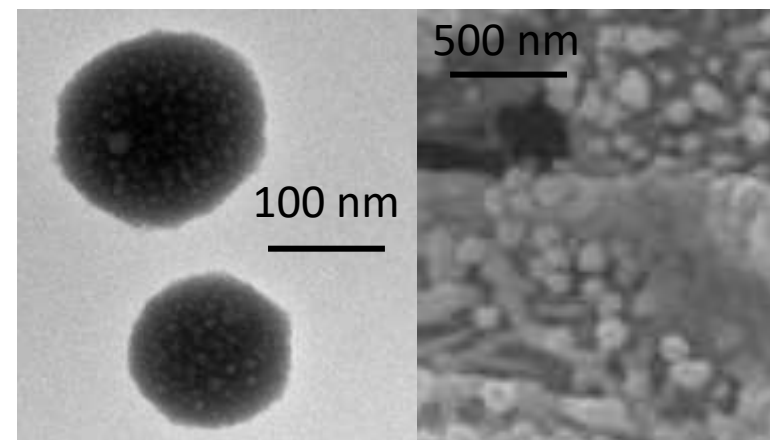
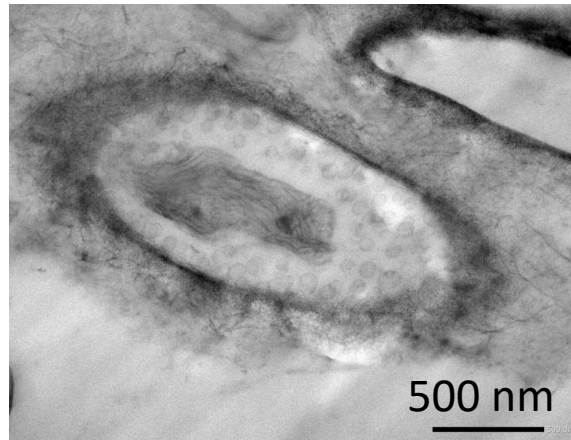
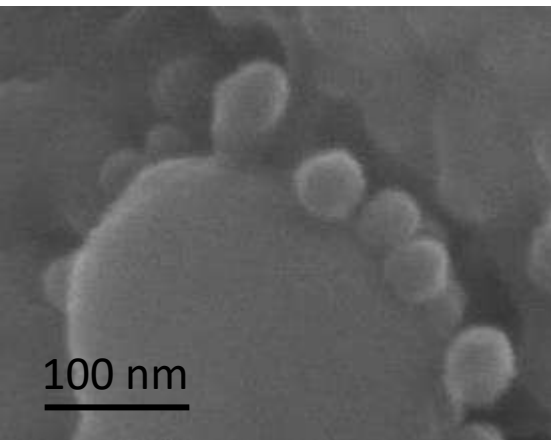
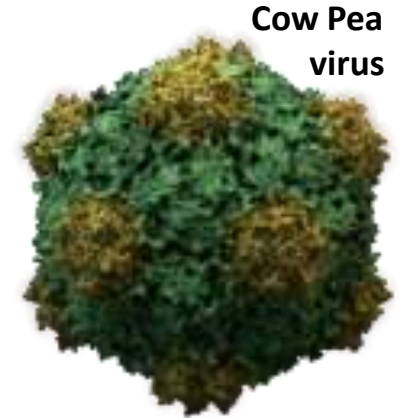
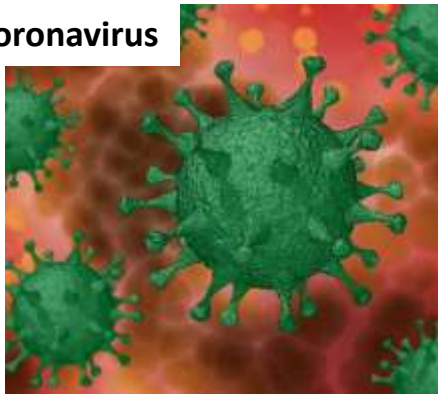


FOSSIL VIRUSES

Maurice Tucker

A current research project with colleagues:

Mirek Słowakiewicz, Andrzej Borkowski, Edoardo Perri, Marcin Syczewski, Filip Owczarek, Anna Sikora, Anna Detman, Fiona Whitaker, Ida Perrotta, Leon Bowen, Lisa Thomas
(Bristol, Warsaw, Krakow, Calabria, Durham, Bath Spa)



Viruses on a bacterium
in a modern microbial mat from Qatar

Viruses in a bacterium
in a modern microbial mat from Qatar

Experimentally mineralised **viruses** Palaeoproterozoic **viruses**

THIS TALK:

Viruses – an introduction

Bacteria and viruses (bacteriophages) and

viruses in microbial mats: their mineralisation, i.e. fossilisation

Experimental precipitation of minerals through viruses

Geological record of viruses

Viruses in natural environments: the good aspects ...

Viruses in sedimentary processes: mineral precipitation

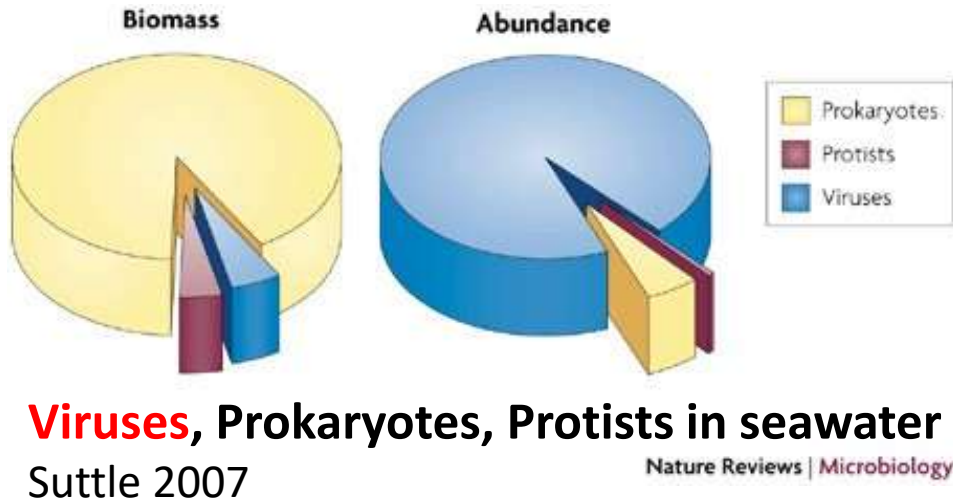
Viruses and extinction

**VIRUSES: THE NEW FRONTIER IN GEOLOGY
DO THEY PLAY A ROLE ?**

VIRUSES

Viruses are everywhere, in greater abundance than bacteria.

In seawater 10 billion per ml, 10-100x more than bacteria.



A gram of soil may contain 10 billion bacteria in the vicinity of plant roots
BUT

the number of **viruses** is 10 or more times these figures.

In terms of numbers, **viruses** are the most abundant biological entities on Earth.

Viruses occur in sediments, like bacteria, shallow and deep, and all environments; hot springs and ice-cold glaciers, the deep subsurface, rain (and Mars, Venus ?!). In terms of volume or mass, however, viruses constitute a much smaller amount than bacteria, just a few %.

AND IN US TOO: For example, the weight of bacteria in the human body is estimated to be 200 gm; the **weight of viruses** would be less than 10 gm.

The **number of viruses** is around 380 trillion (= human virome).

VIRUSES versus BACTERIA

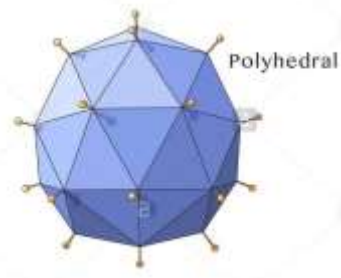
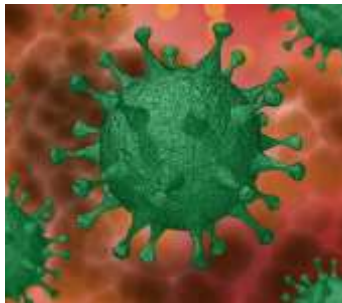
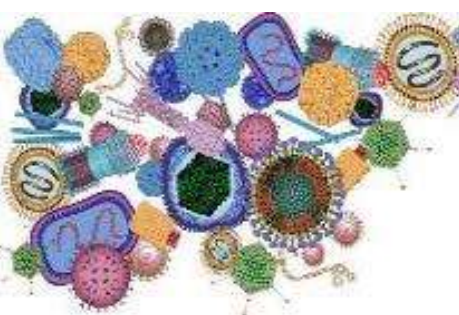
Viruses: 10s-100s nm diameter. Bacteria < micron to 10s of microns. Largest 750 μm .

Bacteria are single cells, prokaryote organisms (i.e. no nucleus), with cell wall within which cytoplasm containing DNA and organelles. They mostly reproduce asexually by binary fission (dividing into 2 daughter cells).

Viruses are somewhere between living and non-living organisms. They consist of a protein shell (called a capsid) within which there is nucleic acid (i.e. RNA or DNA) which carries genetic information. Some **viruses** have an outer membrane (an envelope) which may be spikey, as in a coronavirus.

Virus shapes: spheroidal, rod-shaped or helical (spiral).

Many symmetrical with an **icosahedral** shape, i.e. spheroidal with sides or faces, like an old-fashioned football. Some **viruses** have a 'tail'.



VIRUSES

Viruses are dependent solely on a suitable host for replication; hence they are frequently referred to as *obligate intracellular parasites*.

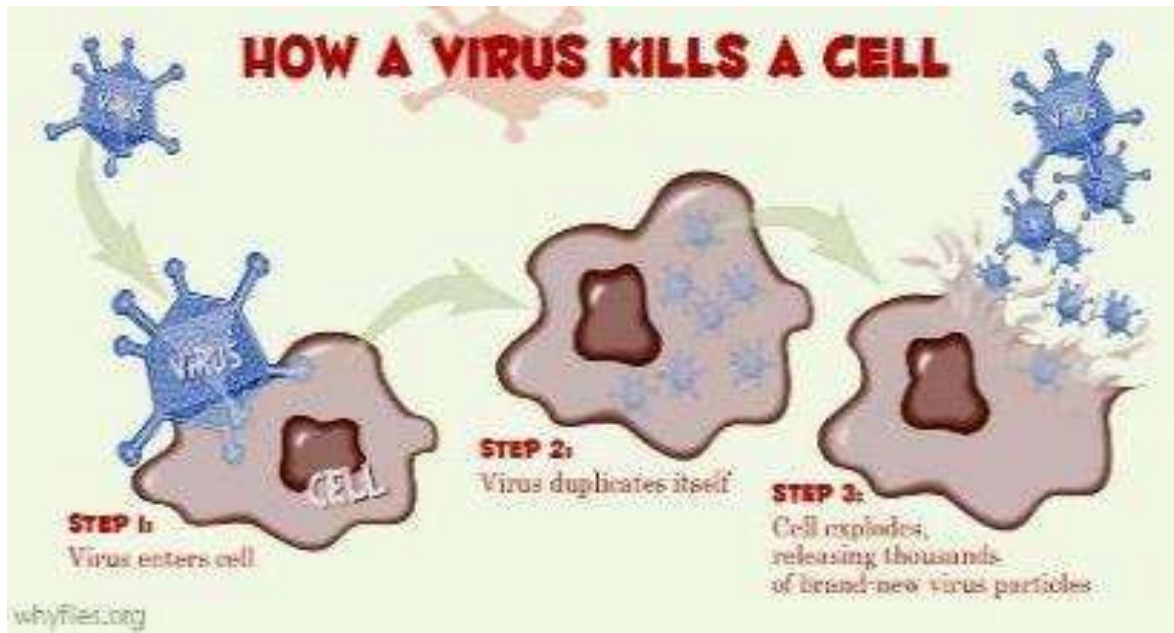
Simple biological entities, indeed are they actually living organisms ?

They only reproduce in host cells.

Viral DNA enters the cell; new **viruses** form; they multiply and burst out (**lysis**), and they may then infect other bacteria/cells.

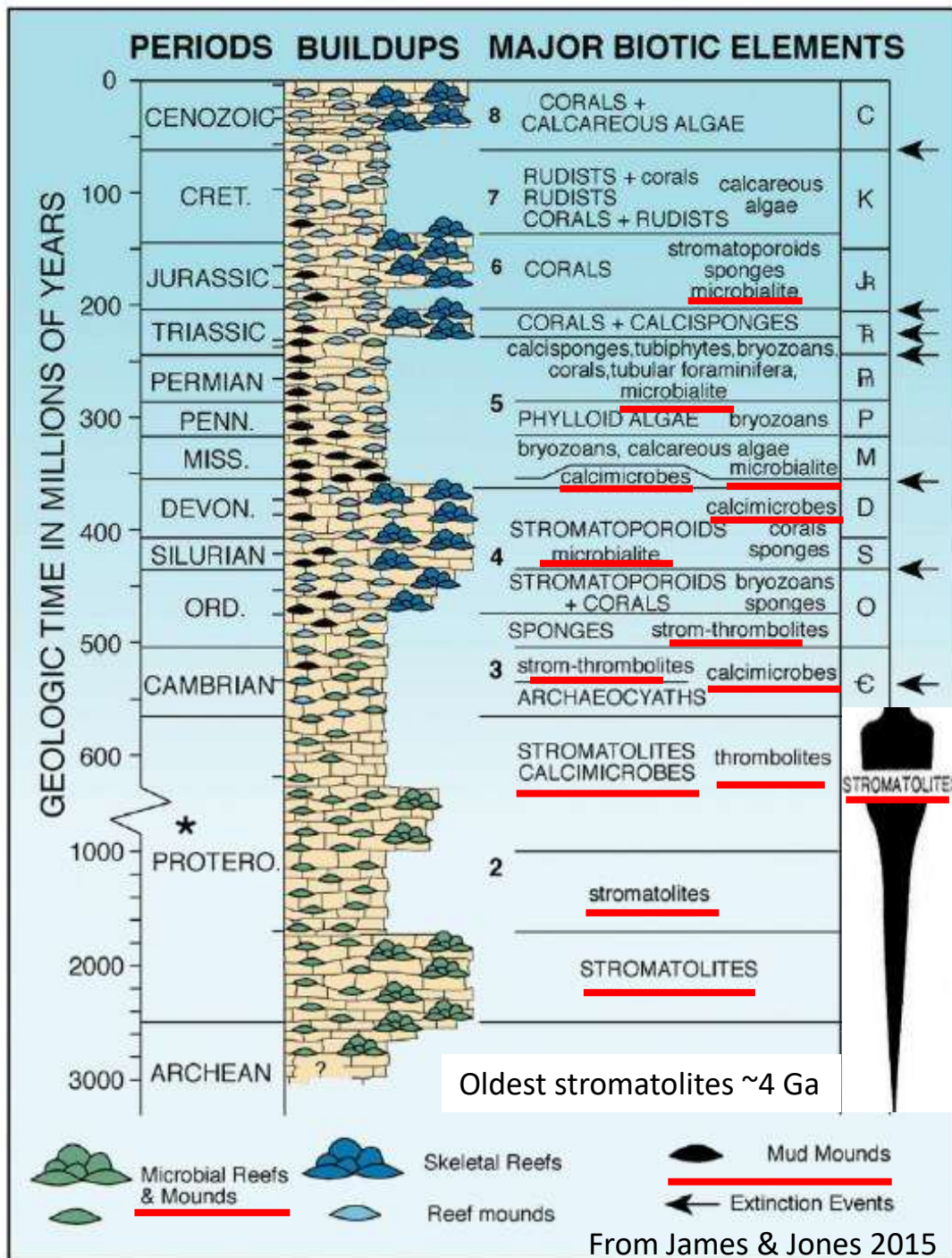
Viruses closely associated with bacteria called bacteriophages or just phages.

Viruses (like EPS, i.e. extracellular polymeric substances) have a negative charge (from hydroxyl and carboxyl functional groups), so can attract cations.



The close association of **viruses** with bacteria is well documented in microbial mats – where many more viruses than bacteria.

Microbialites occur throughout the stratigraphic record.



CARBONATES THROUGH TIME

Microbial carbonates from the early Archean

Donoghue 2020



Fossil bacteria (or pseudofossils?)
Archean (3.4 Ga) Apex Chert, WA.



Eukaryote cells + nuclei. Ruyang, China (1.8 Ga) and Bitter Springs, Oz (1 Ga).

