Passion for Innovation.
Compassion for Patients.™

Daiichi-Sankyo

Development of mRNA vaccines

Oct 5, 2021　Summary from DS seminar

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Vaccine Preparedness

To contribute to society through establishing pharmaceutical technology and manufacturing capability for vaccine preparedness

Stable vaccine supply through in-house manufacturing facility

- DS's marketed vaccines are being stably supplied from a domestic manufacturing facility
  - Seasonal influenza HA vaccine
  - Live vaccines (measles, rubella, and mumpus)

Vaccine R&D by utilizing innovative modality

- Development of DS-5670*
  - **Initiated Ph1/2 study in March 2021**
  - DS-5670 utilizes original LNP that efficiently encapsulates mRNA and confers efficient delivery of mRNA to targets
- To build a platform that streamlines development and manufacturing of a variety of LNP-mRNA vaccines for **future emerging/re-emerging infectious diseases**

To build vaccine manufacturing facilities for future pandemics

- To establish in-house and domestic manufacturing facilities through an enterprise supported by MHLW
- To acquire capability of stable and emergency supply for vaccine preparedness and to become an essential infrastructure for emergency preparedness through collaboration with other organizations in the pharmaceutical industry

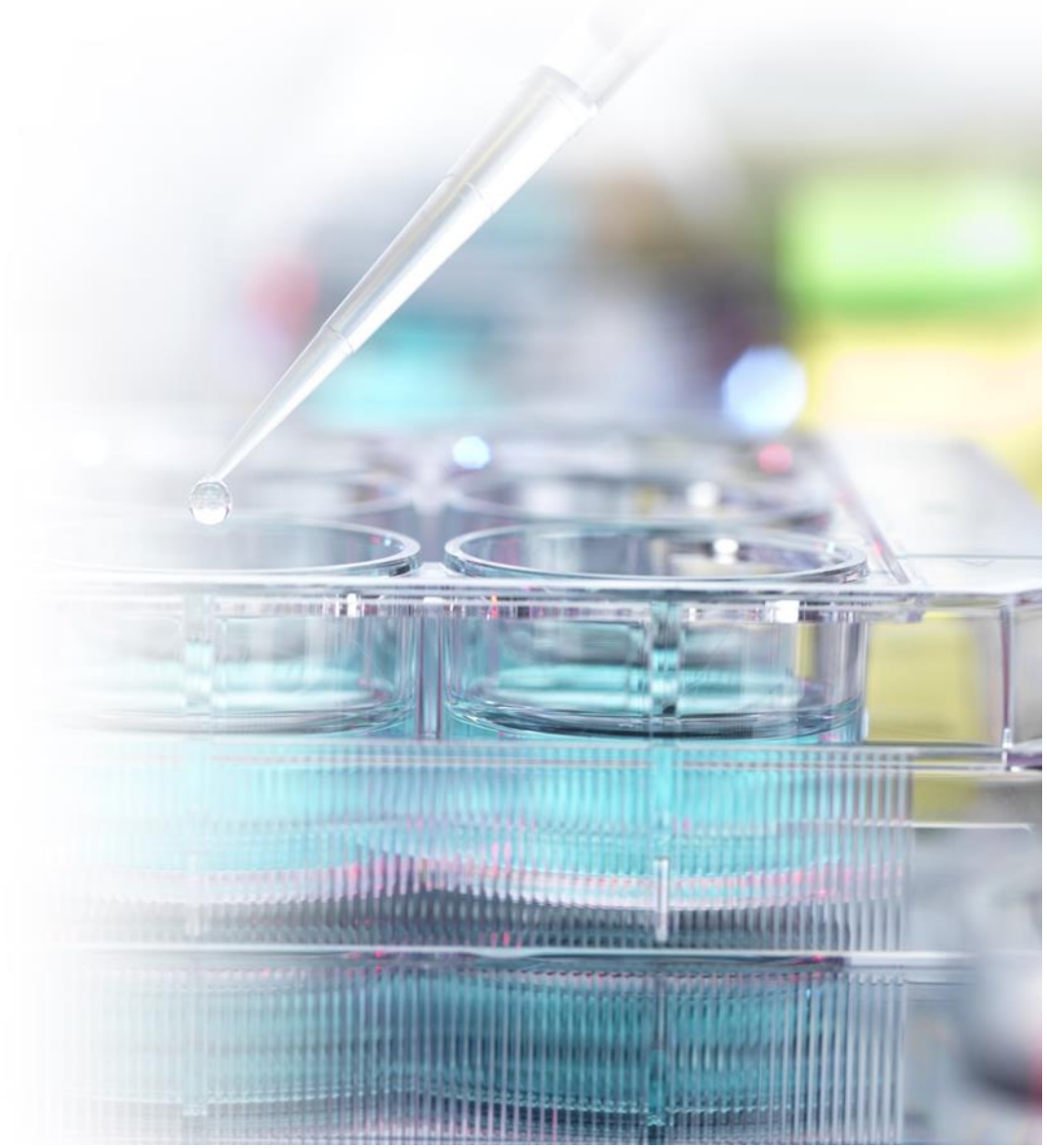
*Development of DS-5670 has been supported by AMED and MHLW

