Supplementary Information for

Investigating Metal-Organic Frameworks Anchors for Giant Unilamellar Vesicle Immobilization

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Chemical Reagents

All reagents were purchased from Sigma Aldrich, Fisher Scientific and Avanti Polar Lipids and used as received, unless otherwise stated.

GUV Lipid Composition

GUVs were synthesized using standard electroformation protocol as reported in our previous study.[1] The lipids 1-Palmitoyl-2-oleoyl-glycero-3-phosphocholine (POPC), 1-palmitoyl-2-oleoyl-sn-glycero3-phospho-(1-rac-glycerol) (sodium salt) (POPG), 1,2-dioleoyl-sn-glycero-3-phosphocholine (DOPC), 1,2-dipalmitoyl-sn-glycero-3-phosphocholine (DPPC) and cholesterol were purchased from Avanti Polar Lipids (dissolved in chloroform to a concentration of 20 mg/mL stock solution). The fluorescent tag Topfluor® cholesterol was also purchased from Avanti Polar Lipids and was made up to 1 mg/mL in chloroform. Three different lipid mixtures were prepared a) POPC, POPG and cholesterol were mixed in a 4:1:1 molar ratio b) POPC and cholesterol were mixed in 7:3 molar ratio c) DOPC, DPPC and cholesterol were mixed in 1:1:20 mol% ratio. Each lipid mixture was made to an overall 2 mg/ml concentration and TopFluor® cholesterol 0.1 mol% of the concentration of cholesterol was added for imaging purpose.

Electroformation Protocol

The non-conductive sides of two ITO coated glass electrodes were marked with a circle of 13 mm diameter. An aliquot of 2 μL for each lipid mixture was gently spread onto the electrically conductive sides of each of the ITO slide on the marked area using a microsyringe. The ITO slides were dried in a vacuum desiccator for at least 30 minutes to fully remove the organic solvent. The slides were loosely covered with aluminum foil to keep the samples in the dark. A 2 mm thick O-ring with a diameter of 14 mm was fixed with minimal amount of silicon grease onto one of the two slides. For each experiment, 160 μL of electroformation buffer solution (200 mM sucrose and 1 mM HEPES pH 7.4 in DI water) with desired MOF approx. 5 mg (a microspatula tip) was vortexed and filled into the chamber. The electroformation solution containing MOF should be left to settle for 5 minutes on ITO slides. The second ITO slide was then put together to form a closed chamber such that the conducting sides of the slides faces each other in NANION Vesicle Prep Pro.

Swelling of POPC:POPG:Chol (4:1:1) and POPC:Chol (7:3) lipid films was done by applying a 10 Hz sinusoidal AC electric field at 35°C. The amplitude of the applied field was linearly increased from 0.1 V - 0.5 V (peak to peak) over 30 minutes. The voltage was then further increased over 15 minutes to 1.6 V and remained constant for 2 hours to grow the vesicles. Finally, the voltage was slowly lowered to 0 V in 5 minutes to peel the vesicles off from the electrodes. For the electroformation of DOPC:DPPC:Chol (1:1:20 mol%) vesicles, a 10 Hz sinusoidal AC electric field at 45°C was applied where the voltage was ramped from 0 V to 2V within first 25 minutes and remained constant at 2V for 2 hours, followed by decrease to 0 V in 5 minutes to end the protocol. The samples were shielded from external light during electroformation. Once electroformed, GUVs were diluted in resuspension buffer solution (200 mM glucose and 1 mM HEPES pH 7.4 in DI water) and transferred to an imaging well for phase contrast microscopy. The GUVs were handled using a plastic pipette with the end cut off to an opening of at least 5 millimetres to prevent lysing of vesicles during the transfer processes.

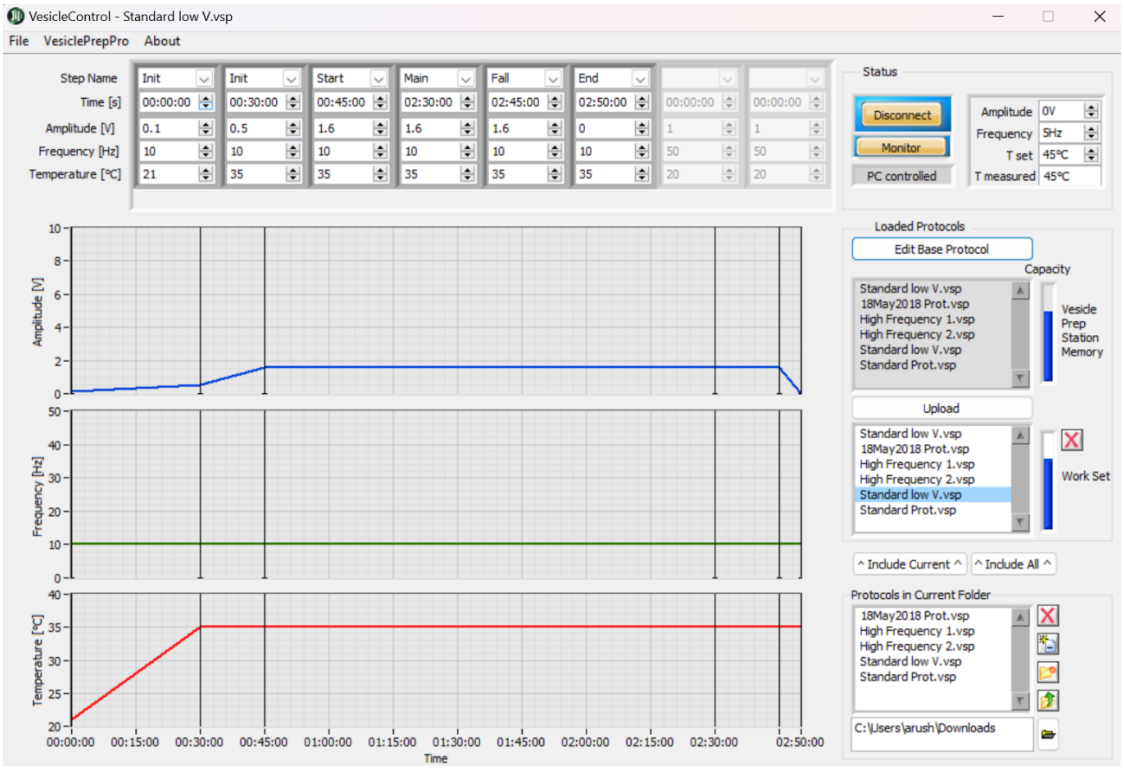

Figure S1. Electroformation protocol parameters displayed on the Nanion Vesicle Prep Pro *VesicleControl* software for POPC:POPG:Chol and POPC:Chol lipid mixtures.

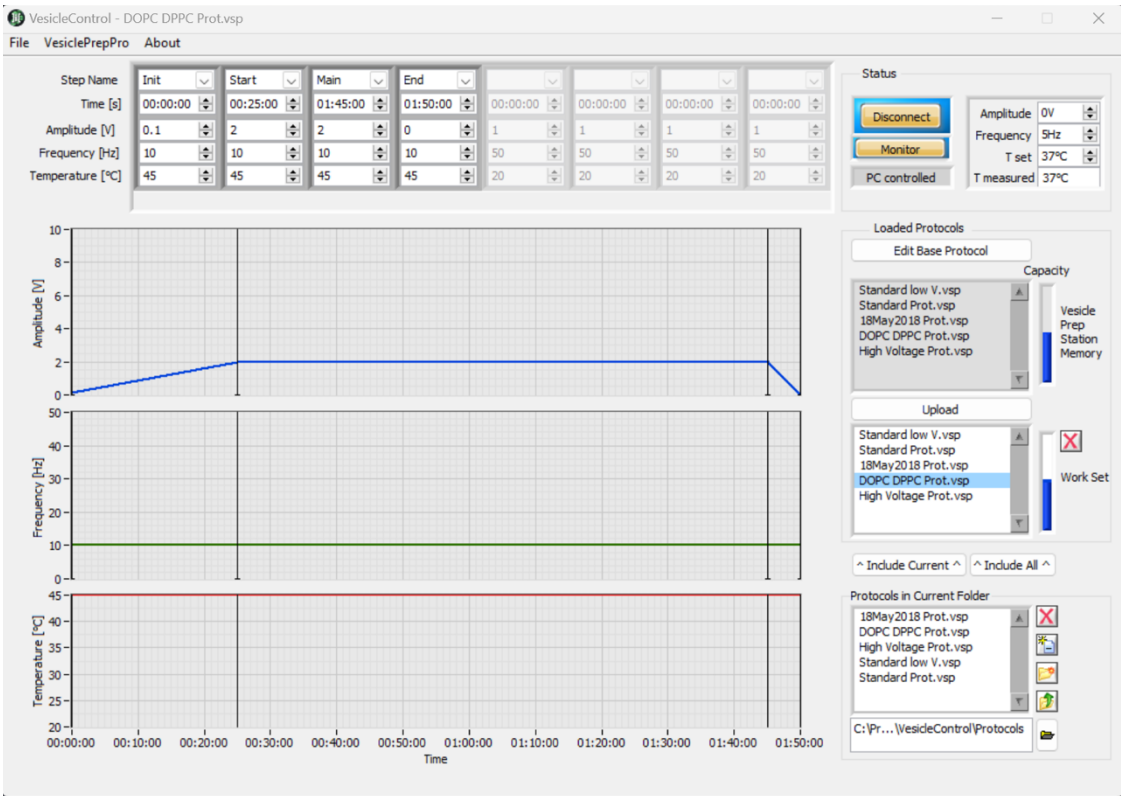

Figure S2. Electroformation protocol parameters displayed on the Nanion Vesicle Prep Pro *VesicleControl* software for DOPC:DPPC:Chol lipid mixture.

GUV Imaging

To assess the influence of various MOFs on GUV formation, we conducted experiments across three independent runs for each lipid mixture associated with each MOF. Following electroformation, the GUVs were suspended in 2 mL of resuspension buffer and subsequently divided into four imaging wells per experiment. Images of the anchored GUVs were captured from each well using Echo Discover Revolve Fluorescent microscope in the FITC channel immediately after they were formed. The micron-sized MOF particles have absorbed TopFluor Chol and hence fluoresce with the GUVs. To determine whether MOFs supported GUV formation, three representative images containing the maximum number of GUVs were selected for each MOF, and the GUVs were counted. This quantification was then plotted to provide a comparative visual representation of GUV yields across the different MOFs. To address the anchoring of GUVs by

respective MOFs, a small amount (200 µL) of the GUV suspension was dispensed on a glass slide, and the vesicles were observed for 5 minutes. The flow of solution made unbound GUVs move while the ones anchored by MOFs remained stationary or attached. (see supplementary videos).

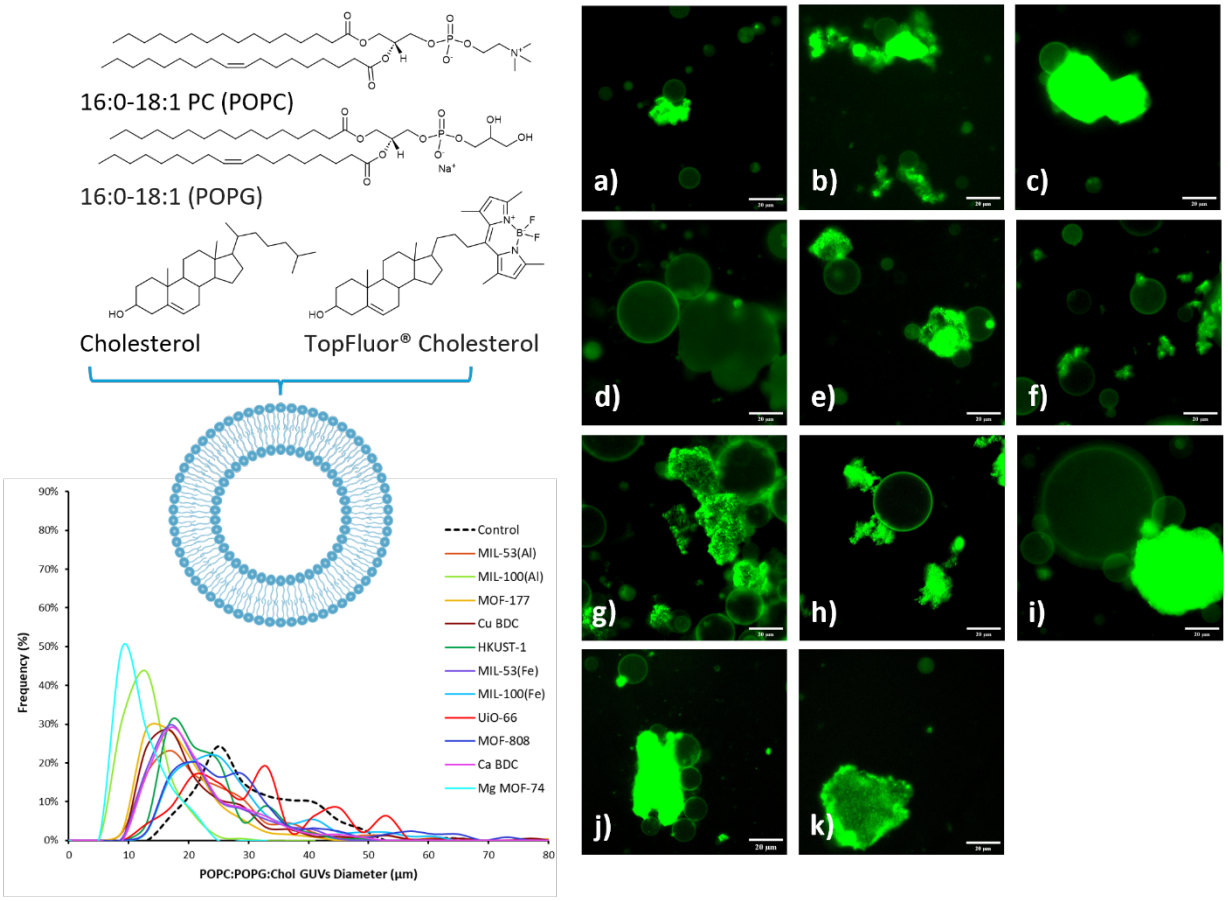

Figure S3. Fluorescence images of POPC:POPG:Chol (4:1:1) GUVs immobilized by MOF particles **a)** GUV/MIL-53(Al)/GUV **b)** GUV/MIL-100(Al) **c)** GUV/MOF-177 **d)** GUV/GUV/CuBDC **e)** GUVs/HKUST-1 **f)** GUVs/MIL-53(Fe) **g)** GUVs/MIL-100(Fe) **h)** UiO-66/GUV/UiO-66 **i)** GUVs/MOF-808 **j)** GUVs/CaBDC **k)** MgMOF-74. Scale bars represent 20 µm.

