Romainville – 3 dec. 2024

ADEBIOTECH
THINK TANK ONE HEALTH

APPEAL

Antivirus **P**andemic **P**reparedness **E**urope**A**n p**L**atform

Olivier Moncorgé

Institut de Recherche en Infectiologie de Montpellier (IRIM)
CNRS – Université de Montpellier
Caroline Goujon's team & Plateau Technique Virus Respiratoires

cnrs

Université de Montpellier

IRIm Institut de Recherche en Infectiologie de Montpellier

www.project-appeal.eu

Funded by
the European Union

The APPEAL consortium

ADEBIOTECH
THINK TANK ONE HEALTH

APPEAL
Antivirus Pandemic Preparedness EuropeAn pLatform

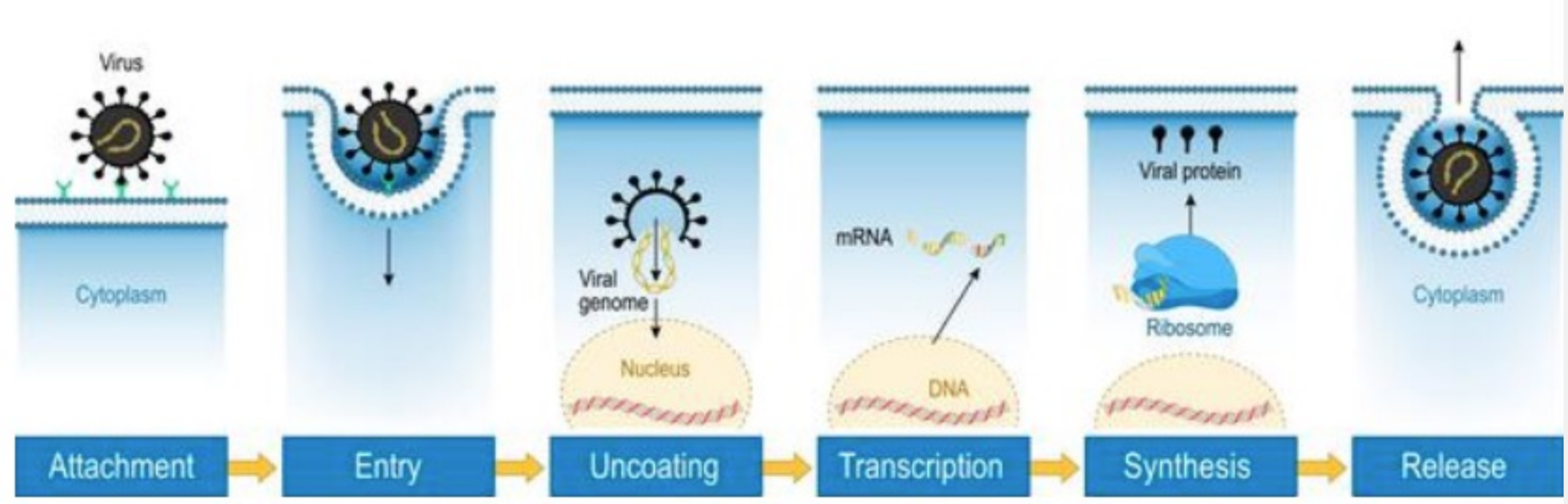
13 partners - 7 countries – 5 year program

UNIVERSITÄTS KLINIKUM jena	GOETHE UNIVERSITÄT FRANKFURT AM MAIN	UNIVERSITÄT HEIDELBERG ZUKUNFT SEIT 1386
Universitätsmedizin GREIFSWALD	cnrs IRIM Institut de Recherche en Infectiologie de Montpellier	erinha European Research Infrastructure on Highly Pathogenic Agents
INSTITUT PASTEUR	ecrin	MATTEK A BICO COMPANY
Welab BARCELONA	ConsulTech.	IMPERIAL
	Erasmus MC	

APPEAL: to identify broad spectrum host-directed antiviral drugs

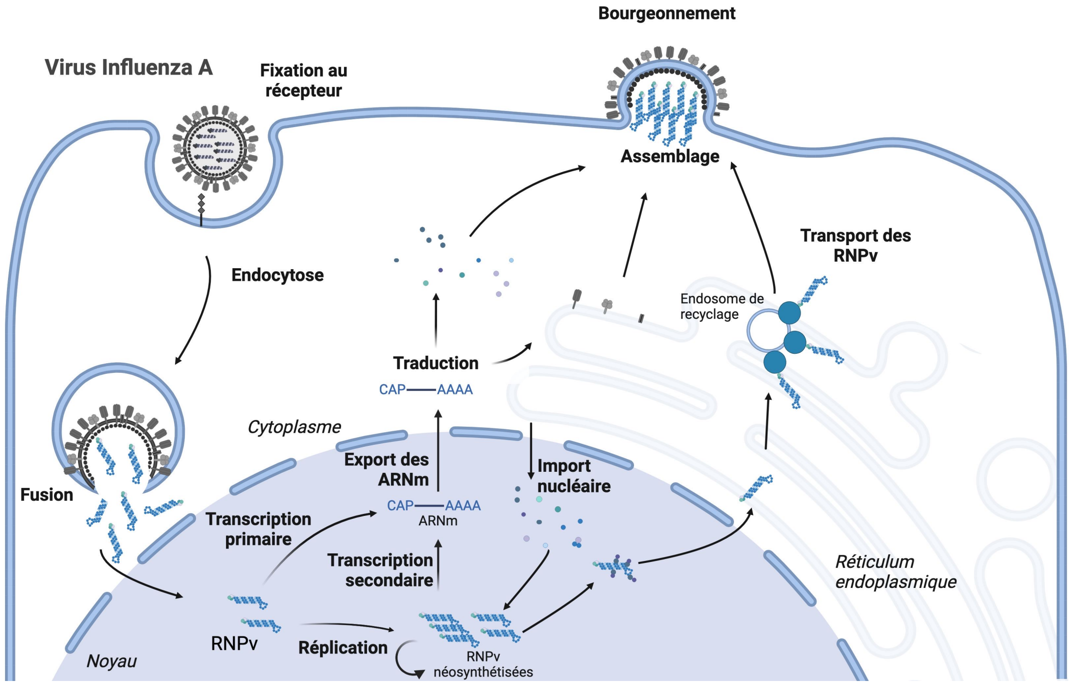
Funded by
the European Union

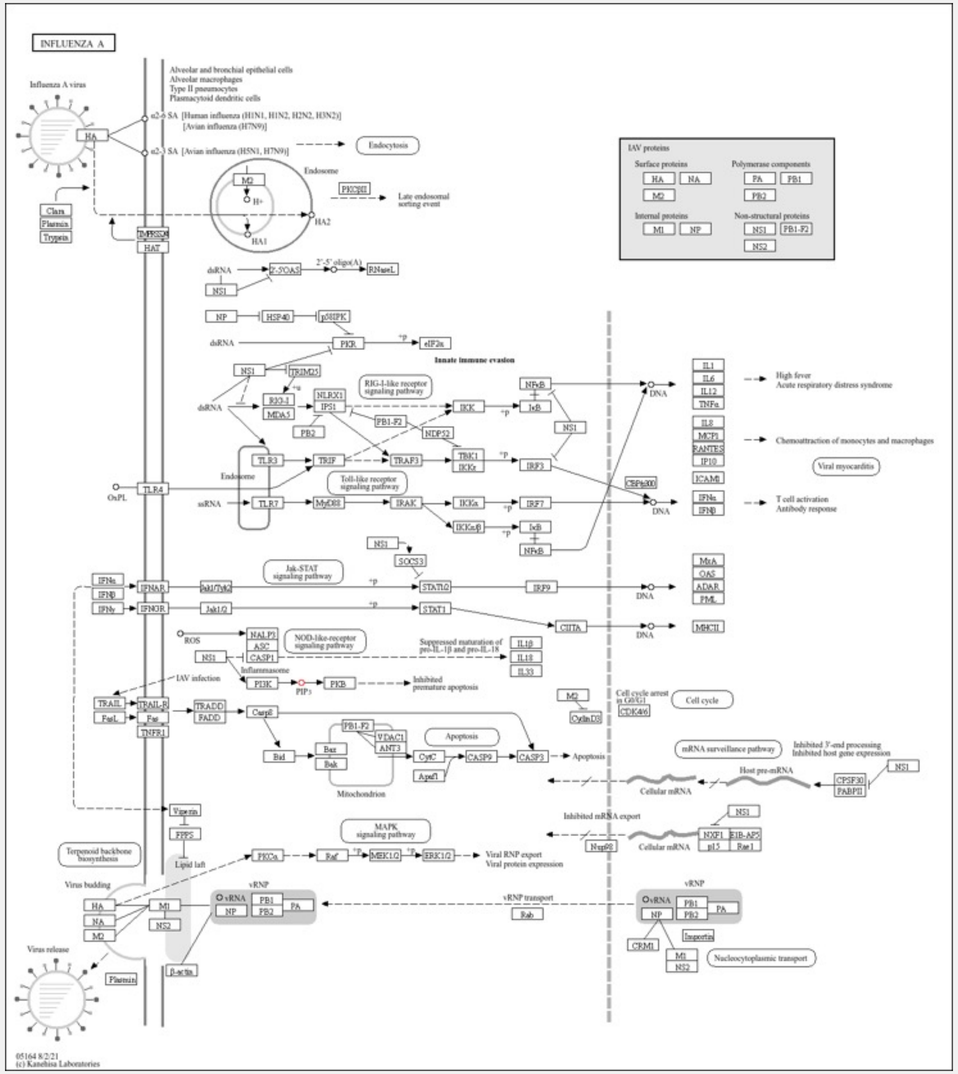
Viruses are obligatory intracellular parasites



Viruses are obligatory intracellular parasites

Influenza virus

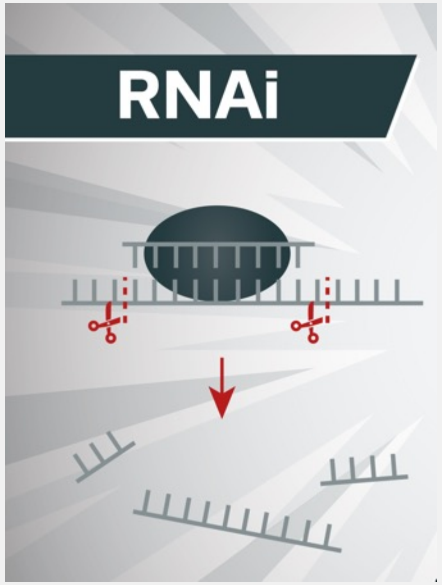


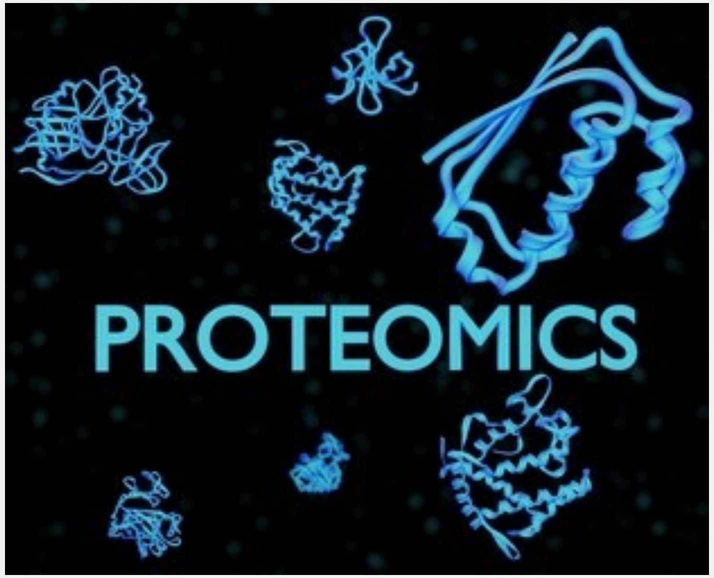


APPEAL: a better knowledge of virus/host cell relations to identify key host factors