Agenda

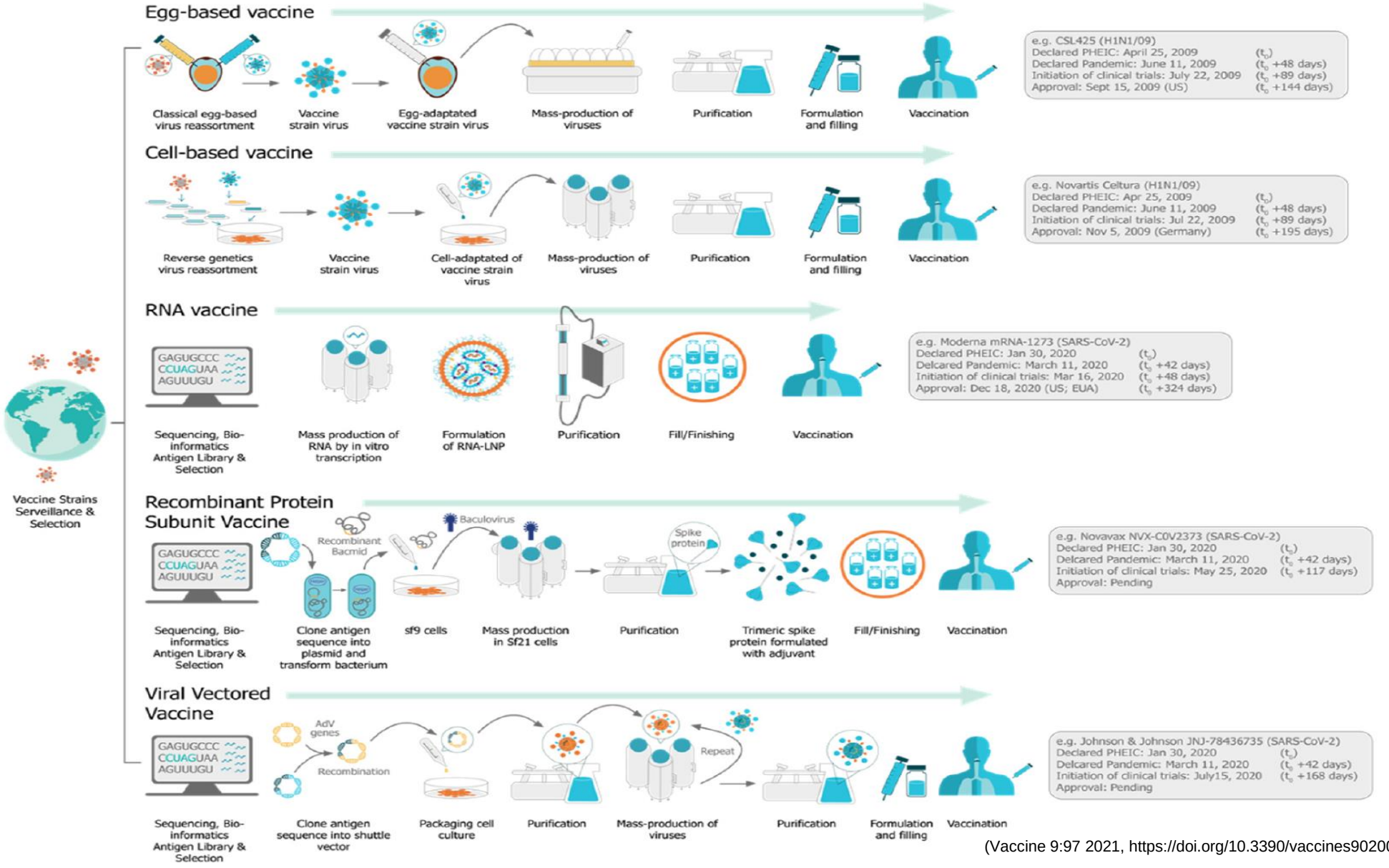
1. LNP-mRNA vaccine technology

2. COVID-19 vaccine（DS-5670）preclinical data

3. Current status of DS-5670 development and future plan

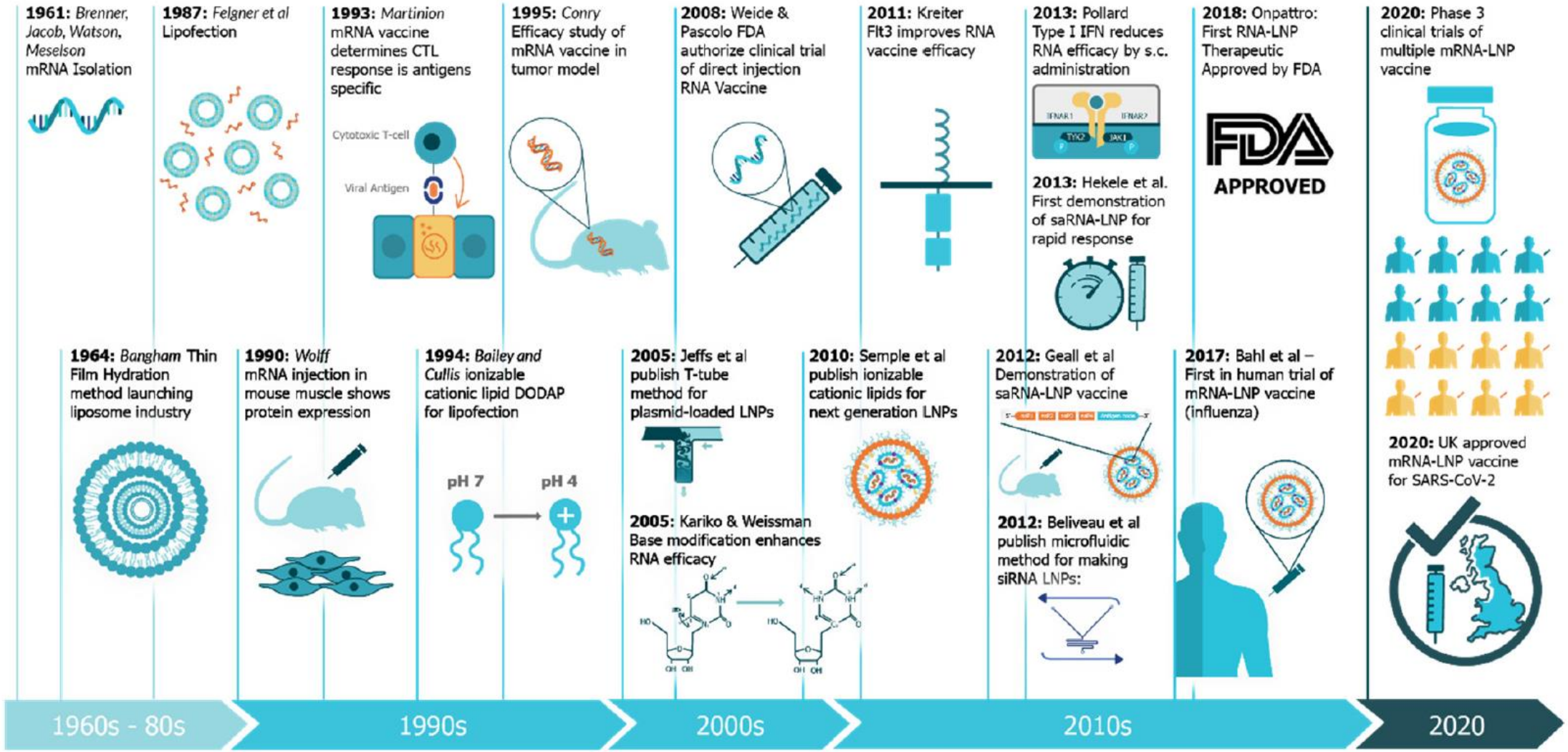


Manufacturing processes for different vaccine platforms



(Vaccine 9:97 2021, https://doi.org/10.3390/vaccines9020097)

History of technology development related to mRNA vaccines



(Vaccine 9:97 2021, https://doi.org/10.3390/vaccines9020097)

The use of mRNA as a treatment modality (as of 2017)

mRNA modality	No. of programs per R&D phase							
	Res	Precl	0	IND	I	II	III	Total
Standardized cancer vaccines		1	1		3	1		6
Individualized cancer vaccines			1		2		1	4
Therapeutic infectious disease vaccines						2		2
Prophylactic infectious disease vaccines & adjuvants	4	4	1	1	6			16
Replicon RNA infectious disease vaccines	3							3
Protein therapeutics for cancer & CV			1	1	1			3
Protein therapeutics for mono-genetic diseases	8	8	3					19
mRNA antibody therapeutics	4	1						5
Ex vivo gene editing	2		2					4
In vivo gene editing	9	1						10
Ex vivo T cell engineering			2					2

(mRNA Vaccines & Therapeutics 2017: an industry analysis of technologies, pipelines, stakeholders and deals released by La Merie Publishing on June 18, 2017)

Clinical studies assessing mRNA vaccines for infectious disease other than COVID-19 (as of Aug 2021)

Disease target	*Study stage*	*Delivery formulation*	*Status*	*Organization*
CMV	Ph-2	LNP	Ongoing	Moderna
RSV	Ph-1	Merck proprietary formulation	Ongoing	Merck/Moderna
RSV	Ph-1	Not disclosed	Completed	Merck/Moderna
RSV	Ph-2	LNP	Ongoing	Moderna
Rabies	Ph-1	Cationic lipid formulation	Ongoing	GSK
Rabies	Ph-1	LNP	Ongoing	CureVac
Rabies	Ph-1	Protamine	Completed	CureVac
Chikungunya	Ph-1	Not disclosed	Ongoing	Moderna
hMPV/PIV3	Ph-1	LNP	Completed	Moderna
Novel Flu (H10N8, H7N9)	Ph-1	LNP	Completed	Moderna
Zika	Ph-1	LNP	Completed	Moderna
Seasonal Flu	Ph-1	LNP	Ongoing	Moderna, TranslateBio/SP, BioNTech/Phizer

(npj Vaccines 5: Article number 11, 2020)

mRNA vaccine candidates in clinical trials for COVID-19
(as of Sep 24, 2021)

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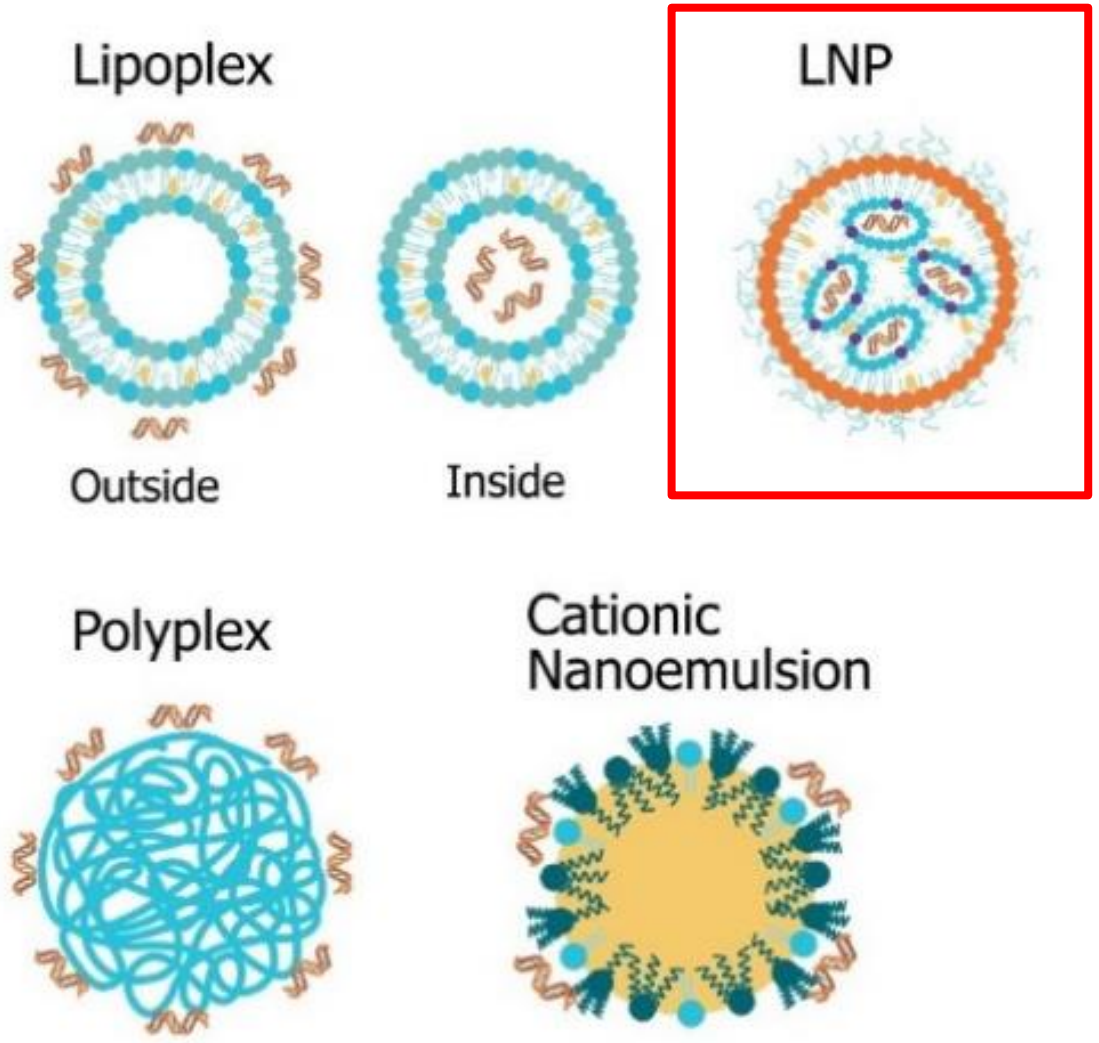
ID	Vaccine platform acronym	Vaccine platform description	Type of candidate vaccine	Number of doses	Schedule	Route of administration	Developers	Phase
9	RNA	RNA based vaccine	mRNA-1273	2	Day 0 + 28	IM	Moderna + National Institute of Allergy and Infectious Diseases (NIAID)	Phase 4
10	RNA	RNA based vaccine	BNT162b2 (3 LNP-mRNAs), also known as "Comirnaty"	2	Day 0 + 21	IM	Pfizer/BioNTech + Fosun Pharma	Phase 4
12	RNA	RNA based vaccine	CVnCoV Vaccine	2	Day 0 + 28	IM	CureVac AG	Phase 3
22	RNA	RNA based vaccine	ARCT-021	NR	NR	IM	Arcturus Therapeutics	Phase 2
38	RNA	RNA based vaccine	LNP-nCoVsaRNA	2	NR	IM	Imperial College London	Phase 1
39	RNA	RNA based vaccine	SARS-CoV-2 mRNA vaccine (ARCoV)	2	Day 0 + 14 or Day 0 + 28	IM	Academy of Military Science (AMS), Walvax Biotechnology and Suzhou Abogen Biosciences	Phase 3
46	RNA	RNA based vaccine	ChulaCov19 mRNA vaccine	2	Day 0 + 21	IM	Chulalongkorn University	Phase 1
71	RNA	RNA based vaccine	PTX-COVID19-B, mRNA vaccine	2	Day 0 + 28	IM	Providence Therapeutics	Phase 1
73	RNA	RNA based vaccine	CoV2 SAM (LNP) vaccine. A self-amplifying mRNA (SAM) lipid nanoparticle (LNP) platform + Spike antigen	2	Day 0 + 30	IM	GlaxoSmithKline	Phase 1
77	RNA	RNA based vaccine	mRNA-1273.351. A lipid nanoparticle (LNP)-encapsulated mRNA-based vaccine that encodes for a full-length, prefusion	3	Day 0 or Day 0 + 28 or Day 56	IM	Moderna + National Institute of Allergy and Infectious Diseases (NIAID)	Phase 4
82	RNA	RNA based vaccine	MRT5500, an mRNA vaccine candidate	2	Day 0 + 21	IM	Sanofi Pasteur and Translate Bio	Phase 2
85	RNA	RNA based vaccine	DS-5670a, mRNA vaccine	2	NR	IM	Daiichi Sankyo Co., Ltd.	Phase 1/2
91	RNA	RNA based vaccine	HDT-301: Self-replicating mRNA vaccine formulated as a lipid nanoparticle.	2	Day 0 + 28	IM	SENAI CIMATEC	Phase 1
93	RNA	RNA based vaccine	mRNA-1283	2	Day 0 + 28	IM	ModernaTX, Inc.	Phase 1
95	RNA	RNA based vaccine	EXG-5003; a temperature-sensitive self-replicating RNA vaccine expressing the receptor binding domain of the SARS-CoV-2 spike protein.	1	Day 0	ID	Elixirgen Therapeutics, Inc	Phase 1/2
98	RNA	RNA based vaccine	mRNA COVID-19 vaccine	2	TBD	IM	Shanghai East Hospital and Stemirna Therapeutics	Phase 1
103	RNA	RNA based vaccine	LNP-nCOV saRNA-02 vaccine; Self-amplifying RNA (saRNA) encapsulated in lipid nanoparticles (LNP)	2	Day 0 + 28	IM	MRC/UVRI and LSHTM Uganda Research Unit	Phase 1
104	RNA	RNA based vaccine	mRNA-1273.211. A multivalent booster candidate combining mRNA-1273 plus mRNA-1273.351.	1	Day 0	IM	ModernaTX, Inc.	Phase 2/3
114	RNA	RNA based vaccine	ARCT-154 mRNA Vaccine	2	Day 0 + 28	IM	Arcturus Therapeutics, Inc.	Phase 2/3
115	RNA	RNA based vaccine	ARCT-165 mRNA Vaccine	2	Day 0 + 29	IM	Arcturus Therapeutics, Inc.	Phase 1/2
116	RNA	RNA based vaccine	ARCT-021 mRNA Vaccine	2	Day 0 + 30	IM	Arcturus Therapeutics, Inc.	Phase 1/2