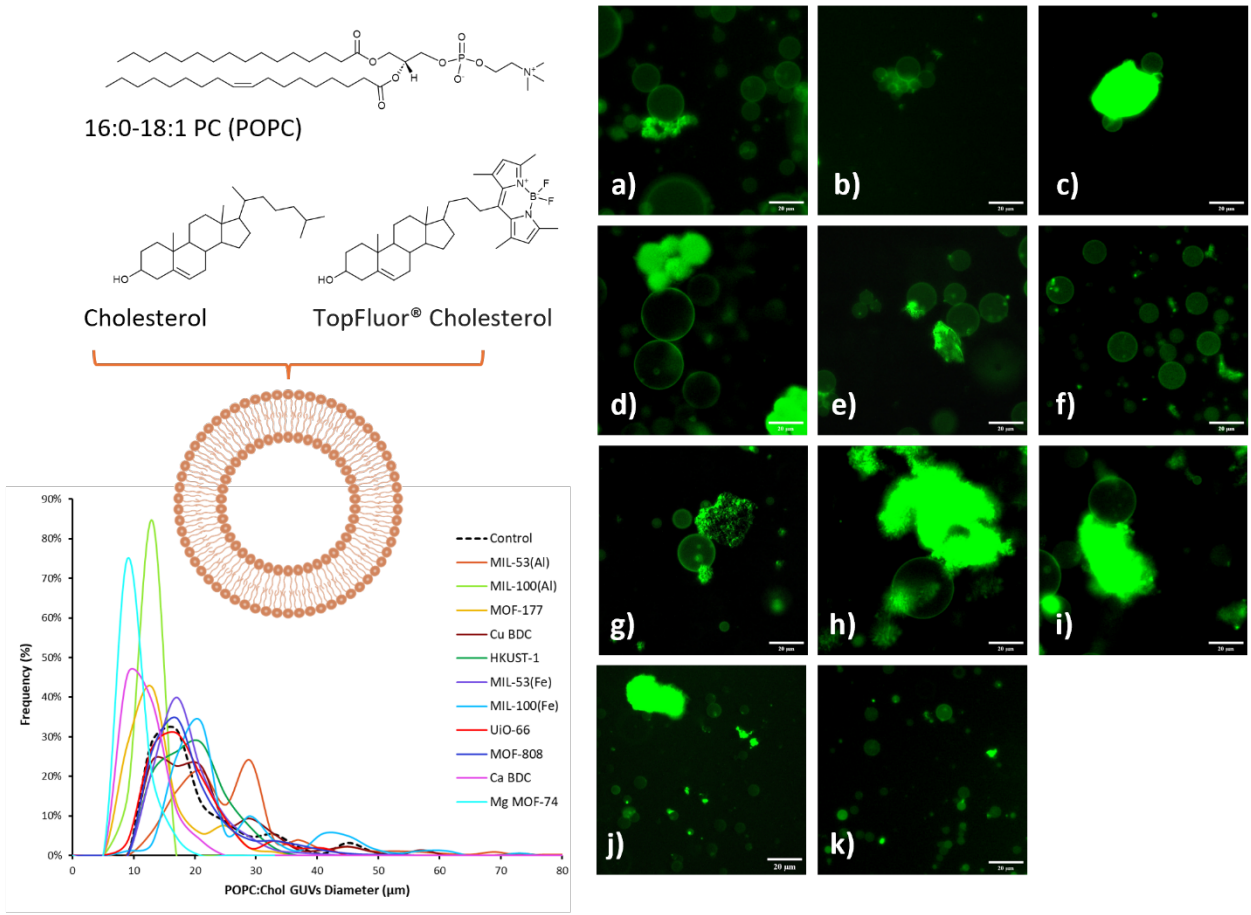

Figure S4. Fluorescence images of POPC:Chol (7:3) GUVs immobilized by MOF particles **a)** GUV/GUV/MIL-53(Al) **b)** GUVs in MIL-100(Al) **c)** GUV/MOF-177/GUV **d)** GUV/GUV/CuBDC **e)** GUVs/HKUST-1 **f)** GUVs/MIL-53(Fe) **g)** MIL-100(Fe)/GUVs/MIL-100(Fe) **h)** GUV/UiO-66 **i)** GUV/MOF-808 **j)** GUVs in CaBDC **k)** GUVs in MgMOF-74. Scale bars represent 20 µm.

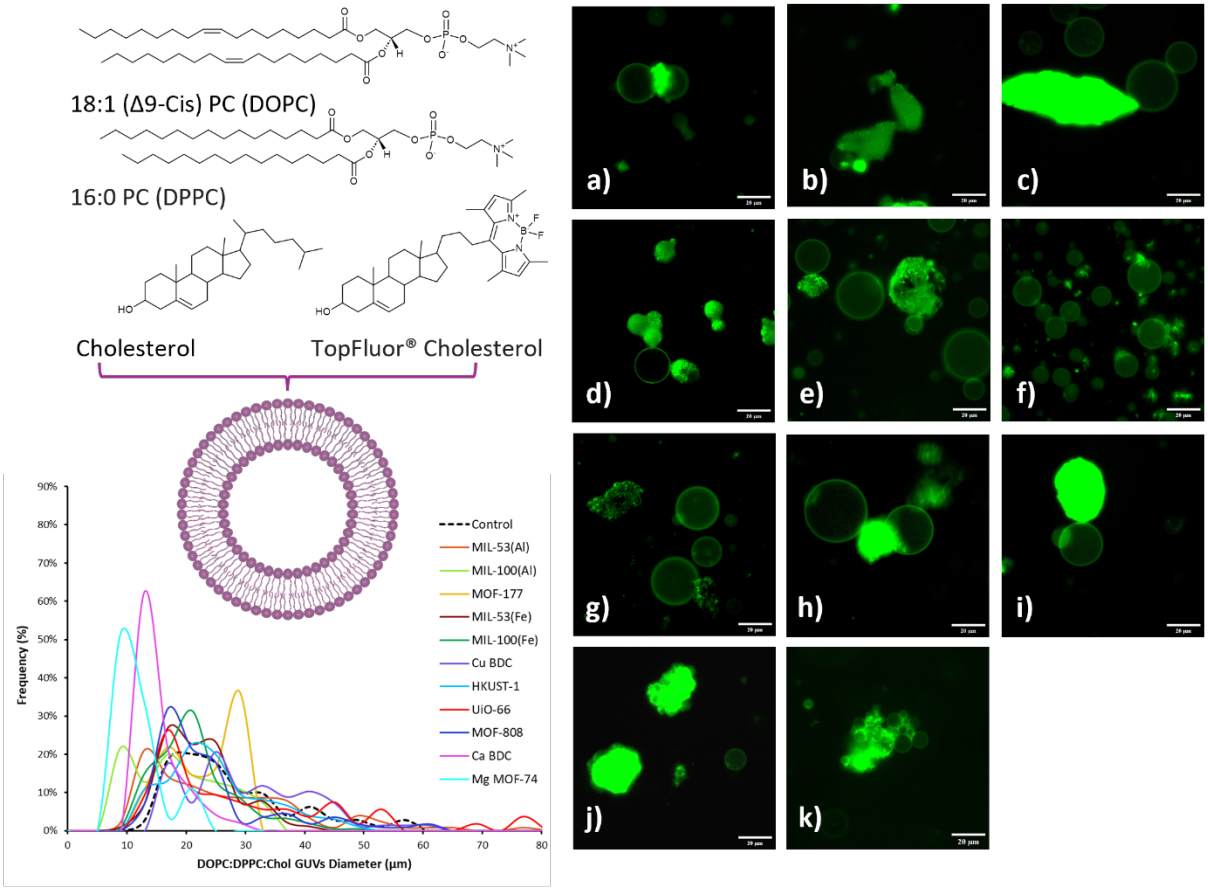

Figure S5. Fluorescence images of DOPC:DPPC:Chol (1:1:20 mol%) GUVs immobilized by MOF particles **a)** GUV/MIL-53(Al) **b)** GUV/MIL-100(Al) **c)** MOF-177/GUV/GUV **d)** CuBDC/GUV/CuBDC **e)** GUVs/HKUST-1 **f)** GUVs/MIL-53(Fe) **g)** GUV/MIL-100(Fe) **h)** GUV/UiO-66/GUV **i)** GUV/MOF-808 **j)** GUV in CaBDC **k)** GUVs in MgMOF-74. Scale bars represent 20 µm.

Vesicle Diameter

The size distribution of GUVs is illustrated based on their lipidic composition; the diameters are rounded to the nearest whole number. The graphs are produced from analyzing GUVs samples from six separate runs (two per lipid mixture) for each MOF.

Aluminium MOFs with Phospholipids

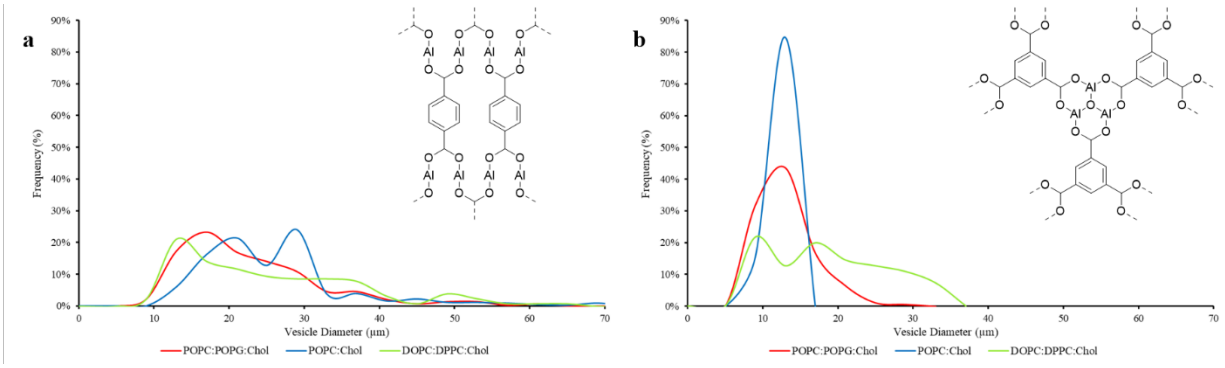

Figure S6. Size distribution of GUVs formed with POPC:POPG:Chol, POPC:Chol and DOPC:DPPC:Chol lipid composition in presence of MOFs **a)** MIL-53(Al) with mean diameter of 28 µm, 18 µm and 26 µm, respectively **b)** MIL-100(Al) with mean diameter of 21 µm, 24 µm and 24 µm, respectively. The graphs are produced from a dataset of 1010 GUVs formed in MIL-53(Al) and 254 GUVs in MIL-100(Al) in total.

Zinc MOF with Phospholipids

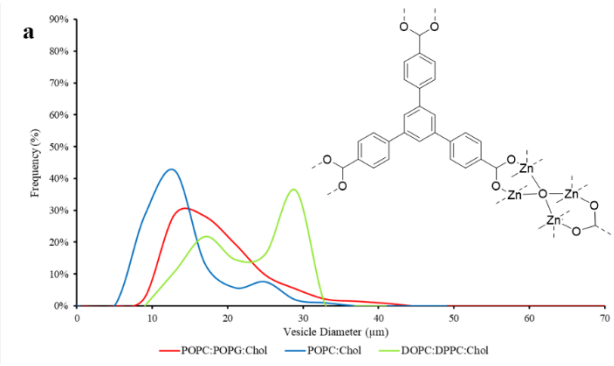

Figure S7. Size distribution of GUVs formed with POPC:POPG:Chol, POPC:Chol and DOPC:DPPC:Chol lipid composition in presence of MOF-177 with mean diameter of 18 µm, 12 µm and 28 µm, respectively. The graph is produced from a dataset of 666 GUVs formed in MOF-177 in total.

