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Accelerated Article Preview

# Rapid reconstruction of SARS-CoV-2 using a synthetic genomics platform

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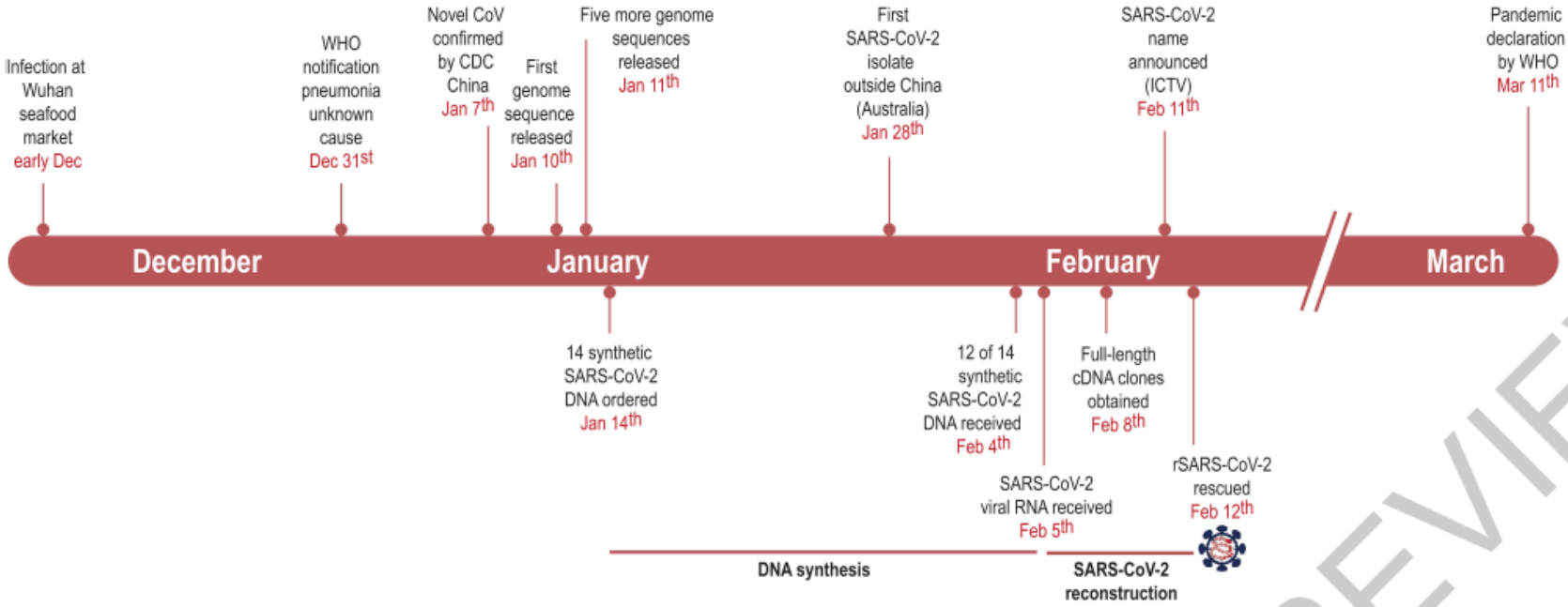
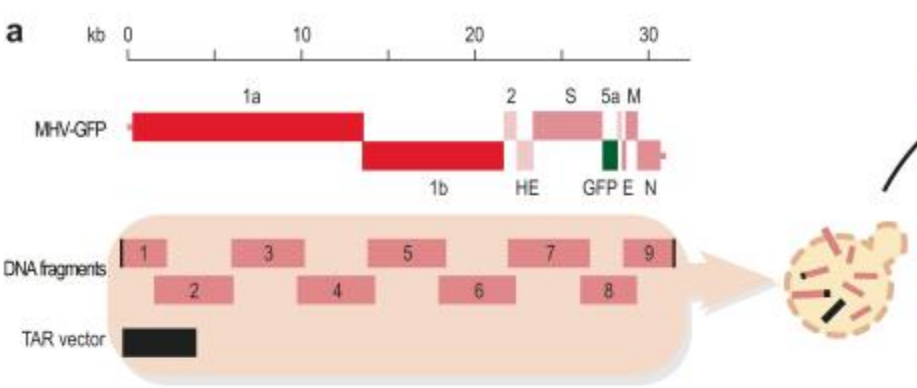


