Nanoparticles and microfluidics for future tuberculosis vaccines

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CURRICULUM VITAE

Born in Copenhagen, Denmark, in 1990, Malene Aaby Neustrup pursued her undergraduate and graduate studies in Pharmacy at the School of Pharmaceutical Sciences, University of Copenhagen, Denmark, from 2011 to 2017. During her academic journey, she gained practical experience as a student assistant in the product quality department at the company Haldor Topsøe.

Her master's thesis research, conducted at the Danish State Serum Institute and the Department of Pharmacy, University of Copenhagen, was supervised by dr. Signe Tandrup Schmidt, prof.dr. Camilla Foged, and dr. Dennis Christensen, and resulted in her thesis titled: "Formulation and Characterisation of Nanoemulsion-Based Adjuvants Mediating Vaccine-Induced $CD8^+$ T-Cell Responses".

In September 2017, Malene Aaby Neustrup commenced her PhD studies at the Leiden Academic Centre for Drug Research, Leiden University, and Leiden University Medical Centre under the guidance of prof.dr. Joke A. Bouwstra, prof.dr. Tom H.M. Ottenhoff, and dr. Koen van der Maaden. Her doctoral research focused on developing a tuberculosis vaccine utilizing nanoparticle formulations and dissolvable microneedle technology. Teaching and mentorship were central components of Malene's studies, demonstrated by her supervision of laboratory courses, guidance of bachelor's students, and mentorship of master's students. In addition, she presented her research at several Dutch conferences.

LIST OF PUBLICATIONS

Systematic investigation of the role of surfactant composition and choice of oil: Design of a nanoemulsion-based adjuvant inducing concomitant humoral and $CD4^+$ T-cell responses
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https://doi.org/10.1007/s11095-017-2180-9

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S. T. Schmidt, G. K. Pedersen, **M. A. Neustrup**, K. S. Korsholm, T. Rades, P. Andersen, D. Christensen, and C. Foged. Front Immunol. 2018;9:898.
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Atomic force microscopy measurements of anionic liposomes reveal the effect of liposomal rigidity on antigen-specific regulatory T-cell responses
N. Benne, R. J. T. Leboux, M. Glandrup, J. van Duijn, F. Lozano Vigario, **M. A. Neustrup**, S. Romeijn, F. Galli, J. Kuiper, W. Jiskoot, and B. Slütter. J Control Release. 2020;318:246-55.
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Stabilin-1 is required for the endothelial clearance of small anionic nanoparticles
G. Arias-Alpizar, B. Koch, N. M. Hamelmann, **M. A. Neustrup**, J. M. J. Paulusse, W. Jiskoot, A. Kros, and J. Bussmann. Nanomedicine. 2021;34:102395.
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Intrinsic immunogenicity of liposomes for tuberculosis vaccines: Effect of cationic lipid and cholesterol
M. A. Neustrup*, M. M. Szachniewicz*, K. E. van Meijgaarden, W. Jiskoot, J. A. Bouwstra, M. C. Haks, A. Geluk, and T. H. M. Ottenhoff. Eur J Pharm Sci. 2024;195:106730.
https://doi.org/10.1016/j.ejps.2024.106730.

Intradermal vaccination with PLGA nanoparticles via dissolving microneedles and classical injection needles
M. A. Neustrup*, J. Lee*, B. Slütter, C. O'Mahony, J. A. Bouwstra, and K. van der Maaden. Pharm Res. 2024;41(2):305-19.
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Evaluation of PLGA, lipid-PLGA hybrid nanoparticles, and cationic pH-sensitive liposomes as tuberculosis vaccine delivery systems in a Mycobacterium tuberculosis challenge mouse model - A comparison
M. M. Szachniewicz, **M. A. Neustrup**, S. J. F. van den Eeden, K. E. van Meijgaarden, K. Franken, S. van Veen, R. I. Koning, R. W. A. L. Limpens, A. Geluk, J. A. Bouwstra, and T. H. M. Ottenhoff. Int J Pharm. 2024;666:124842.
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A versatile, low-cost modular microfluidic system to prepare poly(lactic-*co*-glycolic acid) nanoparticles with encapsulated protein
M. A. Neustrup, T. H. M. Ottenhoff, W. Jiskoot, J. A. Bouwstra, and K. van der Maaden. Pharm Res. 2024;41(12):2347-61.
https://doi.org/10.1007/s11095-024-03792-1

