



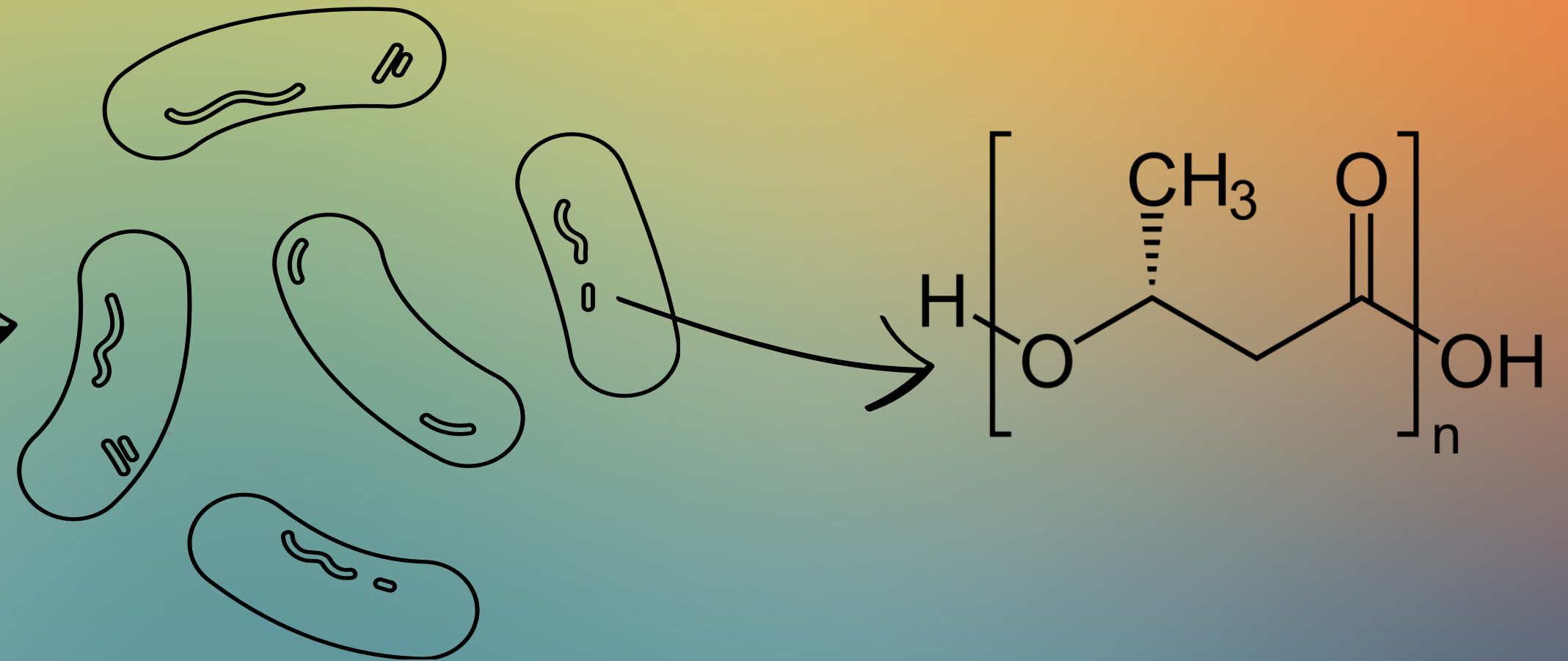
EXPLORING BACTERIAL TOPOGRAPHY WITH ATOMIC FORCE MICROSCOPY

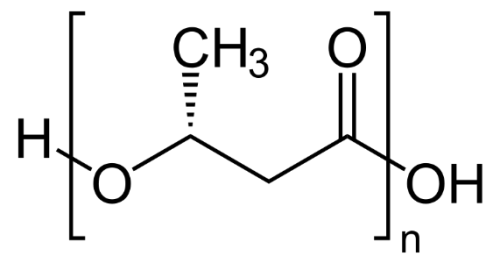
Markéta Khýrová
6.9.2023

INTRODUCTION



Atomic force microscope JPK Nanowizard 4





MODEL MICROORGANISMS

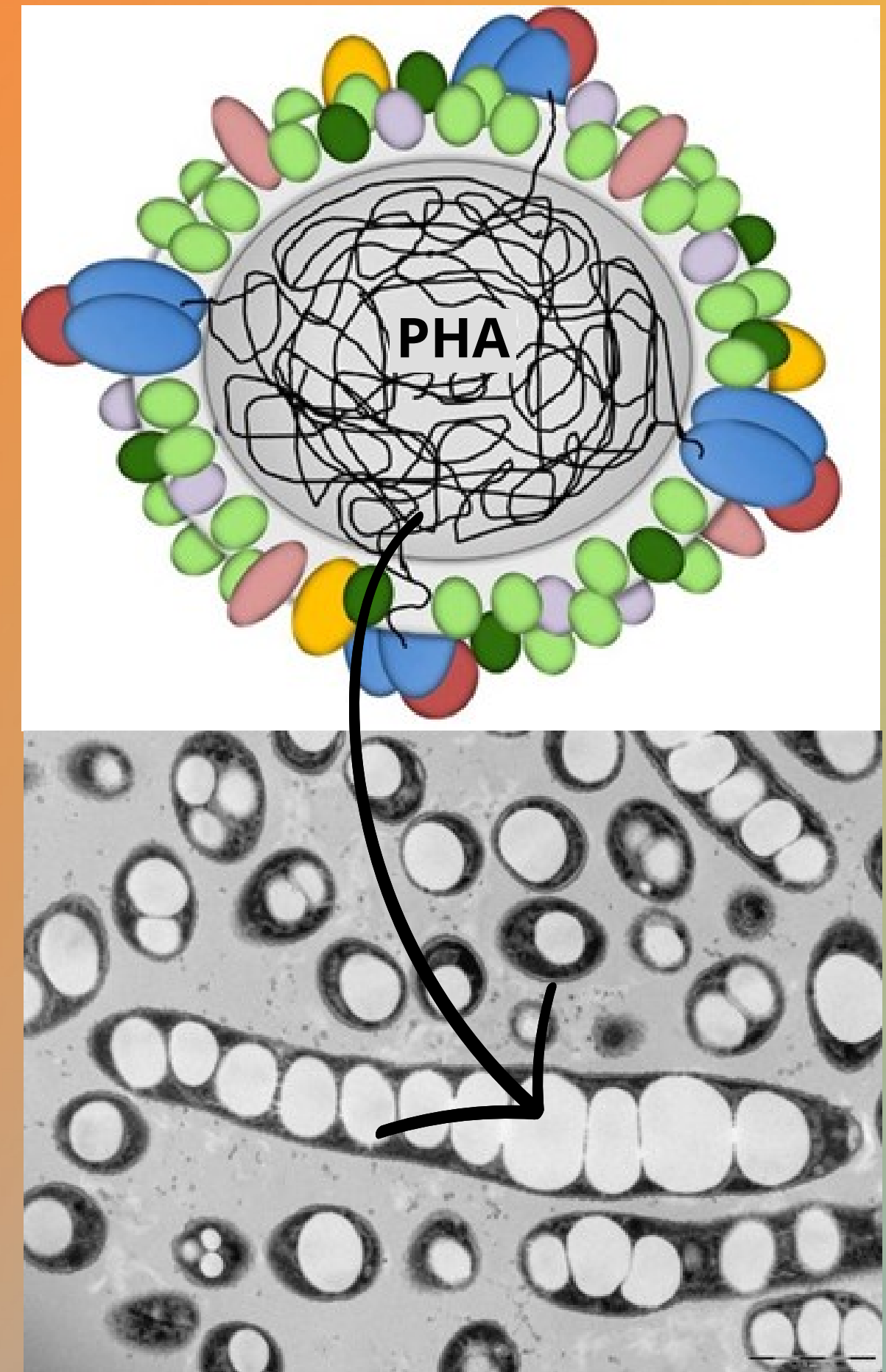
PRODUCERS OF POLYHYDROXYALKANOATES (PHAs)

Cupriavidus necator H16 (CCM 3726)

