

# Anti-PEG Antibodies - Biology, Assays, and Significance

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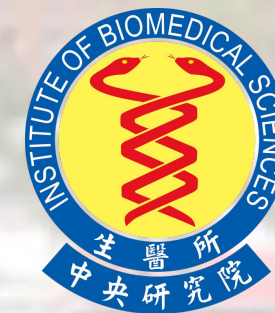
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10 July 2024



JULY 8-12, 2024 • BOLOGNA, ITALY



# The magical world of polyethylene glycol antibodies



Assay PEG drugs

Accelerate drug  
development



Pre-existing Abs

Accelerate clearance

SARS-CoV-2 mRNA  
vaccines

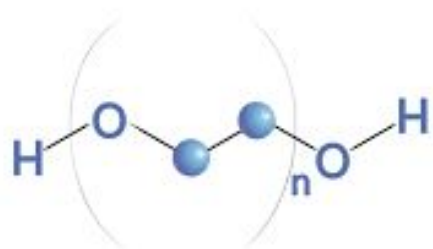


Lipid NP destabilization

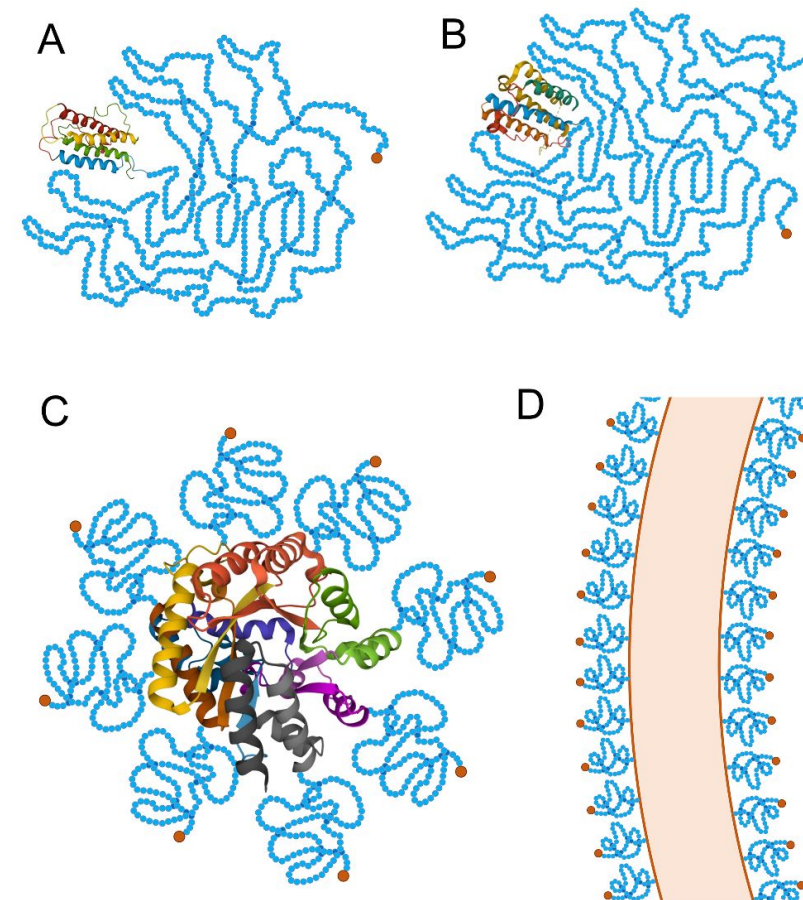
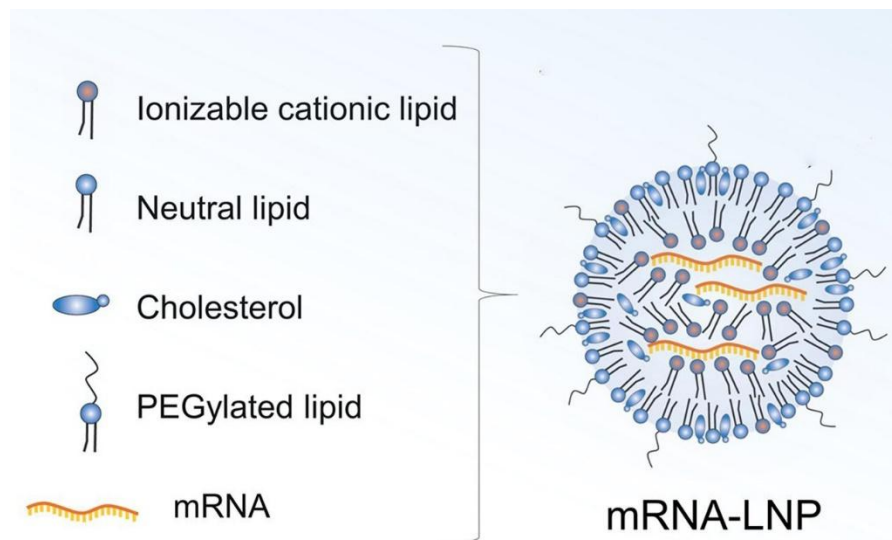
Hypersensitivity



Targeting nanomedicines

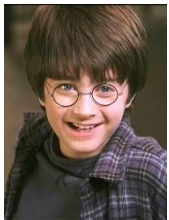
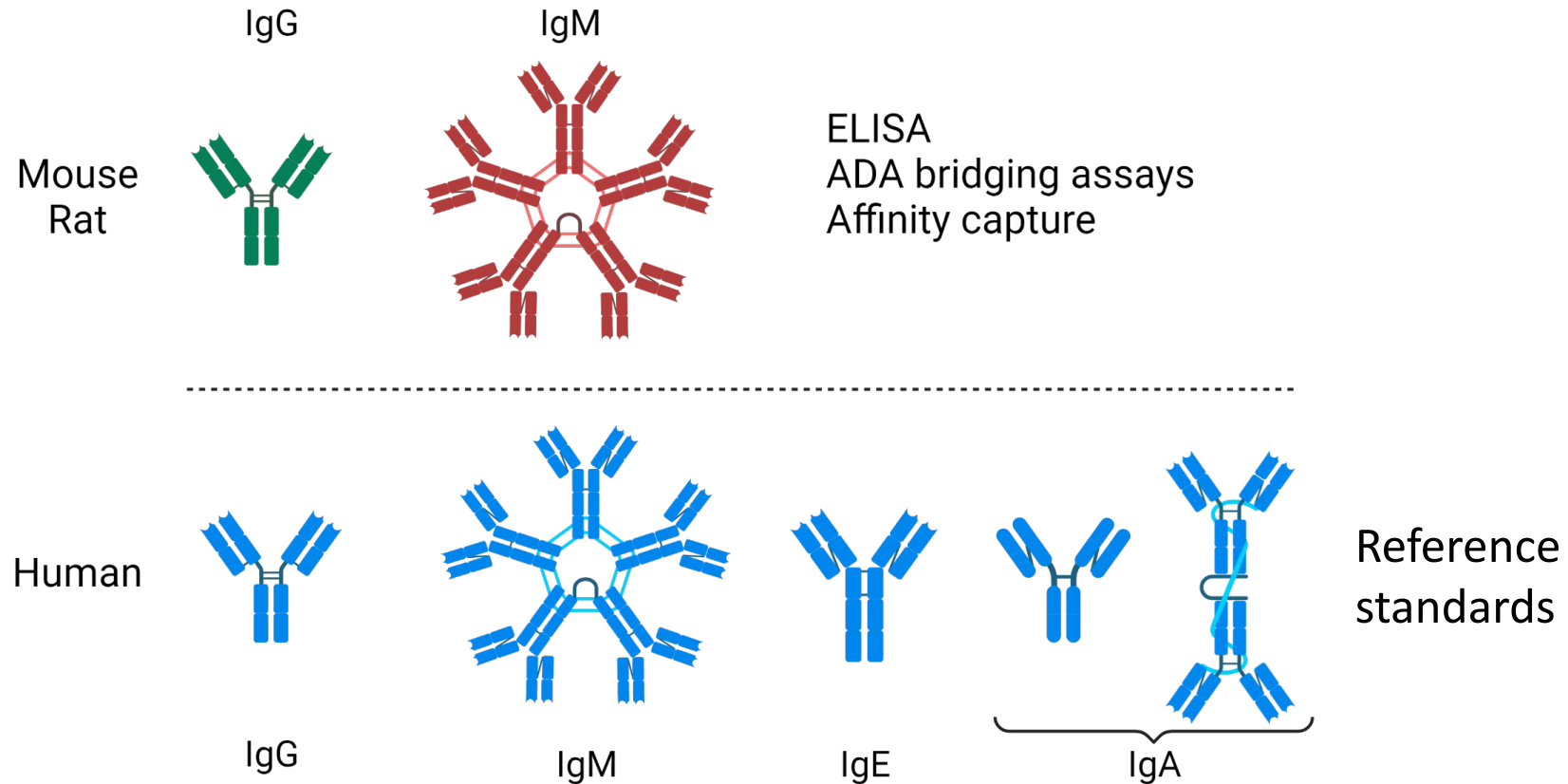