Trucial Coast, Abu Dhabi



Shark Bay



Bahamas



MODERN STROMATOLITES

PRECAMBRIAN STROMATOLITES

Finnmark, Arctic Norway

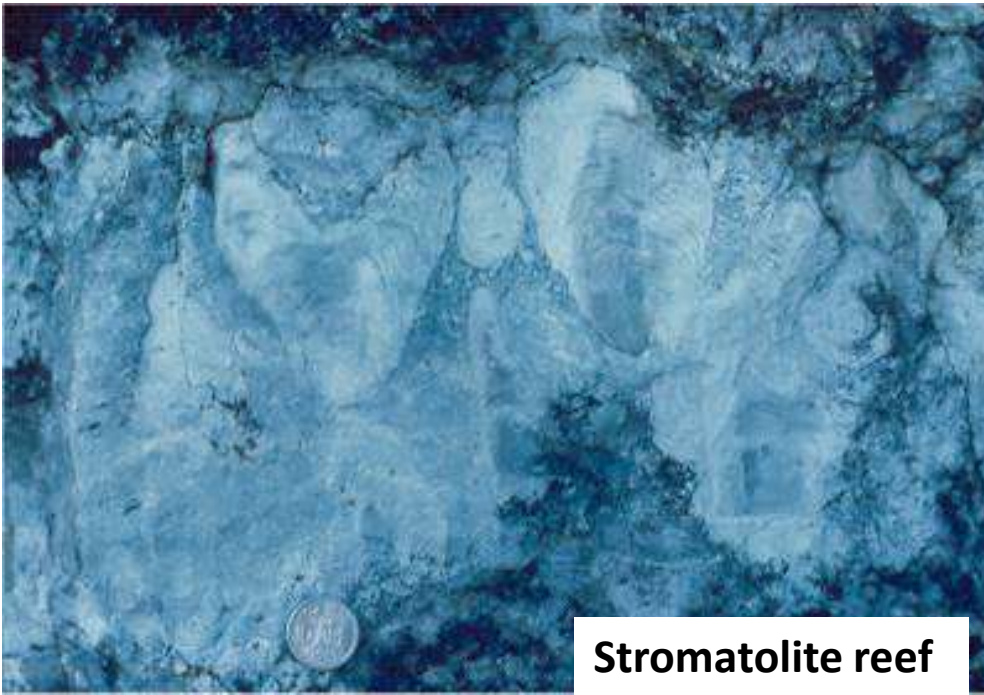
NEOPROTEROZOIC (1000 Ma)
PORSANGER FM, NORWAY



Stromatolite reef and oolite banks



Peritidal microbial facies (Tucker 1976, 77)



Stromatolite reef

MICROBIALITES NEARBY



Cotham Marble ('landscape marble')
Rhaetic, Triassic, Bristol District

**Purbeck – Lulworth
Fossil Forest
(BGS fieldtrip 2017)**

**Also microbialites (stromatolites) in the
Carboniferous (Clifton Down Limestone)**



MICROBIAL MATS and STROMATOLITES

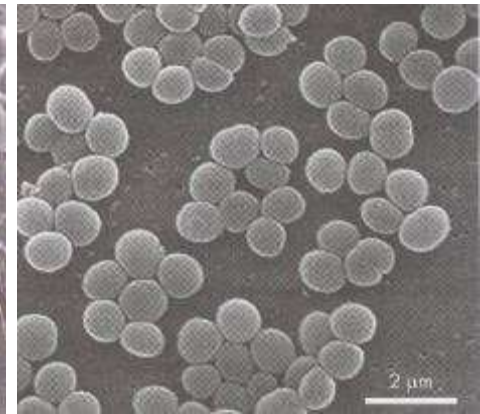
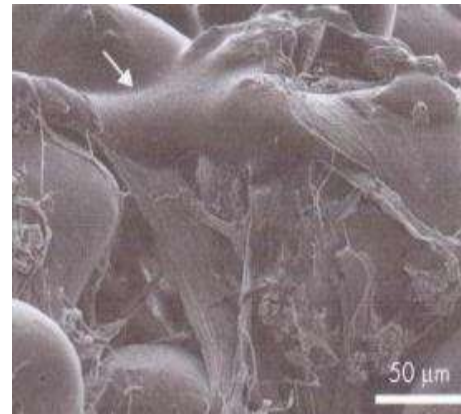
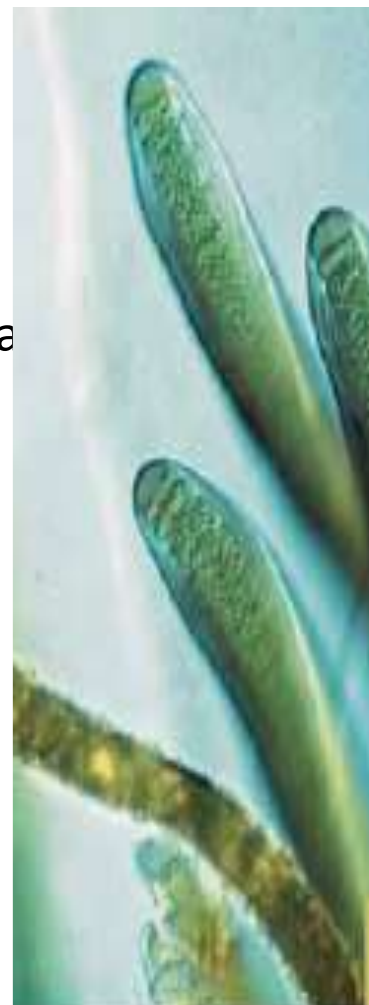
Microbial mats consist of cyanobacteria, other bacteria (SRB), archaea and green algae, plus fungi, diatoms, **EPS** (extracellular polymeric substances, 'mucilage') and **viruses**, all forming a **BIOFILM**.

Microbial precipitates common in mats plus sediment trapped and bound into the mat by the biofilm/EPS and filaments.

Microbes induce precipitation through extraction of CO_2 (in HCO_3^-) from water through **photosynthesis** (+ other processes).

**ALSO: tufa and travertine (springs)
and
planktic microbes with EPS and viruses.**

Coccoid and filamentous cyanobacteria and EPSs (extracellular polymeric substances)



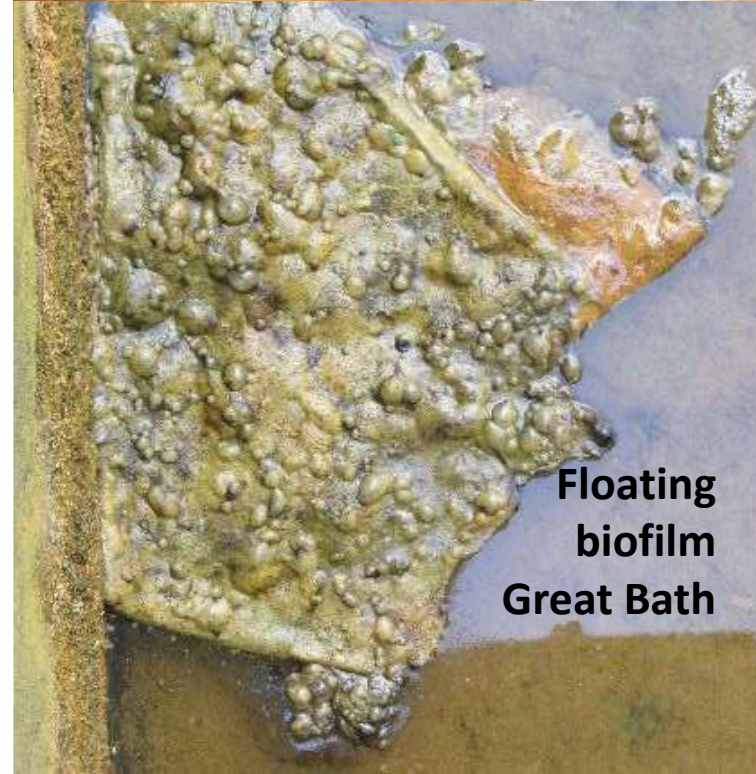
If you want to see biofilms and microbial mats – go to the Roman Baths:



**Mats
and
biofilm
Sacred
Spring /
King's
Bath**



**Mat/
biofilm
edge
of
Great
Bath**



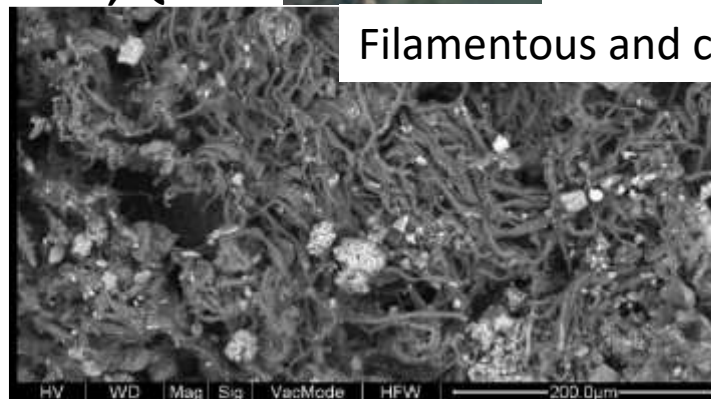
**Floating
biofilm
Great Bath**

MICROBIAL MATS, QATAR

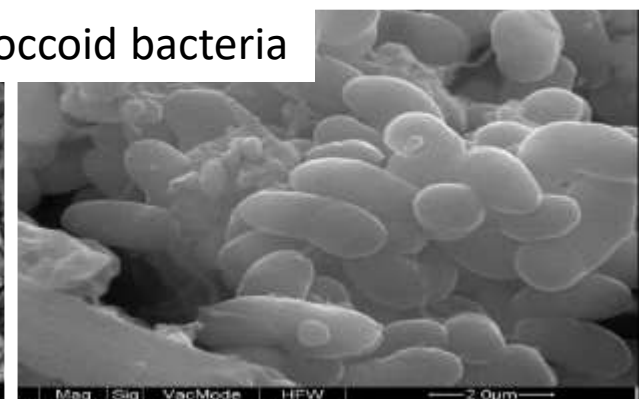
roles of EPS (mucilage), bacteria and **viruses** in mineral precipitation
and HC source-rock potential



Microbial mats at Mesaieed, Qatar

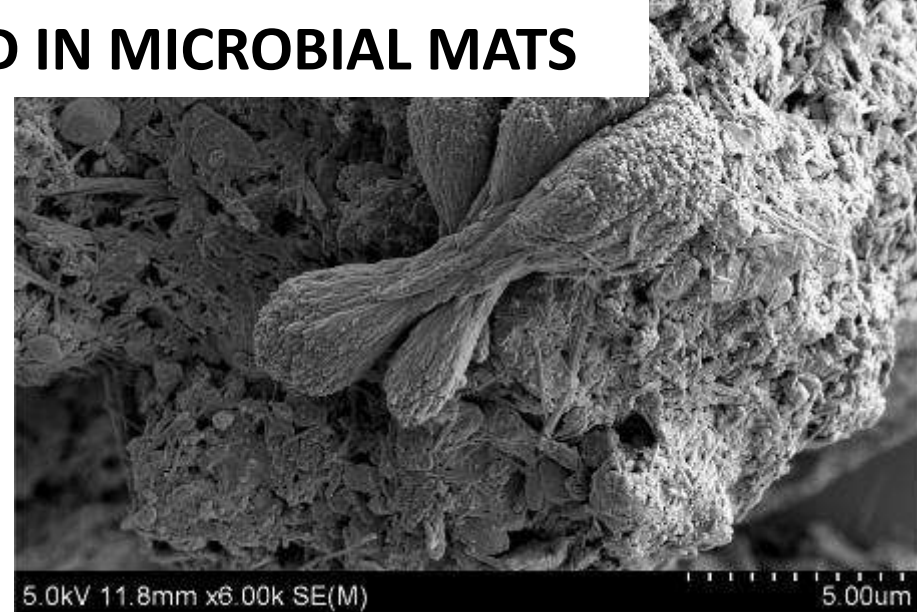
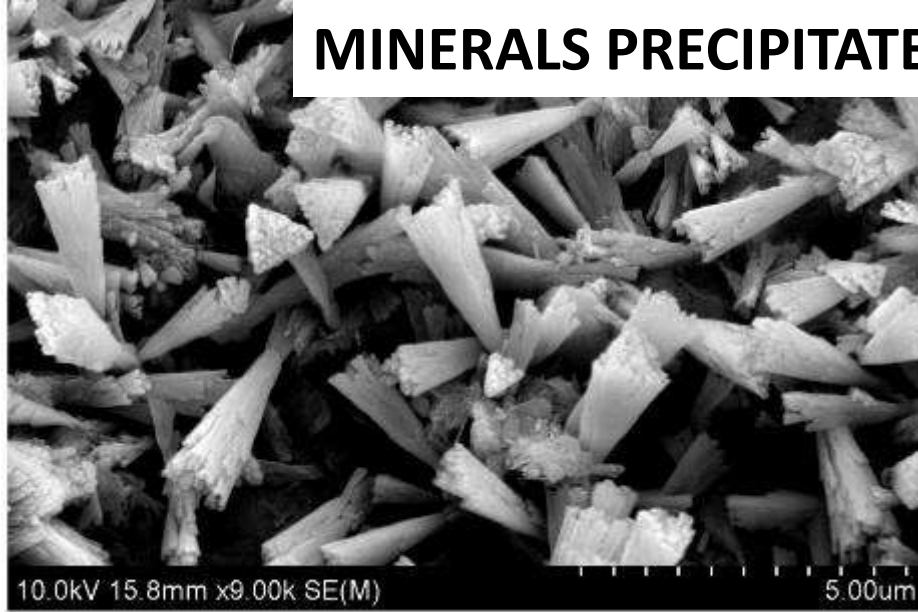


Filamentous and coccoid bacteria



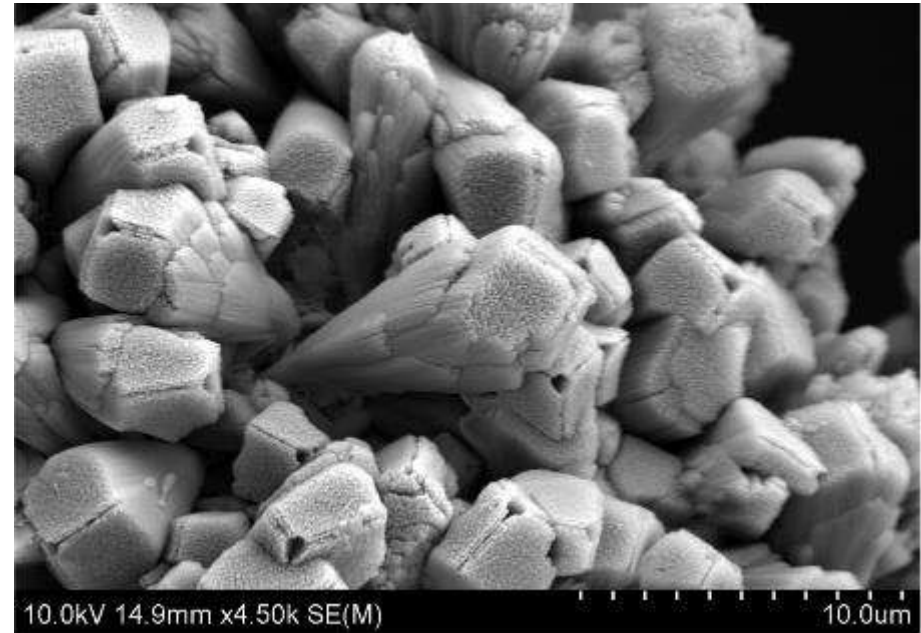
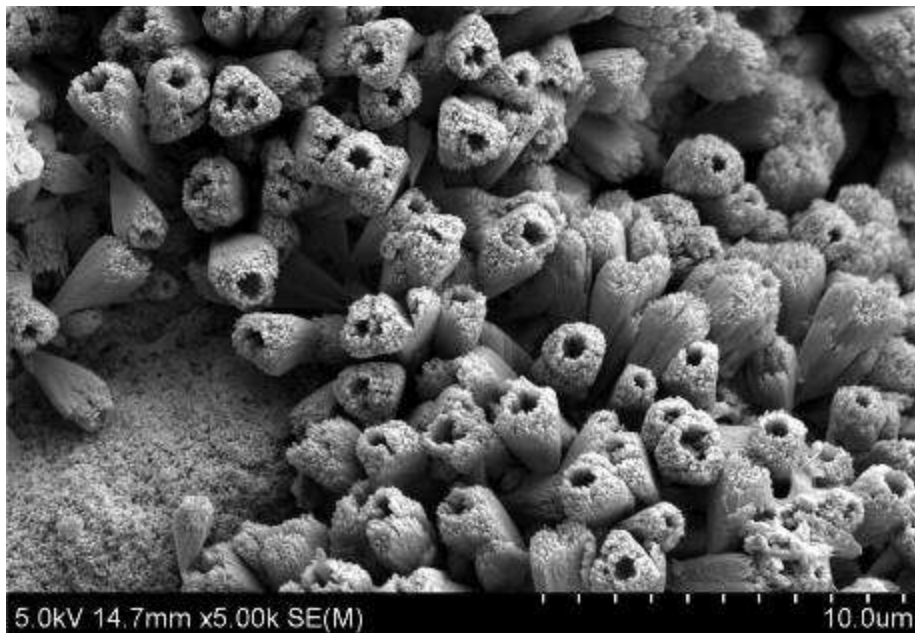
Biomarker study: Slowkiewicz, Tucker, Whitaker et al. (2016) *Organic Geochemistry*
Mineral precipitates in mats: Perri, Tucker, Slowkiewicz et al. (2018) *Sedimentology*.