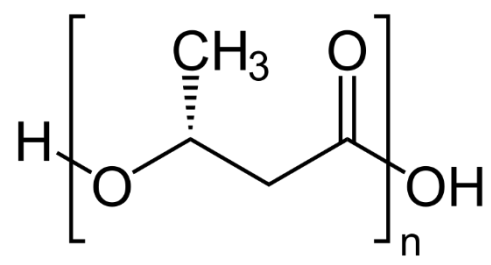
Cupriavidus necator PHB⁻⁴ (DSM 514)

Halomonas halophila (CCM 3662)

Rhodospirillum rubrum (DMS 467)



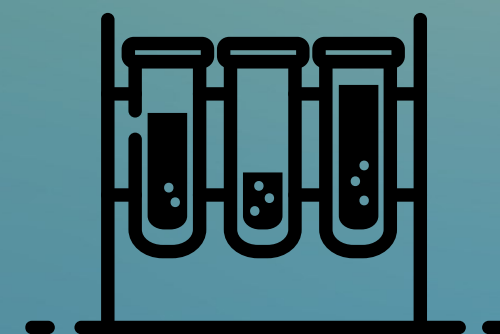
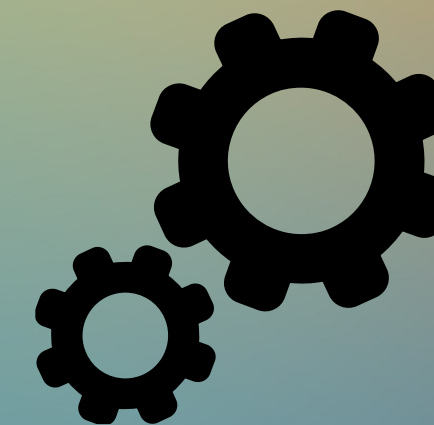
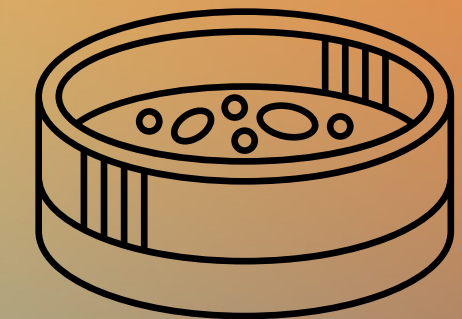
*Scheme of PHA pellet and TEM image of
C. necator H16*

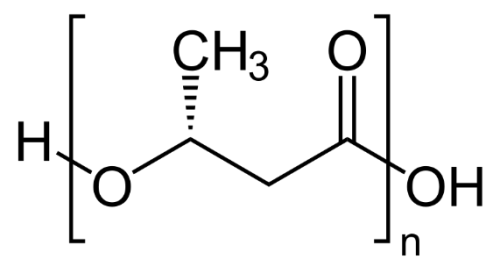


GOALS

MEASUREMENT OPTIMIZATION FOR AIR AND LIQUID

- IMMOBILIZATION TECHNIQUE
- MEASUREMENT MODE SETTING
- CANTILEVER
- SAMPLE DILUTION



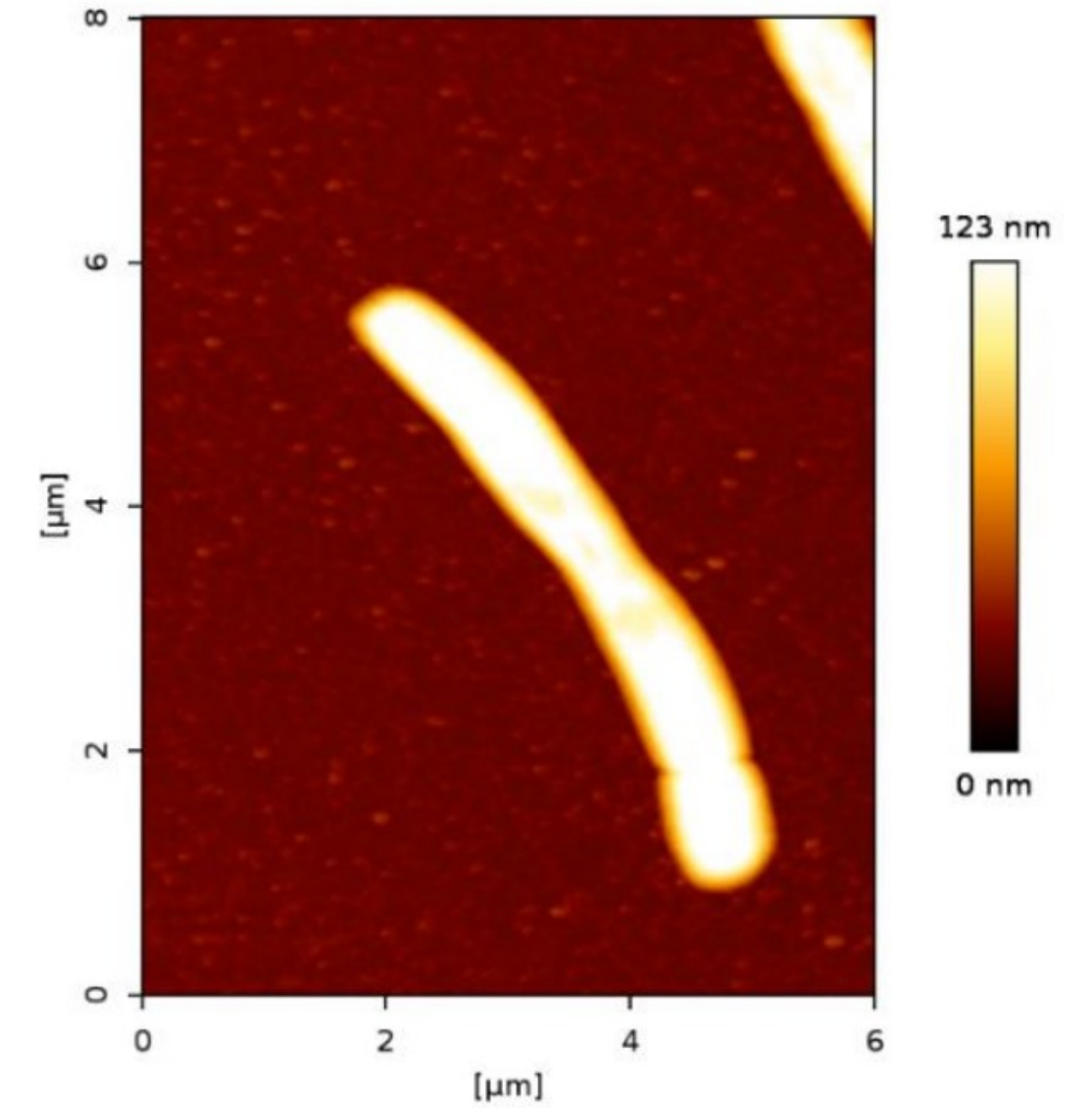
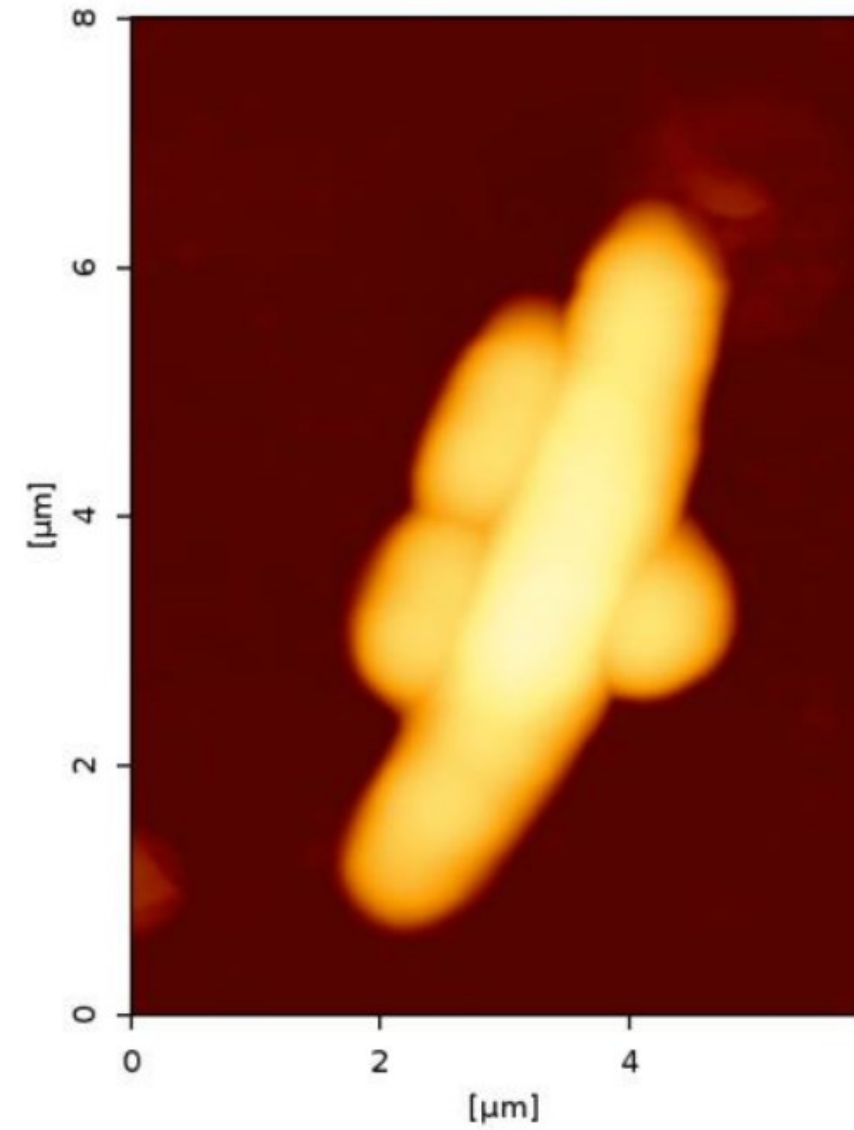


