The Protein Corona of Nanoparticles as a Tool for the Early Detection of Pancreatic Cancer

NANODELIVERY LAB
Smart Nanocarriers for Drug and Gene Delivery

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1. The Protein Corona (PC) of nanoparticles (NPs)

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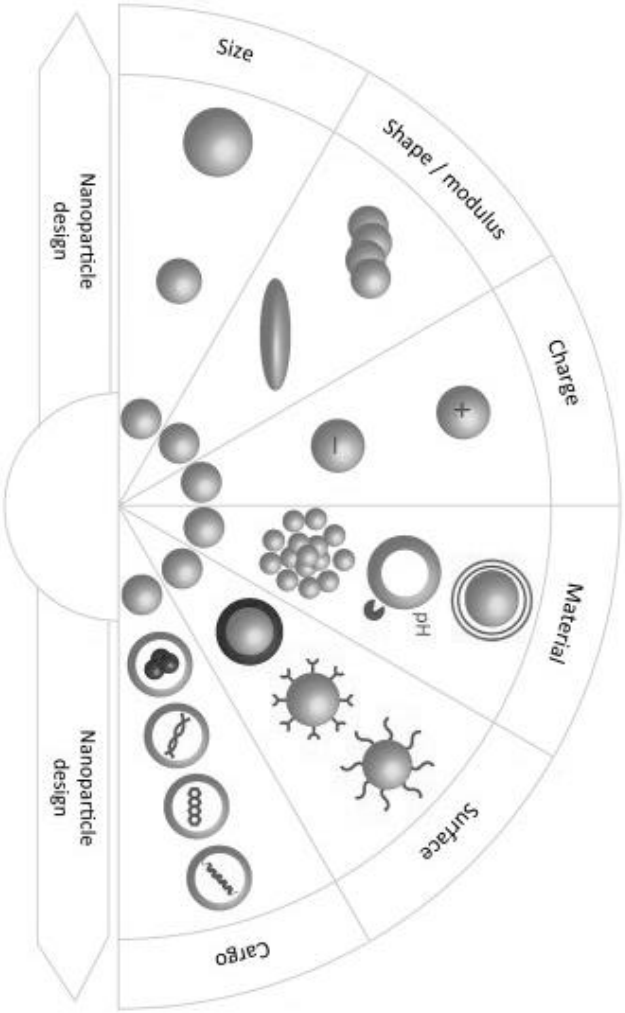
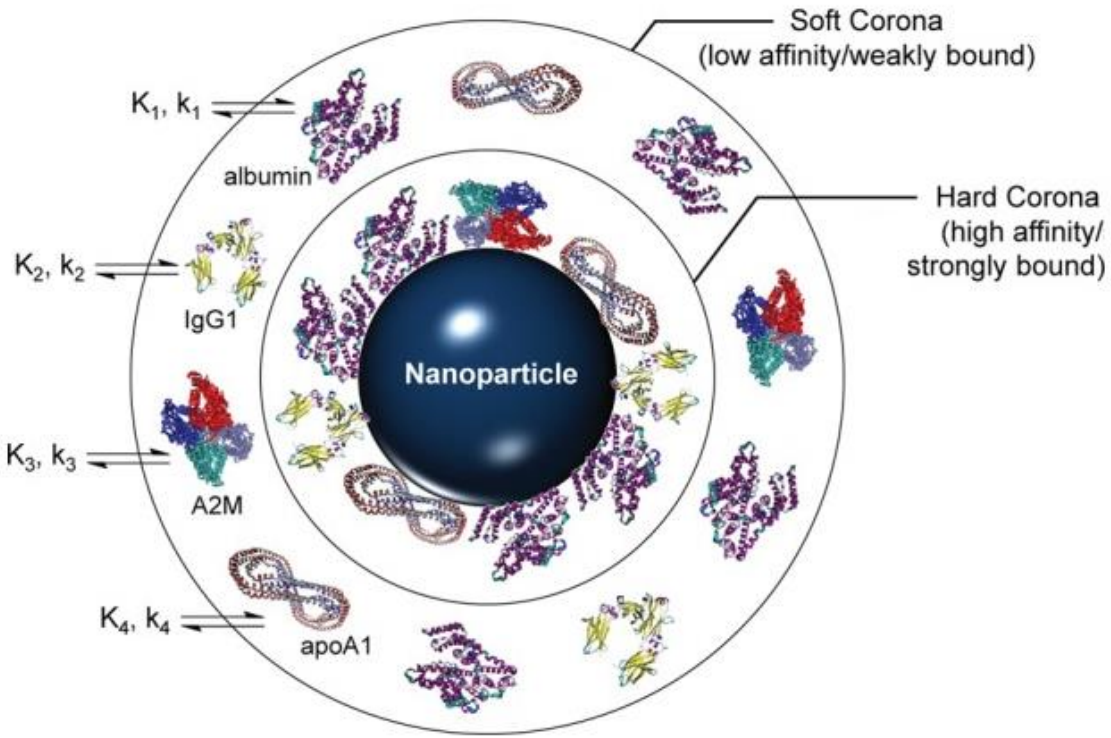
In vitro — Synthetic identity of NPs —(Exposure to biological fluids)→ PC formation → definition of a biological identity of NPs — **In vivo**





Protein corona factors

Nanoparticle	Environment
Material	Composition
Surface	Exposure time
Size	pH
Charge	Temperature
Shape	Shear stress

S. Hauert, S. N. Bhatia. Trends in Biotechnology, 2014

C. C. Fleischer, C. K. Payne. Acc. Chem. Res, 2014.

B. J. L. Van Hong Nguyen. International journal of nanomedicine, 2017

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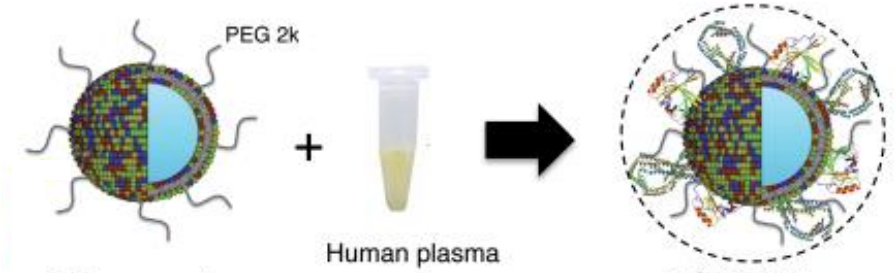
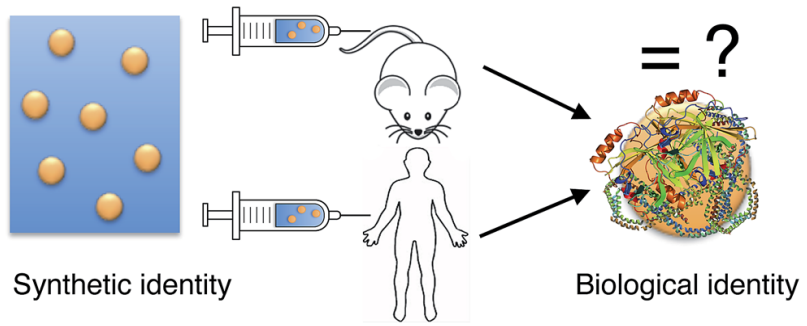
Factors shaping the nanoparticle-protein corona

