme
HIV	human immunodeficiency virus
hKGF	human keratinocyte growth factor
HLA	human leukocyte antigen (MHC)
HMBA	4-hydroxymethylbenzoic acid
HMPB	4-hydroxymethyl-3-methoxyphenoxybutyric acid
HMP-PS	p-benzyloxybenzyl alcohol polystyrene
HMQC	heteronuclear correlation quantum coherence
HOBT	hydroxybenzotriazole
HPLC	high performance liquid chromatography
HPSEC	high-performance size-exclusion chromatography
HQN	hydroxy-quinuclidine
HTE	high-throughput experimentation
HTS	high-throughput screening
HR	high resolution
iPr	isopropyl
IR	infrared
ICR-MS	ion cyclotron resonance mass spectrometry
ICR	ion cyclotron resonance
IRMDP	multiphoton infrared photodissociation
kDa	kilodalton
KHMDS	potassium bis(trimethylsilyl)amide
KR	ketoreductase

KS	ketosynthase
Lau	lauric acid
LC	liquid chromatography
LCAA-CPG	long-chain alkyl amino controlled pore glass
LC-MS	liquid chromatography-mass spectrometry
LD	laser desorption
M-3CR	Mannich – three component reaction
MALDI	matrix assisted laser ionization
MAMP	alpha-methoxy-phenyl (4-methoxy-benzhydryl)
Man	mannose
MAS-NMR	magic angle spinning-nuclear magnetic resonance
MBHA	4-methylbenzhydrylamine
MBP	myelin basic protein
mCPBA	meta-chloroperoxobenzoic acid
MCPS	multiple column peptide synthesis
MCR	multicomponent reaction
MCSL	multiple core structure libraries
Me	methyl
Me-Du-Phos	1,2-bis(2,5-dimethylphospholano)benzene
MHC	major histocompatibility complex
MOBHA	4-methoxybenzhydrylamine
MPR	mannose-phosphate receptor
Ms	mesyl, methanesulfonyl
MS	mass spectrometry
MsCL	methanesulfonylchloride
MTBD	7-methyl-1,5,7-triazabicyclo[4,4,0]elec-5-ene
MTP	microtiter plate
Mtr	4-methoxy-2,3,6-trimethyphenyl-sulfonyl
Nasyl	naphthylsulfonyl
NBA	4-aminomethyl-3-nitrobenzoic acid
Nbb-PS	aminobenzhydryl-polystyrene
NBHA	ortho-nitrobenzylhydrylamino (2-nitrobenzhydrylamine)
NeuAc	N-acetyl-neuraminic acid
NIS	N-iodosuccinimide
NMR	nuclear magnetic resonance
NOE	nuclear Overhauser effect
NOESY	nuclear Overhauser effect spectroscopy
NPE	4-hydroxyethyl-3-nitro-benzoic acid
Npss	2-methoxy-5-(2-(2-nitrophenyl)dithio-1-oxyproplphenylacetic ac
NSG	N-substituted oligoglycine
NBS-Cl	o-nitrobenzenesulfonyl chloride
P	partition coefficient
P-3CR	Passerini – three component reaction
PAAs	poly-N-acylated amines

PAL	peptide amide linker: 5-[4[9-fluorenylmethoxycarbonylaminomethyl-]-3,5-dimethoxyphenoxy]valeric acid
PAM	phenylacetamidomethyl
PBS	phosphate buffered saline
PCA	principal component analysis
PCR	polymerase chain reaction
PDC	pyridinium dichromate
PDTs	pharmacophor definition triplets
PEG	polyethyleneglycol
PEGA	polyethylene glycol-polyacrylamide co-polymer
PEG-PS	polyethylene glycol-polystyrene
PET	photo-induced electron transfer effect
Pfp	pentafluorophenyl
Ph	phenyl
PhFl Resin	9-phenylfluorene-9-yl-PS/DVB
Pip	piperidine
PK	pharmacokinetics
PKSs	polyketide synthases
PNAs	peptide nucleic acids
PNP	para-nitrophenyl
POEPOP	polyoxyethylene-epichlorohydrine derivative
POEPS	polyoxyethylene-cross-linked-oligostyrene
Ppa	phenylpropionic acid
PS	polystyrene
PS-DVB	polystyrene-divinylbenzene copolymer
PSD	post-source decay
PS-PEG	polystyrene-polyethyleneglycol graft polymer
PyBroP	bromo-tris-pyrrolidino-phosphonium-hexafluorophosphate
QSAR	quantitative structure-activity relationship
R	alkyl
RCM	ring closure metathesis
relSI	relative stimulation index
REMPI	laser-induced resonant multiphoton ionization
Rev	reverse transcriptase (HIV)
RIfS	reflectometric interference spectroscopy
RP-HPLC	revers phase-high performance liquid chromatography
RRE	Rev-responsive element
S-3CR	Strecker – three component reaction
SAR	structure-activity relationship
SASRIN	super acid sensitive resin
SDS-PAGE	sodium dodecylsulfate-polyacrylamide gel electrophoresis
SI	stimulation index
S-Le	sialyl – Lewis
SMSI	string metal support interaction
SPOC	solid-phase organic chemistry

SPOCC	polyoxyethylene-oxetane derivative
SPOS	solid phase organic synthesis
SPPS	solid phase peptide synthesis
ssRNA	single stranded ribonucleic acid
TAR RNA	trans-activation response region
TAP	transporter associated with antigen processing
TBAF	tetrabutylammonium fluoride
Tbf	tetrabenzo[a,c,g,i]fluorene
tBu	tert.-butyl
TCC	$CD4^+$ T-cell clones
TCR	T-cell receptor
TE	thioesterase
TEA	triethyl amine
Tf	trifluoromethylsulfonyl
TFA	trifluoroacetic acid
TFE	trifluoroethanol
TFMSA	trifluoromethanesulfonic acid
THF	tetrahydrofurane
THP	tetrahydropyranyl
TIC	total ion current
TIPS	triisopropylsilane (TIS)
TIS	triisopropylsilane (TIPS)
TLC	thin layer chromatography
TMS	trimethylsilyl
TMOF	trimethylsilyl trifluoromethanesulfonate
TMSOTf	trimethylsilyl trifluoromethanesulfonate
Tn	tags for labeling
TOCSY	total correlation spectroscopy
TOF	time-of-flight
TPAP	thrombin receptor activating peptide
Ts	tosyl, para-toluenesulfonyl
U-4CR	Ugi – four component reaction
uHTS	ultra high-throughput screening
UV	ultra-violet
UVDD	ultra-violet photodissociation
VEGF	vascular endothelial growth factor
VLSIPS	very large-scale immobilized polymer synthesis
VPF	vascular permeability factor
WHIM	weighted holistic invariant molecular (indices)
Z	benzyloxycarbonyl

Further special abbreviations of linkers and resins are found in Chapter 5.