Biomimetics: Learning from Nature for an Innovative Future

23. Japanese-German Symposium

April 20 and 21, 2018 in Frankfurt

11:00-11:30

The NanoSuit®, Imaging living organisms in a scanning electron microscope

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Convener: Prof. Dr. Olaf Karthaus

Humans are called as a vision animal.

Paradigm shift by new innovation.

1600～ Light microscope Netherland

Paradigm shift life science （Organisms are composed of cells.）

1838
M.J.Shleiden

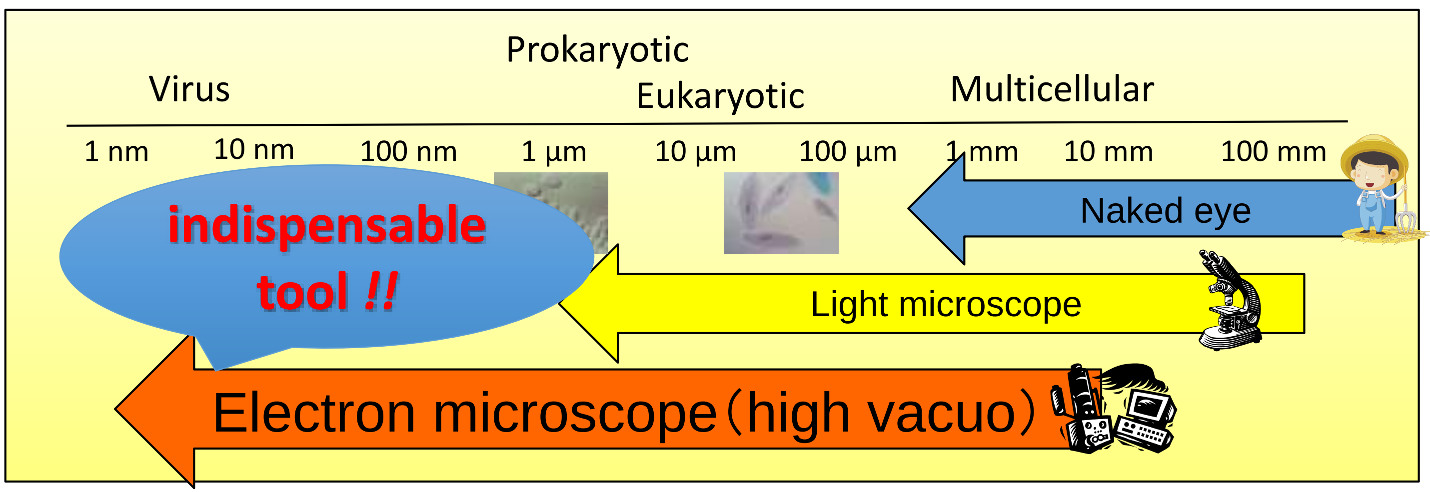
1839
T.Shwann

1930～ Electron microscope 1950～ popular equipment for science.

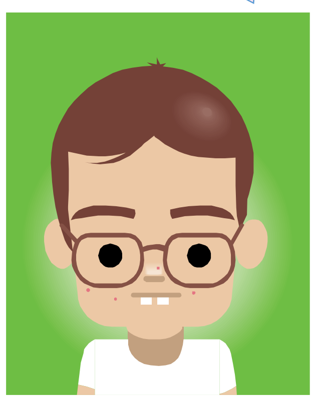
1986
Ernst Ruska

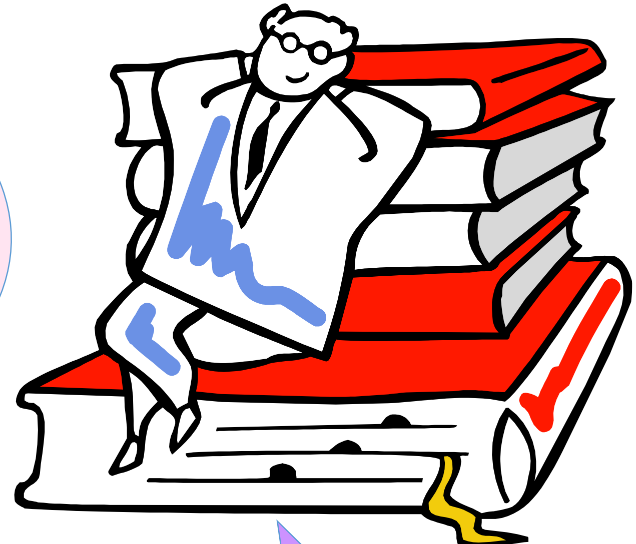
Paradigm shift

Paradigm shift life science （Virus, Organelles）

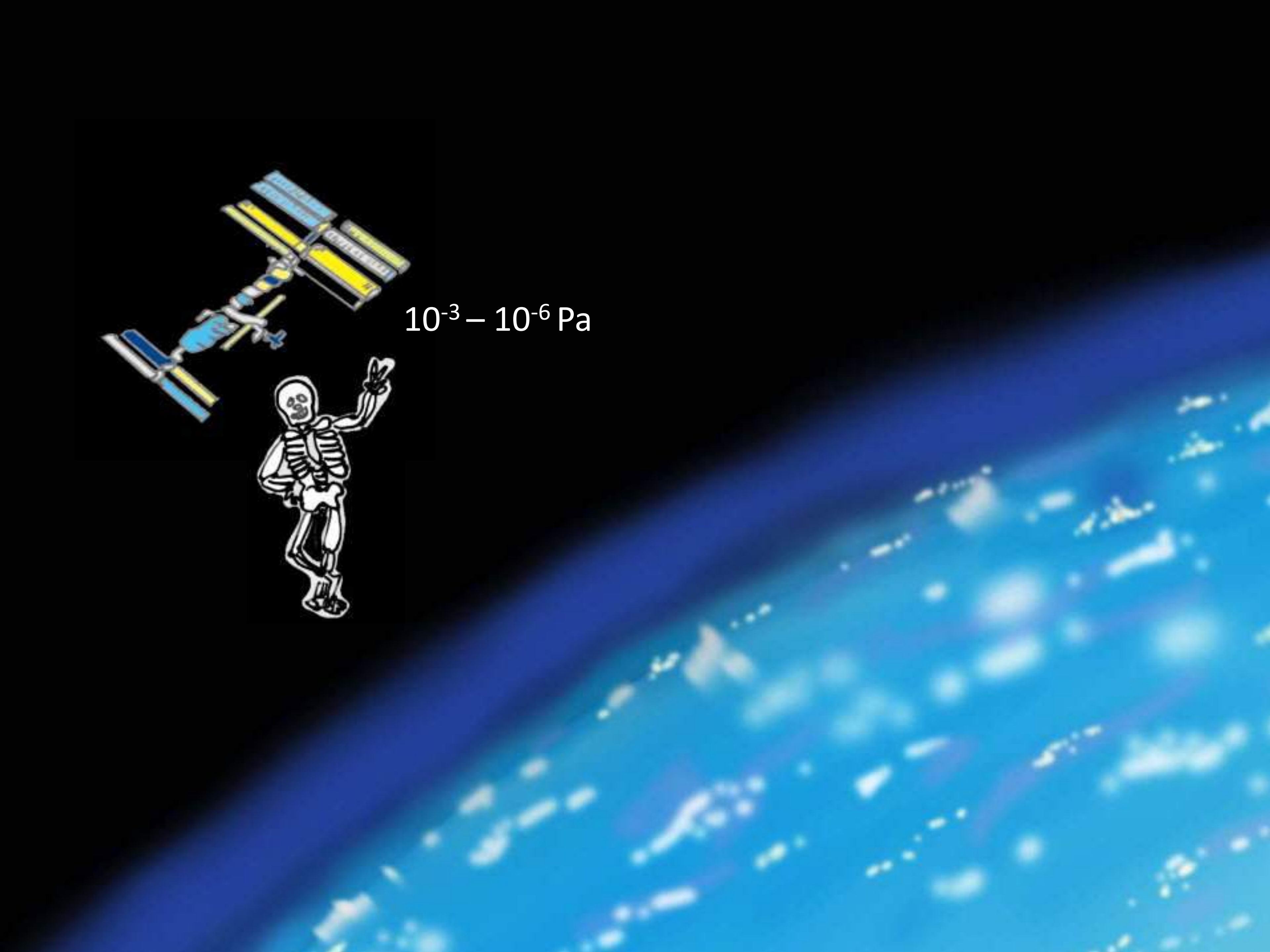


I do want to observe the real structure of organism in an electron microscope!