PEGYLATION



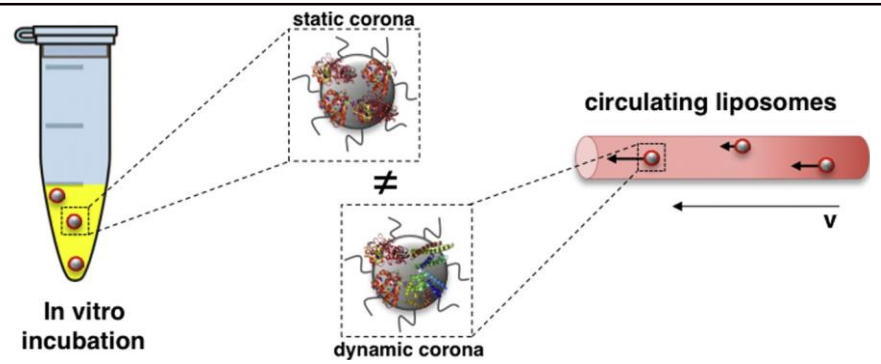
D. Pozzi et al. Nanoscale, 2014

PROTEIN SOURCE



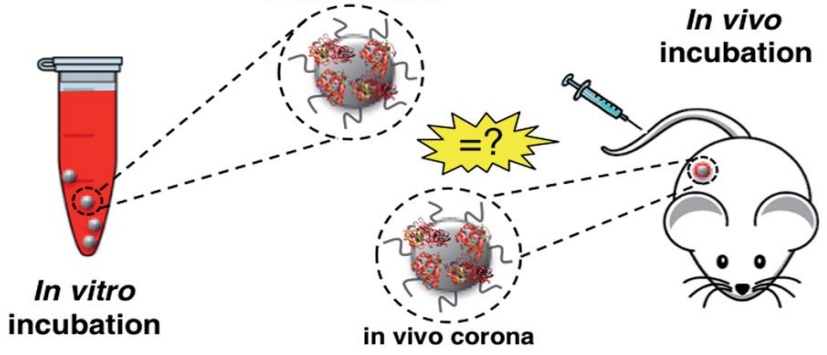
G. Caracciolo et al. Journal of Materials Chemistry B, 2014

SHEAR STRESS



S. Palchetti et al. BBA Biomembranes, 2016

IN VIVO INCUBATION



A. Amici et al. RSC Advances, 2017

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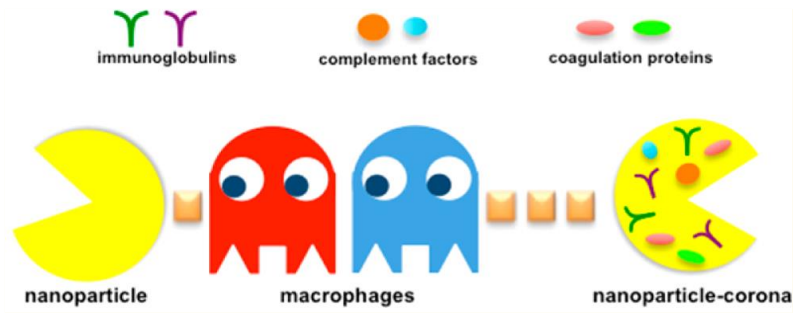
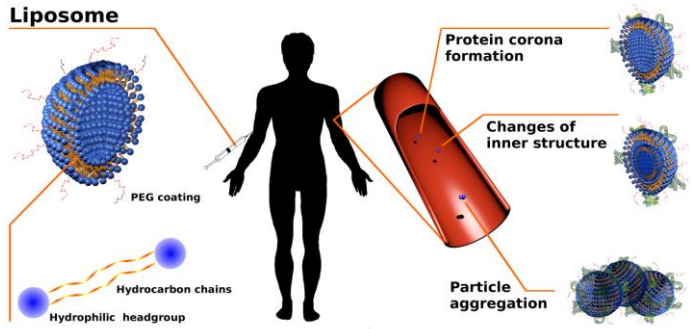
Biological effects of the nanoparticle-protein corona

STEALTH EFFECT ON PARTICLE UPTAKE BY IMMUNE CELLS



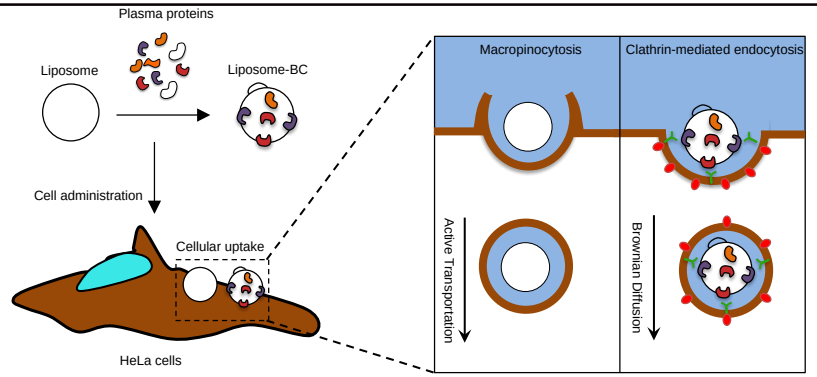
G. Caracciolo et al. Langmuir 2015

PARTICLE AGGREGATION IN VIVO



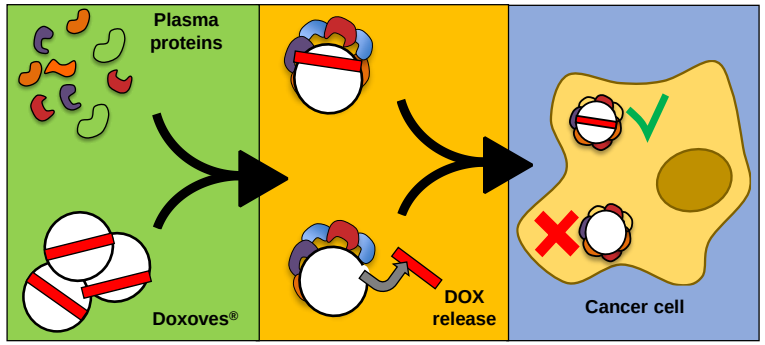
L Digiacomo et al. Biomaterial Sciences, 2017