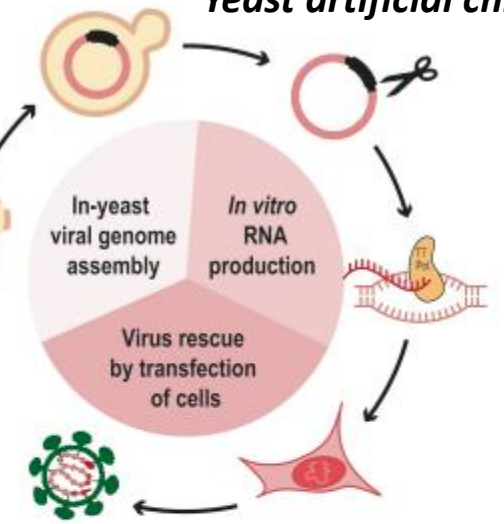

**Fig. 2 | Timeline of the reconstruction and recovery of rSARS-CoV-2 in relation to key events of the SARS-CoV-2 pandemic.**

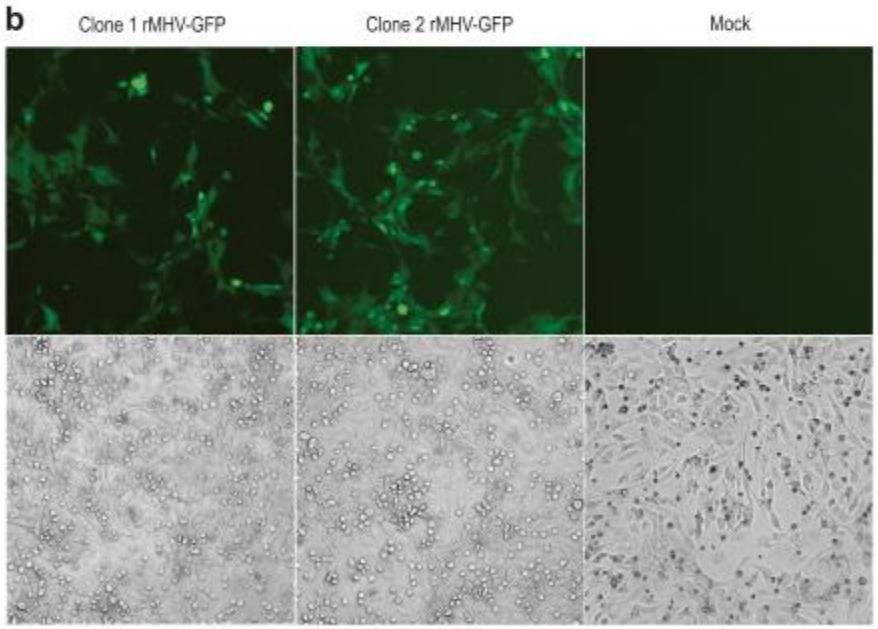
# Molekularbiologische Rekonstruktion von rekombinantem Maus Hepatitis Virus durch TAR (Transformation-associated recombination)-Klonierung in Hefe