Early 21st century: the OMICS era

Since 2000: Multi-omics and genome-wide studies





…

OMICS applied to the study of virus/host cell interactions

i.e.: RNAi screens and influenza virus to unravel host-dependency factors (HDRs) involved in viral replication

- Brass et al. Cell 2009 [21]
- Hao et al. Nature 2008 [22]
- Karlas et al. Nature 2010 [23]
- König et al. Nature 2010 [24]
- Shapira et al. Cell 2009 [25]

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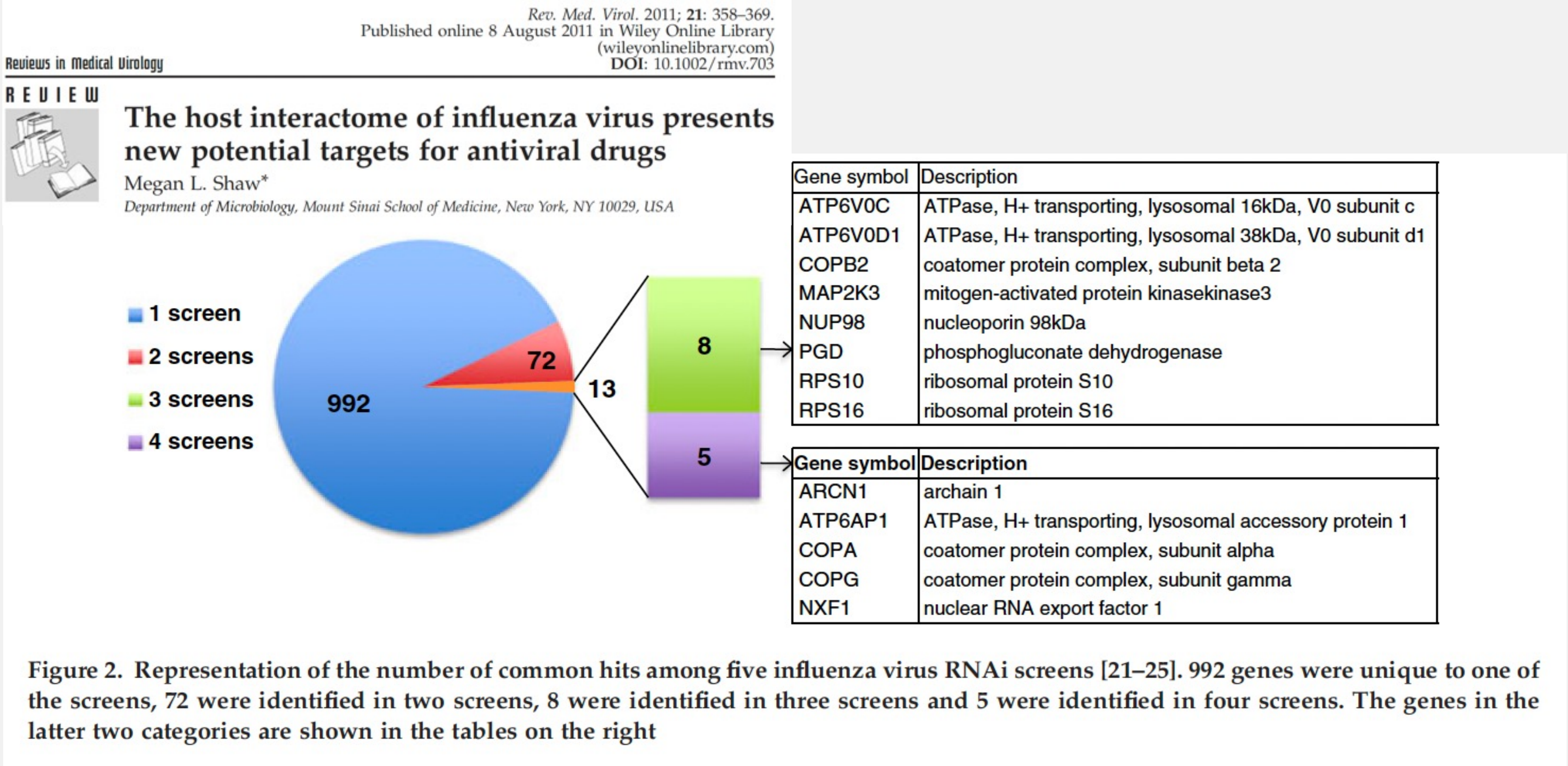


Rev. Med. Virol. 2011; **21**: 358–369.
Published online 8 August 2011 in Wiley Online Library
(wileyonlinelibrary.com)
DOI: 10.1002/rmv.703

Reviews in Medical Virology

REVIEW

The host interactome of influenza virus presents new potential targets for antiviral drugs

Megan L. Shaw*
Department of Microbiology, Mount Sinai School of Medicine, New York, NY 10029, USA

Gene symbol	Description
ATP6V0C	ATPase, H+ transporting, lysosomal 16kDa, V0 subunit c
ATP6V0D1	ATPase, H+ transporting, lysosomal 38kDa, V0 subunit d1
COPB2	coatomer protein complex, subunit beta 2
MAP2K3	mitogen-activated protein kinasekinase3
NUP98	nucleoporin 98kDa
PGD	phosphogluconate dehydrogenase
RPS10	ribosomal protein S10
RPS16	ribosomal protein S16

Gene symbol	Description
ARCN1	archain 1
ATP6AP1	ATPase, H+ transporting, lysosomal accessory protein 1
COPA	coatomer protein complex, subunit alpha
COPG	coatomer protein complex, subunit gamma
NXF1	nuclear RNA export factor 1

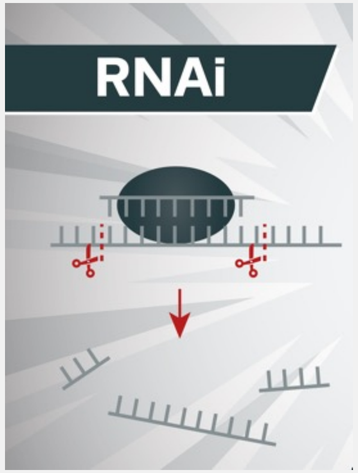
Figure 2. Representation of the number of common hits among five influenza virus RNAi screens [21–25]. 992 genes were unique to one of the screens, 72 were identified in two screens, 8 were identified in three screens and 5 were identified in four screens. The genes in the latter two categories are shown in the tables on the right

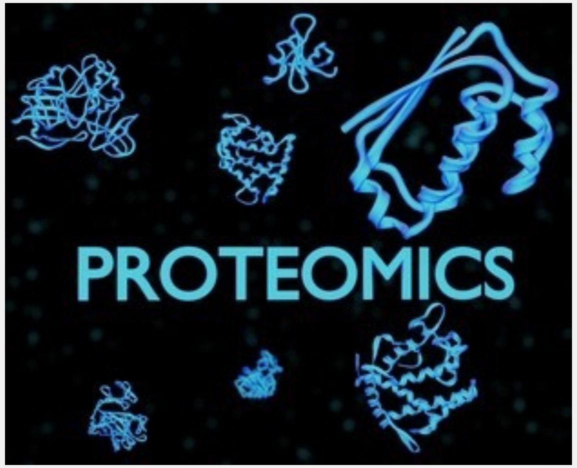
… not a single gene was commun to all 5 studies!

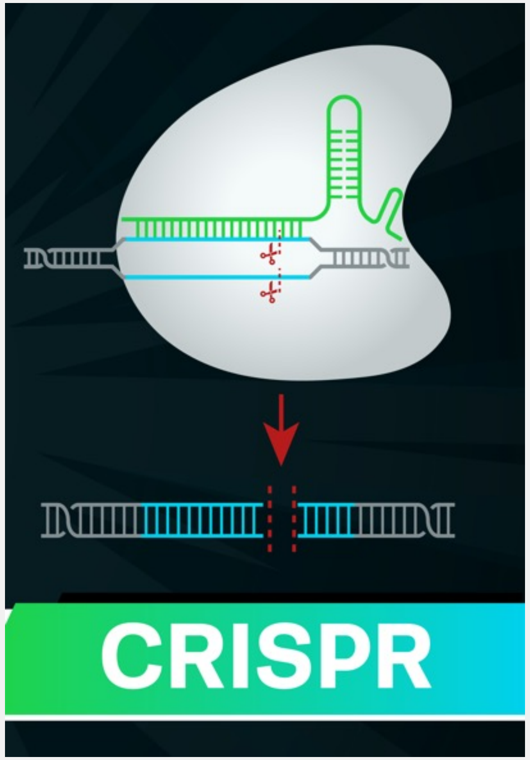
Early 2010: the CRISPR revolution

2000

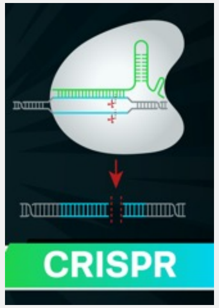
2012: CRISPR
(Clustered Regularly Interspaced Short Palindromic Repeats)



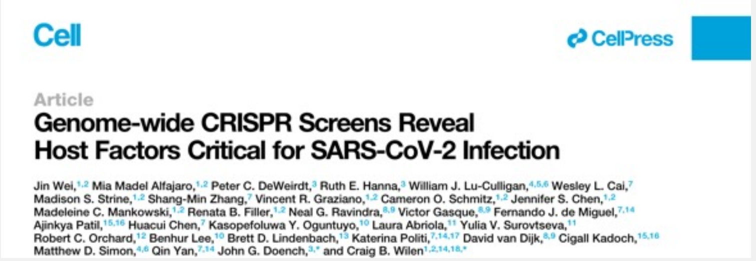




Whole genome CRISPR screens and SARS-CoV-2



Vero E6 (online 20/10/2020)


Cell

CellPress

Article

Genome-wide CRISPR Screens Reveal Host Factors Critical for SARS-CoV-2 Infection

Jin Wei,[1,2] Mia Madel Alfajaro,[1,2] Peter C. DeWeirdt,[3] Ruth E. Hanna,[3] William J. Lu-Culligan,[4,5,6] Wesley L. Cai,[7] Madison S. Strine,[1,2] Shang-Min Zhang,[7] Vincent R. Graziano,[1,2] Cameron O. Schmitz,[1,2] Jennifer S. Chen,[1,2] Madeleine C. Mankowski,[1,2] Renata B. Filler,[1,2] Neal G. Ravindra,[8,9] Victor Gasque,[8,9] Fernando J. de Miguel,[7,14] Ajinkya Patil,[15,16] Huacui Chen,[7] Kasopefoluwa Y. Oguntuyo,[10] Laura Abriola,[11] Yulia V. Surovtseva,[11] Robert C. Orchard,[12] Benhur Lee,[10] Brett D. Lindenbach,[13] Katerina Politi,[7,14,17] David van Dijk,[8,9] Cigall Kadoch,[15,16] Matthew D. Simon,[4,6] Qin Yan,[7,14] John G. Doench,[3,*] and Craig B. Wilen[1,2,14,18,*]

A549-ACE2 (online 24/10/2020)


Cell

CellPress

Article

Identification of Required Host Factors for SARS-CoV-2 Infection in Human Cells

Zharko Daniloski,[1,2,7] Tristan X. Jordan,[3,7] Hans-Hermann Wessels,[1,2] Daisy A. Hoagland,[3] Silva Kasela,[1,4] Mateusz Legut,[1,2] Silas Maniatis,[1] Eleni P. Mimitou,[5] Lu Lu,[1,2] Evan Geller,[1,2] Oded Danziger,[3] Brad R. Rosenberg,[3] Hemali Phatnani,[1,6] Peter Smibert,[5] Tuuli Lappalainen,[1,4] Benjamin R. tenOever,[3,*] and Neville E. Sanjana[1,2,8,*]

Huh-7.5 (online 09/12/2020)


CellPress

Cell

Article

Genome-Scale Identification of SARS-CoV-2 and Pan-coronavirus Host Factor Networks

