REGULATION OF TUMOR IMMUNE MICROENVIRONMENT USING ADVANCED NANO-IMMUNOTHERAPY

Helena F. Florindo

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CRS 2022 Annual Meeting & Expo

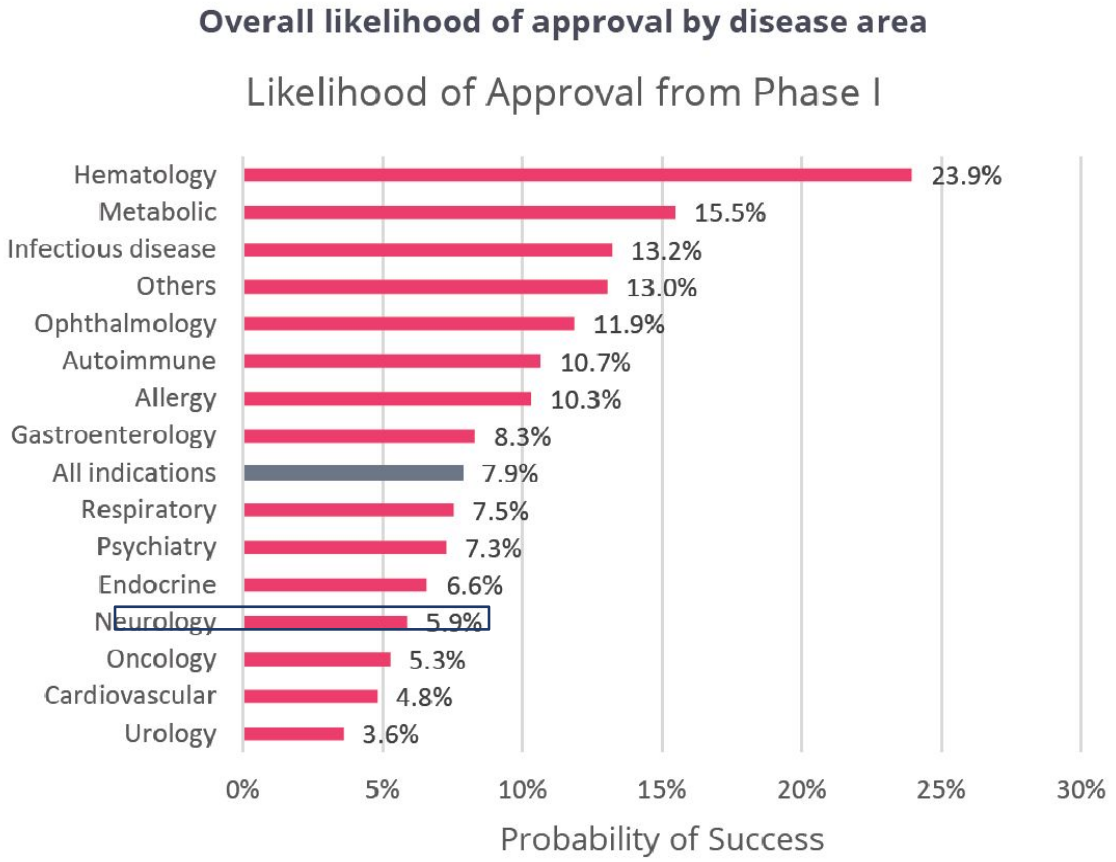
July 11 – 15, 2022 | Montreal Congress Center, Montreal Canada

Advanced Delivery Science

Clinical Development Success Rates

The overall likelihood of approval (LOA) from Phase I for all developmental candidates over 2011–2020 was 7.9%.

Unmet clinical needs in cancer, especially the advanced stage of this disease

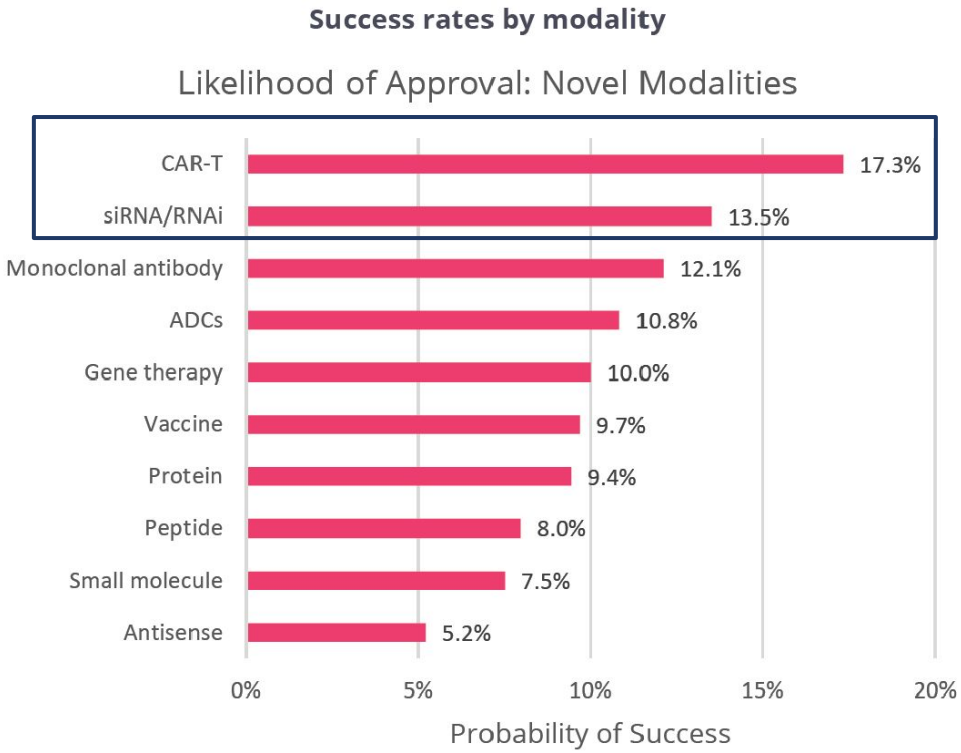


Biomedtracker® and Pharmapremia®, 2020

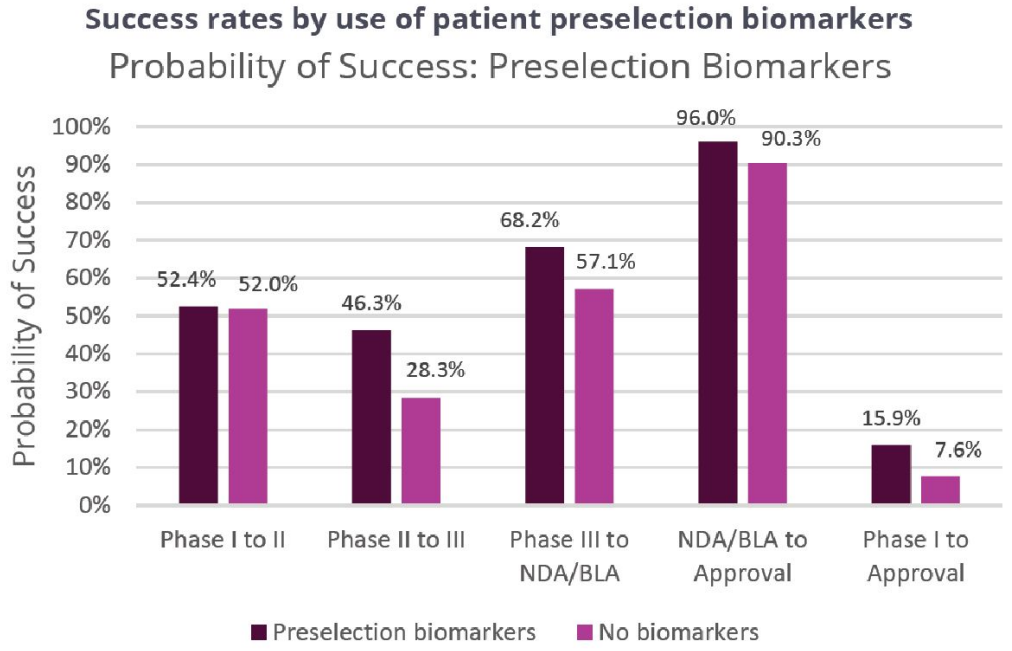
Clinical Development Success Rates

Immuno-oncology therapies success in oncology R&D with an overall LOA of 12.4% *vs* 5.3% for all oncology approaches.

Biological complexity in drug modalities leads to higher LOA:

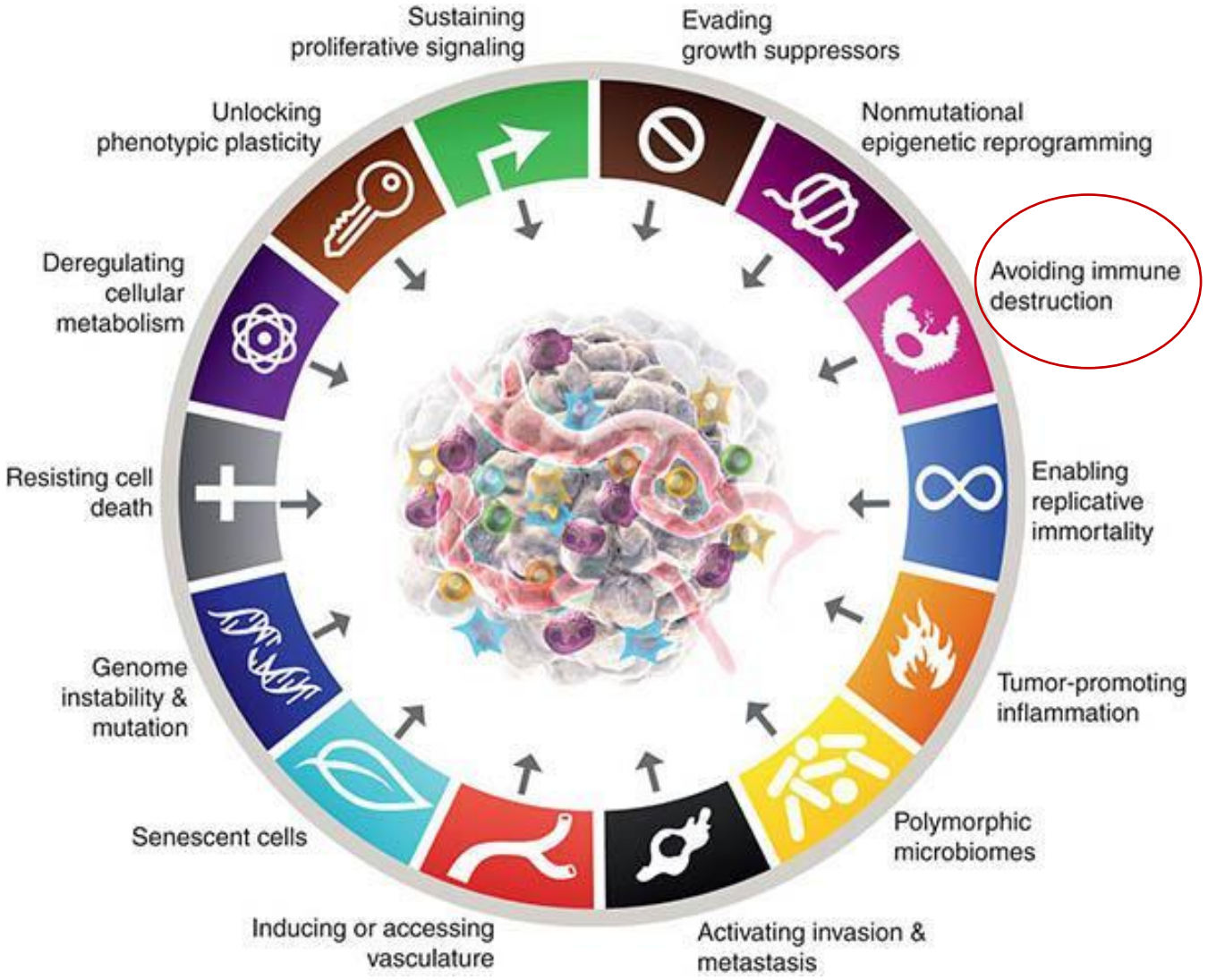


Trials with patient preselection biomarkers have two-fold higher LOAs (15.9%), with a Phase II success rate of nearly 50%.



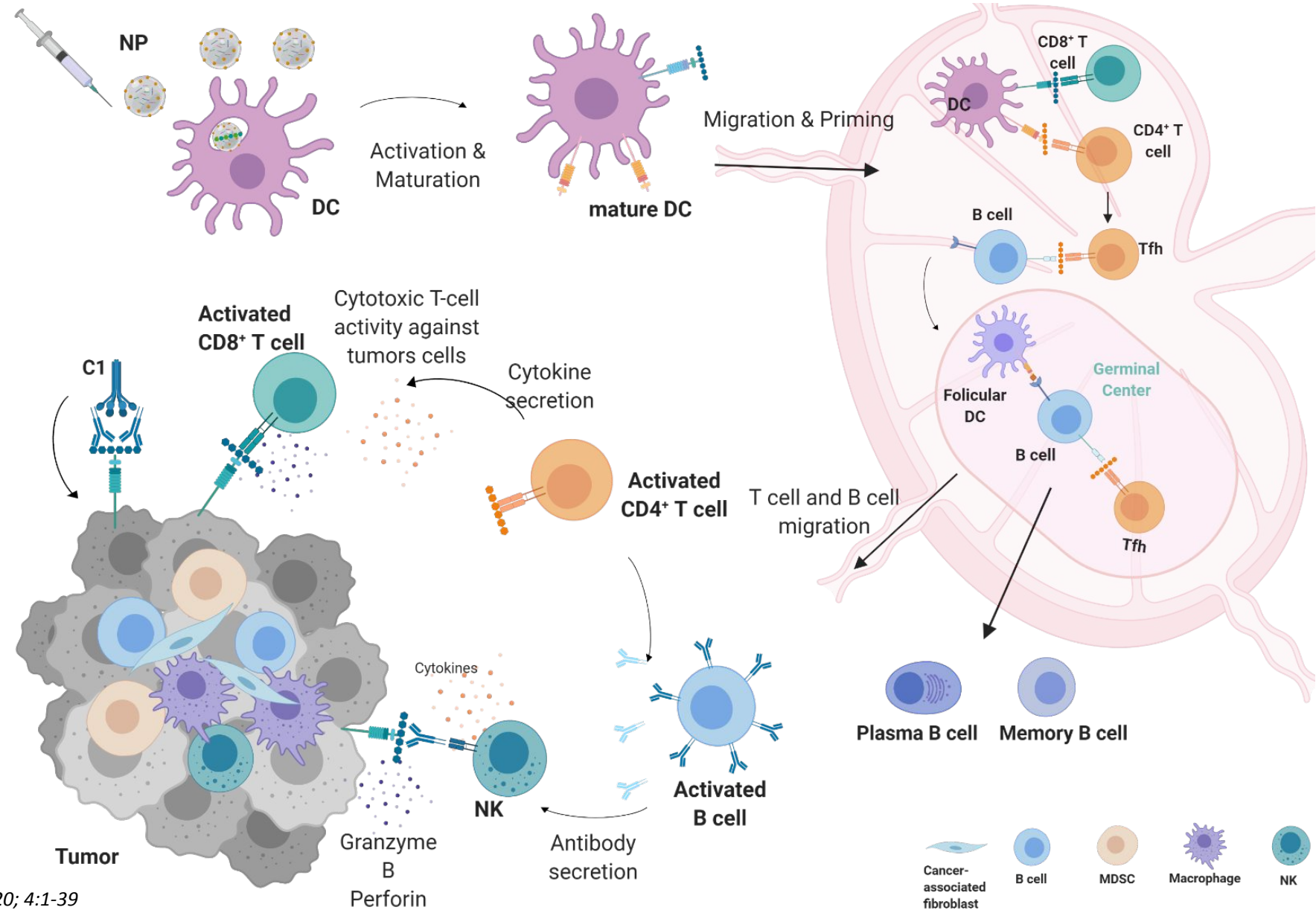
Biotechnology Innovation organization, Informa Pharma Intelligence report on "Clinical development success rates and contributing factors 2011-2020, 2021.