(https://www.who.int/publications/m/item/draft-landscape-of-covid-19-candidate-vaccines)

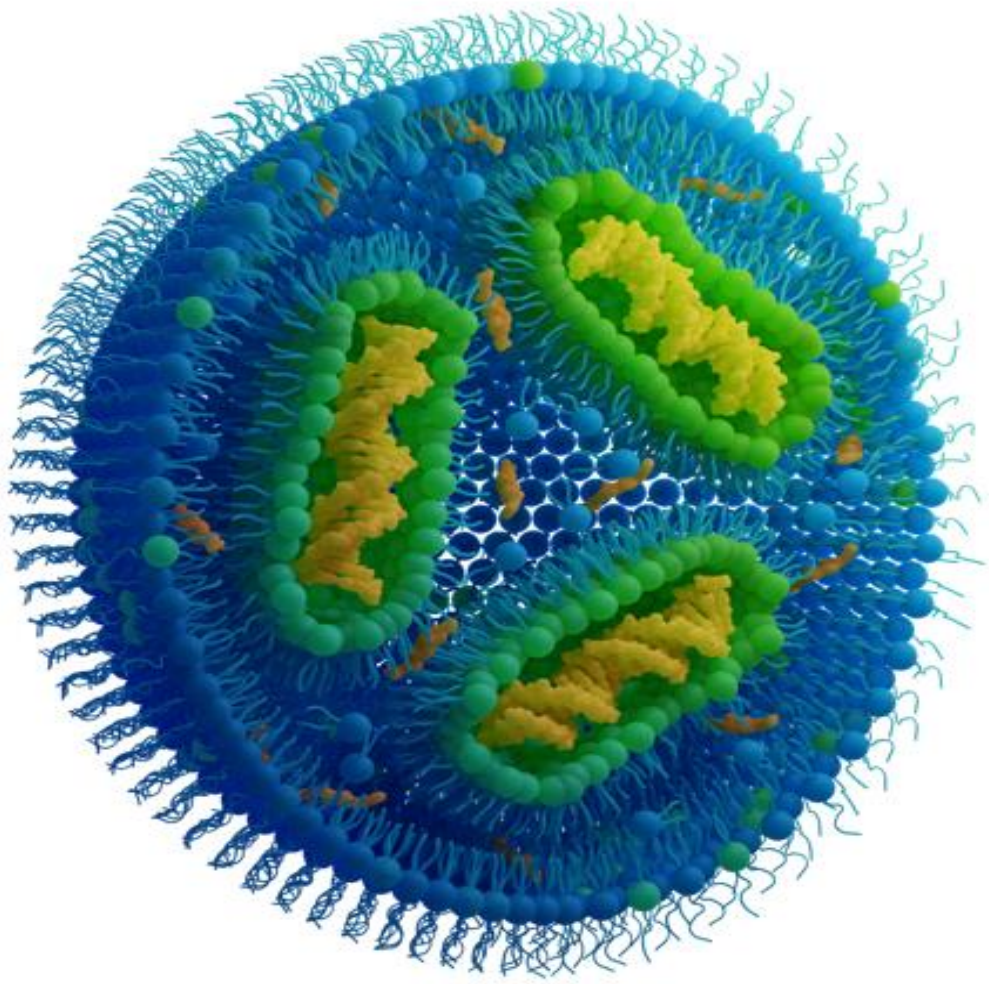
Delivery systems for mRNA



Non-viral mRNA delivery systems. Lipid-, polymer-, and emulsion-based delivery systems all use cationic groups to mediate condensation of the anionic RNA as well as delivery across the cell membrane.

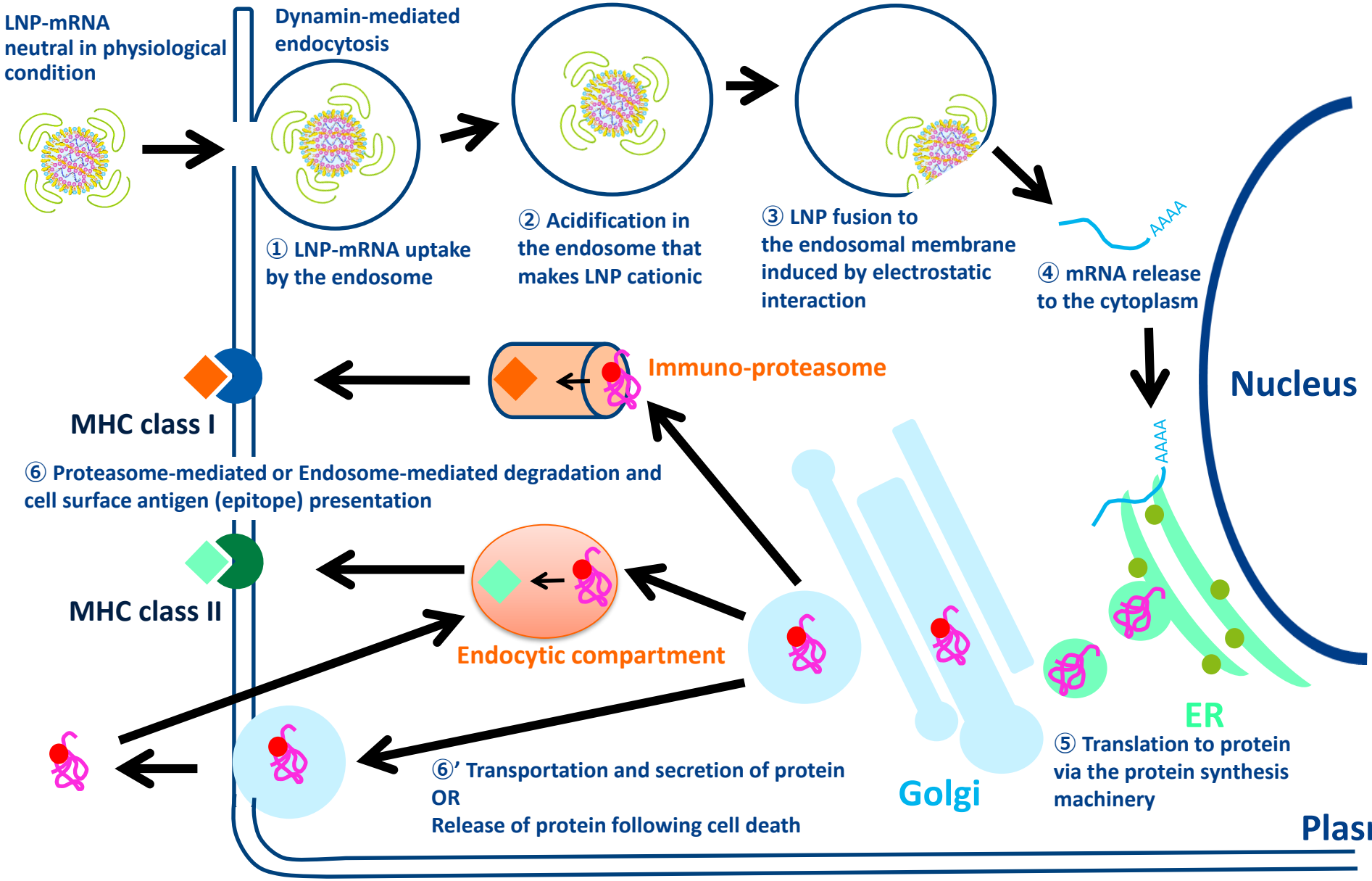
(Vaccine 9:97 2021, https://doi.org/10.3390/vaccines9020097)

