# Progress in corona virus studies using SR

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Cockcroft Institute, 1st April 2020

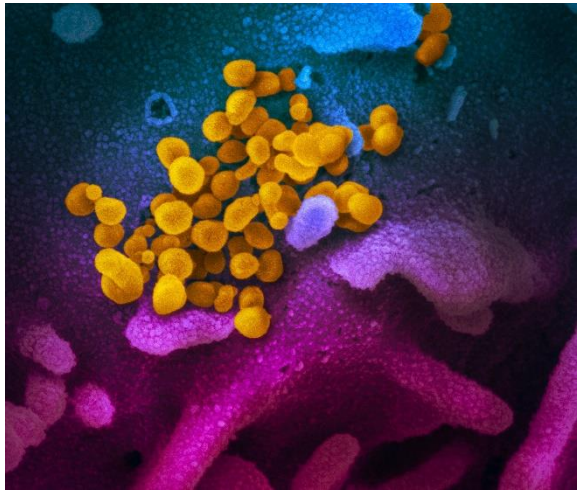
# First just a few basics

Corona viruses are a family which includes the common cold, SARS, MERS as well as the current outbreak of the disease COVID-19, caused by the **SARS-CoV-2 virus**.

- 3rd bat derived coronavirus to cause outbreaks of disease in humans in less than 20 years and most serious

- Some just affect animals

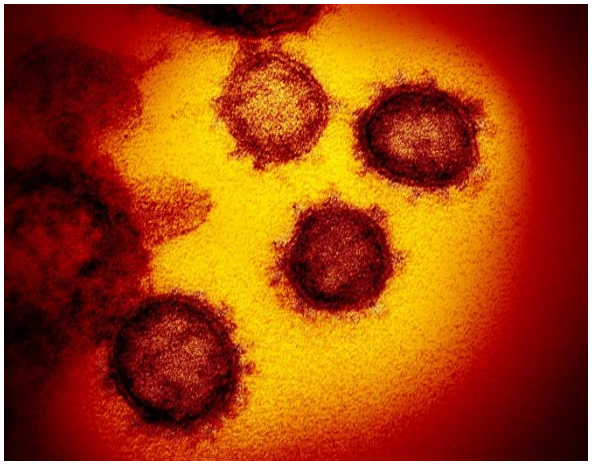
# EM images of SARS-CoV-2 (False Colour)



SEM image of SARS-CoV-2 emerging from the surface of a cell.

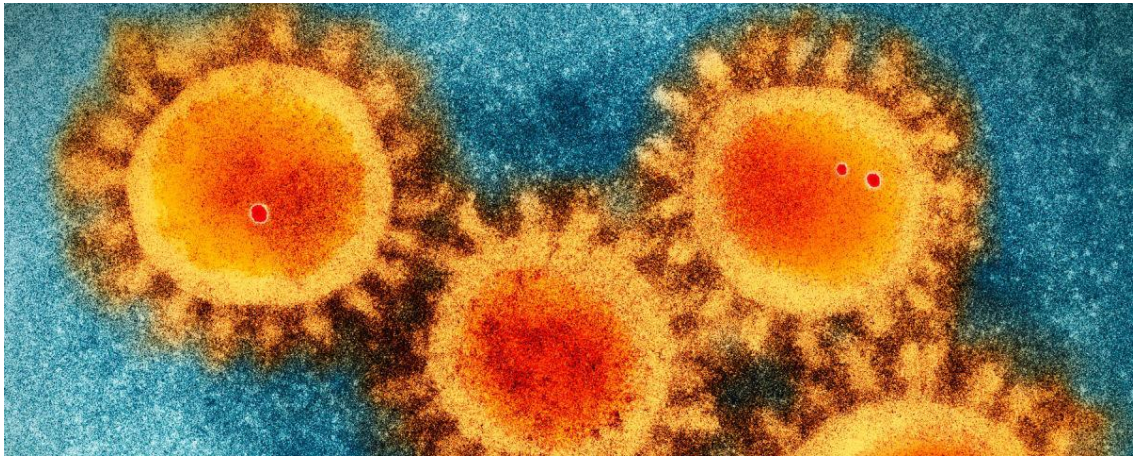
Viral particles - yellow
Cell surface - blue and pink

RML investigator Emmie de Wit provided the virus, microscopist Elizabeth Fischer produced the images.