Hallmarks of cancer: rethinking how to defeat cancer



Hanahan D. Cancer Discov 2022;12:31–46

Hallmarks of anti-tumor immune response and nanotechnology



Carreira B, et al.
Advanced Therapeutics, 2020; 4:1-39

Nanotechnology Integration with Pharmacology, Immunology and Pathophysiology

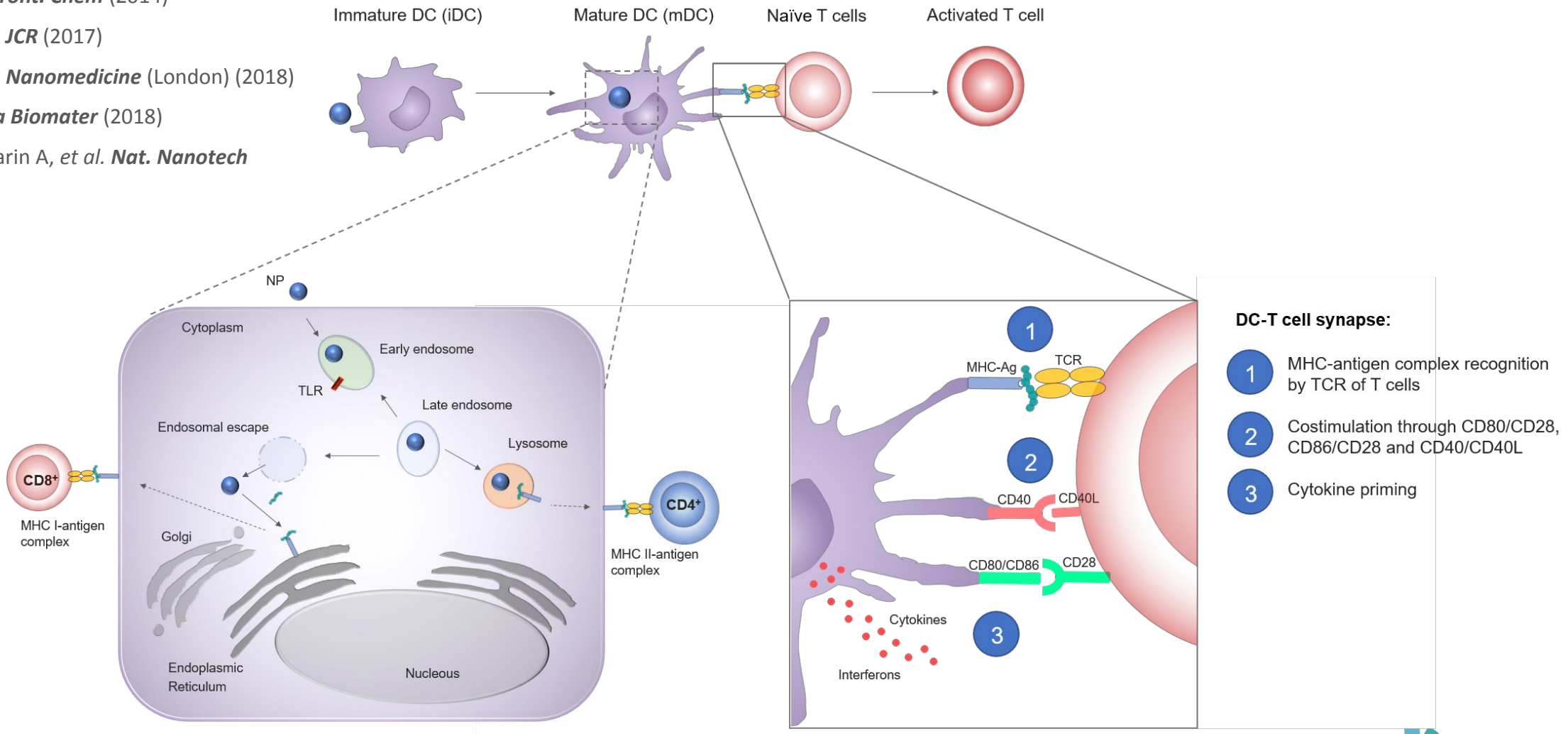
Conniot J, *et al.*, ***Front. Chem*** (2014)

Zupančič E, *et al.* ***JCR*** (2017)

Zupančič E, *et al.* ***Nanomedicine*** (London) (2018)

Sainz V, *et al.* ***Acta Biomater*** (2018)

Conniot J, Scomparin A, *et al.* ***Nat. Nanotech*** (2019)

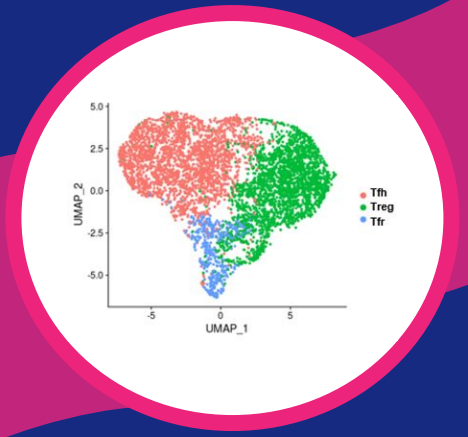


imed Research Institute for Medicines

Nano-Immunotherapeutic Approaches

Cancer nanovaccine
(Melanoma)

Cancer nanovaccine
(Germinal Center)



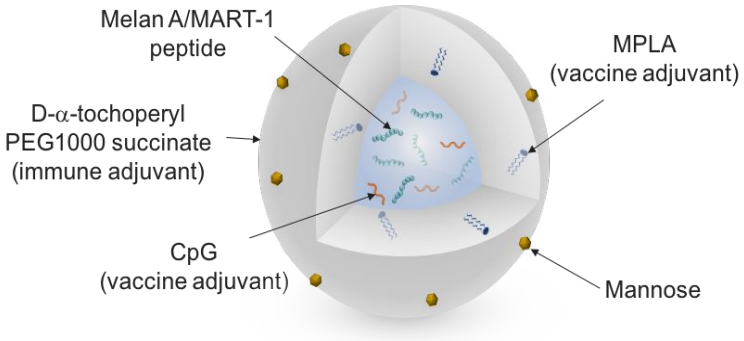
Cancer nanovaccine
(Triple Negative Breast Cancer)

Cancer nanovaccine
(Breast Cancer – Luminal B)

No breakthrough is too small

Our Approach – A Multifunctional Nanovaccine Against Melanoma

Schematic representation of man-NP



\+

Anti-PD-1, for immunosuppression blockade

Anti-OX40, for T-cell stimulation and expansion

⬇

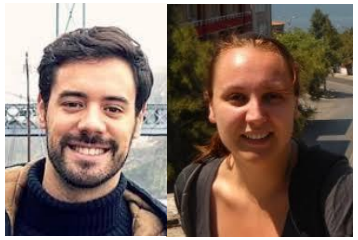
Better outcomes ?

nature nanotechnology ARTICLES
https://doi.org/10.1038/s41565-019-0512-0

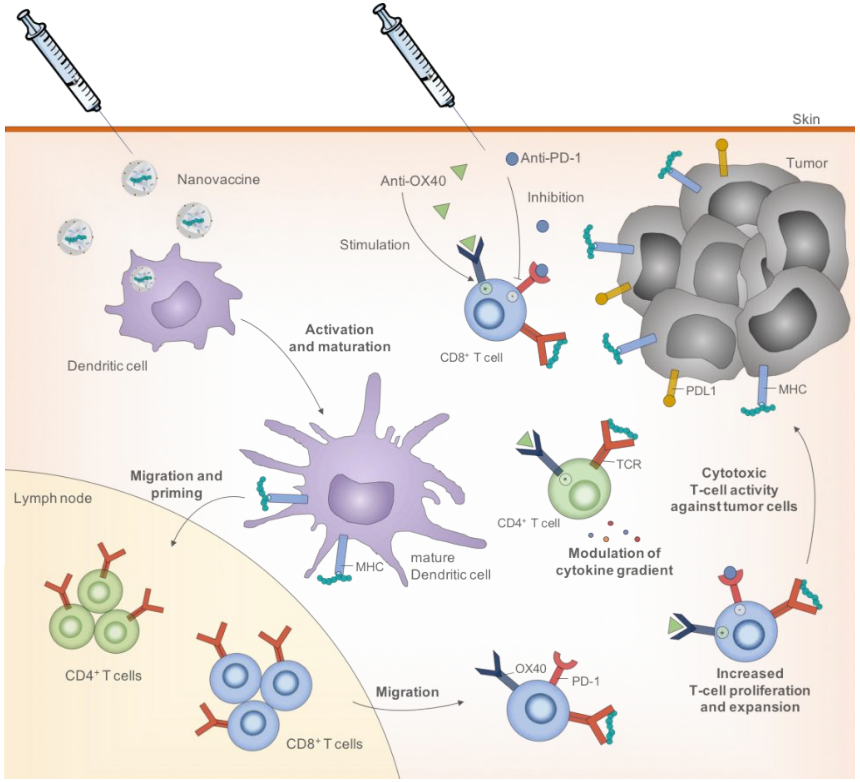
Immunization with mannosylated nanovaccines and inhibition of the immune-suppressing microenvironment sensitizes melanoma to immune checkpoint modulators

João Conniot[1,2,7], Anna Scomparin[1,3,7], Carina Peres[2], Eilam Yeini[1], Sabina Pozzi[1], Ana I. Matos[2], Ron Kleiner[1], Liane I. F. Moura[2], Eva Zupančič[2,4], Ana S. Viana[5], Hila Doron[6], Pedro M. P. Gois[2], Neta Erez[6], Steffen Jung[4], Ronit Satchi-Fainaro[1*] and Helena F. Florindo[2*]

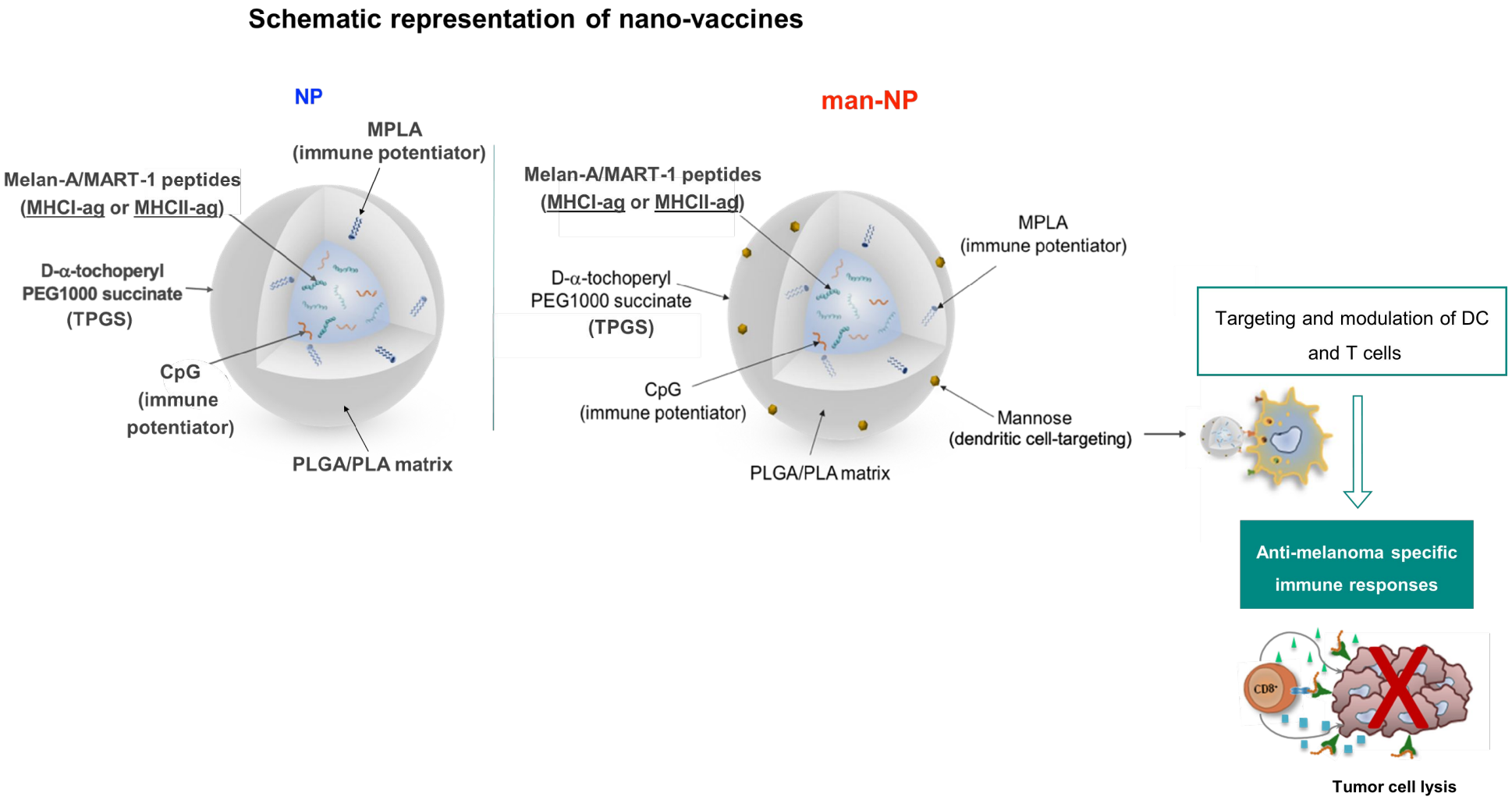
Conniot J, Scomparin A. *et al* ***Nat Nanotech*** *2019, 14 (9): 891-901*
Satchi-Fainaro, Florindo HF, Conniot J, Scomparin A.
WO/2020/136657 (July 2020)



Ronit Satchi-Fainaro



Our Approach – A Multifunctional Nanovaccine Against Melanoma



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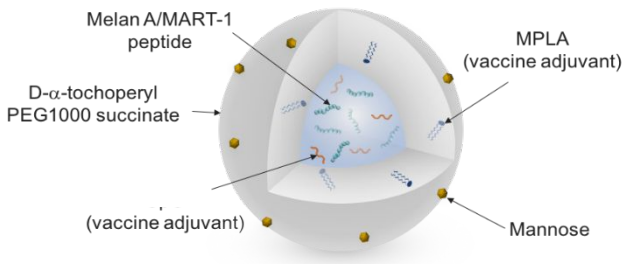
Our Approach – A Multifunctional Nanovaccine Against Melanoma