Copper MOFs with Phospholipids

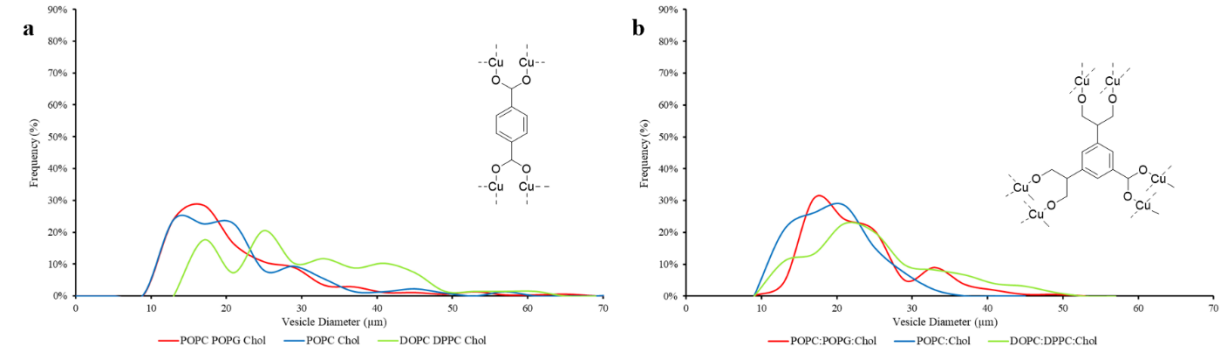

Figure S8. Size distribution of GUVs formed with POPC:POPG:Chol, POPC:Chol and DOPC:DPPC:Chol lipid composition in presence of **a)** CuBDC MOF with mean diameter of 20 µm, 20 µm and 28 µm, respectively **b)** HKUST-1 with mean diameter of 21 µm, 18 µm and 23 µm, respectively. The graphs are produced from a dataset of 687 GUVs formed in CuBDC and 742 GUVs in HKUST-1 in total.

Iron MOFs with Phospholipids

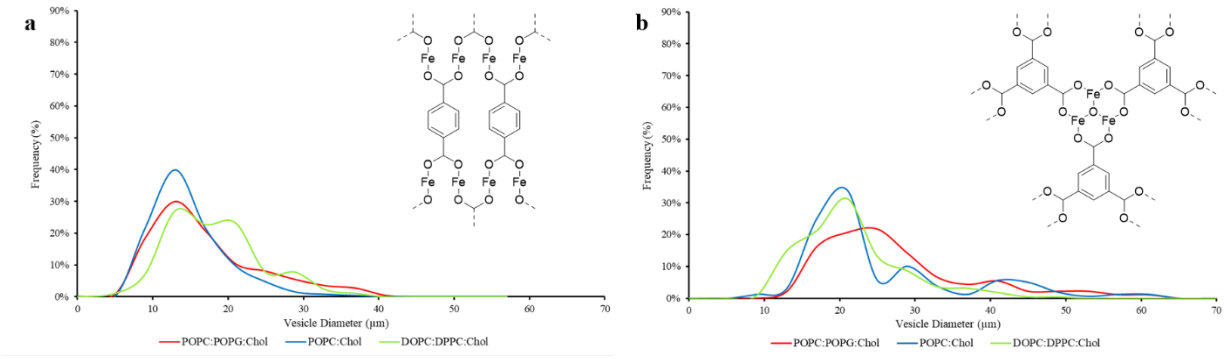

Figure S9. Size distribution of GUVs formed with POPC:POPG:Chol, POPC:Chol and DOPC:DPPC:Chol lipid composition in presence of **a)** MIL-53(Fe) with mean diameter of 19 μm, 17 μm and 20 μm, respectively **b)** MIL-100(Fe) with mean diameter of 25 μm, 23 μm and 20 μm, respectively. The graphs are produced from a dataset of 720 GUVs formed in MIL-53(Fe) and 471 GUVs in MIL-100(Fe) in total.

Zirconium MOFs with Phospholipids

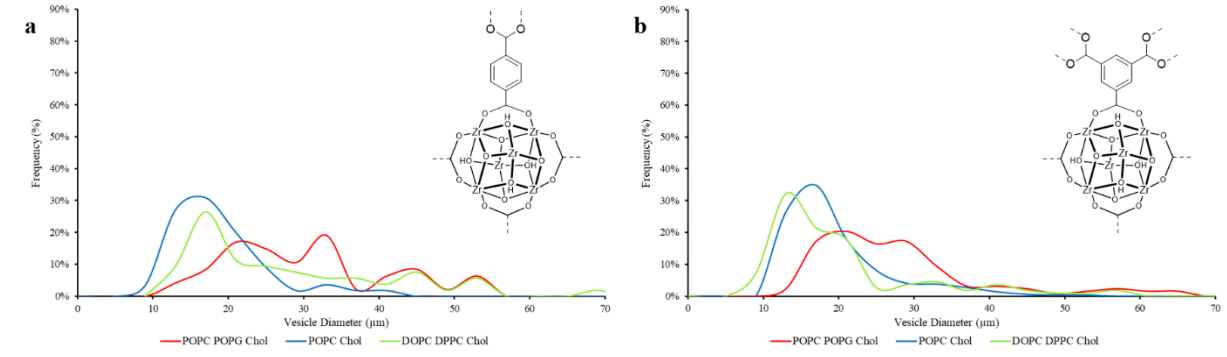

Figure S10. Size distribution of GUVs formed with POPC:POPG:Chol, POPC:Chol and DOPC:DPPC:Chol lipid composition in presence of **a)** UiO-66 with mean diameter of 28 μm, 17 μm and 28 μm, respectively **b)** and MOF-808 with mean diameter of 26 μm, 18 μm and 22 μm, respectively. The graphs are produced from a dataset of 155 GUVs formed in UiO-66 and 614 GUVs in MOF-808 in total.

Calcium and Magnessium MOFs with Phospholipids

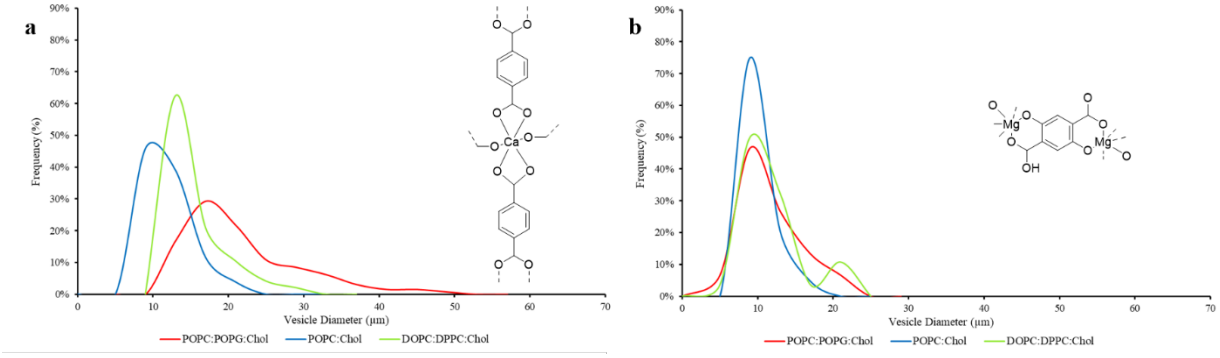

Figure S11. Size distribution of GUVs formed with POPC:POPG:Chol, POPC:Chol and DOPC:DPPC:Chol lipid composition in presence of **a)** CaBDC with mean diameter of 20 µm, 10 µm and 14 µm, respectively **b)** MgMOF-74 with mean diameter of 10 µm, 8 µm and 10 µm, respectively. The graphs are produced from a dataset of 335 GUVs formed in Ca BDC and 105 GUVs in Mg MOF-74 in total.

GUV Immobilization and Viability Images

The GUVs were imaged at intervals of 6 hours and 12 hours, and the following section presents the results, highlighting the immobilized GUVs observed during these time periods.

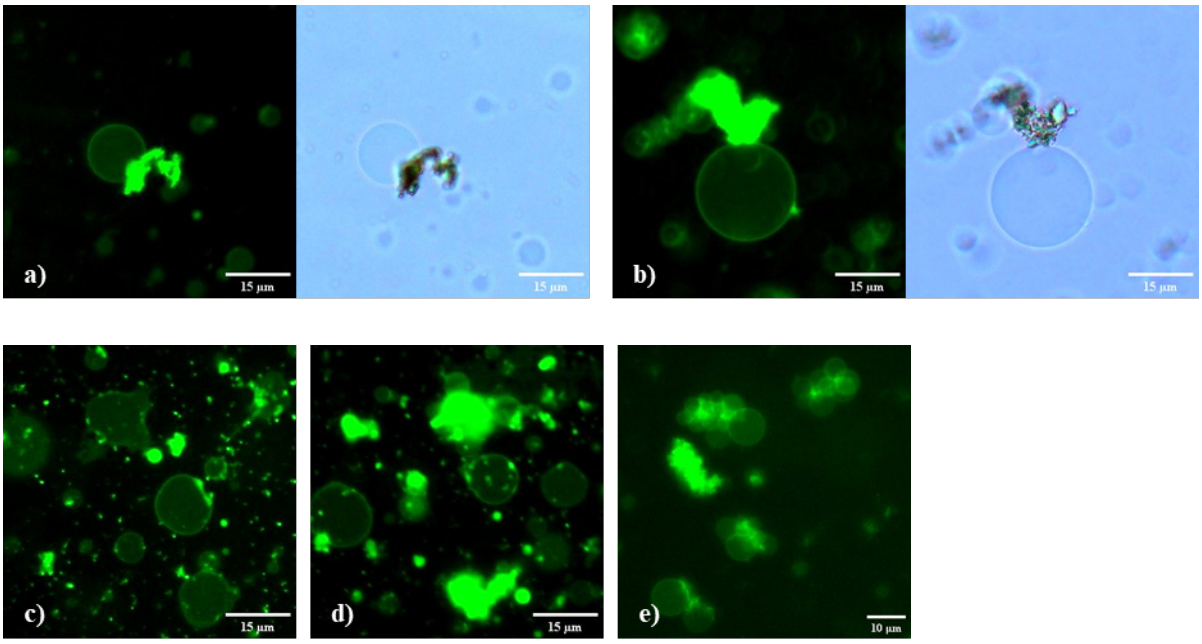

Figure S12 MIL-100(Al) immobilized GUVs **a)** after 6 hours **b)** 12 hours **c)** deformed GUVs after 12 hours **d)** deformed GUVs **e)** GUV aggregates.

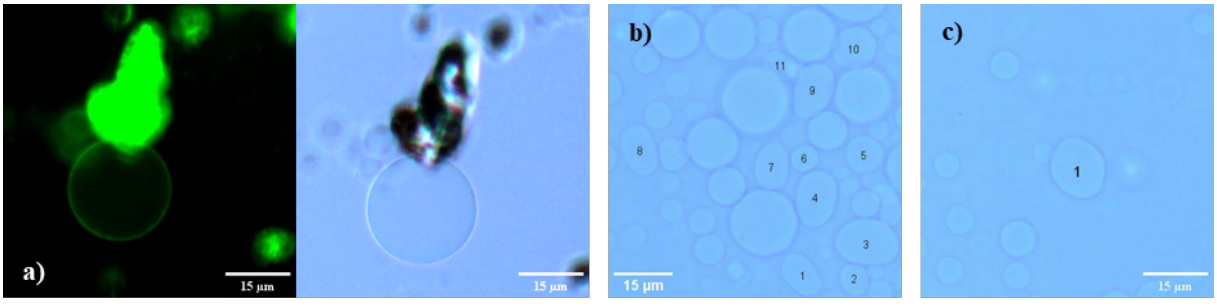

Figure S13 MOF-177 immobilized GUVs **a)** after 6 hours **b)** oblong shaped GUVs seen after electroformation **c)** oblong shaped GUV.

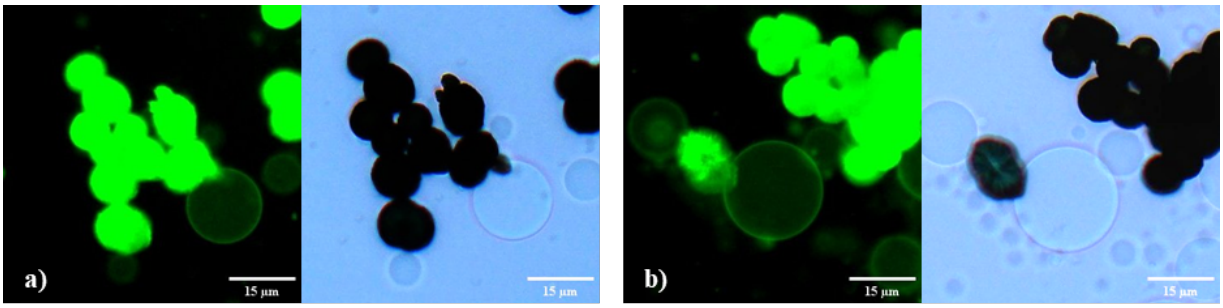

Figure S14 CuBDC immobilized GUVs **a)** after 6 hours **b)** after 12 hours.