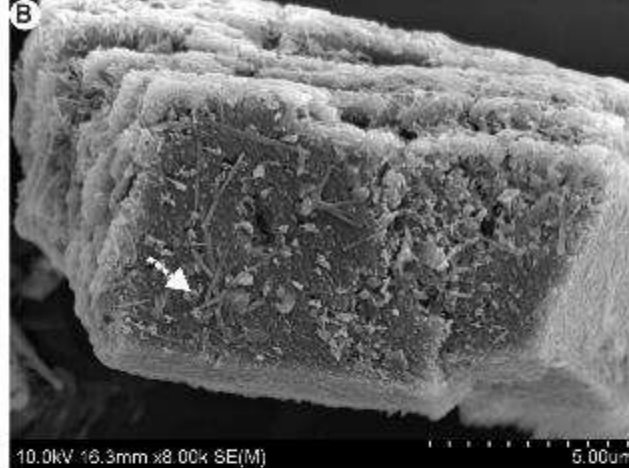
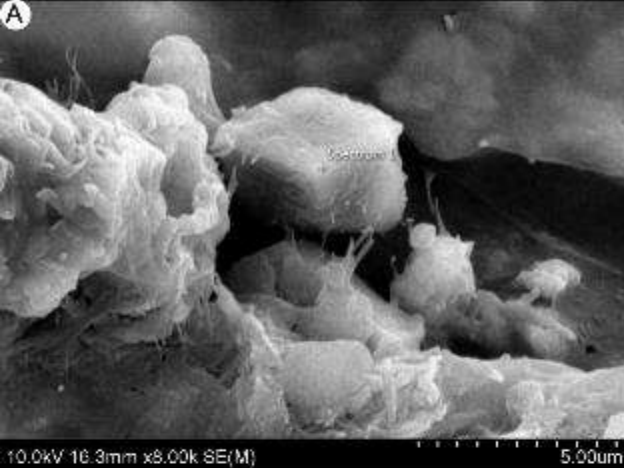
MINERALS PRECIPITATED IN MICROBIAL MATS



CALCITE CRYSTALLITE BUNDLES TO CRYSTALS

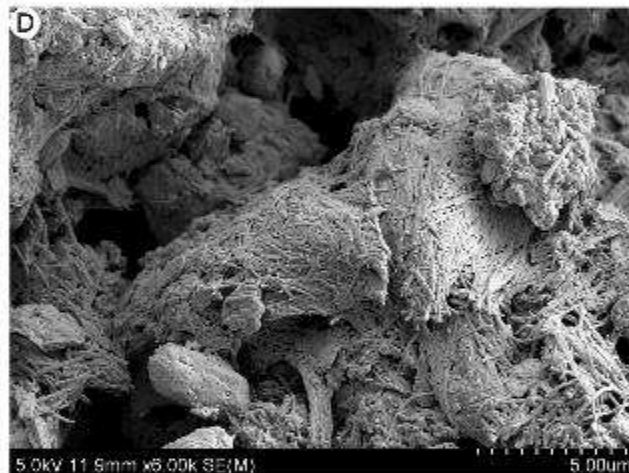
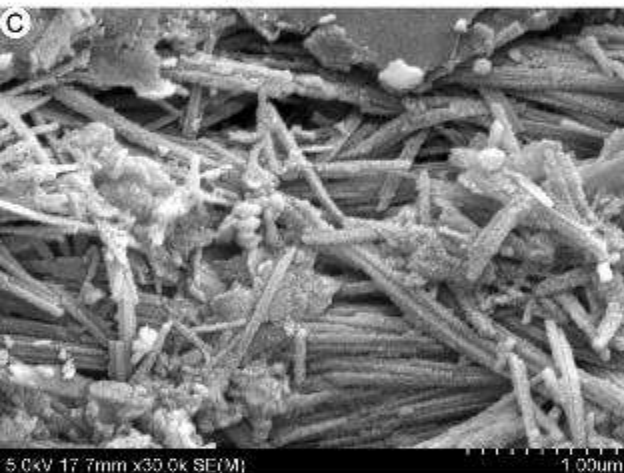
Perri, Tucker, Slovakiewicz et al. (2018) *Sedimentology*.





ALSO PRESENT:

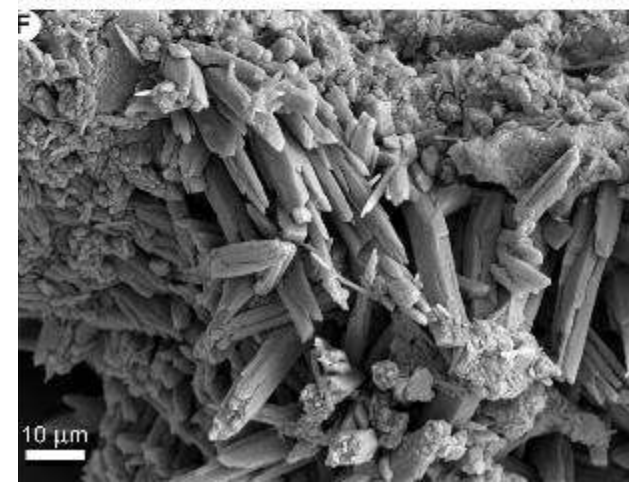
VHMC-DOLOMITE
rhombs (A, B)



PALYGORSKITE (C, E)

Clay fibres Mg 8.5%, Al 1.9 %,
Si 8.4%, Ca 5.4% O 61.7 %.
Felted mat of clay fibres
(?permineralised EPS).

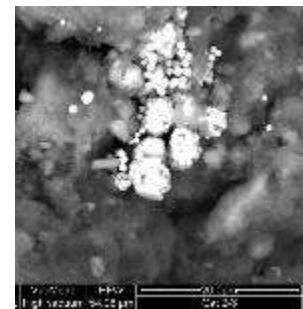
ARAGONITE (F)
needles



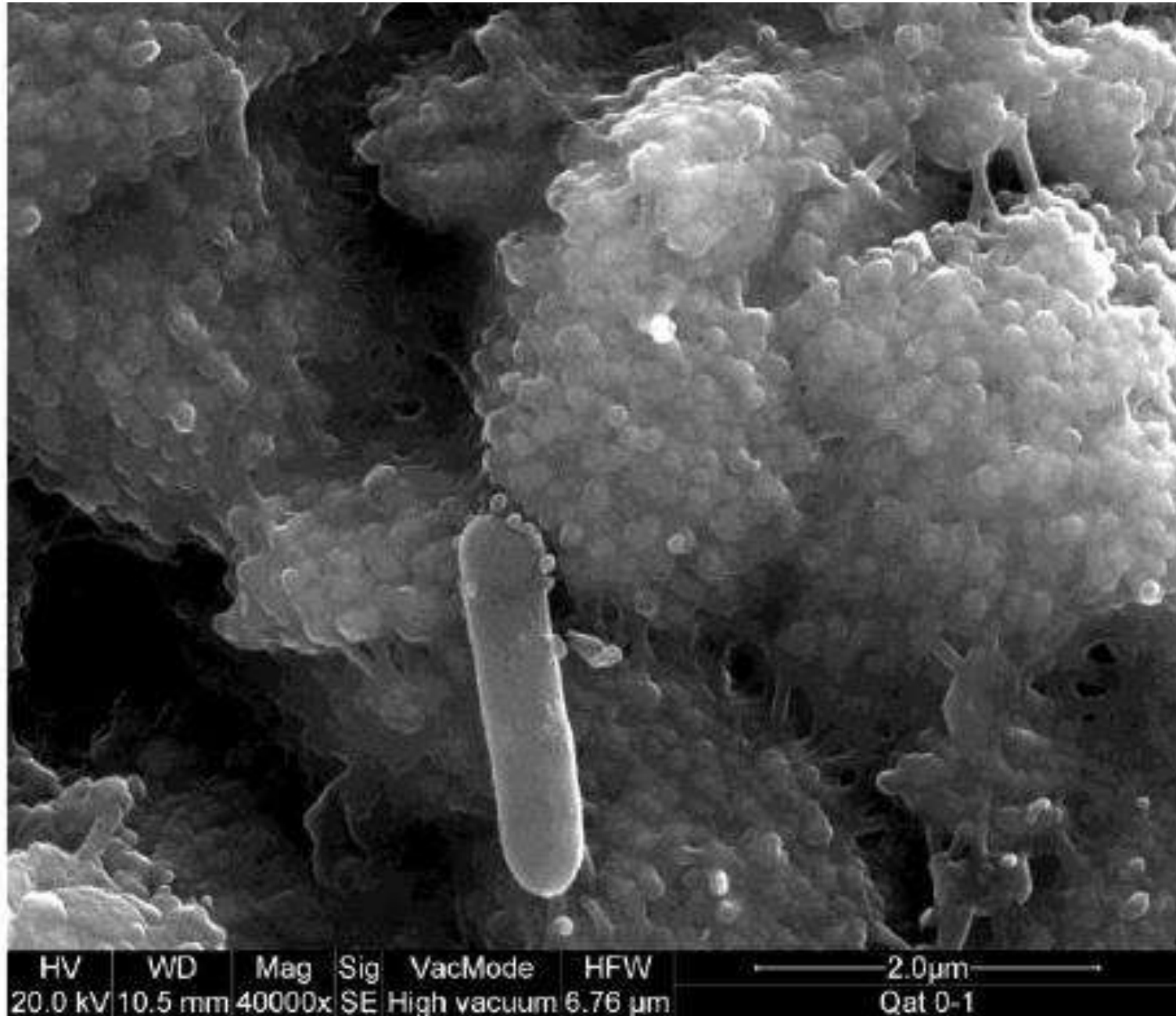
**MINERAL
PRECIPITATION
THROUGH MICROBIAL
PROCESSES
BUT HOW CRYSTALS
NUCLEATED ?**

**AND
PYRITE**

Perri, Tucker,
Slovak. et al. (2018)
Sedimentology.



SEM: NANOSPHERES IN UPPER MM OF MICROBIAL MAT



Nanospheres
enveloped in
EPSs forming
clusters,
i.e. peloids,
and attached
to bacterium.

Uniform size:
~100 nm.
Notice shape.

**Are these
viruses ?**

Other possible
origins –
bacterial vesicles
or intracellular
precipitates
or abiotic.

NANOSPHERES

cluster of nanospheres
(**calcified viruses**)

**Yes
we
are
viruses**

bacterium

Nanospheres
attached to
bacterium
and in a
cluster,
where they
appear to
amalgamate
into larger
features.

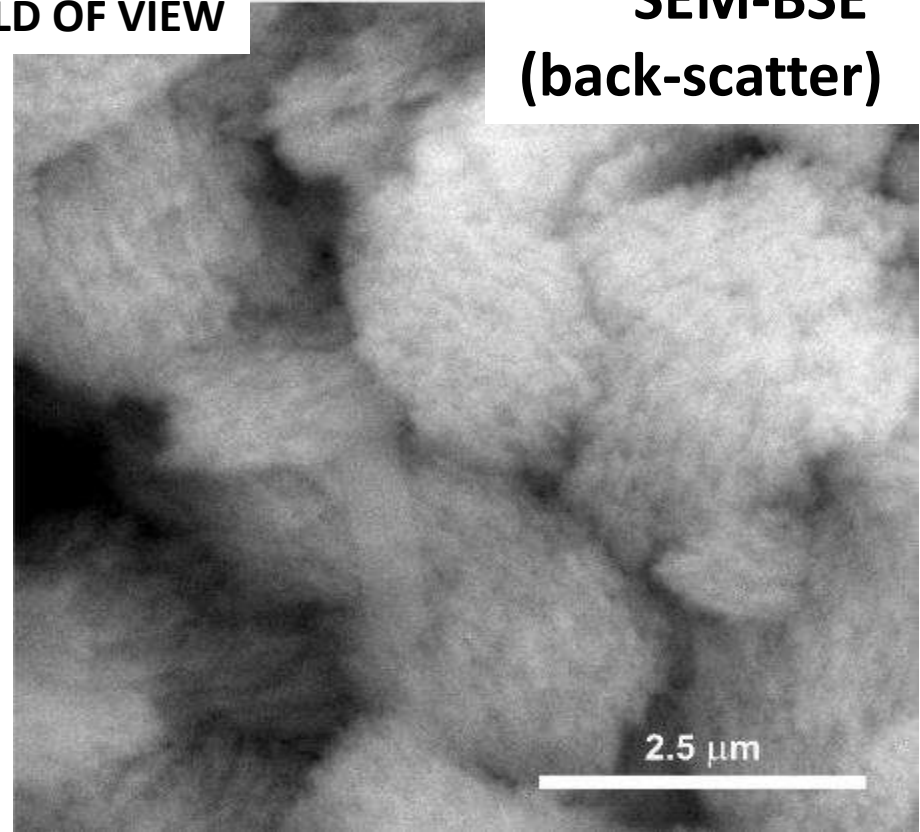
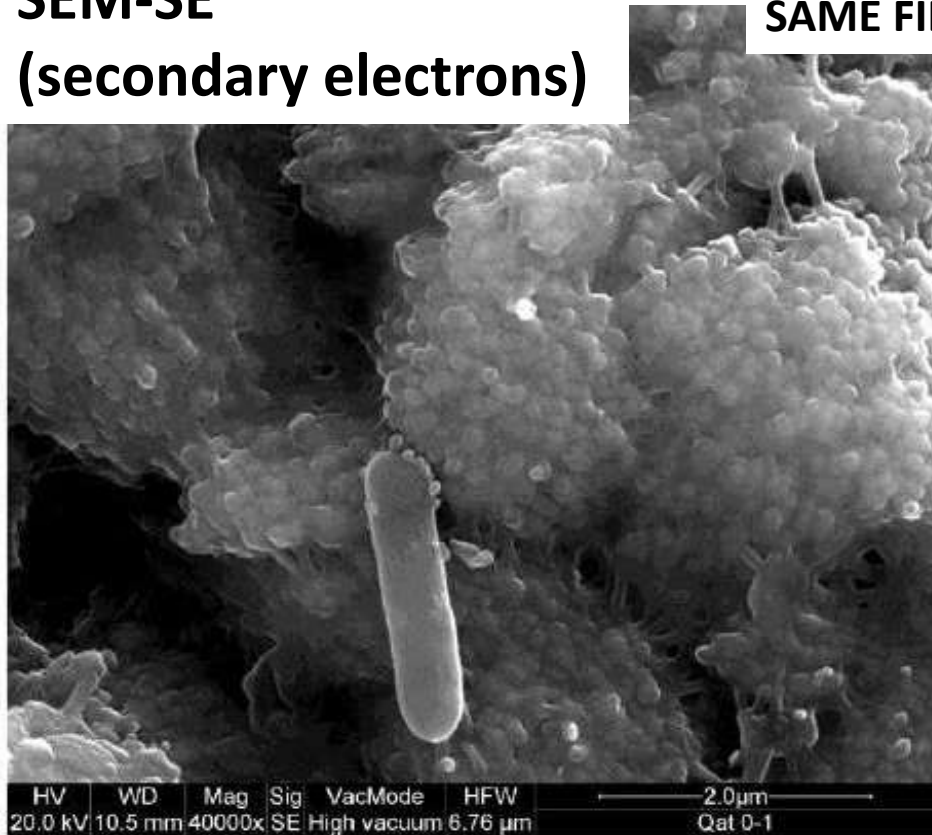
Notice shape:
icosahedral,
and
similar size.
**Very likely to
be viruses**
caught in the
act of
infecting.