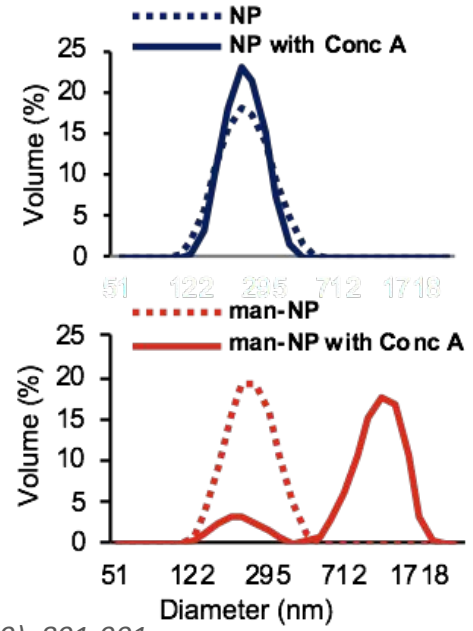
Size, polydispersity index (PdI), Zeta Potential, Entrapment Efficiency (EE) and Loading Capacity (LC)

Particles[d]	Size[a] (nm ± SD[b])	PdI ± SD[b]	Zeta Potential (mV ± SD[b])	EE (% ± SD[b])	LC (mg/mg ± SD[b])
NP (empty)	168 ± 10	0.15 ± 0.05	-2.17 ± 0.40	-	-
NP MHCI-ag	178 ± 6	0.16 ± 0.03	-3.11 ± 0.50	99.1 ± 0.1	49.6 ± 0.05
NP MHCII-ag	170 ± 5	0.18 ± 0.03	-2.34 ± 0.65	82.4 ± 0.6	41.2 ± 0.3
man-NP (empty)	169 ± 16	0.13 ± 0.05	-2.11 ± 0.40	-	-
man-NP MHCI-ag	181 ± 8	0.15 ± 0.04	-3.02 ± 0.46	97.5 ± 0.2	48.8 ± 0.1
man-NP MHCII-ag	166 ± 5	0.18 ± 0.04	-1.72 ± 0.47	74.6 ± 3.5	37.3.1 ± 1.7

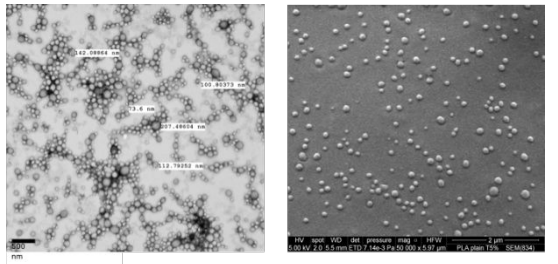
Schematic representation of man-NP

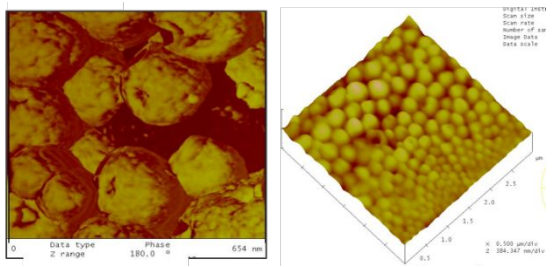


Detection scheme of mannose on the surface of NP



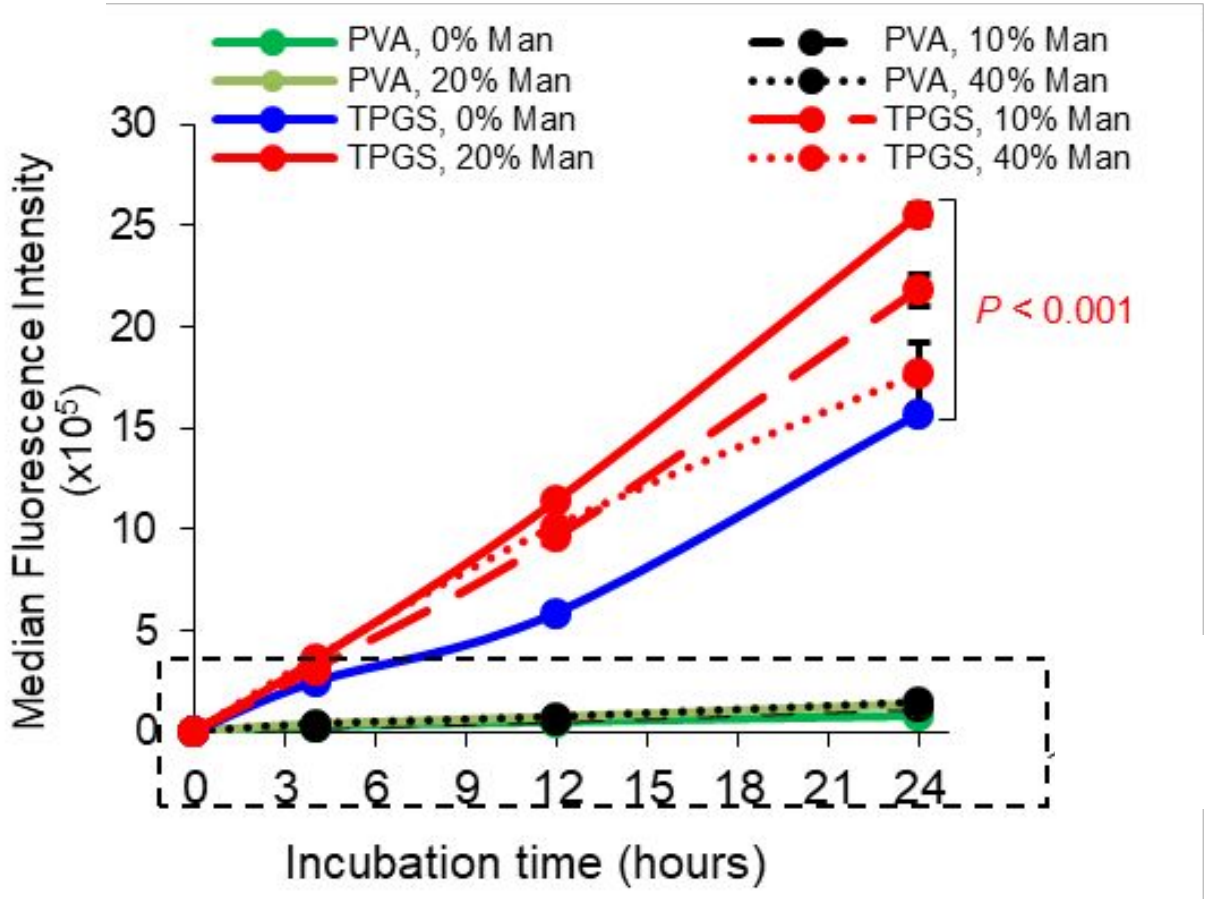
Morphological characterization

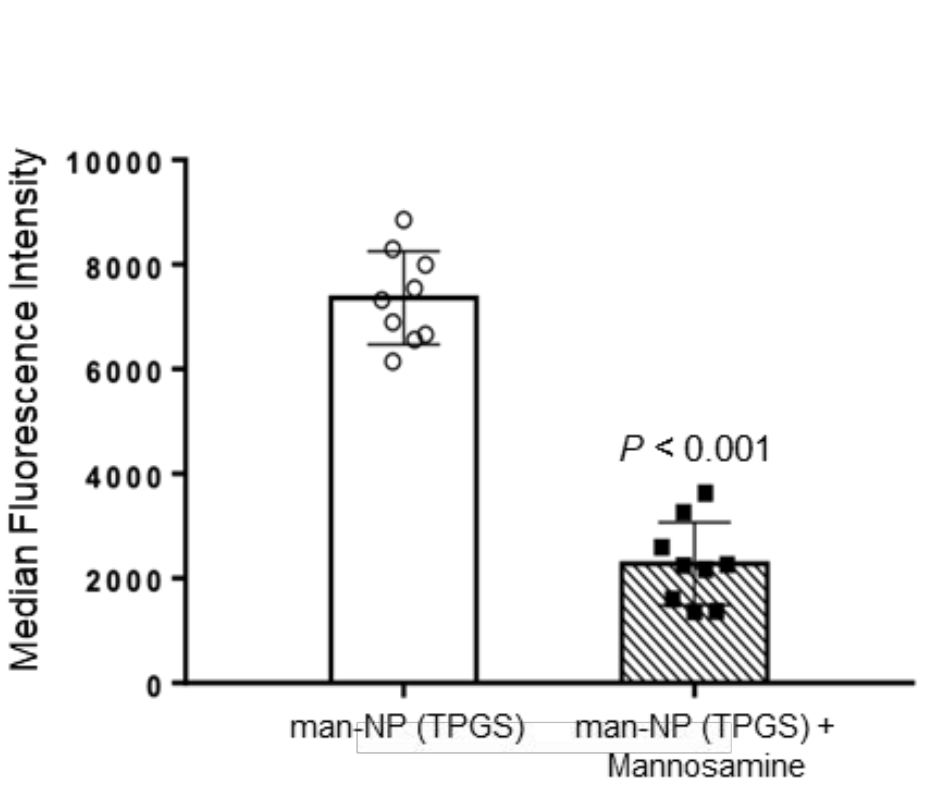




Conniot J, Scomparin A. *et al* ***Nat Nanotech*** *2019, 14 (9): 891-901*

Our Approach – A Multifunctional Nanovaccine Against Melanoma





Cy5.5-labeled particles

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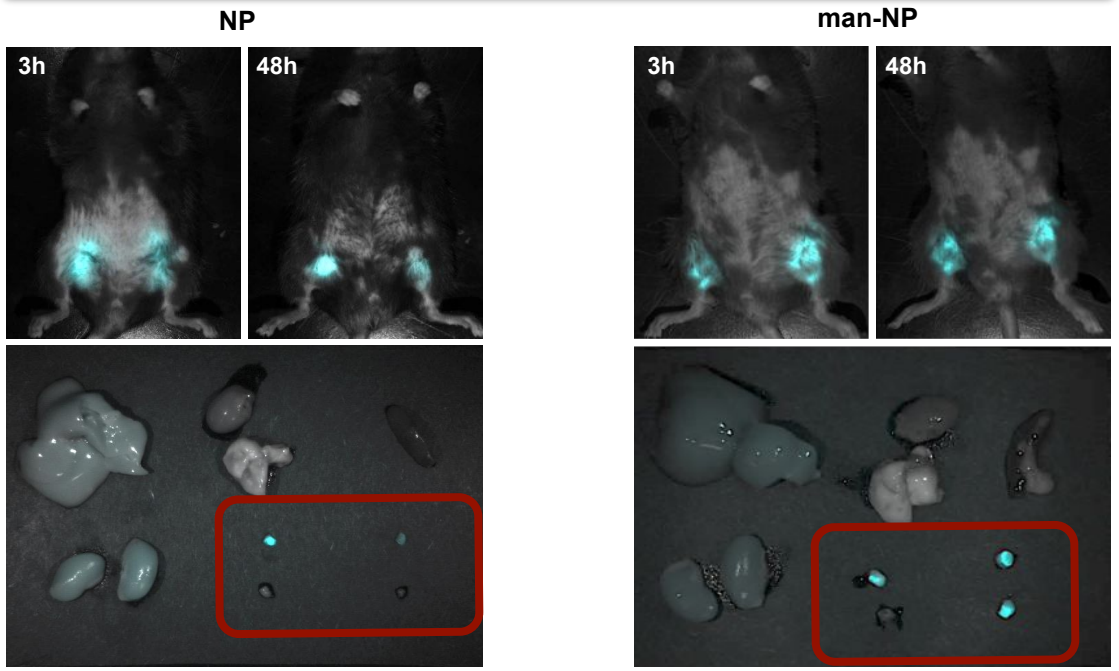
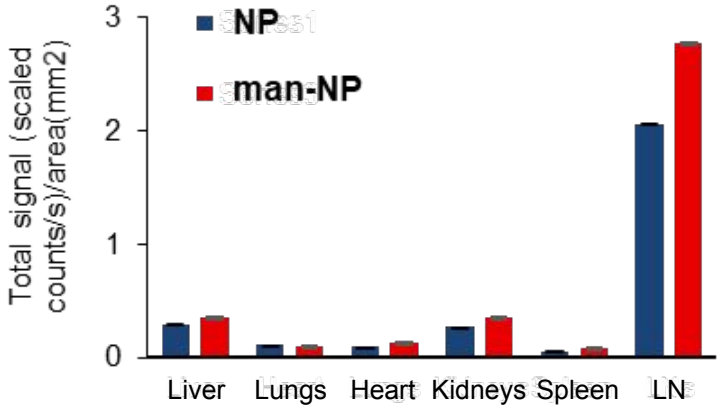
Our Approach – A Multifunctional Nanovaccine Against Melanoma

***In vivo* biodistribution profile**



Cy5.5-labeled particles



NP and man-NP preferentially accumulate in the LN

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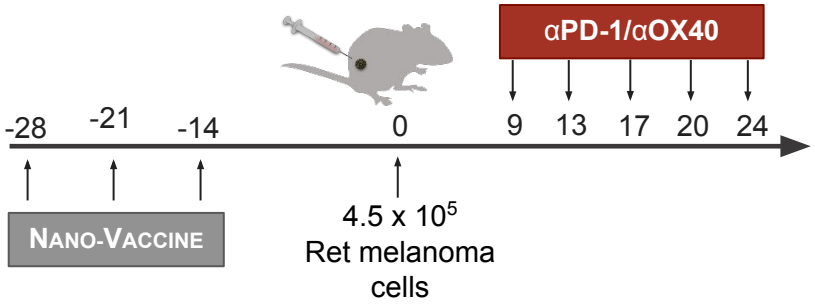

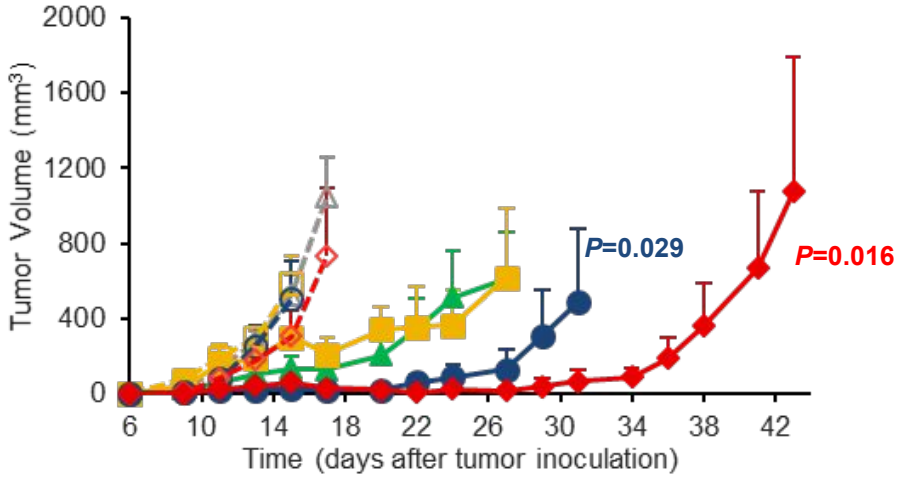

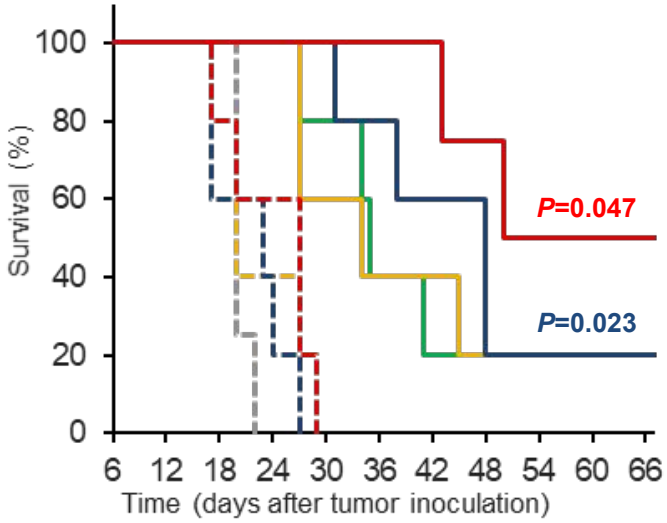

PBS
MHCI-ag/MHCII-ag (free)
NP MHCI-ag/NP MHCII-ag
man-NP MHCI-ag/man-NP MHCII-ag
αPD-1/αOX40
MHCI-ag/MHCII-ag (free) + αPD-1/αOX40
NP MHCI-ag/NP MHCII-ag + αPD-1/αOX40
man-NP MHCI-ag/man-NP MHCII-ag + αPD-1/αOX40

mean ± SEM (*N*=4-5/group)