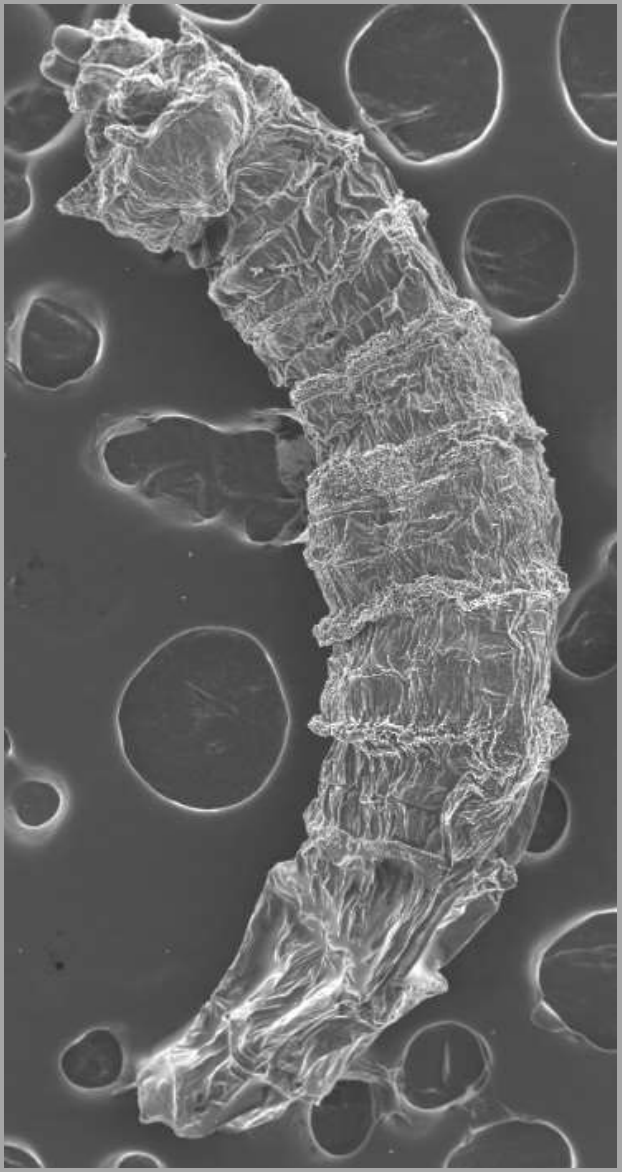


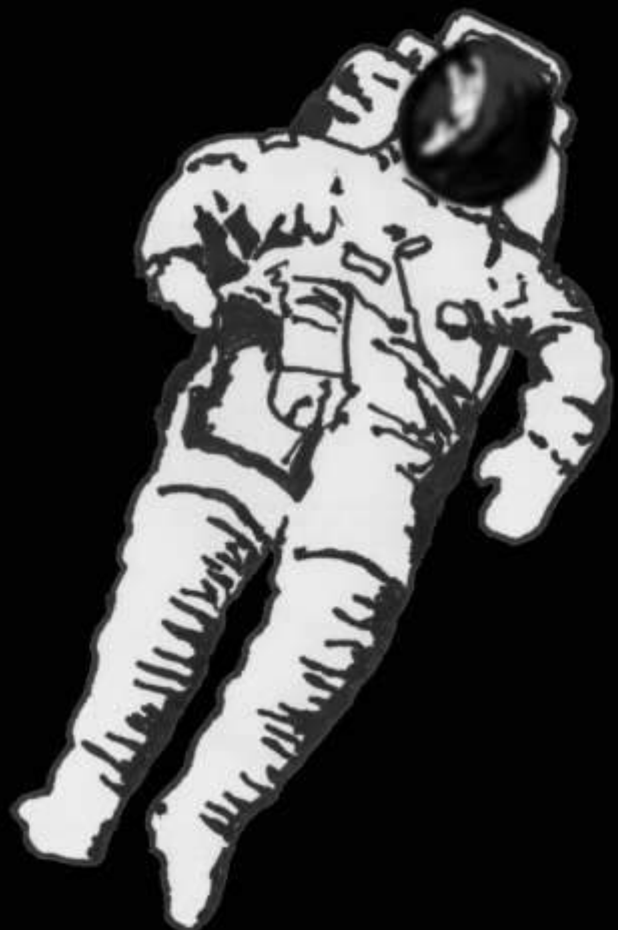


Absolutely impossible!


$10^{-3} – 10^{-6}$ Pa


Arthropoda
Drosophila larva





Pease R.F., T.L. Hayes, A.S. Camp and N.M. Amer (1966) Electron microscopy of living insects. *Science* 154, 1185–1186.

Electron micrographs of living specimens of the various developmental stages of the insect *Tribolium confusum* have been obtained with a scanning electron microscope. In most cases the specimens resumed their normal activity after being examined with the electron microscope and under went metamorphosis into the next stage.