NANOSPHERES IN UPPER MM OF THE MICROBIAL MAT

SEM-SE
(secondary electrons)

SAME FIELD OF VIEW

SEM-BSE
(back-scatter)



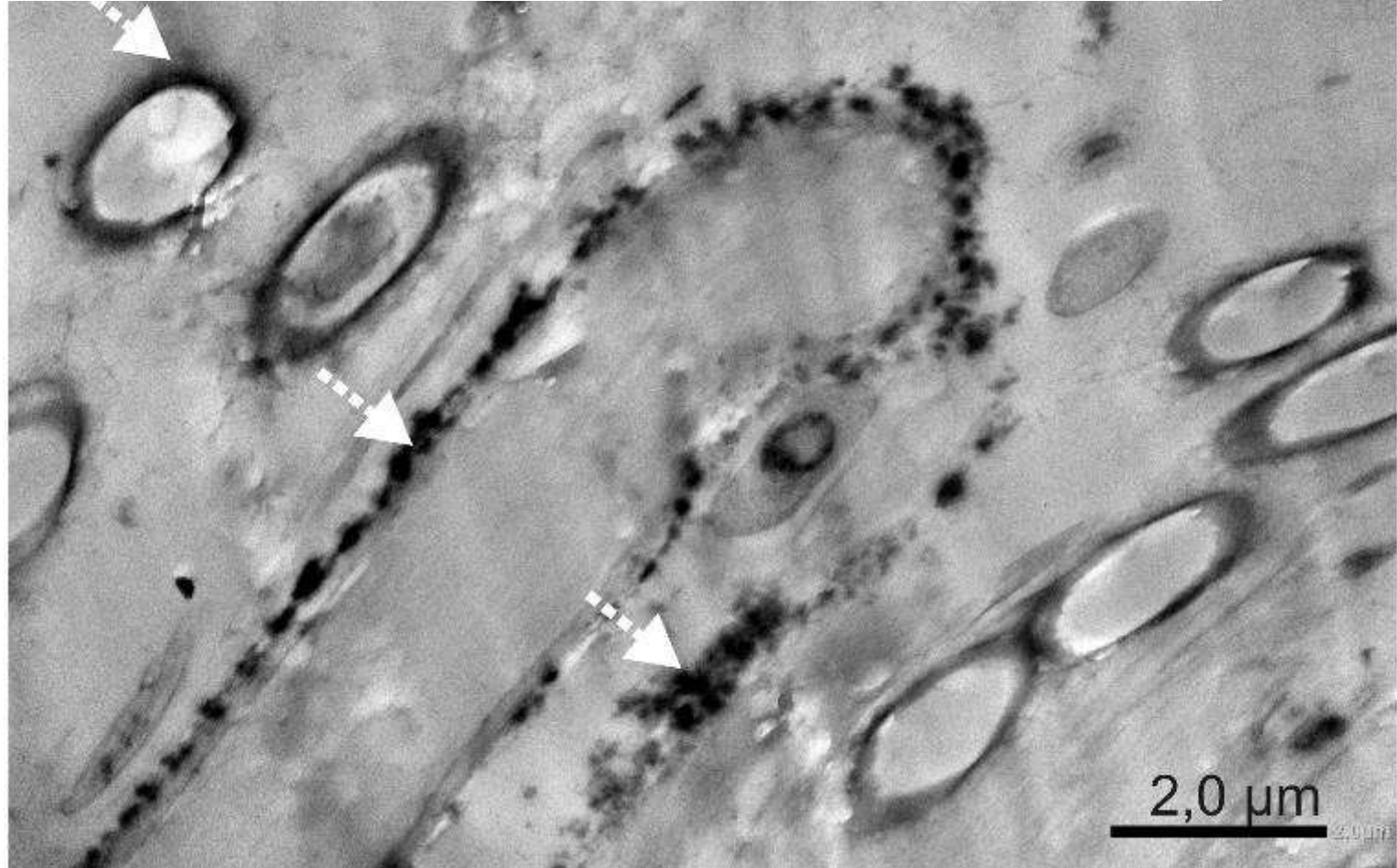
With BSE, organic matter (EPSs, bacteria) almost transparent (low atomic no. C,H,O), but where higher atomic no. elements (Ca, Mg, Si, Fe) see a lighter shade.

i.e. THEY ARE PER-MINERALISED

Nanospheres in clusters composed of nanocrystallite mineral bundles (=peloids)

BUT NOTE bacterium and attached **viruses** not permineralised (yet).

TRANSMISSION ELECTRON MICROSCOPE IMAGE

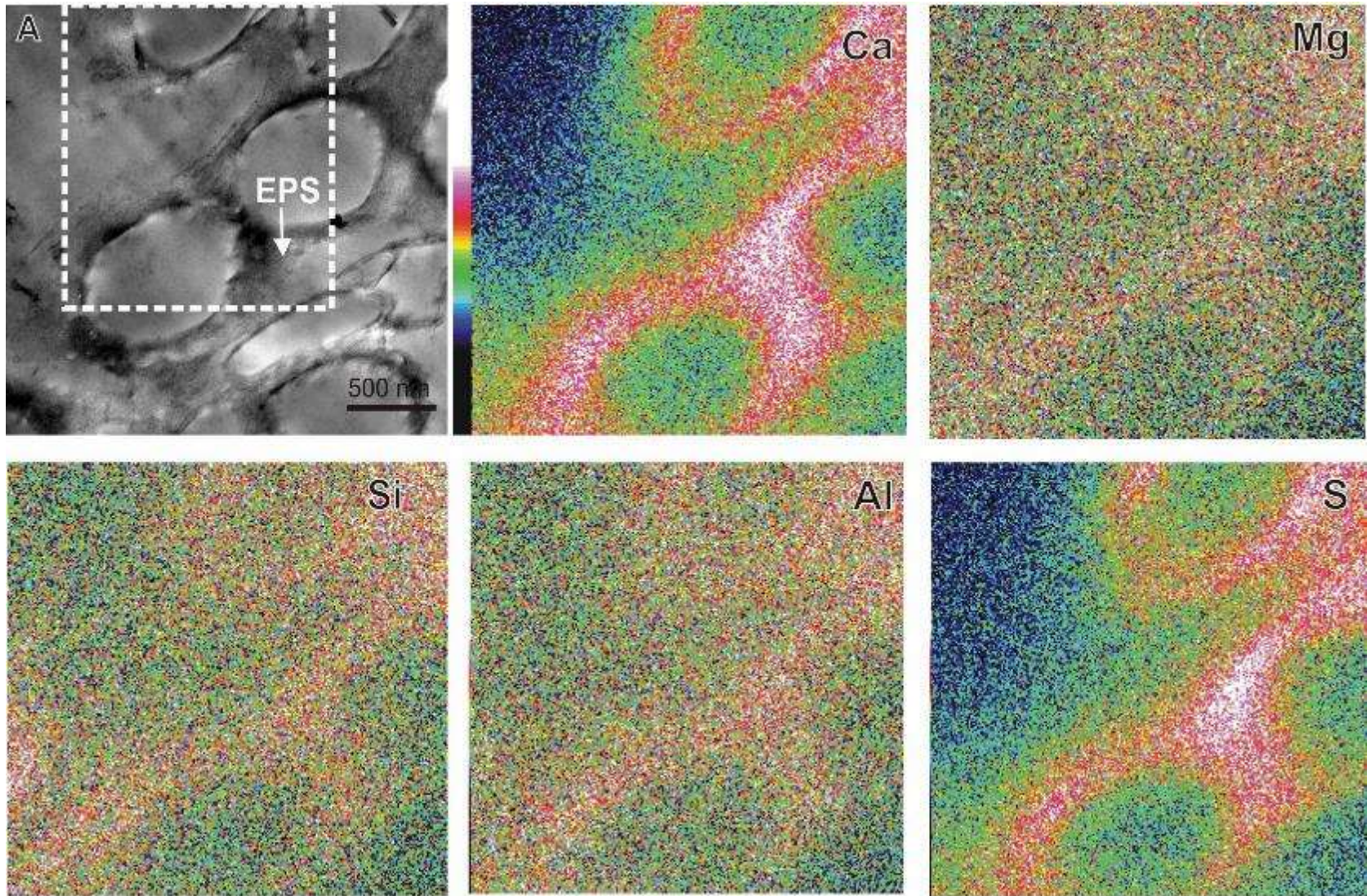


With TEM, electron-dense / darker areas (dashed arrows) = minerals. Shows early stage of mineral formation with initial sites of nucleation within the EPS that surround the bacterial cells.

SO - ARE THESE MINERALISED VIRUSES ?

Top mm of microbial mat, Mesaieed, Qatar.

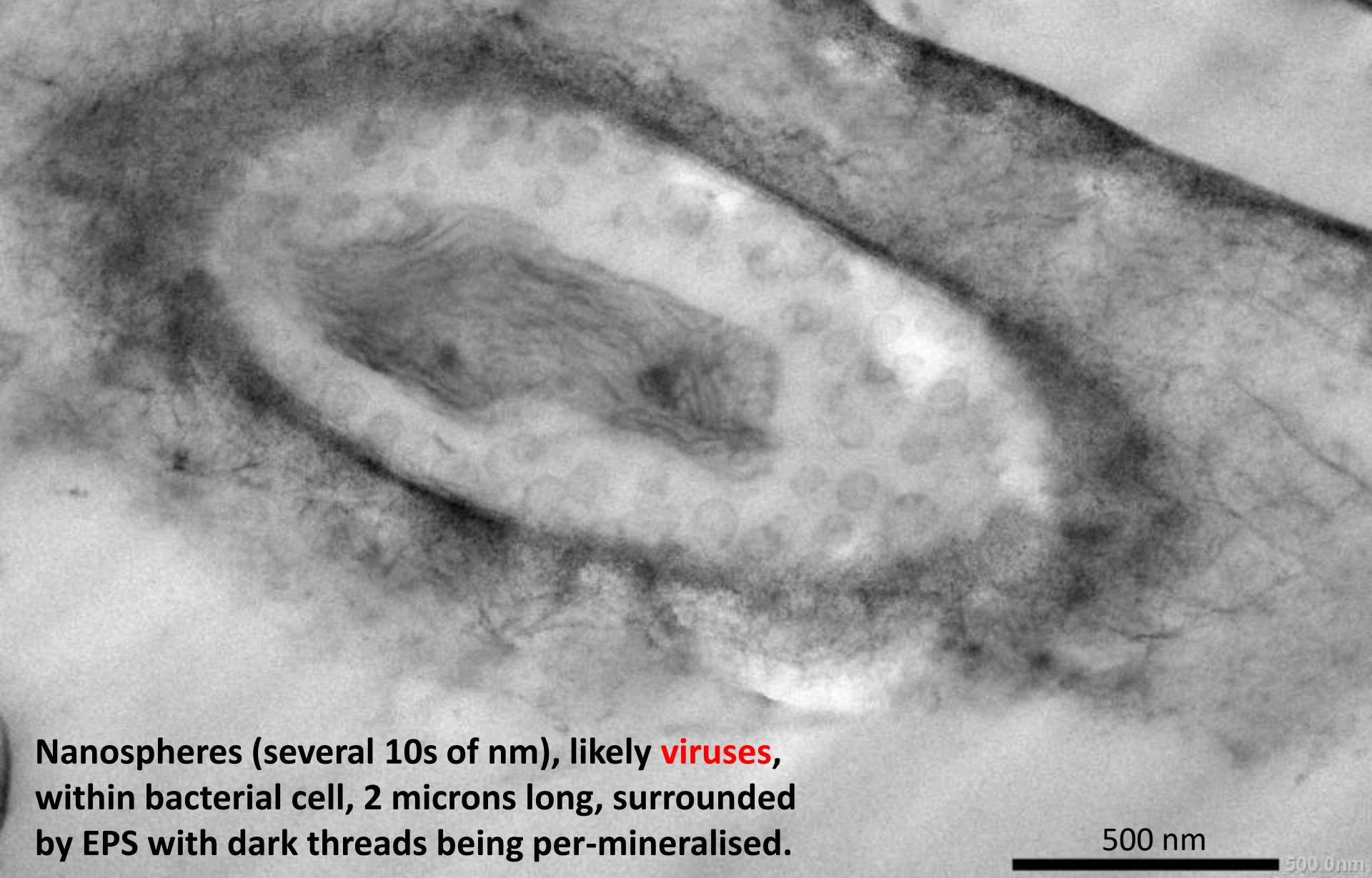
COMPOSITION OF AMORPHOUS MINERAL PRECIPITATES IN EPS



Elemental compositional maps of EPS around coccoid bacterial cells being mineralised: Ca, Mg, Si, Al, S..

C, O and N are not shown but form 95-98 % of the total element composition.

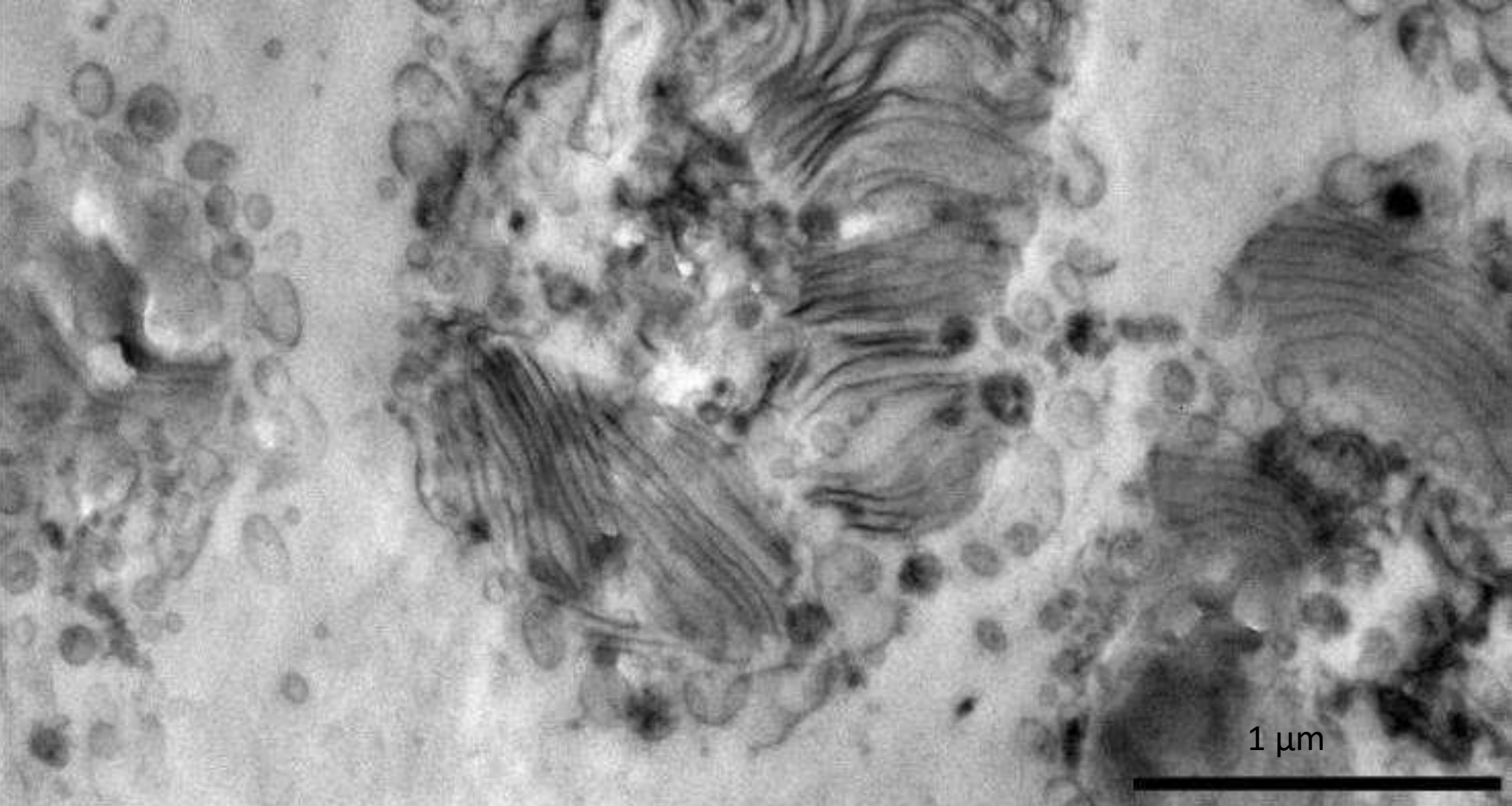
UPPER MM OF MICROBIAL MAT (TEM): **VIRUSES** PRESENT IN COCCOID BACTERIA



Nanospheres (several 10s of nm), likely **viruses**, within bacterial cell, 2 microns long, surrounded by EPS with dark threads being per-mineralised.

