*Soft Matter*

Electronic Supplementary Information on

# Time lapse AFM on vesicle formation from mixed lipid bilayers induced by the membrane-active peptide melittin

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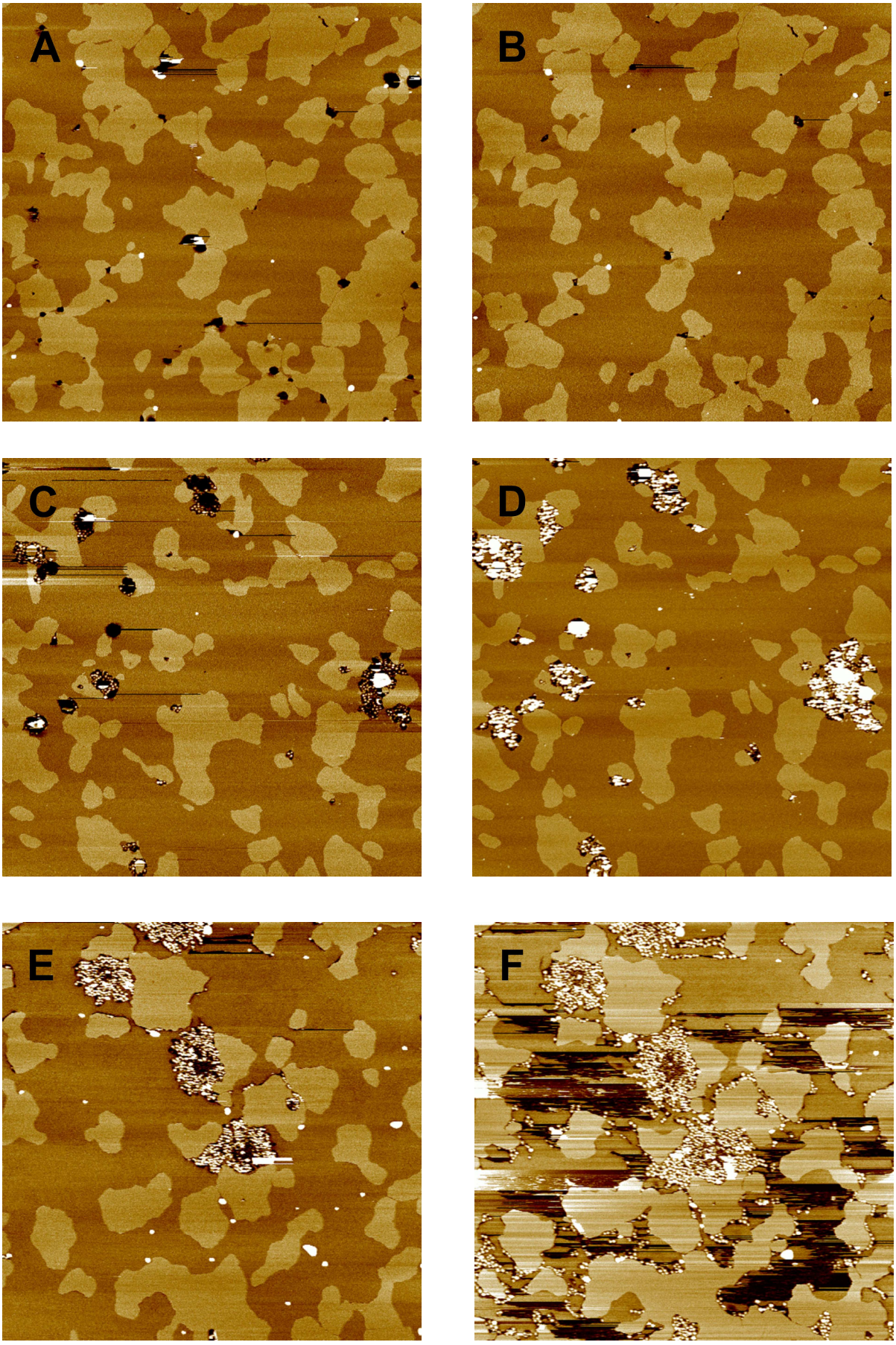

A
B
C
D
E
F

**Figure S1.** Time lapse AFM on supported DOPC-DPPC bilayer on mica after addition of melittin. **(A, B)** 0.1 µM melittin, where (A) is taken 7 min, (B) 35 min after addition of melittin; no bilayer restructuring can be observed. **(C, D)** 0.5 µM melittin, where (C) is taken 8 min, (D) 19 min after addition of melittin; vesicular structures aggregate in membrane defects. **(E, F)** 5.0 µM melittin, where (E) is taken 9 min, (F) 17 min after addition of melittin; high concentration of melittin leads to a strong and fast degradation of DOPC domains and vesicle formation. Image sizes 5 x 5 $\mu m^2$, z-scale 7 nm.