Red flour beetle
Tribolium castaneum

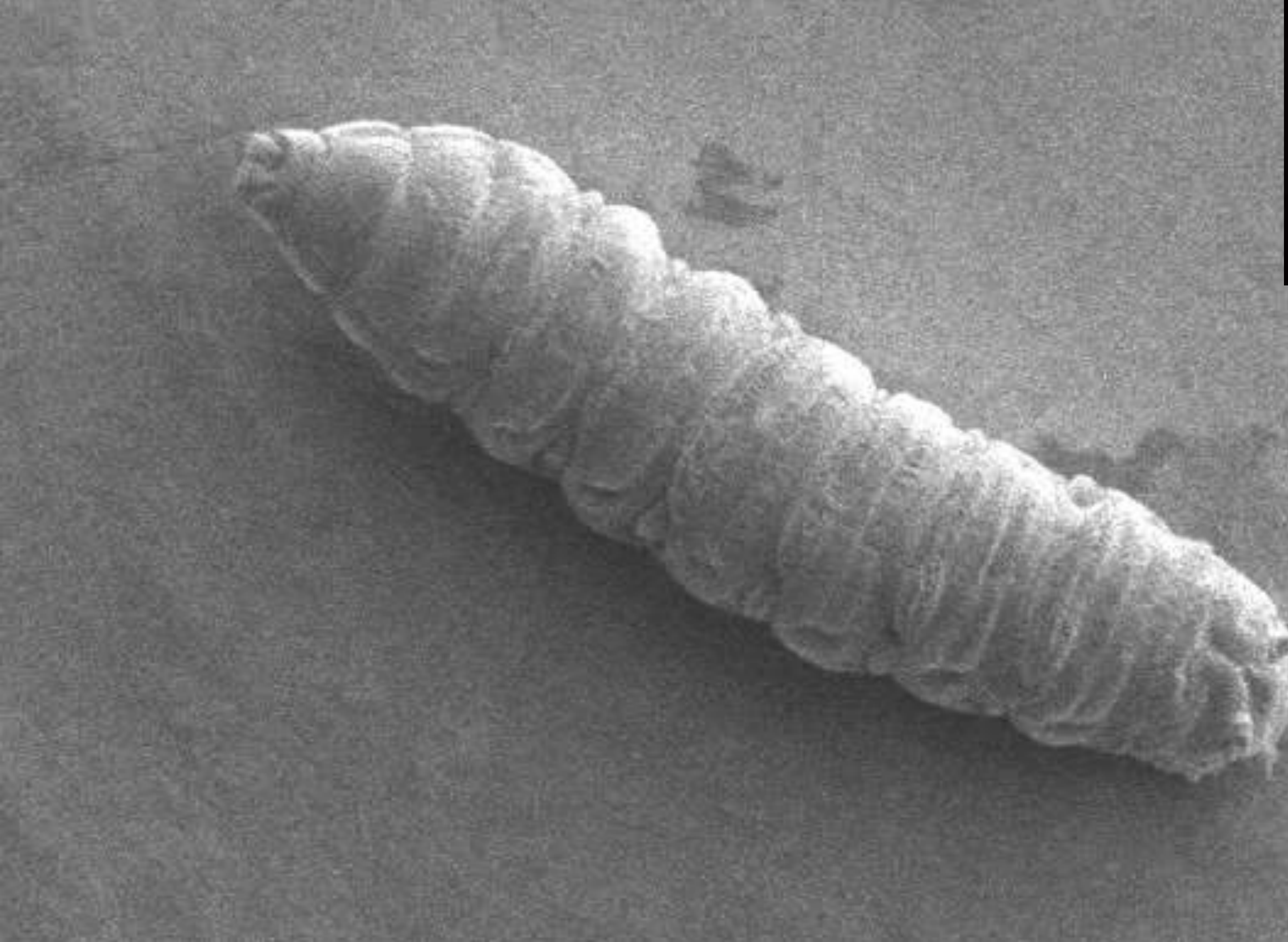
Young scientists encouraged me!

The pressure level of the SEM which used in the former report was 10^{-2} Pa.
We would like to observe several living specimens with an FE-SEM (10^{-3}-10^{-7} Pa).

Our Aim

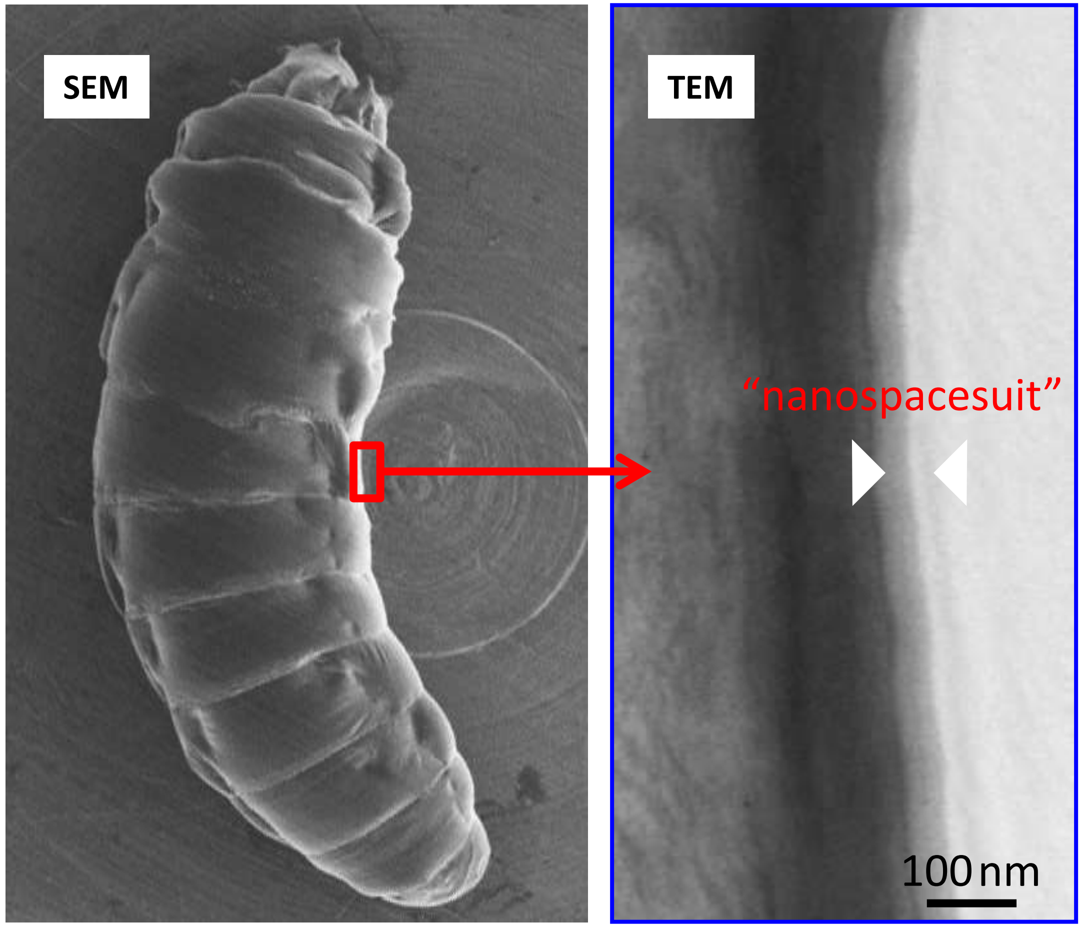
Observe several organism/tissures/cells in a FE-SEM.