Polyethylene glycol



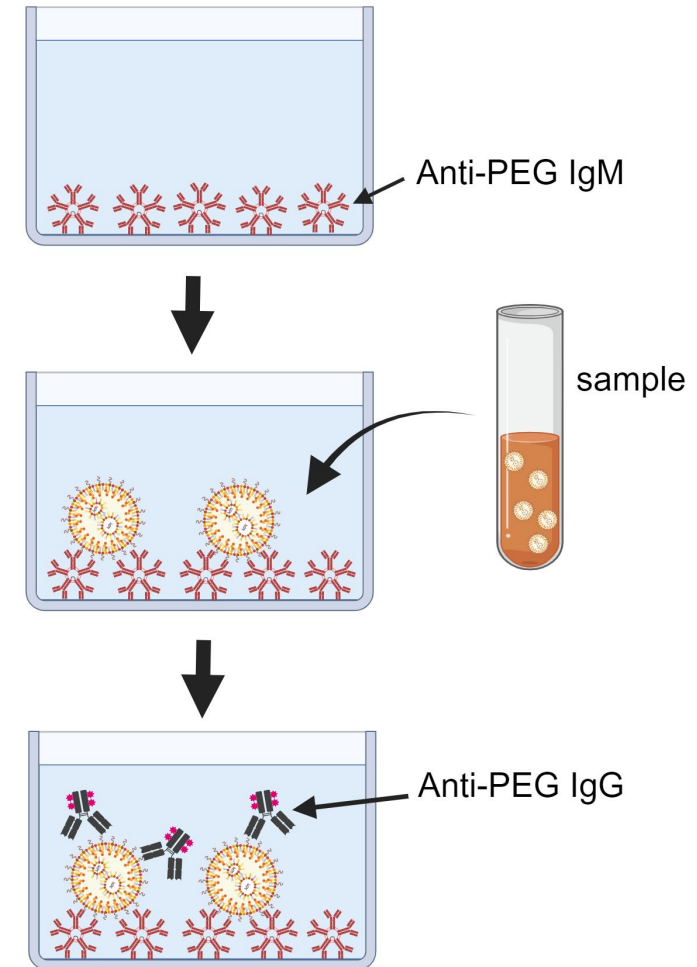
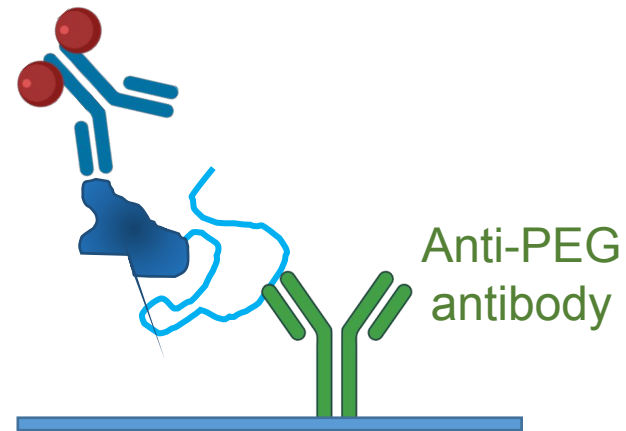
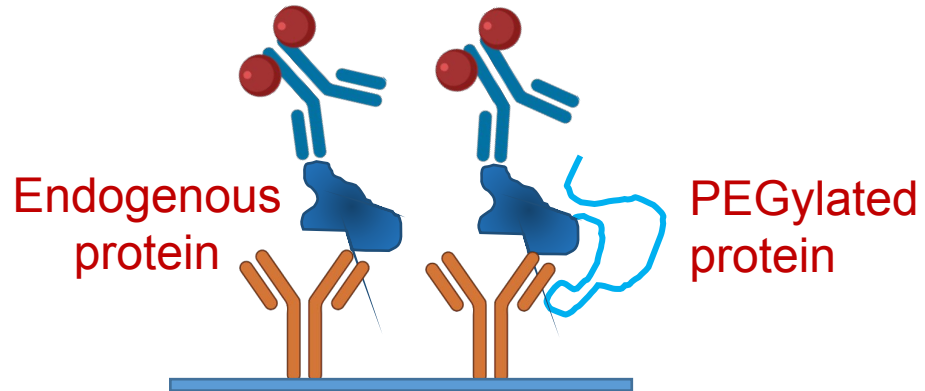


<https://www.ibms.sinica.edu.tw/~sroff/anti-PEG/index.html>



Cheng *et al.*, Bioconj. Chem., 10: 520-528 (1999)

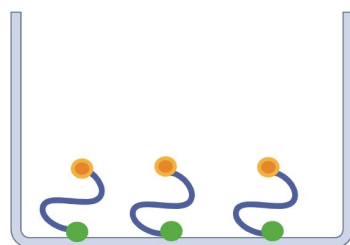
# Assay of PEGylated drugs



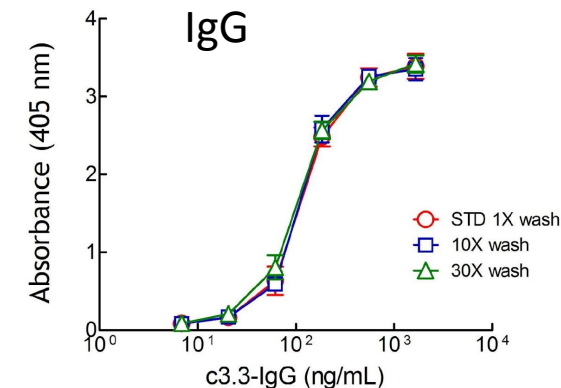
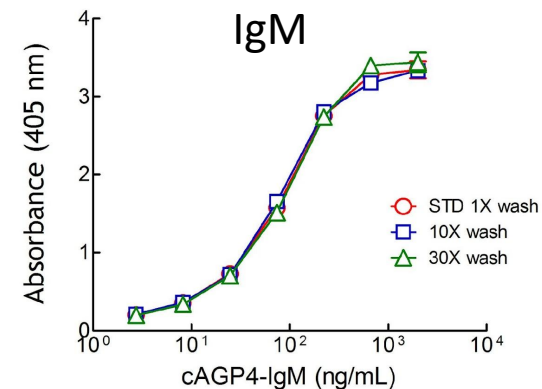
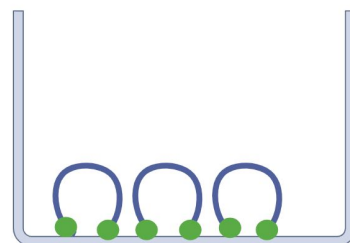


# Key points of anti-PEG assay

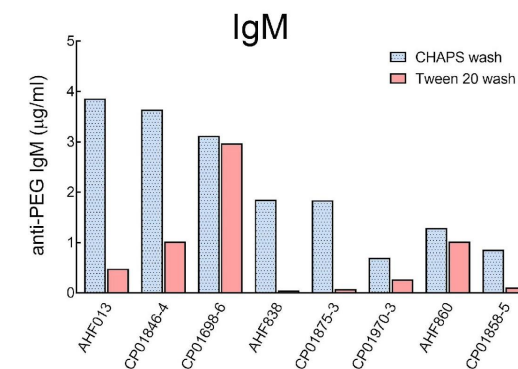
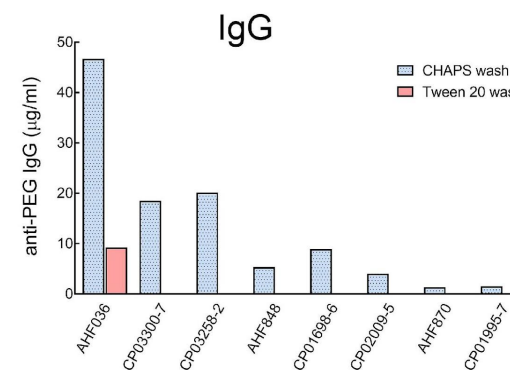
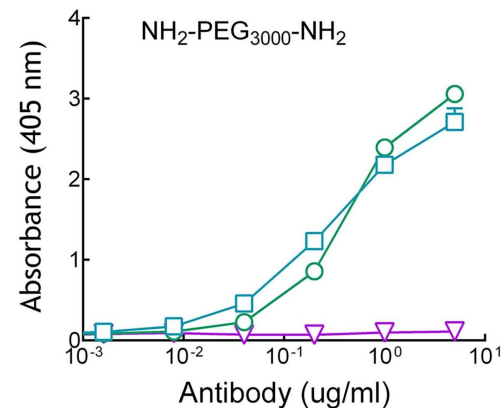
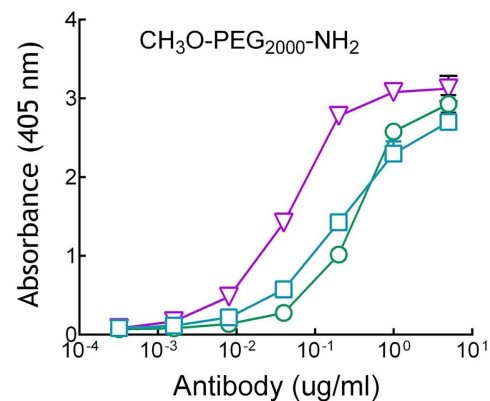
(backbone  
+ methoxy)



(backbone)