William M. Schneider,[1,5] Joseph M. Luna,[1,5] H.-Heinrich Hoffmann,[1,5] Francisco J. Sánchez-Rivera,[2,5] Andrew A. Leal,[3,6] Alison W. Ashbrook,[1,6] Jérémie Le Pen,[1,6] Inna Ricardo-Lax,[1] Eleftherios Michailidis,[1] Avery Peace,[1] Ansgar F. Stenzel,[1,4] Scott W. Lowe,[2] Margaret R. MacDonald,[1] Charles M. Rice,[1,*] and John T. Poirier[3,7,*]

Huh-7.5.1-ACE2-TMPRSS2 (online 09/12/2020)

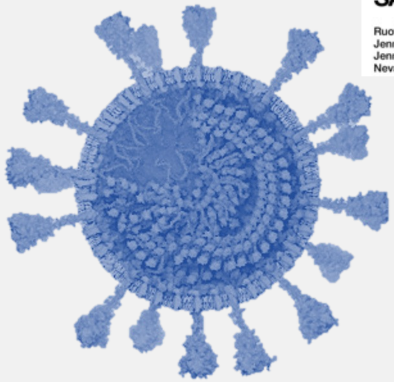

CellPress

Cell

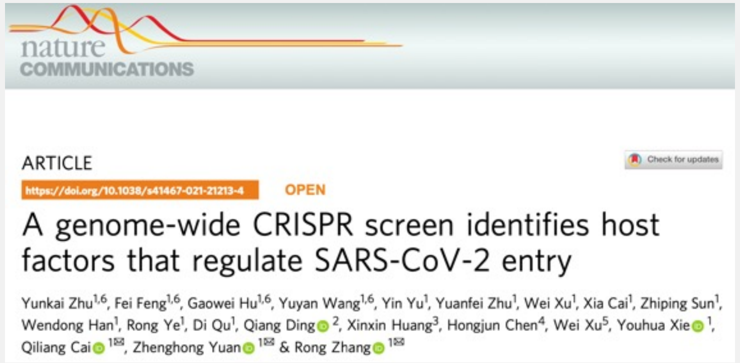
Article

Genetic Screens Identify Host Factors for SARS-CoV-2 and Common Cold Coronaviruses

Ruofan Wang,[1,13] Camille R. Simoneau,[2,3,4,5,13] Jessie Kulsuptrakul,[1] Mehdi Bouhaddou,[2,4,6,7] Katherine A. Travisano,[1] Jennifer M. Hayashi,[2,3,4] Jared Carlson-Stevermer,[8] James R. Zengel,[9] Christopher M. Richards,[9] Parinaz Fozouni,[2,3,4,5,10] Jennifer Oki,[8] Lauren Rodriguez,[11] Bastian Joehnk,[12] Keith Walcott,[12] Kevin Holden,[8] Anita Sil,[12] Jan E. Carette,[9] Nevan J. Krogan,[2,4,6,7] Melanie Ott,[2,3,4,*] and Andreas S. Puschnik[1,14,*]



A549-ACE2 (online 11/02/2021)


nature COMMUNICATIONS

ARTICLE

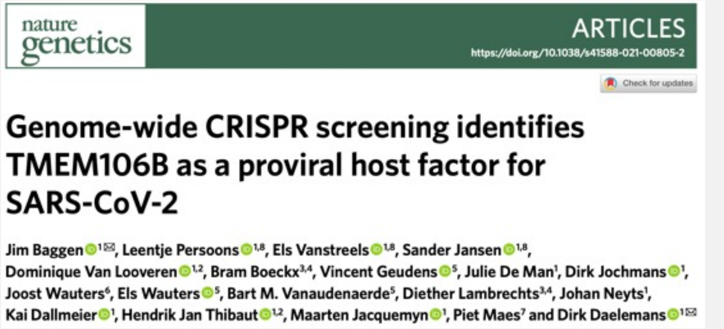
Check for updates

https://doi.org/10.1038/s41467-021-21213-4 OPEN

A genome-wide CRISPR screen identifies host factors that regulate SARS-CoV-2 entry

Yunkai Zhu[1,6], Fei Feng[1,6], Gaowei Hu[1,6], Yuyan Wang[1,6], Yin Yu[1], Yuanfei Zhu[1], Wei Xu[1], Xia Cai[1], Zhiping Sun[1], Wendong Han[1], Rong Ye[1], Di Qu[1], Qiang Ding[2], Xinxin Huang[3], Hongjun Chen[4], Wei Xu[5], Youhua Xie[1], Qiliang Cai[1✉], Zhenghong Yuan[1✉] & Rong Zhang[1✉]

Huh7 (online 08/03/2021)


nature genetics

ARTICLES

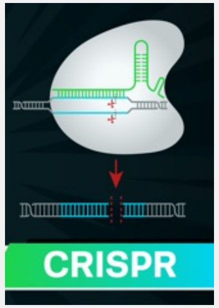
https://doi.org/10.1038/s41588-021-00805-2

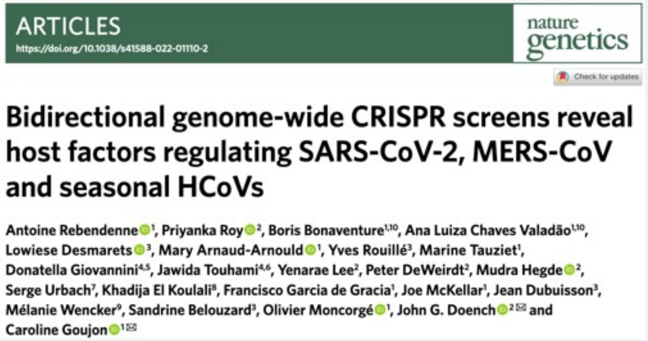
Check for updates

Genome-wide CRISPR screening identifies TMEM106B as a proviral host factor for SARS-CoV-2

Jim Baggen[1✉], Leentje Persoons[1,8], Els Vanstreels[1,8], Sander Jansen[1,8], Dominique Van Looveren[1,2], Bram Boeckx[3,4], Vincent Geudens[5], Julie De Man[1], Dirk Jochmans[1], Joost Wauters[6], Els Wauters[5], Bart M. Vanaudenaerde[5], Diether Lambrechts[3,4], Johan Neyts[1], Kai Dallmeier[1], Hendrik Jan Thibaut[1,2], Maarten Jacquemyn[1], Piet Maes[7] and Dirk Daelemans[1✉]

Whole genome CRISPR screens and SARS-CoV-2




ARTICLES
https://doi.org/10.1038/s41588-022-01110-2
nature genetics
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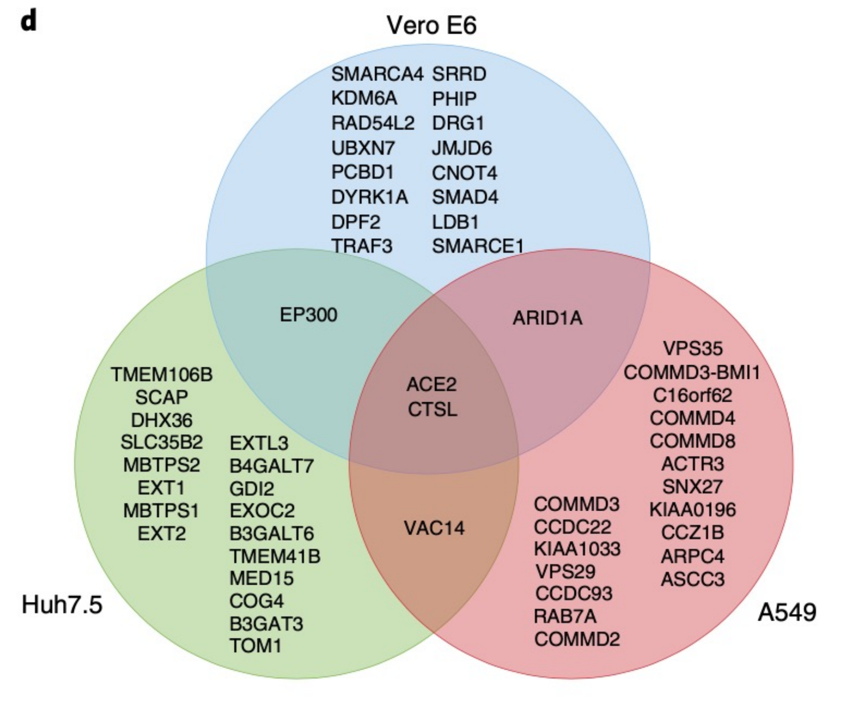
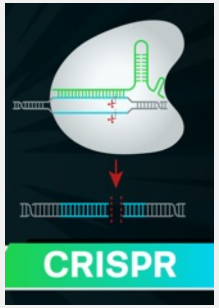
Bidirectional genome-wide CRISPR screens reveal host factors regulating SARS-CoV-2, MERS-CoV and seasonal HCoVs

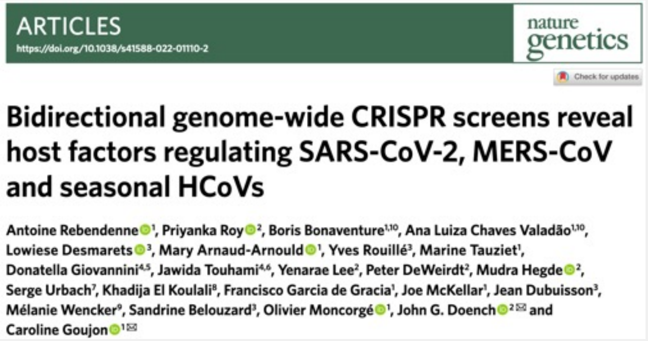
Antoine Rebendenne[1], Priyanka Roy[2], Boris Bonaventure[1,10], Ana Luiza Chaves Valadão[1,10], Lowiese Desmarets[3], Mary Arnaud-Arnould[1], Yves Rouillé[3], Marine Tauziet[1], Donatella Giovannini[4,5], Jawida Touhami[4,6], Yenarae Lee[2], Peter DeWeirdt[2], Mudra Hegde[2], Serge Urbach[7], Khadija El Koulali[8], Francisco Garcia de Gracia[1], Joe McKellar[1], Jean Dubuisson[3], Mélanie Wencker[9], Sandrine Belouzard[3], Olivier Moncorgé[1], John G. Doench[2] ✉ and Caroline Goujon[1] ✉

Priyanka Roy



Whole genome CRISPR screens and SARS-CoV-2





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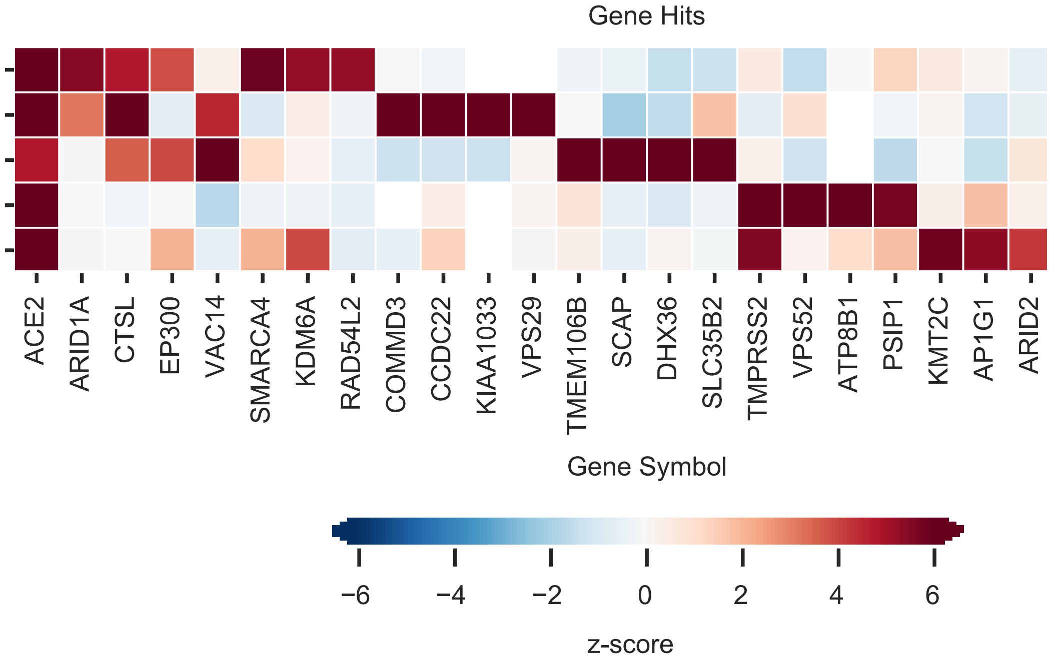
Caroline Goujon