CELLULAR UPTAKE AND INTRACELLULAR FATE



L. Digiacomo et al. Nanoscale 2017

DRUG/GENE RELEASE IN VIVO



G. Caracciolo et al. ACS Applied Materials & Interfaces, 2018

2. Personalized Protein Corona

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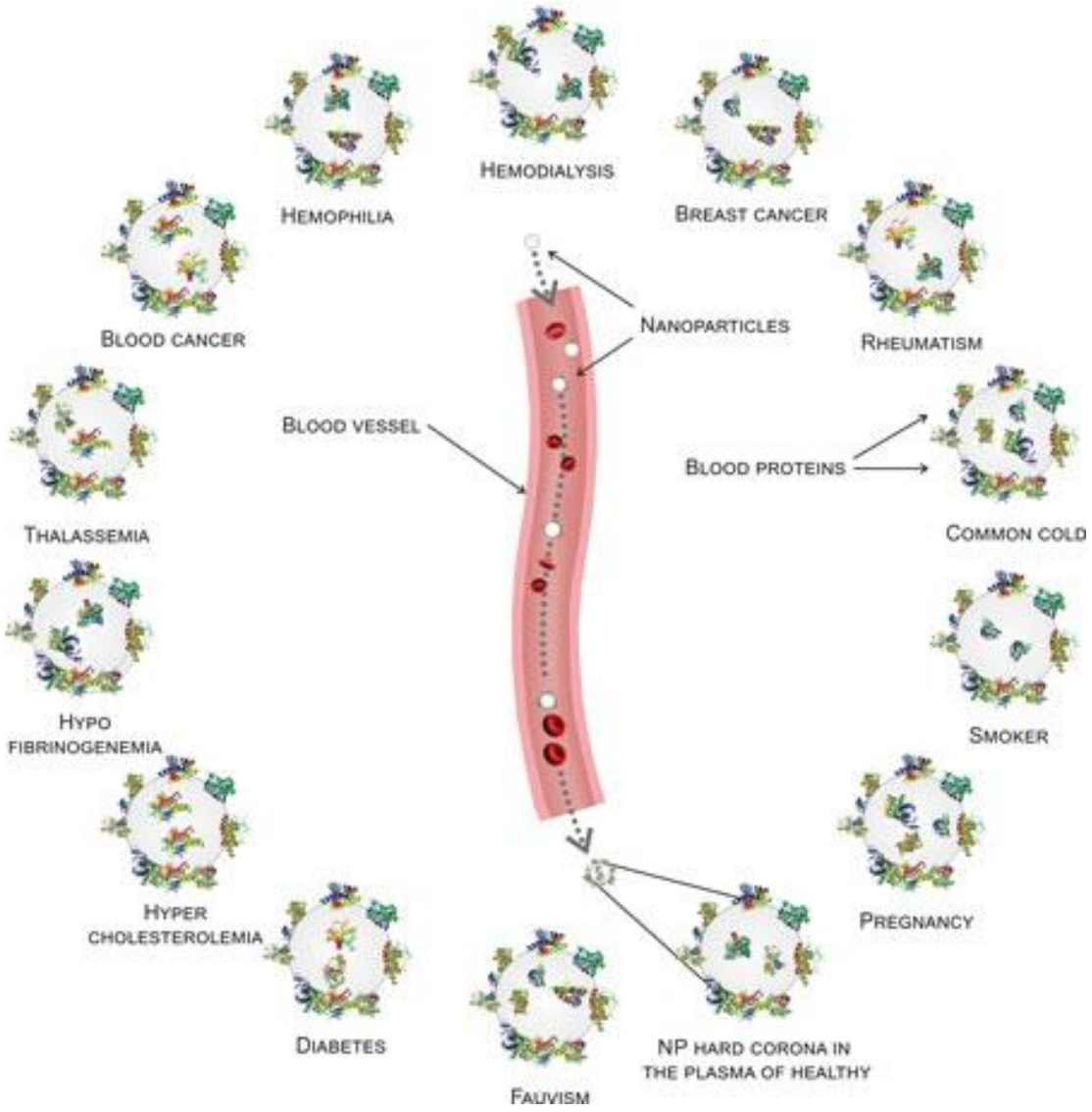
PCs from different subjects are different

NPs were introduced into human plasma taken from subjects exhibiting a range of physical states (including cancer, diabetes, hypercholesterolemia and pregnancy).

The resulting PCs were altered by disease and also by disease severity.

M. J. Hajipour et al. Biomaterials Science, 2014

The personalized protein corona is a determinant factor in nano-biomedical science.



M. J. Hajipour et al. Biomaterials Science, 2014

2. Personalized Protein Corona

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PCs from different protein sources are vastly different

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M. J. Hajipour et al. Biomaterials Science. 2014

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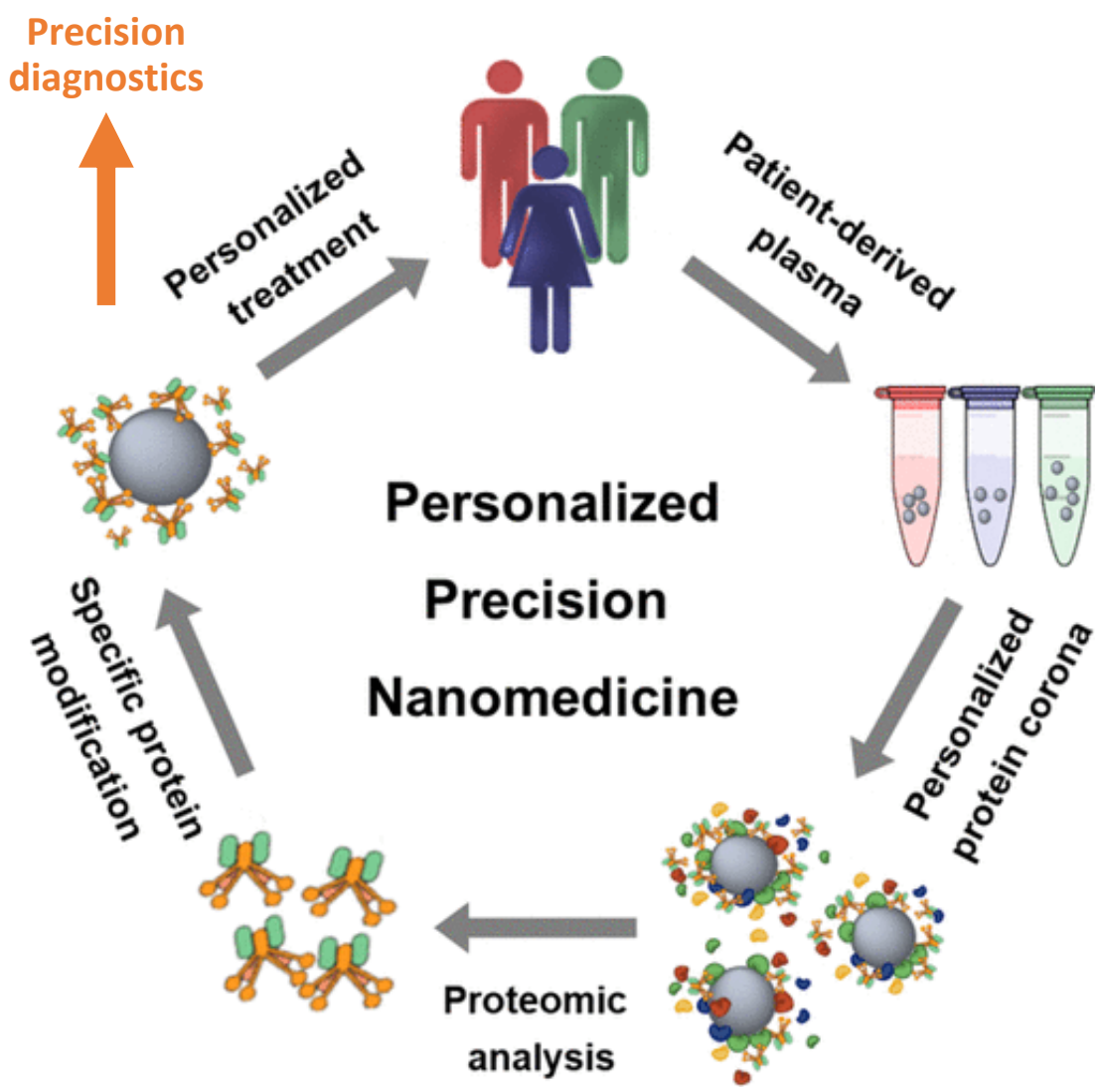