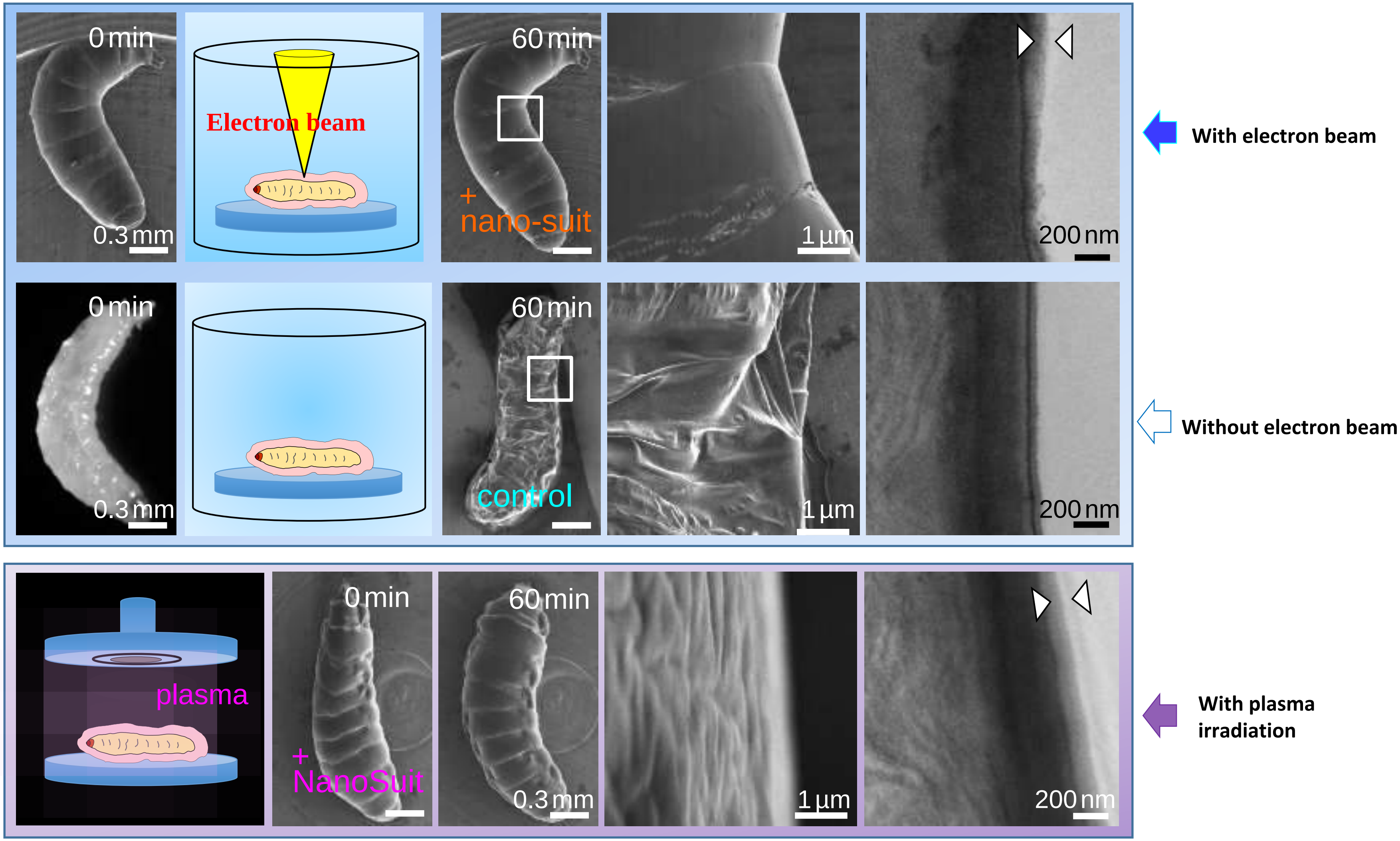
Larva of *Drosophila melanogaster* in FE-SEM



Using ECS with electron or plasma irradiation,

we could make "nanospacesuit" · · ·





0 min
0.3 mm
Electron beam
60 min
+
nano-suit
1 µm
200 nm
With electron beam
0 min
0.3 mm
60 min
control
1 µm
200 nm
Without electron beam
plasma
0 min
+
NanoSuit
60 min
0.3 mm
1 µm
200 nm
With plasma irradiation

When the animal possesses ECS and when irradiated by electrons or plasma, they can survive in high *vacuo*.