Antoine Rebendenne

Priyanka Roy

	Cell lines	**Origin**
Wilen / **our study**	Vero E6 average	Kidney (monkey)
Sanjana / Zhang	A549 average	**Lung**
Poirier / Puschnik / Daelemans	Huh7 average	Liver
Our study	Caco-2	Intestine
	Calu-3 KO	**Lung**



Meta-analysis done using:
Wei et al. Cell 2021
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and **our data**

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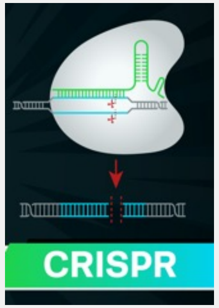

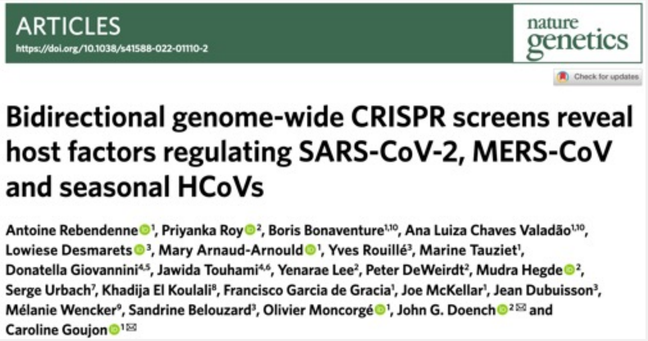

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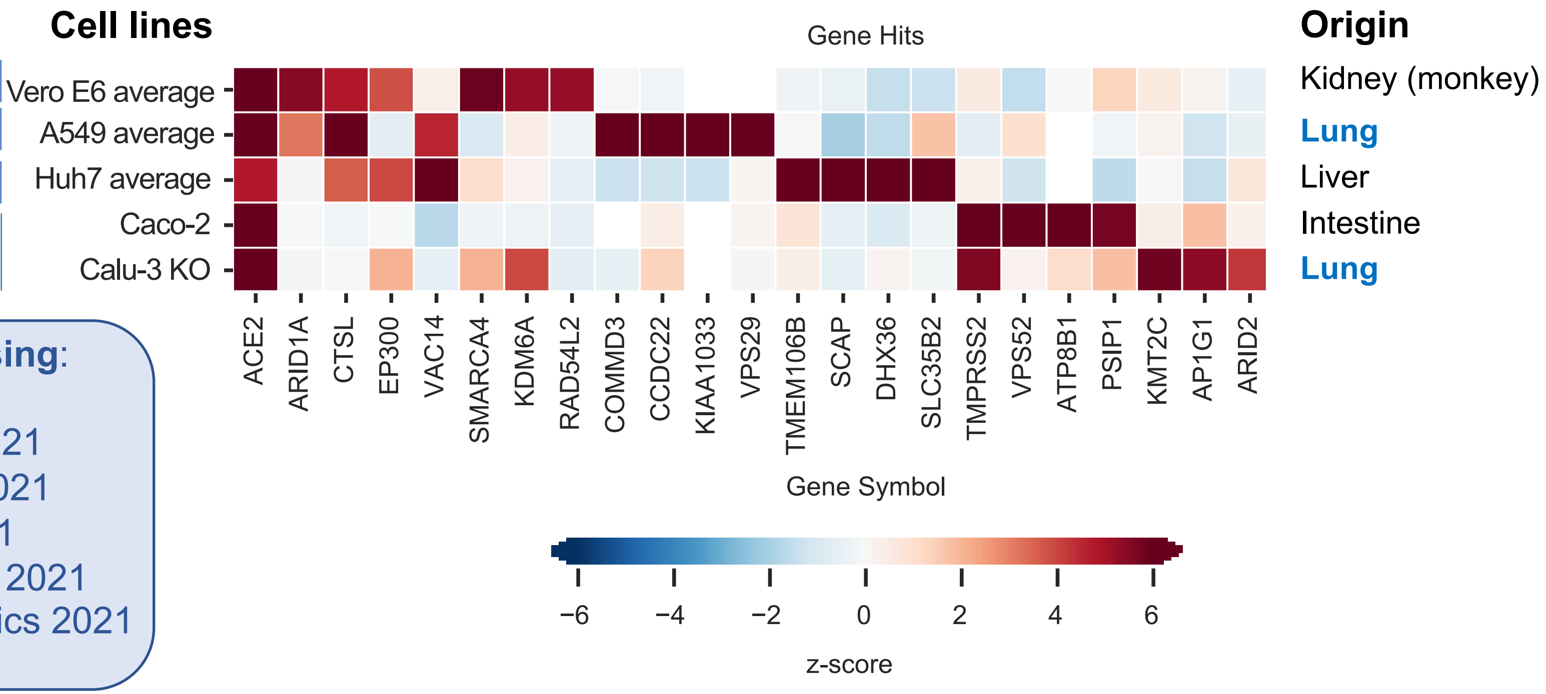
Caroline Goujon

Antoine Rebendenne

Priyanka Roy

Wilen / **our study**
Sanjana / Zhang
Poirier / Puschnik / Daelemans
Our study



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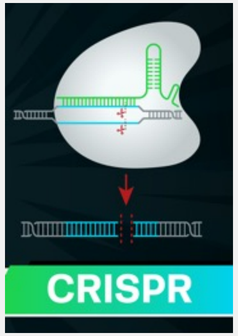
Baggen et al. Nature Genetics 2021
and **our data**

The cellular model is the primary source of variability

Whole genome CRISPR screens and SARS-CoV-2



-The hits identified in CRISPR genome-wide screens aiming at defining the landscape of genes modulating SARS-CoV-2 replication are highly dependent on the cell line used

- Scientific relevance of certain factors?

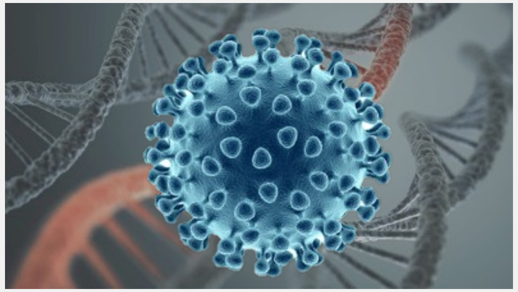
- Cost! Between $100,000 and $500,000 for a scientific publication!

- This flaw is not only true for CRISPR or siRNA screens

The OMICS era (in cell lines): good or bad?

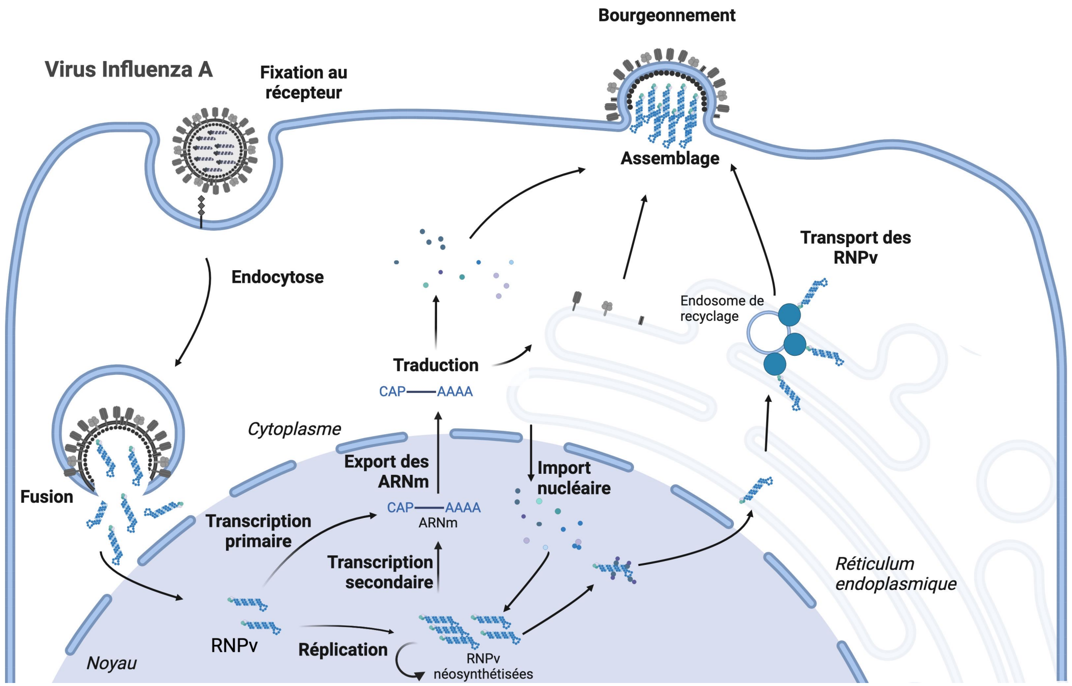
- **Transcriptomics**: Analysis of the transcriptome (all RNA transcripts)
- **Proteomics**: Examination of the proteome (all proteins)
- **Metabolomics**: Study of the metabolome (all metabolites)
- **Epigenomics**: Study of epigenetic modifications
- **Lipidomics**: Analysis of cellular lipids
- **Glycomics**: Study of the complete set of glycans
- **Interactomics**: Analysis of molecular interactions
- **Ubiquitinomics**: Examination of ubiquitin modifications
- **…**

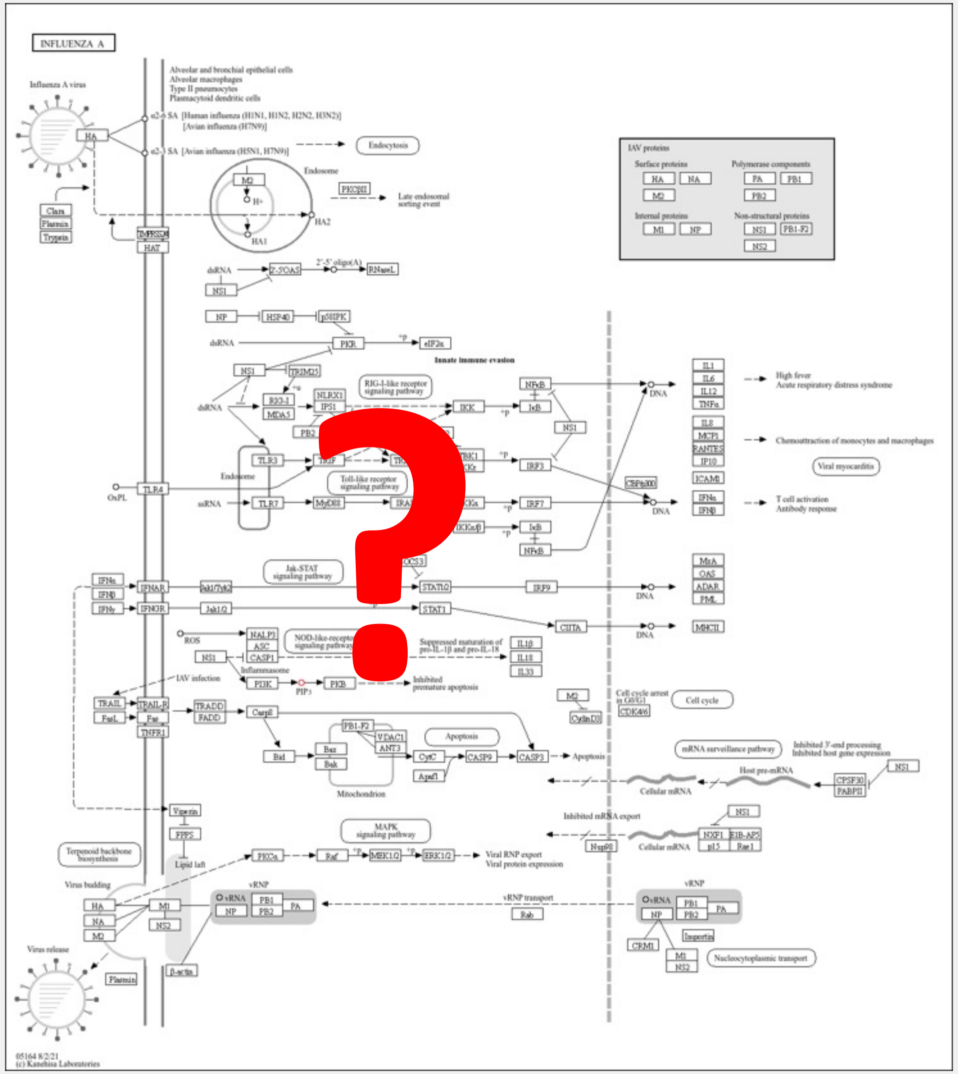
- **Multi-omics**: Integration of multiple omics data types



The OMICS era (in cell lines): good or bad?