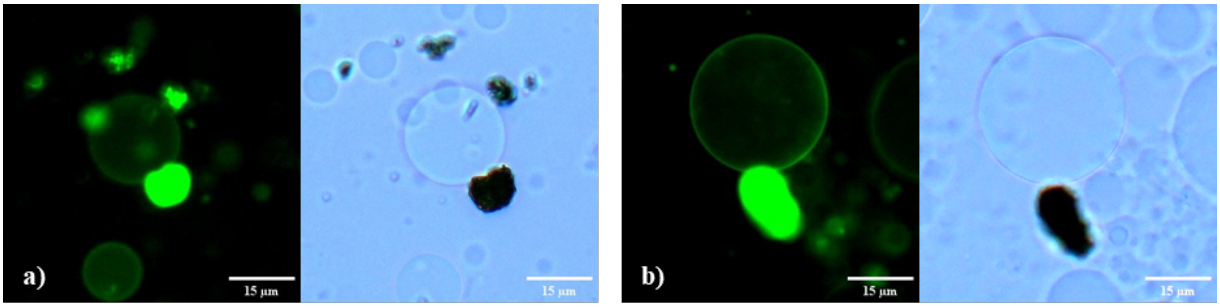

Figure S15 HKUST-1 immobilized GUVs **a)** after 6 hours **b)** after 12 hours.

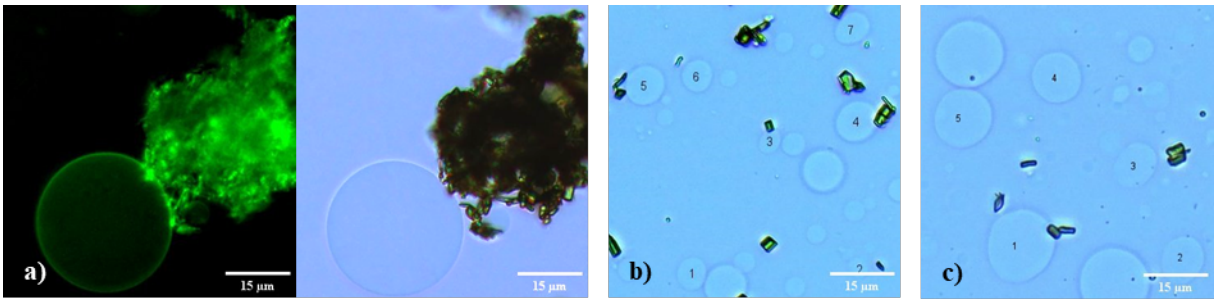

Figure S16 MIL-53(Fe) immobilized GUVs **a)** GUV anchored to cluster after 6 hours **b)** unbound mobile GUVs **c)** oblong shaped GUVs.

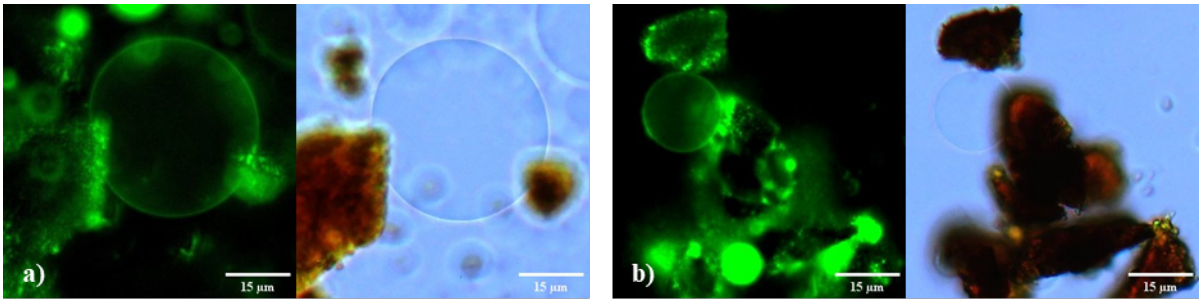

Figure S17 MIL-100(Fe) immobilized GUVs **a)** after 6 hours **b)** after 12 hours.

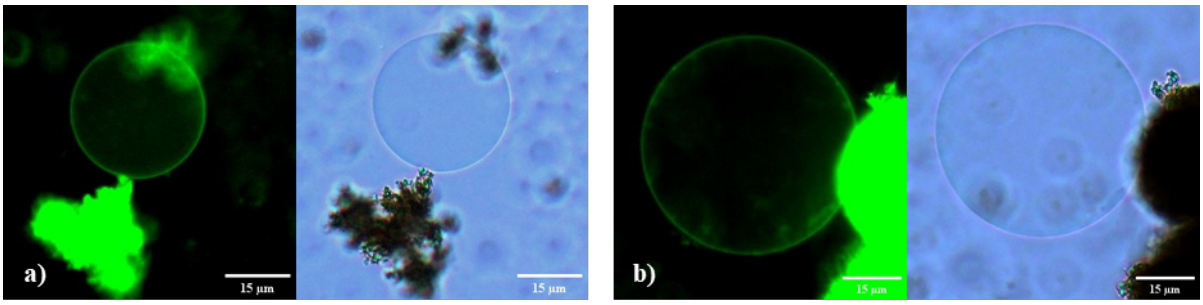

Figure S18 UiO-66 immobilized GUVs **a)** after 6 hours **b)** after 12 hours.

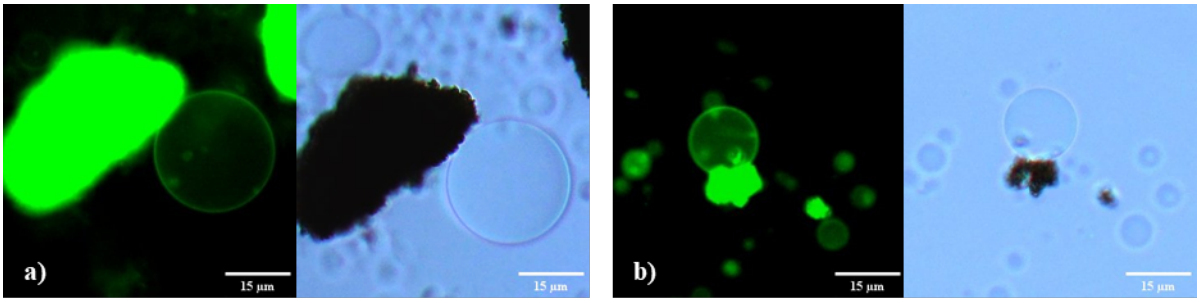

Figure S19 MOF-808 immobilized GUVs **a)** after 6 hours **b)** after 12 hours.

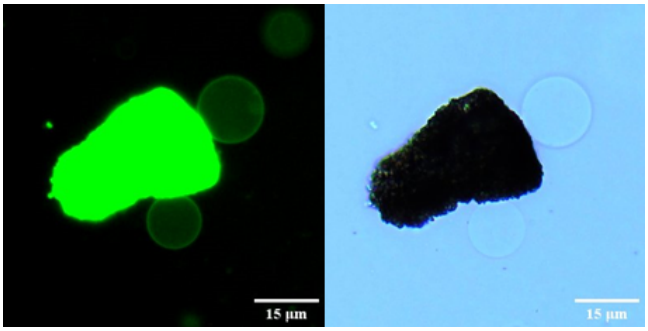

Figure S20 CaBDC immobilized GUVs after 6 hours

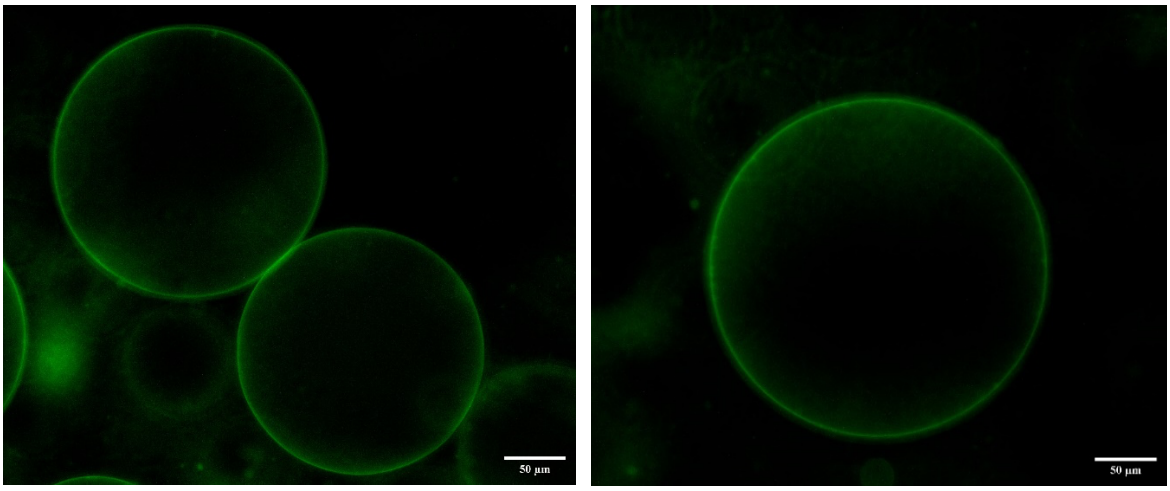

Figure S21 Giant vesicles in MIL-53(Al)

MOF Synthesis Protocols

All MOF particles were synthesized based on literature procedures and were characterized using SEM, EDS, and PXRD.

Powder x-Ray diffractograms were measured on a Malvern P'Analyrical Empyeran Powder X-Ray diffractometer using a copper source. The scan step size was set to 0.008356 with a time per scan of 10.795 seconds.

The SEM images were collected at the UNB Microscopy and Microanalysis Facility with a JEOL JSM-6400 Scanning Electron Microscope using an accelerating voltage of 15 kV. Images were acquired using a Digiscan II operated by Gatan Digital Micrograph software. The SEM images of UiO-66 And Mg MOF-74 were captured by ThermoScientific Scios 2 Dualbeam SEM system. The MOF samples were attached to mounting stubs using a carbon tape and coated with gold for conductivity by sputtering using an Edwards S150A coater.

The EDS analysis of MOFs was also performed at the UNB Microscopy and Microanalysis Facility with a JEOL JSM-6400 Scanning Electron Microscope equipped with an EDAX Genesis 4000 Energy Dispersive X-ray (EDS) analyser. The MOF samples were carbon coated using an Edwards 306A carbon coater prior to observation. EDS analysis was performed at an accelerating voltage of 15 kV and a beam current of 1.5 nA, with a working distance of 14 mm. Collection time was 50 seconds per analysis point. The EDS of UiO-66 was performed on ThermoScientific Scios 2 Dualbeam, equipped with an Oxford Ultim Max 170 EDS detector, and an Oxford Symmetry EBSD detector controller by the Aztec software using similar conditions with beam current 3.2 nA.

MIL-53(Al)

2.5835 g (8.048 mmol) of $Al(NO_3)_3 \cdot 6H_2O$ and 5.7200 g (34.45 mmol) of terephthalic acid were combined in a 50 mL flask, followed by 10 mL of water. The solution was sonicated for 5 minutes, then transferred to a 50 mL Teflon lined autoclave and heated at 150ºC overnight.[2]

Once removed from heat the reaction vessel was allowed to cool to room temperature. The solution and precipitate were transferred into a centrifuge tube, centrifuged for 5 minutes, and the solution was decanted. 10 mL of water was added to the centrifuge tube and the precipitate was shaken to

redistribute. The mixture was then centrifuged for 5 minutes, and the solution decanted. This process was completed 3 times with water, and 3 with methanol.

The remaining precipitate was transferred to a 50 mL round bottom flask and 30 mL of DMF was added, followed by refluxing overnight. Once reflux was completed, the mixture was cooled to room temperature and transferred to a centrifuge tube, centrifuged for 5 minutes, and the solution was decanted. 10 mL of methanol was added, centrifuged for 5 minutes, and the liquid was decanted. The remaining precipitate was placed in an 80ºC oven for 3 hours to dry.

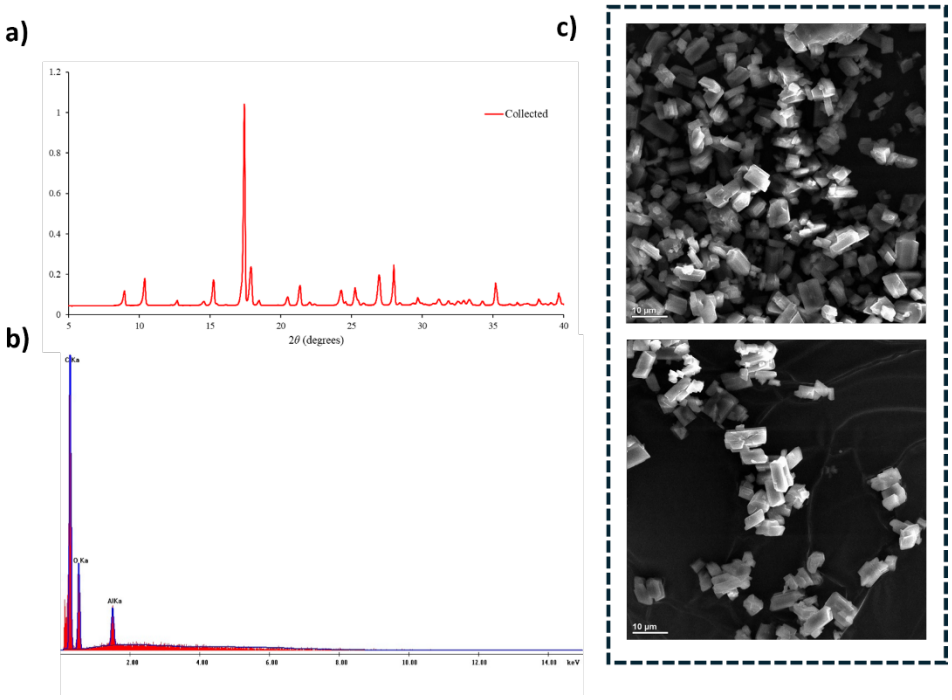

Figure S22. a) pxrd collected b) EDS c) SEM of MIL-53(Al)

MIL-100(Al)

0.4020 g (1.252 mmol) of $Al(NO_3)_3 \cdot 6H_2O$ and 0.0.1827 g (0.8694 mmol) of 1,3,5-tirmethy benzene dicarboxylate were combined in a 50 mL flask, followed by 5 mL of water and 0.06 mL of DMF. The solution was sonicated for 5 minutes, then transferred to a 50 mL Teflon lined autoclave and heated at 220ºC for 4 hours.[3]

Once removed from heat the reaction vessel was allowed to cool to room temperature. The solution and precipitate were transferred into a centrifuge tube, centrifuged for 5 minutes, and the solution was decanted. 10 mL of water was added to the centrifuge tube and the precipitate was shaken to redistribute. The mixture was then centrifuged for 5 minutes, and the solution decanted. This process was completed 3 times with water, and 3 with methanol.