Characteristics of DS's LNP-mRNA

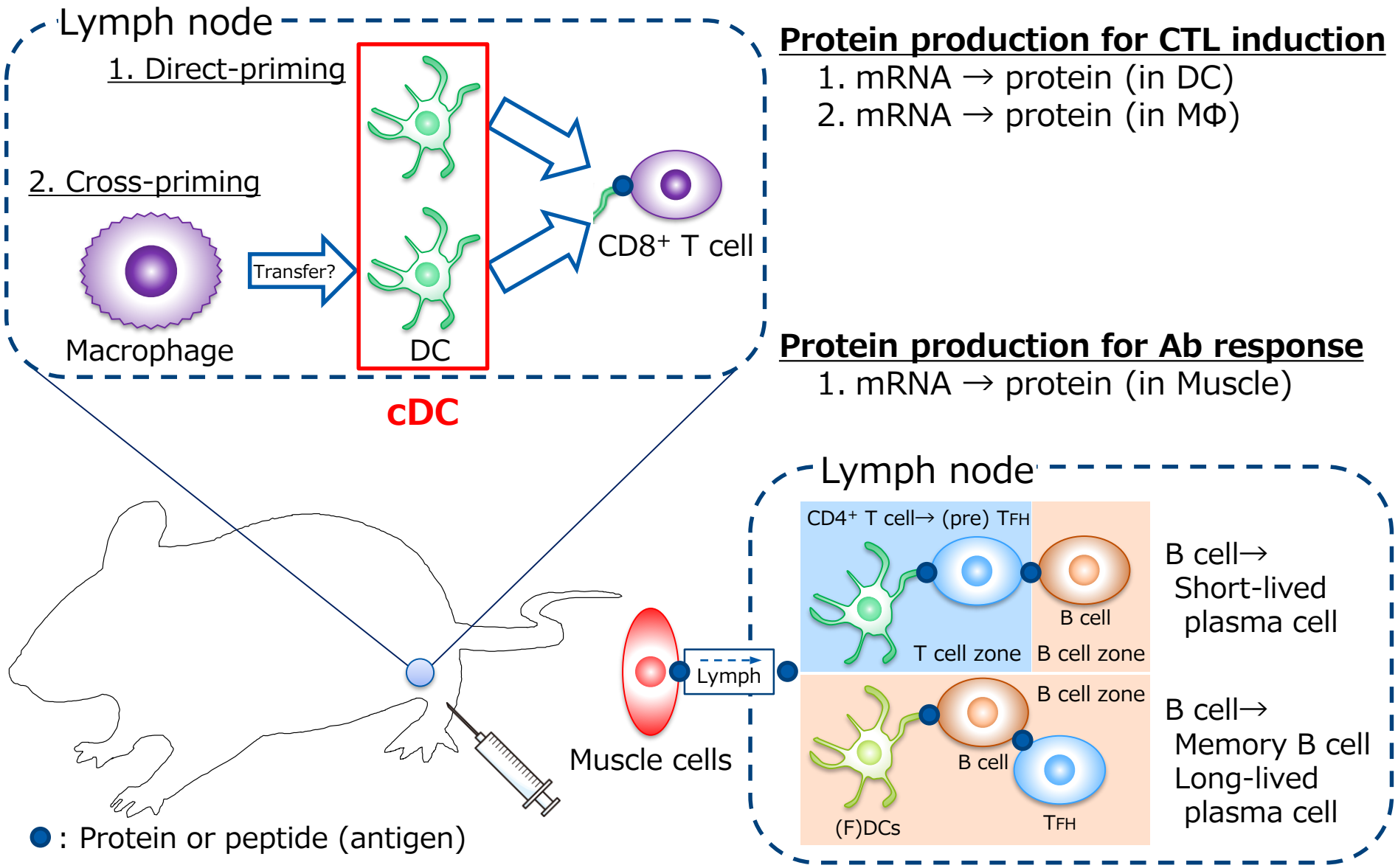


- DS original cationic lipid
- Efficient encapsulation of mRNA in nanoparticles, and efficient delivery of mRNA to targets
- Applicable to pandemic and other vaccines

Proposed mechanism of LNP-mediated subcellular mRNA delivery and process of antigen protein



Proposed immunogenic pathways of LNP-mRNA vaccine



CTL: cytotoxic T lymphocyte, DC: dendritic cells, MΦ: macrophage

Concept of LNP-mRNA vaccine (1/2)

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【Pharmacological and safety profiles】

1. High-level, broad-spectrum antigen-specific immune responses are induced as compared with inactivated or recombinant protein antigens. In addition to antibody and helper T cell responses, cytotoxic T cells, which are necessary to eliminate intracellular pathogens, can be induced.
2. No interfering effect by existing immunity to vaccine formulations such as those observed in live attenuated vaccines and viral vectored vaccines confers stable boosting effects.
3. Due to high quality of antigen proteins produced in vivo, from the viewpoint of post-translational modification and conformation, induced immune responses are qualitatively superior to heterologously expressed antigen protein such as those produced in eggs, insects, or plants.
4. The risk of genetic injury in vaccines, such as carcinogenicity, immune deficiency, and transgenerational transmission, which poses a challenge to other types of genetic vaccines, is expected to be low.

Concept of LNP-mRNA vaccine (2/2)

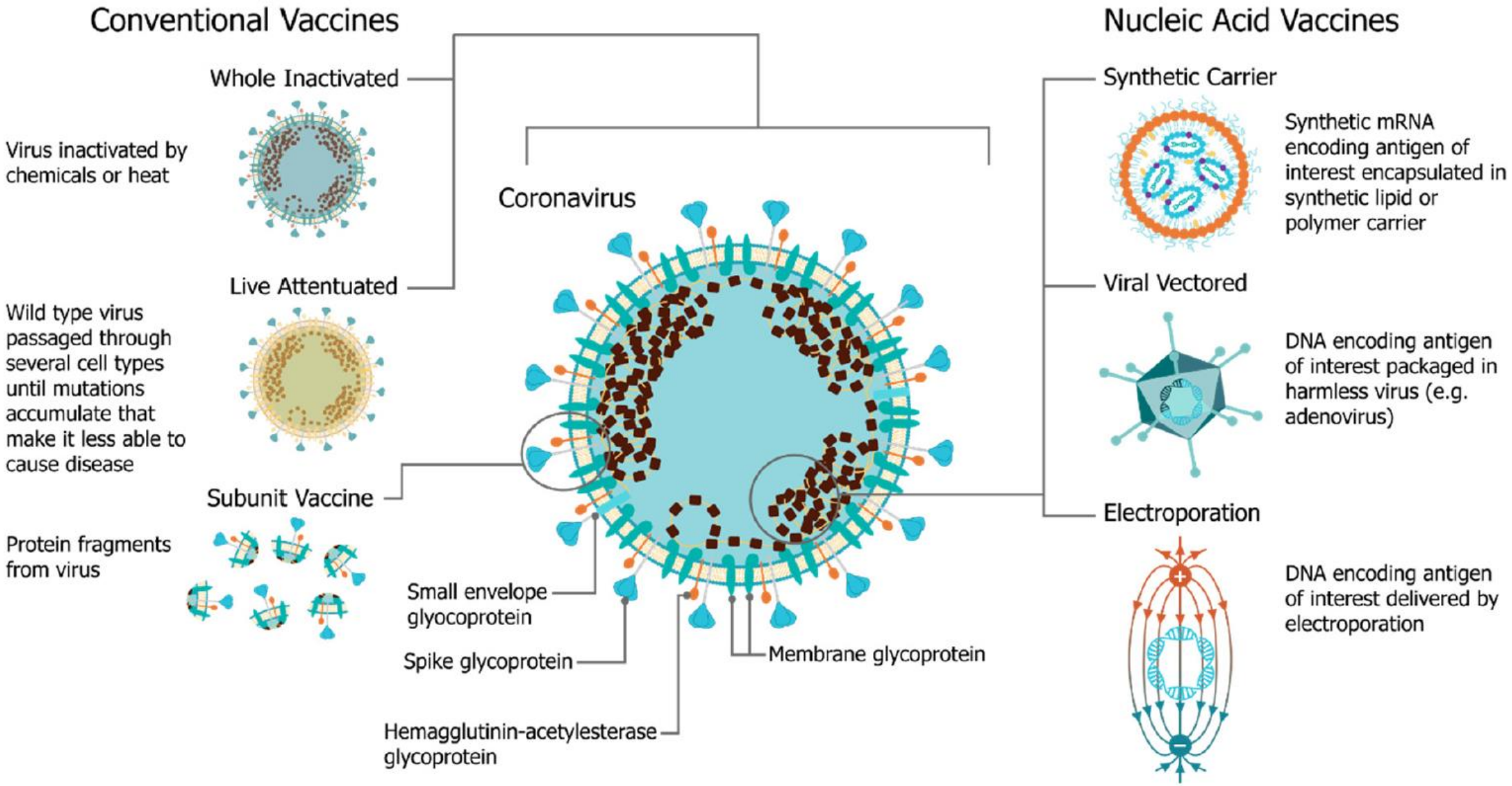
【Quality and manufacturing profiles】

1. Lower risk in quality and manufacturing related to biologics, compared with live vaccines:
   - Non-pathogenic and relatively easy to handle in manufacturing
   - No requirement of bulky facilities for culture of cells or pathogens
   - The lack of in vivo replication ability makes it easier to determine dose
2. Once the platform has been established, development and manufacturing of a variety of LNP-mRNA vaccines can be streamlined

Significance of developing LNP-mRNA vaccines

- Original antigen design
- Feasible to supply vaccines containing novel antigens for breakthrough variants supposed to emerge in the future
- Having experiences in R&D for mRNA vaccine pipeline
- Expected to be superior in domestic development and distribution as compared with other leading mRNA vaccines developed in foreign countries
- To acquire capability of stable and emergency supply for vaccine preparedness and to become an essential infrastructure for emergency preparedness through collaboration with other organizations in the pharmaceutical industry