500 nm

500 nm

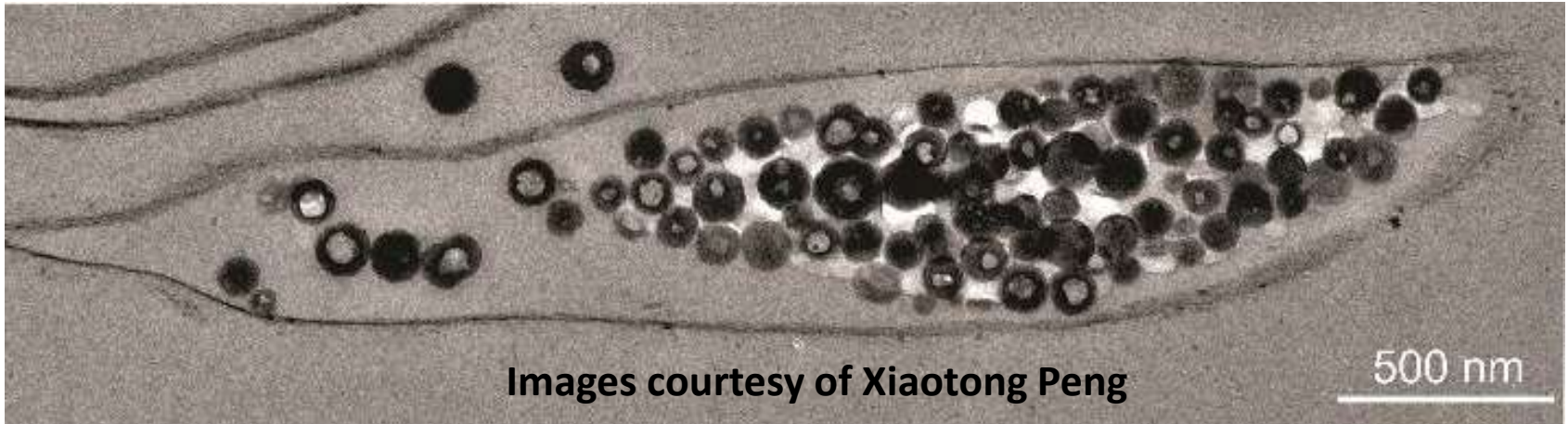


THUS: Nanospheres – which we interpret as **viruses - are becoming per-mineralised. Here seen within and *between* bacteria (with thylakoids – photosynthetic apparatus).**

Mineralised viruses i.e. FOSSILISED VIRUSES

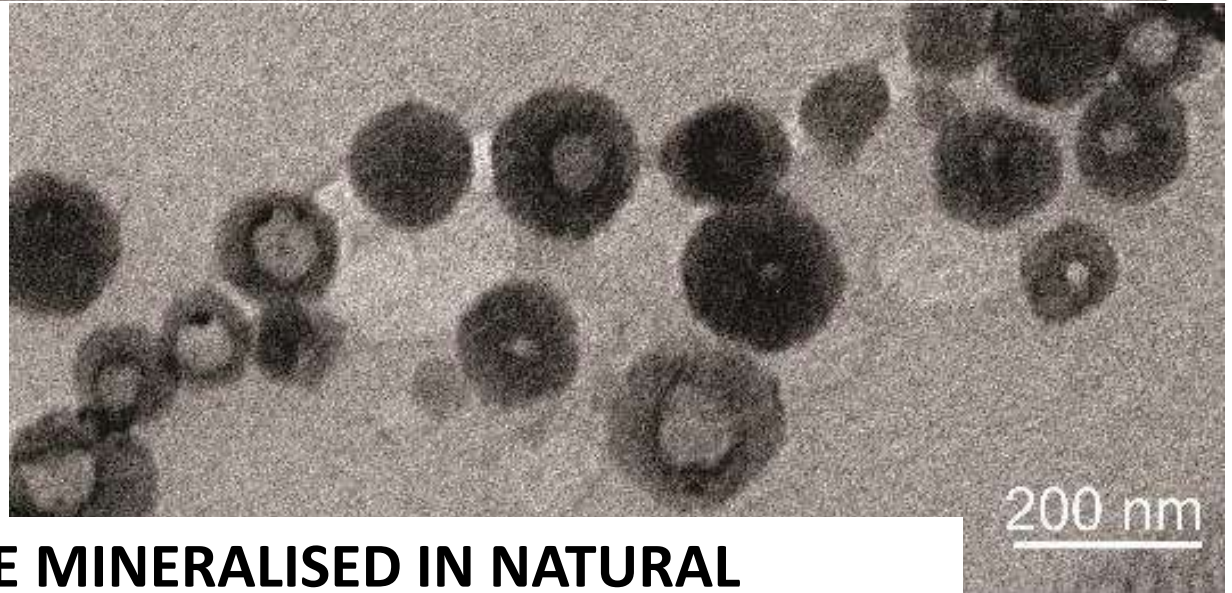
Permineralised viruses also reported by Pacton et al. 2014, 2015, and De Wit et al. 2015.

VIRUSES ALSO BEING MINERALISED IN HOT SPRINGS



Here **viruses** in a bacterium
being mineralised and
coated by silica.

Sinter, hot spring in China
Peng & Jones (2013)

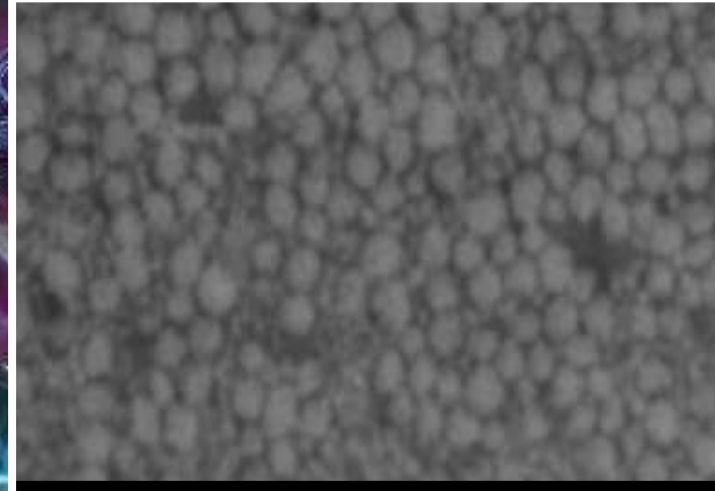
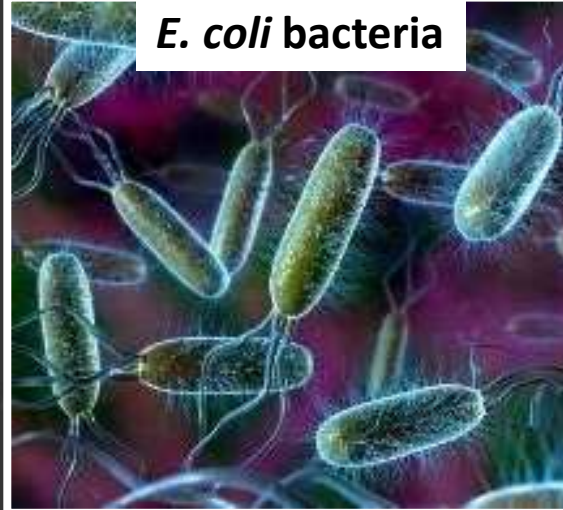


THUS **VIRUSES** CAN BE MINERALISED IN NATURAL
ENVIRONMENTS, **THAT IS FOSSILISED – NOW SOME EXPERIMENTS**

OUR EXPERIMENTS WITH **VIRUSES** AND CARBONATE PRECIPITATION

Viruses (size 100 nm) extracted from *Escherichia coli* bacteria (size 1-5 μm)
E. coli common bacteria – in sewage, rivers, lakes, coasts... and stomachs !

STEM images



200 nm — WD=3.6 mm EHT=30.00 kV Signal=STEM

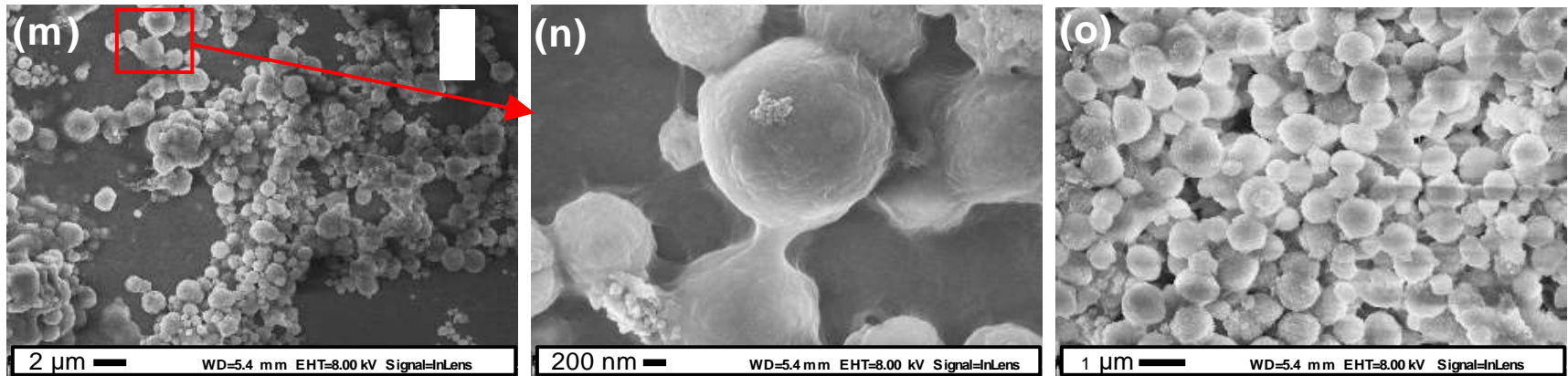
200 nm — WD=3.6 mm

Before experiments: phage capsids +/- tails, often coalescing to form larger accumulations, with loose quasi-regular structure.

Experiments conducted by adding more and more Na_2CO_3 to CaCl_2 solution with a billion **viruses** per ml (also control experiments with no phages).

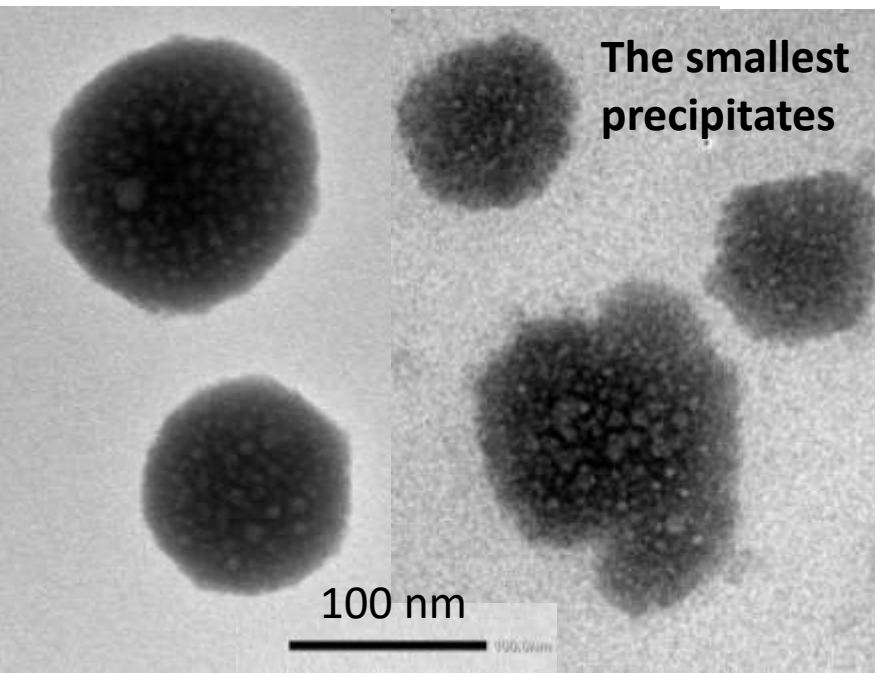
Results just published in *Geochim Cosmochim Acta* (Slovakiewicz et al. 2021).

phage concentration of 10^{11} mL⁻¹



PRECIPITATES OBTAINED DURING EXPERIMENTS WITH **VIRUSES**

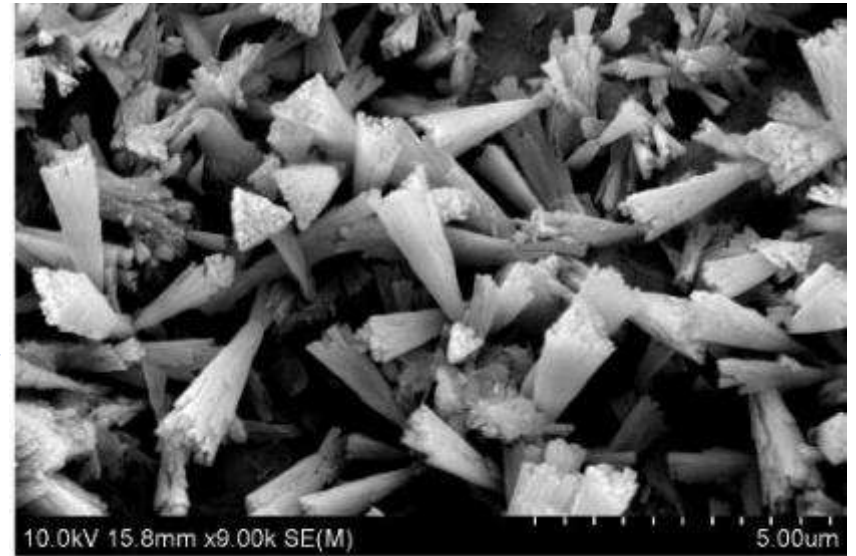
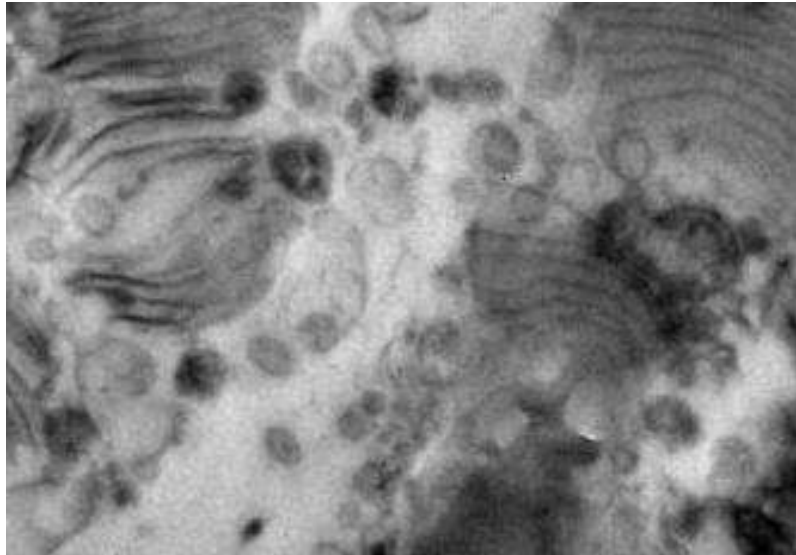
we get nano-micro spheroids, dumbbells, tiny particles (80-100 nm) resembling permineralized capsids. Also coalescing of particles.