The remaining precipitate was transferred to a 50 mL round bottom flask and 30 mL of DMF was added, followed by refluxing overnight. Once reflux was completed, the mixture was cooled to room temperature and transferred to a centrifuge tube, centrifuged for 5 minutes, and the solution was decanted. 10 mL of methanol was added, centrifuged for 5 minutes, and the liquid was decanted. The remaining precipitate was placed in an 80ºC oven for 3 hours to dry.

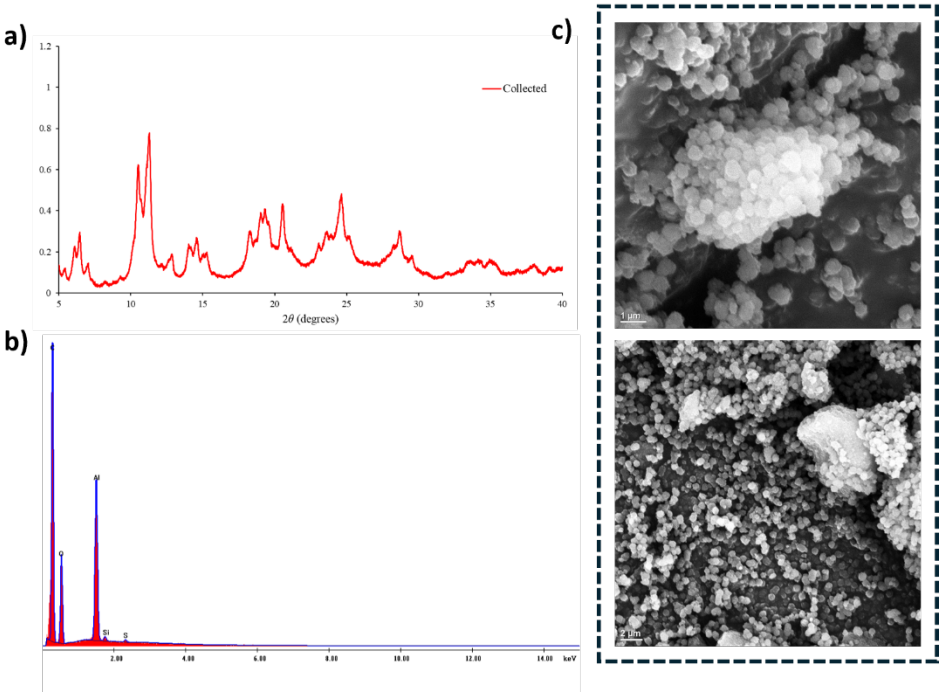

Figure S23. a) pxrd collected b) EDS c) SEM of MIL-100(Al)

MOF-177

MOF-177 was synthesized following a modified literature procedure.[4] 0.6005 g (2.018 mmol) of $Zn(NO_3)_2 \cdot 6H_2O$ and 0.1201 g (0.2739 mmol) of 1,3,5-Tris(4-carboxyphenyl)benzene were added

to a 100 mL flask followed by 40 mL DMF. The solution was sonicated for 5 minutes and then placed in an oven at 70 °C for 7 days.

The solution and precipitate were cooled to room temperature and then centrifuged for 5 minutes. The solution was decanted, and the precipitate was washed with 10 mL DMF and chloroform thrice. The resulting crystals were dried in the oven at 70 °C for 3 hours.

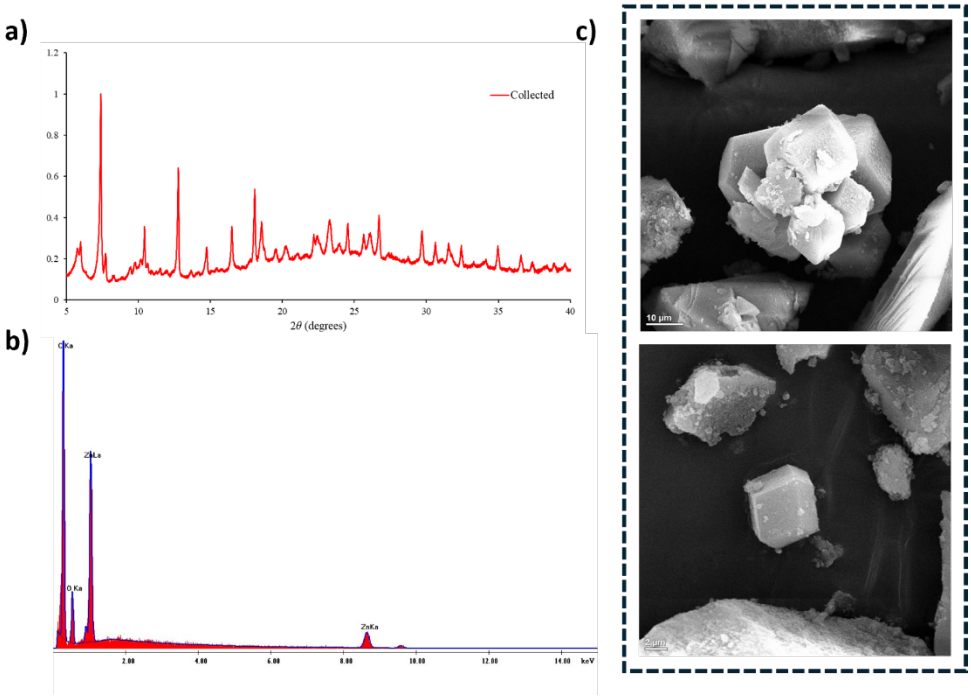

Figure S24. a) pxrd collected b) EDS c) SEM of MOF-177.

CuBDC

1.3205 g (5.694 mmol) of $Cu(NO_3)_2 \cdot 2.5H_2O$ and 0.3287 g (1.980 mmol) of terephthalic acid were combined in a 50 mL flask followed by 24 mL of DMF/ethanol (2:1) solution. The solution was sonicated for 5 minutes, then transferred to a 50 mL Teflon lined autoclave and heated at 120ºC for 16 hours.[5]

Once removed from heat the reaction vessel was allowed to cool to room temperature. The solution and precipitate were transferred into a centrifuge tube, centrifuged for 5 minutes, and the solution

was decanted. 10 mL of DMF was added to the centrifuge tube and the precipitate was shaken to redistribute. The mixture was then centrifuged for 5 minutes, and the solution decanted. This process was completed 3 times with DMF and 3 times with ethanol. The remaining precipitate was placed in an 80ºC oven for 3 hours to dry.

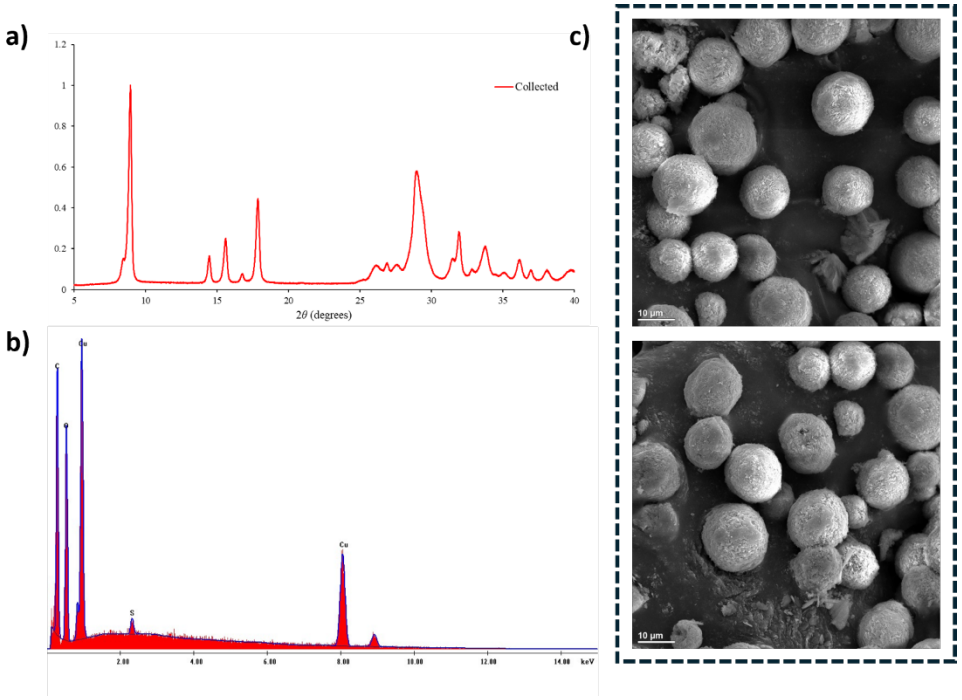

Figure S25. a) pxrd collected b) EDS c) SEM of CuBDC.

HKUST-1

0.3034 g (1.308 mmol) of $Cu(NO_3)_2 \cdot 2.5H_2O$ and 0.2119 g (1.009 mmol) of 1,3,5-benzenetricarboxylate were combined in a 50 mL flask, followed by 15 mL of water/ethanol (1:1) mixture. The solution was sonicated for 5 minutes, then transferred to a 50 mL Teflon lined autoclave and heated at 110ºC for 16 hours.[6]

Once removed from heat the reaction vessel was allowed to cool to room temperature. The solution and precipitate were transferred into a centrifuge tube, centrifuged for 5 minutes, and the solution was decanted. 10 mL of ethanol was added to the centrifuge tube and the precipitate was shaken

to redistribute. The mixture was then centrifuged for 5 minutes, and the solution decanted. This process was completed 3 times.

The remaining precipitate was transferred into an 80ºC oven for 3 hours to dry. The solid was then transferred to a 25 mL round bottom flask and heated to 150ºC overnight under vacuum.

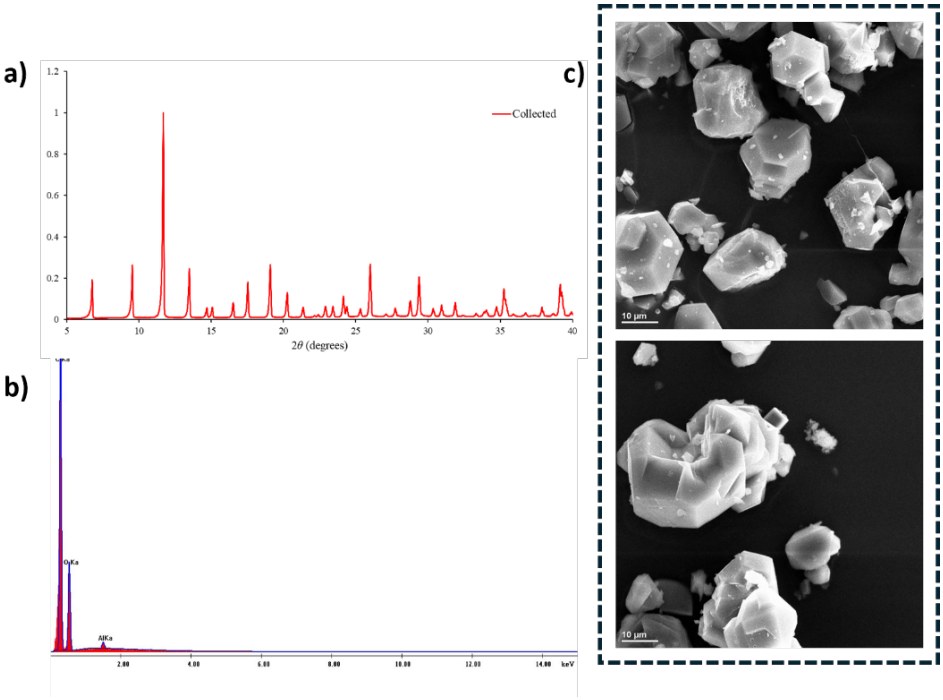

Figure S26. a) pxrd collected b) EDS c) SEM of HKUST-1

MIL-100(Fe)

0.9934 g (3.694 mmol) of $FeCl_3 \cdot 6H_2O$ and 0.4361 g (2.055 mmol) of 1,3,5-benzenetricarboxylate were added to a 50 mL flask, followed by 10 mL of water and 0.12 mL of concentrated HNO_3. The solution was sonicated for 5 minutes. The solution was transferred to a 50 mL Teflon lined autoclave and heated at 150ºC overnight.[7]

Once removed from heat the reaction vessel was allowed to cool to room temperature. The solution and precipitate were transferred into a centrifuge tube, centrifuged for 5 minutes, and the solution was decanted. 10 mL of water was added to the centrifuge tube and the precipitate was shaken to

redistribute. The mixture was then centrifuged for 5 minutes, and the solution decanted. This process was completed 3 times with water, and 3 with methanol.