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ABBREVIATION LIST

Abbreviation	Meaning
ACK	ammonium-chloride-kalium
AER	Ag85B-ESAT6-Rv2034
Ag	antigen
Ag85B	antigen 85B
APC	antigen-presenting cell
BCA	bicinchoninic acid
BCG	*Mycobacterium bovis* Bacille Calmette-Guérin
BSA	bovine serum albumin
CCD	the Netherlands's Central Authority for Scientific Procedures on Animals
CCL	chemokine (C-C motif) ligand
CCR	C-C chemokine receptor type
CD	cluster of differentiation
CFU	colony forming unit
CpG ODN	cytosine-phosphate-guanine oligodeoxynucleotide
CXCL	chemokine (C-X-C motif) ligand
CXCR	C-X-C motif chemokine receptor
DC	dendritic cell
DC-cholesterol	3ß-[N- (N',N'-dimethylaminoethane)-carbamoyl]cholesterol hydrochloride
DDA	dimethyldioctadecylammonium bromide, bromide salt
DLS	dynamic light scattering
dMNA	dissolvable microneedle array
DMSO	dimethyl sulfoxide
DOBAQ	N-(4-carboxybenzyl)-N,N-dimethyl-2,3-bis(oleoyloxy)pro-pan-1-aminium
DODMA	1,2-dioleyloxy-3-dimethylaminopropane
DOPC	1,2-dioleoyl-*sn*-glycero-3-phosphocholine
DOPE	1,2-dioleoyl-*sn*-glycero-3-phosphoethanolamine
DOTAP	1,2-dioleoyl-3-trimethylammonium-propane, chloride salt
DSPC	1,2-distearoyl-sn-glycero-3-phosphocholine
EE%	encapsulation efficiency
ELISA	enzyme-linked immunosorbent assay

Abbreviation	Meaning
EPC	1,2-dioleoyl-*sn*-glycero-3-ethylphosphocholine, chloride salt
ESAT-6	the 6 kDa early secretory antigenic target
EU	endotoxin unit
FBS	fetal bovine serum
FDR	false discovery rate
FRR	flow rate ratio
GL-67	N^4-cholesteryl-spermine hydrochloride
GM-CSF	granulocyte-macrophage colony-stimulating factor
GMP	Good Manufacturing Practice
His	N-terminal hexahistidine
HLA	human leukocyte antigen
HRP	horse radish peroxide
i.n.	intranasal
ID	inner diameter
IFN	interferon
Ig	immunoglobulin
IL	interleukin
IMDM	Iscove's Modified Dulbecco's Medium
IQR	interquartile range
KLRG1	killer cell lectin-like receptor subfamily G member 1
LAL	limulus amebocyte lysate
LN	lymph node
MACS	magnetic cell isolation
M-CSF	macrophage colony-stimulating factor
MDDC	monocyte-derived dendritic cell
MDMF	monocyte-derived macrophages
MDR-TB	multidrug-resistant tuberculosis
MHC	major histocompatibility complex
MPLA	monophosphoryl lipid A
Mtb	*Mycobacterium tuberculosis*
MVL5	N1-[2-((1S)-1-[(3-aminopropyl)amino]-4-[di(3-amino-propyl) amino]butylcarboxamido)ethyl]-3,4-di[oleyloxy]-benzamide
NP	nanoparticle

Abbreviation	Meaning
OD	outer diameter
OVA	ovalbumin
PAMP	pathogen-associated molecular pattern
PB	phosphate buffer
PBMC	peripheral blood mononuclear cell
PBS	phosphate-buffered saline
PCR	polymerase chain reaction
PD-1	programmed cell death protein 1
PDI	polydispersity index
PDMS	polymethylmethacrylate
PE	phosphatidylethanolamine
PEEK	polyether ether ketone
PLGA	poly(D,L-lactic-*co*-glycolic acid)
PPD	purified protein derivative
PRR	pattern-recognition receptor
PTFE	polytetrafluoroethylene
PVA	poly(vinyl alcohol)
PVP	polyvinylpyrrolidone
QC	Quality Control
rpm	rounds per minute
RPMI	Roswell Park Memorial Institute
s.c.	subcutaneous
SA	stearylamine
SDGs	Sustainable Development Goals
SOI	site of injection
TB	tuberculosis
TCR	α:β T-cell receptor
T_{FH} cell	T follicular helper cell
TFR	total flow rate
Th	T helper
Th1	T helper type 1
Th1/Th2/Th17	T-helper type 1/2/17 cell
TLR	Toll-like receptor
TMB	tetramethylbenzidine

Abbreviation	Meaning
TNF	tumor necrosis factor
T_{reg} cell	regulatory T cell
Ultrapure water	Spectra-Por® Milli-Q® water
UMAP	uniform manifold approximation and projection
WHO	World Health Organization