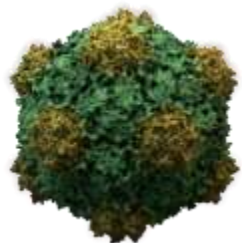
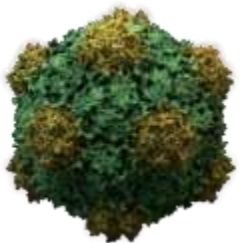
Control experiment precipitates only calcite, BUT with **viruses: 70 % vaterite**, 30 % calcite.
(vaterite unstable CaCO_3)

THUS **VIRUSES** DO HAVE AN INFLUENCE IN CARBONATE PRECIPITATION in terms of crystal/particle size, shape, coalescence and mineralogy.

**SO VIRUSES BEING MINERALISED AND
THEY INFLUENCE CARBONATE PRECIPITATION.
WE SUGGEST THEY PROVIDE **NUCLEI** FOR FURTHER
CARBONATE (& other) PRECIPITATION**

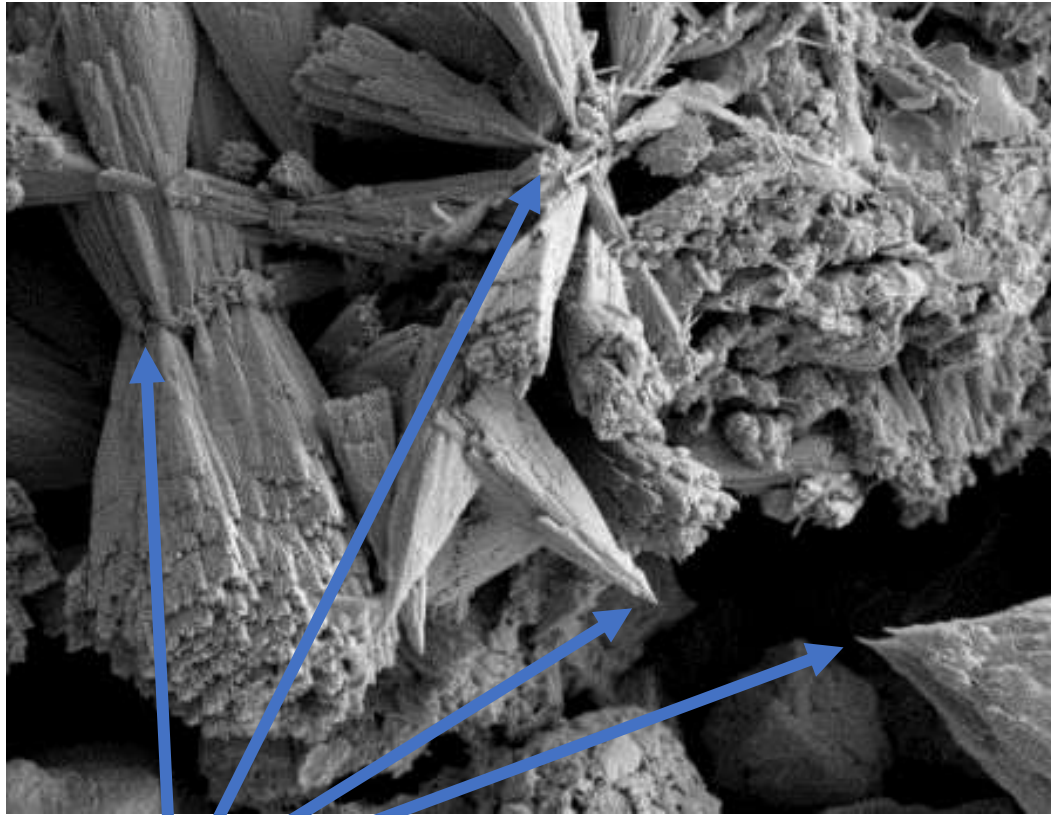


**VIRUSES THE SEEDS FOR
CRYSTAL GROWTH ?**



HOW DO CRYSTALS GROW ?

HOW NUCLEATED ? SPONTANEOUS OR SEEDED ?



Can see nucleation / initiation points for calcite. But what started them off ?

Direct precipitation (*homogeneous/spontaneous nucleation*) not simple;
(some say impossible) seeds/nuclei are needed (that is *heterogenous nucleation*).

**We suggest the initial precipitates are
MINERALISED VIRUSES (& EPS): THE NUCLEI FOR PRECIPITATION.**

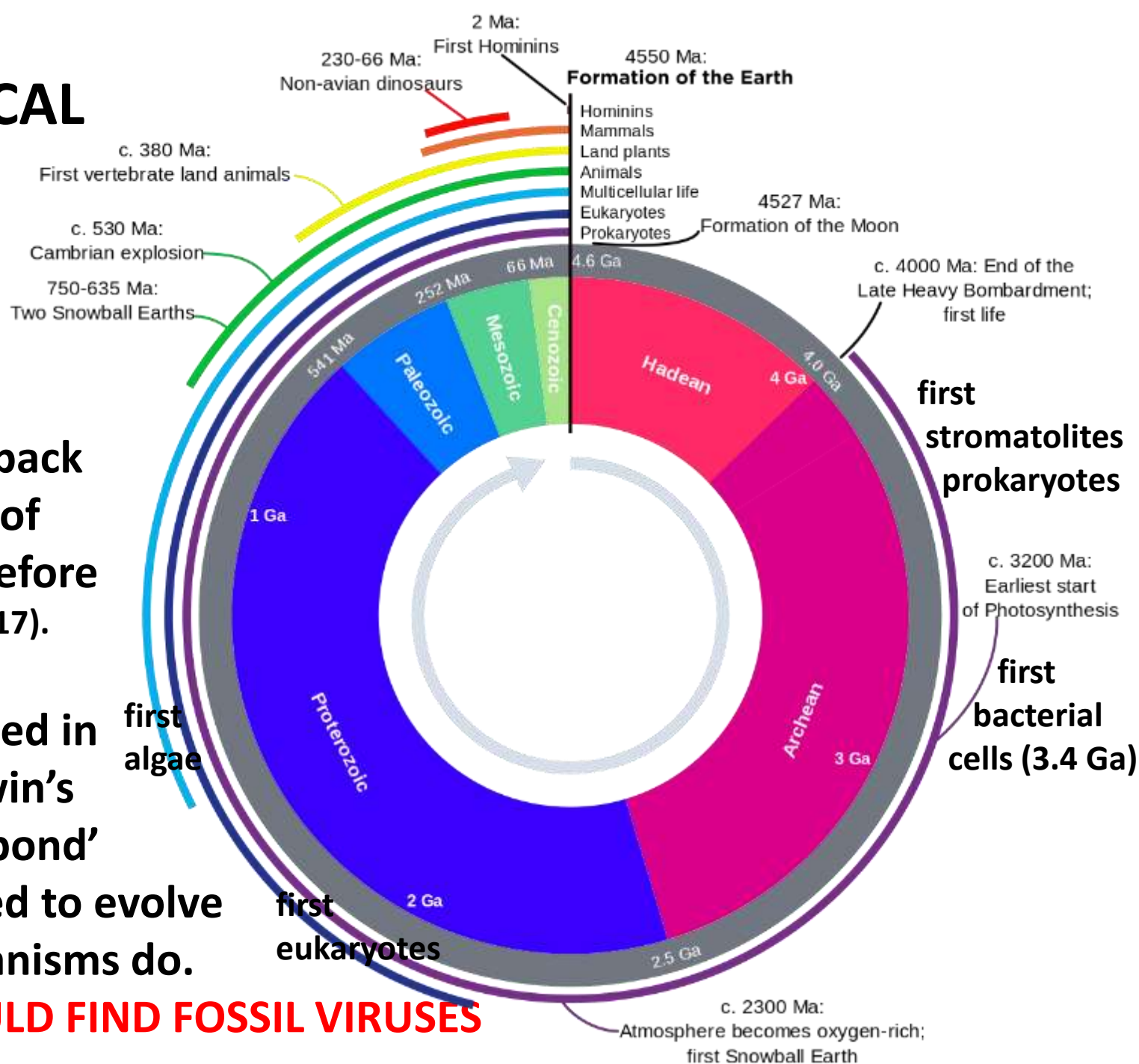
GEOLOGICAL RECORD OF LIFE

VIRUSES

probably go back
to the origin of
life – if not before
(e.g. Koonin 2017).

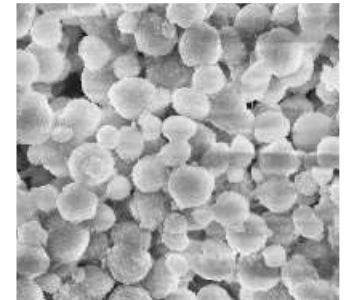
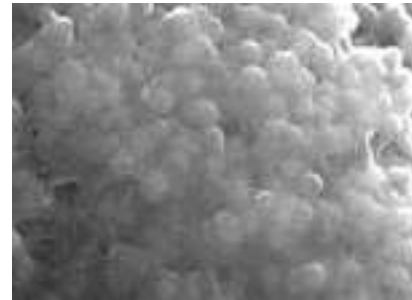
Viruses created in
Charles Darwin's
'warm little pond'
and continued to evolve
as other organisms do.

SO WE SHOULD FIND FOSSIL VIRUSES



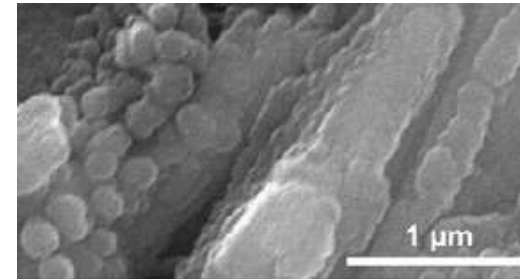
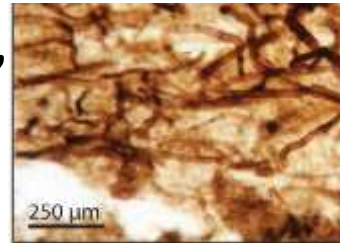
SO HOW ARE WE GOING TO RECOGNISE **FOSSIL VIRUSES** ?

SIZE: range 50-300 nm
BUT large numbers
all of a similar size



SHAPE: nanospheres, **icosahedral**, but there is a preservation issue:
mineralised viruses may act as nuclei for further precipitation of the
mineral fossilising them **AND** they coalesce

CONTEXT: associated with bacteria,
so look in stromatolites

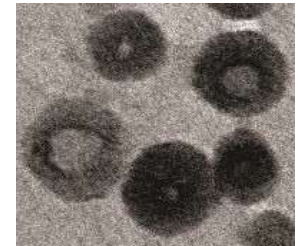


MINERALOGY:

carbonate: calcite, aragonite, dolomite **BUT** recrystallisation and
overgrowth, so may be difficult to recognize.

silica: resistant to recrystallisation, sinter, hot springs.

phosphate: lots of microbes, high nutrient supply.



iron minerals: BSR, suboxic-anoxic, siderite, ferrihydrite, pyrite

Viruses should be present throughout the geological record.

EXAMPLES

NANOSPHERES PRESENT IN PRECAMBRIAN ROCKS

Here in
Palaeoproterozoic
chert.

Silicified microfossils
Labrador, Canada, 1.88
Ga, showing microbial
filaments and numerous
nanospheres.