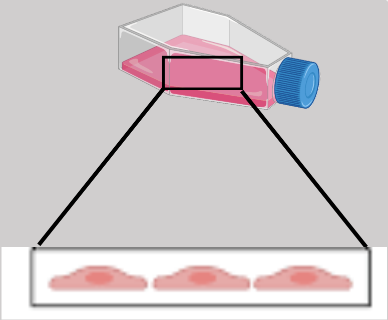
Influenza virus



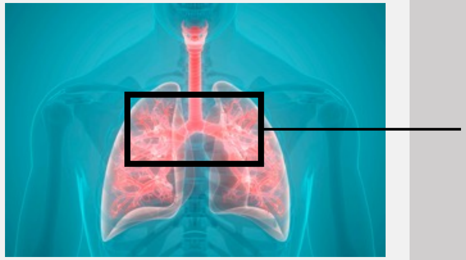


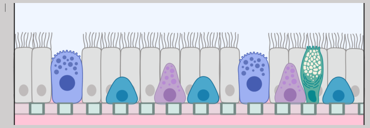
What are the real key cellular factors?

Need for more relevant cellular models



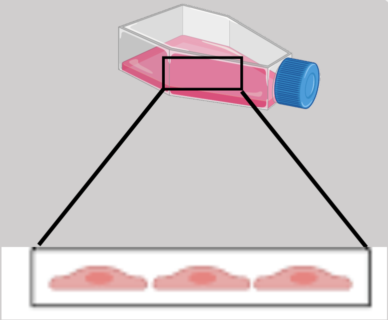
Cancer cell lines
(2D culture)





Primary epithelium
(3D culture)
Complex architecture, true target of respiratory viruses

Need for more relevant cellular models



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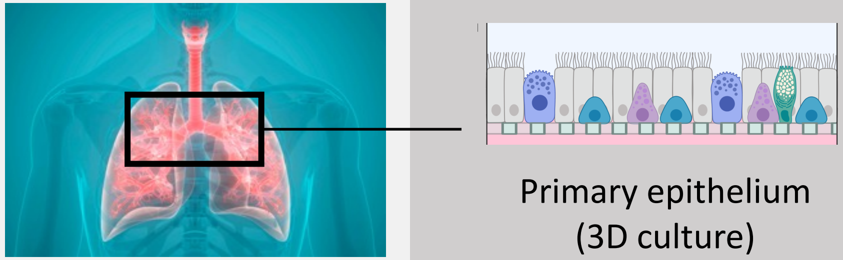

Primary epithelium
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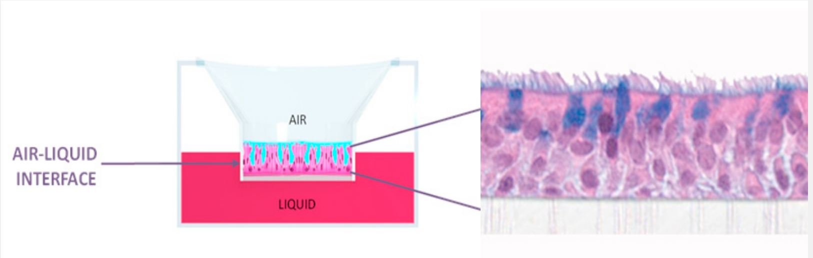
Complex architecture, true target of respiratory viruses

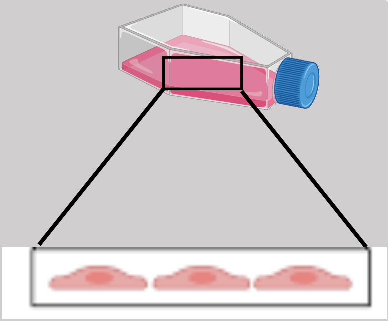
ALI (Air Liquid Interface) cultures



EPITHELIX

MATTEK
A BICO COMPANY

Need for more relevant cellular models



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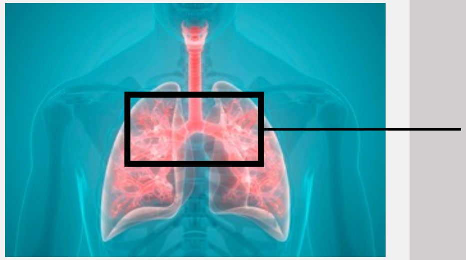

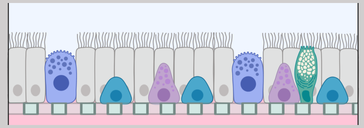

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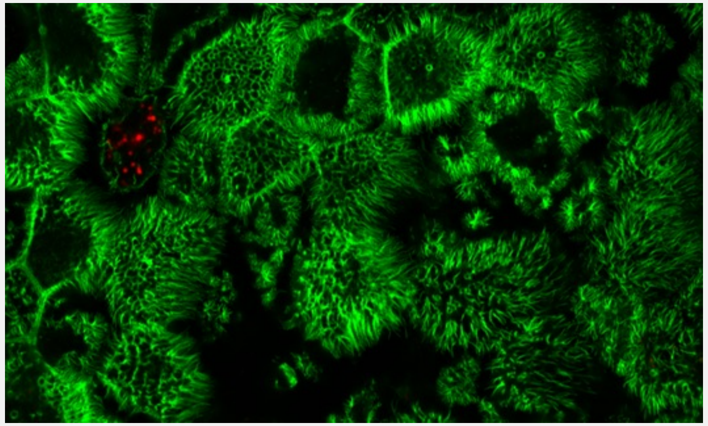
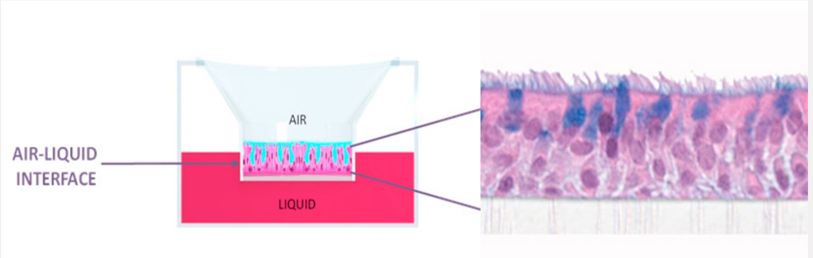
(3D culture)
Complex architecture, true target of respiratory viruses