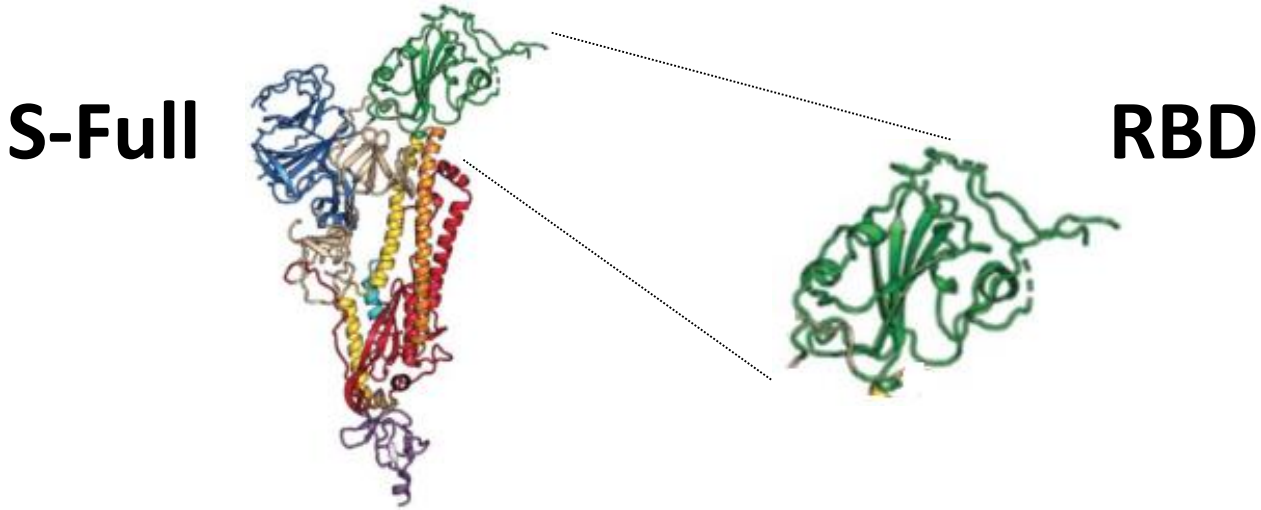
Structure of SARS-CoV-2 and vaccine modalities



(Vaccine 9:97 2021, https://doi.org/10.3390/vaccines9020097)

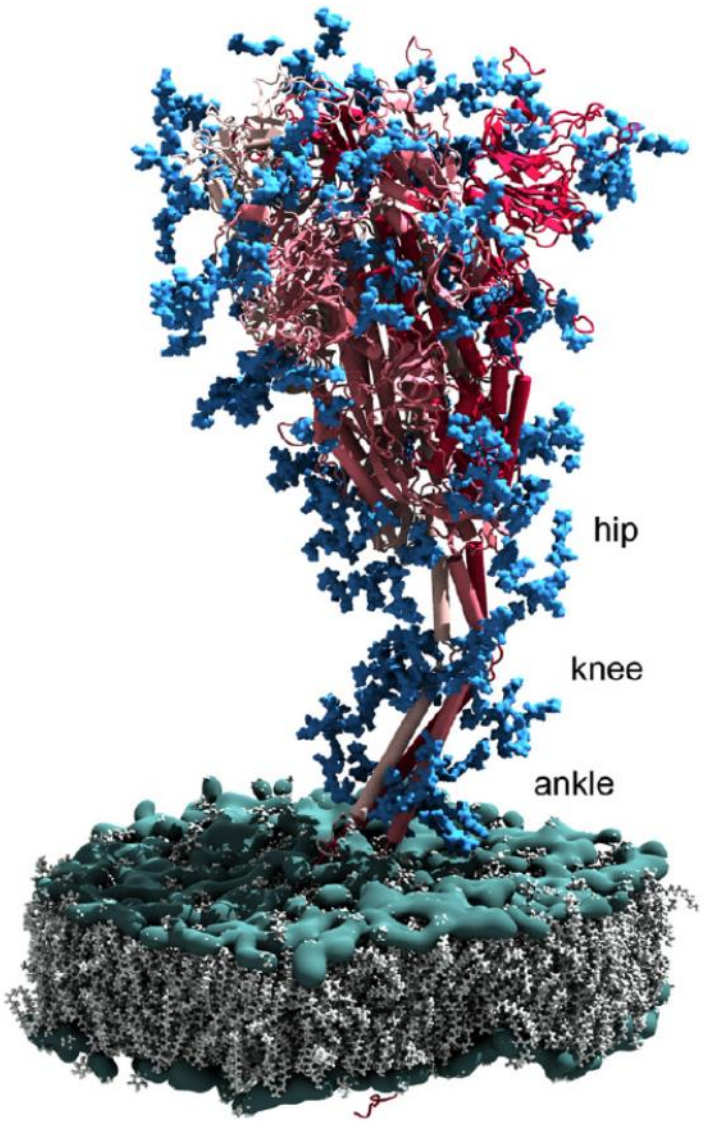
Design of SARS-CoV-2 spike protein (S) antigen for DS-5670

	Full length (S-Full)	Receptor-binding domain (RBD)
Length of mRNA	• 4.1 kb	• 1.0 kb
Proposed advantages	• May contain additional neutralization epitopes and T cell epitopes other than those present in RBD	• Efficient and stable encapsulation of mRNA into LNP because ORF of RBD is shorter than that of S-Full • Lower risk of enhanced disease because potentially pathogenic epitopes are less as compared with S-Full (CELL 12060 https://doi.org/10.1016/j.cell.2021.05.032P NAS 117:8218 2020、Vaccine 25:2832 2007)

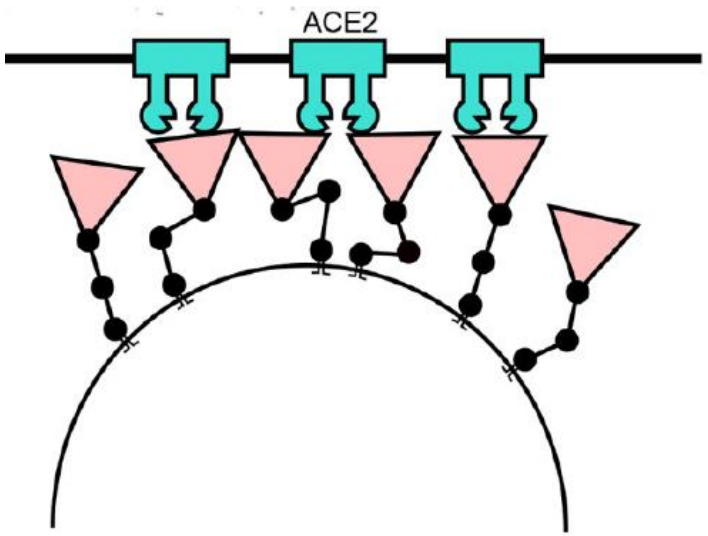


ORF: open reading frame

Superiority of RBD antigen to S-Full antigen



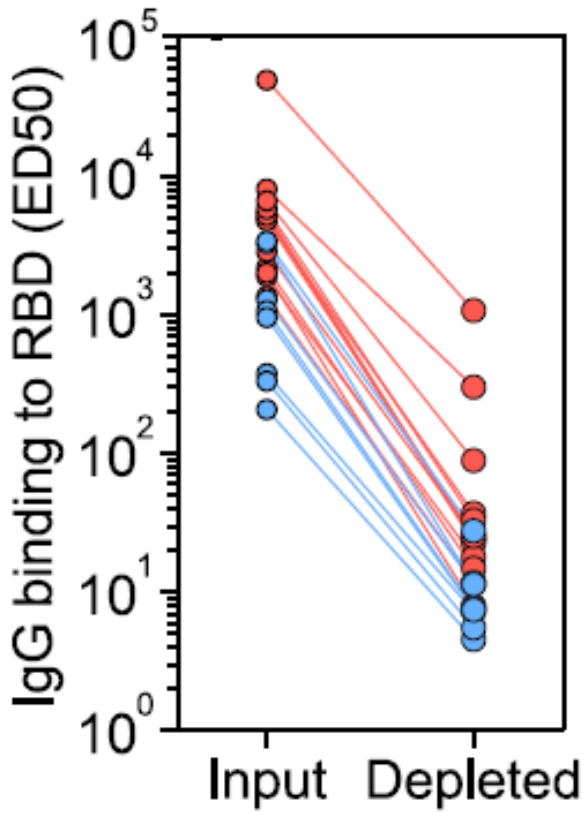
(B. Turoňová et al., Science 10.1126/science.abd5223 (2020).)