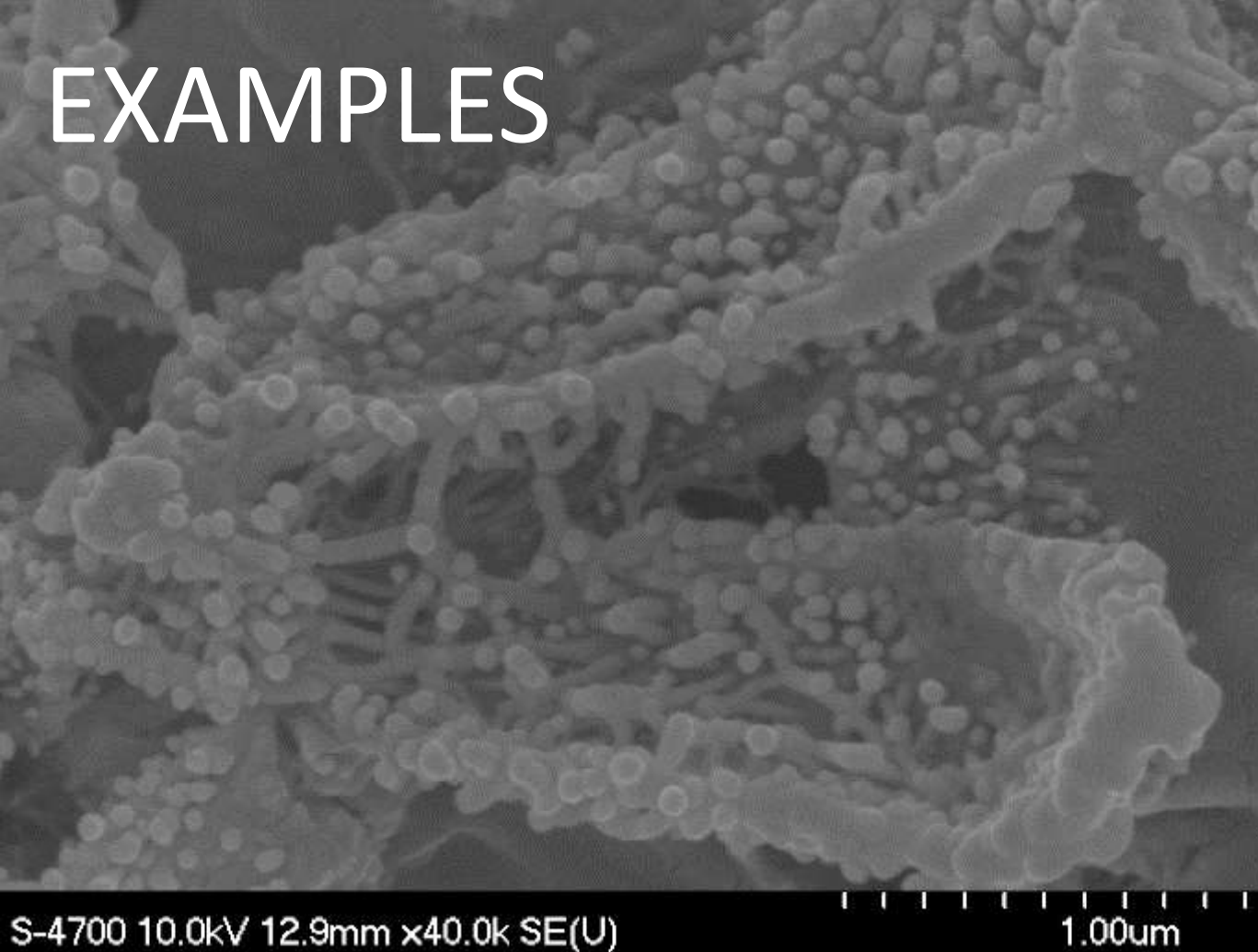


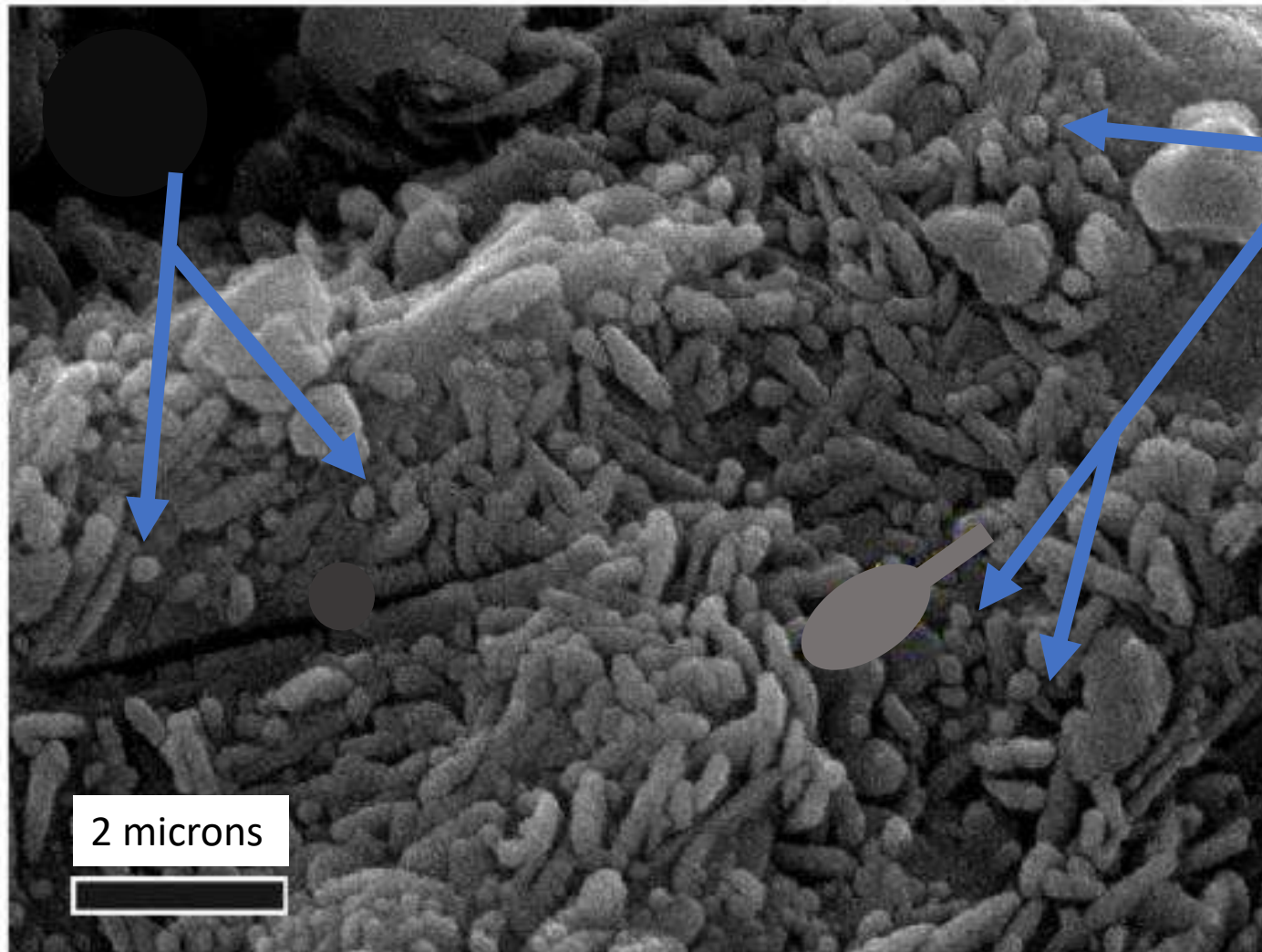
Image courtesy of Cole Edwards

THESE NANOSPHERES COULD BE SILICIFIED VIRUSES.

Note their size – really tiny, 50-70 nm, and shape (not perfect spheres).

Edwards et al. (2012) Precambrian Research.

AND HERE IN A PALAEOPROTEROZOIC PHOSPHORITE:



Numerous
nanospheres
amongst the
relics of
microbial
filaments.

**ARE THESE
PHOSPHATISED
VIRUSES ?**

Notice their
shape and
similar size.

1.85 Ga Michigamme Fm, Labrador, Canada. Palaeoproterozoic tidal-flat phosphorite. Hiatt et al. (2015).

BUT: IS THERE A SIGNIFICANCE FOR VIRUSES BEYOND MICROBIAL MATS and PANDEMICS ?



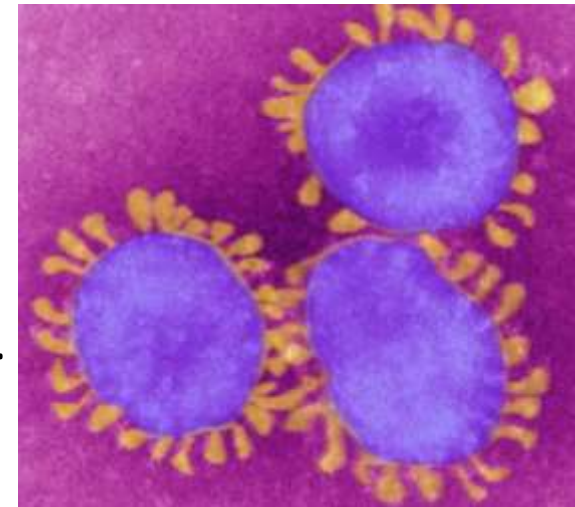
VIRUSES ARE NOT ALL BAD: THEY DO MUCH GOOD IN THE WORLD
in terms of biogeochemical cycles and biological diversity

Recycling of organic matter: Through infection and cell lysis, viruses are responsible for the death of nearly 80% of all prokaryotes (i.e. bacteria and archaea), so releasing nitrogen, carbon and phosphorus, producing dissolved organic matter, which stimulates further microbial growth.

Nutrient supply: returning organic matter to the base of the food chain.

Marine viruses appear to **prevent bacterial population explosions**, allowing a broad diversity of species to coexist.

Viruses inducing evolution of their hosts, so improving the gene pool; signatures of that evolution present in the genetic code of organisms.



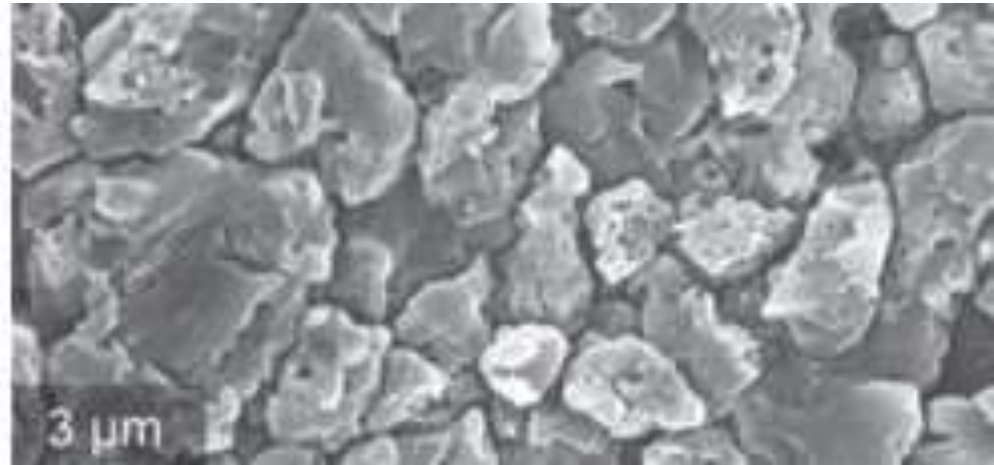
BUT WHAT ELSE – IN EARTH SCIENCES ?

FINE-GRAINED LIMESTONES COMMON THRU GEOLOGICAL RECORD:

BUT ORIGIN OF THE LIME MUD ?

Planktic cyanobacterial blooms and their **associated viruses** providing the seeds for carbonate pptn.

SOLNHOFEN



Solnhofen Limestone Upper Jurassic Bavaria.

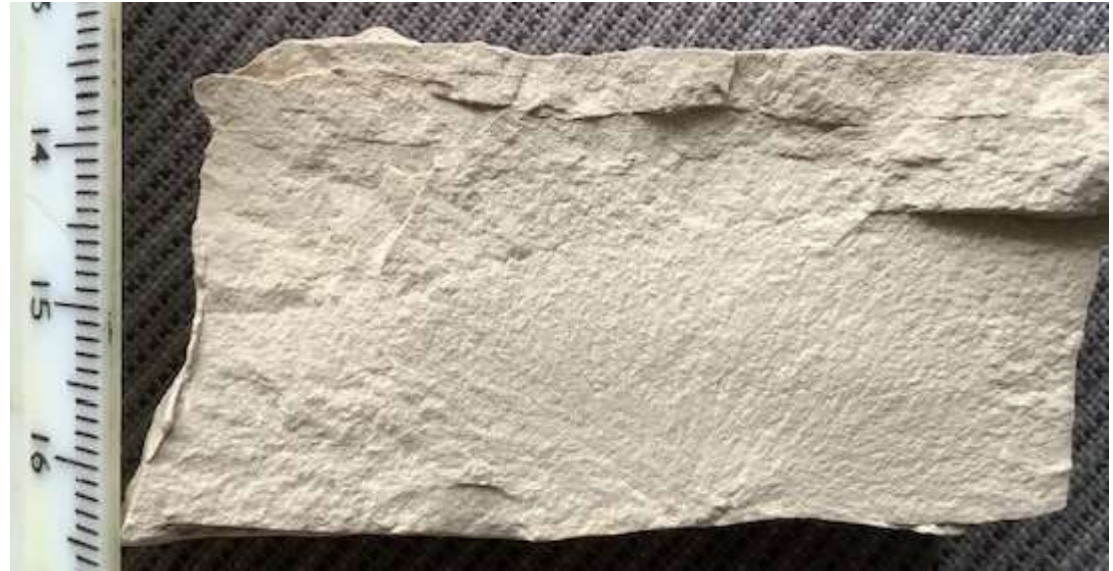
PALAEOPROTEROZOIC, CHATTISGARH BASIN, INDIA



LIME MUD PRECIPITATED VIA PLANKTIC MICROBES AND THEIR VIRUSES

HERE IN BATH: Rhaetic, Upper Triassic.

White Lias – Saltford, Keynsham – fine grained micritic limestone



Temporary exposure Keynsham; also Saltford, and old Hartwell Garage, Upper Bristol Road, Bath. Useful building stone (and Roman tesserae).

An extremely fine limestone: but how precipitated ?

simply out of seawater – unlikely – would need a trigger.

Could that be a bloom of microbes with their viruses ?

induced by climate, dust, upwelling, river flood increasing nutrients ?

ARE THESE VIRALITES ?

WHITINGS

Viruses could provide the seeds for mineral precipitation
as in lime mud precipitation

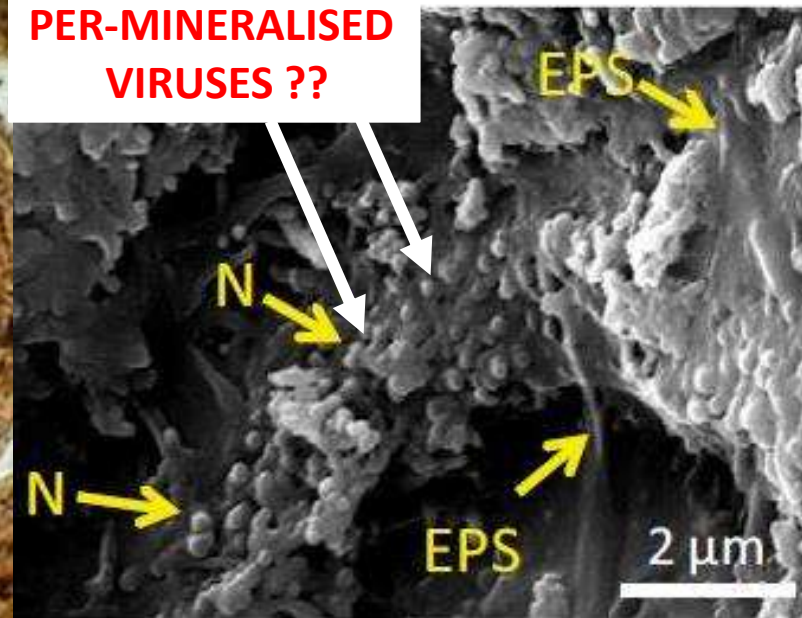


WHITINGS in the Bahamas: off-platform currents bringing supersaturated seawater on to GBB
(Purkis et al. 2017) AND/OR effect of wind-blown Saharan dust (Swart et al. 2014),
inducing CaCO_3 precipitation through photosynthetic activity of planktic cyanobacteria
PLUS THEIR ASSOCIATED VIRUSES.

AND OIDS



**PER-MINERALISED
VIRUSES ??**

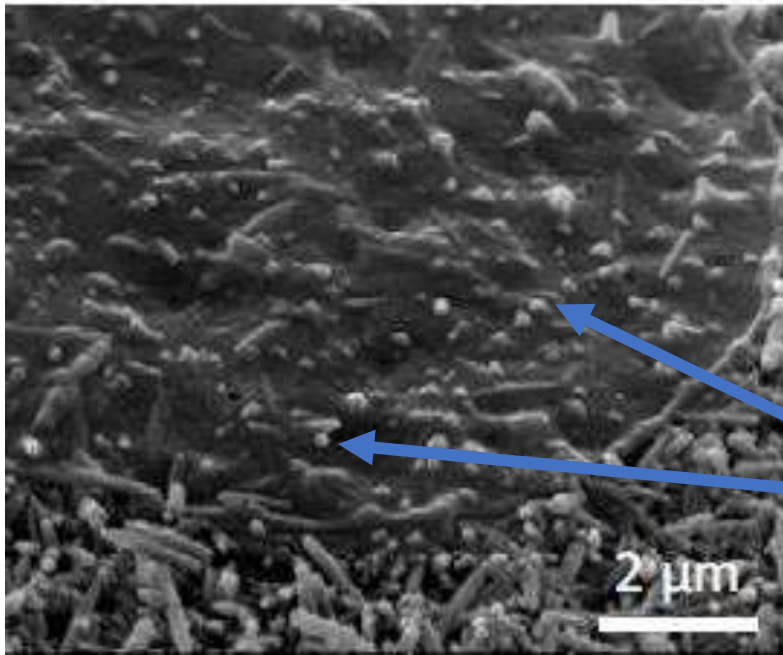


OIDS NOW SUGGESTED AS MICROBIAL IN ORIGIN (Diaz et al. 2017, 2019)
because bacteria and EPS found.

BUT:

Ooids are full of nanospheres (N):

Scattered and clusters of nanograins (N) associated with mucus (EPS). Composition is ACC (amorphous calcium carbonate), converts to calcite-aragonite.



**ARE THESE PER-MINERALISED
VIRUSES ??**

From Diaz et al. (2017, 2019) Geology, ESR.

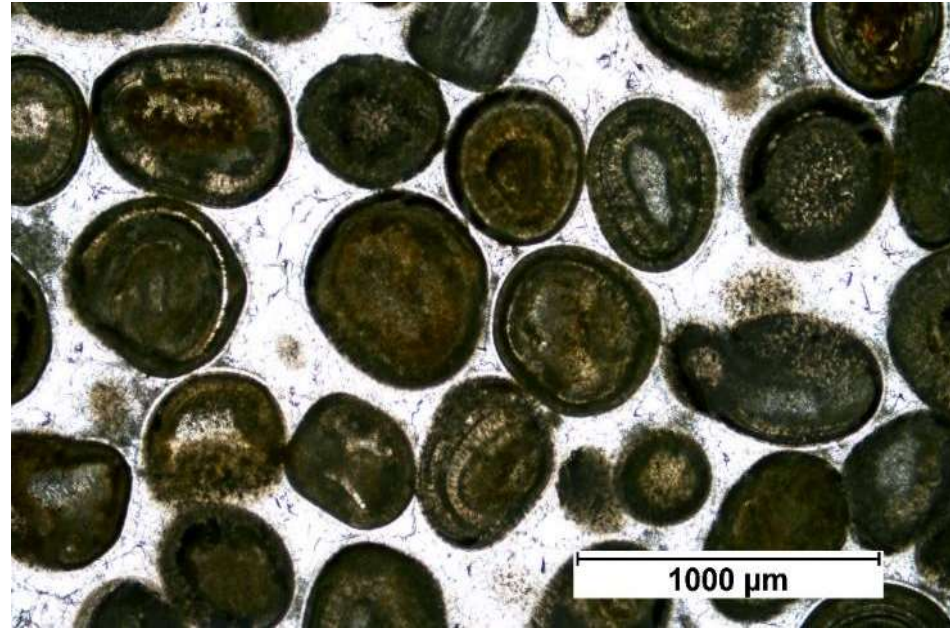
AND MANY OOLITES IN THE JURASSIC HERE:

BATH STONE / GREAT OOLITE (& INFERIOR OOLITE)

ALSO IN THE CARBONIFEROUS – GULLY OOLITE (AVON GORGE)



**BATH OOLITE oolitic grainstone
with few bioclasts (shells)**

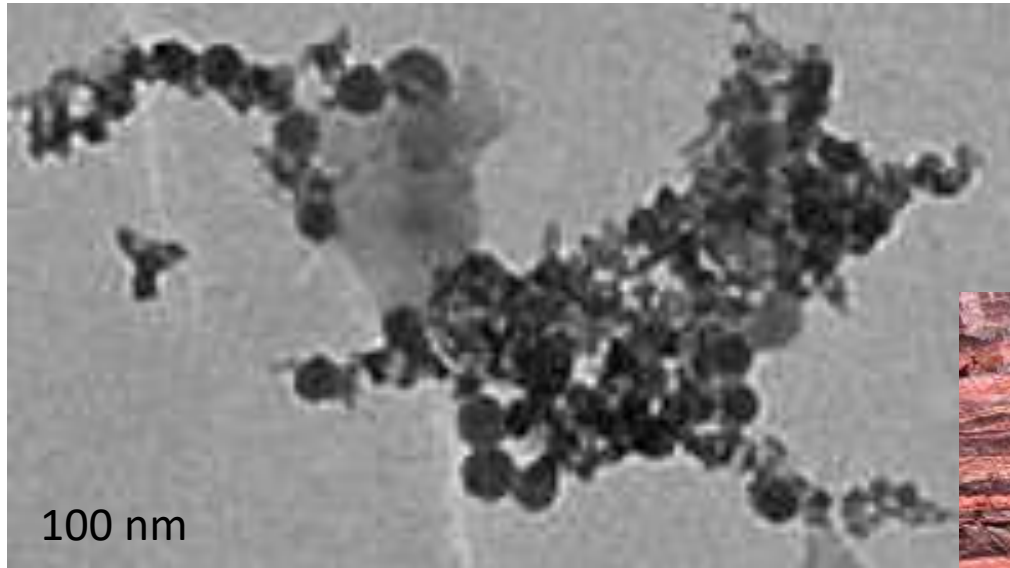


**Another
VIRALITE ?**

**Cross-bedded shelly oolitic
grainstone + burrows**



AND **VIRUSES** ASSOCIATED WITH IRON PRECIPITATION



TEM images of iron mineral particles associated with *Siphoviridae* viral capsids.