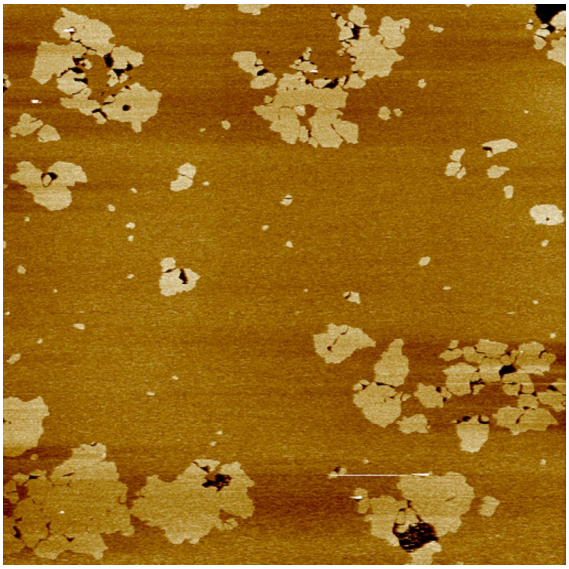
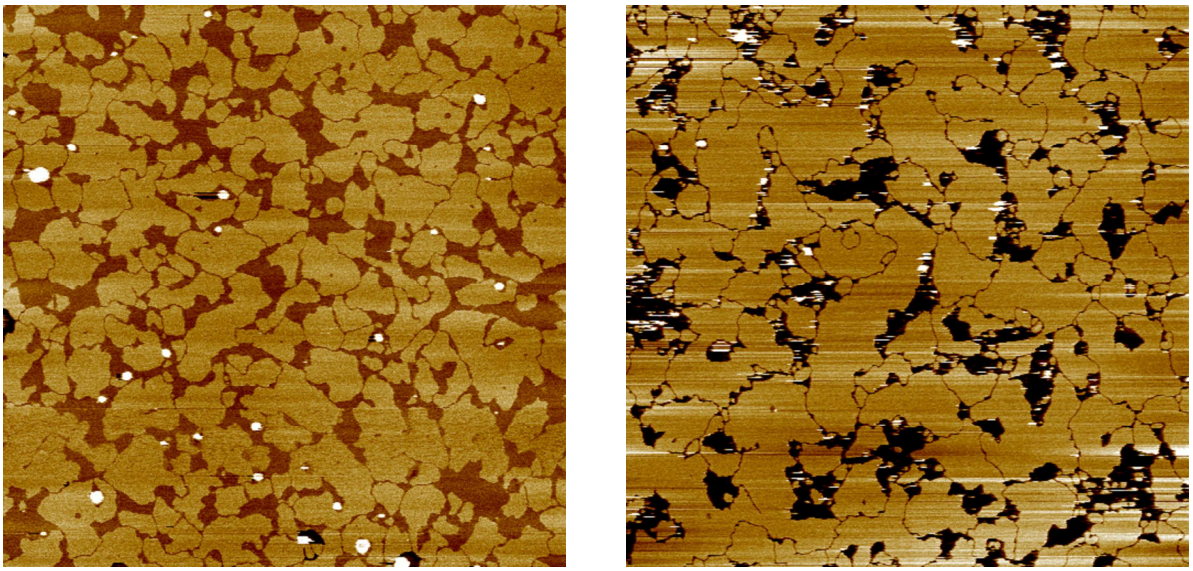


**Figure S2.** AFM on supported DOPS-DPPC (50%:50%) bilayer on mica 31 min after the addition of 2 µM melittin. This is about the same time scale and melittin concentration as in Fig. 1D. Compared to the bilayer with uncharged lipids in Fig 1D no (or only very few) effects of the action of melittin can be observed. Image size 4 x 4 $\mu m^2$, z-scale 7 nm.



F**igure S3.** AFM on supported DOPS-DPPC (15%:85%) bilayer on mica. (Left) without melittin, (Right) 36 min after addition of a high concentration of melittin (34 µM). A removal of the lower domains can be observed, but less vesicles can be seen. Many of the higher DPPC domains fused together. Image sizes 5 x 5 $\mu m^2$, z-scale 7 nm.