DRYED SAMPLES

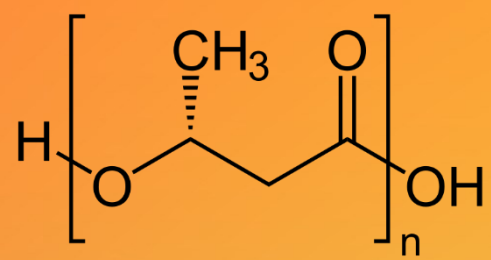
TESPA-V2

QI mode

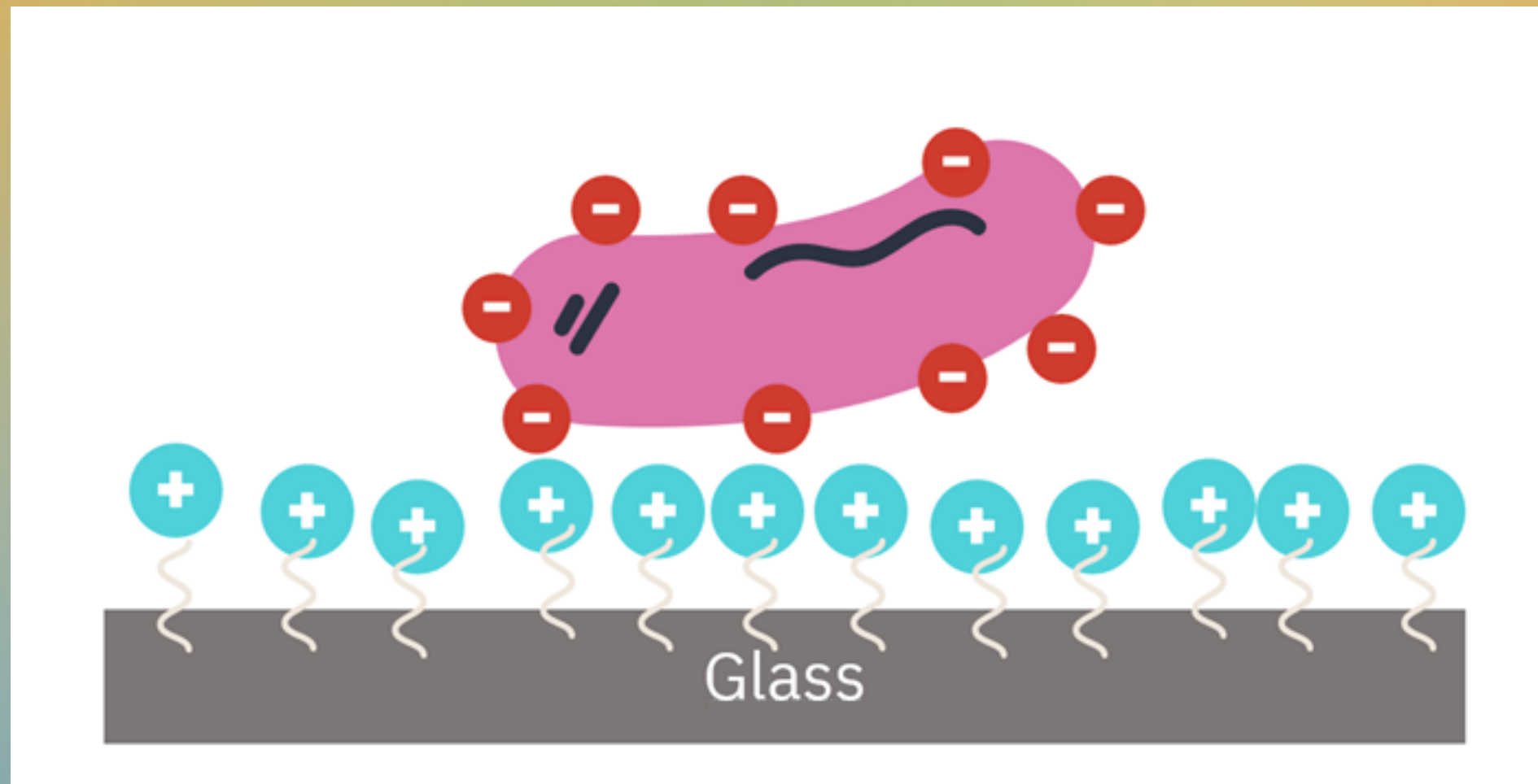
10x DILUTION



Dried Cupriavidus necator H16 (left) and *C. necator* PHB-4 (right)