Virus mineralization at low pH in the Rio Tinto, Spain (Kyle et al. 2008).

Viral capsids with iron-bearing minerals sorbed to their surfaces.

IMPLICATIONS FOR ALL IRON FORMATIONS

e.g. BIFs IN THE PRECAMBRIAN

Viruses and ore deposits

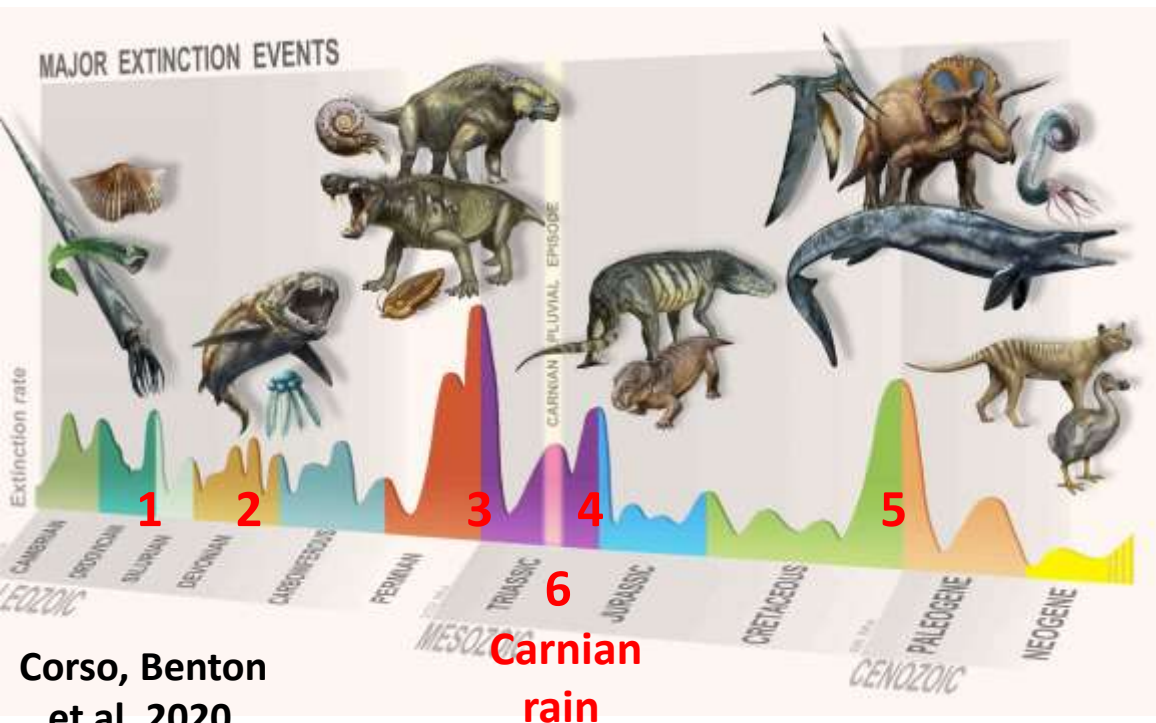


ANOTHER VIRALITE !!

AND FINALLY: WHAT ABOUT **VIRUSES AND EXTINCTION** ? COULD THEY PLAY A ROLE THERE ?

Two types of extinction: mass extinctions and background extinctions.

With many organisms, extinction events every few million years: coccoliths, foraminifera, ammonites etc: hence used in BIOSTRATIGRAPHY. Suggested by Emiliani (1993) background extinctions caused by a host-specific viral action, a fundamental part of the process of evolution, related to an environmental 'upset'.



Mass extinctions: catastrophic events: meteorite, volcanism, climate, sixth just recognised. BUT Background extinctions more subtle, more species-specific, environment-driven, ? toxins, **viruses** very likely involved.

THE FIELD IS OPEN – FOR FURTHER RESEARCH

CONCLUSIONS

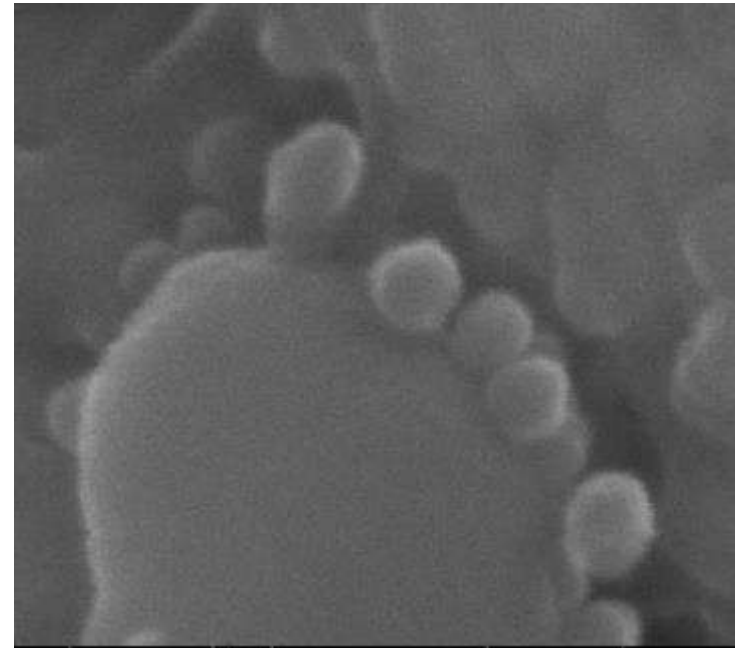
Viruses are completely neglected in Earth Sciences – they must have played many roles...

Viruses do become fossilised and can be found back to Precambrian rocks

Viruses can be the seeds for carbonate (and other mineral) precipitation – **viruses can form rocks**

The role of **viruses** in species extinction needs to be explored

VIRUSES are the new frontier in Earth Sciences ...



**And this is why I am a
sedimentologist !!**

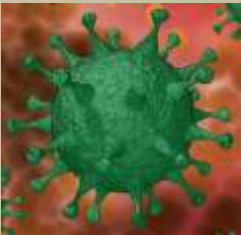
Fieldwork, Los Roques, Venezuela

THANK YOU

FOR FURTHER INFORMATION SEE:

Tucker, M.E. (2020) Fossil viruses. *Geology Today*, 36, 56-60.

Slowakiewicz et al. (2021) Expts with viruses. *GCA* 292.





**An icosahedral dice –
from Ptolemaic Egypt,
a Hellenistic dynasty
founded 305 BC, until 30 BC
when Cleopatra took over.**

Background vs Mass Extinction

- **Mass Extinctions:** Only 4%

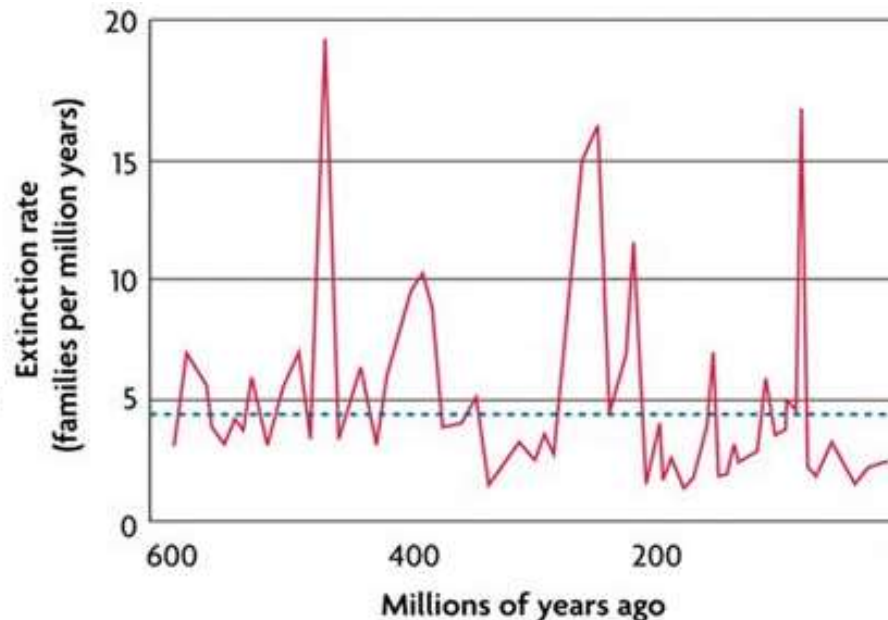
- Mass Extinctions kill off a very large % of the species on the planet at a given time, but because they are short in duration, they comprise a small % of animals that ever lived on the planet

- **Background Extinctions: 96%**

- Extinctions are always happening in the background, and over a long long period of time, these extinctions add up to a lot of species extinctions

- **99% of species that have lived are extinct**

----- Approximate background extinctions
 — Extinction rate



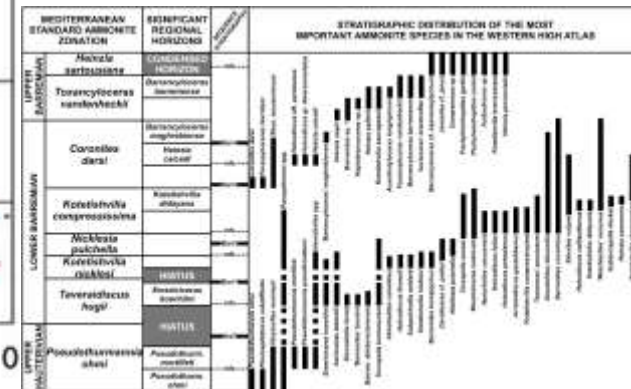
When extinction rate is plotted against time, mass extinctions appear as periodic peaks rising above background extinction levels.

Background Extinctions:

- **Cause:** Local changes in environment
 - Forest fires, habitat destruction.
- Affects a few species in a small area
 - Less severe.
- Occur at roughly the same rate as speciation

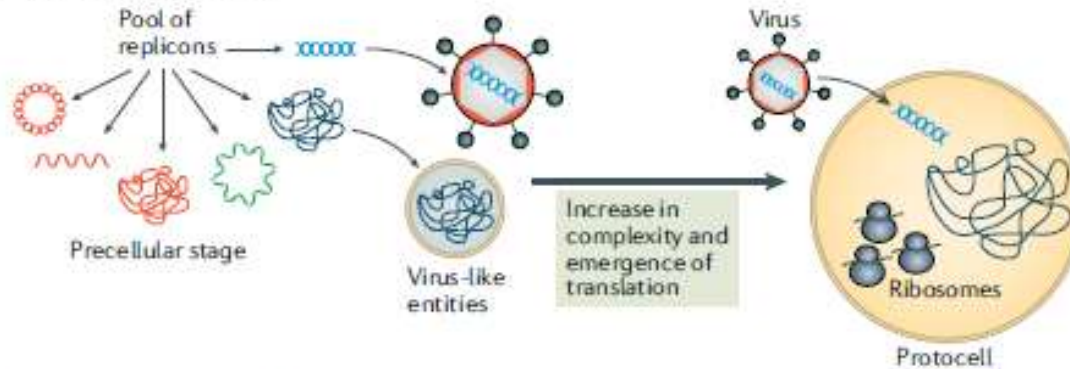
Mass Extinctions:

- **Cause:** Catastrophic events
 - Ice Age, Meteorites
 - Destroys many species at global level
 - Very severe
 - Rare but much more intense
 - At least 5 mass extinctions in last 600 million years
- 

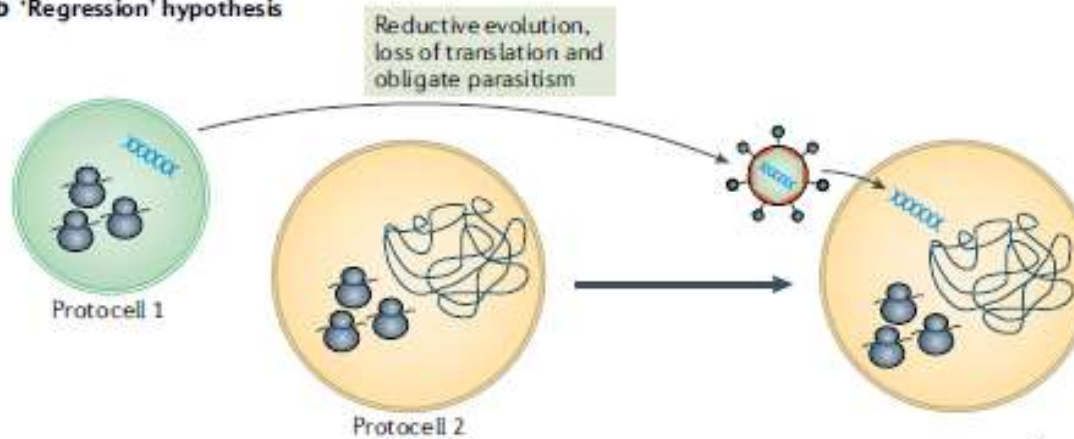


EXTINCTIONS – AN EVERYDAY STORY IN EARTH HISTORY

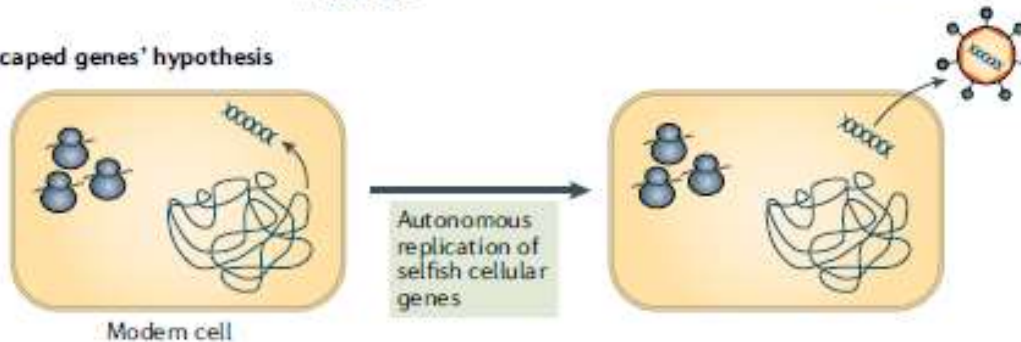
a 'Virus early' hypothesis



b 'Regression' hypothesis



c 'Escaped genes' hypothesis



The three major scenarios for the origin of viruses.

A: The 'virus early' hypothesis assumes that viruses evolved from early replicative elements that preceded the first cellular life forms.

B: The 'regression' hypothesis suggests that viruses emerged through the degeneration of cells that then assumed a parasitic lifestyle.

C: Finally, the 'escaped genes' hypothesis proposes that cellular genes acquired the ability for 'selfish' replication and spread.