



#### University of Groningen

Dynamic combinatorial and	d protein-templated	click chemistry is	n medicinal	chemistry
Mondal, Milon				

IMPORTANT NOTE: You are advised to consult the publisher's version (publisher's PDF) if you wish to cite from it. Please check the document version below.

Document Version Publisher's PDF, also known as Version of record

Publication date: 2016

Link to publication in University of Groningen/UMCG research database

Citation for published version (APA):

Mondal, M. (2016). Dynamic combinatorial and protein-templated click chemistry in medicinal chemistry. University of Groningen.

Other than for strictly personal use, it is not permitted to download or to forward/distribute the text or part of it without the consent of the author(s) and/or copyright holder(s), unless the work is under an open content license (like Creative Commons).

The publication may also be distributed here under the terms of Article 25fa of the Dutch Copyright Act, indicated by the "Taverne" license. More information can be found on the University of Groningen website: https://www.rug.nl/library/open-access/self-archiving-pure/taverneamendment.

If you believe that this document breaches copyright please contact us providing details, and we will remove access to the work immediately and investigate your claim.

Downloaded from the University of Groningen/UMCG research database (Pure): http://www.rug.nl/research/portal. For technical reasons the number of authors shown on this cover page is limited to 10 maximum.

Download date: 08-06-2022

# Dynamic Combinatorial and Protein-Templated Click Chemistry in Medicinal Chemistry

Milon Mondal



## Dynamic Combinatorial and Protein-Templated Click Chemistry in Medicinal Chemistry

Milon Mondal Ph.D Thesis University of Groningen, The Netherlands

The research described in this thesis was carried out within the Stratingh Institute for Chemistry, University of Groningen, The Netherlands.

In compliance with the requirements of the Graduate School of Science of the Faculty of Mathematics and Natural Sciences, University of Groningen, The Netherlands.

This work was financially supported by the University of Groningen and the Netherlands Organisation for Scientific Research (NWO).

The cover was designed by Milon Mondal and Jonas Lohse.

Printed by Ipskamp Printing BV, Enschede, The Netherlands.

ISBN: 978-90-367-8663-8 (printed) 978-90-367-8662-1 (digital)



# Dynamic Combinatorial and Protein-Templated Click Chemistry in Medicinal Chemistry

#### **PhD Thesis**

to obtain the degree of PhD at the University of Groningen on the authority of the Rector Magnificus Prof. E. Sterken and in accordance with the decision by the College of Deans.

This thesis will be defended in public on

Friday 12 February 2016 at 12.45 hours

by

**Milon Mondal** 

born on 5 March 1988 in Swarupnagar, India

**Supervisors** Prof. A.K.H. Hirsch Prof. A.J. Minnaard

### **Assessment committee**

Prof. A.S.S. Dömling Prof. J.G. Roelfes Prof. C. Schofield

Ma and Baba (my parents)

### Table of contents

Chaj	pter 1.		c Combinatorial Chemistry: a Tool to te the Identification of Inhibitors for Protein	1
1.1	Structi	ure-based	drug design	2
1.2	Fragm	ent-based	drug design	5
1.3	Dynar	nic combii	natorial chemistry	6
	1.3.1	General	features of DCC applied to protein targets	7
	1.3.2	Reversit	ole covalent bond formation for protein targets	9
		1.3.2.1	C=N bond formation	9
			1.3.2.1.1 Imine formation	9
			1.3.2.1.2 Hydrazone formation	11
			1.3.2.1.3 Acylhydrazone formation	13
		1.3.2.2	C-C bond formation: Alkene cross metathesis	16
		1.3.2.3	C-S bond formation	18
			1.3.2.3.1 Hemithioacetal formation	18
			1.3.2.3.1 Thioether formation	20
		1.3.2.4	S-S bond formation: Disulfide-bond formation	23
		1.3.2.5	B-O bond formation: Boronate ester formation	25
1.4	Aspar	tic proteas	ees	28
	1.4.1	Endothia	apepsin	29
1.5	Strate	gic combir	nations	30
	1.5.1	Overvie	W	32
1.6	Refere	ences		32