J. Ren et al. Nano Letters, 2019.

2. Personalized Protein Corona

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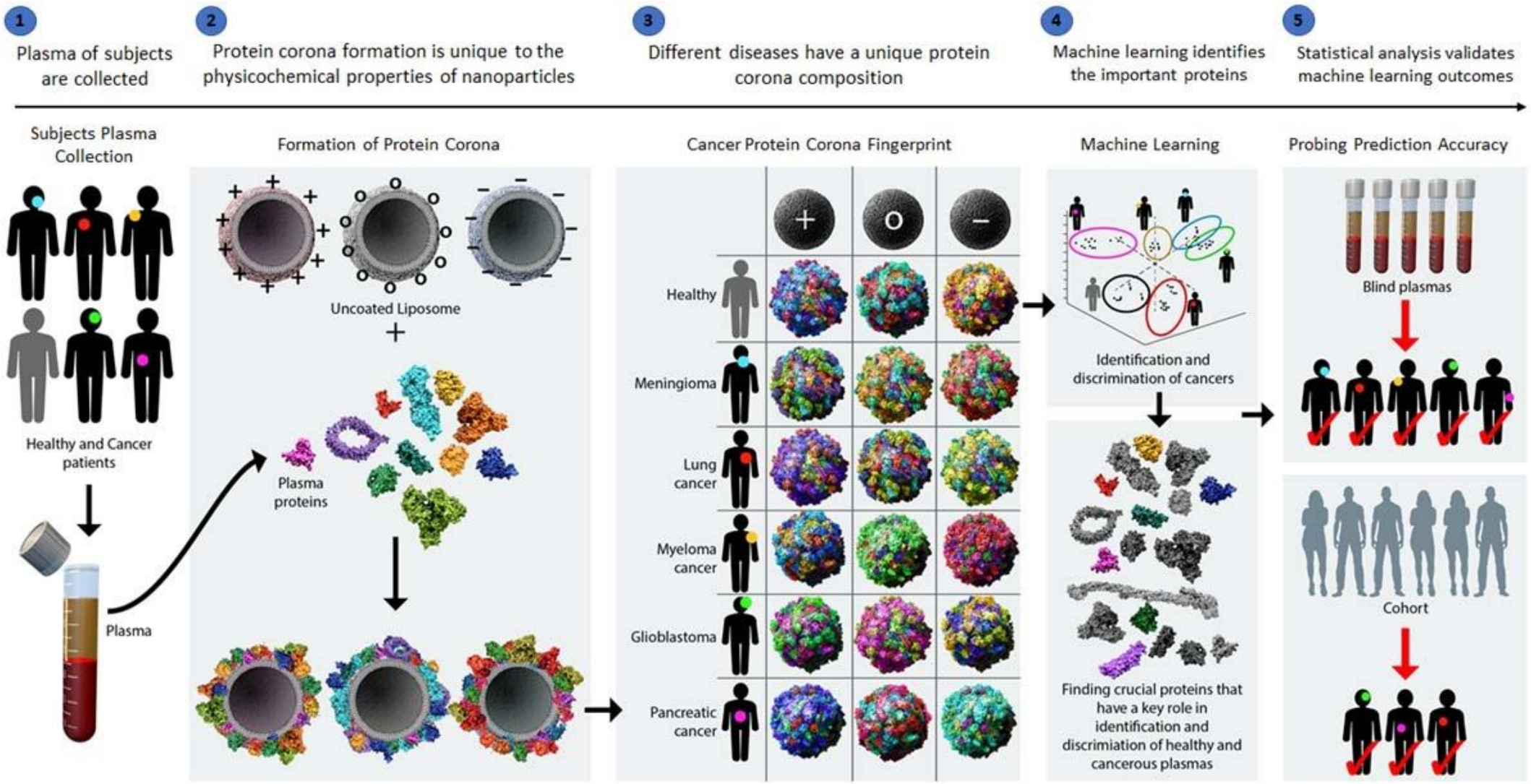
Exploiting the nanoparticle-protein corona for cancer biomarker discovery