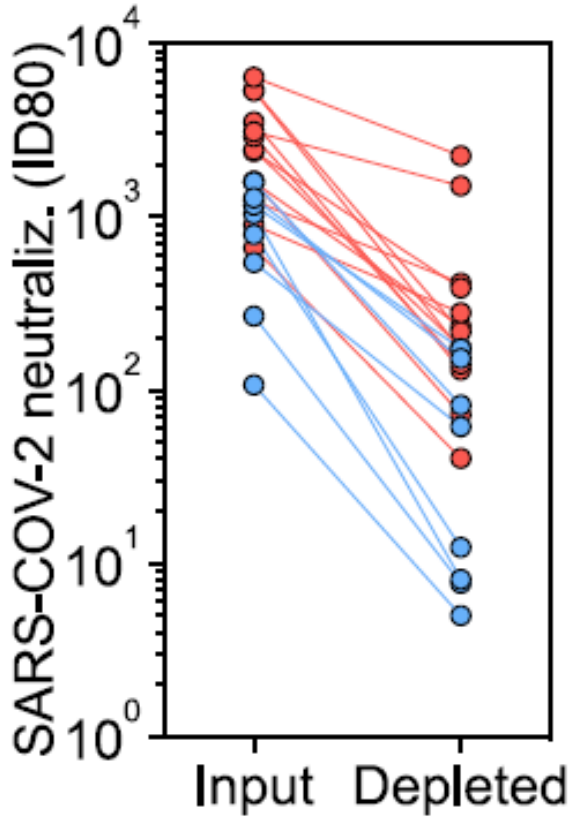


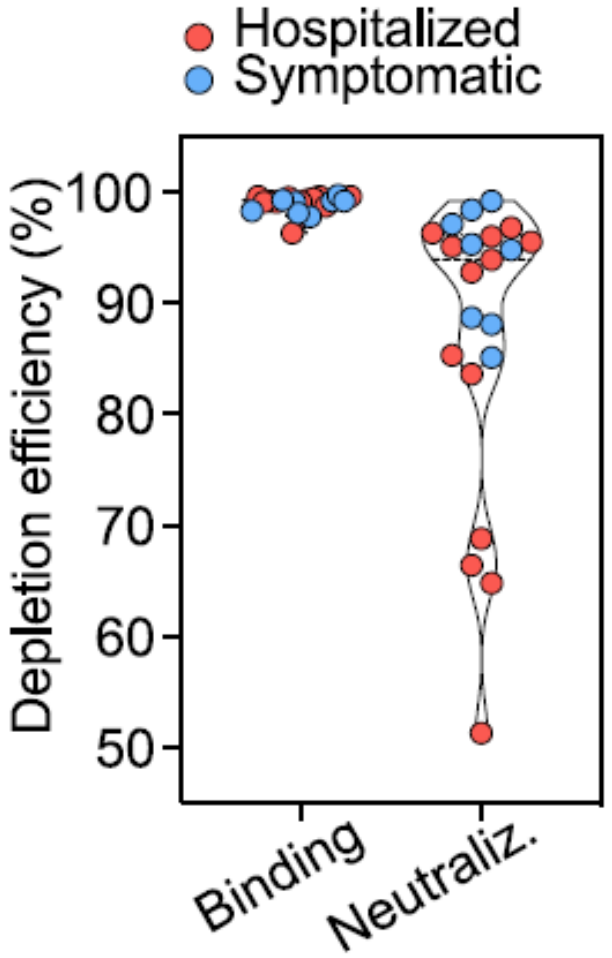
(B. Turoňová et al., Science 10.1126/science.abd5223 (2020).)

- Binding of RBD to ACE2 is cis-regulated by domains other than RBD, so-called 'hip', 'knee', and 'ankle'.
- When using the S-Full of variants as vaccine antigen, mutations in 'hip', 'knee', and 'ankle' may affect the immunogenicity of RBD (may be evolutionally less immunogenic, enabling viral escape from host immune responses).
- In contrast, novel RBD antigens appropriate for emerging variants would be more simply designed and would be predictable.

Critical role of RBD for inducing neutralizing activity





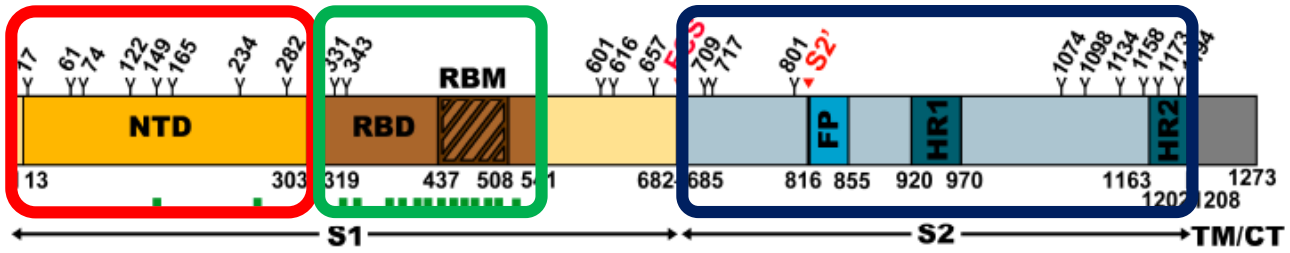


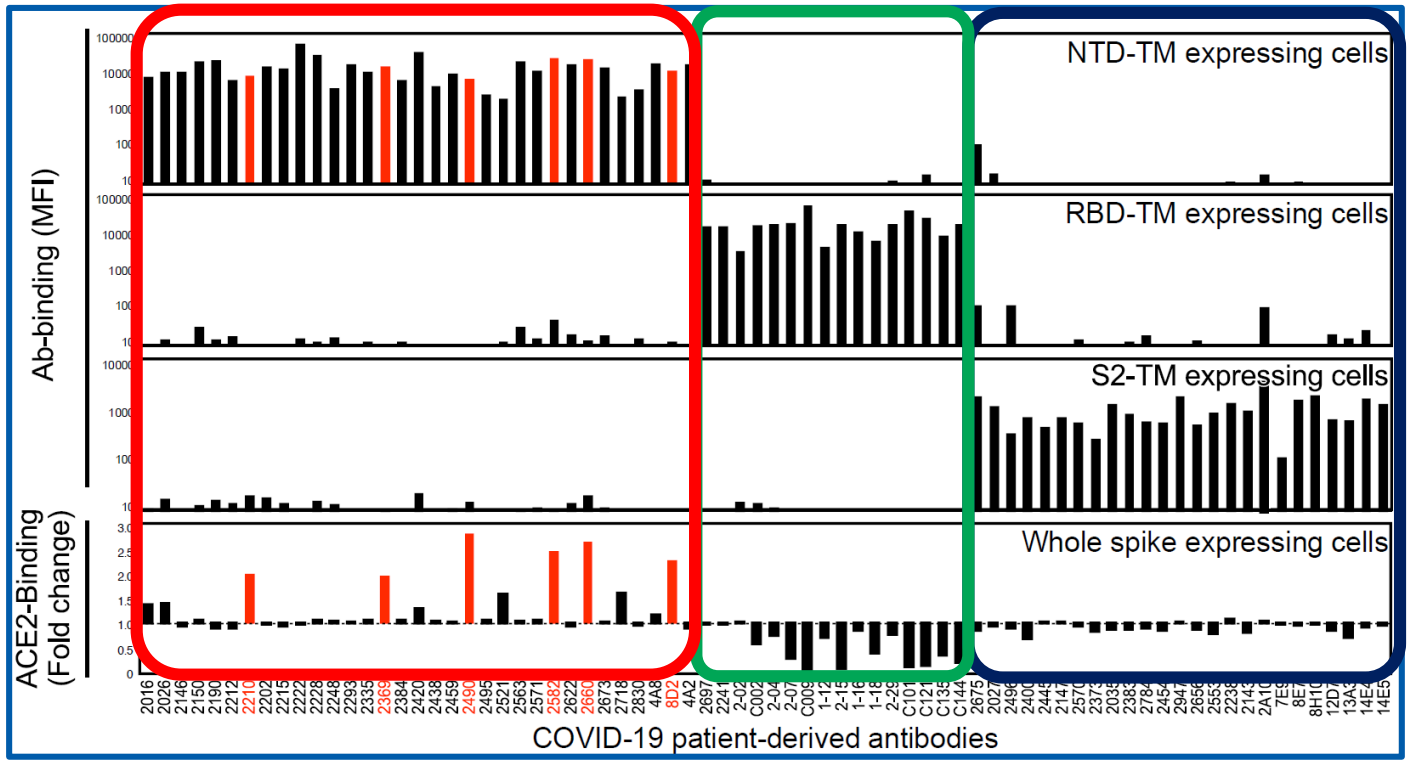
By analyzing more than 650 serum samples from COVID-19 patients, it was suggested that more than 90% of neutralizing antibodies targeted RBD

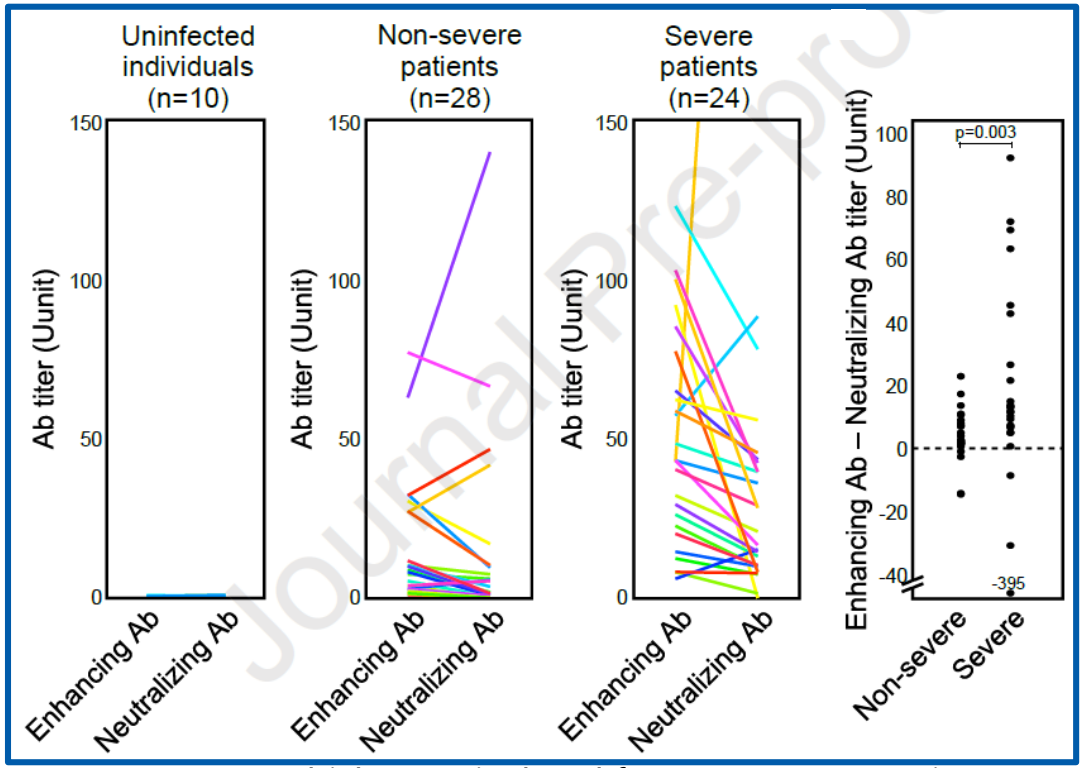
(The diagram shows the result of 21 samples, Cell 183:1024 2020)