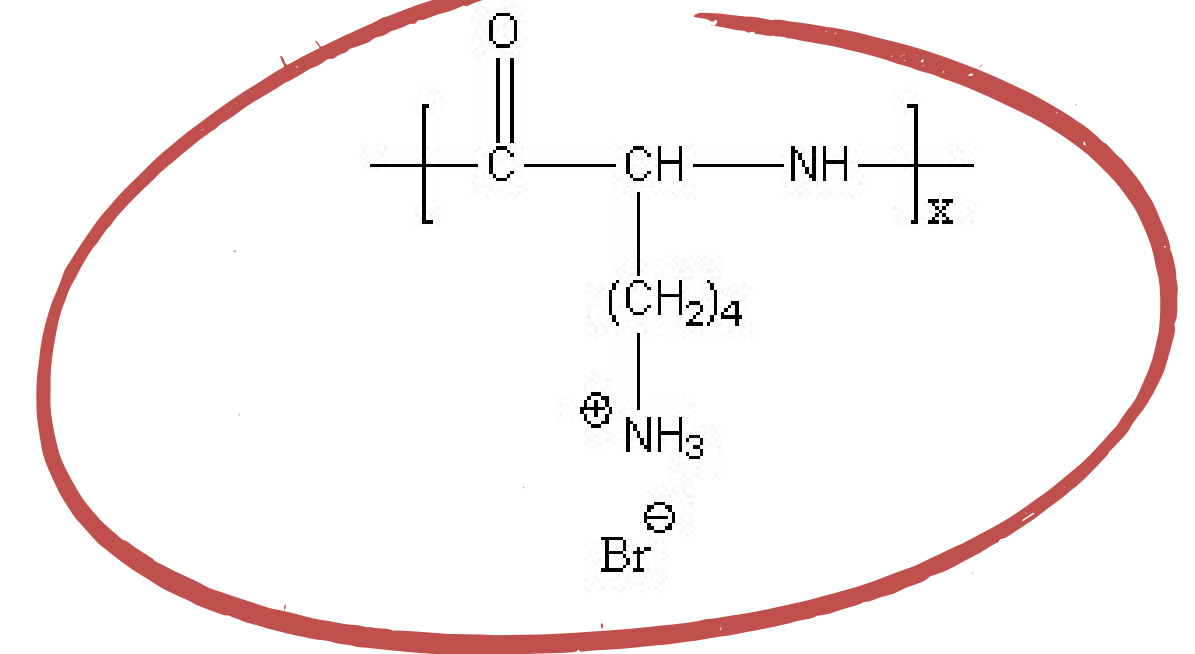


LIQUID MEASUREMENTS

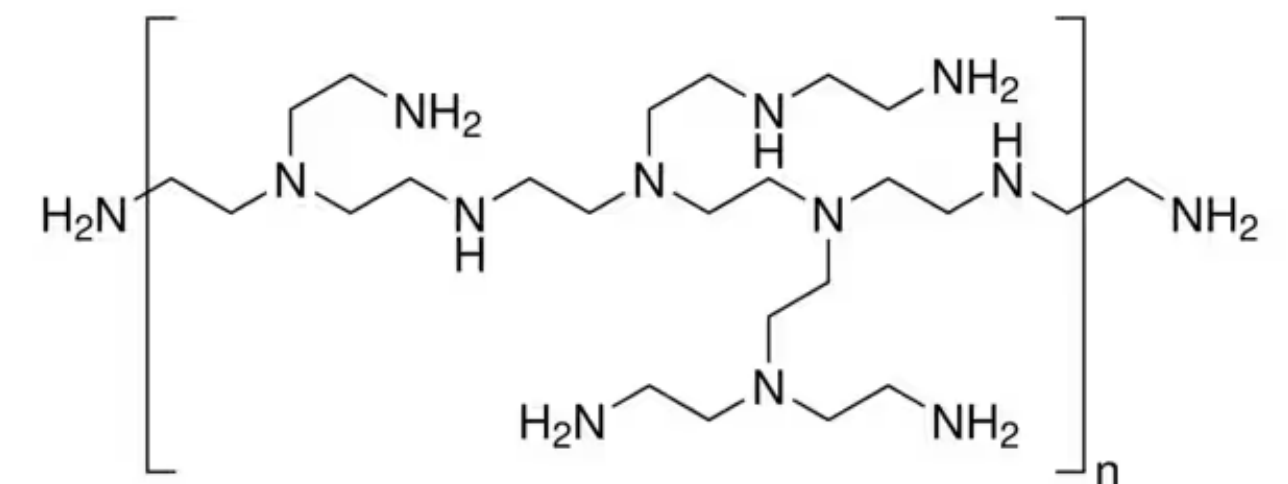


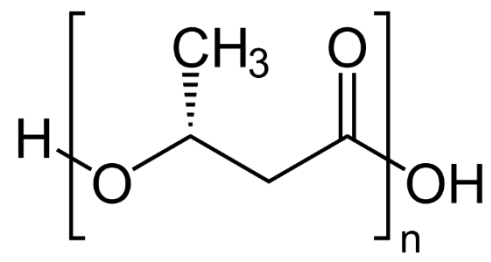
Adsorption of negatively charged bacteria on positively charged glass surface

POLY-L-LYSINE HYDROBROMIDE -



POLY(ETHYLENIMINE) - PEI





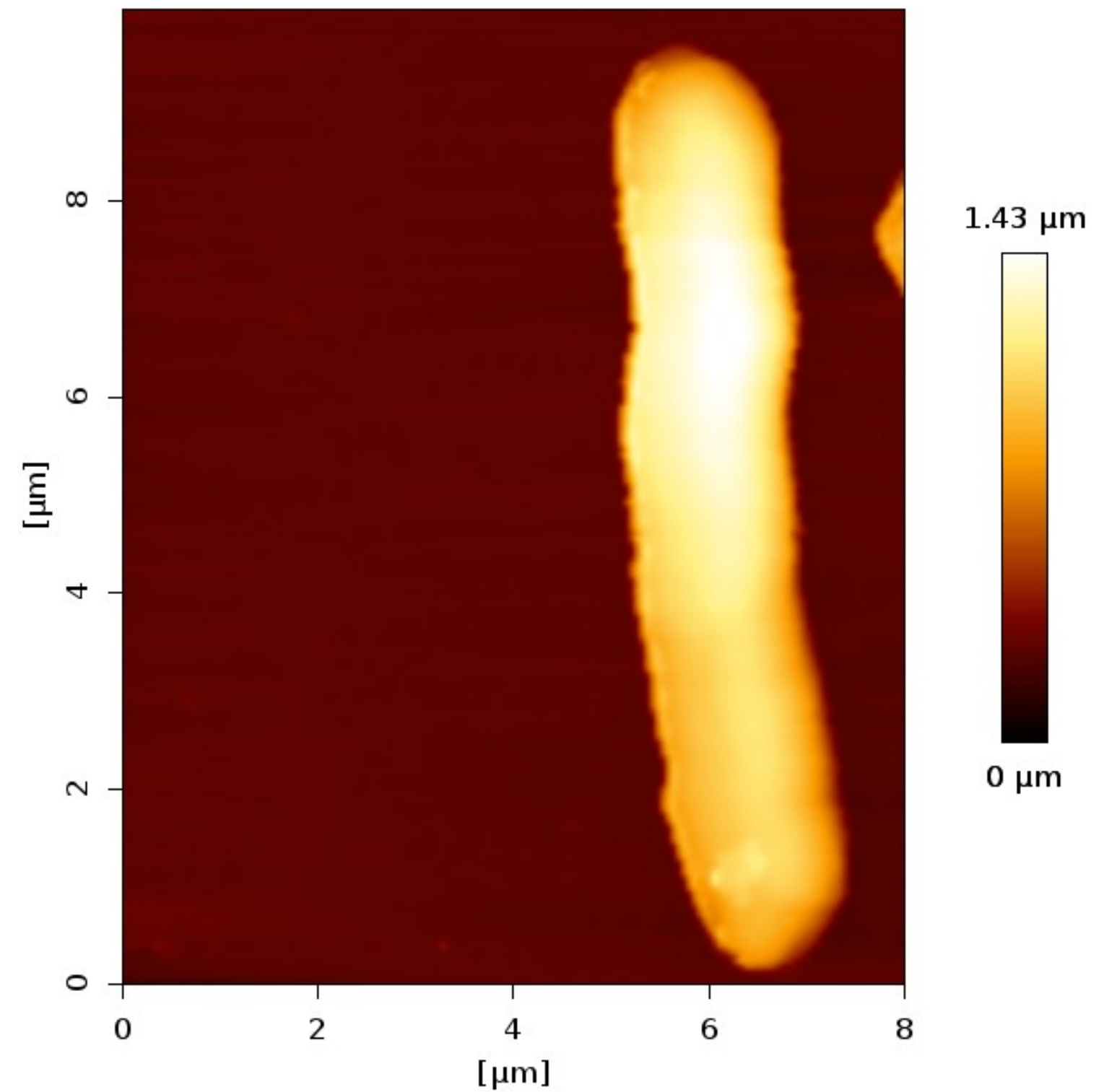
LIVE SAMPLES

MLCT – A, SNL – B
(spring constant around 0,1 N/m)