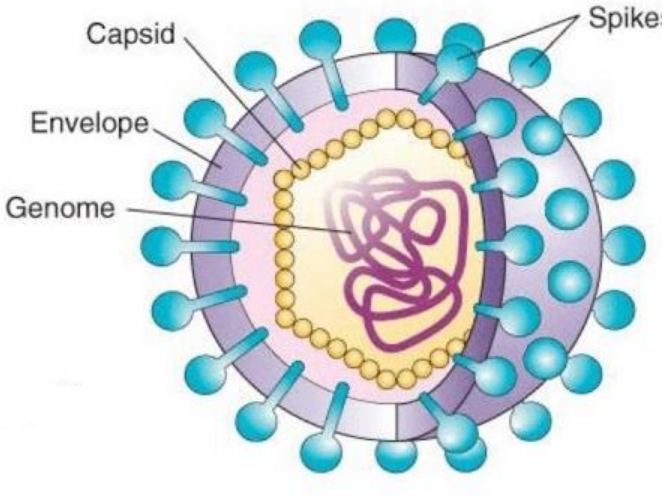
TEM image

Corona viruses derive their name from the spikes on their surface. Corona Latin for crown.



TheScientist, Feb24, 2020.

# Viral structure schematic




The shapes vary a lot

Envelope – lipid bilayer

Capsid – within which is the viral genome
(for CoV helically symmetrical
like a small can)

Viral genome – Single stranded,
+ve sense RNA
– 26,000 to 32,000 bases
very large