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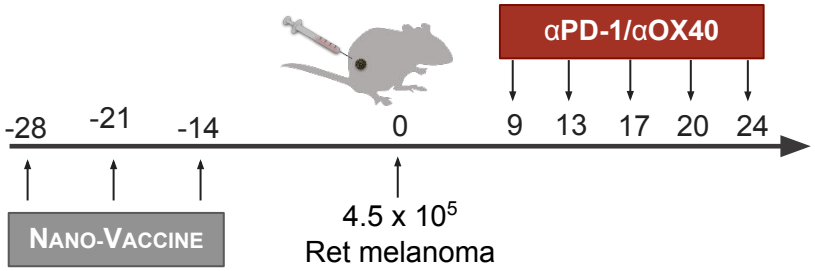
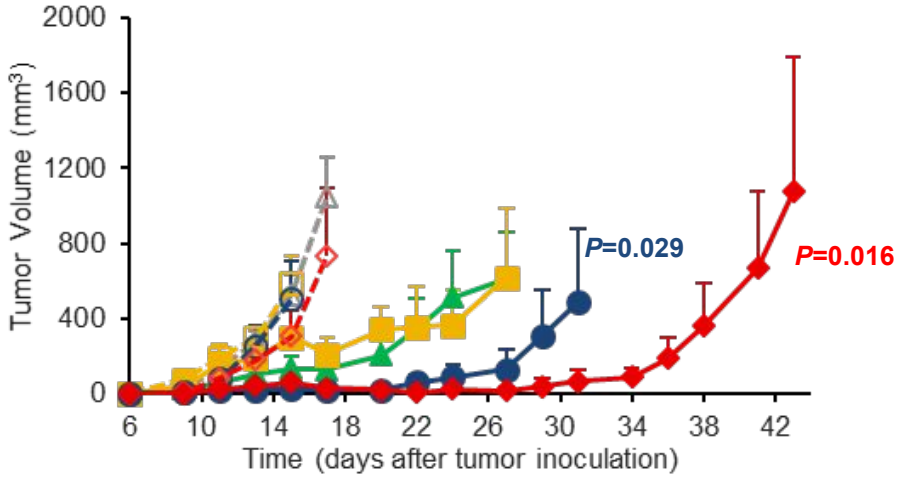
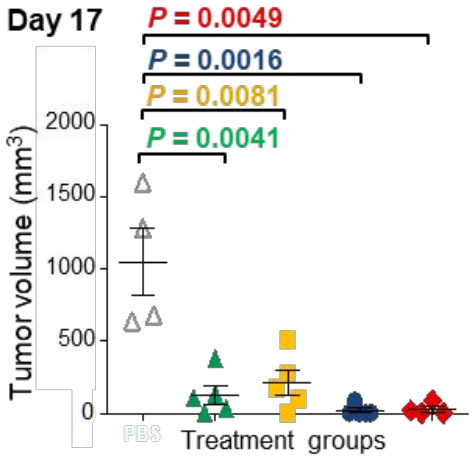
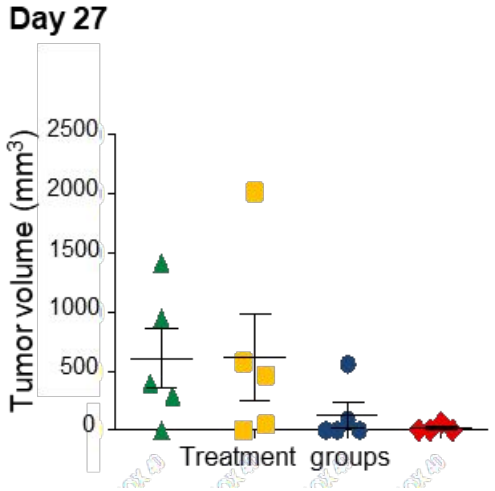






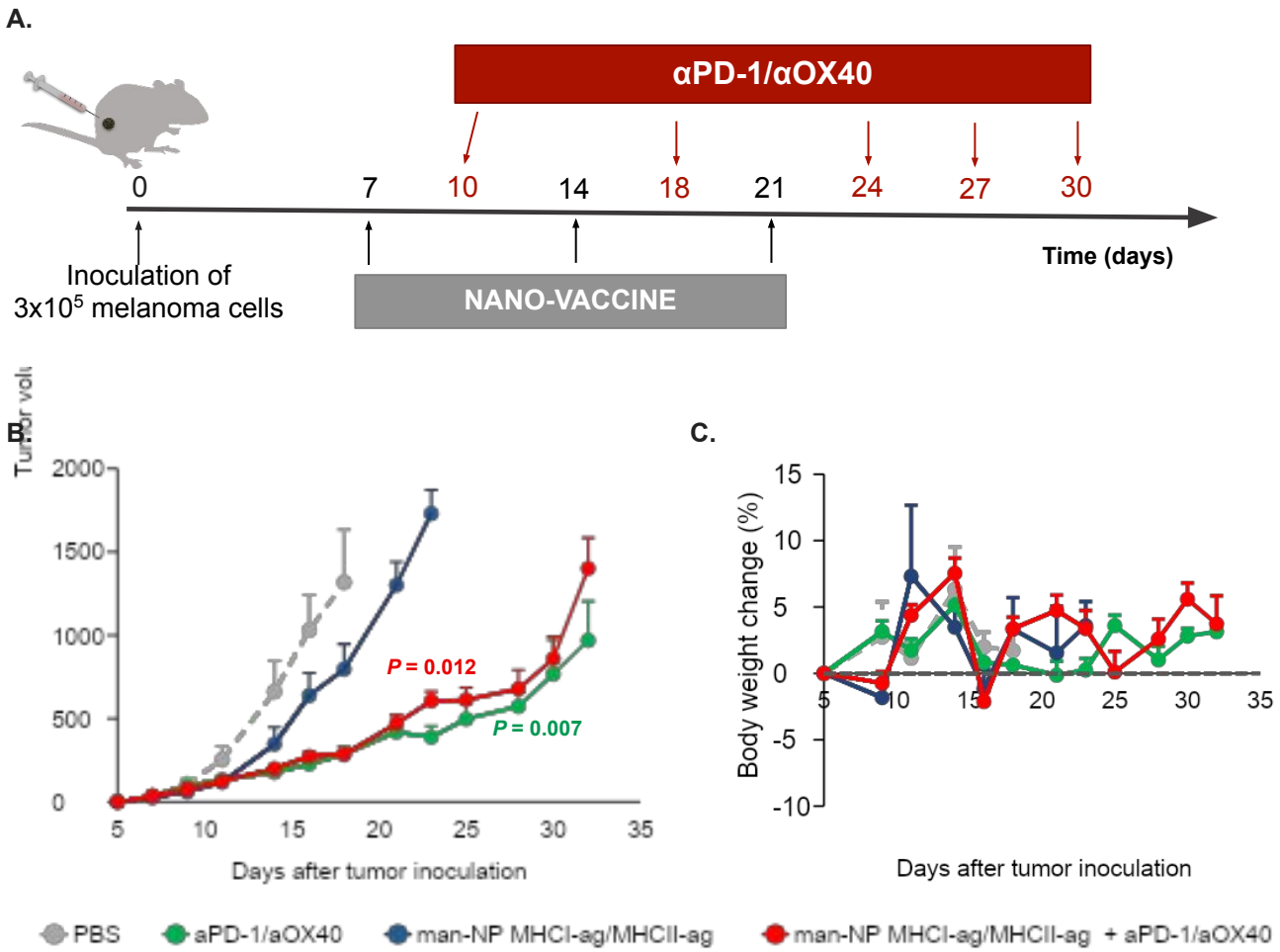


PBS
MHCI-ag/MHCII-ag (free)
NP MHCI-ag/NP MHCII-ag
man-NP MHCI-ag/man-NP MHCII-ag
αPD-1/αOX40
MHCI-ag/MHCII-ag (free) + αPD-1/αOX40
NP MHCI-ag/NP MHCII-ag + αPD-1/αOX40
man-NP MHCI-ag/man-NP MHCII-ag + αPD-1/αOX40

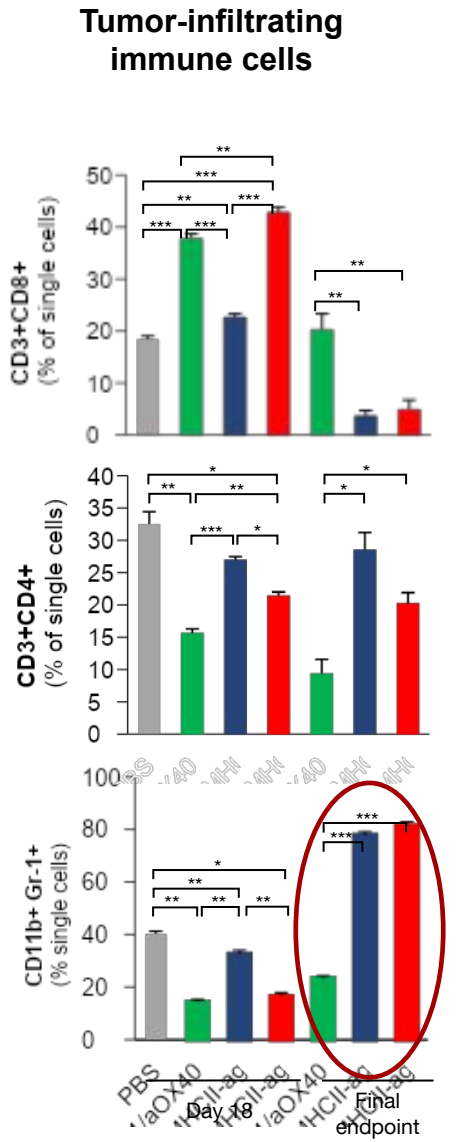
Conniot J, Scomparin A. *et al* ***Nat Nanotech*** *2019, 14 (9): 891-901*

mean ± SEM (*N*=4-5/group) *** *P* < 0.001

Our Approach – A Multifunctional Nanovaccine Against Melanoma



Data presented as mean SEM (N=7/group)



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Our Approach – A Multifunctional Nanovaccine Against Melanoma

MICROENVIRONMENT AND IMMUNOLOGY | AUTHOR CHOICE | APRIL 14 2016

Myeloid-Derived Suppressor Cells Express Bruton's Tyrosine Kinase and Can Be Depleted in Tumor-Bearing Hosts by Ibrutinib Treatment FREE

Andrew Stiff; Prashant Trikha; Robert Wesolowski; Kari Kendra; Vincent Hsu; Sarvani Uppati; Elizabeth McMichael; Megan Duggan; Amanda Campbell; Karen Keller; Ian Landi; Yiming Zhong; Jason Dubovsky; John Harrison Howard; Lianbo Yu; Bonnie Harrington; Matthew Old; Sean Reiff; Thomas Mace; Susheela Tridandapani; Natarajan Muthusamy; Michael A. Caligiuri; John C. Byrd; William E. Carson, III

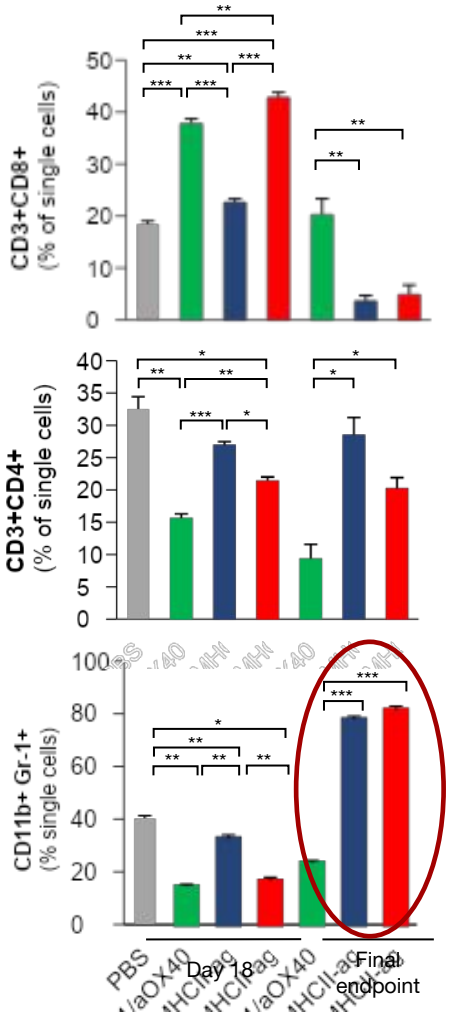
Check for updates

+ Author & Article Information

Cancer Res (2016) 76 (8): 2125–2136.

https://doi.org/10.1158/0008-5472.CAN-15-1490 **Article history**

Tumor-infiltrating immune cells

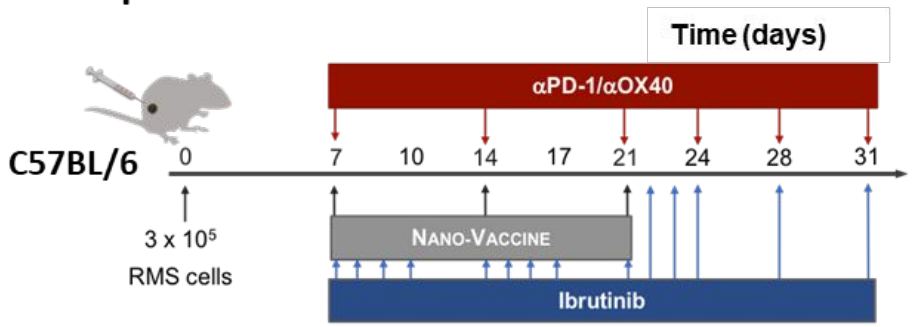


* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$

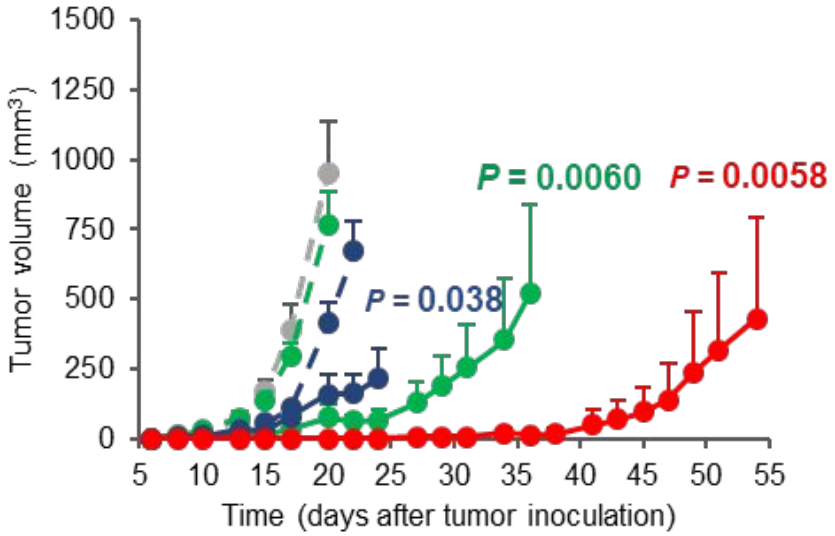
Conniot J, Scomparin A. *et al* ***Nat Nanotech*** *2019, 14 (9): 891-901*