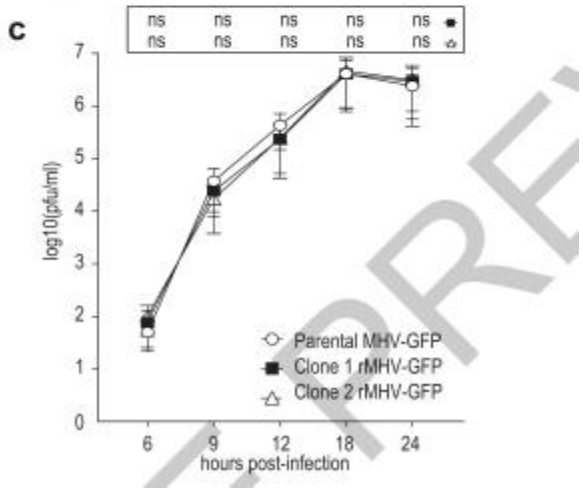
**9 PCR-Fragmente**




*Yeast artificial chromosome*



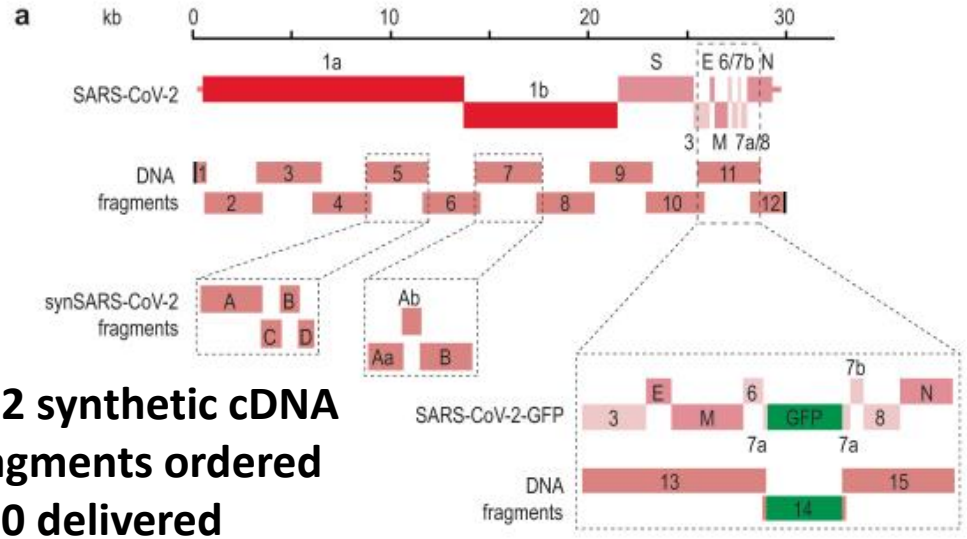



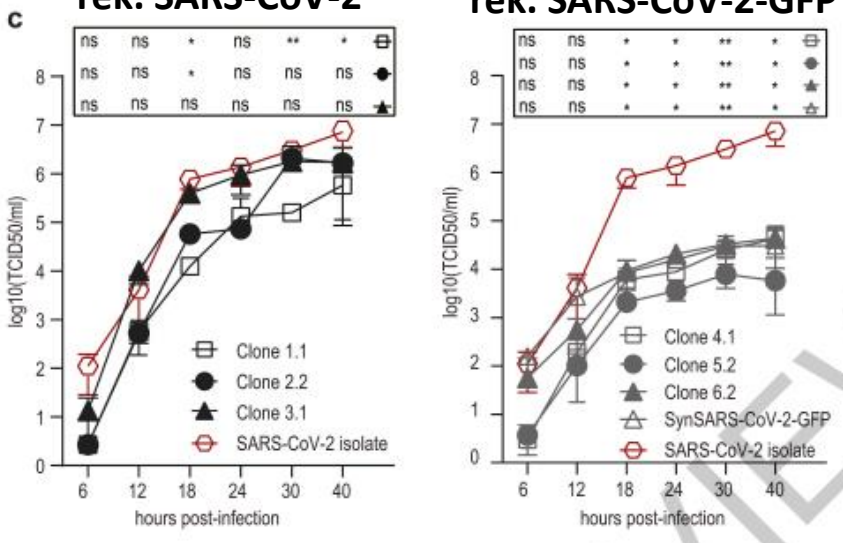

-----Zytopathischer Effekt -----

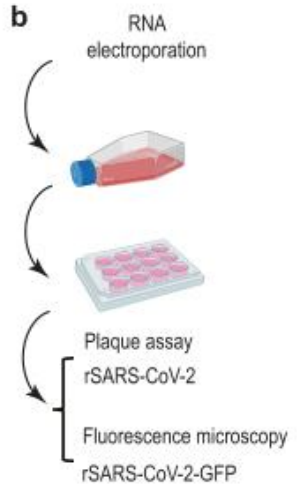
**Rekombinanter und WT-MHV-GFP: gleiche Vermehrungseigenschaften**

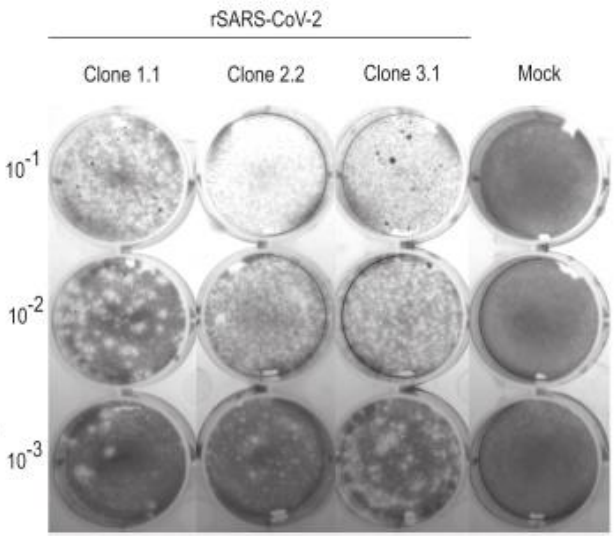
# *De novo*-Erzeugung von rekombinanten SARS-CoV-2 und SARS-CoV-2-GFP