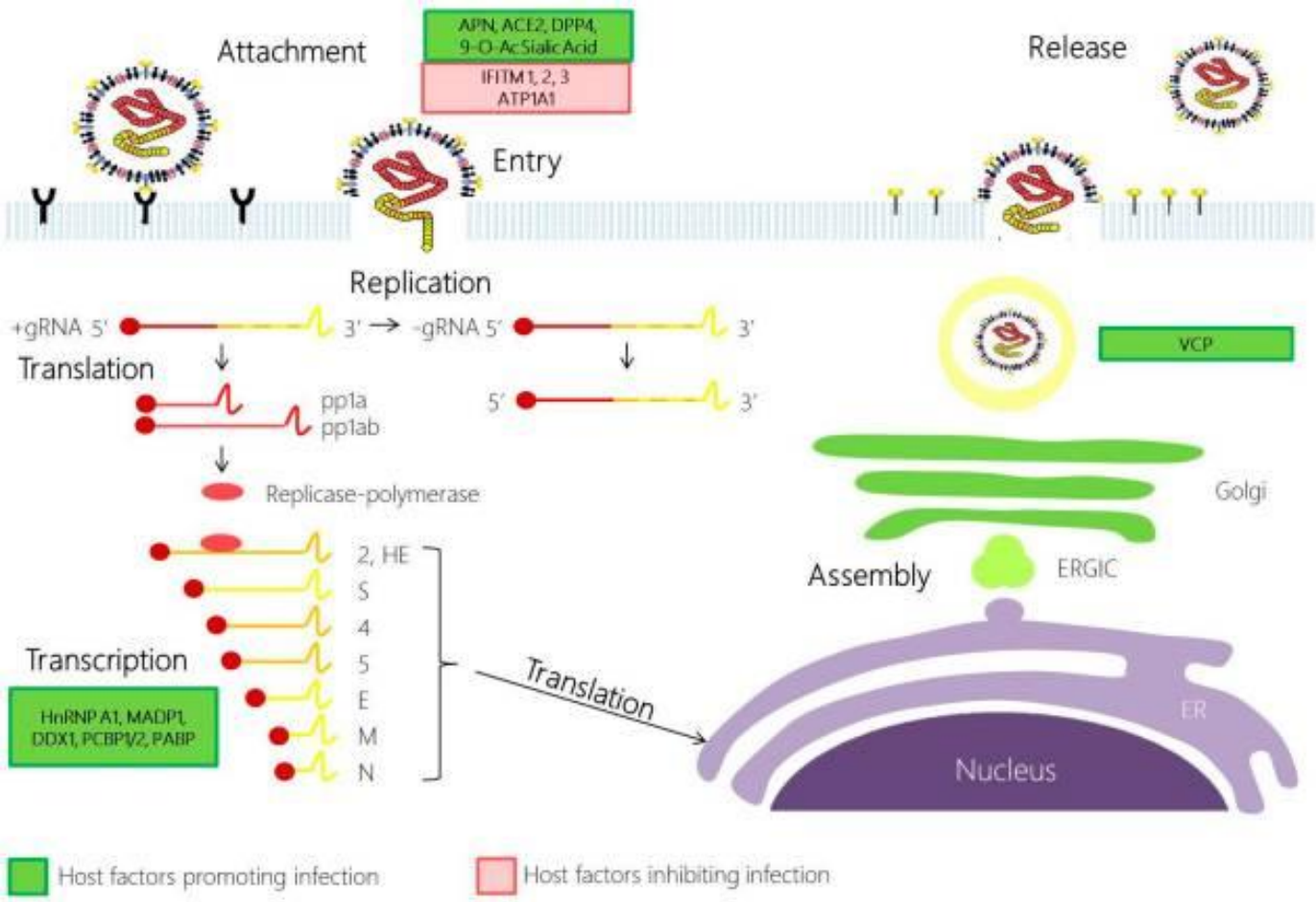
Spike – protein

Lim, Ng, Tam and Liu, Diseases. 2016 Sep; 4(3): 26.
https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5456285/

HCoVs exploit the host cell machinery for their own replication and spread.

# CoV infection cycle:

- Attachment – S1 domain of spike protein attaches to the ACE-2 receptor of cell
- Fusion of cell walls and entry into the host cell
- Translation of viral gRNA to form replicase-polymerase
- Replication of
  - the viral genomic RNA
  - formation of structural proteins
- Assembly in the ERGIC
- Budding of newly packaged viruses




Lim, Ng, Tam and Liu, Diseases. 2016 Sep; 4(3): 26.

# Structural Studies

To find a vaccine against, or cure for, a viral infection it is imperative to have as much detailed knowledge as possible at the molecular level of how the virus operates.

Techniques:

- Protein crystallography
  conducted at DLS, Elettra, Soleil, ESRF, etc.

- Cryo-Electron Microscopy
  ESRF Grenoble

Not by any means the only techniques but currently the most important.

# Structure guided drug discovery
# AIDS early 80s

In the **years** after AIDS was identified hundreds of structures were solved for key viral enzymes *i.e.* potential drug candidates. This work lead to the development of potent new compounds - now part of effective therapy.

Lessons:

- Even rapidly evolving viruses can be checked in their tracks by highly active multi-drug therapy.

- Timescale for drug discovery **long** compared with the time for a pandemic to sweep the planet.

**So what is being done differently now?**

# Many things…

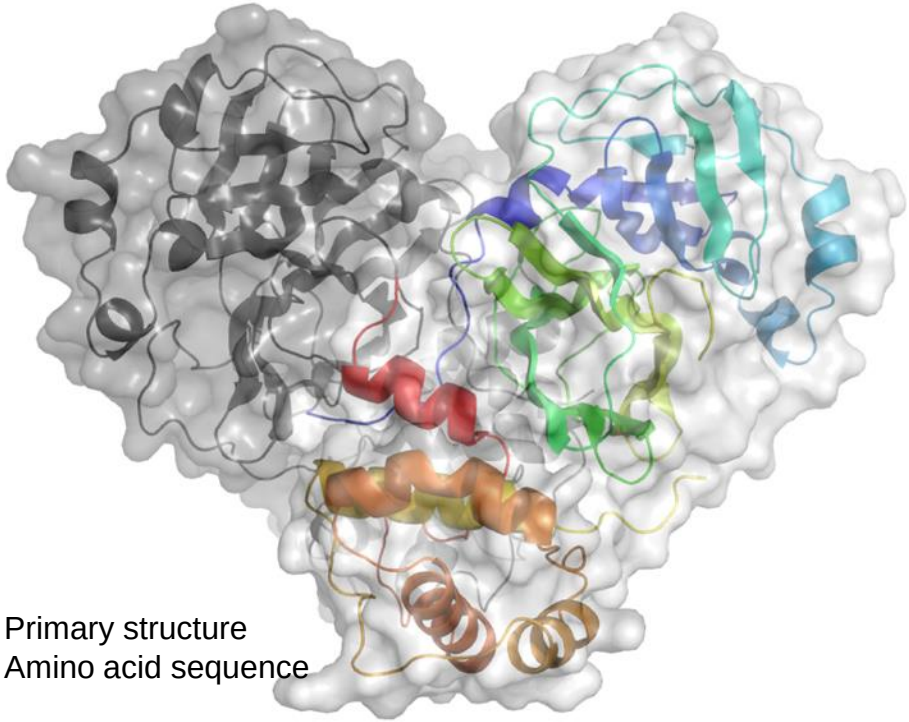
- Automation
  - Data acquisition
  - Data analysis
- AI
- Rapid sharing of information and results
  - Open access to data
  - Analysis of results immediately available without peer review
- Huge Cooperation between teams at synchrotrons rather than competition
  - Global not just Europe