Mannosylated Nanoaccine with Inhibition of MDSC Sensitizes Melanoma to Immune Checkpoint Therapies

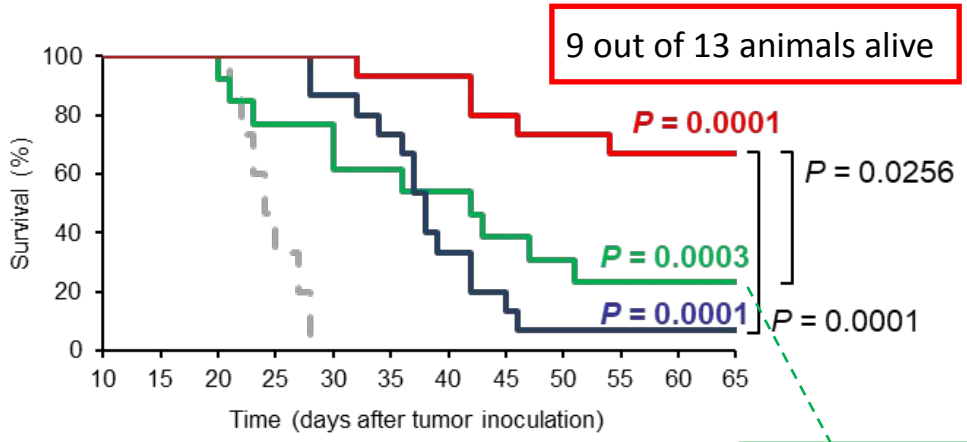
Therapeutic scheme



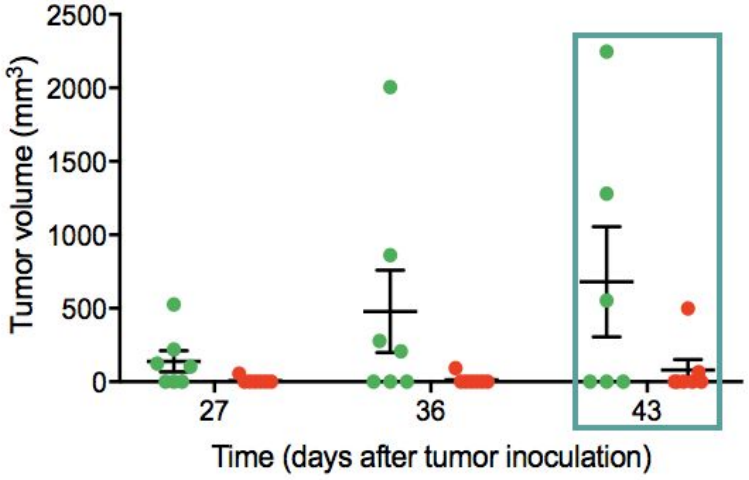
PBS
aPD-1/aOX40 + MDSC Inhibitor
man-NP + aPD-1/aOX40
MDSC Inhibitor
man-NP + MDSC Inhibitor
man-NP + aPD-1/aOX40 + MDSC Inhibitor



Overall survival



mean ± SEM (*N*=13/group)
replicated in 3 independent experiments



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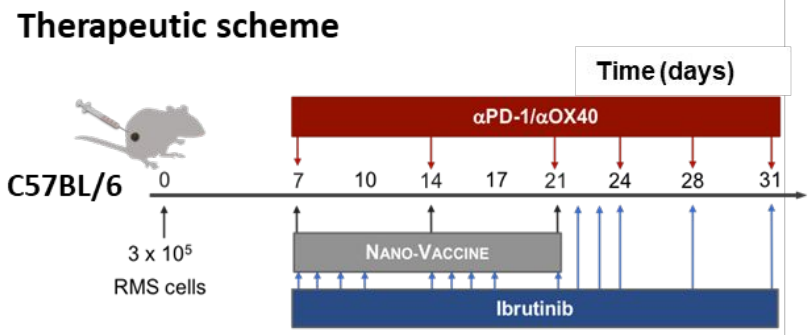
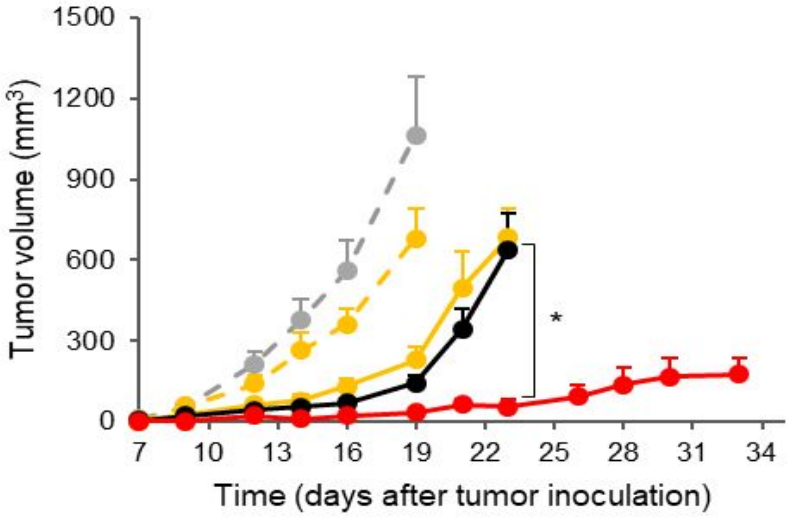
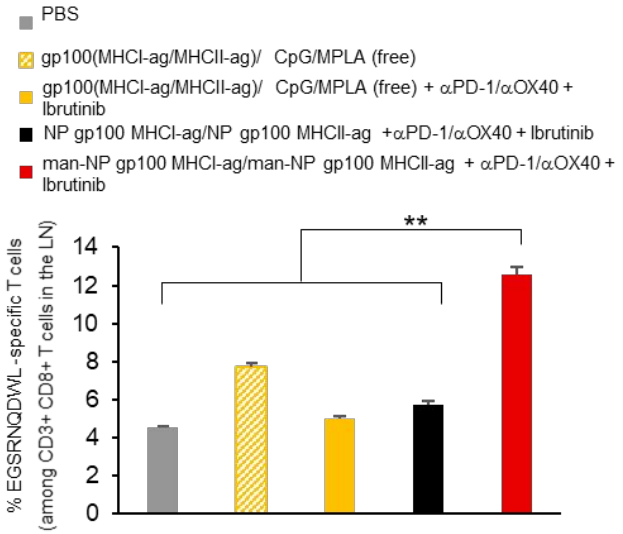
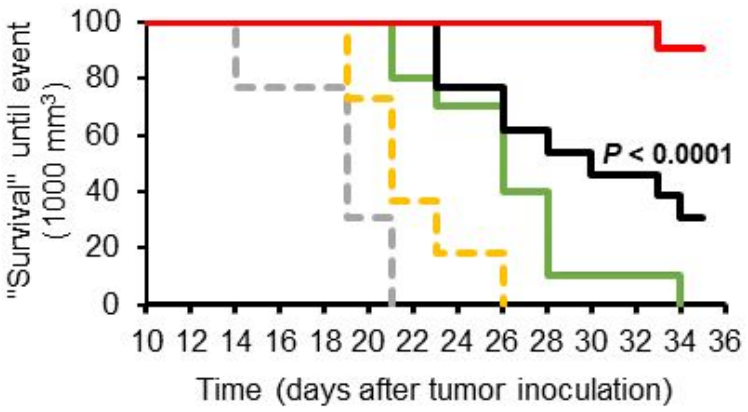
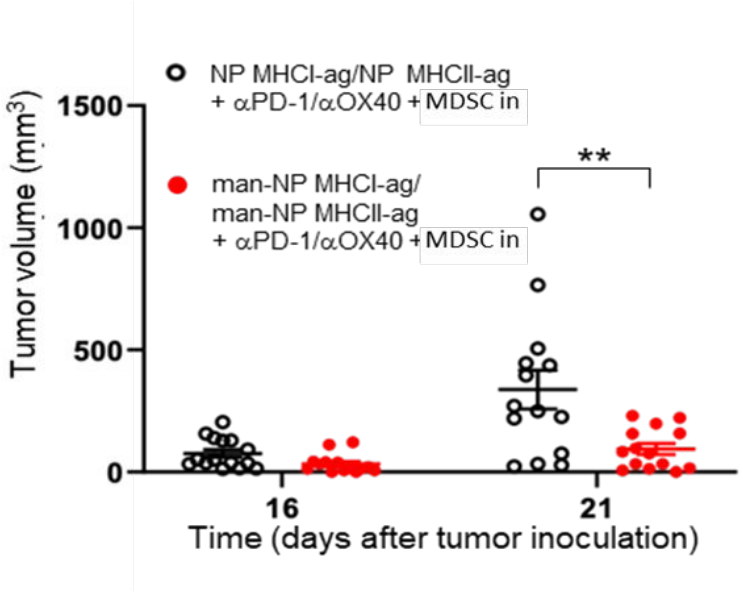
Mannosylated Nanoaccine with Inhibition of MDSC Sensitizes Melanoma to Immune Checkpoint Therapies

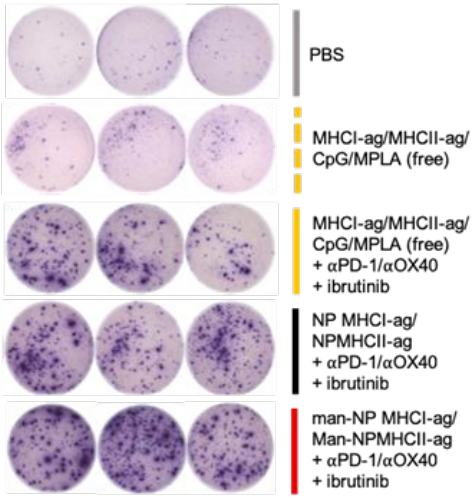








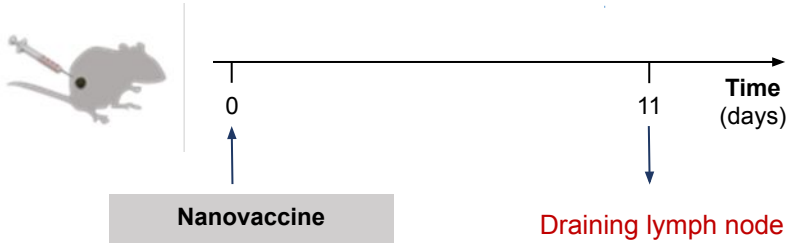




N=13-15/group; * $P < 0.05$; ** $P < 0.001$
replicated in 3 independent experiments

Conniot J, Scomparin A. *et al* ***Nat Nanotech*** *2019, 14 (9): 891-901*

Nanomaterials Change the Characteristics of the Germinal Center Responses



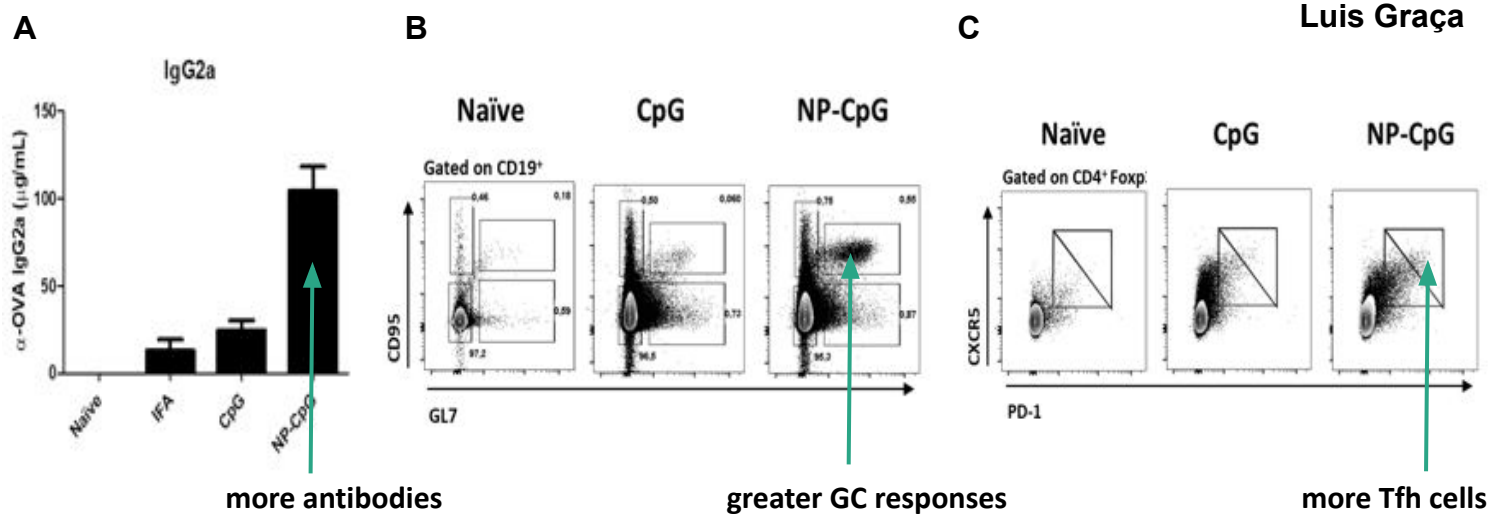
T follicular helper cells (Tfh)