G. Caracciolo et al. Nanoscale Horizon, 2019

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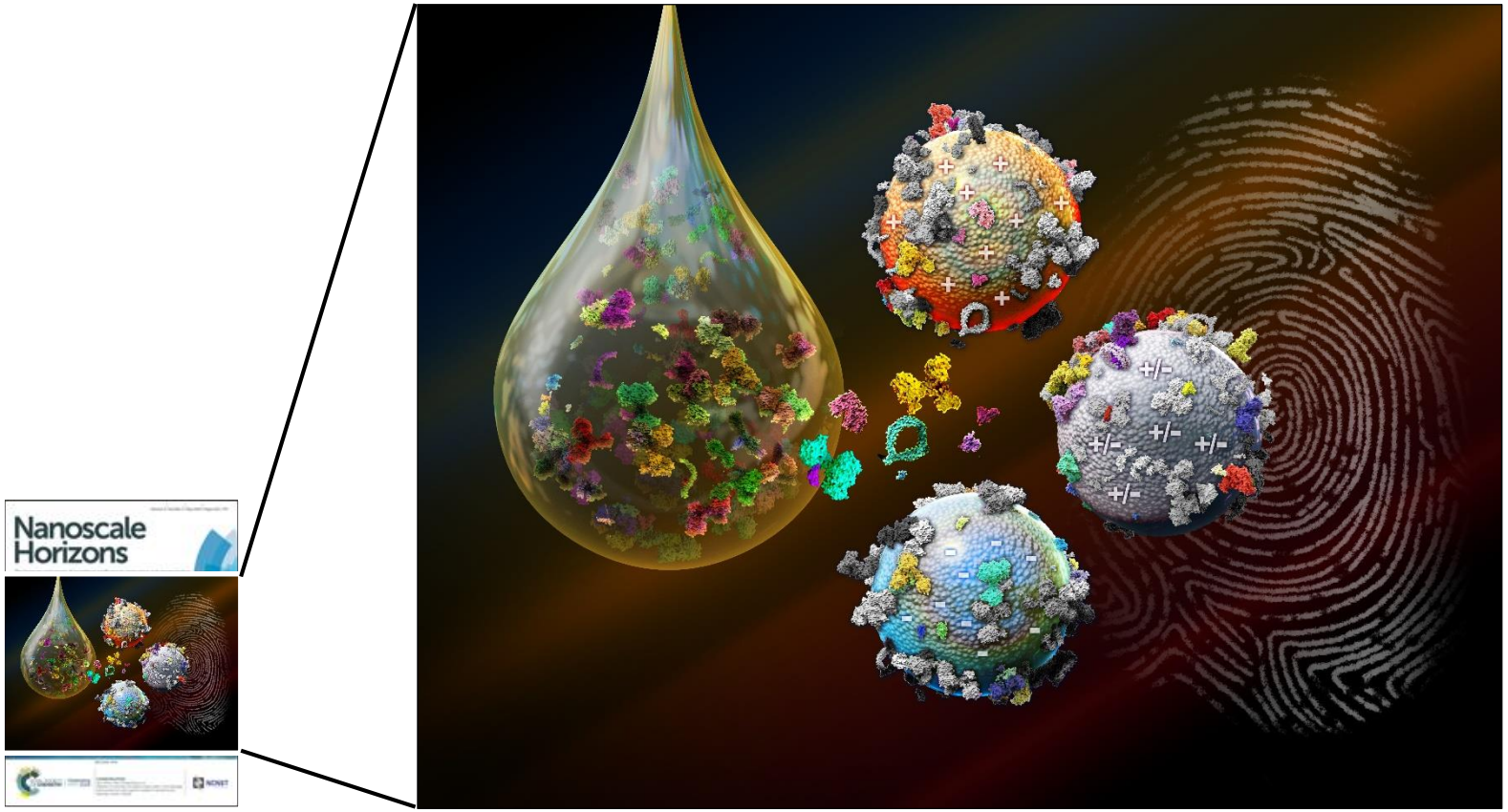

PC sensor arrays have disease detection capacity, are **potentially disrupting** but **expensive** and time consuming.

They do not fit yet the general guidelines of World Health Organization for cancer screening procedures.

G. Caracciolo et al. Nanoscale Horizon, 2019

3. Nanoparticle-enabled blood test

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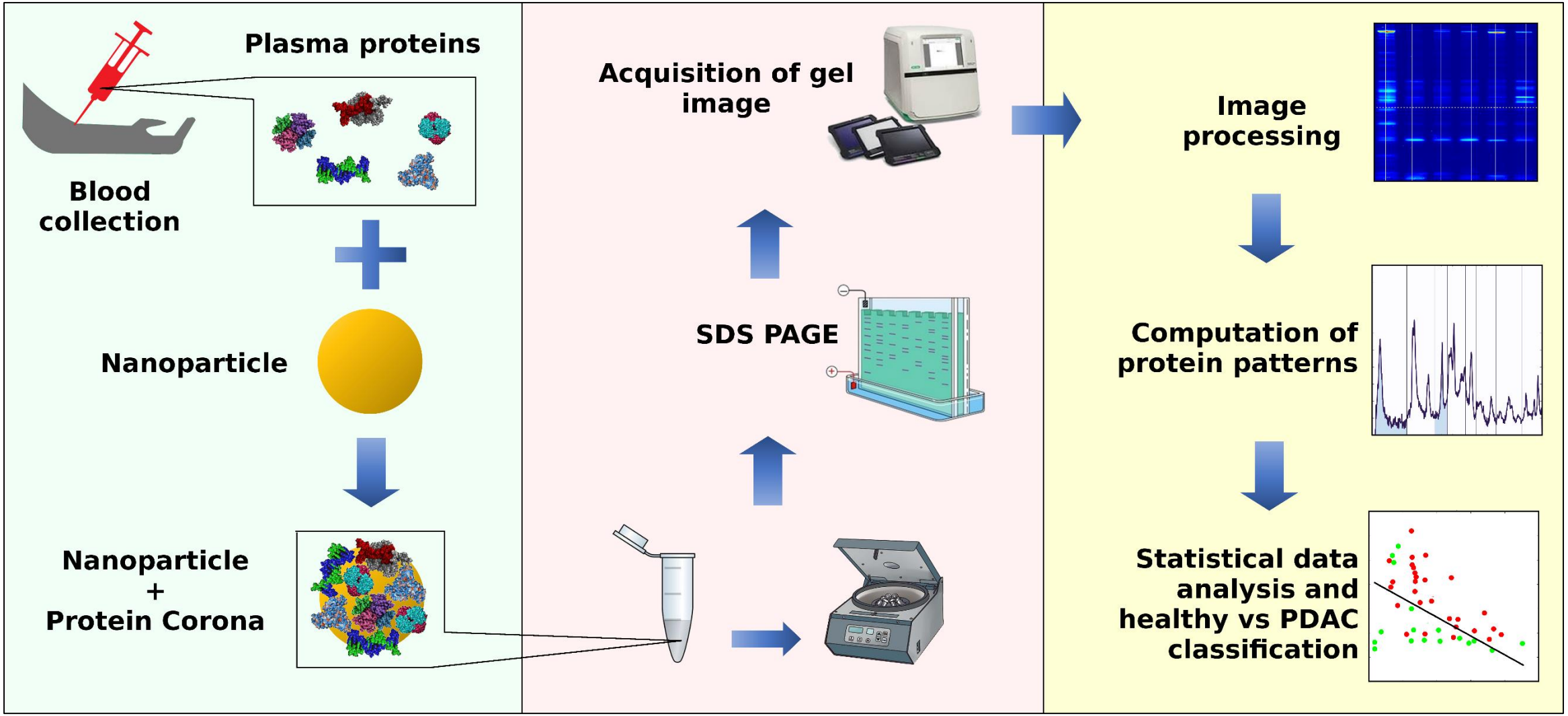