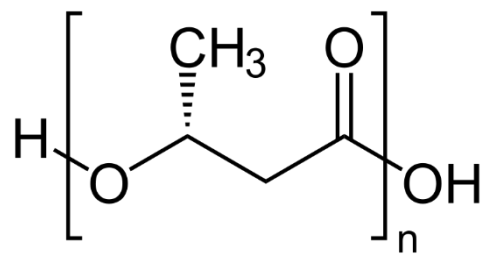
QI mode

DILUTION ACCORDING OPTICAL
DENSITY

POLY-L-LYSINE



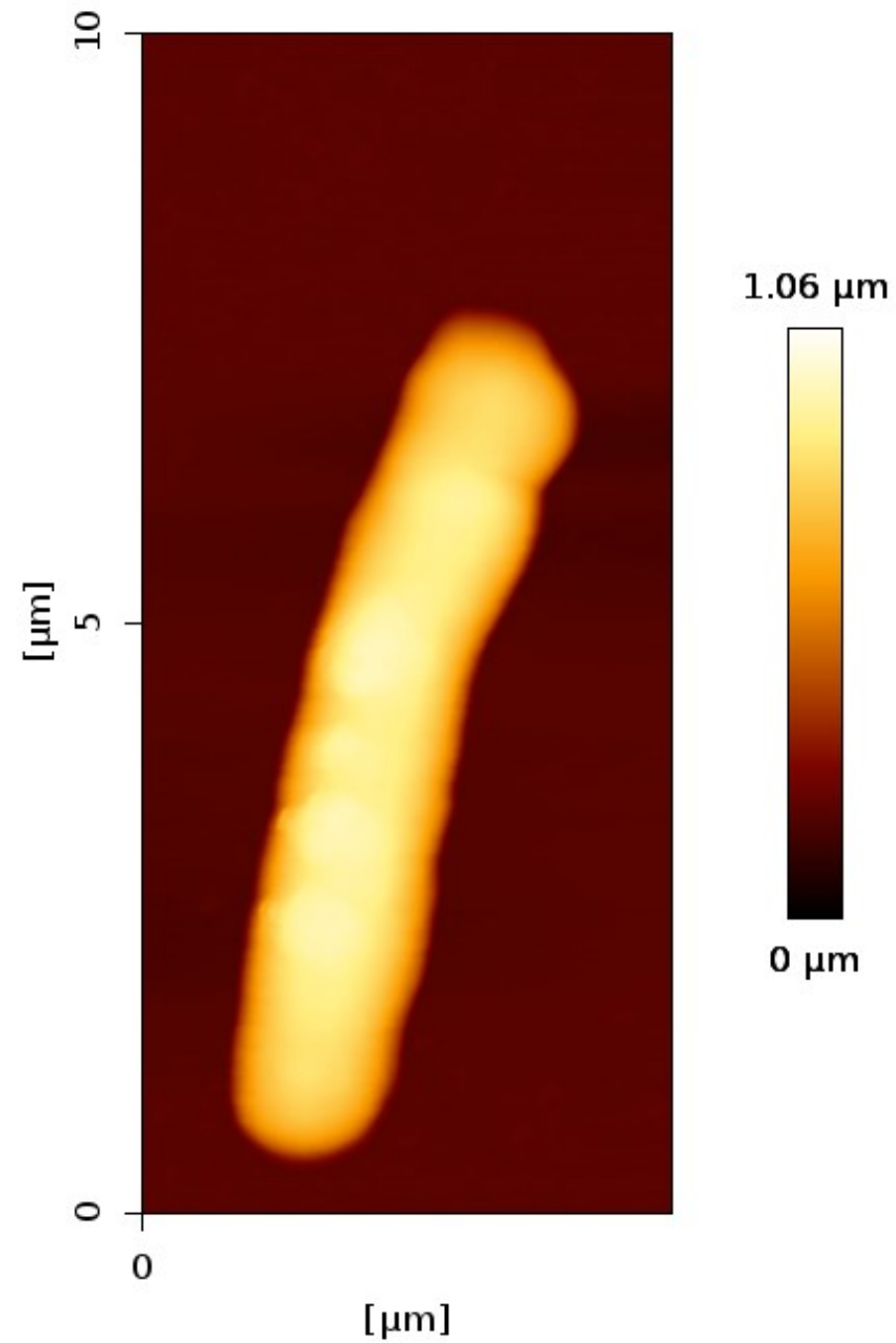
C. necator H16 measured in liquid



Cupriavidus necator H16

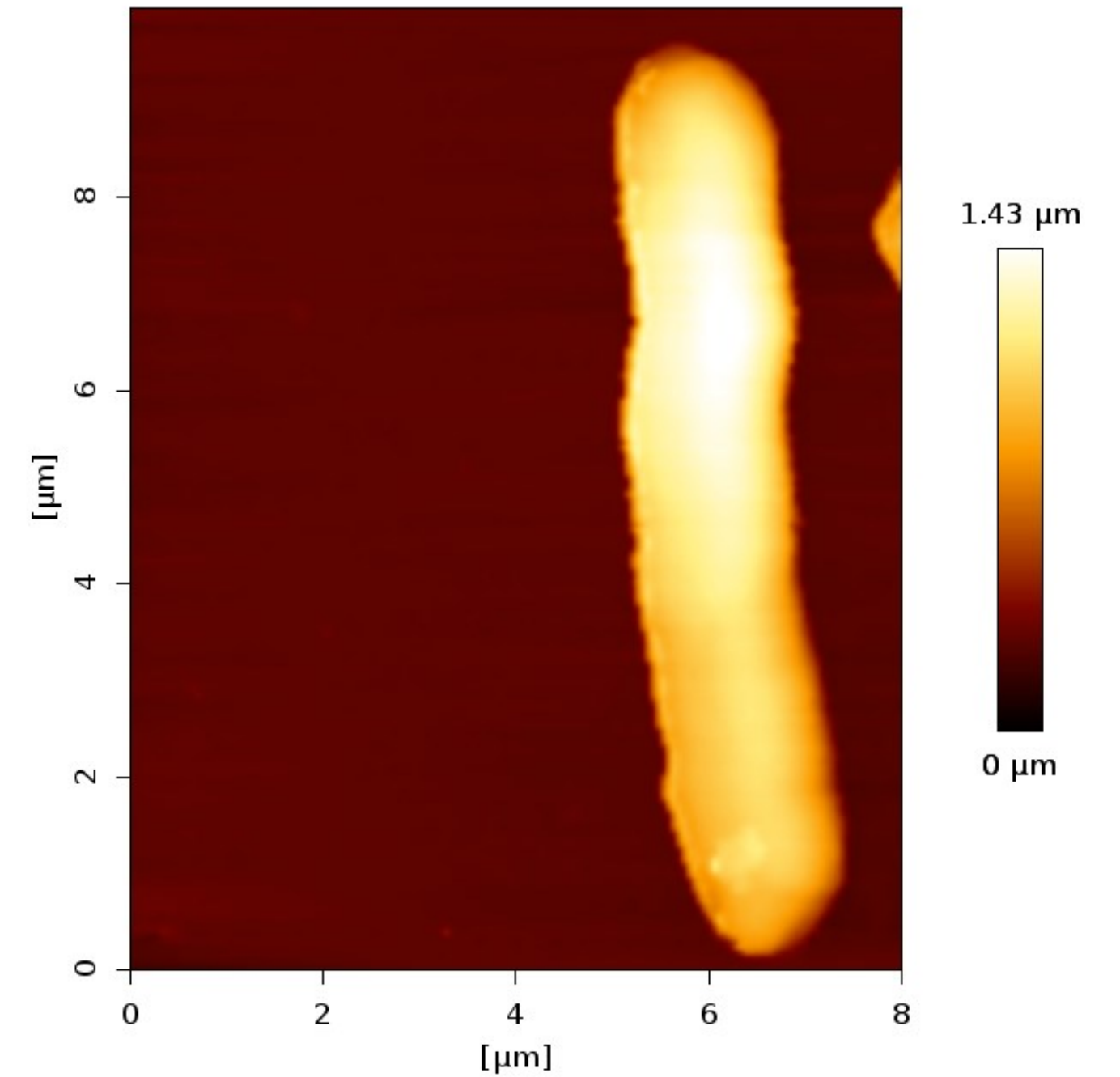
AIR

(94,1 % PHA)

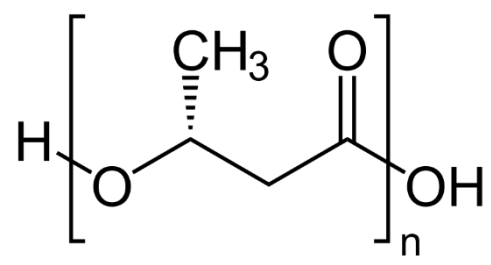


PBS

(83,4 % PHA)