T follicular regulatory (Tfr) cells

IFA- Incomplete Freund's Adjuvant

Nanomaterials to improve vaccine effectiveness

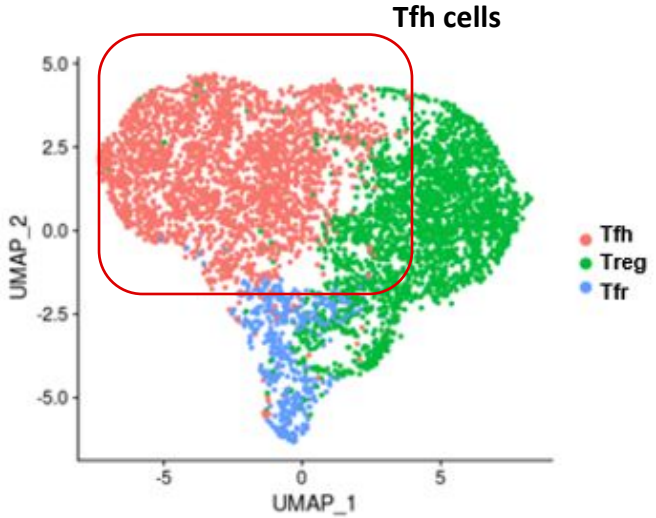
Luis Graça

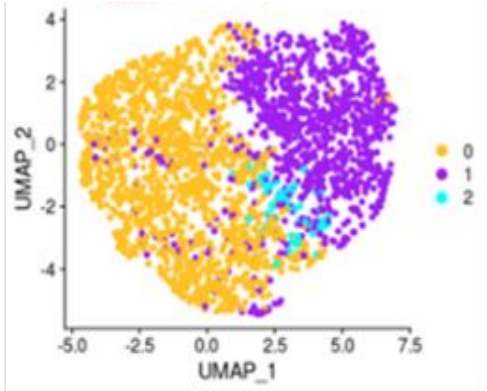


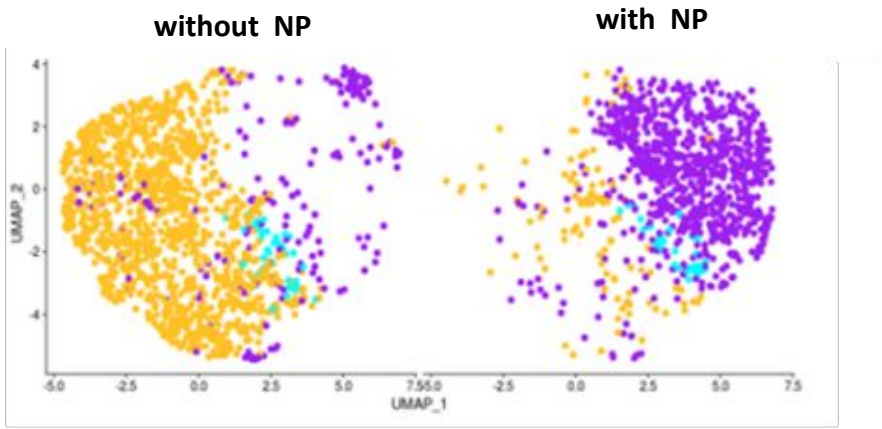
more antibodies

greater GC responses

more Tfh cells







Rodrigues et al., unpublished; ***DO NOT POST***

N = 2; *n* = 3; Mean ± SD (*N*=10/group) *** *P* < 0.001

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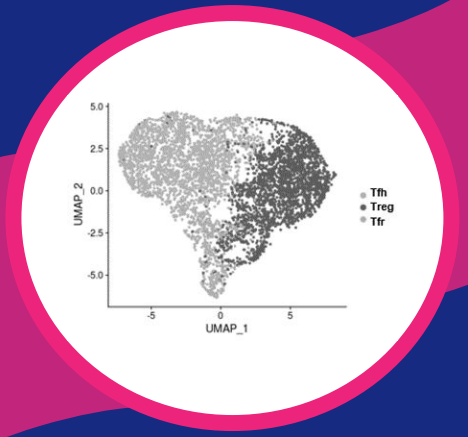
Nano-immunotherapeutic approaches

Cancer nanovaccine
(Melanoma)

Cancer nanovaccine
(Germinal Center)

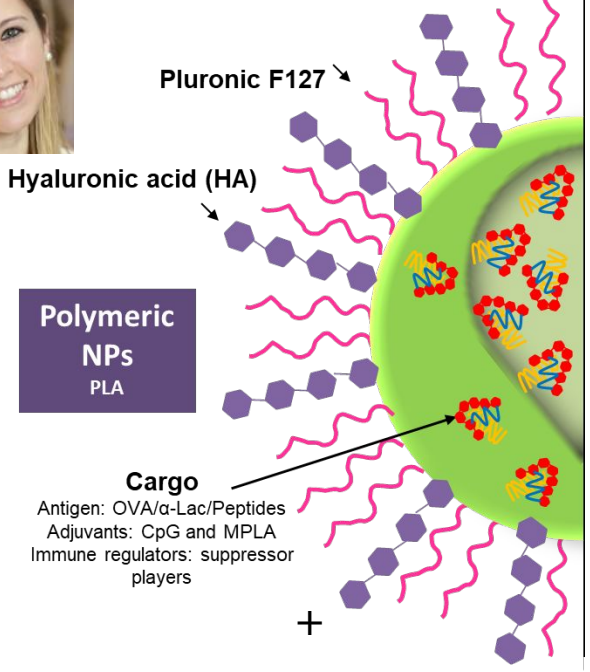


Cancer nanovaccine
(Triple Negative Breast Cancer)

Cancer nanovaccine
(Breast Cancer – Luminal B)

No breakthrough is too small

Our Approach – A Multifunctional Nanovaccine against Breast Cancer

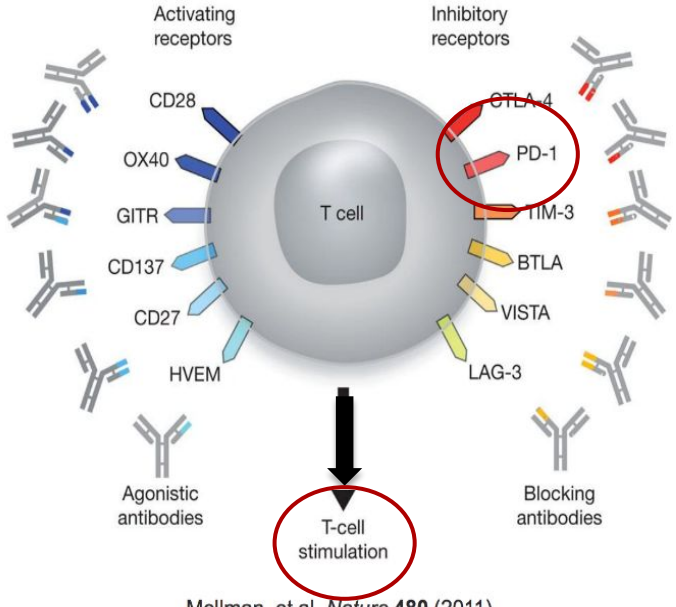


Anti-OX40 (agonist), for T-cell stimulation and expansion

Triple-negative breast cancers (TNBC) account for ~15–20% of all diagnosed breast cancer cases.

Complex molecular landscape (heterogenous 4-6 molecular types)
Low detection rate
Highly proliferative

Worst prognosis and multiple drug resistant compared to non-TNBC
Advanced TNBC - Unmet medical need



Mellman, et al. *Nature* **480** (2011)

Modest immune cytolytic activity
Tumor mutations
Modest load of neo-epitopes

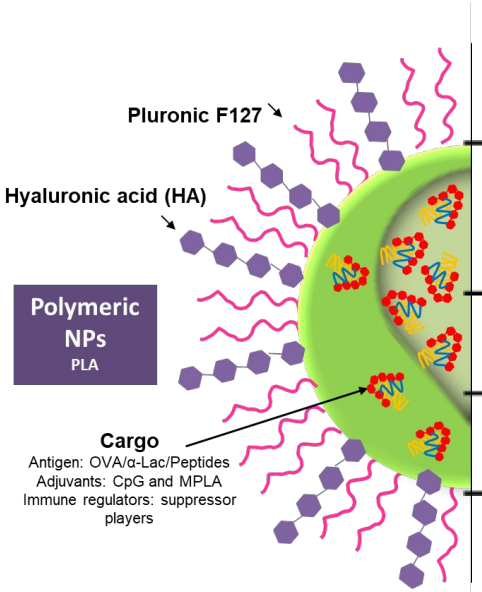
12% - 19% response rates to α-PD-1/ α-PD-L1 in Advanced TNBC

Peres C, et al., unpublished.
Please do not post.

Ronit Satchi-Fainaro

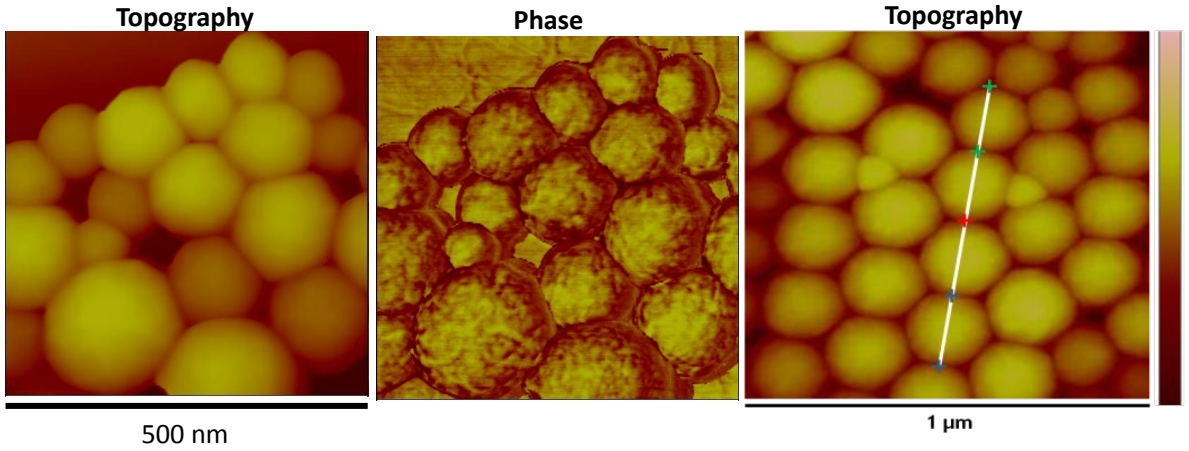
Véronique Préat

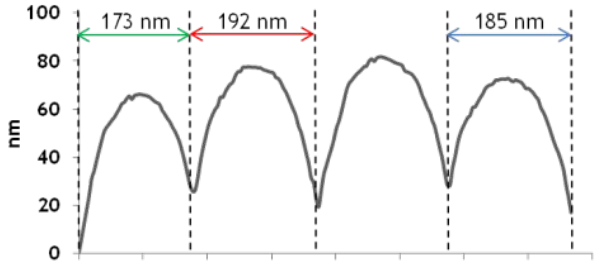
Breast Cancer Nanovaccine Physicochemical Characterization



Nanovaccines	Z-ave (nm)	PdI	ZP (mV)	Antigens EE (%)	Adjuvants EE (%)	Reg. EE (%)
NPs + Ag + Adj + Reg.	208 ± 8	0.13 ± 0.02	-4.65 ± 0.47	86.2 ± 10.9	95.1 ± 1.2	93.0 ± 0.93
NPs + Adj + Reg.	206 ± 6	0.15 ± 0.03	-5.69 ± 0.89	-	93.9 ± 2.3	95.1 ± 0.54

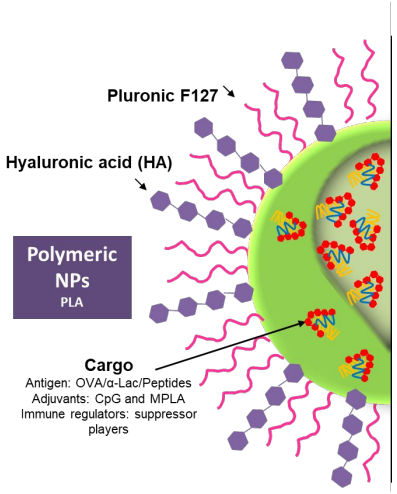
Z-ave: Z-average hydrodyamic diameter; PdI: polydispersity index; ZP: ζ-potential; EE: entrapment efficiency

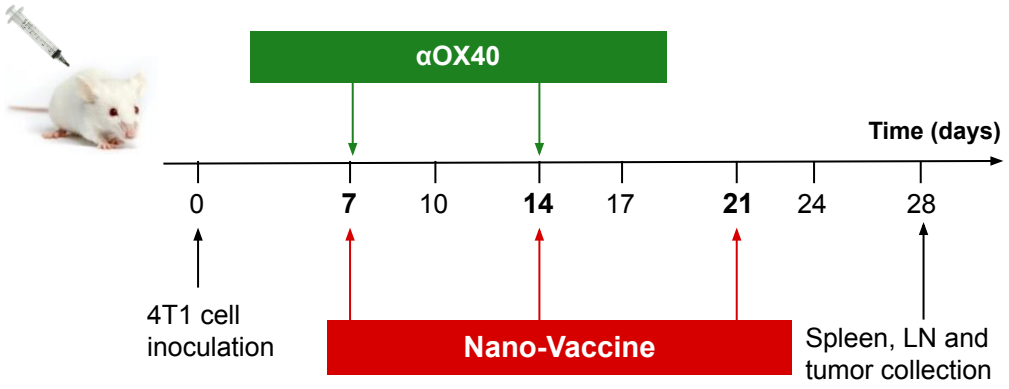


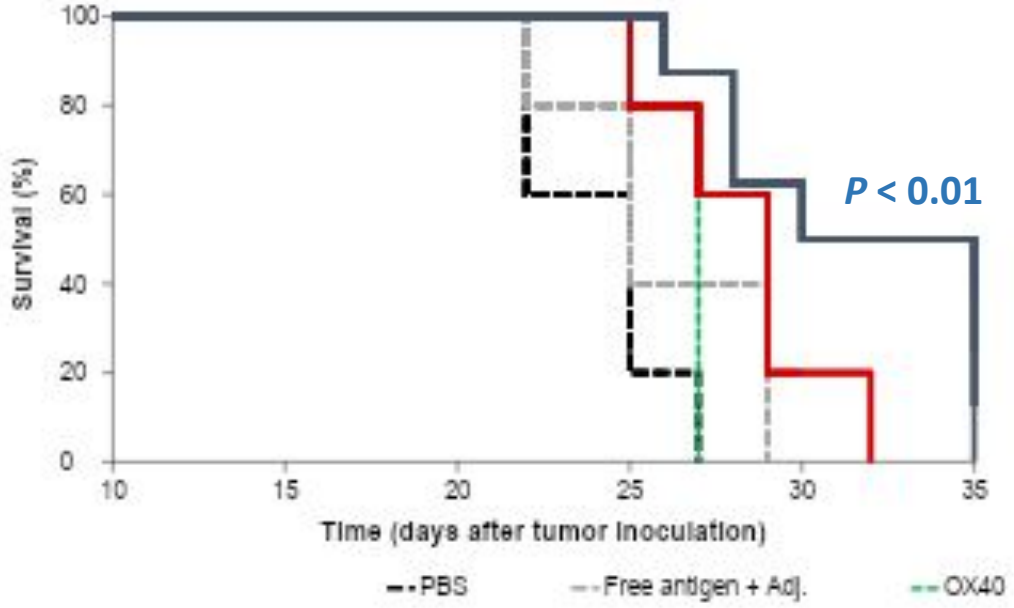


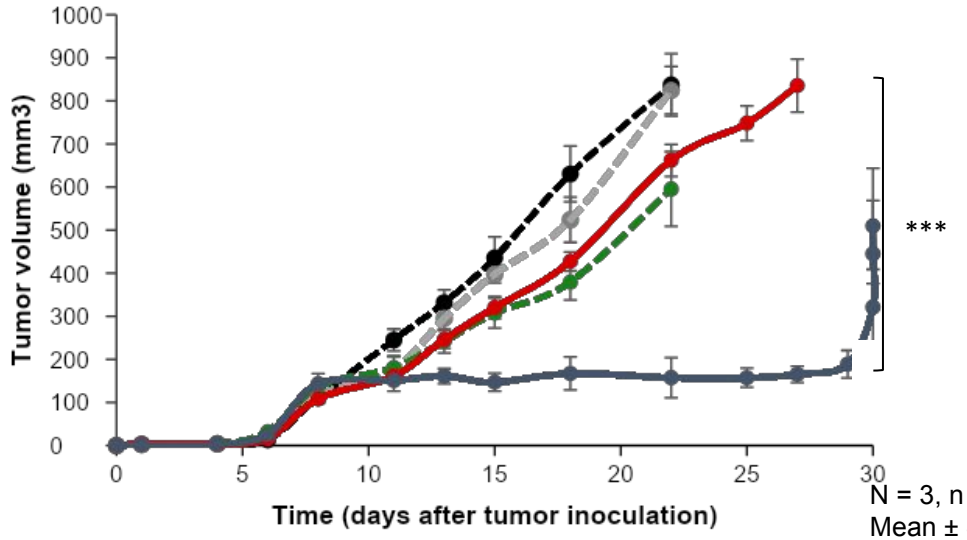
Peres C. et al., unpublished. ***PLEASE DO NOT POST.***

Proof-of-concept – *Therapeutic efficacy of nanovaccine in combination with OX40 modulation*