The remaining precipitate was transferred to a 50 mL round bottom flask and 15 mL of DMF was added, followed by refluxing overnight. Once reflux was completed, the mixture was cooled to room temperature and transferred to a centrifuge tube, centrifuged for 5 minutes, and the solution was decanted. 10 mL of methanol was added, centrifuged for 5 minutes, and the liquid was decanted. The remaining precipitate was placed in an 80ºC oven for 3 hours to dry.

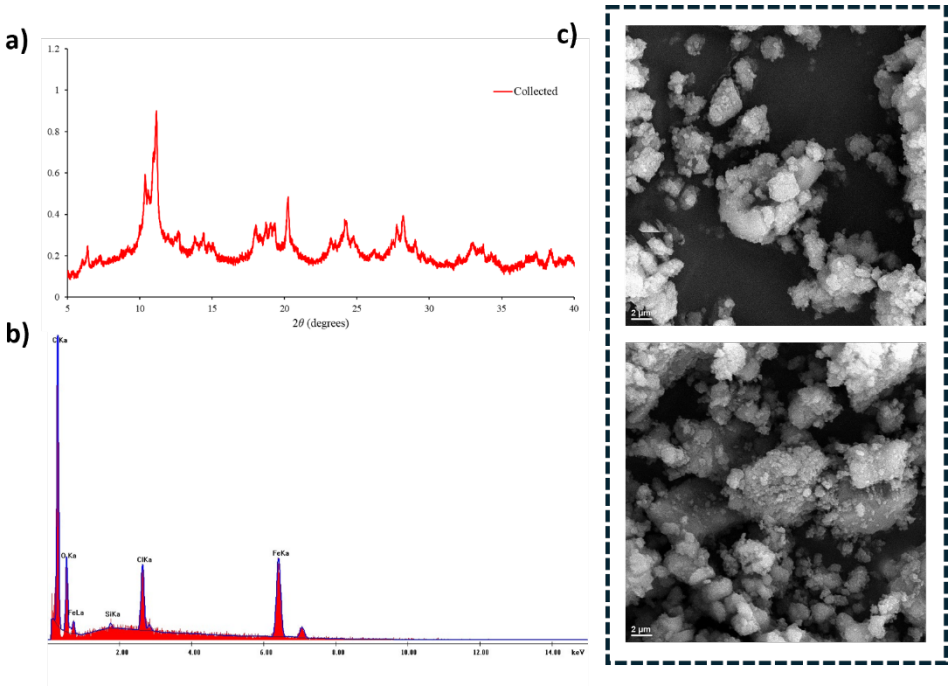

Figure S27. a) pxrd collected b) EDS c) SEM of MIL-100(Fe).

MIL-53(Fe)

In a modified literature prep, 0.5428 g (2.019 mmol) of $FeCl_3 \cdot 6H_2O$ 0.3366 g (2.027 mmol) of terephthalic acid were added to a 50 mL flask, followed by 11 mL of DMF. The solution was sonicated for 5 minutes. The solution was transferred to a 50 mL Teflon lined autoclave and heated at 150ºC overnight.[2]

Once removed from heat the reaction vessel was allowed to cool to room temperature. The solution and precipitate were transferred into a centrifuge tube, centrifuged for 5 minutes, and the solution was decanted. 10 mL of water was added to the centrifuge tube and the precipitate was shaken to redistribute. The mixture was then centrifuged for 5 minutes, and the solution decanted. This process was completed 3 times with water, and 3 with methanol.

The remaining precipitate was transferred to a 50 mL round bottom flask and 15 mL of DMF was added, followed by refluxing overnight. Once reflux was completed, the mixture was cooled to room temperature and transferred to a centrifuge tube, centrifuged for 5 minutes, and the solution was decanted. 10 mL of methanol was added, centrifuged for 5 minutes, and the liquid was decanted. The remaining precipitate was placed in an 80ºC oven for 3 hours to dry.

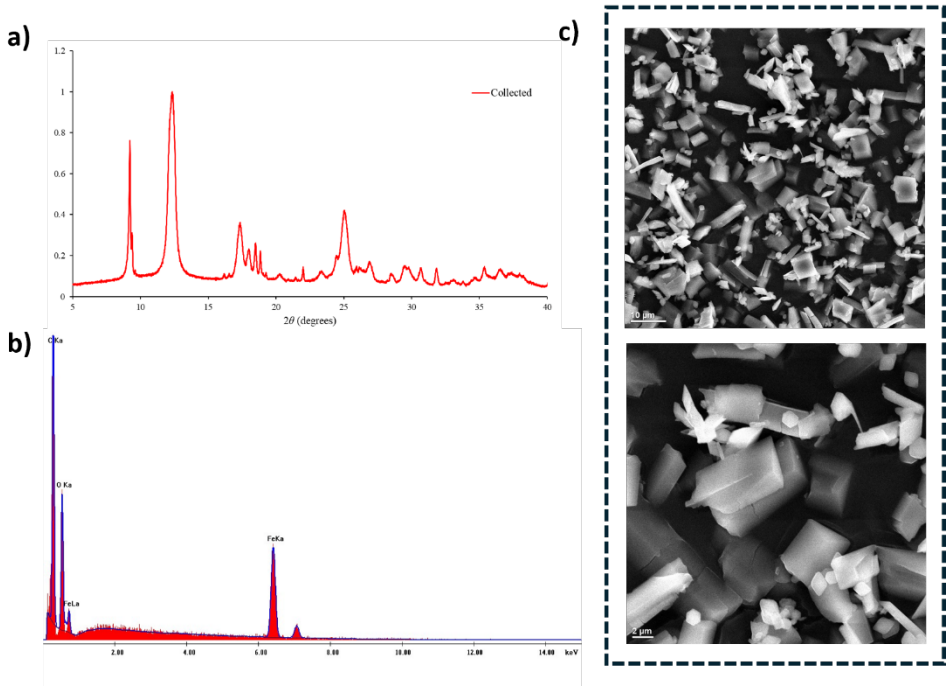

Figure S28. a) pxrd collected b) EDS c) SEM of MIL-53(Fe).

UiO-66

UiO-66 was synthesized using a modified literature procedure.[8] 0.1250 g (0.5363 mmol) $ZrCl_4$, 0.1250 g (0.7524 mmol) of terephthalic acid was added to a 8 dram vial followed by 15 mL DMF

and 1 mL HCL. The mixture was sonicated for 5 minutes. The reaction was then placed in an oven at 80ºC for 24 hours. The vial was cooled to room temperature, the solution and precipitate was transferred to a centrifuge tube. After centrifugation for 5 minutes the solution was decanted. The precipitate was washed by centrifugating for 5 minutes with 10 mL DMF and ethanol, each for three times. Following washes the precipitate was placed in an 80ºC oven for 3 hours to dry.

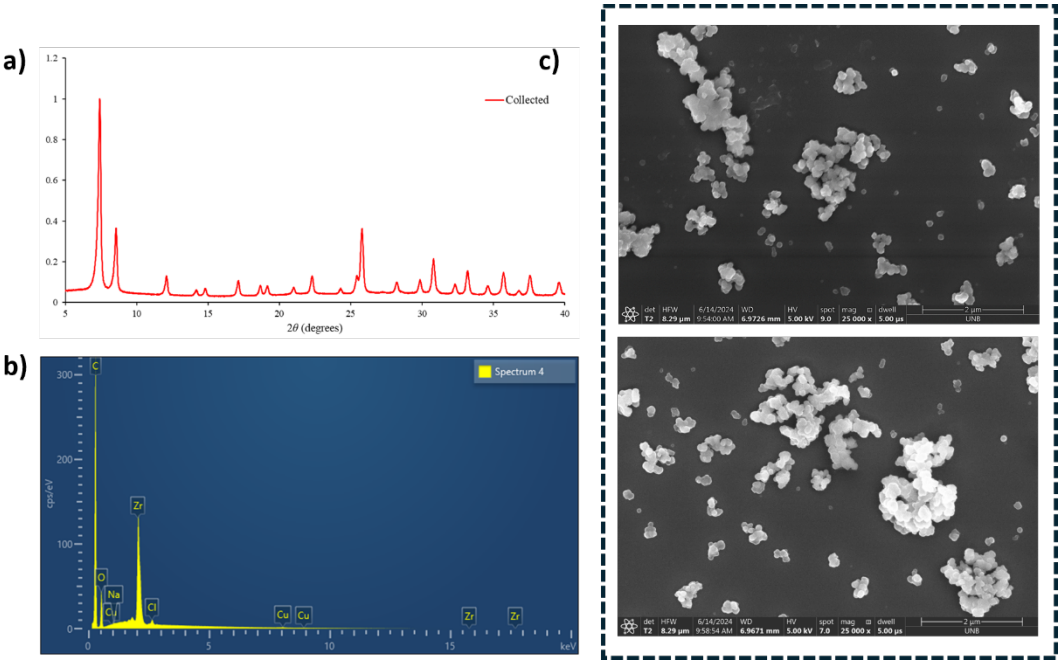

Figure S29. a) pxrd collected b) EDS c) SEM of UiO-66.

MOF-808

0.4320 g (1.350 mmol) of $ZrOCl_2 \cdot 8H_2O$ and 0.3063g (1.458 mmol) of 1,3,5-benzenetricarboxylate were added to a 100 mL flask followed by 25 mL DMF and 26 mL of formic acid. The mixture was sonicated for 5 minutes then transferred to a 100 mL flask, capped and placed in a 110ºC oven for 48 hours.

Once removed from heat the reaction vessel was allowed to cool to room temperature. The solution and precipitate were transferred into a centrifuge tube, centrifuged for 5 minutes, and the solution

was decanted. 15 mL of DMF was added and the resulting solution was transferred to a round bottom flask and refluxed overnight. Once reflux was completed, the mixture was cooled to room temperature and transferred to a centrifuge tube, centrifuged for 5 minutes, and the solution was decanted. 10 mL of methanol was added, centrifuged for 5 minutes, and the liquid was decanted. The remaining precipitate was placed in an 80ºC oven for 3 hours to dry.[9]

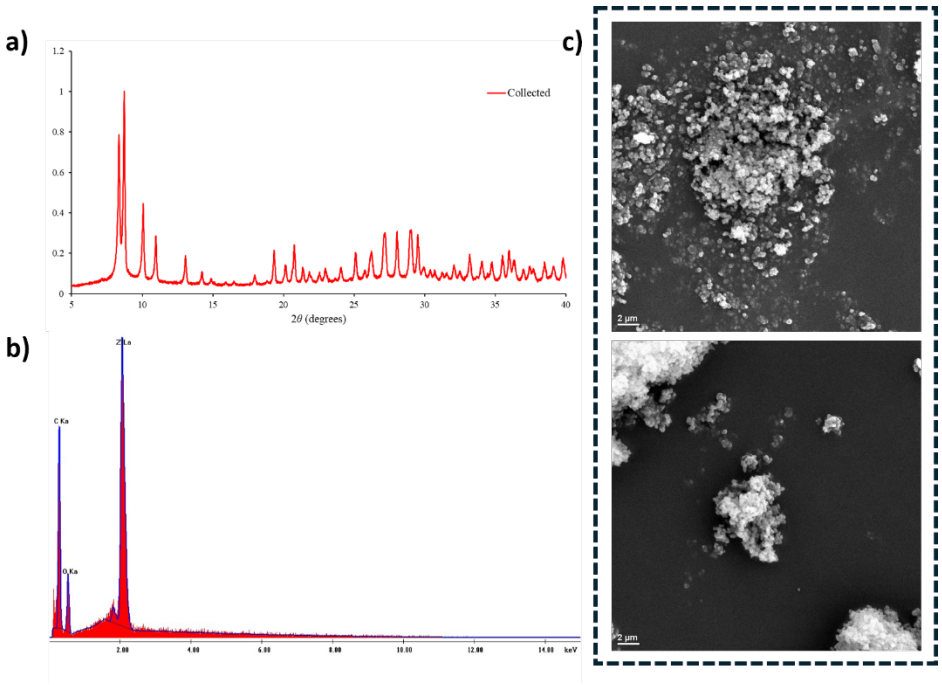

Figure S30. a) pxrd collected b) EDS c) SEM of MOF-808.

CaBDC

0.2362 g (1.001 mmol) of $Ca(NO_3)_2 \cdot 4H_2O$ and 0.0836 g (0.5035 mmol) of terephthalic acid were added to a 20 mL glass vial followed by 7 mL of DMF. The solution was sonicated for 5 minutes then placed in a 120ºC oven for 3 days. Once heating was completed the vial was cooled to room temperature.