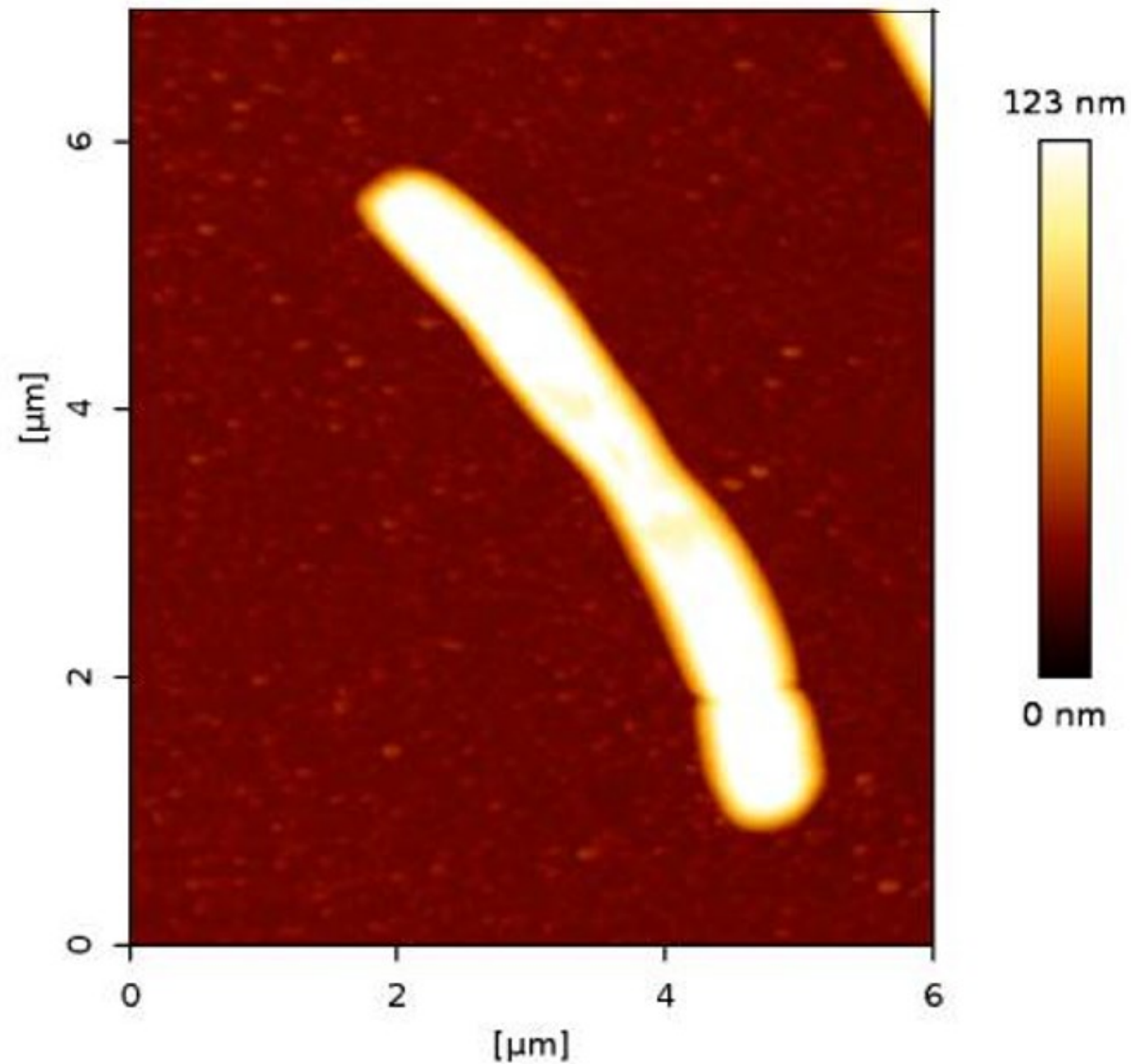
Comparison of C. necator H16 measured on air (left) and in liquid (right)



Cupriavidus necator PHB-4

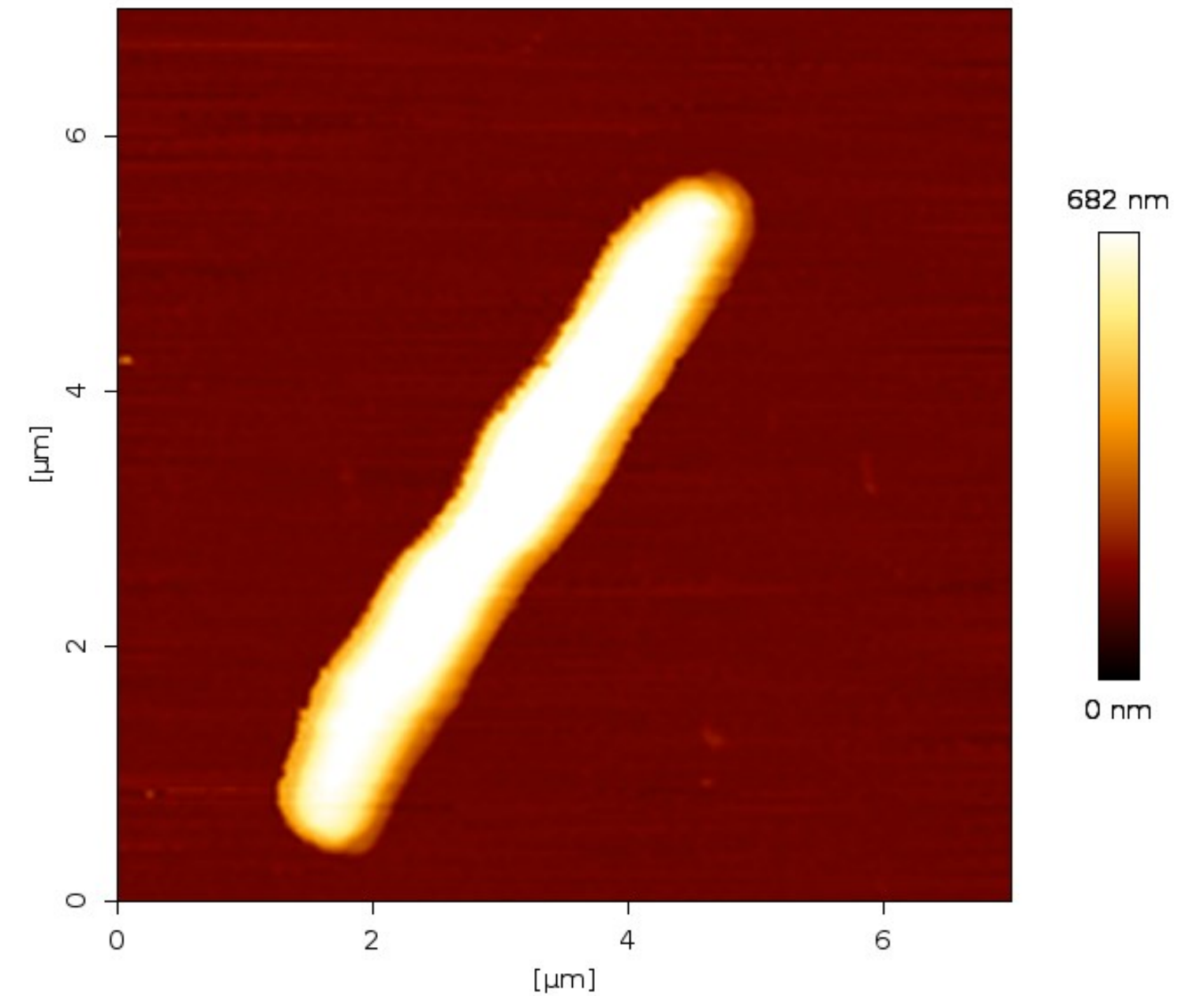
AIR

(0,7 % PHA)

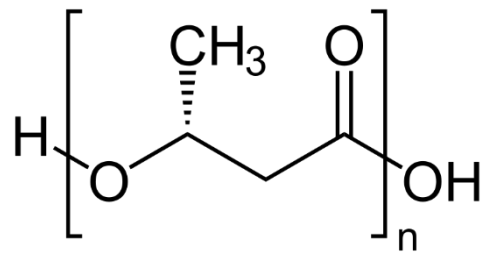


PBS

(0,7 % PHA)