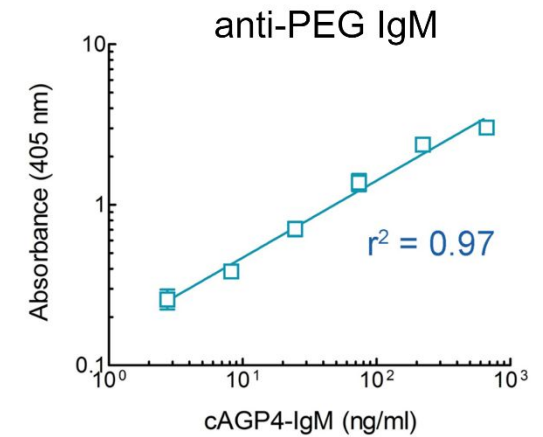
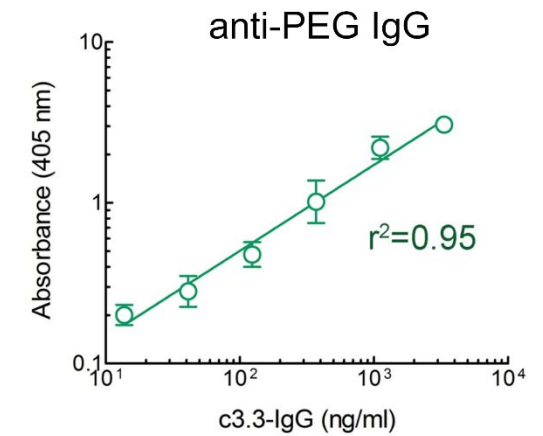
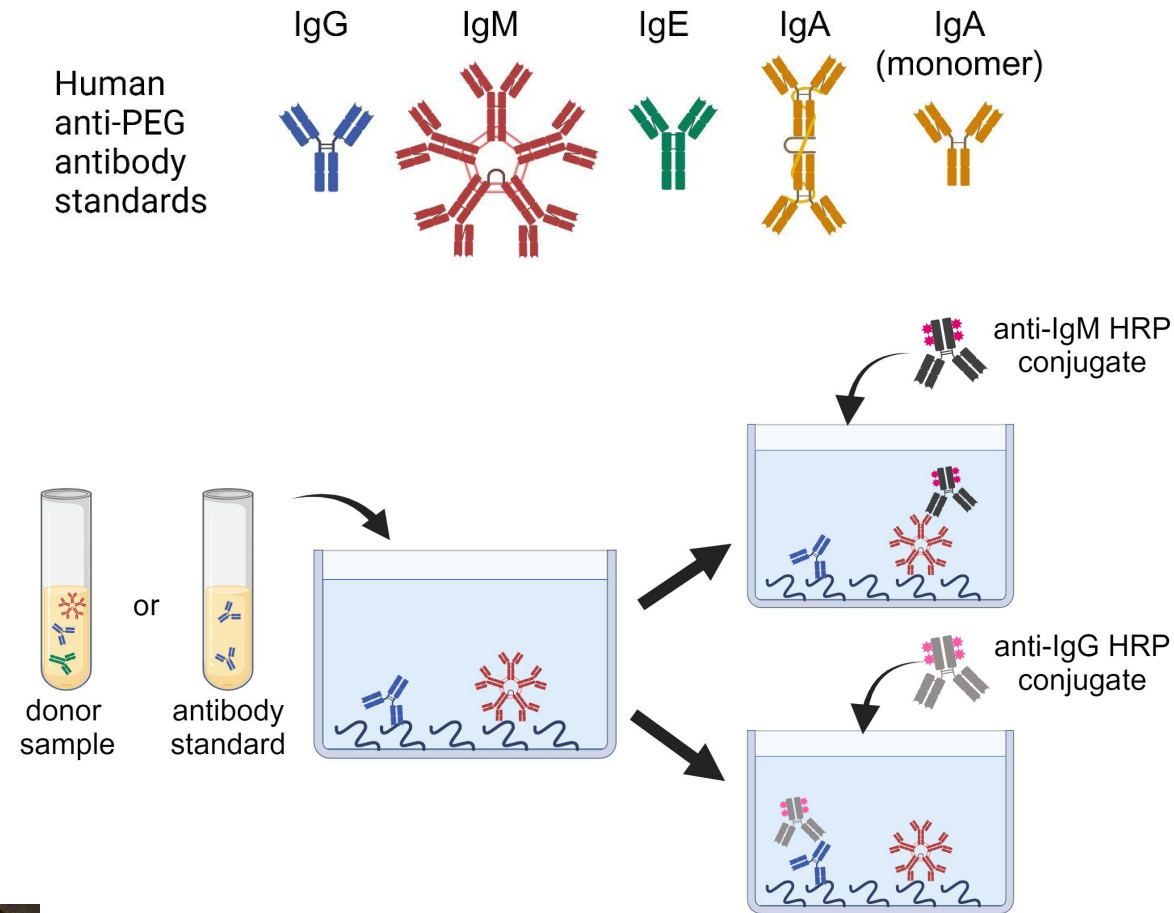
▽ 15-2b (anti-mPEG IgG)  
○ 3.3 (anti-PEG IgG)  
□ AGP4 (anti-PEG IgM)



Chen *et al.*, *Analytical Chem.* 88:10661–10666 (2016)



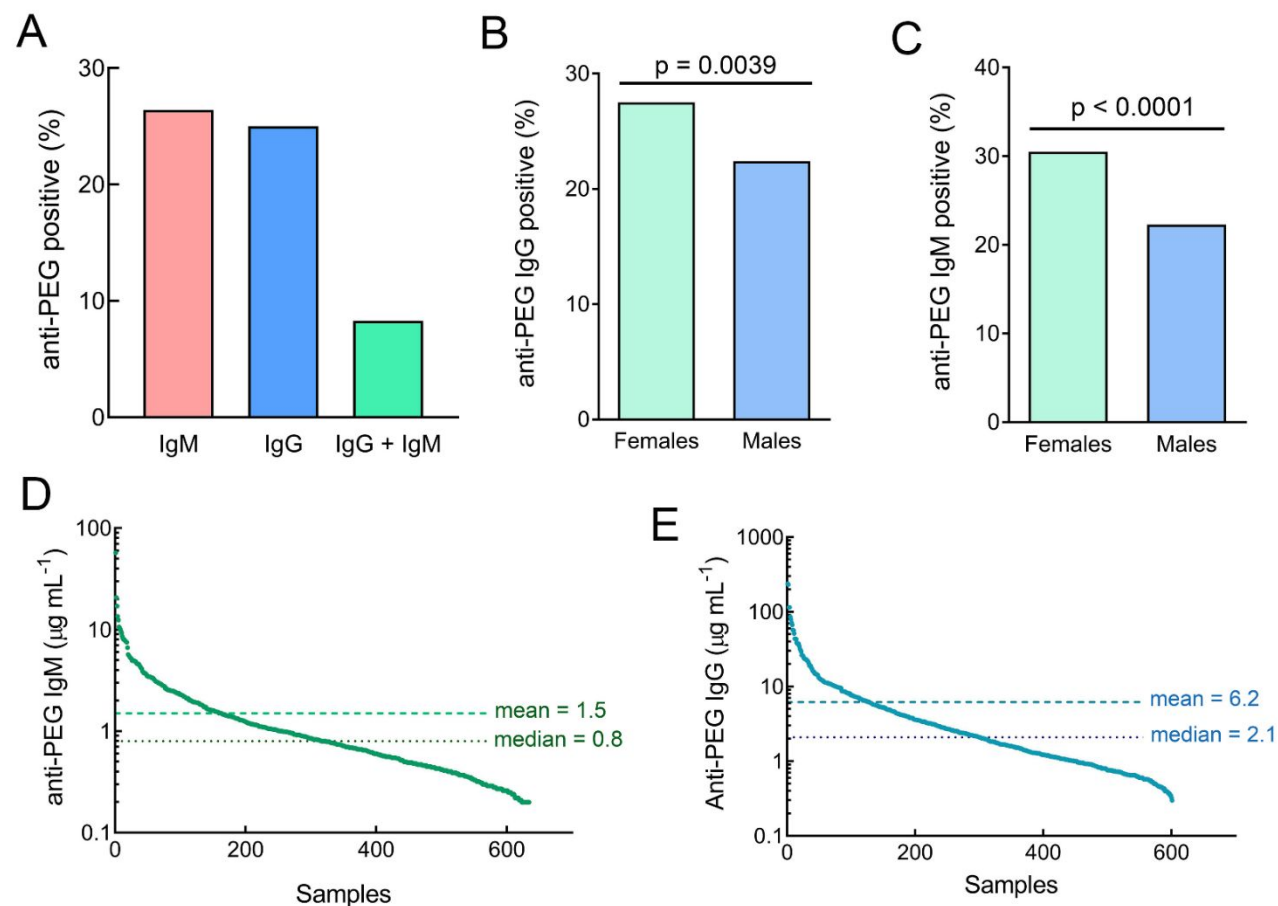
# Anti-PEG antibody assay (ADA)



Standardize assay results between labs



N=2404 normal  
donors from Taiwan



Prescreen serum

Chang *et al.*, *Nat Commun.* 8:522 (2017)  
Chen *et al.*, *Analytical Chem.* 88:10661–10666 (2016)  
Chen *et al.*, *ACS Nano* 15:14022–48 (2021)

# SARS-CoV-2 mRNA vaccines increase anti-PEG antibody titers

| Vaccine                | Anti-PEG IgG | Anti-PEG IgM |
|------------------------|--------------|--------------|
| BNT1622b2              | 1.8-fold     | 2.6-fold     |
| mRNA-1273<br>(Moderna) | 13-fold      | 69-fold      |



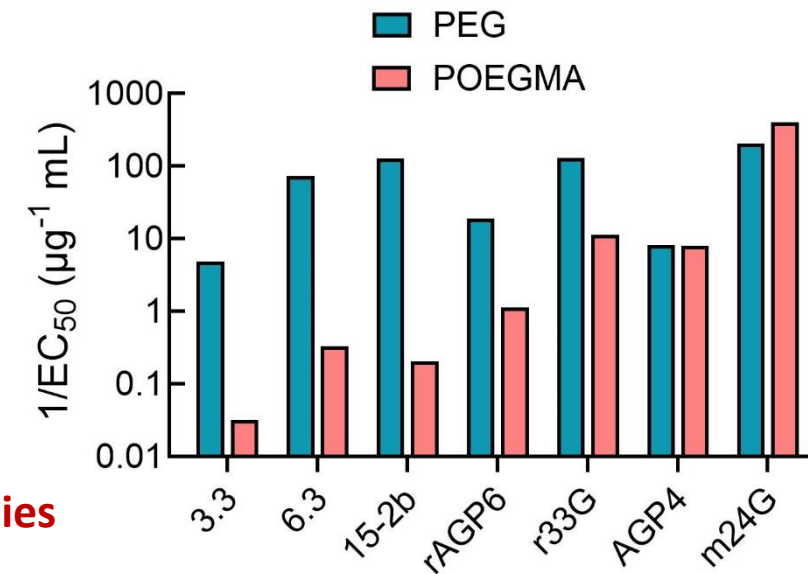
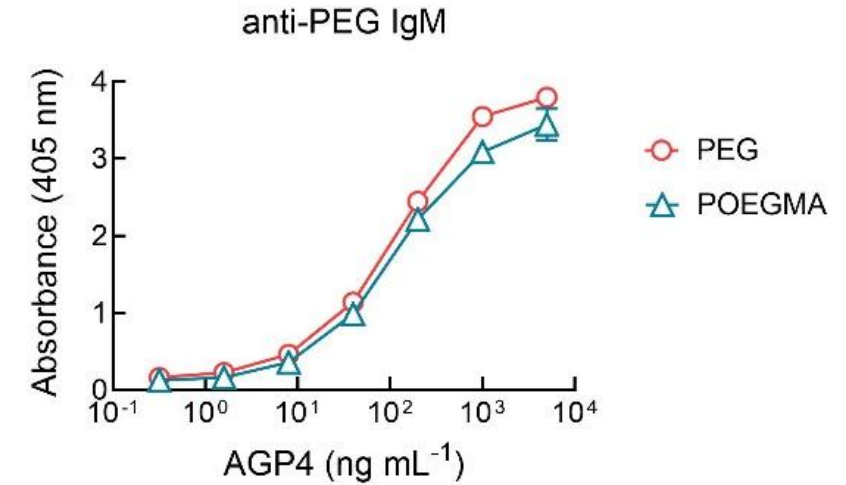
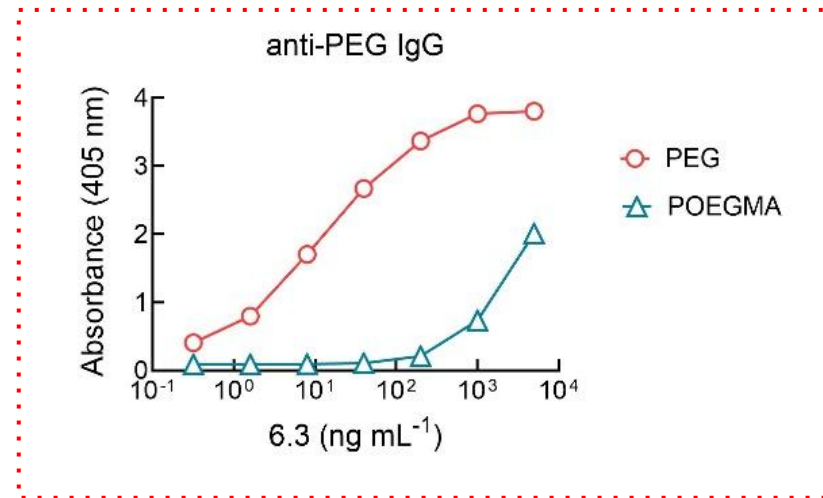
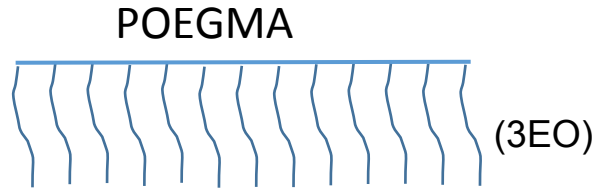
Ju et al., *ACS Nano* 2022, **16**: 11769-11780