Next, we must keep living animal which do not possess ECS on their surface・・・

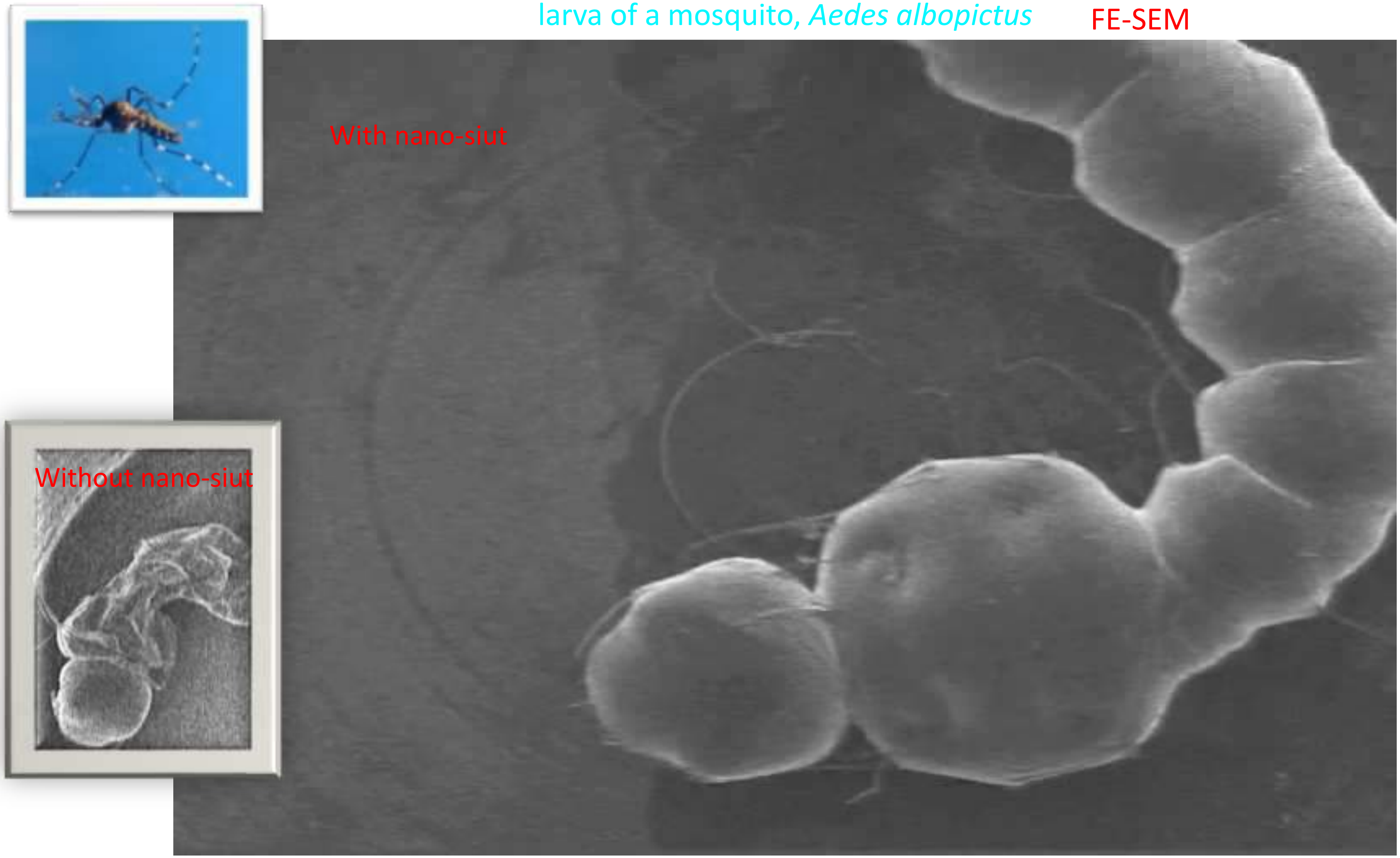
Biomimetics

ECS mimetic substance

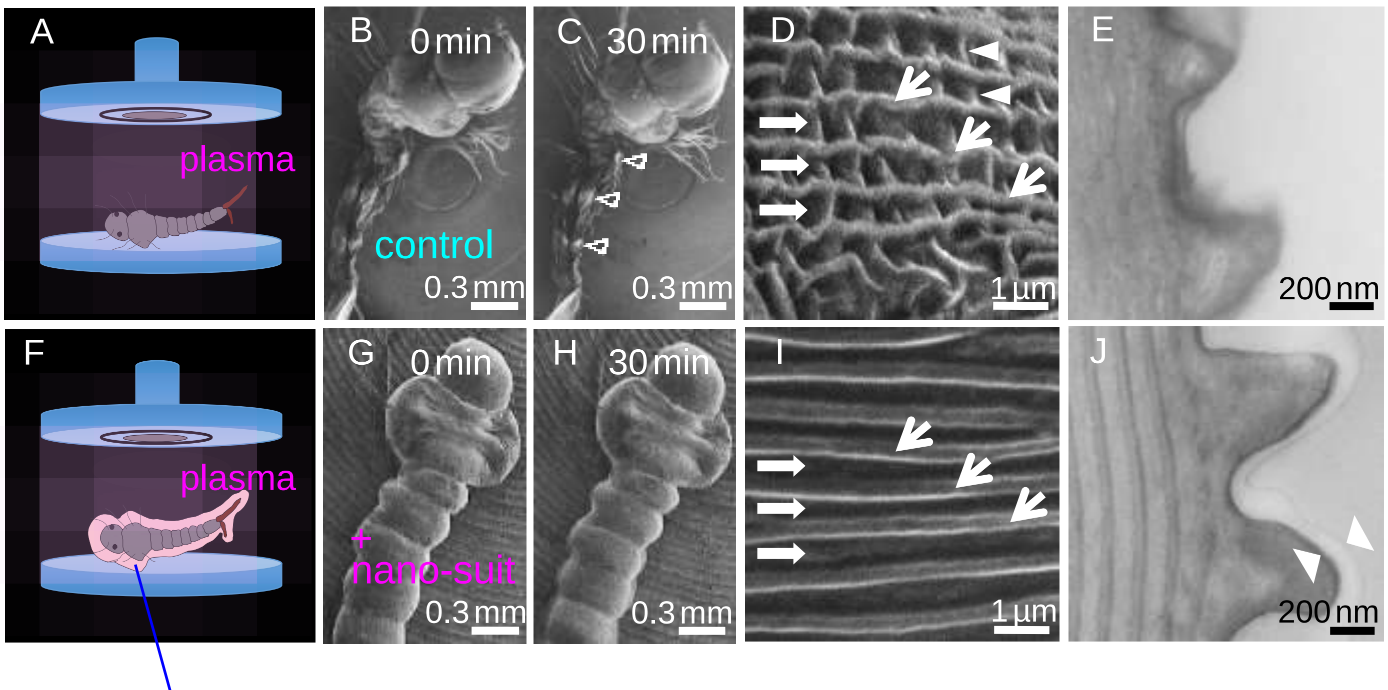
We chose amphiphilic molecules.
（1% Tween20） and irradiated.

Polysorbate 20（Tween 20）

larva of a mosquito, *Aedes albopictus* FE-SEM

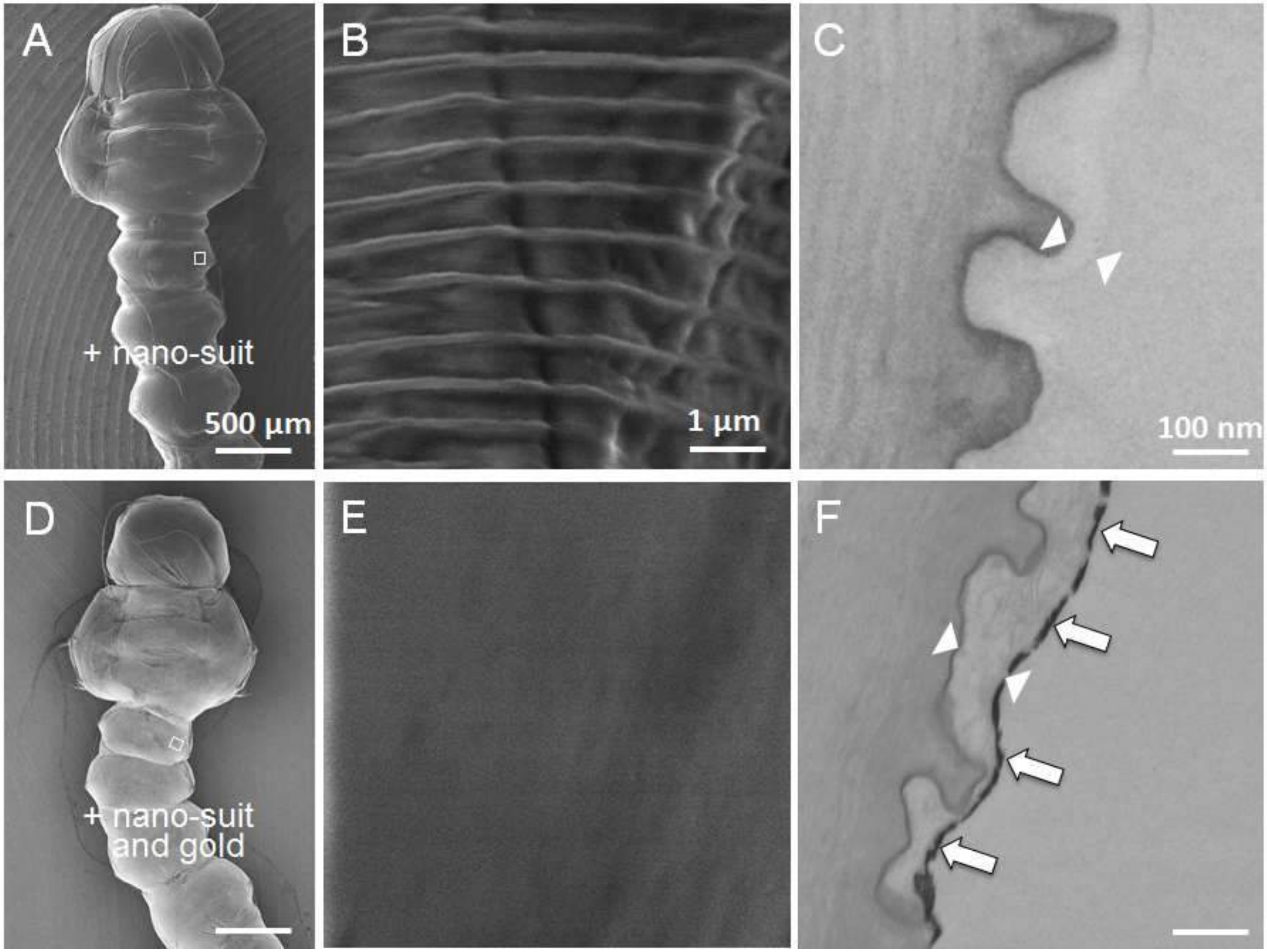


By only the plasma irradiation there was no NanoSuit, but with the additional amphiphilic molecules and the plasma irradiation, we found the NanoSuit on the surface of the cuticle, and the fine structure of the mosquito larvae were ordered..



ECS mimetic substance

nano-siut



nanospacesiut


PNAS

A thin polymer membrane, nano-suit, enhancing survival across the continuum between air and high vacuum

conventional SEM method

First Fixation — GA and/or PFA

↓

Second Fixation — O_sO_4

↓

Dehydration

↓

Desiccation

↓

Covered by metal — Gold, O_sO_4

↓

observation

Need at least a whole day.