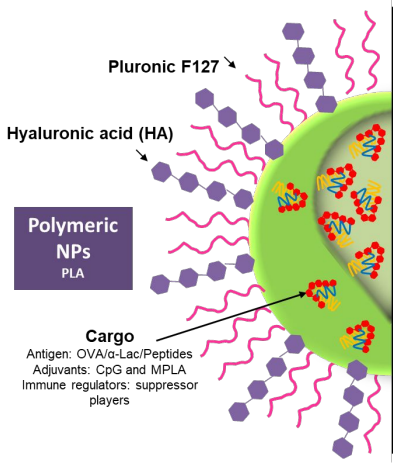


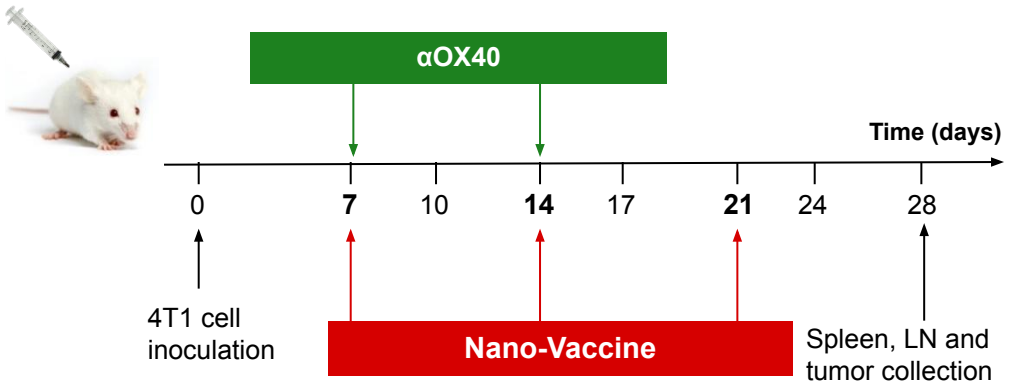


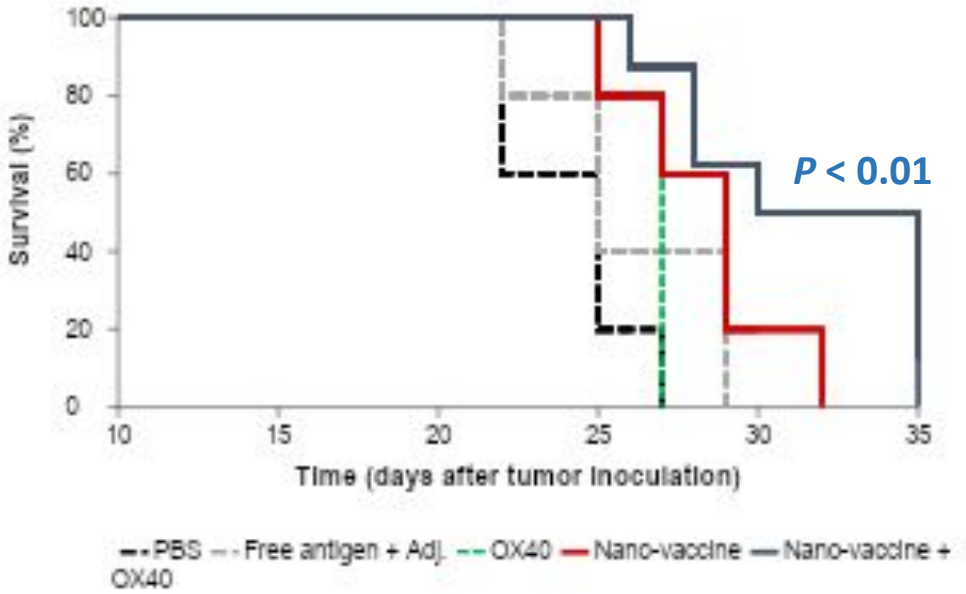
N = 3, n = 12,
Mean ± SEM *** *P*< 0.001

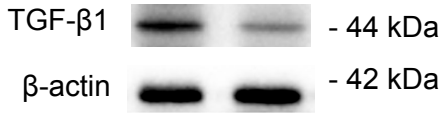
Peres C. et al., unpublished. ***PLEASE DO NOT POST.***

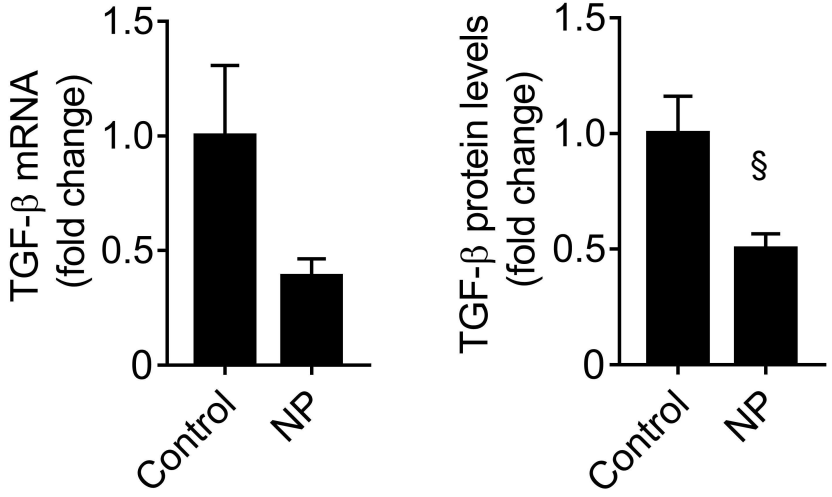
Proof-of-concept – *Therapeutic efficacy of nanovaccine in combination with OX40 modulation*







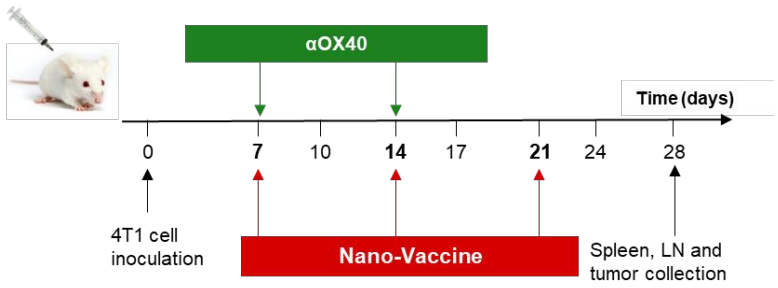


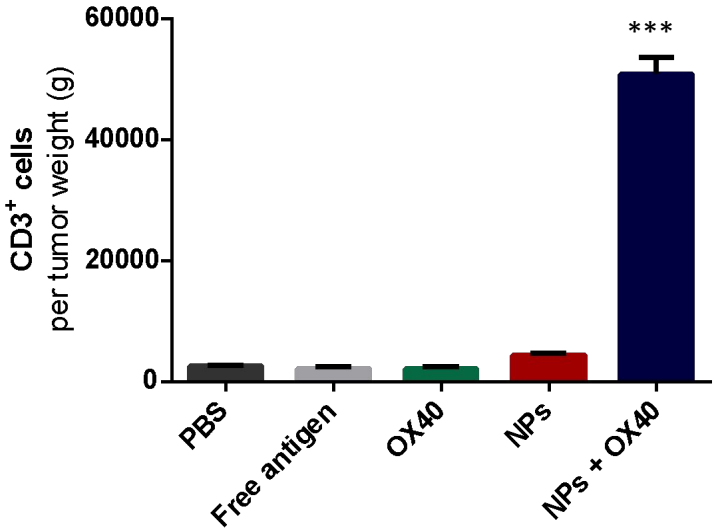


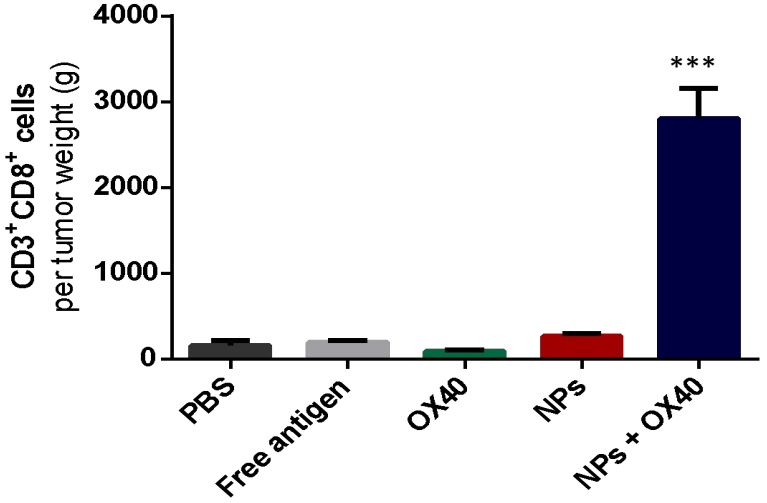
Peres C. et al., unpublished. ***PLEASE DO NOT POST.***

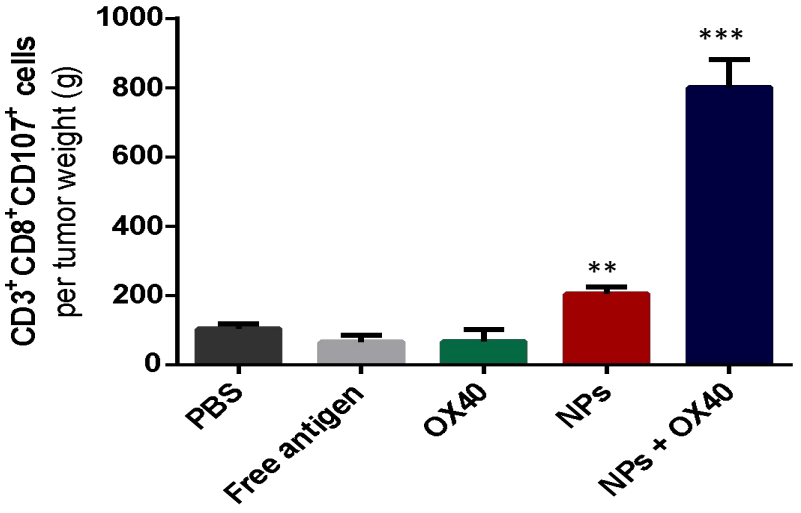
N = 3, n = 3, Mean ± SEM $P < 0.05$

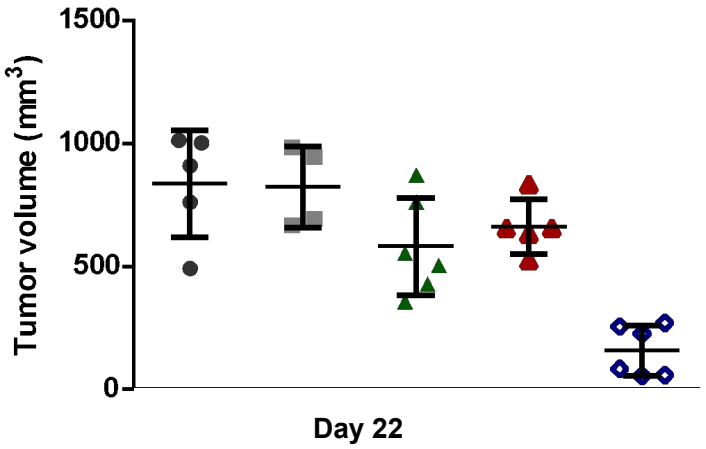
Proof-of-concept – *Therapeutic efficacy of nanovaccine in combination with OX40 modulation*

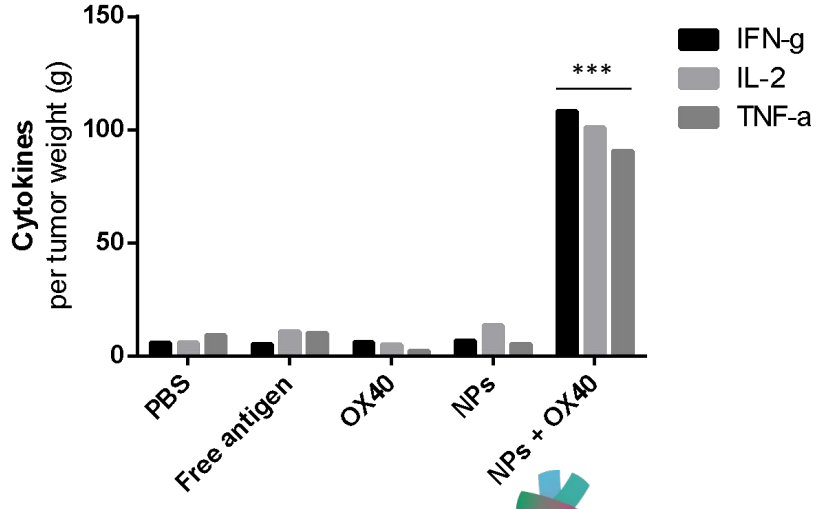








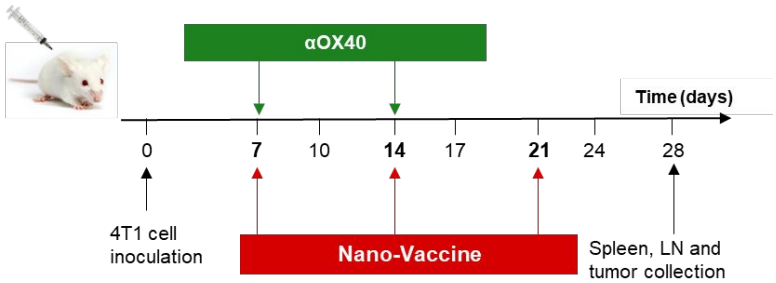


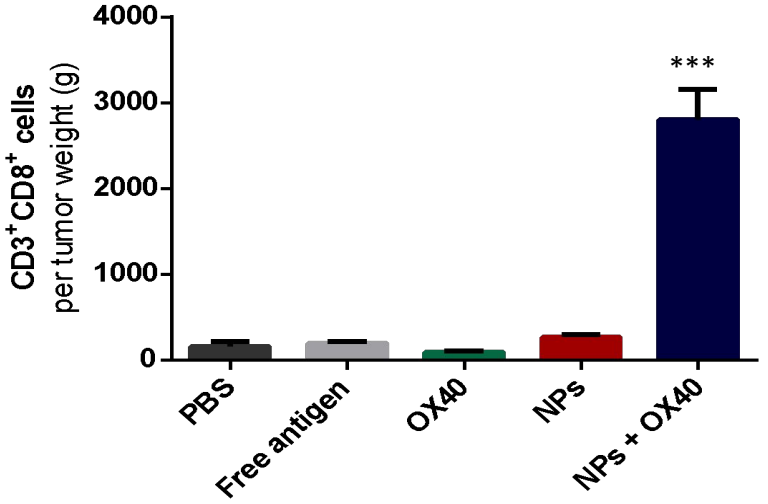


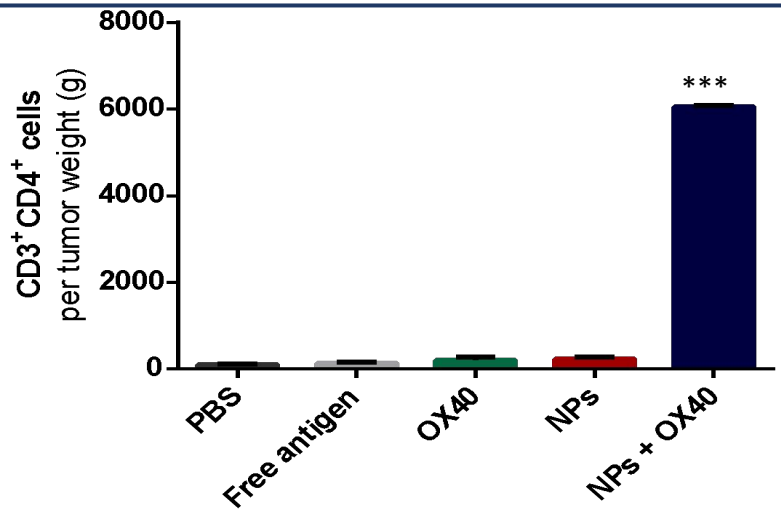
N = 3, n = 3, Mean ± SEM $^{**}P < 0.01$, $^{***}P < 0.001$