SARS-CoV-2 / Actin

J. McKellar – C. Goujon Lab

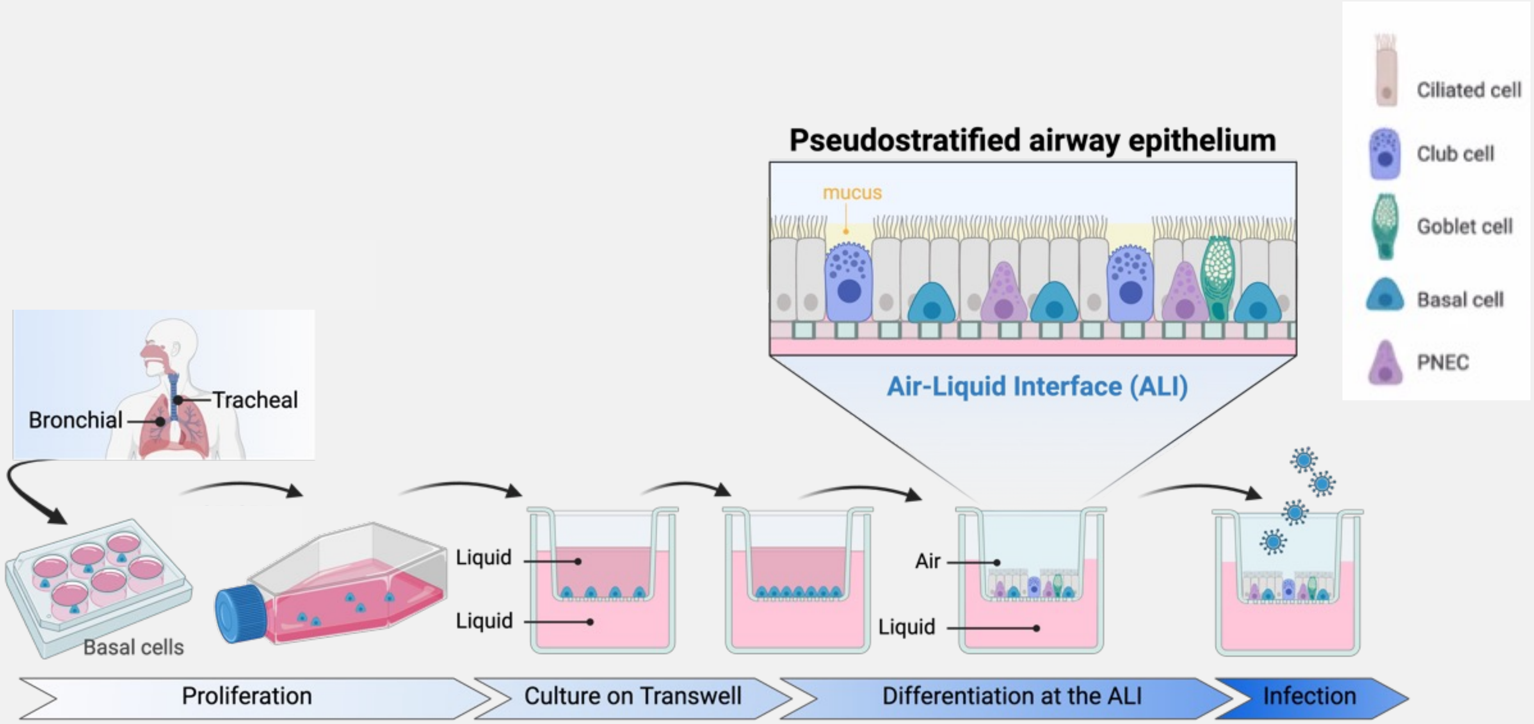
ALI (Air Liquid Interface) cultures



EPITHELIX

MATTEK
A BICO COMPANY

Need for more relevant cellular models

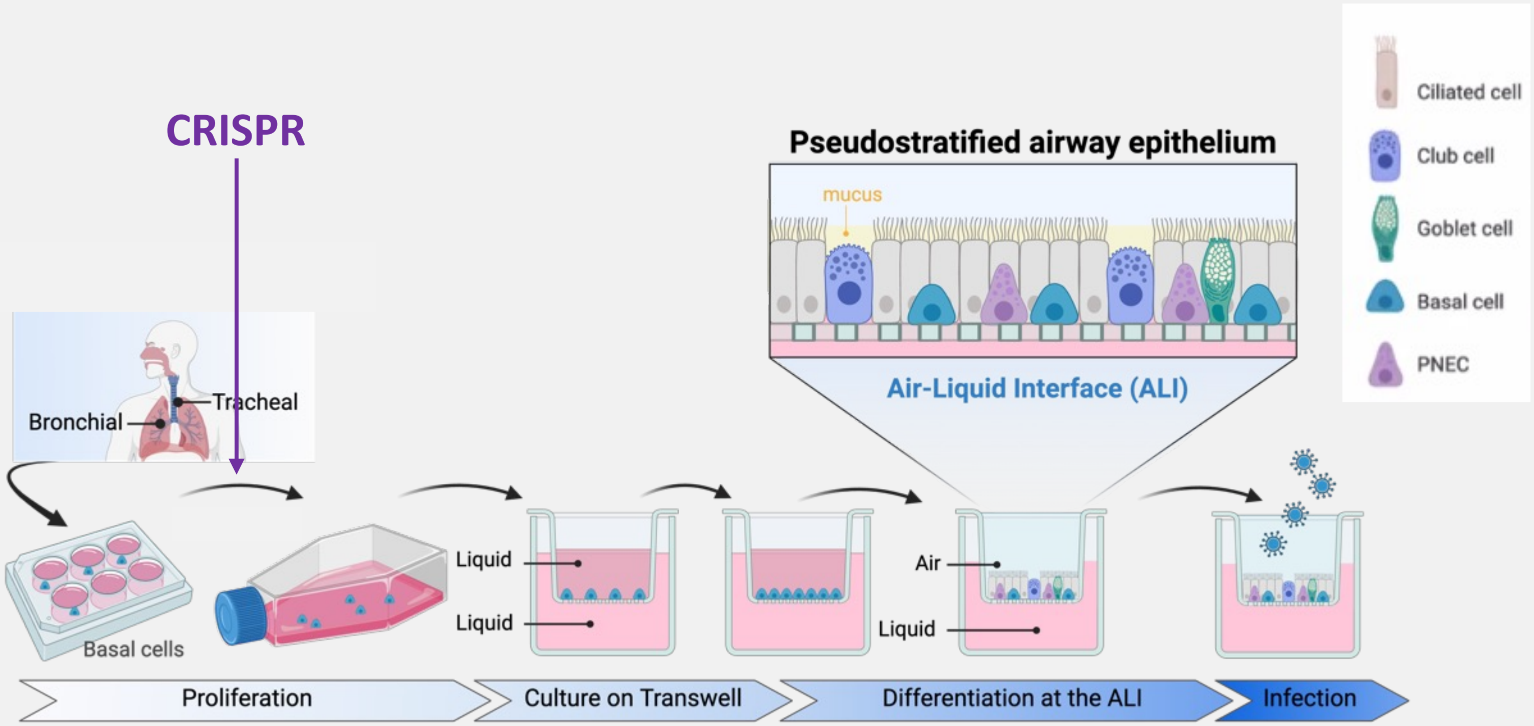


5 days

21 days

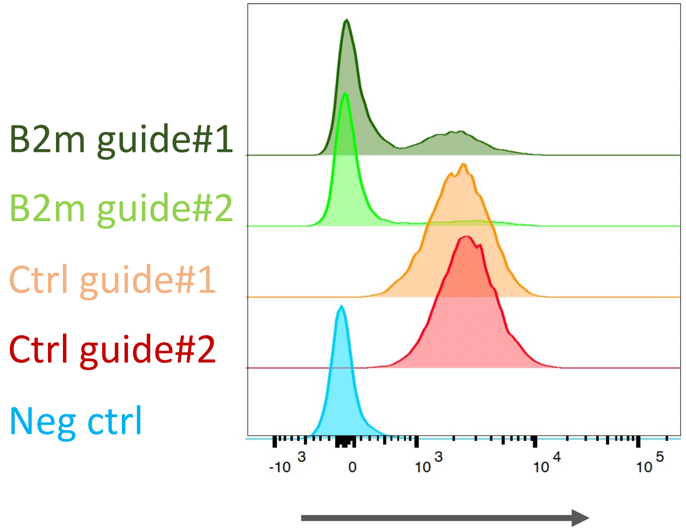
Need for more relevant cellular models

Antoine Rebendenne



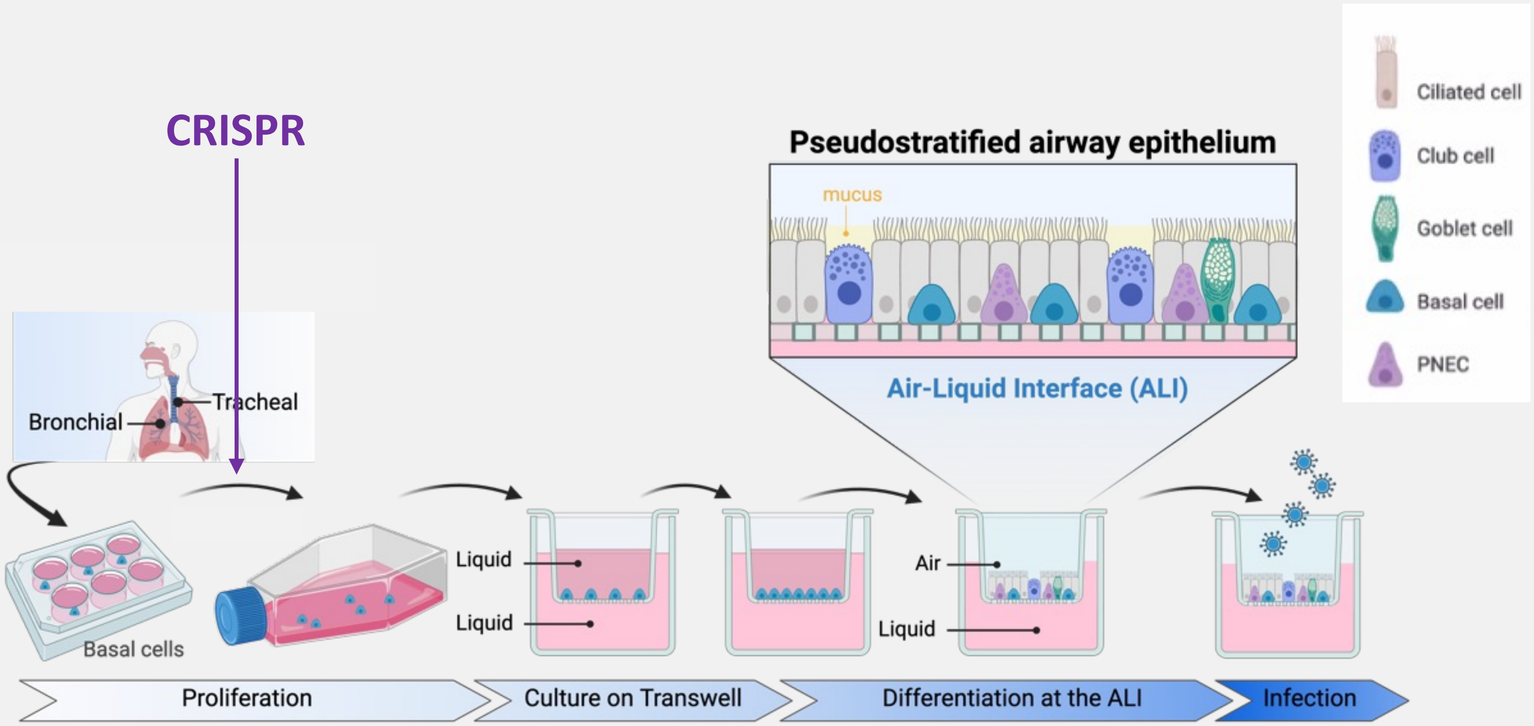
5 days

21 days



B2m expression level in differentiated cells (FACS analysis)