Sample preparation | PC isolation | Data analysis

4. Pancreatic Ductal AdenoCarcinoma (PDAC)

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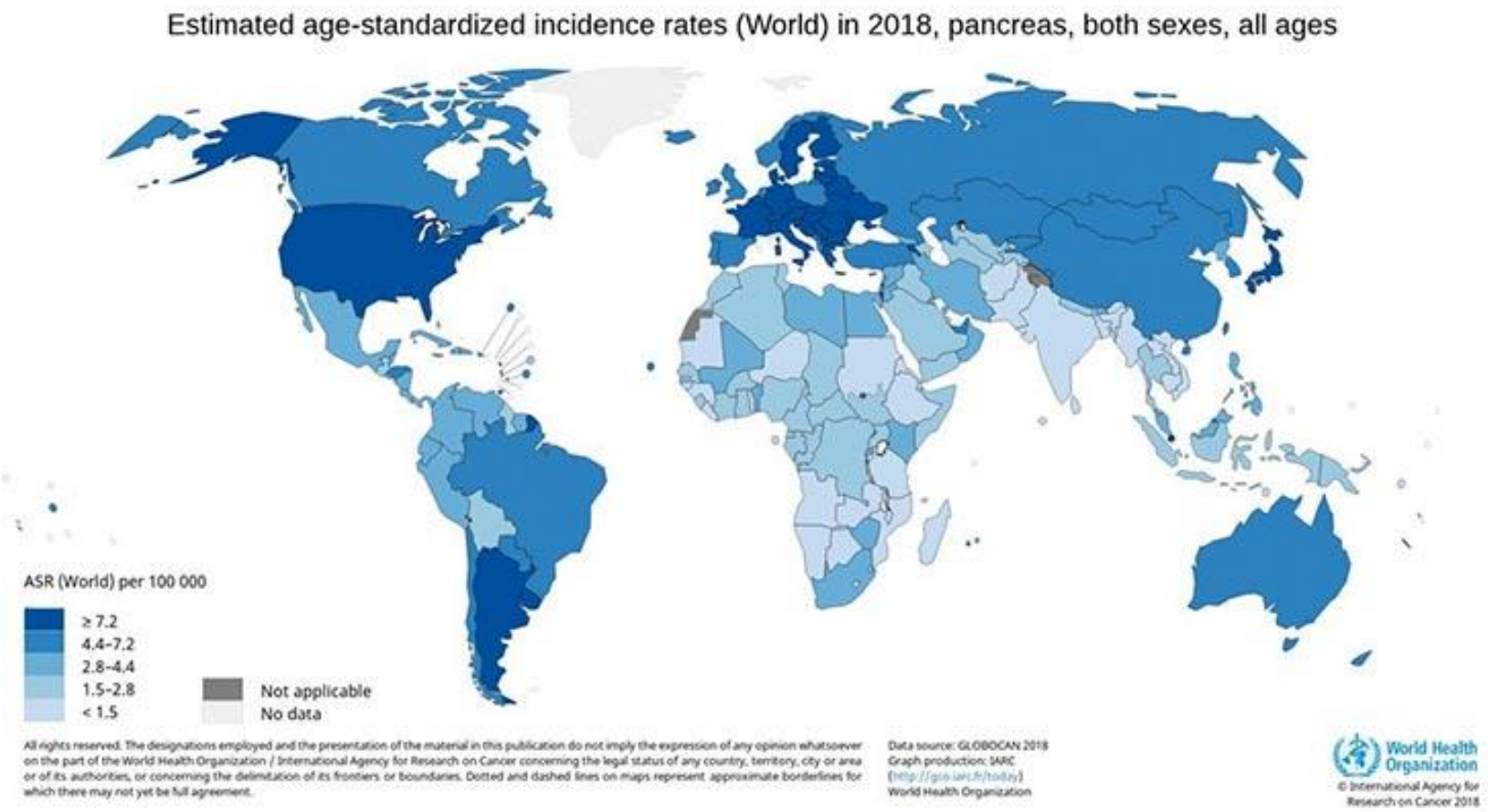
PDAC is the most common malignancy of pancreas,
it is particularly aggressive and difficult to treat.



- Dismal prognosis of PDAC is related to the advanced stage of the disease at the moment of the diagnosis.
- To date, a non-biopsy-based-test (CA19-9) is routinely used but fails to be specific and is poorly sensitive (71%).
- Therefore, **novel diagnostic tools for the early detection of PDAC** are urgently needed.

Pancreatic Cancer Action Network (PanCAN), 2018

5. Results

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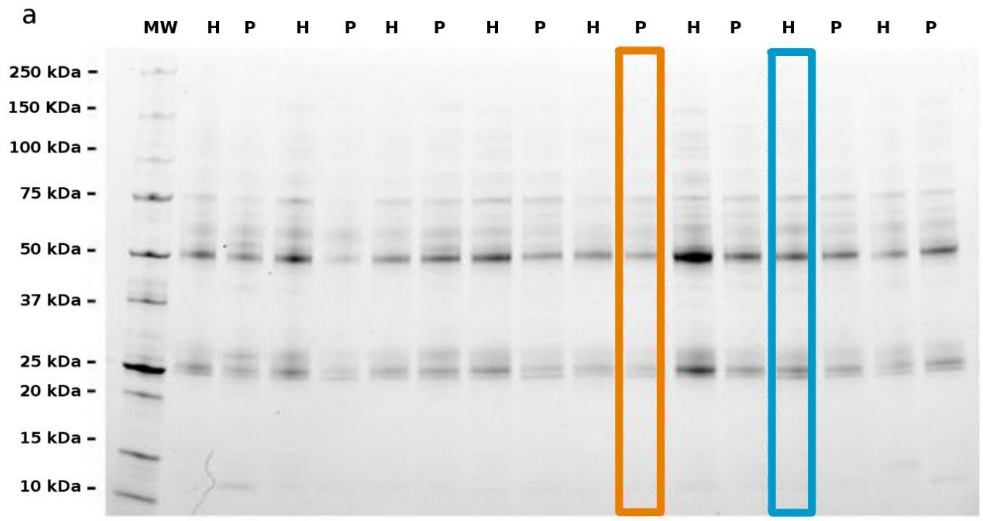
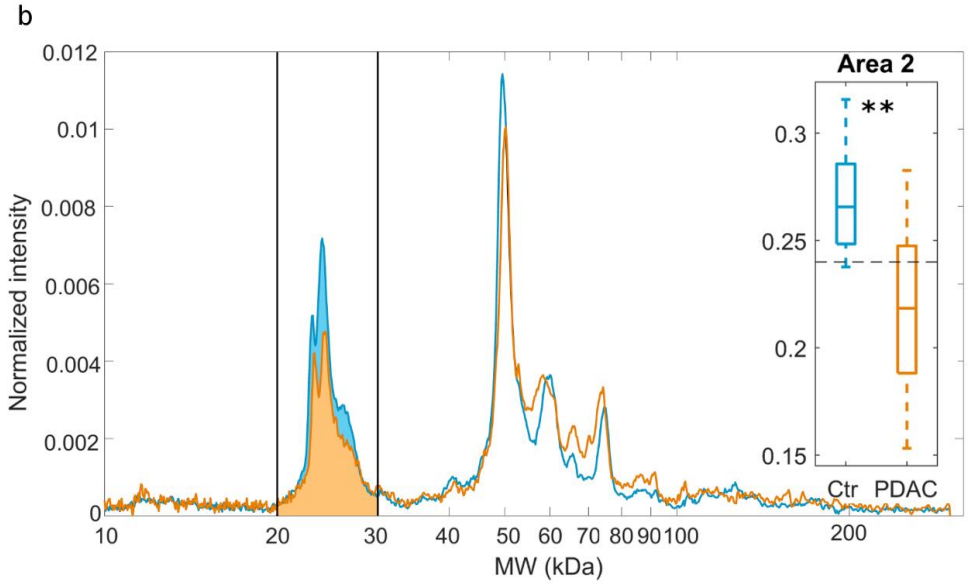
Graphene Oxide (GO) + Human Plasma (HP) from PDAC and non-oncological subjects