Bavli et al., *J Control Release* 2023, **354**: 316-322

Guerrini et al., *Int J Mol Sci* 2022, **23**: 8838

Carreño et al., *Vaccine* 2022, **40**: 6114-6124

# POEGMA: Poly(oligo(ethylene glycol) methyl ether methacrylate)

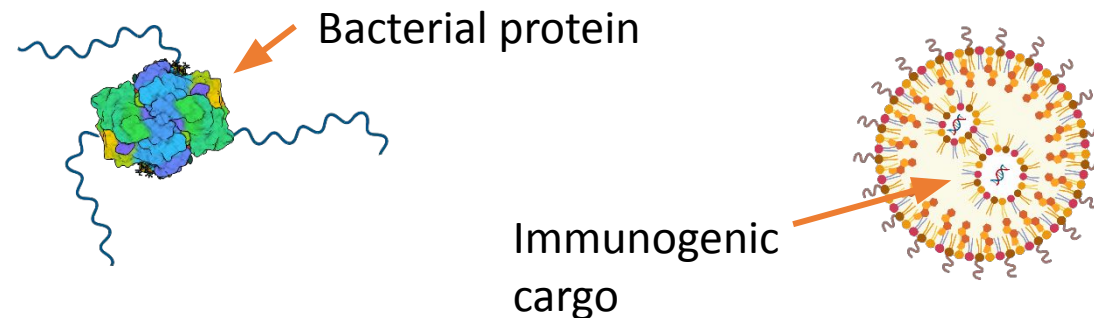


**Anti-PEG antibodies can bind POEGMA**

**POEGMA conjugates induce anti-POEGMA antibodies**



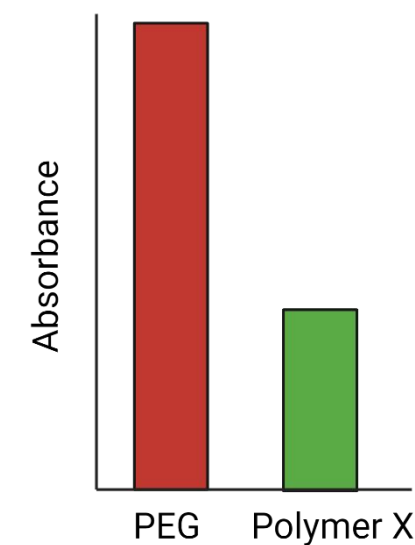
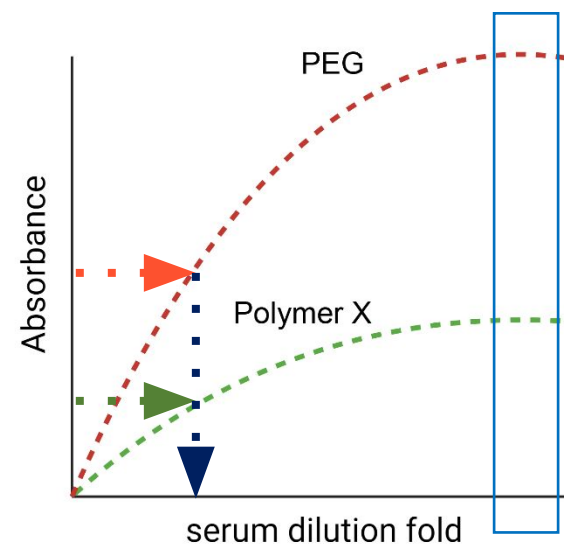
1. Use immunogenic conjugates