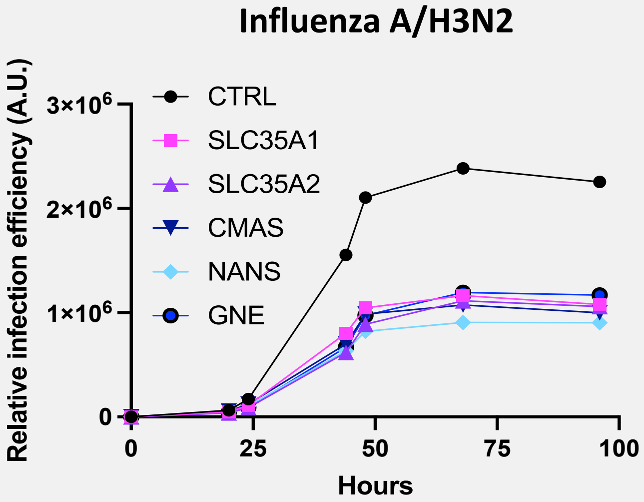
Need for more relevant cellular models

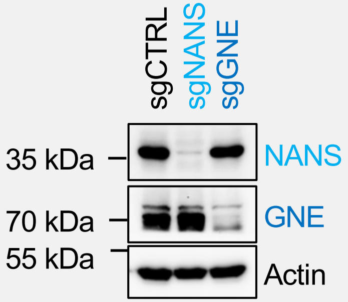


5 days

21 days

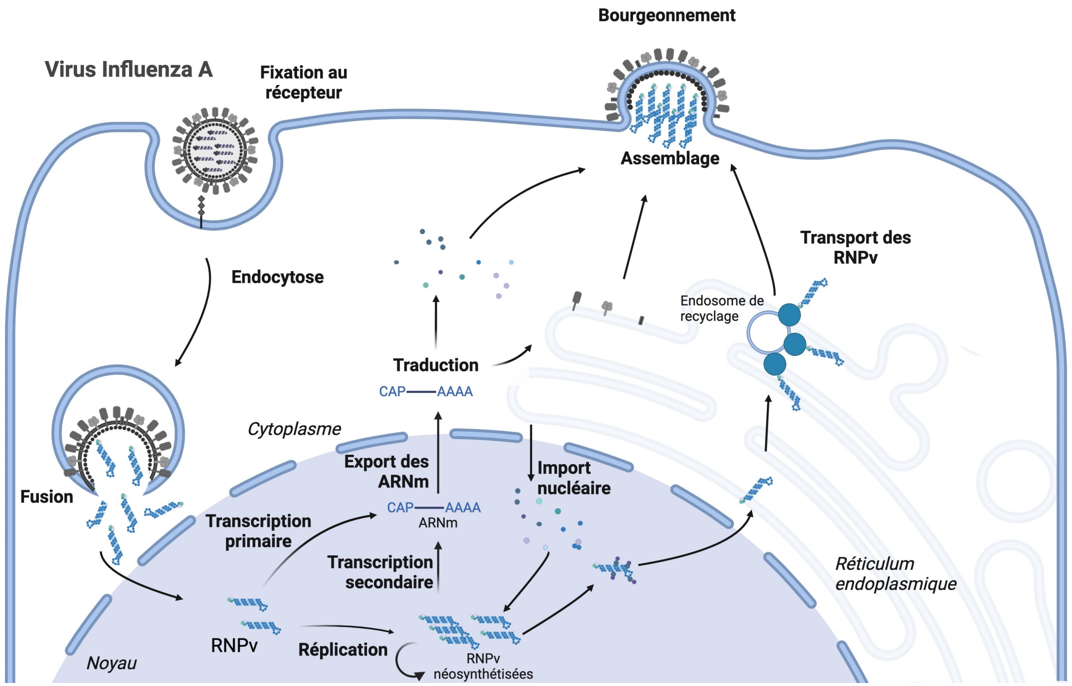
Caroline Goujon

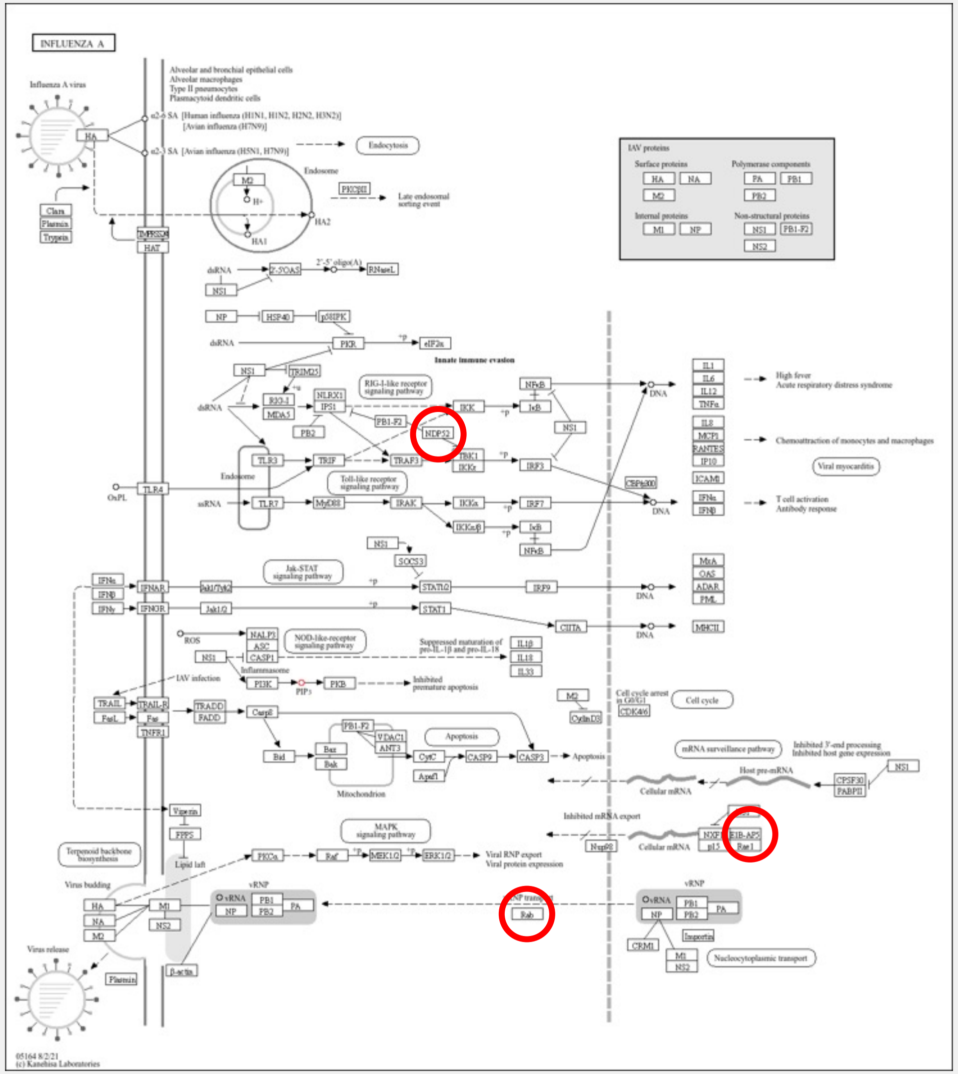




Atlas of key Host Dependency Factors (HDF)

Influenza virus

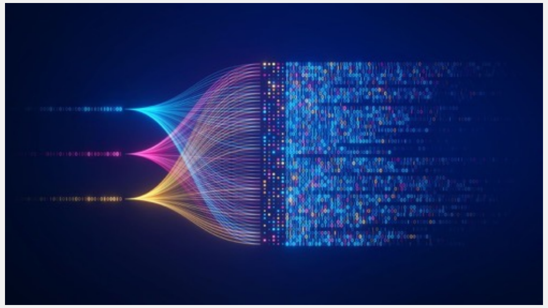




And identification of inhibitors for therapeutical intervention

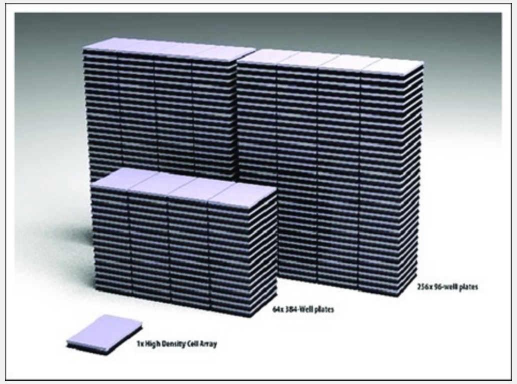
Drug selection pipeline

- Selection of broad-spectrum candidate genes using **machine learning** based on available data and our data



- Emerging, re-emerging viruses, pandemic potential: BSL2 (influenza A and B), BSL3 (influenza A H5N1, SARS-CoV-2, SARS-CoV, MERS) and **BSL4** (Lassa, Nipah) viruses

- Validations on **High Density Cell Arrays** (HDCA)



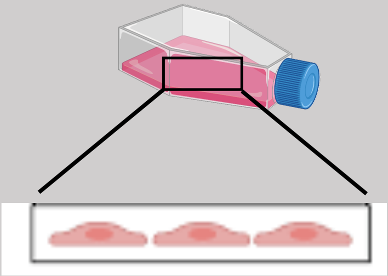
HDCA

- Use of **primary cell cultures** (MatTek)

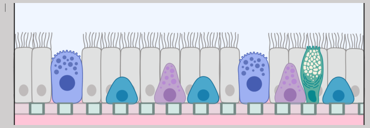
- **Drug repurposing** strategy targeting key cellular gene products/pathway

Poor compound efficacy prediction in immortalized cell lines

Example #1: hit compound against influenza virus

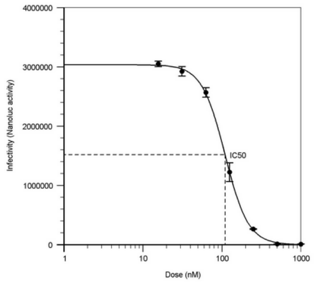


Cancer cell lines
(A549)



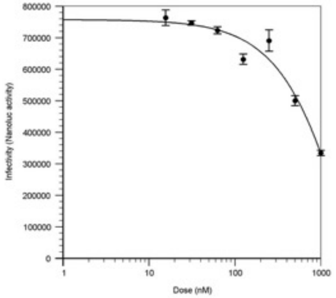
Primary epithelium
(3D culture)

EC-IR(-)



IC50: 110nM

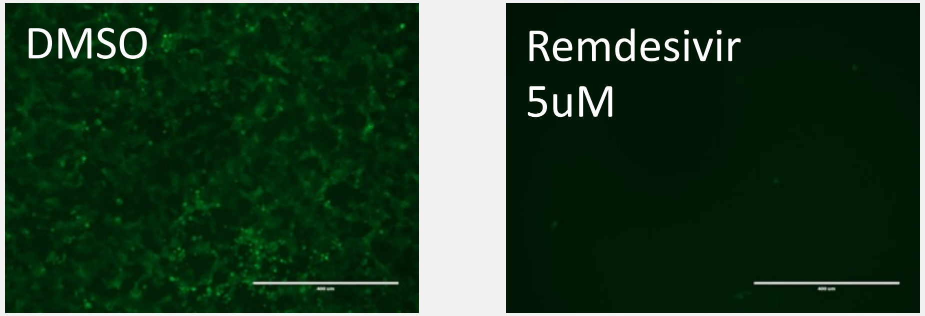
EC-IR(-)



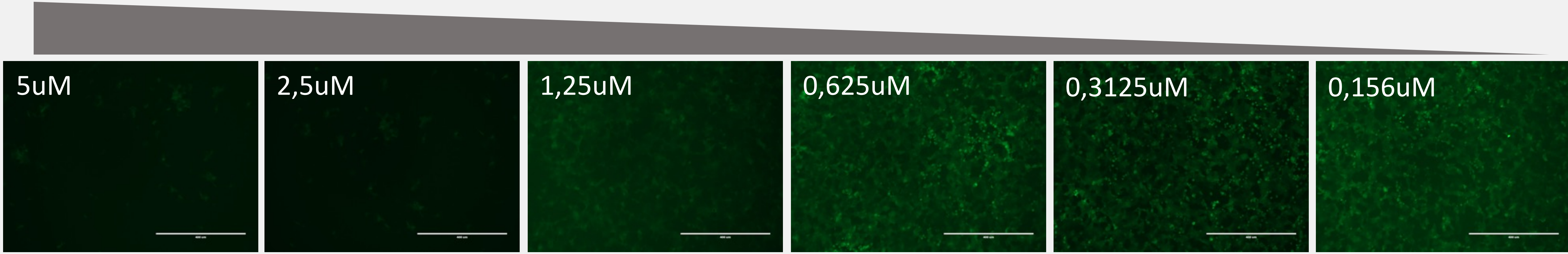
IC50: >3uM

Poor compound efficacy prediction in immortalized cell lines

Example #2: hit compound against SARS-CoV-2 in Vero E6 cell line



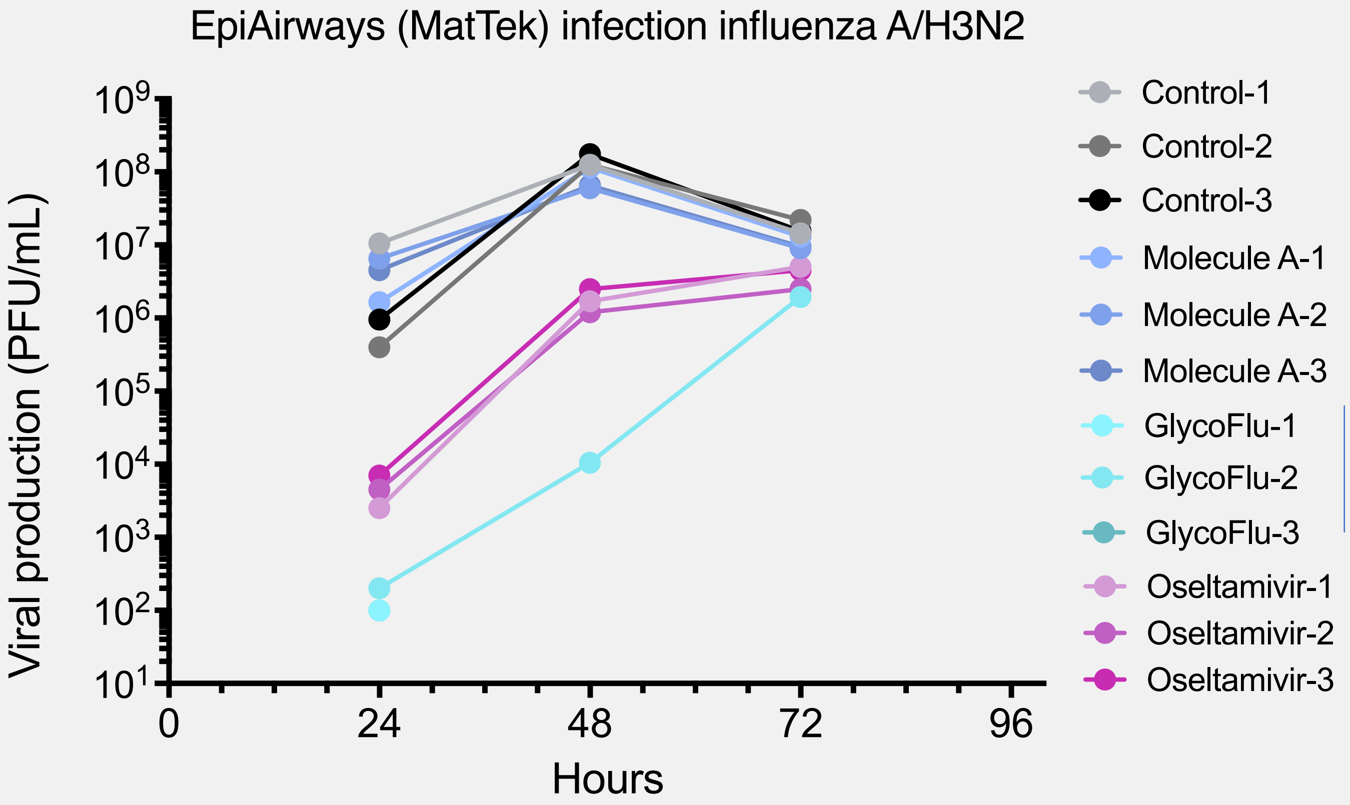
Inhibiteur AZ :