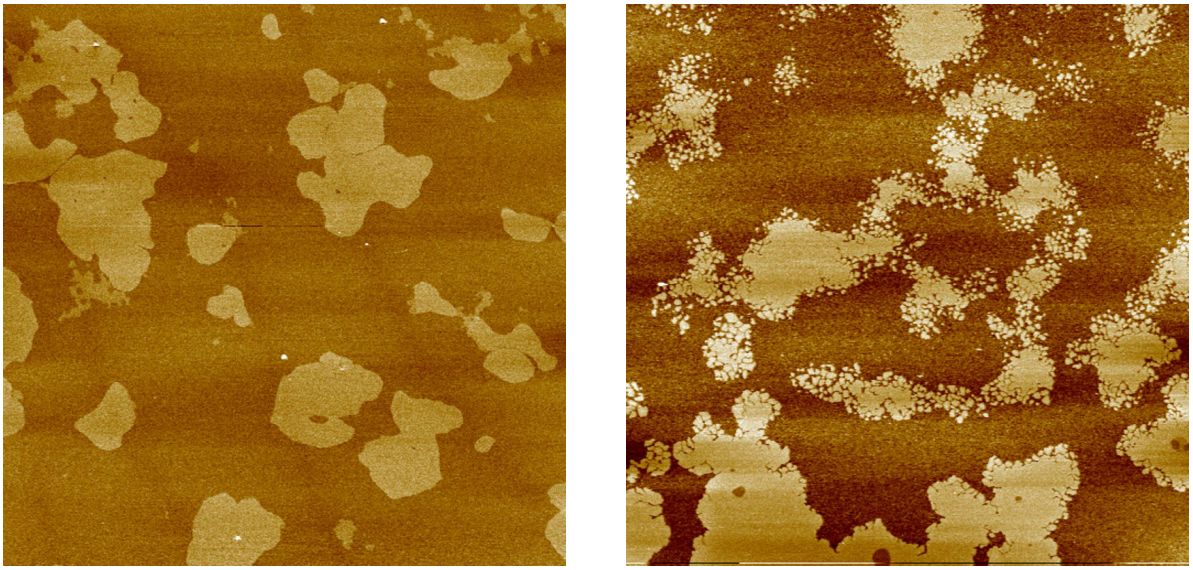


**Figure S4.** AFM on supported DOPC-DPPC bilayer on mica after addition of phospholipase $A_2$ (Left) in imaging buffer without $Ca^{2+}$ after 30 min; domains are still intact. (Right) in imaging buffer with $Ca^{2+}$ after 10 min; a degradation of domains can be observed. None of the observed effects resembles the action of melittin as seen in Figures 1 - 3. Image sizes 5 x 5 $\mu m^2$, z-scale 7 nm.

**Table S1.** Domain Size Expansion for Figures 1A - 1C. Domains are assigned as shown in the figure below.

| Domain Number | 1 | 2 | 3 | 4 | 5 | 6 | 7 |
|---|---|---|---|---|---|---|---|
| Area Increase [%] | 54 | 49 | 54 | 25 | 54 | 14 | 19 |
| **Domain Number** | **8** | **9** | **10** | **11** | **12** | **13** | **14** |
| Area Increase [%] | 45 | 0 | 13 | 150 | 18 | 19 | 15 |

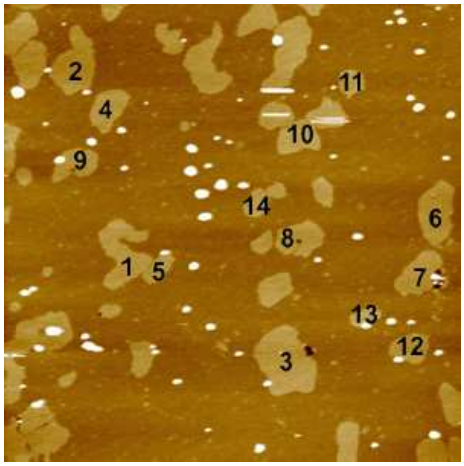



**Table S2**. Domain Size Expansion for Figures S1E - S1F. Domains are assigned as shown in the figure below.

| Domain Number | 1 | 2 | 3 | 4 | 5 | 6 |
|---|---|---|---|---|---|---|
| Area Increase [%] | 4 | 0 | 6 | -4 | 0 | -18 |
| **Domain Number** | **7** | **8** | **9** | **10** | **11** | **12** |
| Area Increase [%] | 2 | 28 | 18 | 56 | 58 | 18 |

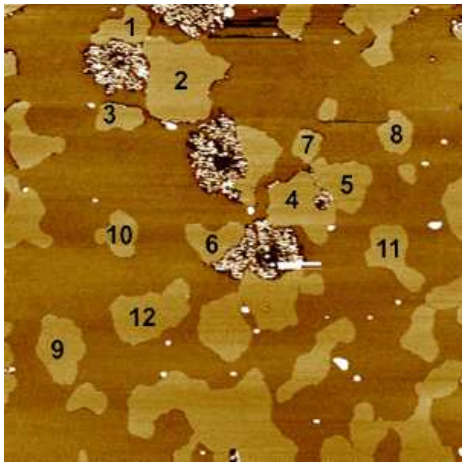


**Table S3**. Defect area increase for the experiment shown in Figure 1 (image for 16 min not shown). A linear time dependence resulted in a fit with an $R^2$ value (coefficient of determination) of 0.9986. The area increases by about 0.08 $\mu m^2$ per minute.

| Time | 8 min | 16 min | 24 min | 32 min |
|---|---|---|---|---|
| **Area absolute [$\mu m^2$]** | 0.34 | 0.89 | 1.54 | 2.18 |