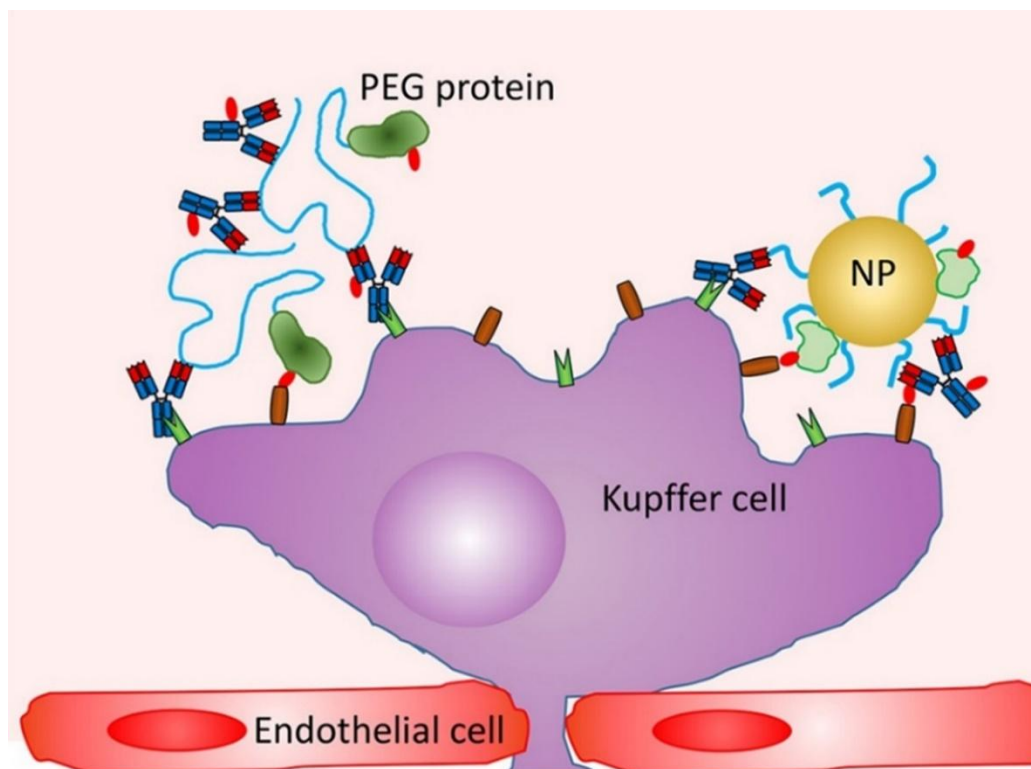


2. Inject female BALB/c mice

3. Multiple injections

4. Compare antibody response not OD





ABC when antibody > PEG compound

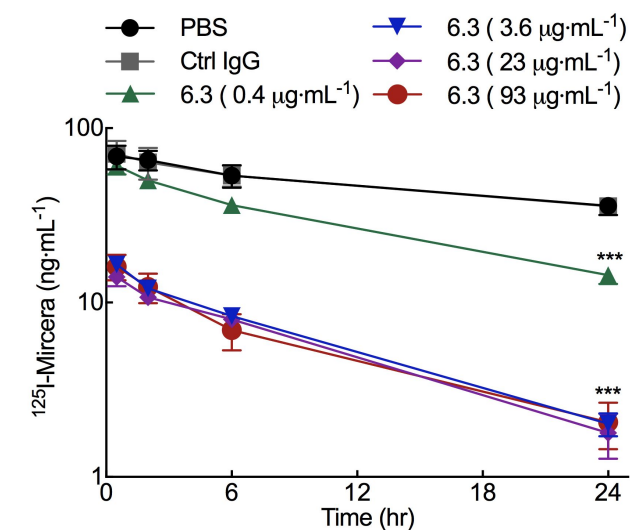
~ 3:1 on a molar basis



Shelly Chang

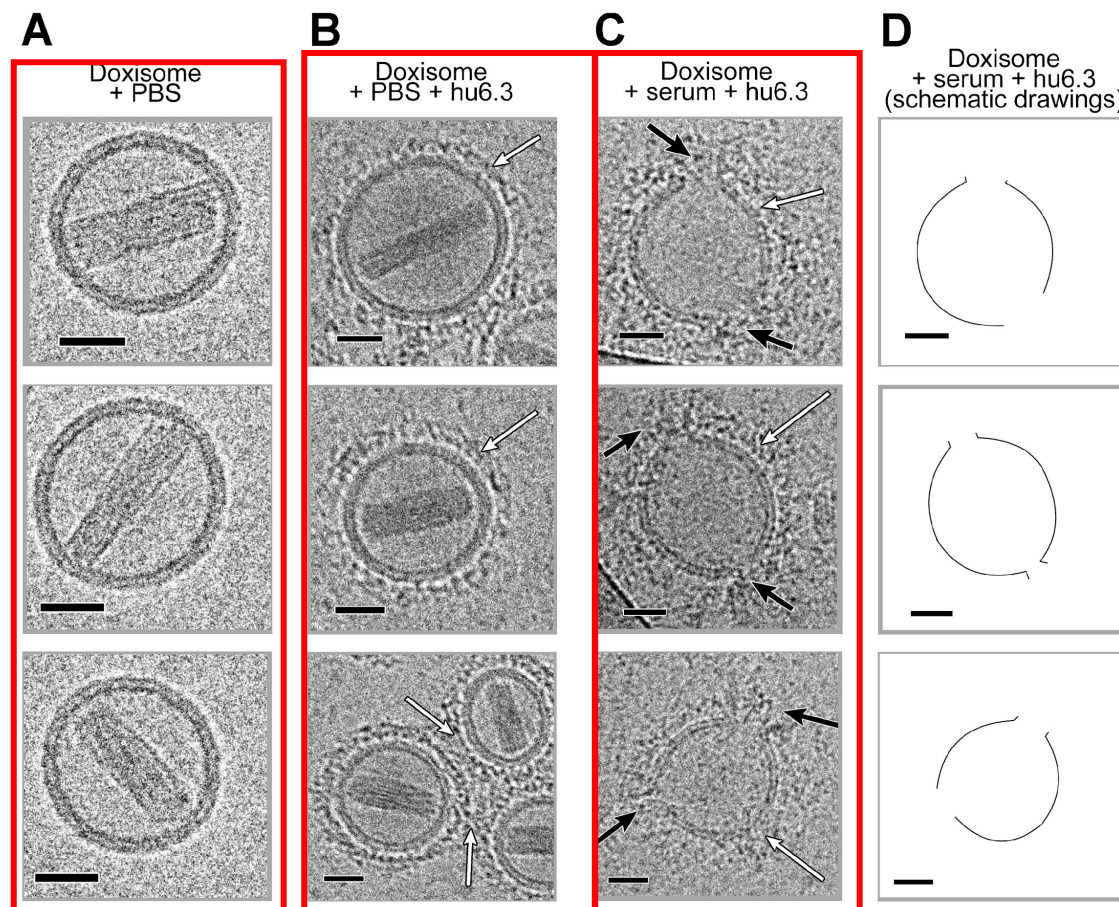
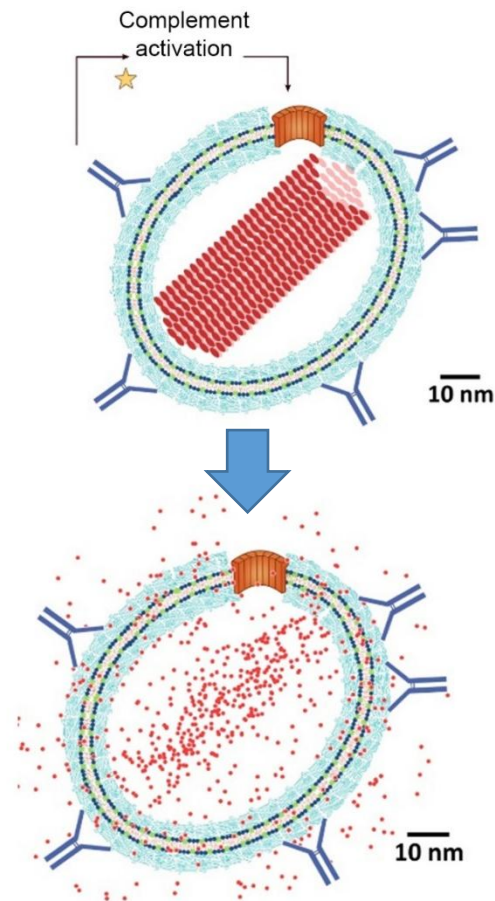


Ronald Lin



YC Lin et al., J Control Release 354: 354-367 (2023)  
 BM Chen et al., ACS Nano 15:14022–14048 (2021)  
 TC Chang et al., Pharmaceutics 12:15 (2020)  
 TL Cheng et al., Bioconj. Chem. 10: 520-528 (1999)

# Anti-PEG antibodies can activate complement to destabilize PLD



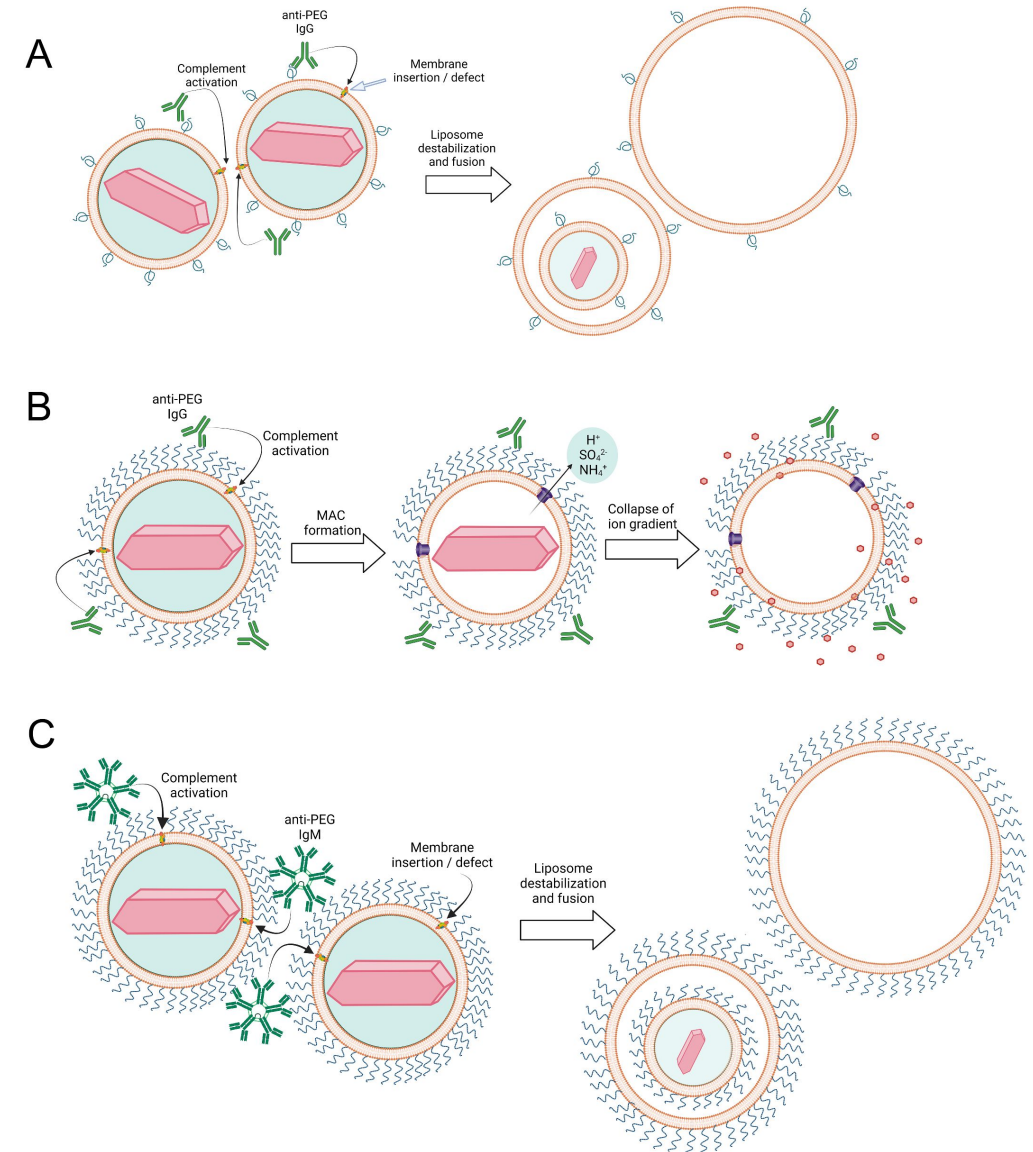
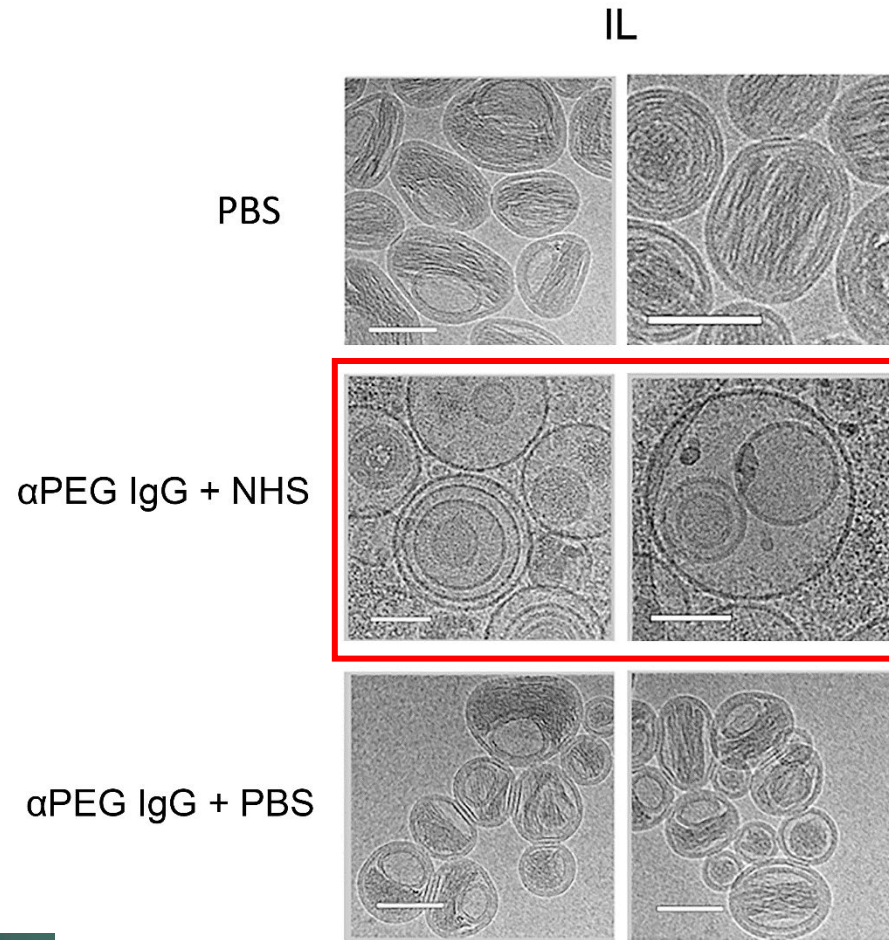
Even Chen



Chen *et al*, ACS Nano 14: 7808-7822 (2020)  
Senti *et al*, J Control Release 341: 475-486 (2022)