The solution and resulting precipitate were transferred to a centrifuge tub and centrifuged for 5 minutes. The remaining solution was decanted, and 10 mL of DMF added to the centrifuge tube and the precipitate was shaken to redistribute. The mixture was then centrifuged for 5 minutes, and

the solution decanted. This process was completed 3 times with DMF, and 3 with methanol. The remaining precipitate was transferred into an 80ºC oven for 3 hours to dry.[10]

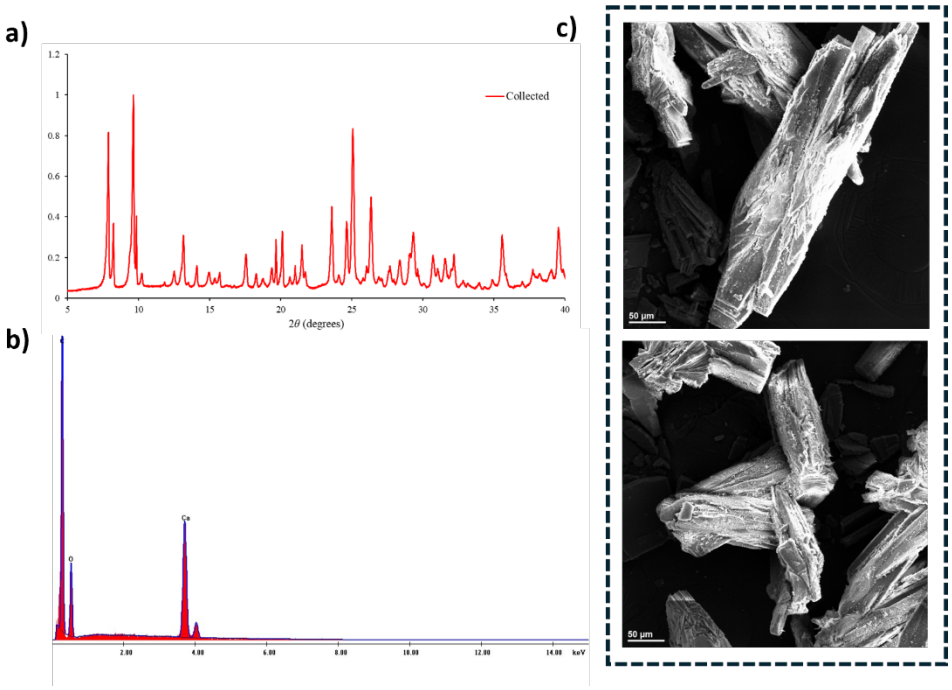

Figure S31. a) pxrd collected b) EDS c) SEM of CaBDC.

MgMOF-74

0.7120 g (2.438 mmol) of $Mg(NO_3)_2 \cdot 8H_2O$ and 0.1680 g (0.8484 mmol) of 2,5-dihydroxybenzoic acid were added to a 50 mL flask followed by 15 mL of DMF, 1 mL of ethanol and 1 mL of water. The mixture was sonicated for 5 minutes. After sonication the solution was transferred to a 50 mL Teflon lined autoclave and heated at 125ºC overnight. Once heating was completed the reaction vessel was cooled to room temperature.[11]

The mixture was transferred into a centrifuge tube, centrifuged for 5 minutes, and the solution was decanted. 10 mL of DMF was added to the centrifuge tube and the precipitate was shaken to redistribute. The mixture was then centrifuged for 5 minutes, and the solution decanted. This process was completed 3 times with DMF and 3 times with ethanol. The remaining precipitate was placed in an 80ºC oven for 3 hours to dry.

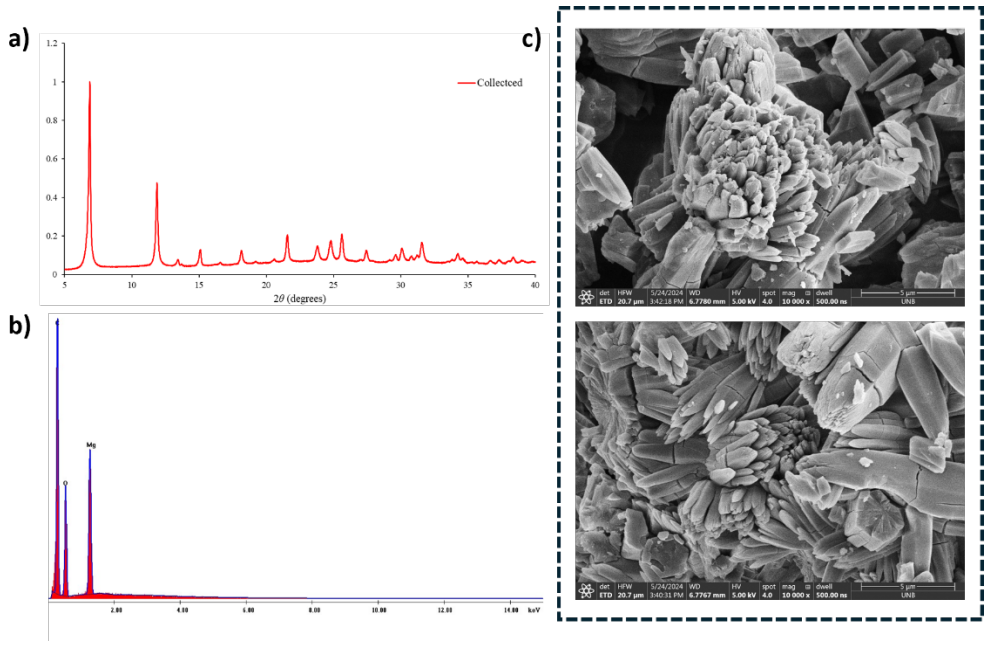

Figure S32. a) pxrd collected b) EDS c) SEM of MgMOF-74

ICP-OES Analysis

To monitor the decomposition of MOF-177 in solution a series of Inductively coupled plasma optical emission spectroscopy (ICP-OES) experiments were preformed. A small amount of MOF-177 (1.5-1.8 mg) was placed in the GUV solution used throughout the imaging experiments (0.7 mL). 0.1 mL of this solution was removed every 2 hours over the course of 12 hours. Caution was taken to ensure that no MOF was removed during removal of the supernatant. These aliquots were brought up to a total volume of 5 mL using deionized water and concentrated nitric acid (1 part in 20). The solutions were then analysed on a Varian Vista MPX CCD equipped with simultaneous ICP OES. As a control the experiments were repeated under same conditions with a zirconium MOF UiO-66 which is known to be a water stable framework.

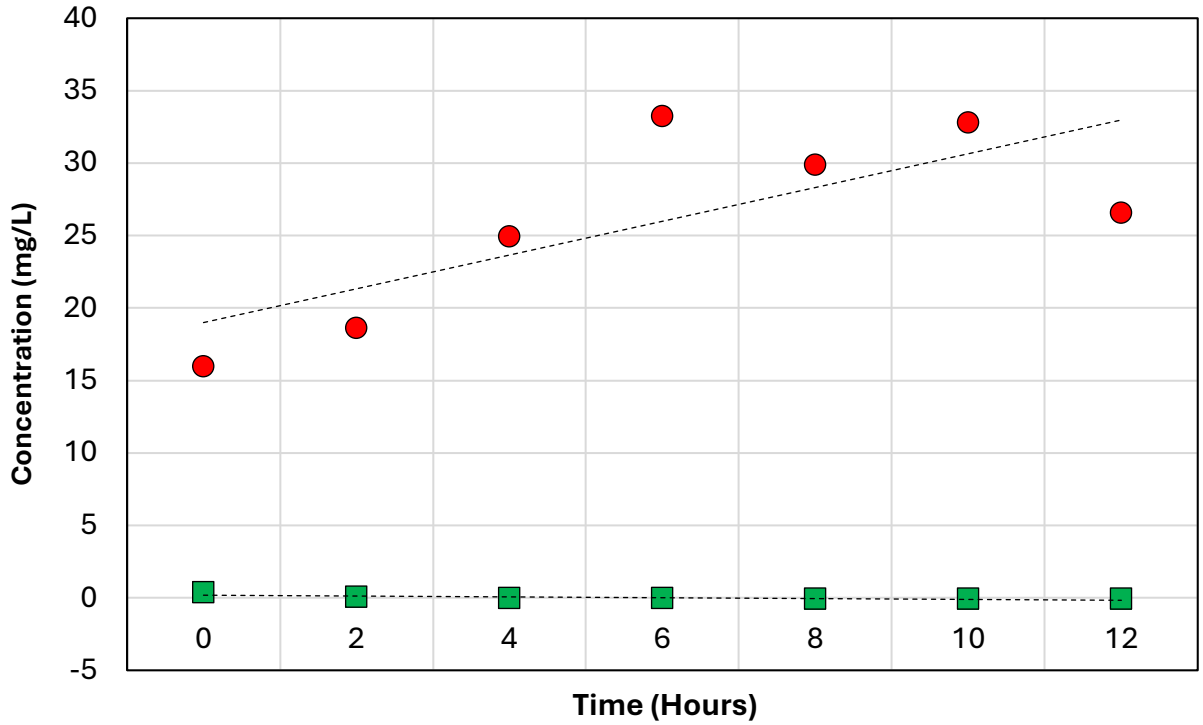

Figure S33. Concentration of zinc ions (red circles), and zirconium ions (green squares) from GUV solutions containing MOF-177 and UiO-66 respectively.

To further reflect on the dissolution of MOF-177, we electroformed POPC:POPG:Chol GUVs with MOF-177 and imaged MOF particles after 2 hours for 12 hours. The images showed obvious structural changes over time, indicative of gradual dissociation of MOF particles. Though complete dissolution was not observed, these morphological observations aligned with the results from ICP-OES analysis, which demonstrated an increase in zinc ion concentration in the supernatant over time.

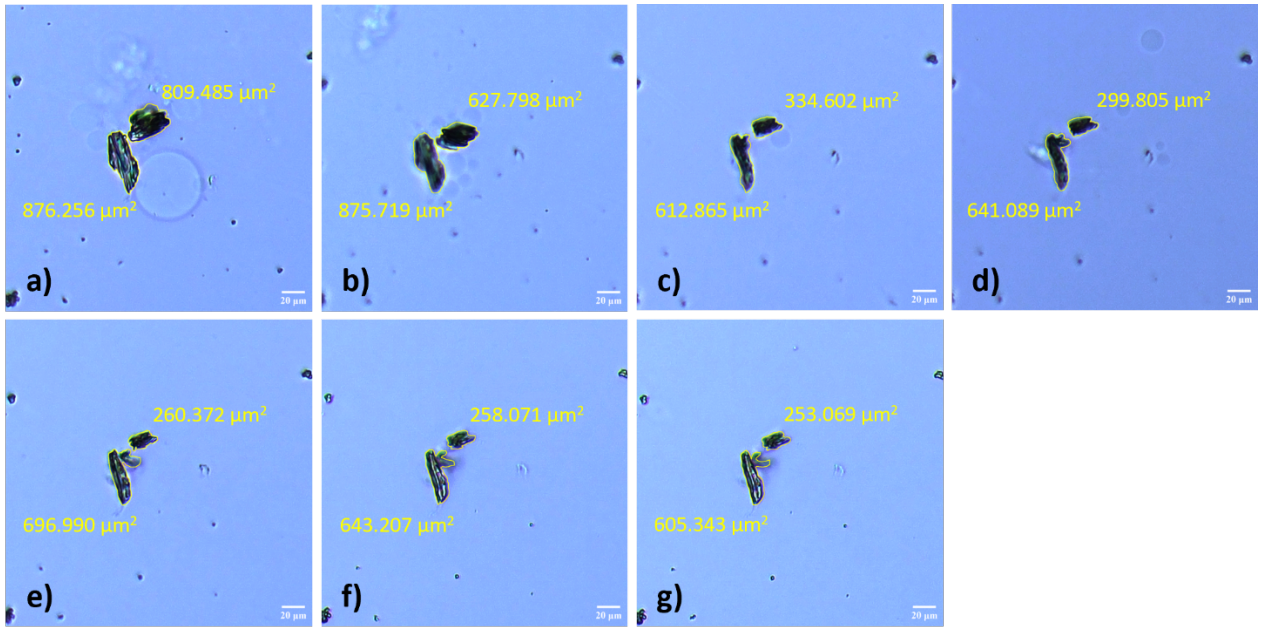

Figure S34. The brightfield images of GUV@MOF-177 imaged for 2 hour for 12 hours indicating changes in MOF structure.

Confocal Z-stacks

Confocal z-stacks were collected to examine the interaction of GUVs membrane immobilized by different MOFs. Imaging was performed using 0.1 mol% TopFluor® cholesterol labeled POPC:POPG:Chol (4:1:1) lipid mixture. The z-stacks demonstrated the spatial interaction between GUVs and MOFs (supplementary videos 11-14). The 3D reconstructions of these z-stacks are shown below.