- GO. Lateral size = 500 nm; Zeta Potential = -30 mV; concentration = 1 mg/mL.
- HP from 25 PDAC and 25 non-oncological subjects.
- 1h incubation, PC isolation and evaluation of protein patterns by 1D SDS PAGE experiments

Final outcomes

Specificity: 84.6%

Sensitivity: 84.2%

Overall correctness: 84.4%

5. Results

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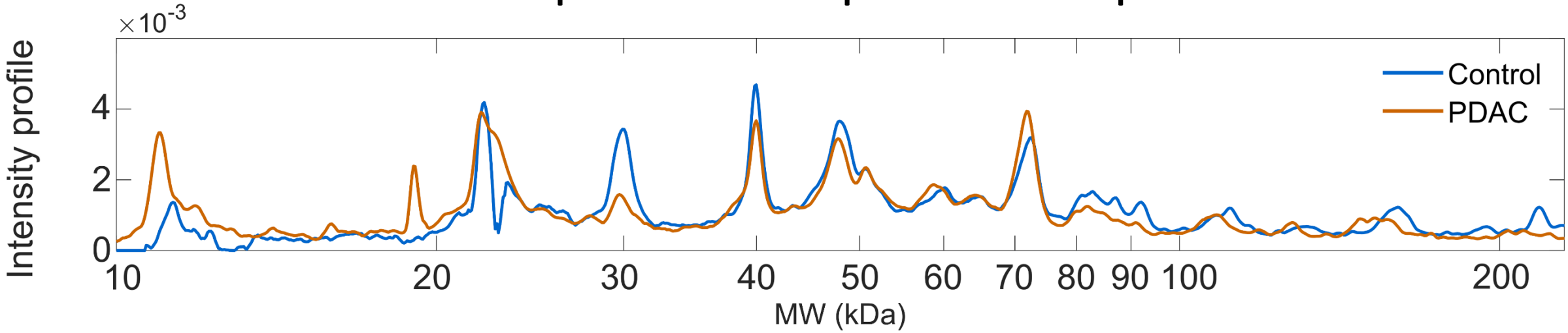
Gold NPs + Human Plasma (HP) from PDAC and non-oncological subjects

- Au NPs. Size = 120 nm; Zeta Potential = -21 mV; concentration = 4 10^9 particles/mL.
- HP from 49 PDAC and 28 non-oncological subjects.
- 1h incubation, PC isolation and evaluation of protein patterns by 1D SDS PAGE experiments

1D patterns of two representative samples



5. Results

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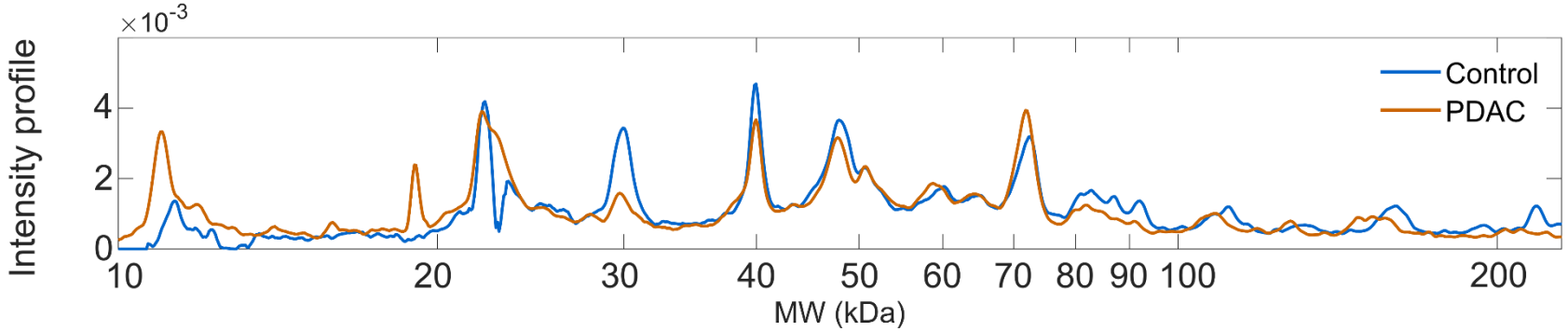
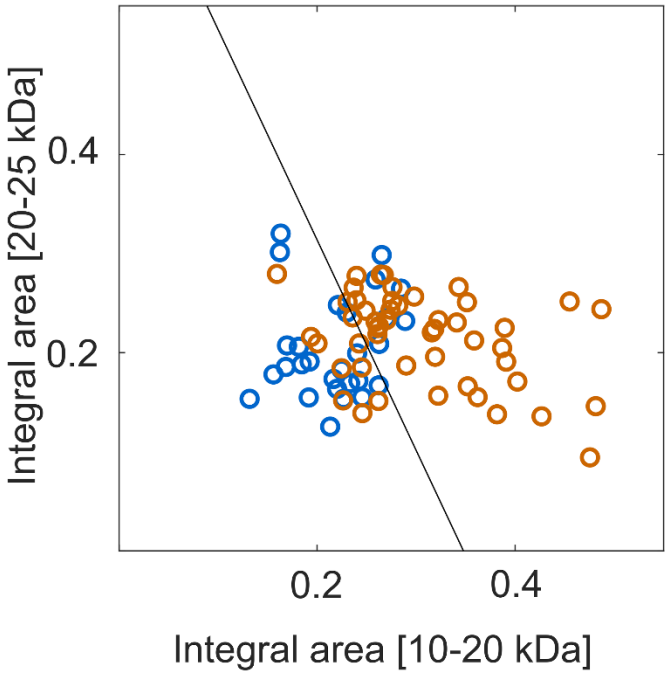
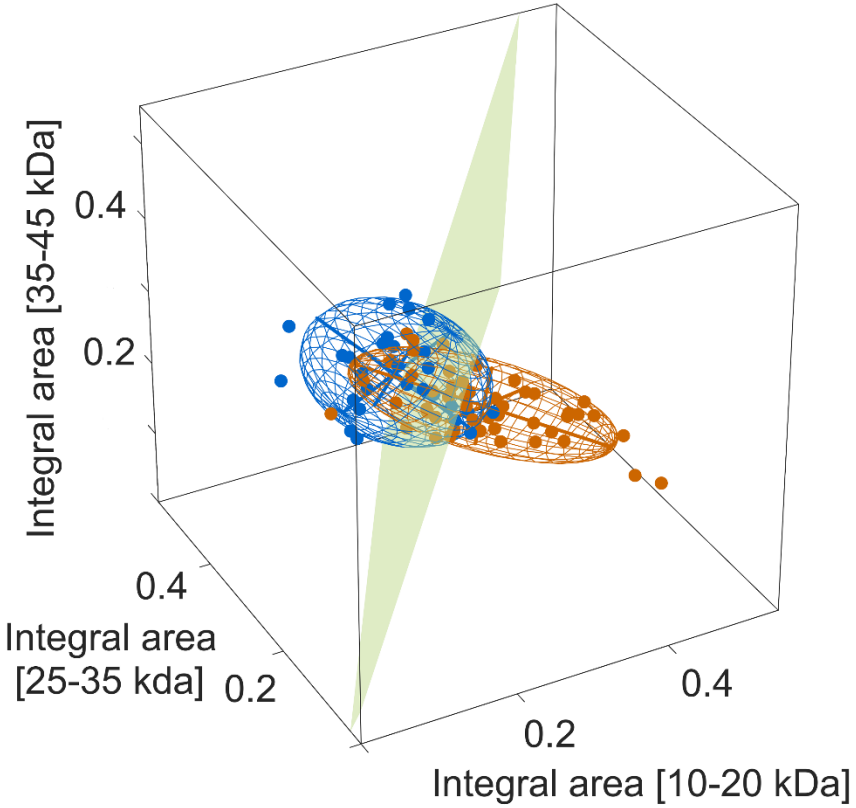
Gold NPs + Human Plasma (HP) from PDAC and non-oncological subjects