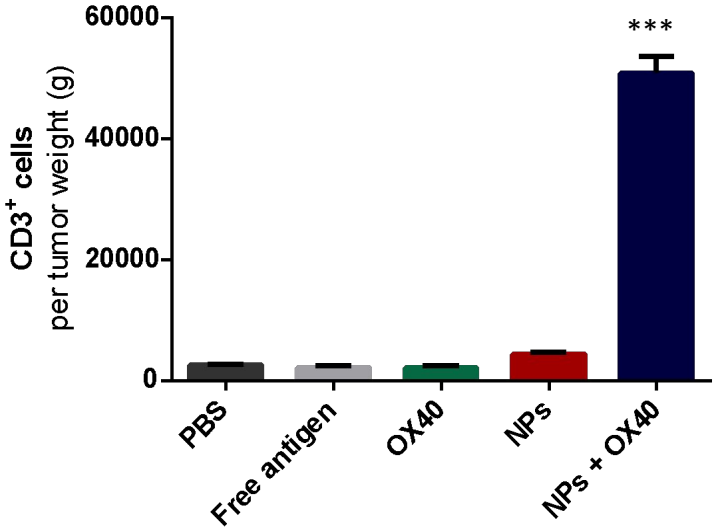
Peres C. et al., unpublished. ***PLEASE DO NOT POST.***

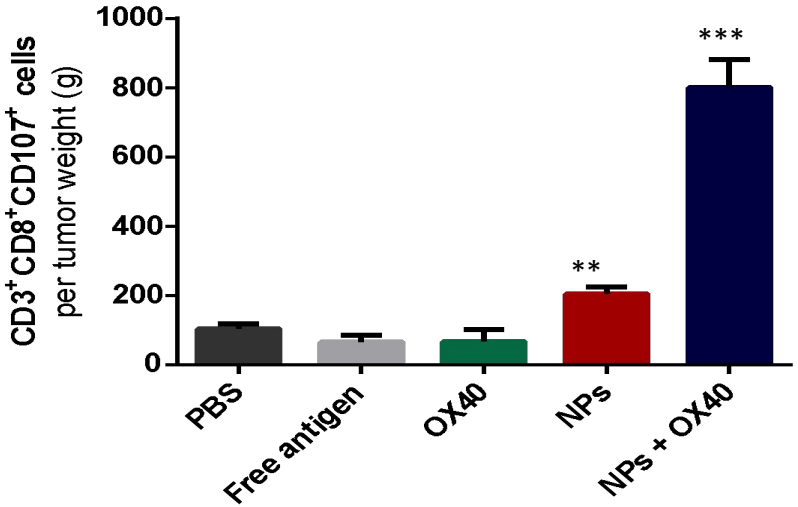
Proof-of-concept – *Therapeutic efficacy of nanovaccine in combination with OX40 modulation*

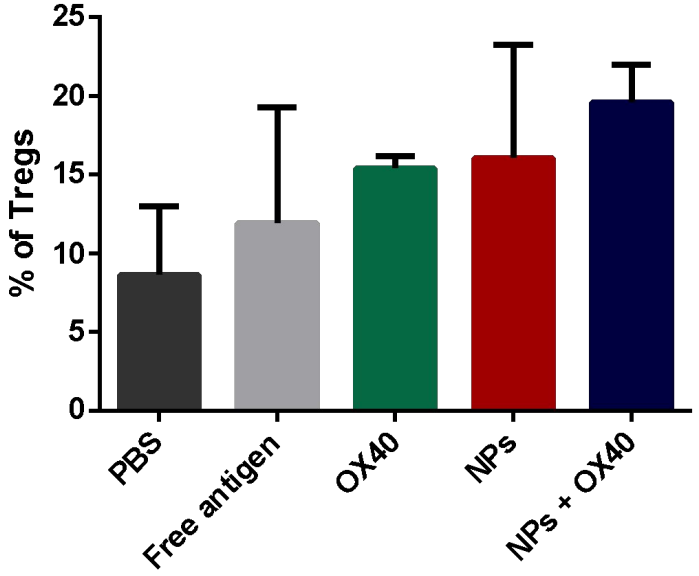








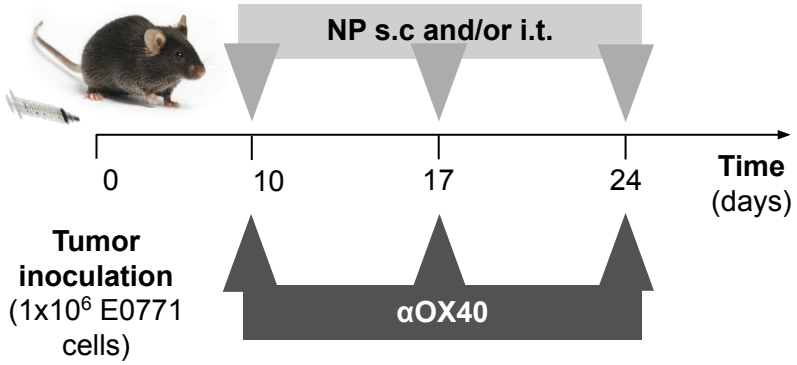


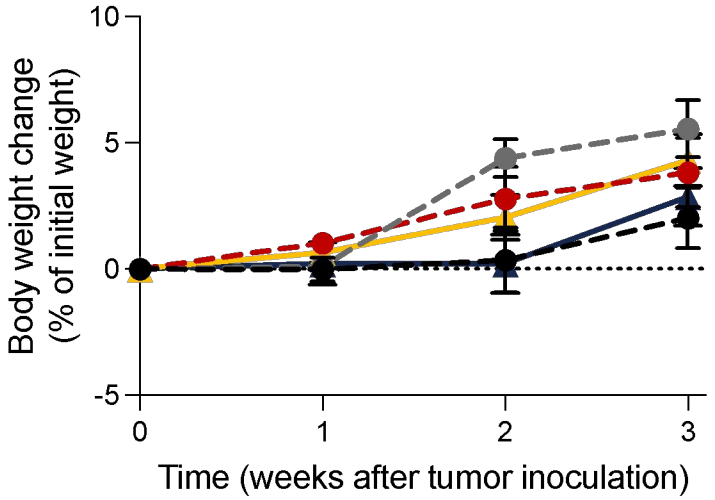


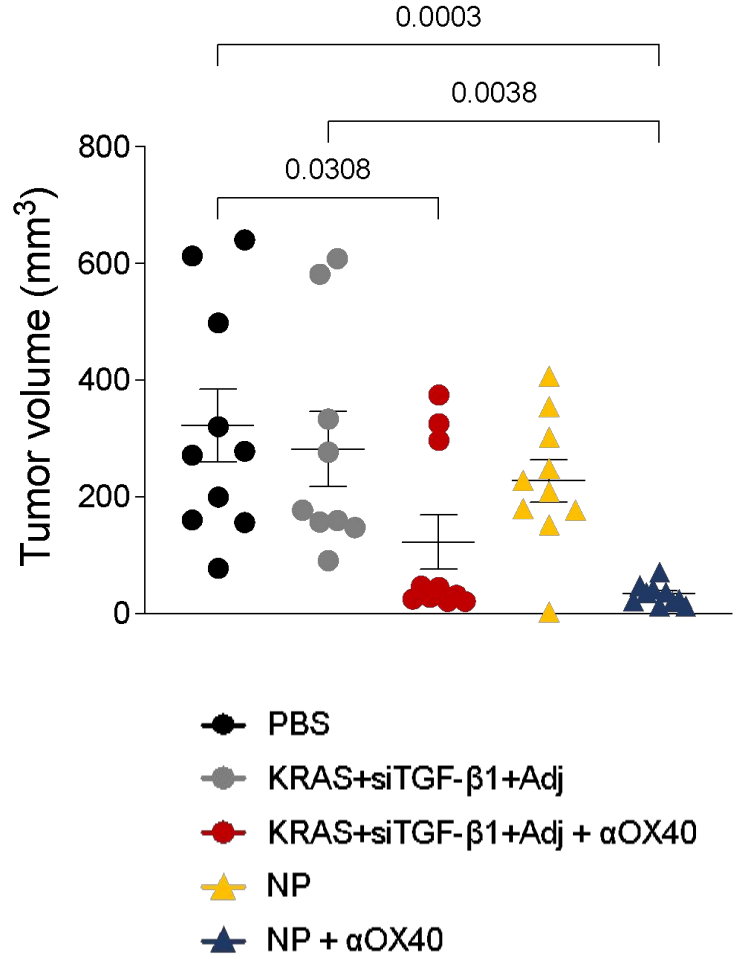
N = 3, n = 3, Mean ± SEM $^{**}P < 0.01$, $^{***}P < 0.001$

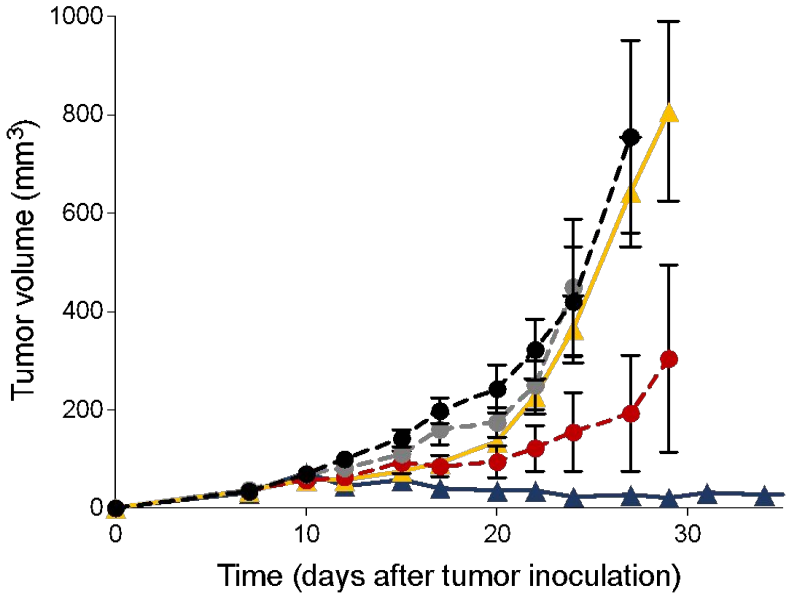
Peres C. et al., unpublished. ***PLEASE DO NOT POST.***

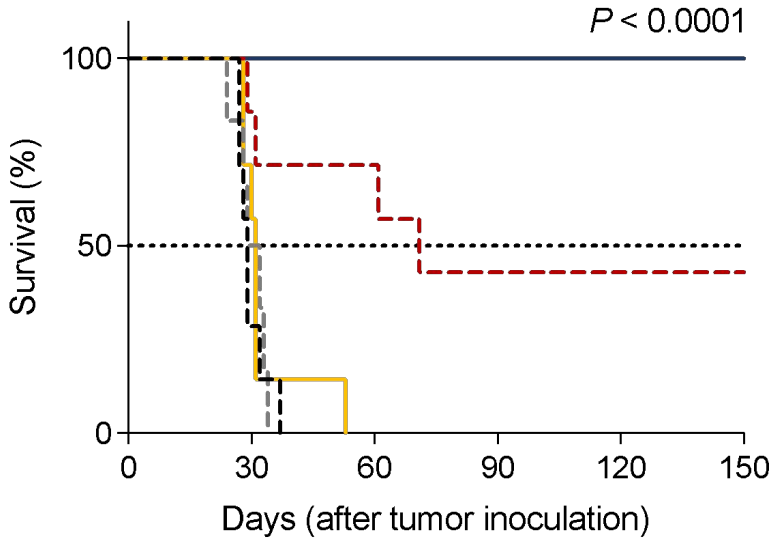
Proof-of-concept – *Therapeutic efficacy of nanovaccine in combination with OX40 modulation*











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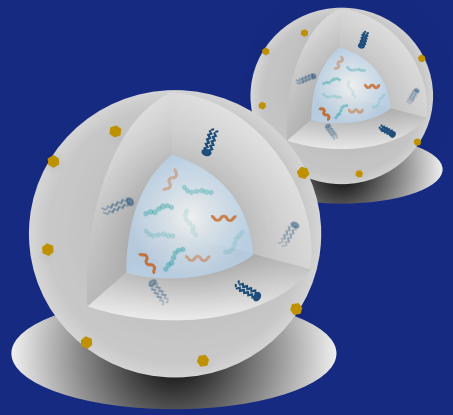

NANO-IMMUNOTHERAPY FOR SOLID TUMORS:

- **Antigen-specific cellular immunity triggered by combining the delivery of shorth (MHC class I)** and long (**MHC class II) T cell peptide** sequences with adjuvants (e.g., TLR ligands):
  - Activation of IFN-γ producing $CD4^+$ T helper cells;
  - Reduction or non-activation of Treg cells
  - Effector Cytotoxic T Lymphocytes
- **Modulation of tumor immunosuppressive environment**
- **Humoral immunity** leading the secretion of **high-affinity antibodies and memory immune response.**
- **Suitable for different routes of administration –** modulation of lymphoid structures at distinct locations.

Acknowledgments

BioNanoSciences – Drug Delivery & Immunoengineering

- **Bárbara Carreira**
- **Carina Peres**
- Rita Acúrcio
- Liane Moura
- Ana I. Matos
- Mariana Bento
- Andrícia Bonomo
- Cláudio Ferro
- Nicole Mendes
- Jéssica Luís
- Ana Carolina Santos
- Raquel Gouveia

Prof. R. Satchi-Fainaro
A. Somparin, Ph.D
E. Yeini, M.Sc.
S. Pozzi, M.Sc.

Prof. S. Jung lab
Prof. L. Eisenbach
C. Curato, Ph.D

Prof. R. Jordan Lab
E. Wegener, Ph.D

Dr. Marta Pojo lab
Joaquim Brito, M.D.

Prof. V. Préat Lab

Vitor Farricha, M.D.

Prof. L. Graça Lab
Afonso Bastos, Ph.D.
Filipa Ribeiro, M.Sc.

Prof. Luis Costa lab
Sandra Casimiro, PhD

Prof. J. P. Conde Lab
Eduardo Brás, M.Sc

Prof. J. Gonçalves lab
Prof. C. Rodrigues lab
Marta Afonso, Ph.D
Prof. Rita Guedes
Prof. Ana Paula Leandro

Prof. Maria Vicént
Alessio Malfanti, Ph.D

PTDC/BTM-SAL/4350/2021
UTAP-EXPL/NPN/0041/2021
EXPL/MED-QUI/1316/2021

LCF/TR/CD20/52700005
LCF/PR/HR19/52160021

UTAP-EXPL/NPN/0041/2021

No breakthrough is too small

Thank you!!

CRS 2022 Annual Meeting & Expo

July 11 – 15, 2022 | Montreal Congress Center, Montreal Canada

Advanced Delivery Science