Oxidation, aggregation and immunogenicity of therapeutic proteins

Torosantucci, R.

Appendix 2

Abbreviations

Abbreviations

ABI	ammonium bicarbonate
ABS	4-(aminomethyl) benzenesulfonic acid
ADAs	anti-drug antibodies
Ag	antigen
ANOVA	analysis of variance
Asn	asparagine
AUC	area under the curve
BABs	binding antibodies
BAM	benzylamine
CB	sodium citrate buffer
CD	circular dichroism
CDRs	complementarity determining regions
CFA	complete Freund's adjuvant
CHO	Chinese hamster ovary
CSB	sodium citrate sucrose buffer
CT	calcitonin
Cu^{2+}	bivalent copper ion
Cys	cysteine
DHB	dihydroxybenzoic acid
DLS	dynamic light scattering
DNA	deoxyribonucleic acid
DNPH	2,4-dinitrophenylhydrazine
DO	dissolved oxygen
DOCH	2-amino-3-(3,4-dioxocyclohexa-1,5-dien-1-yl) propanoic acid
DOPA	3,4-dihydroxyphenylalanine
DTT	DL-dithiothreitol
E. coli	Escherichia coli
EDAC	1-(3-dimethylaminopropyl)-3-ethylcarbodiimide
EDTA	ethylenediaminetetraacetic acid
ELISA	enzyme-linked immunosorbent assay
ESI-MS	electrospray ionization mass spectrometry
ESI-ToF	electrospray ionization time of flight
Fab	fragment antigen-binding
Fc	fragment crystallisable
FcRn	neonatal Fc receptor
FDA	Food and Drug Administration

FT-ICR MS	Fourier transform ion cyclotron resonance mass spectrometry
G-CSF	granulocyte colony stimulating factor
GH	growth hormone
GM-CSF	granulocyte macrophage colony stimulating factor
GnHCl	guanidine hydrochloride
GPC	gel permeation chromatography
GSH	reduced glutathione
His	histidine
HMWO	high-molecular-weight oligomers
HSA	human serum albumin
IAM	iodoacetamide
IFNα-2a	interferon alpha-2a
IFNα-2b	interferon alpha-2b
IFNβ-1a	interferon beta-1a
IgG	immunoglobulin G
i.m.	intramuscular
i.p.	intraperitoneal
i.v.	intravenous
LOPC	light obscuration particle counting
Lys	lysine
mAbs	monoclonal antibodies
MCO	metal catalyzed oxidation
MeO-PEG-NHS	Alpha-metoxy omega-carboxylic acid succinimidyl ester poly(ethylene glycol)
MES	2-N-morpholino-ethanesulfonic acid
Met	methionine
Nab	neutralizing antibody
NHS-sulfo	N-hydroxysulfosuccinimide sodium salt
NP	polystyrene nanoparticles
NTA	nanoparticle tracking analysis
NTG	non transgenic
OD	optical density
PAGE	polyacrylamide gel electrophoresis
PB	sodium phosphate buffer
PBS	phosphate buffered saline
PCR	polymerase chain reaction
PDI	polydispersity index
PEG	polyethylene glicol

Phe	phenylalanine
PTH	parathyroid hormone
PVDF	polyvinylidene difluoride
ROS	reactive oxygen species
RP-HPLC	reversed-phase high pressure liquid chromatography
rtPCR	real time PCR
SA	sinapinic acid
s.c.	subcutaneous
SDS	sodium dodecyl sulfate
SEC	size-exclusion chromatography
SEM	standard error of the mean
Td	T-cell dependent
TETA	triethylenetetramine
TFA	trifluoroacetic acid
TG	transgenic
Ti	T-cell independent
TMB	3,3′,5,5′-tetramethylbenzidine
Trp	tryptophan
Tyr	tyrosine
USP	United States Pharmacopeia
UV	ultraviolet
v/v	volume/volume
w/w	weight/weight
Z-ave	Z-average diameter

Appendix 3

List of publications

List of publications

2013

R. Torosantucci, V. S. Sharov, M. van Beers, V. Brinks, C. Schöneich, W. Jiskoot. Identification of oxidation sites and covalent cross-links in metal catalyzed oxidized interferon beta-1a: potential implications for protein aggregation and immunogenicity. *Molecular Pharmaceutics* **2013**, *10*, 2311-2322.

R. Torosantucci, D. Weinbuch, R. Klem, W. Jiskoot. Triethylenetetramine prevents insulin aggregation and fragmentation during copper catalyzed oxidation. *European Journal of Pharmaceutics and Biopharmaceutics* **2013**, *84,* 464-471.