Antibodies specific to N-terminal domain of spike protein and immune enhancement

Structure of spike protein





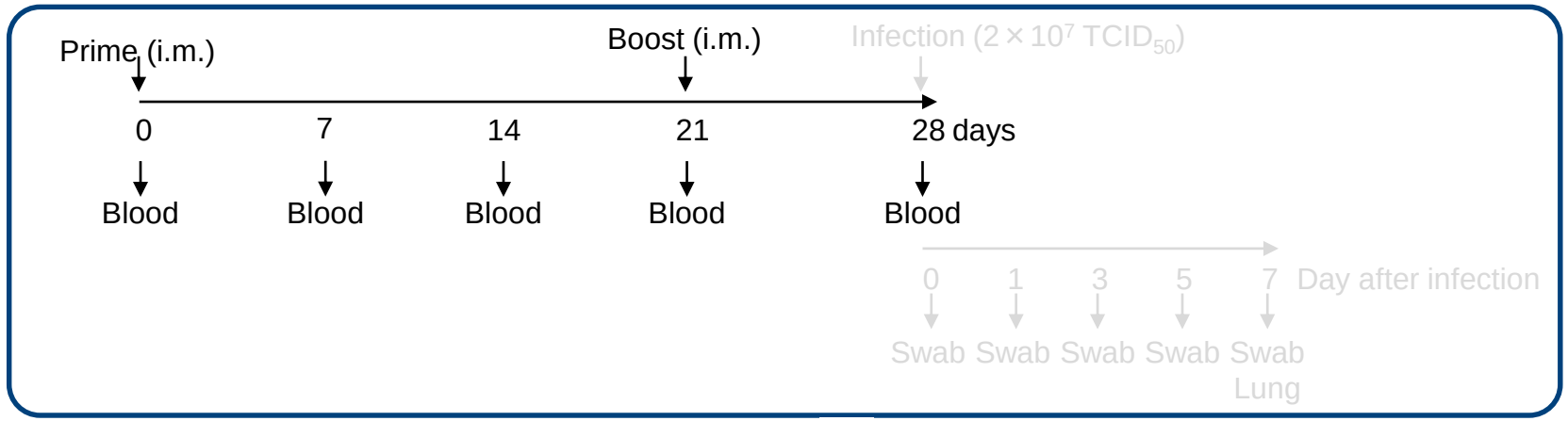


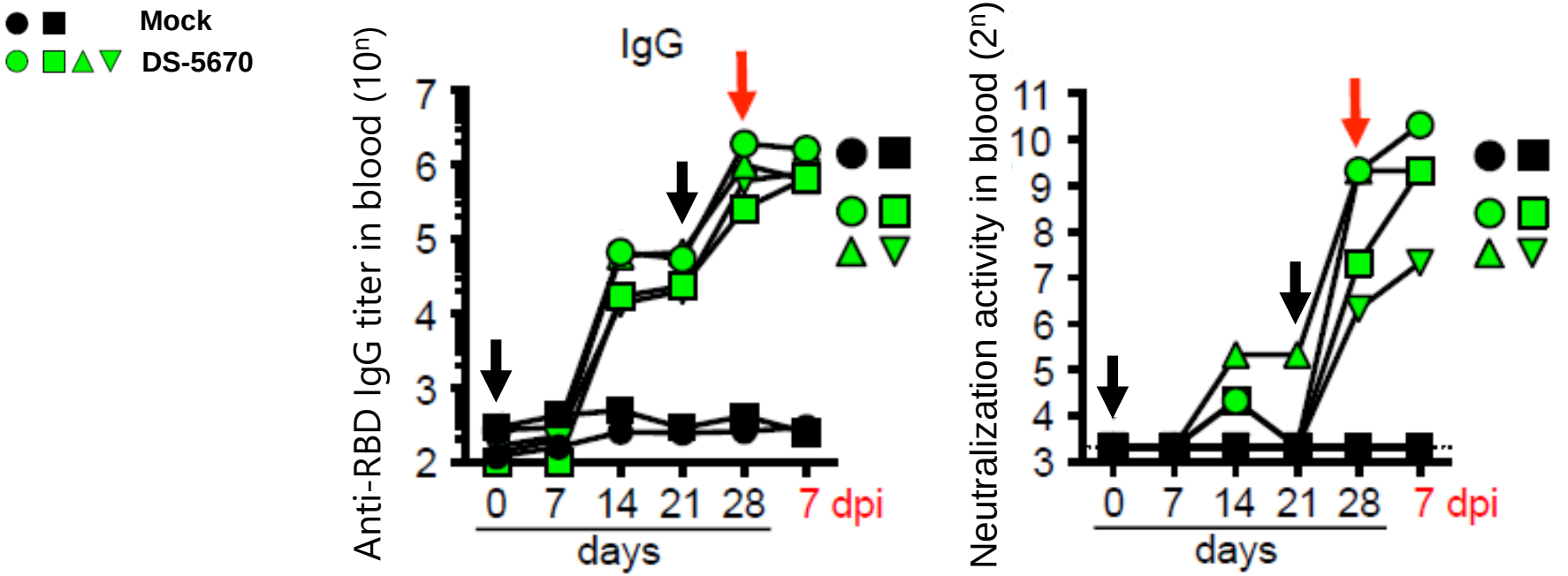
Left: Characterization of monoclonal antibodies specific to different domain in S (NTD, RBD, or S2-TM), which were isolated from COVID-19 patients. The lowest panel shows effects of monoclonal antibodies on S-Full binding to ACE2.
Right: Enhanced and neutralizing antibody titers in serum obtained from COVID-19 patients.

(CELL 12060 https://doi.org/10.1016/j.cell.2021.05.032)

Immunogenicity and protective efficacy of DS-5670 in cynomolgus monkeys (1/3)

Results of research collaboration with the University of Tokyo and Shiga Medical University*

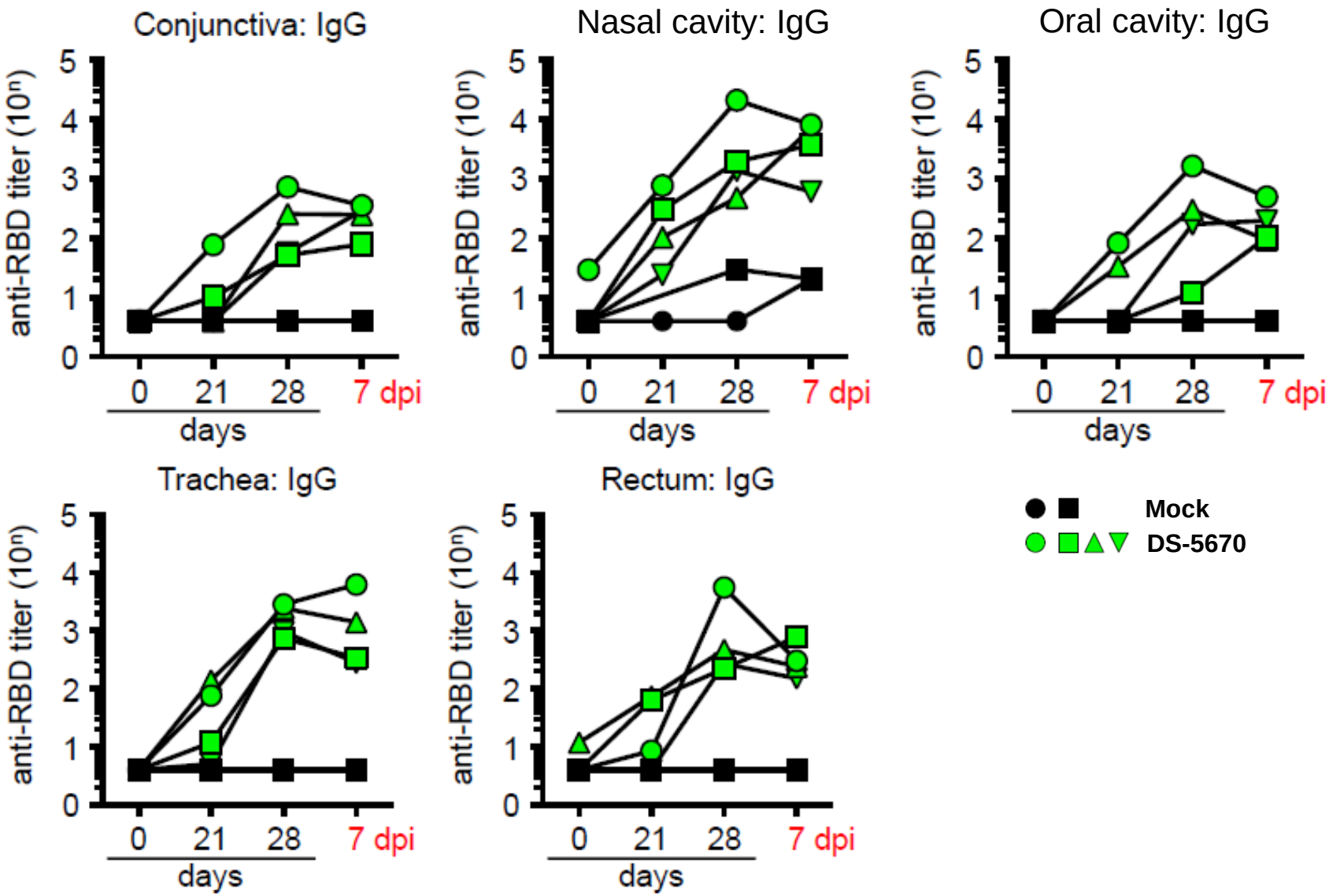




*This data was acquired in the "Fundamental Research on the Control of a Novel Coronavirus (2019-nCoV), which is an initiative supported by the Japan Agency for Medical Research and Development (AMED). (Principal investigator: Prof. Yoshiro Kawaoka, Institute of Medical Sciences, The University of Tokyo

Immunogenicity and protective efficacy of DS-5670 in cynomolgus monkeys (2/3)

Results of research collaboration with the University of Tokyo and Shiga Medical University*