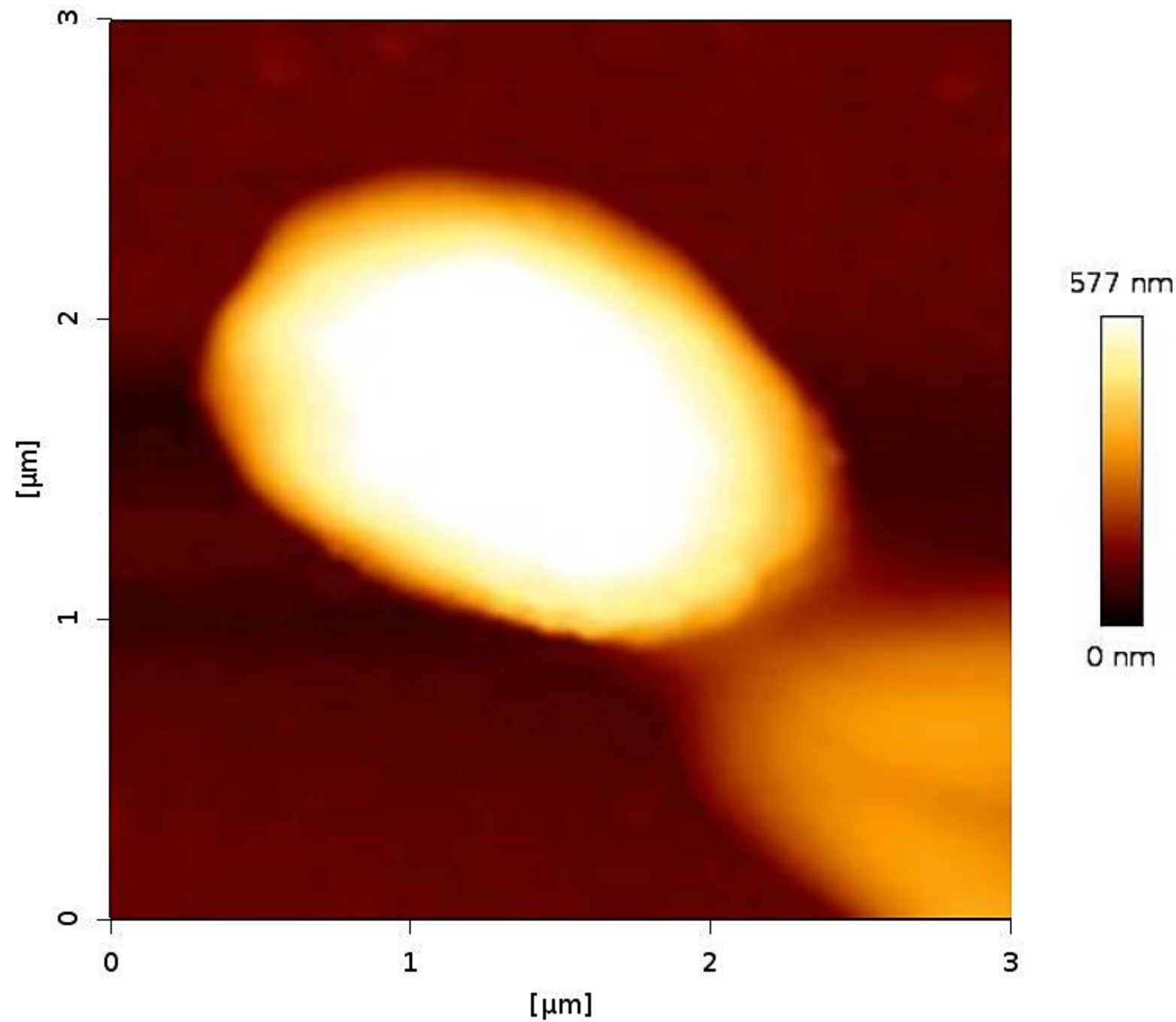
Comparison of C. necator PHB-4 measured on air (left) and in liquid (right)



Halomonas halophila

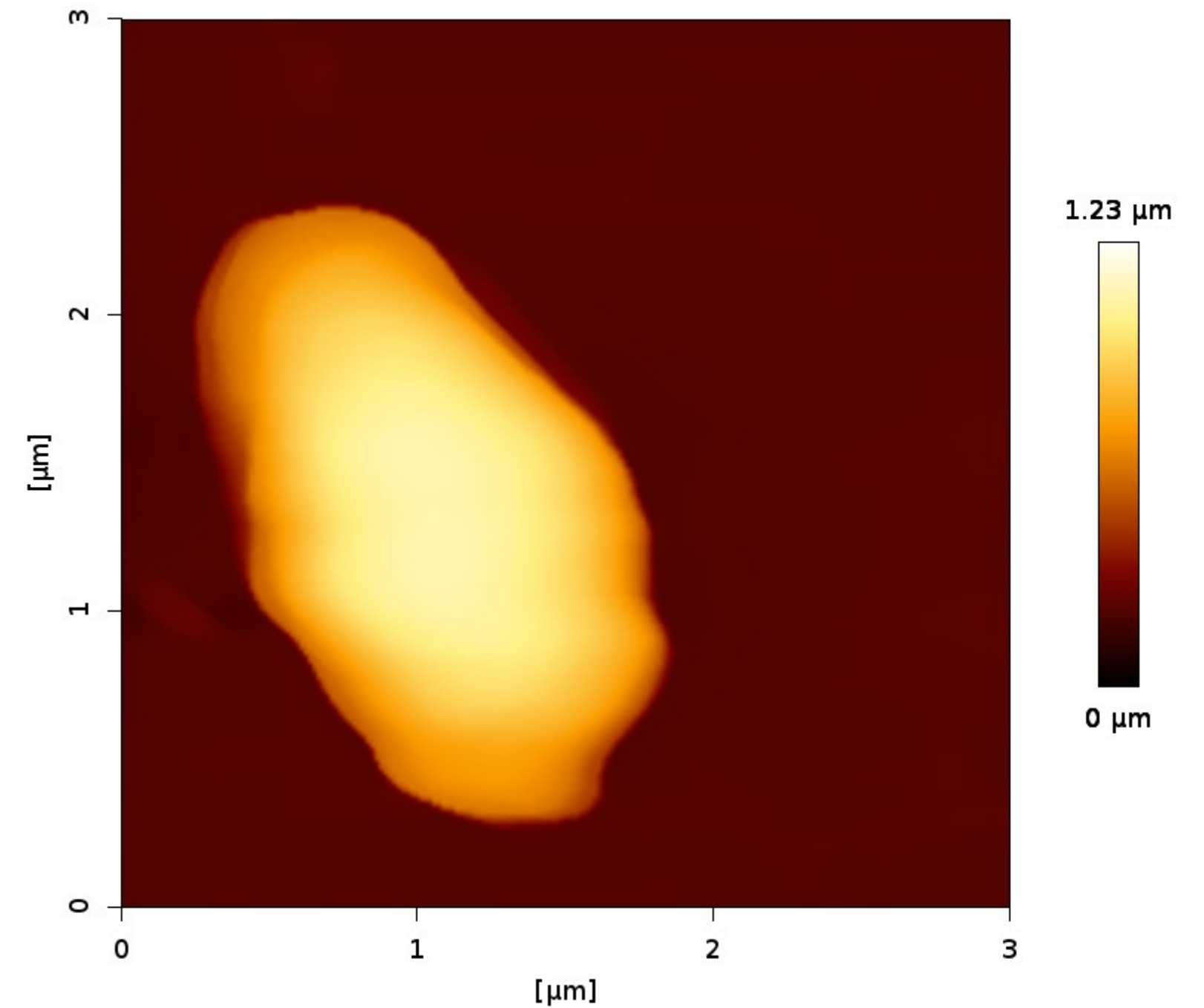
AIR

(43,6 % PHA)

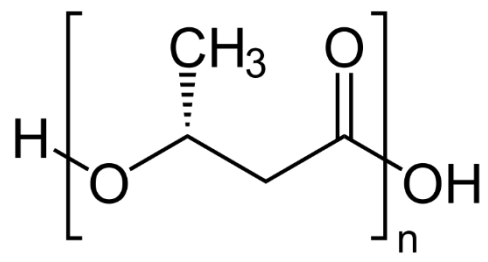


PBS

(43,6 % PHA)