# Anti-PEG antibodies destabilize IL (Onivyde) with 0.3% PEG



Chen *et al* 2024, ACS Nano (under revision)



# Vaccine adverse events

| Samples   | Vaccine   | Anaphylaxis | Reference  |
|-----------|-----------|-------------|------------|
| 25,929    | BNT1622b2 | 0.027%      | Blumenthal |
| 38,971    | mRNA-1273 | 0.023%      | Blumenthal |
| 1,892,269 | BNT1622b2 | 0.001%      | CDC        |



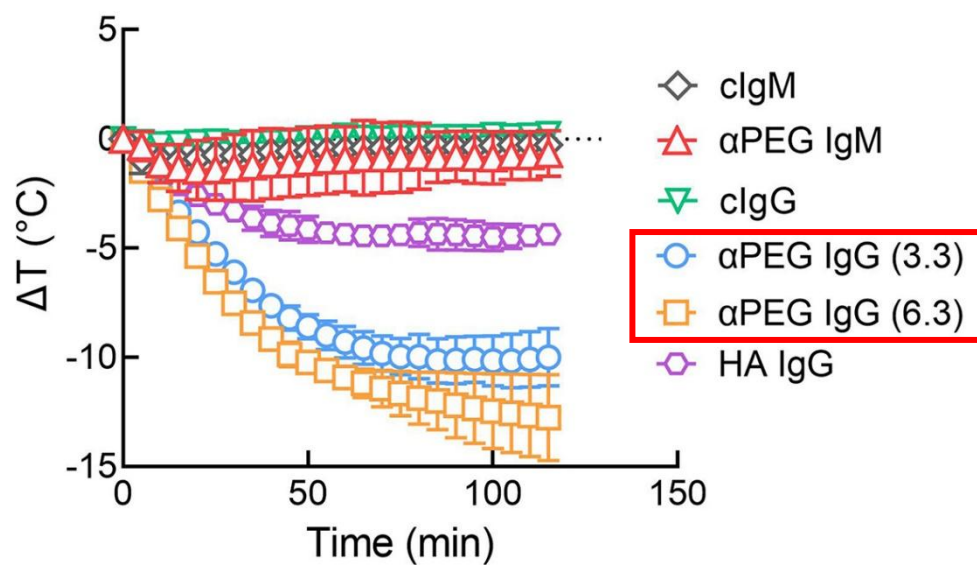
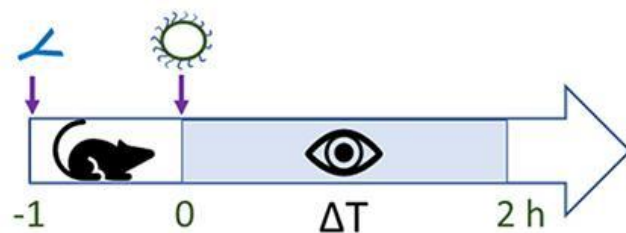
KG Blumenthal *et al.*, *Jama* 325.15:1562-1565 (2021)  
CDC, *Morbidity and mortality weekly report* 70.2: 46 (2021)



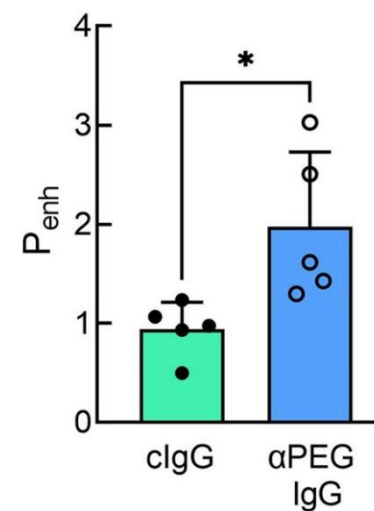
Vivian Chen



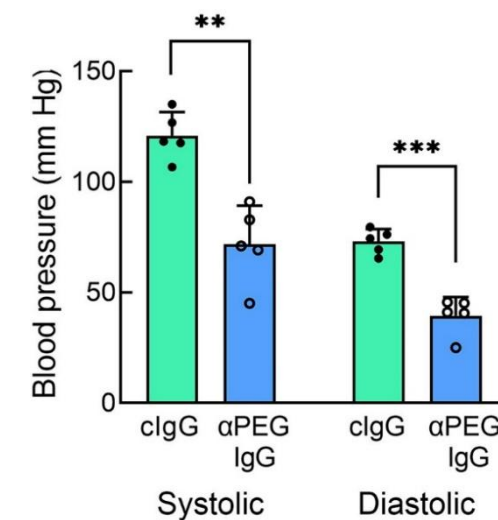
Peter Chang



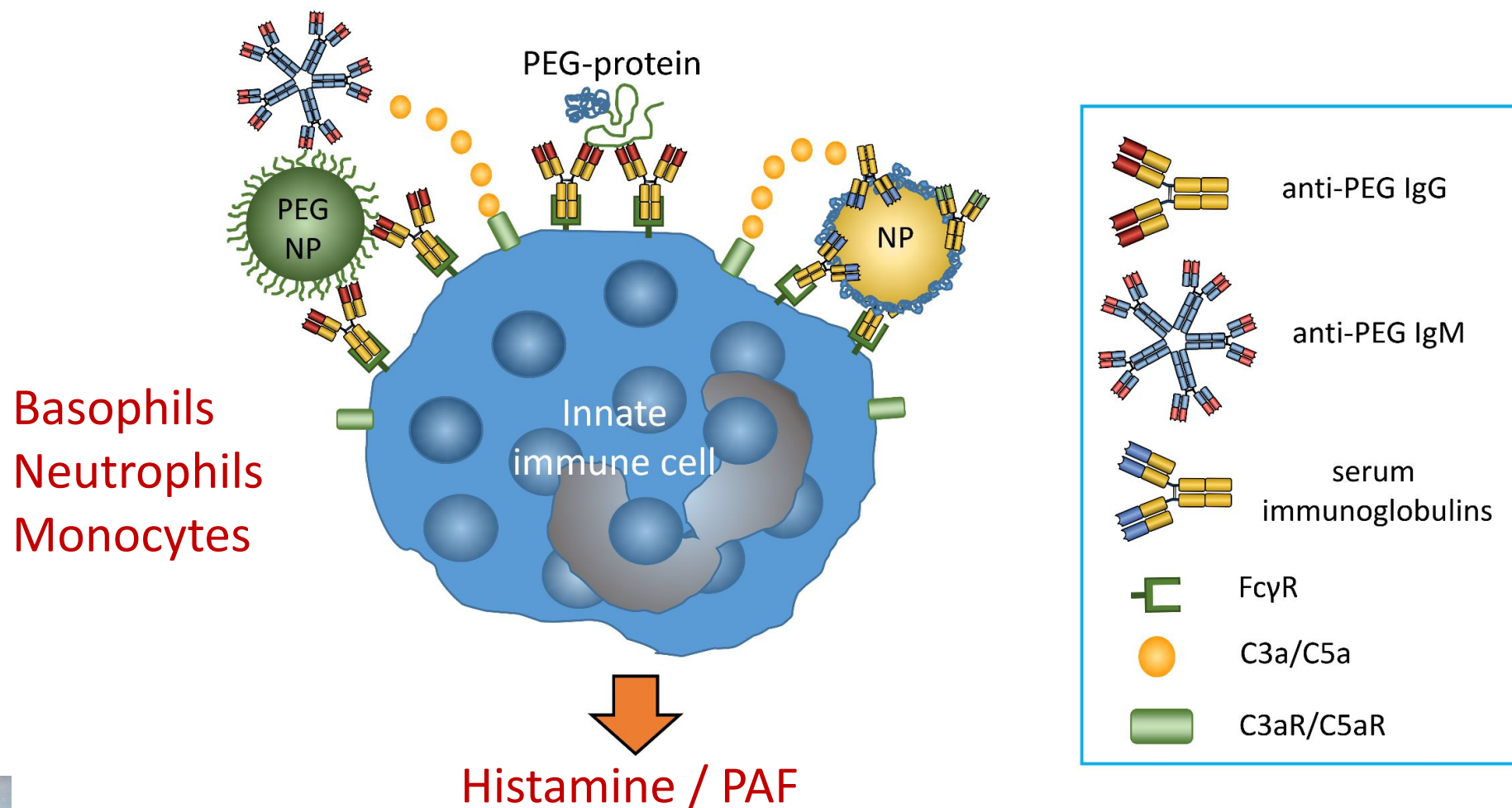
## Lung function



## Blood pressure



Chen *et al.*, *ACS Nano* 17:5757–5772 (2023)



Stavnsbjerg *et al.*, J. Control Release 342: 337-344 (2022)  
Chen *et al.*, ACS Nano 17:5757–5772 (2023)