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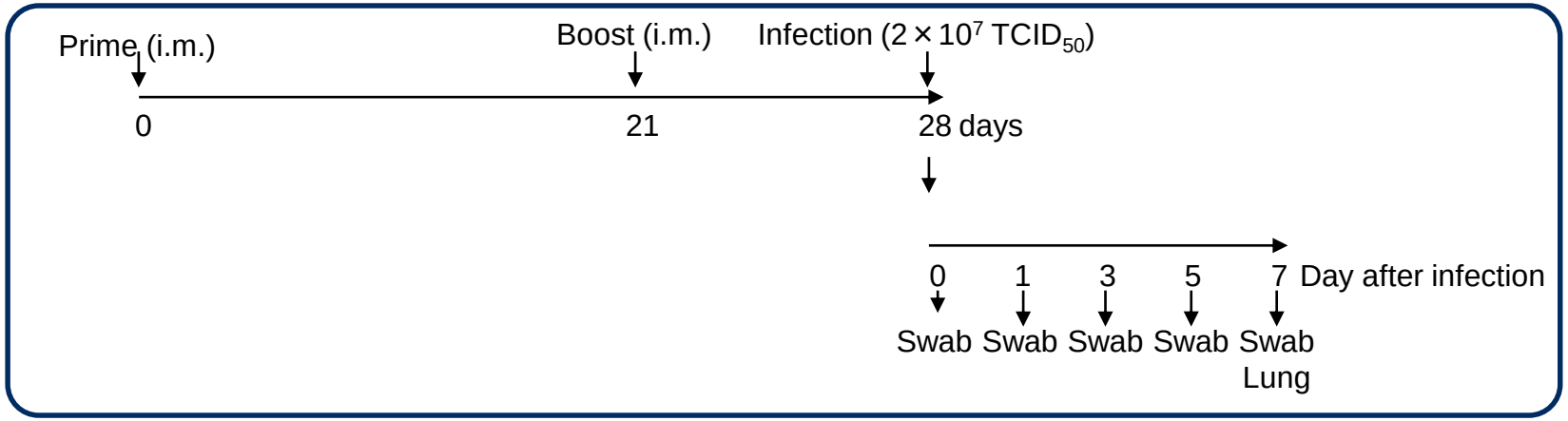
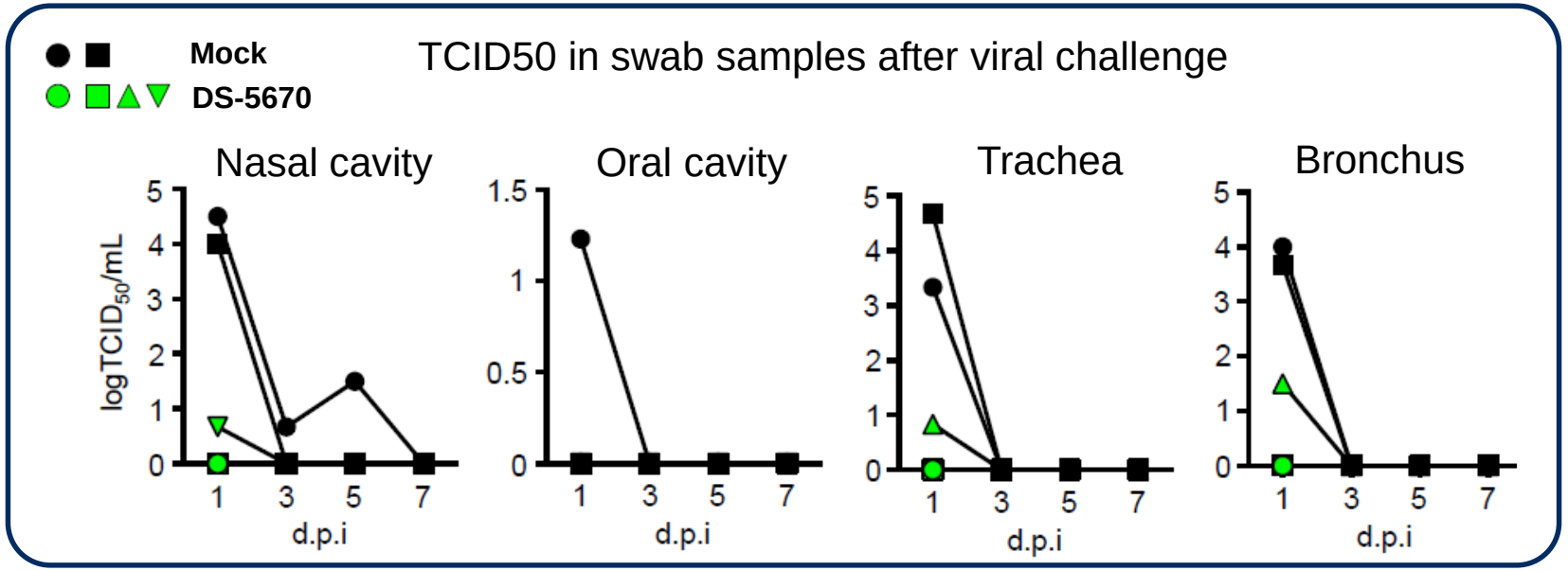
Immunogenicity and protective efficacy of DS-5670 in cynomolgus monkeys (3/3)

Results of research collaboration with the University of Tokyo and Shiga Medical University*





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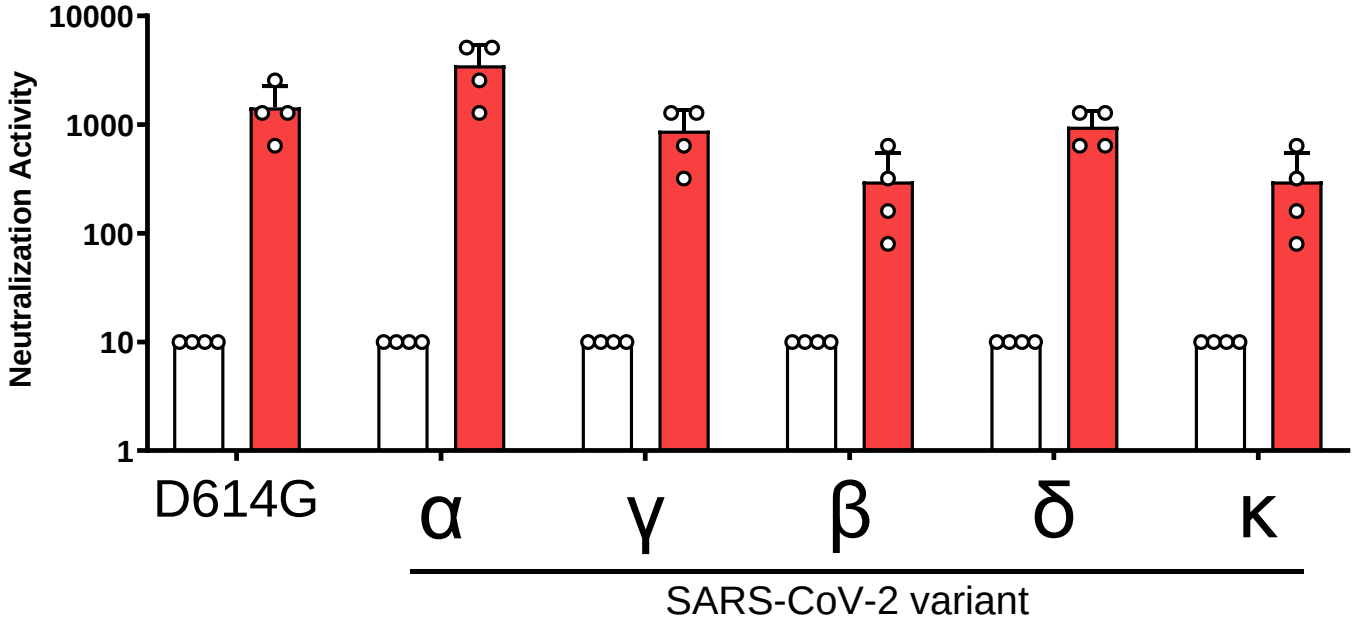
Cross-neutralizing activity against recently emerged variants

- Cynomolgus monkey
- 50 μg/body of DS-5670 by mRNA conversion
- Dosed in brachial deltoid muscle q2w, total 3 times (4 monkeys/group)
- Measured neutralizing activity using plasma collected 2 weeks after the third dose (AMED Kawaoka group)



Variant	Mutation in RBD
α	N501Y
γ	K417T/E484K/N501Y
β	K417N/E484K/N501Y
δ	L452R/T478K
κ	L452R/E484Q

	SARS-CoV-2 variant					
Monkey ID	D614G	α	γ	β	δ	κ
#1	640	2560	640	160	1280	160
#2	2560	5120	1280	640	640	640
#3	1280	5120	1280	320	1280	320
#4	1280	1280	320	80	640	80

*This data was acquired in the AMED's drug discovery support program "Development of a Vaccine for COVID-19 Vaccines".

Current status of DS-5670 development and future plan

- Selected to be a provider for the MHLW's "Emergent Initiative to Build Production Capacity for COVID-19 Vaccines*1 (First Round)"
- Selected to be a company for the AMED's drug discovery support program "Development of a Vaccine for COVID-19 Vaccines*2 (Second Round)"
- Initiated **Ph1/2 study** in March 2021 and data expected around autumn 2021. Currently evaluating the safety, immunogenicity, and recommended dose.
- To initiate **active-controlled, non-inferiority confirmatory study** this year, in which several thousand subjects will be enrolled
- **BLA and commercialization expected in CY2022** when all regulatory requirements are satisfied
- A clinical trial for booster vaccination also being planned and considered
- The overall development plan and designs of further studies being continuously discussed with the Health Authority

*1 The project aims to swiftly develop an actual (large-scale) production system for biologics, including vaccines, in order to ensure that the vaccines necessary for the prevention of the spread and severity of unexpected epidemics, including COVID-19, are produced as soon as possible, and that their supply is secured for the Japanese people.
*2 The project aims to support the development of a vaccine against COVID-19, for which R&D is already underway, and aims to ensure the early commercialization of safe and effective vaccines.