NanoSuit method

Living organisms

↓

Covered with ECS mimic solution

↓

Irradiated by
electron beam or plasma

↓

observation

Need three minutes. カップラーメン　Cup RAMEN noodle

nano-siut

nature

Nano-suit shields bugs in the void

Submit today!

Nuclear Reprogramming and the Cancer Genome

Register today!

Science

Nano-Suit Protects Bugs From Space-Like Vacuums

POPULAR

FROM THE MAGAZINE

WEBINAR

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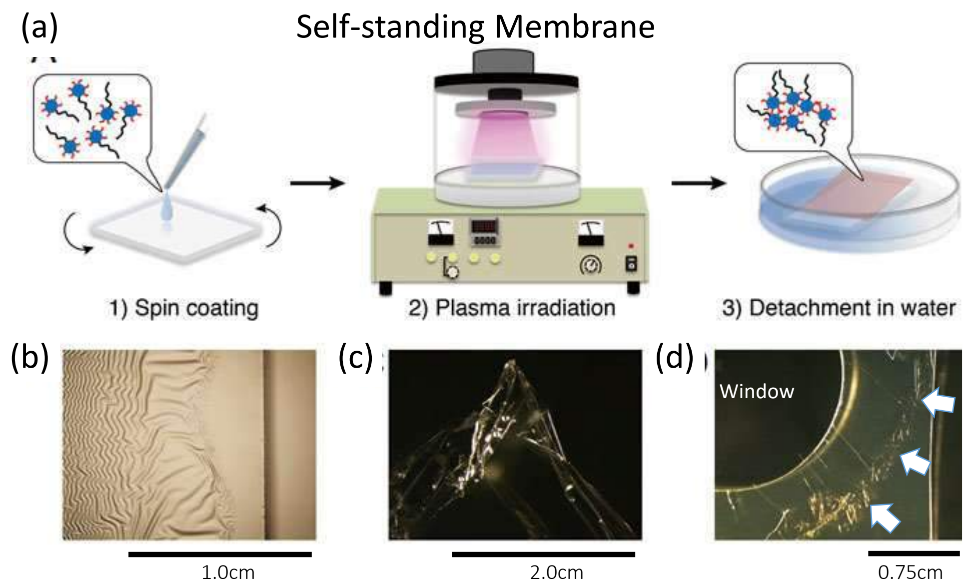
After the report of PNAS ・・・

We could make self-standing ultra-thin films from water soluble amphiphilic molecules.