R. Torosantucci, V. Brinks, G. Kijanka, L. Andhyk Halim, M. Sauerborn, H. Schellekens and W. Jiskoot. Development of a transgenic mouse model to study the immunogenicity of recombinant human insulin. *Submitted for publication.*

R. Torosantucci, C. Schöneich, W. Jiskoot. Oxidation of therapeutic proteins and peptides: structural and biological consequences. *Submitted for publication.*

M. Sauerborn#, **R. Torosantucci**#, L. Boon, W. Jiskoot, H. Schellekens, V. Brinks. Immune mechanisms underlying immunogenicity of aggregated recombinant human interferon alpha 2a in immune tolerant mice. *Submitted for publication.*

# authors contributed equally

2012

R. Torosantucci, O. Mozziconacci, V. Sharov, C. Schöneich, W. Jiskoot. Chemical modifications in aggregates of recombinant human insulin induced by metal-catalyzed oxidation: covalent cross-linking via michael addition to tyrosine oxidation products. *Pharmaceutical Research* **2012**, *29*, 2276-2293.

2011

R. Torosantucci, B. Kükrer, A. Mero, M. Van Winsen, R. Tantipolphan, W. Jiskoot. Plain and mono-pegylated recombinant human insulin exhibit similar stress-induced aggregation profiles. *Journal of Pharmaceutical Sciences* **2011**, *100*, 2574-2585.

2010

R. Tantipolphan, S. Romeijn, J. Engelsman, **R. Torosantucci**, T. Rasmussen, W. Jiskoot. Elution behavior of insulin on high-performance size exclusion chromatography at neutral pH. *Journal of Pharmaceutics and Biomedical Analysis*, **2010**, *52*, 195-202.

2009

F. De Simone, J. Andres, **R. Torosantucci**, J. Waser. Catalytic formal homo-Nazarov cyclization. *Organic Letters* **2009**, *11*, 1023-1026.

2008

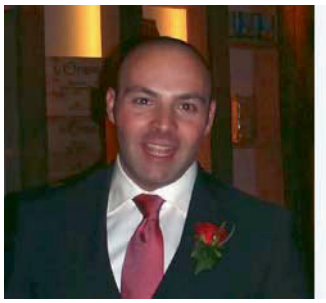
M. Faccini, M. Balakrishnan, M. B. J. Diemeer, **R. Torosantucci**, A. Driessen, D. N. Reinhoudt, W. Verboom. Photostable nonlinear optical polycarbonates. *Journal of Materials Chemistry* **2008**, *18*, 5293-5300.

M. Faccini, M. Balakrishnan, **R. Torosantucci**, A. Driessen, D. N. Reinhoudt, W. Verboom. Facile attachment of nonlinear optical chromophores to polycarbonates. *Macromolecules* **2008**, *41*, 8320-8323.

Appendix 4

Curriculum Vitae

Curriculum Vitae



Riccardo Torosantucci was born in Rome, Italy, 22nd September 1980. In July 2008 he received his master degree in Chemistry and Pharmaceutical Technologies from the University of Rome "La Sapienza". He completed the first internship of his master studies working for six months in supramolecular chemistry, in particular on the synthesis of calixarenes at the Department of Biologically Active Substances of University of Rome "La Sapienza" under the supervision of Prof. Dr. Bruno Botta. Next, in July 2007, Riccardo was awarded the "Master's thesis abroad travel grant" and he joined for six months the group of Supramolecular Chemistry and Technology (SMCT) directed by Prof. Dr. David Nicholas Reinhoudt at Twente University, The Netherlands. During this period he worked on the synthesis of new polymers for second-order nonlinear optics. After his master thesis defence in July 2008, from October until December 2008, Riccardo worked at the Laboratory of Catalysis and Organic Synthesis (LCSO) of the École Polytechnique Fédérale de Lausanne (EPFL), Switzerland, under the supervision of Prof. Dr. Jérôme Waser, focusing mainly on the synthesis of heterocyclic compounds. In March 2009 Riccardo started his PhD at the Division of Drug Delivery Technology at Leiden University, The Netherlands, under the supervision of Prof. Dr. Wim Jiskoot, Prof. Dr. Huub Schellekens and Dr. Vera Brinks, on a project entitled "Unwanted immunogenicity of therapeutic proteins". In May 2011, as a part of his PhD project, he spent 5 months at the Department of Pharmaceutical Chemistry of Kansas University, Lawrence, USA, in the group directed by Prof. Dr. Christian Schöneich, where he worked on mass spectrometric characterization of therapeutic protein aggregates induced via metal catalyzed oxidation. Since May 2013 Riccardo is working as a project manager at Coriolis-Pharma in Munich, Germany.

Grants & Awards

2012

- First prize for oral presentation at the ninth biennial meeting of the Globalization of Pharmaceutics Education Network, Monash University, Melbourne, Australia.
- GlaxoSmithKline Housing Grant
- Merck & Co. Travel Grant
- Nederlandse Stichting voor Farmacologische Wetenschappen (NSFW) travel grant *2012*

2011

- CASSS Separation Science Society travel grant
- Leids Universiteits Fonds travel grant (Leiden University, NL)
- Nederlandse Stichting voor Farmacologische Wetenschappen (NSFW) travel grant *2011*

2007

- “Master’s thesis abroad travel grant”, University of Rome "La Sapienza"

2006

- Scholarship at the Department of Pharmaceutical Chemistry, University of Rome "La Sapienza"

Appendix 5

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