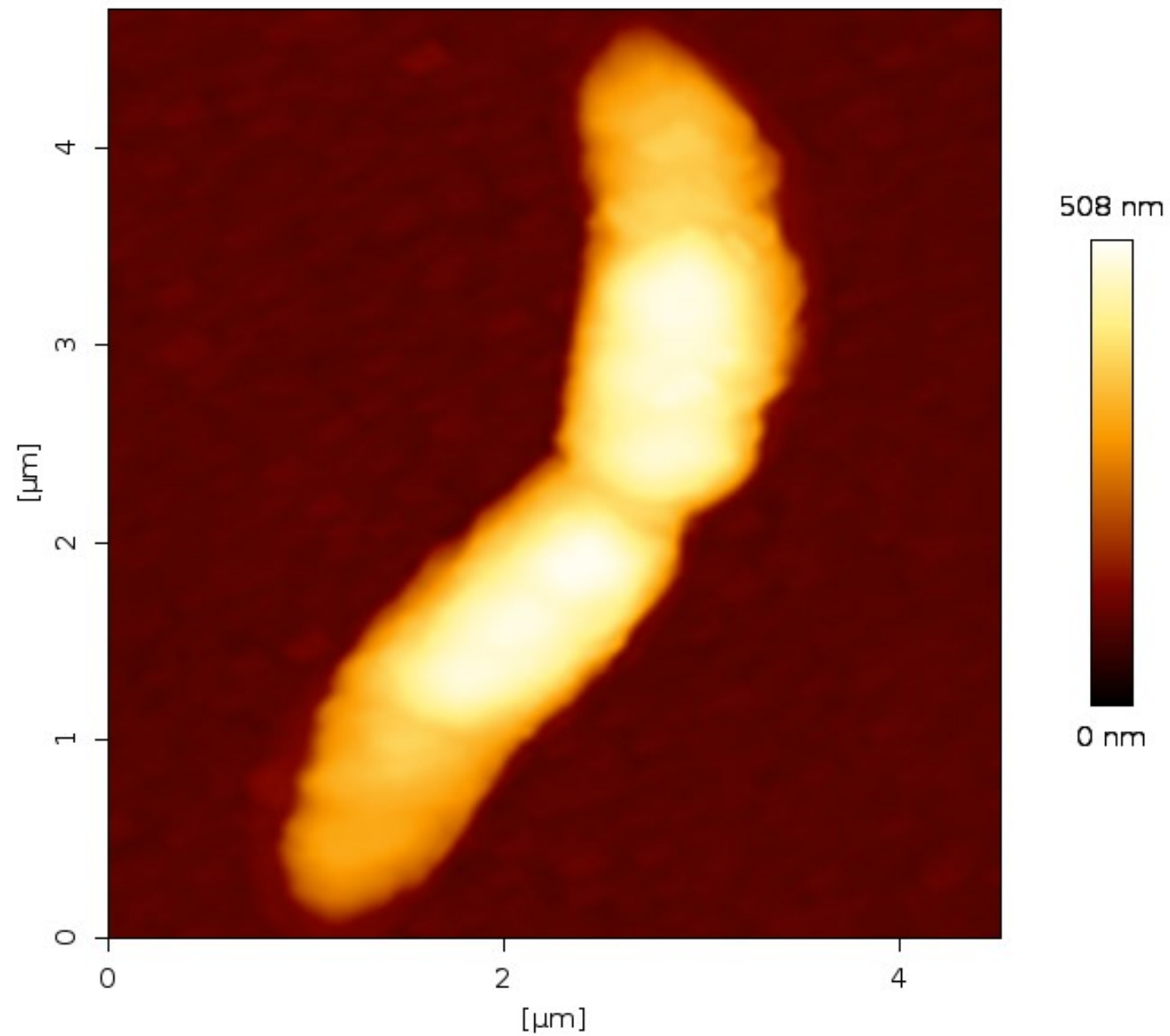
Comparison of Halomonas halophila measured on air (left) and in liquid (right)



Rhodospirillum rubrum

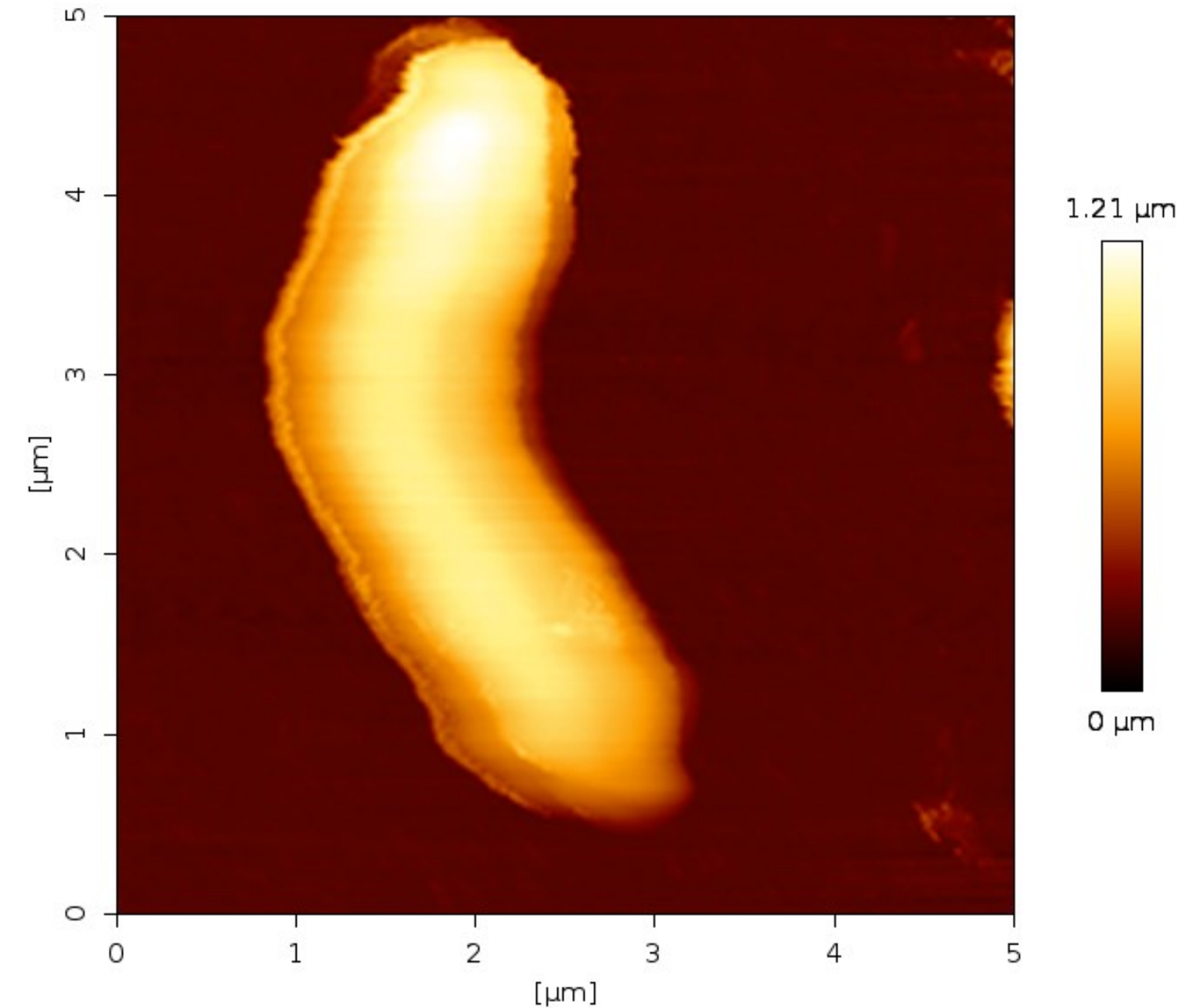
AIR

(28,6 % PHA)

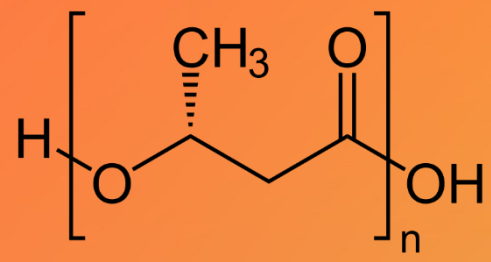


PBS

(28,6 % PHA)



Comparison of Rhodospirillum rubrum cultivated 24 hours on acetate measured on air (left) and in liquid (right)



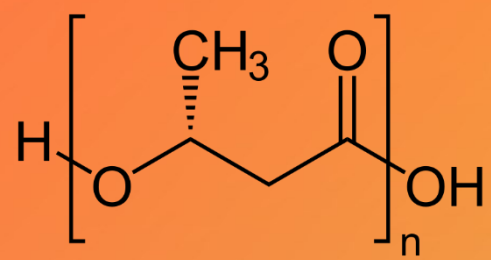
JUST TO SUM IT UP

AIR

- TESPA-V2
- FIXATION BY DRYING
- QI MODE
- DILUTION 10×

LIQUID

- SNL – B, MLCT – A
- FIXATION WITH POLY-L-LYSINE
- QI MODE
- DILUTION ACCORDING TO OPTICAL DENSITY
(2×, 10×, 20× a 50×)



SPECIAL THANKS TO:

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