Final outcomes

Specificity: 78.6%

Sensitivity: 83.7%

Overall correctness: 81.8%

5. Results

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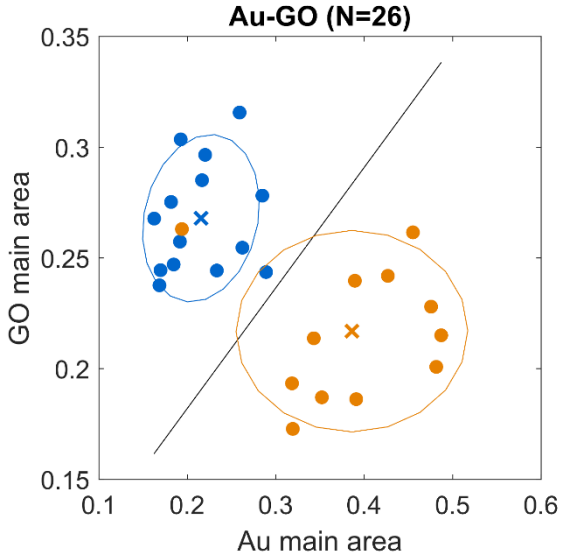
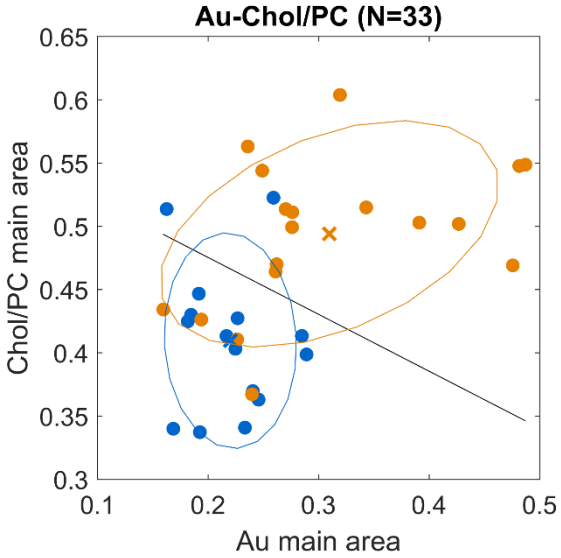
Library of NPs + Human Plasma (HP) from PDAC and non-oncological subjects

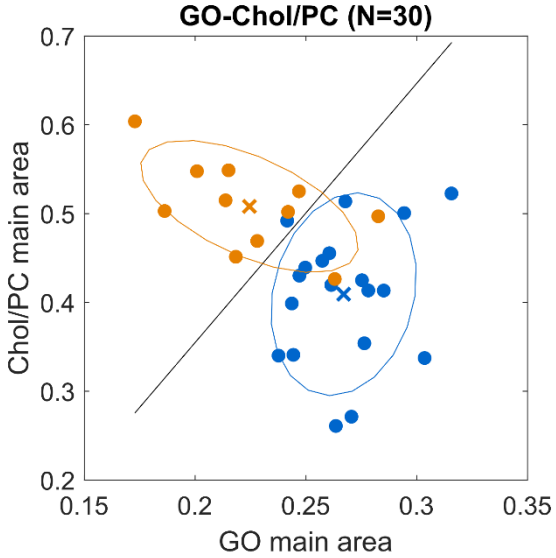
Outcomes from single-NP electrophoretic analysis

Material	DOTAP/DOPE	Ag nanoparticles	CHOL/PC	Au nanoparticles	Graphene Oxide
Number of samples	51	99	48	77	50
Specificity	80.0%	64.1%	80.0%	78.6%	84.6%
Sensitivity	52.0%	83.3%	82.6%	83.7%	84.2%
Global correctness	**66.0%**	**75.8%**	**81.3%**	**81.8%**	**84.4%**

Outcomes for multiplatform-enabled blood test







Coupling	Au-GO	Au-Chol/PC	GO-Chol/PC
Number of samples	26	33	30
Specificity	100%	86.6%	96.5%
Sensitivity	91.7%	77.8%	81.8%
Global correctness	**96.0%**	**81.8%**	**90.0%**

6. Conclusions and perspectives

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We exploited the protein corona of nanoparticles to design a safe, accurate and non-invasive tool for pancreatic cancer detection.

The test is aligned to the **ASSURED** (Affordable, Sensitive, Specific, User-friendly, Rapid and robust, Equipment-free and Deliverable to end-users) **criteria** stated by the World Health Organization for cancer screening and detection

Further studies will aim

- to distinguish the **cancer's stage** by the protein corona technology
- to couple the information arising by multiple parameters (e.g. inflammatory response elements and other clinical markers) from different techniques for the development of **multiplexed point-of-care testing systems.**

Acknowledgments

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Valentina Palmieri
Giordano Perini

University Campus-Biomedico di Roma

Damiano Caputo
Roberto Coppola
Chiara Cascone

AIRC

Thank you