# PEG bispecific Abs and engagers

Additional  
function

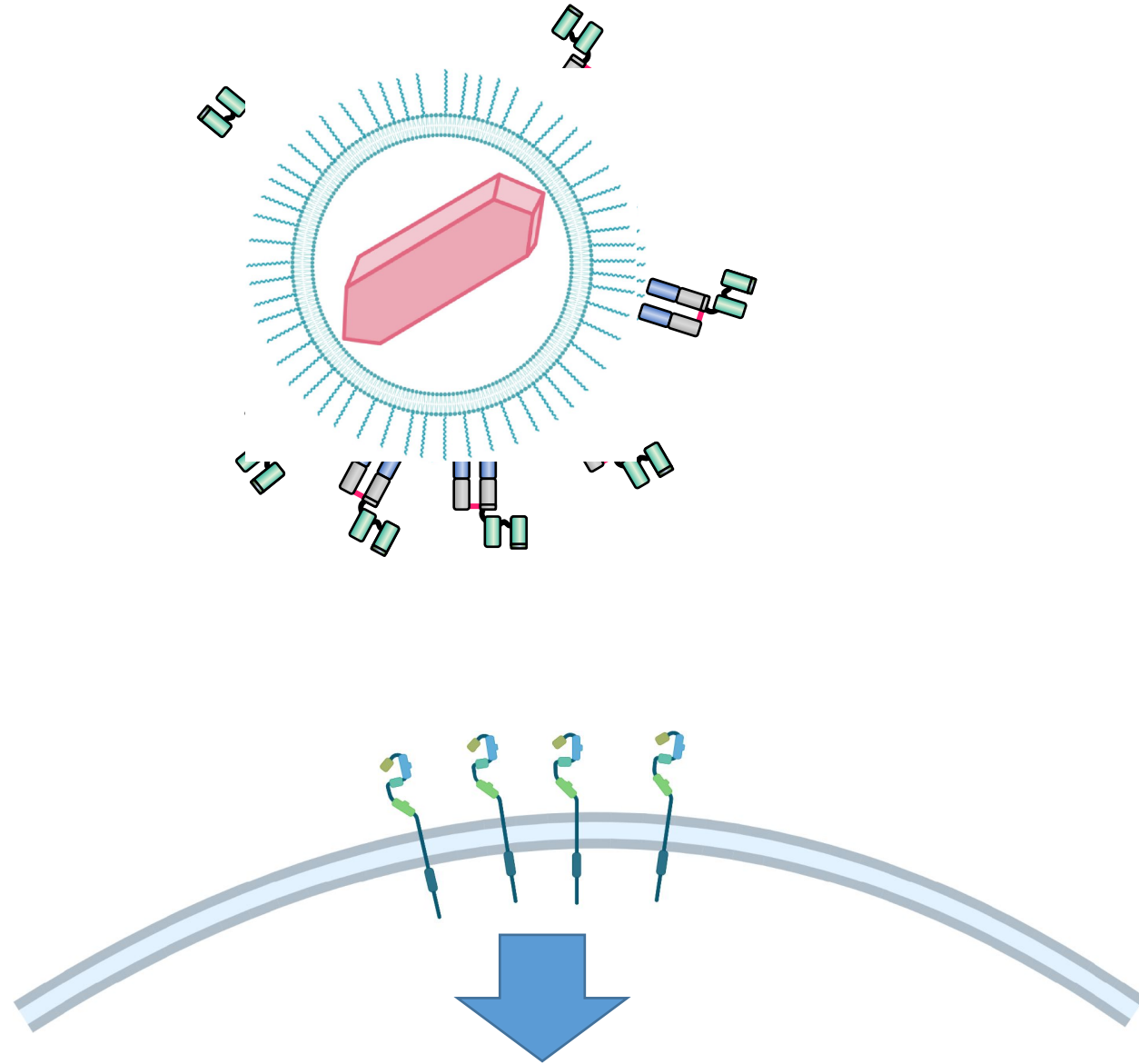
anti-cancer  
antibody

anti-PEG  
scFv

anti-PEG  
Fab

Targeting

anti-cancer  
scFv



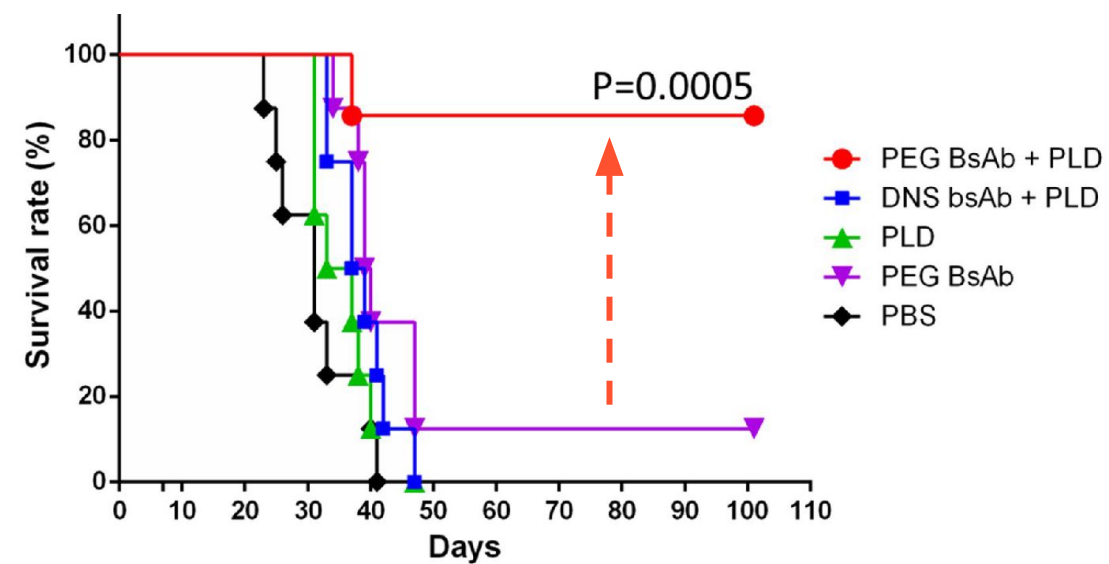
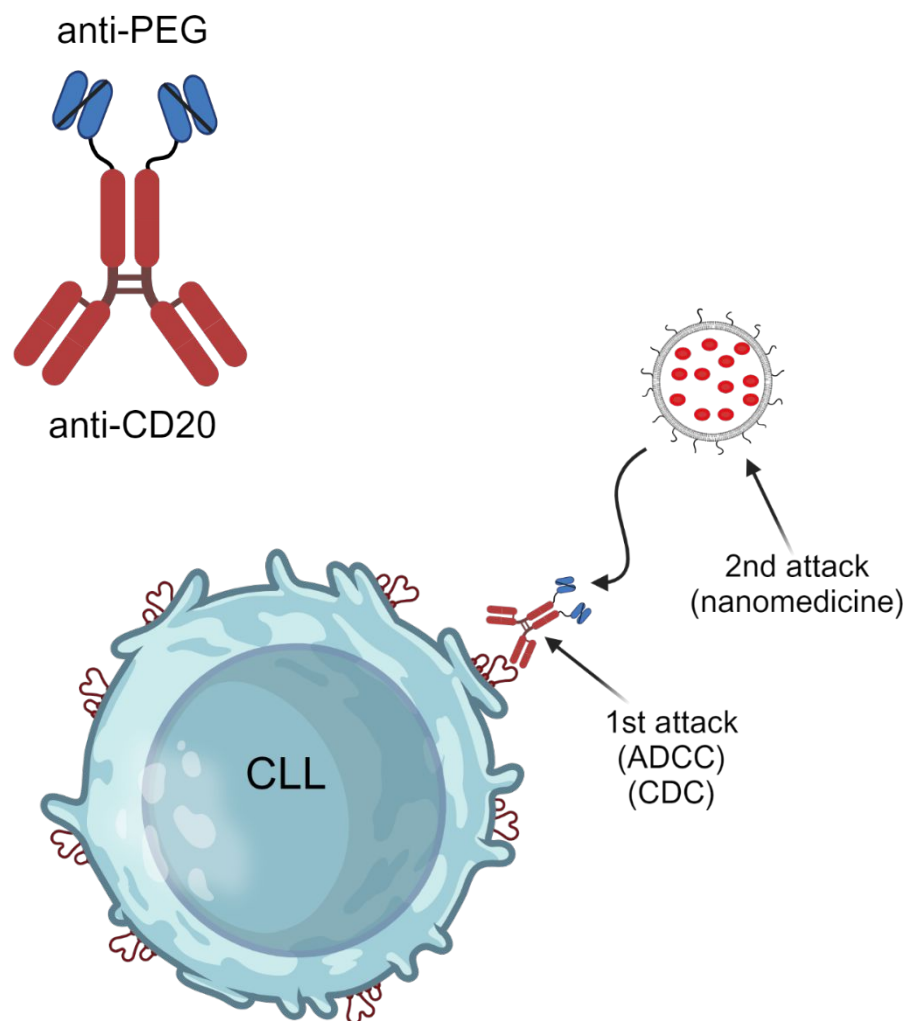