Chapter 2.		Structure-Based Drug Design of Inhibitors of the Aspartic Protease Endothiapepsin by Exploiting Dynamic Combinatorial Chemistry		39	
2.1	Introd	luction		40	
2.2	Results and discussion		40		
	2.2.1	De novo	structure-based design	40	
	2.2.2	Synthesis of building blocks		42	
	2.2.3	Formation	on and analysis of DCLs using <sup>1</sup> H-STD-NMR	43	
	2.2.4	Synthesi	is of identified acylhydrazones	46	
	2.2.5	Biochemical evaluation		47	
	2.2.6	X-ray crystallography			
2.3	Conclu	usions		51	
2.4	Experi	Experimental section			
	2.4.1	General procedure for the formation and analysis of the DCLs		51	
		<sup>1</sup> H-STD		52	
	2.4.2	Fluoresc	ence-based inhibition assay	52	
	2.4.3	Modelin	g and Docking	53	
	2.4.4	Crystalli	zation, data collection and processing	53	
	2.4.5	General Experimental Details		54	
	2.4.6	Synthesis of the monomers		55	
		2.4.6.1	General procedure for hydrazide formation to form the corresponding methyl ester (GP1)	55	
		2.4.6.2	General procedure for acylhydrazone formation (GP2)	57	
2.5	Refere	ences		62	

Chapter 3.		Fragment Linking and Optimization of Inhibitors of the Aspartic Protease Endothiapepsin: Fragment- Based Drug Design Facilitated by Dynamic Combinatorial Chemistry		65		
3.1	Introd	uction		66		
3.2	Result	Results and Discussion		67		
	3.2.1	Fragmen	nt-based drug design	67		
	3.2.2	Synthesi	Synthesis of building blocks			
	3.2.3	Formatic	Formation and analysis of DCLs using LC-MS			
	3.2.4	Synthesi	s of identified bis-acylhydrazones	72		
	3.2.5	Biochem	nical evaluation	73		
	3.2.6	X-ray cr	X-ray crystallography			
3.3	Conclu	usions		75		
3.4	Experi	Experimental Section 7				
	3.4.1	General DCLs HPLC c	procedure for the formation and analysis of the onditions	76 76		
	3.4.2		ence-based inhibition assay	77		
	3.4.3	·		77		
	3.4.4					
		3.4.4.1	General Experimental Details	77		
		3.4.4.2	Synthesis of the Hydrazides	77		
3.5	Refere	ences	·			
Cha	pter 4.		re-Based Optimization of Inhibitors of artic Protease Endothiapepsin	83		
4.1	Introdu	uction		84		
4.2	Result	Results and Discussion				
	4.2.1	Structure	e-based drug design	84		
	4.2.2	Synthesi	s of Acylhydrazone derivatives	87		

	4.2.3	Biochemical evaluation	87
	4.2.4	Discussion	89
4.3	Conclu	usions	91
4.4	Experi	imental Section	91
	4.4.1	Fluorescence-based inhibition assay	91
	4.4.2	Modeling and Docking	91
	4.4.3	General experimental details	91
	4.4.4	Synthesis	91
4.5	Refere	ences	96
Cha	pter 5.	Fragment Growing Exploiting Dynamic Combinatorial Chemistry of Inhibitors of the Aspartic Protease Endothiapepsin	99
5.1	Introd	uction	100
5.2	Result	s and Discussion	100
	5.2.1	FBDD: fragment growing	100
	5.2.2	Synthesis of building blocks	102
	5.2.3	Formation and analysis of DCL	102
	5.2.4	Synthesis and biochemical evaluation of identified acylhydrazones	105
5.3	Conclu	• •	107
5.4	Experi	imental Section	107
	5.4.1	Fluorescence-based assay for DCL screening	107
	5.4.2	Fluorescence-based inhibition assay	108
	5.4.3	Modeling and Docking	108
	5.4.4	General experimental details	108
	5.4.5	Synthesis	108
5.5	Refere	ences	112

Chapter 6.		Fragment-Based Drug Design Facilitated by Protein- Templated Click Chemistry: Fragment Linking and Optimization of Inhibitors of the Aspartic Protease Endothiapepsin	
6.1	Introdu	uction	116
6.2	Results and discussion		118
	6.2.1	Fragment-based drug design	118
	6.2.2	Synthesis of building blocks (azides and alkynes)	119
	6.2.3	Generation of library	121
	6.2.4	Synthesis of identified triazoles	123
	6.2.5	Biochemical evaluation	125
	6.2.6	Discussion	126
6.3	Conclu	asions	128
6.4	Experimental Section		
	6.4.1	PTCC experiments	128
	6.4.2	Fluorescence-based inhibition assay	130
	6.4.3	Modeling and Docking	130
	6.4.4	General experimental details	130
	6.4.5	Synthesis of azides, alkynes and triazoles	130
	6.4.6	General procedure for azide synthesis	130
6.5	References		136
Con	clusions	s and Perspectives	139
7.1	Introd	uction	140
7.2	Resea	rch overview	141
7.3	Perspe	ectives	143
Sam	e nvattir	ng	147
Ack	nowle dg	gments	151