… but no activity in human primary airway epithelia …

Poor compound efficacy prediction in immortalized cell lines

Example #3: Molecule "A" predicted to be active against influenza A virus

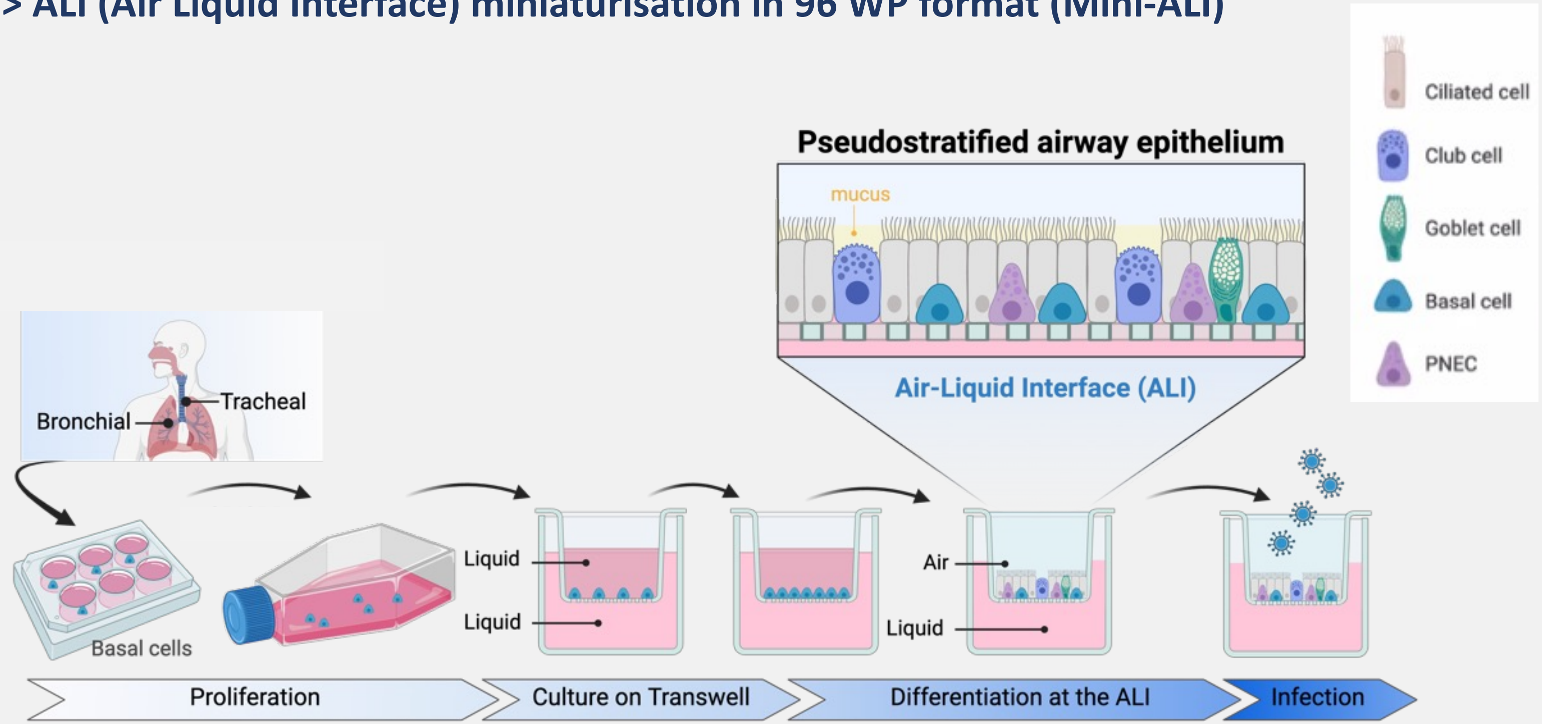


Need to develop the use of more relevant cellular models

For drug screening and development

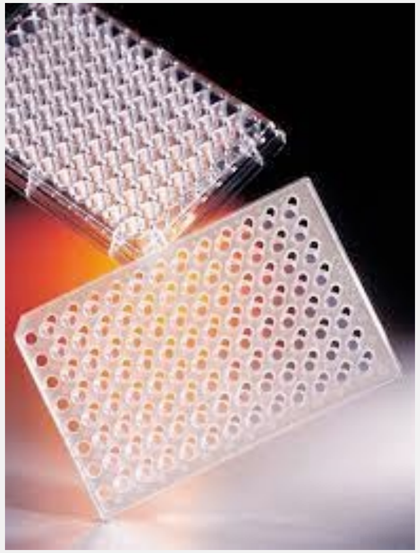
-> ALI (Air Liquid Interface) miniaturisation in 96 WP format (Mini-ALI)

Alice Trausch



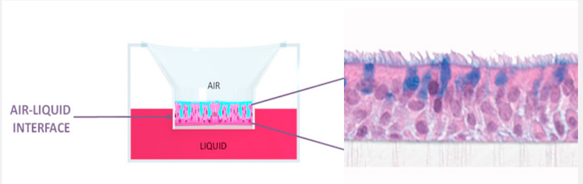
5 days

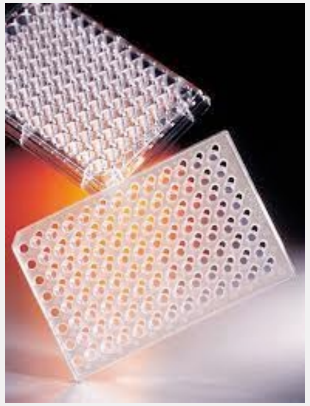
21 days



Need to develop the use of more relevant cellular models

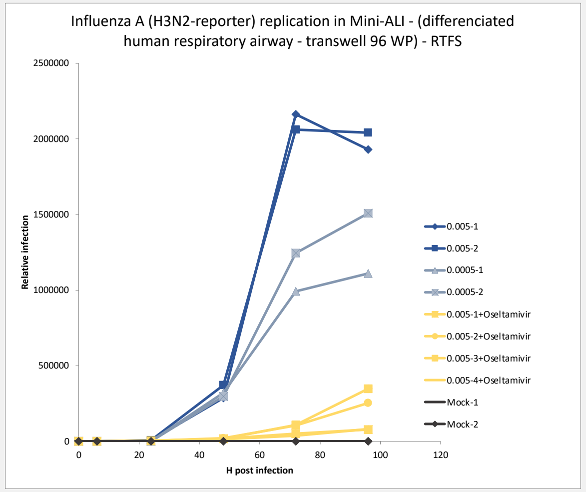
ALI (Air Liquid Interface) miniaturisation in 96 WP format (Mini-ALI)



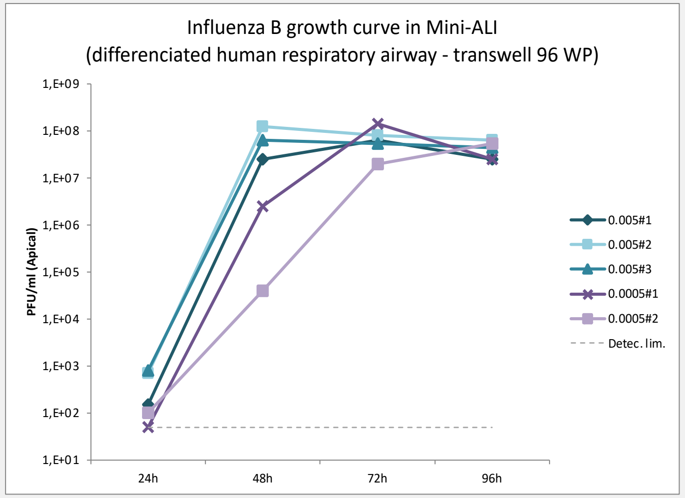


Viral replication in Mini-ALI

Influenza A/H3N2



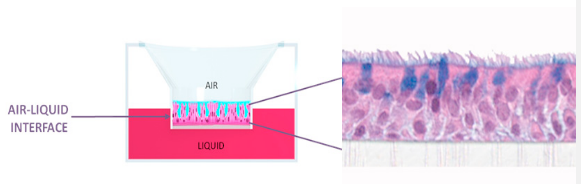
Influenza B

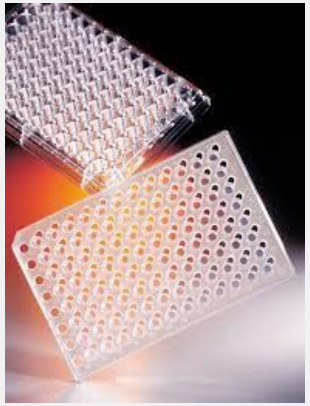


In collaboration with Aurélie Fort (MedBioMed) and Arnaud Bourdin (CHU Montpellier)

Need to develop the use of more relevant cellular models

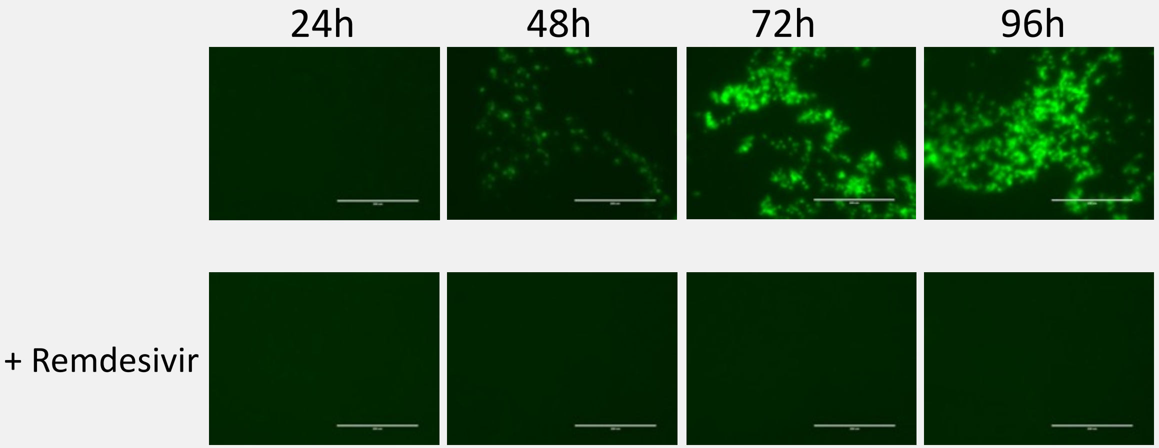
ALI (Air Liquid Interface) miniaturisation in 96 WP format (Mini-ALI)



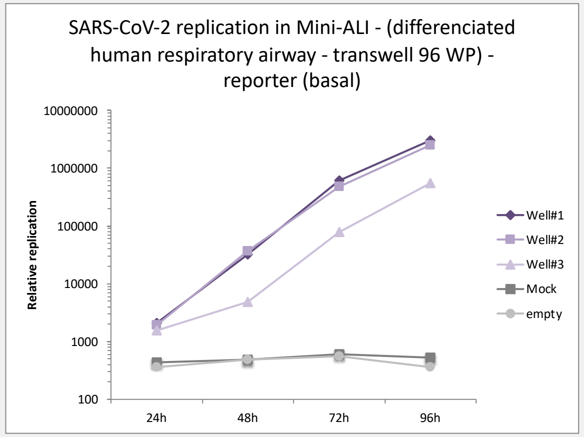


Viral replication in Mini-ALI

SARS-CoV-2 (mNeonGreen)



SARS-CoV-2 (NanoLuc)



In collaboration with Aurélie Fort (MedBioMed) and Arnaud Bourdin (CHU Montpellier)

See Alice's poster for more details

Take home messages

- Careful with OMICS data / data obtained with only 1 cell line

- Use more relevant cellular models (primary, organoids, iPSC, …)

- Particularly true for respiratory viruses

- Primary cells are still a model… and a model is never perfect

- By using machine learning, assays in primary cells, we hope to identify key cellular factors whose inhibition will prevent replication of several viruses

Thank you for your attention

The next Negative-Strand RNA Virus meeting (NSV) is in France!

www.project-appeal.eu

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