Kai-Wen Ho  
何愷文

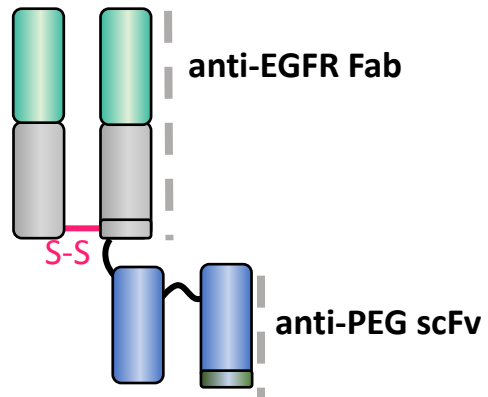


鄭添祿

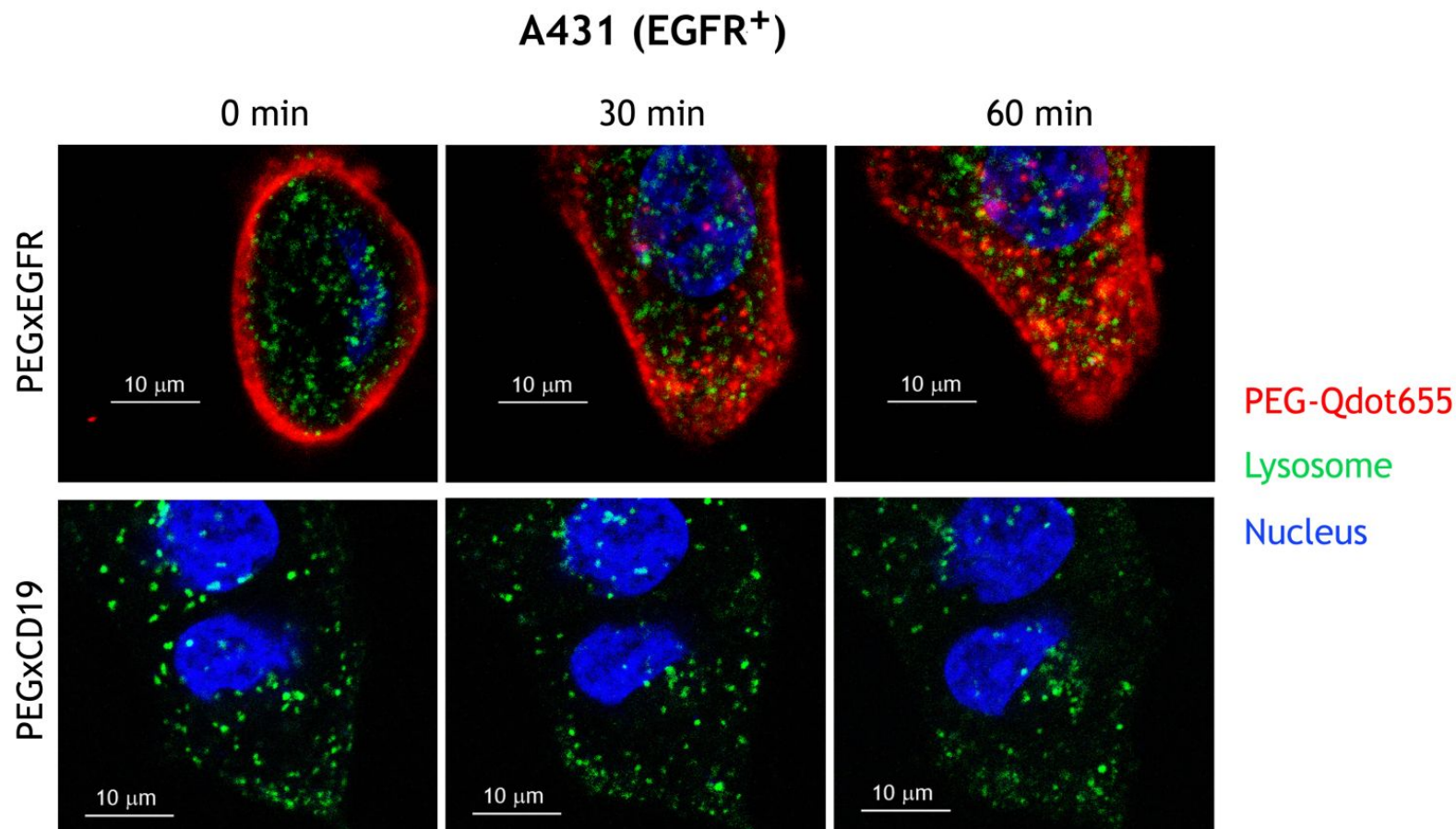


Ho et al, J. Nanobiotechnol. (2021)

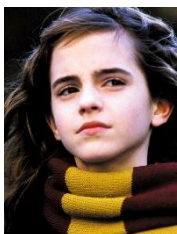
# PEG engagers can increase NP endocytosis



PEG-Qdot655



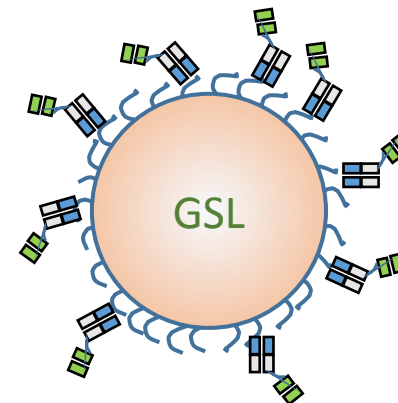
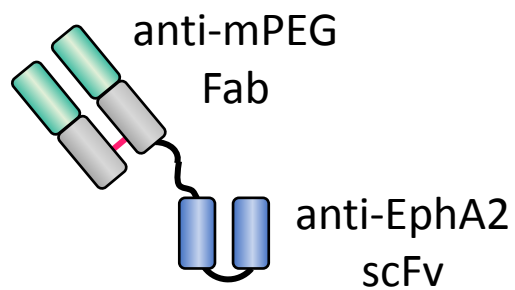
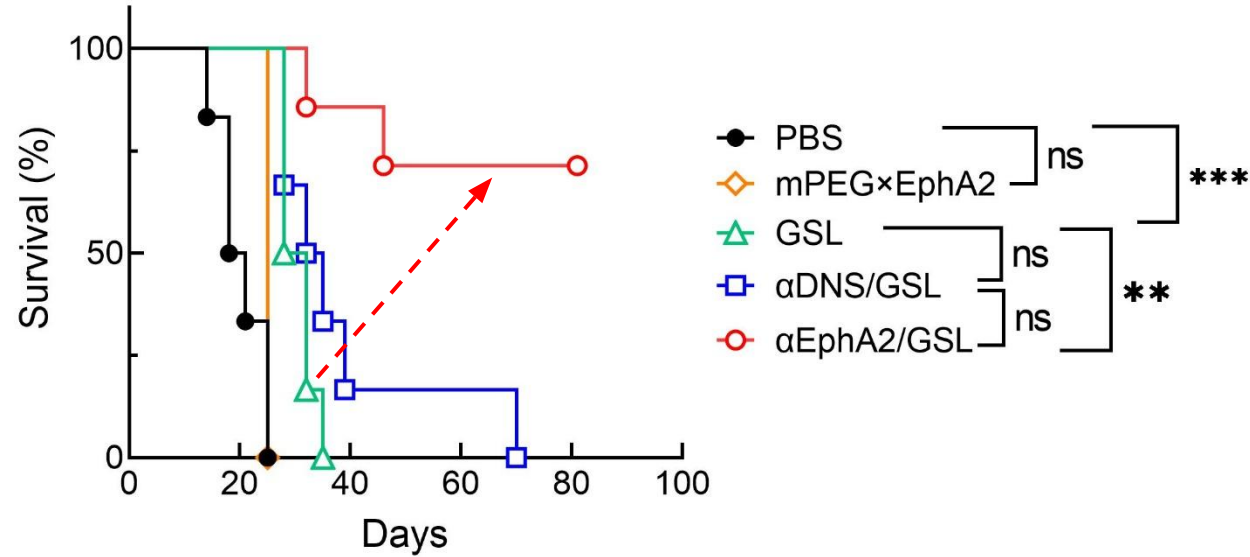
Johnny Su  
蘇昱誠



Su et al., Nat Commun., 8:15507 (2017)



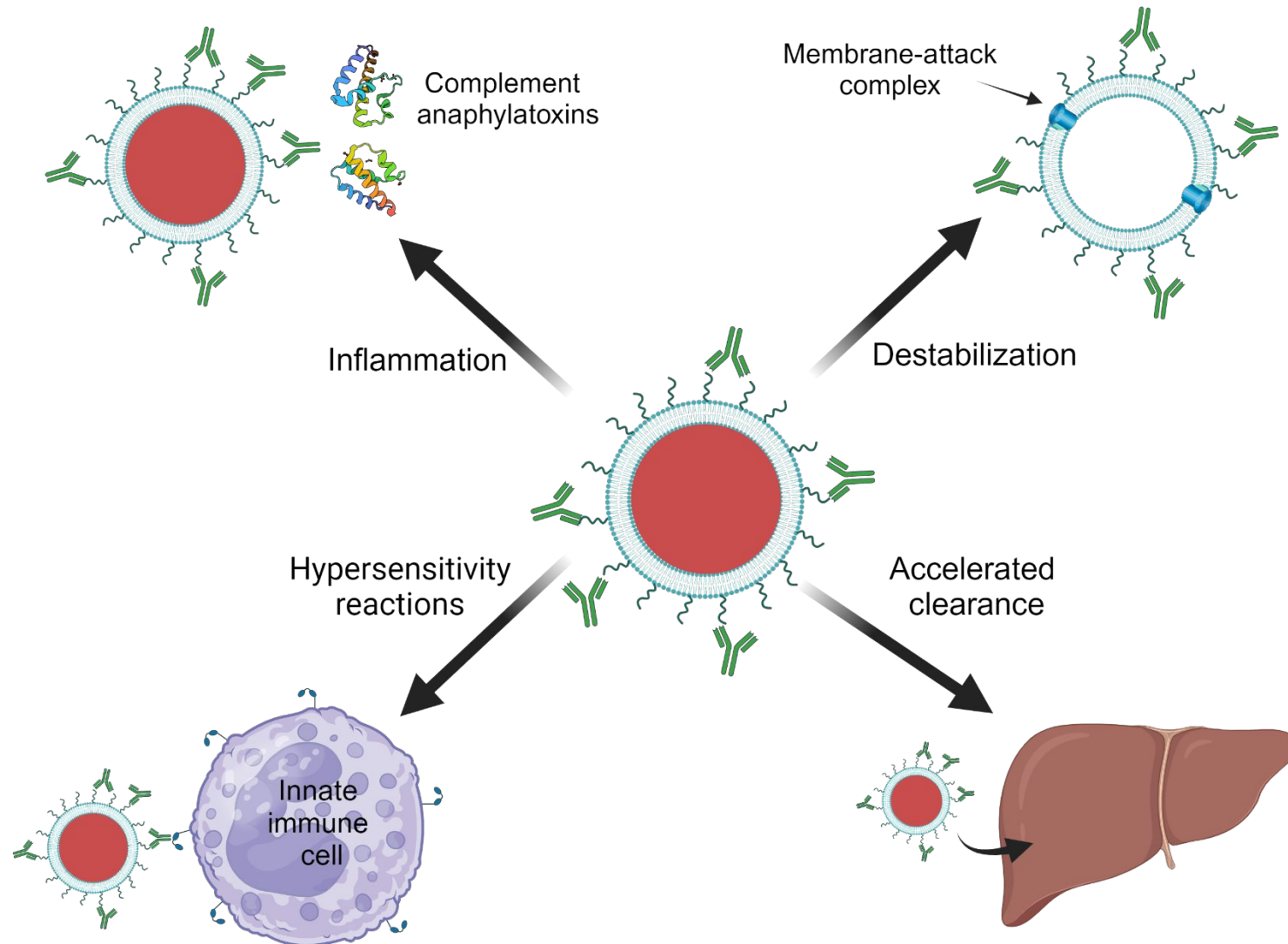
Kai-Wen Ho 何愷文  
Graduate Institute of Medicine  
Kaohsiung Medical University



Burnouf *et al.*, Nature Comm., 9:1843 (2018)  
Ho et al., Submitted



# Effects of anti-PEG antibodies





Anti-PEG IgG is important for many in vivo effects

Anti-PEG antibody response depends on cargo immunogenicity

Pre-existing anti-PEG Ab concentrations are low in most people

Covid-19 mRNA vaccines increase anti-PEG antibody concentrations  
(Moderna > BNT)

Duration of mRNA vaccine induced anti-PEG antibodies?

Anti-PEG antibody impact on new mRNA therapeutics?

Will mRNA vaccines or therapies affect other pegylated medicines?

Role of anti-PEG IgE or IgA antibodies?



# Acknowledgements



鄭添祿



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Department of Biomedical Science and Environmental Biology



呂玉玲



嘉南藥理大學  
Chia Nan University  
of Pharmacy & Science



林俊宏



生醫轉譯研究中心  
Biomedical Translation Research Center



Yechezkel  
Barenholz



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THE HEBREW UNIVERSITY OF JERUSALEM



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TAIWAN