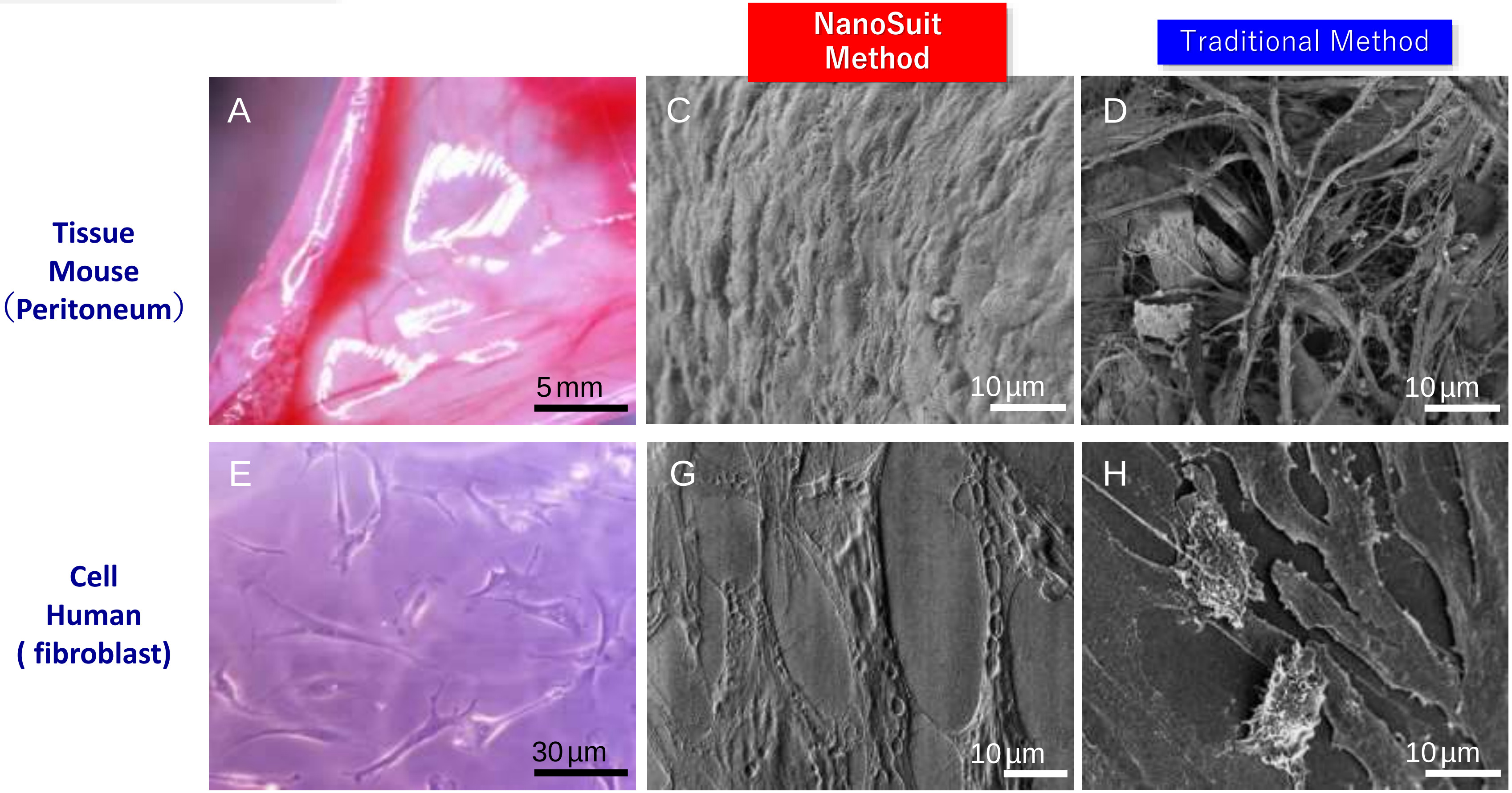


Sample	Monomer	Polymerization site	Solvent	State
1	Tween 20	PEO chain	Water	◎
2	Tween 40	PEO chain	Water	◎
3	Tween 60	PEO chain	Water	◎
4	Tween 80	PEO chain	Water	◎
5	Brij 35	PEO chain	Water	◎
6	Triton X-100	PEO chain	Water	◎
7	Poly(ethylene oxide)	PEO chain	Water	◎
8	Pluronic F-127	PEO chain	Water	◎
9	Pluronic F-68	PEO chain	Water	◎
10	Lecithin (from soy bean)	Multiple OH	Ethanol	◎
11	Tannic acid	Multiple OH	Ethanol	◎
12	Tetraethoxysilane	Multiple OH	Ethanol	◎
13	Span 20	Multiple OH	Ethanol	◎
14	D-Maltose	Multiple OH	Water	◎
15	Trehalose C12	Multiple OH	Water	◎
16	D-Glucose	Multiple OH	Water	◎
17	n-Dodecyl-β-D-maltoside	Multiple OH	Water	◎
18	MEGA-8	Multiple OH	Water	○
19	CHAPS	Multiple OH	Water	○
20	D-Trehalose	Multiple OH	Water	○
21	Sodium cholate	Multiple OH	Water	○
22	n-Octyl-β-D-glucoside	Multiple OH	Water	○
23	Inulin	Multiple OH	Water	△
24	Pullulan	Multiple OH	Water	△
25	D-Sorbitol	Multiple OH	Water	△
26	L-Tyrosine	Multiple OH	Water	△
27	L-Glutamic acid	Multiple OH	Water	△
28	L-Aspartic acid	Multiple OH	Water	△
29	Lauric acid	Single OH	Ethanol	○
30	Stearic acid n-dodecyl ester	Single OH	Ethanol	○
31	Docosanoic acid	Single OH	Ethanol	○
32	L-Proline	Single OH	Water	△
33	L-Lysine	Single OH	Water	△
34	L-Histidine	Single OH	Water	△
35	Linolenic acid	OH & C=C double bond	Ethanol	◎
36	Linoleic acid	OH & C=C double bond	Ethanol	◎
37	Oleic acid	OH & C=C double bond	Ethanol	◎
38	Erucic acid	OH & C=C double bond	Ethanol	◎
39	Methacroylcholine chloride	OH & C=C double bond	Ethanol	◎
40	L-Glutamine	OH & C=C double bond	Water	○
41	L-Arginine	OH & C=N double bond	Water	○
42	1,3-Diallylimidazolium bromide	C=C double bond	Ethanol	◎
43	1,3-Diallylimidazolium tetrafluoroborate	C=C double bond	Ethanol	◎
44	1,3-Diallylimidazolium bis(trifluoromethanesulfonyl)imide	C=C double bond	Ethanol	◎
45	1-Butyl-3-methylimidazolium tetrafluoroborate	C=C double bond	Ethanol	○
46	1-Butyl-3-methylimidazolium bis(trifluoromethanesulfonyl)imide	C=C double bond	Ethanol	○

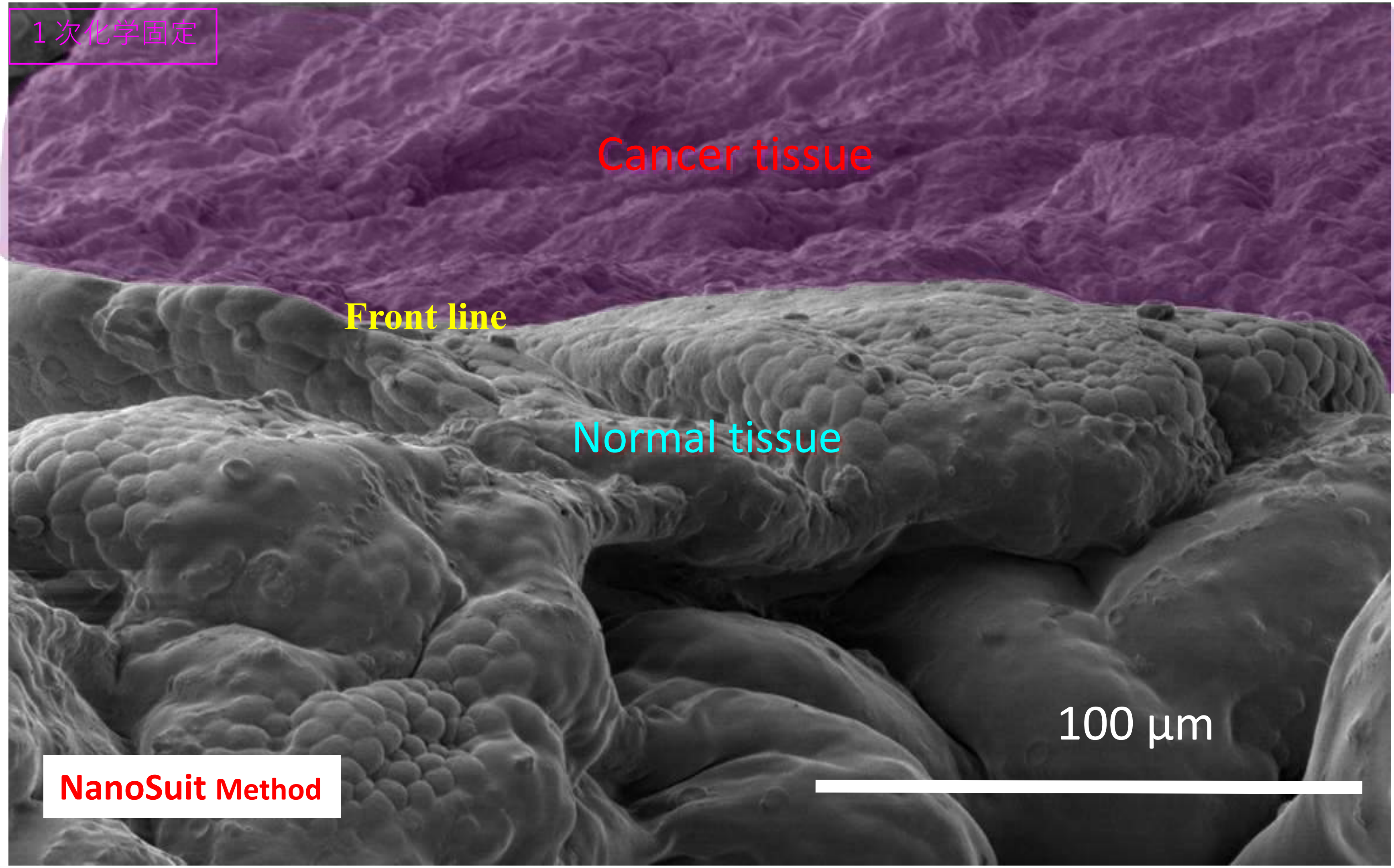
◎: Large, thick, stable film. ○: Small, thin, stable film. △: Unstable film. SEM observation of living organisms

Wako Pure Chemical Co., Tokyo Kasei Kogyo Co., Sigma-Aldrich Japan Co., Kanto Chemical Co., Dojindo Co.