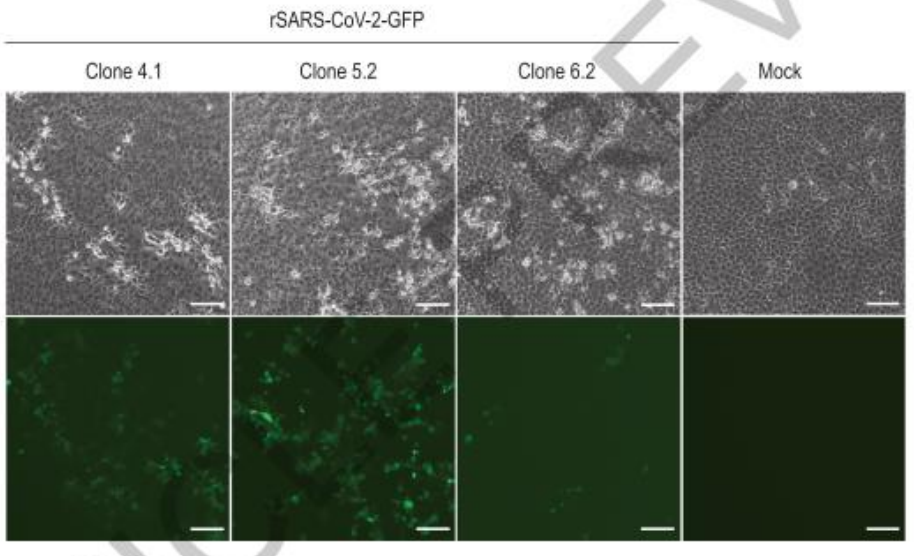



- 12 synthetic cDNA fragments ordered
- 10 delivered



# Synthetische Erzeugung von rekombinanten Viren mit großen Genomen – Anwendungen:

- **SARS-CoV-2-GFP: Entwicklung von Zell-basierten Screening-Testen**
- **Phänotypische Charakterisierung von Mutationen in zirkulierenden SARS-CoV-2**
- **Gezielte Attenuierung von SARS-CoV-2 im Rahmen der Impfstoff-Entwicklung (?):**

**Table 1 | RNA virus genomes cloned using the synthetic genomics platform**

| Virus | Family | Size (kb) | Template | Fragment generation | Fragment [No]* | Virus rescue |
|---|---|---|---|---|---|---|
| MHV-GFP | Coronaviridae | 31.9 | Viral RNA; DNA clone | RT-PCR/PCR | 9 | yes |
| MERS-CoV | Coronaviridae | 30.1 | DNA clone | PCR | 8 | yes |
| MERS-CoV-GFP | Coronaviridae | 30.7 | DNA clone; GFP plasmid DNA | PCR | 10 | yes |
| HCoV-229E | Coronaviridae | 27.3 | Viral RNA; DNA clone | RT-PCR/PCR | 13 | not attempted |
| HCoV-HKU1 | Coronaviridae | 29.9 | Synthetic DNA; viral RNA | PCR/RT-PCR | 11 | not attempted |
| MERS-CoV-Riyadh-1734-2015 | Coronaviridae | 30 | Viral RNA | RT-PCR | 8 | not attempted |
| ZIKA virus | Flaviviridae | 10.8 | Viral RNA | RT-PCR | 6 | not attempted |
| Human RSV-B | Paramyxoviridae | 15 | Clinical sample | RT-PCR | 4 | not attempted |
| SARS-CoV-2 | Coronaviridae | 30 | Synthetic DNA; viral RNA | plasmid/RT-PCR | 12 | yes |
| SARS-CoV-2-GFP | Coronaviridae | 30.5 | Synthetic DNA; viral RNA | plasmid/RT-PCR/PCR | 14 | yes |
| synSARS-CoV-2-GFP | Coronaviridae | 30.5 | Synthetic DNA | plasmid/PCR | 19 | yes |

*excluding the TAR vector fragment