Results that may have taken months or years to see the light of day before are happening in **days**

First reports of an unknown pneumonia – **31st December 2019**

# SARS-CoV-2 protease

Protease is an **enzyme** that catalyzes the breakdown of proteins into smaller polypeptides or single amino acids. SARS-Cov-2 has 2 proteases. Both are essential for viral replication and are therefore attractive targets.



Primary structure
Amino acid sequence

Secondary structure
Coils – alpha helices
Ribbons – beta sheets

Tertiary structure
Overall 3D shape

**6 virus sequences made available 11th January**

Experiments at the Shanghai SR Facility (SSRF) in China enabled the structure of the main SARS-CoV-2 protease to be solved by structural biologists Rao & Yang.

**Solved 5th February** - Info freely available on the WorldWide Protein Data Bank.

# In silico screening

- Based on the structure, the Shanghai team screened, in silico, a large number of existing drugs and biologically active natural products to assess their potential as therapeutics.

- 30 candidates selected

- However, scheduled shutdown of Shanghai SR Facility stops further experimental work.

- Rao contacts Life Sciences Director at Diamond (Dave Stuart) 26th January about collaborating to use the X-Chem Facility.

So what exactly is going on at DLS?

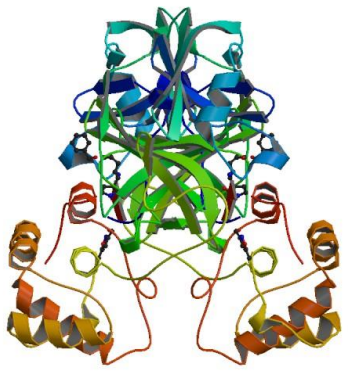
# Fragment-based screening

A ‘fragment’ is a smaller and simpler molecule than most drug molecules.

Experiments at DLS are using highly automated fragment-based screening at the active site of the main protease to select potential drug candidates.

By **early March** 1500 crystals and fragments analysed.

**Over recent weeks ~50 compounds have been identified.**



# SR studies

**Diamond Light Source: Suspension of Operations** **except for COVID-19 work**
Given the rapid spread of the COVID-19 virus Diamond Light Source has decided to suspend user operations from our facilities until 31st May. A further extension of that period might become necessary, and we will keep users informed during the coming weeks.

SR sources **globally** are encouraging rapid access applications for COVID-19 work.

# In parallel

Collection of licenced drugs with potential antiviral activity identified and the info made available publicly

- Clinical trials underway
- First results due soon
- These are re-purposed compounds and tailor-made molecules will take longer

**SO WATCH THIS SPACE**

# Many thanks for listening in …

# Key Words

Protease
Protease is an **enzyme** that catalyzes proteolysis, the breakdown of proteins into smaller polypeptides or single amino acids. They do this by cleaving the peptide bonds within proteins by hydrolysis, a reaction where water breaks bonds.

Glycosylation
**Glycosylation** (see also chemical **glycosylation**) is the reaction in which a carbohydrate, i.e. a glycosyl donor, is attached to a hydroxyl or other functional group of another molecule (a glycosyl acceptor).

ERGIC
The vesicular-tubular cluster (VTC), also referred to as the ER-Golgi intermediate compartment (or **ERGIC**), is an organelle in eukaryotic cells. This compartment mediates trafficking between the endoplasmic reticulum and Golgi complex, facilitating the sorting of cargo

Replication polymerase