Tissue and Cells

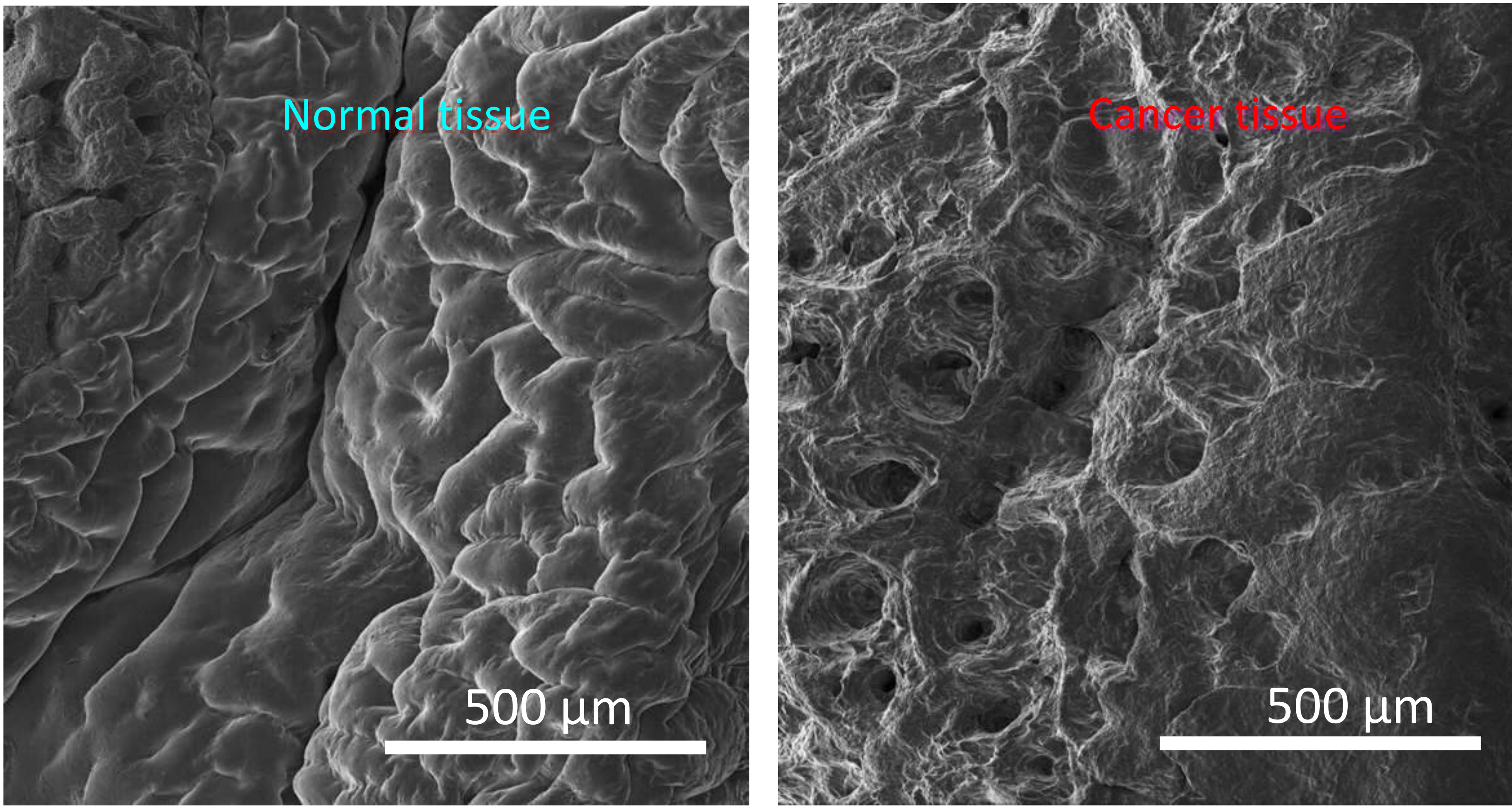


Human Stomach Cancer （Front line between cancer and normal tissue）



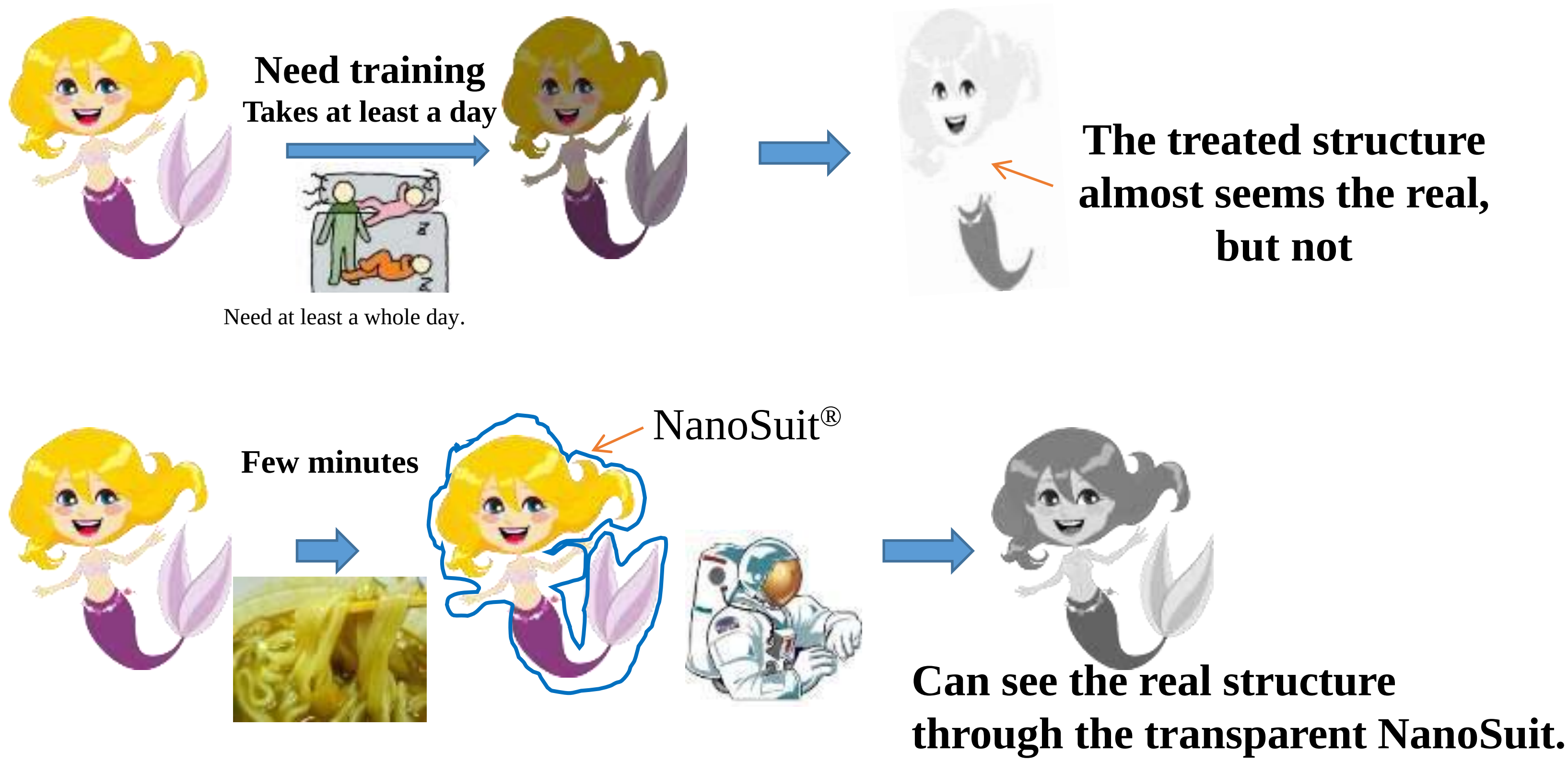
Human Stomach Cancer （normal and cancer tissue）

SSE / nanosuit method



Human Stomach Cancer

The “NanoSuit®” enabled the direct observation of hydrous biological samples.




Don't you want to see
what you cannot see?
We can now observe
subcellular size
organic substances
as it is.
For Biomimetics!

NanoSuit®

Members

Yasuharu Takaku

Hiroshi Suzuki

Isao Ohta

Hiroyuki Konno

Hirotoshi Kikuchi

Hiroshi Watanabe

Takanori Hiraide

Daisuke Ishii

Masatsugu Shimomura

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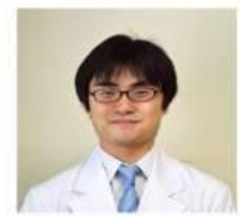
Hideya Kawasaki

Tomohiro Matsumoto



Yusuke Ozaki

Yuji Hirai

Yoshinori Muranaka

Takahiko Hariyama

Satoshi Hirakawa

Takeshi Nokuo

Vielen Dank!