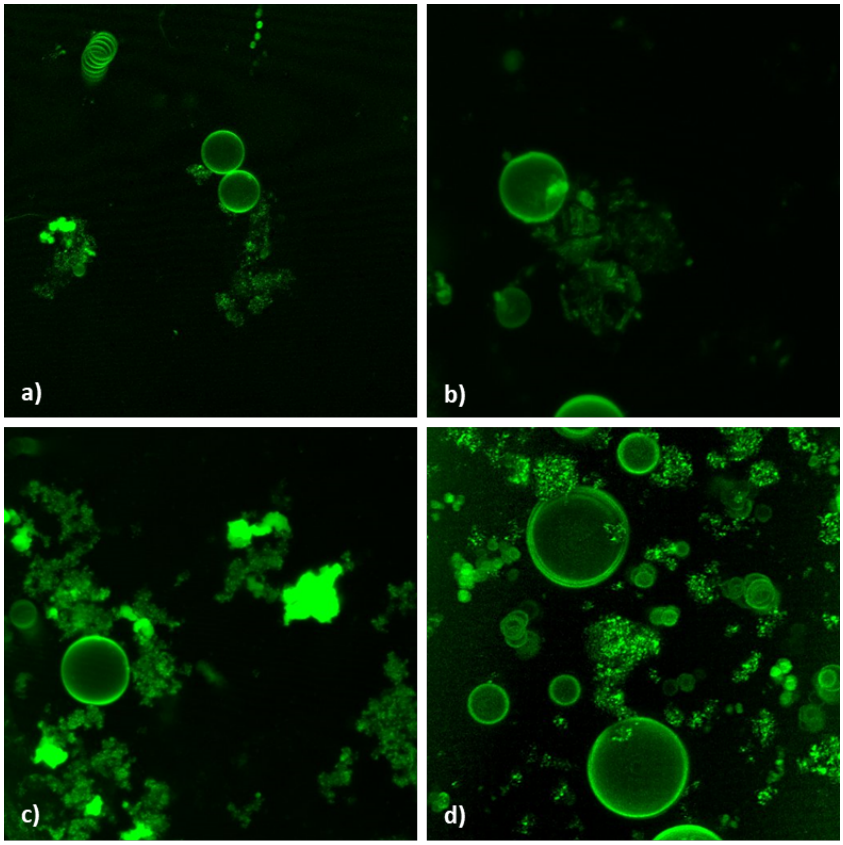

Figure S35. Confocal z-stack images a) GUV@MIL-100(Fe) b) HKUST-1 c) UiO-66 d) MOF-808

GUV@MOF SEM

The SEM images of POPC:POPG:Chol (4:1:1) GUV@MOF adduct were collected at the Digital Microscopy Facility at Mount Allison University using Hitachi SU3500 SEM operating at 10 kV, 10 mm working distance, and 0.1 nA beam current. The EDS spectra were recorded by Oxford Instruments AZtec/X-Max 20 EDS system. Spectra acquired from 0-10 keV into 1024 channels, 100 second dead-time corrected acquisitions from areas indicated by bounding-boxes in the screen shots. The GUV@MOF samples with HKUST-1 and MOF-808 were vapor fixed with 2% OsO4 for 4 hours and deposited onto 1 µm pore-size polycarbonate filters, mounted onto SEM support with double-side tape, rimmed with colloidal carbon and coated with ca. 10 nm gold in a Hummer 6.2 sputtering system.

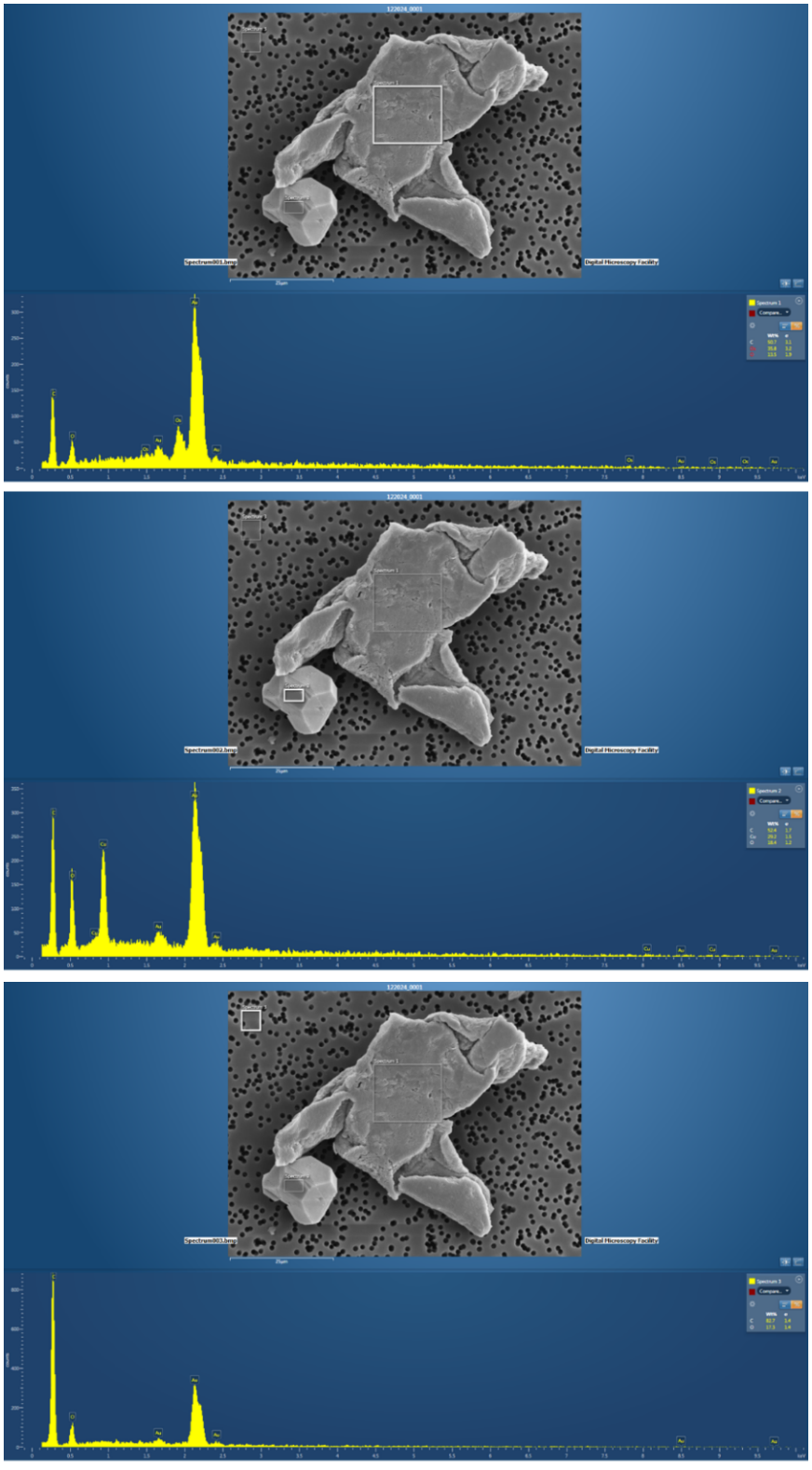

Figure S36. GUV@HKUST-1 EDS analysis

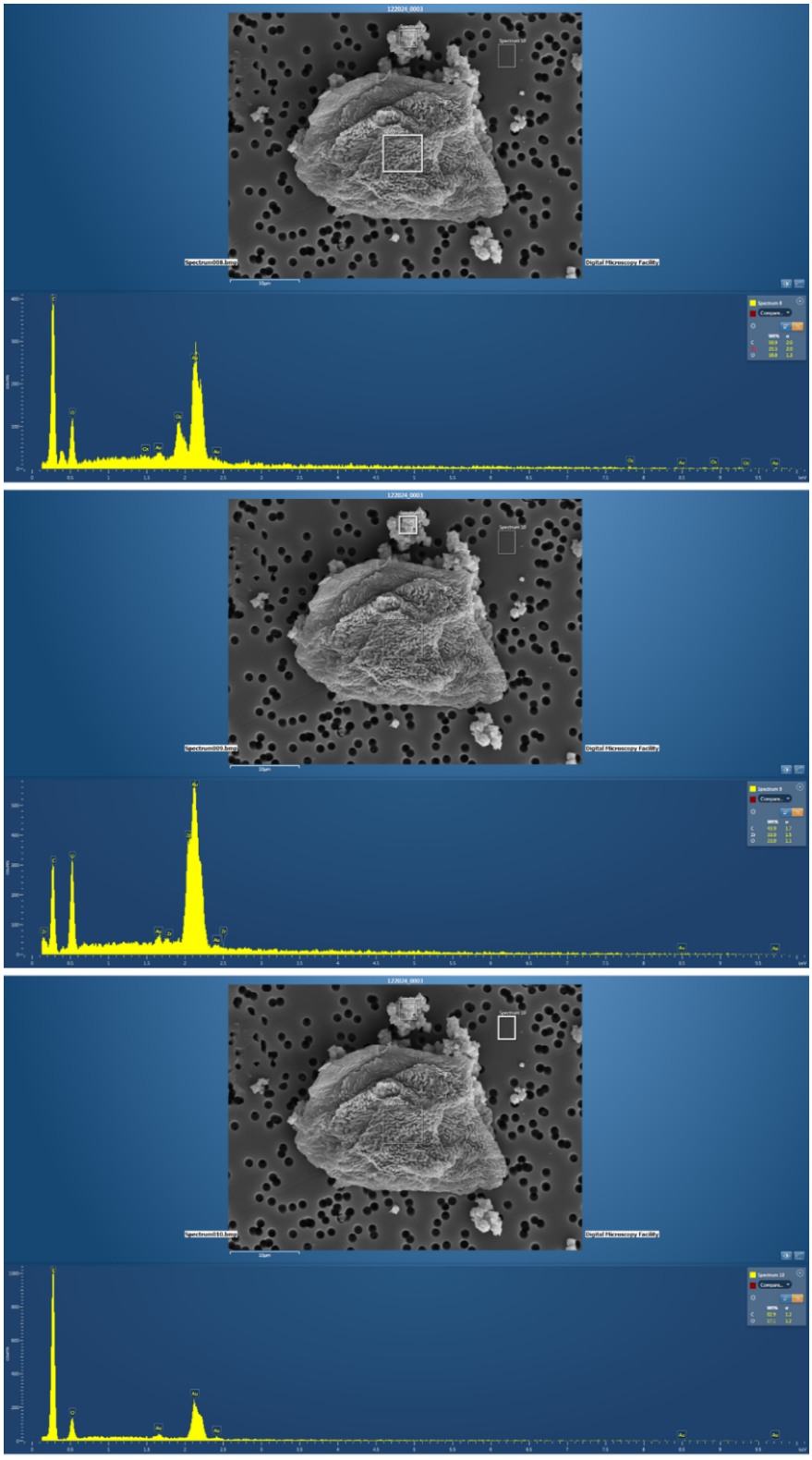

Figure S37. GUV@MOF-808 EDS analysis

Atomic Force Microscopy (AFM)

The AFM was utilized to investigate the interaction between GUV membrane and MOF surfaces, the results show high-resolution images of the physical interface confirming the immobilization of GUVs with MOF particles under dry-stage conditions. The droplets of the GUV@HKUST-1 MOF suspension were introduced to freshly cleaved mica sheets such that they covered the substrates, and after ~ 1 min, dried under a gentle stream of nitrogen and imaged immediately. The samples were scanned in intermittent-contact mode using a Park Systems XE-100 atomic force microscope equipped with a silicon cantilever (f0~300 kHz, Park Systems). Topographic and phase images were recorded simultaneously at a resolution of 256 x 256 pixels, at a scan rate of 1 Hz. Image processing (i.e. deglitching, cropping, and flattening) was performed using the Park Systems XEI software.AFM imaging of the GUV@HKUST-1 revealed bubble-like textures across the film when imaged immediately after drying with a gentle stream of N2 gas (Figure S38a, green arrow), with heights typically at least 400 nm. Synchronous phase imaging supports the assignment of vesicles to regions of large negative phase contrast, indicating a softer material in those regions compared to the mica substrate. Often two types of features were imaged as attached to the vesicle structures: nearly flat features with strong phase contrast to the vesicle (blue arrow), and very rough and tall features (red arrow). Further imaging of the solution containing glucose and HEPES in absence of the GUV@MOF (Figure S39b) shows features mainly with low topography (i.e. 2-10 nm in height). The corresponding phase images, however, display large phase contrast between these features and the substrate. Figure S39b supports the assignment of the bubble features to vesicles given their unique height and morphology compared to the glucose and HEPES control images. These images also enable the assignment of the large features in Figure S39a to that of the “vesicle-HKUST-1 MOF” given their amorphous and pronounced topography. Overall, figure S38 demonstrates that intimate contact is made between the MOF and the giant lamellar vesicle.

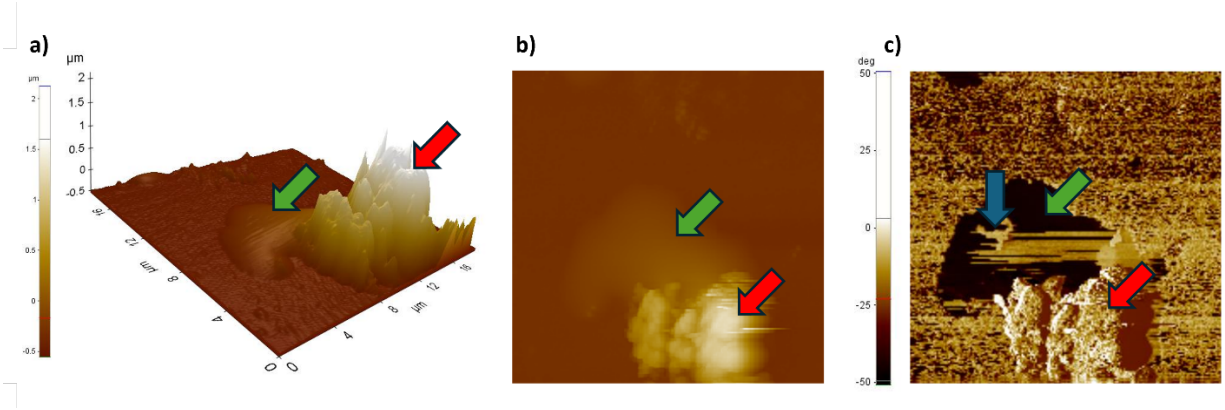

Figure S38. The renderings of AFM topography images a) 3D rendering b) 2D rendering and c) phase image corresponding to a GUV@HKUST-1 adduct adsorbed onto mica substrate. The arrows highlight features corresponding to a vesicle (green), MOF (red), and adsorbed crystallite, most likely corresponding to glucose.

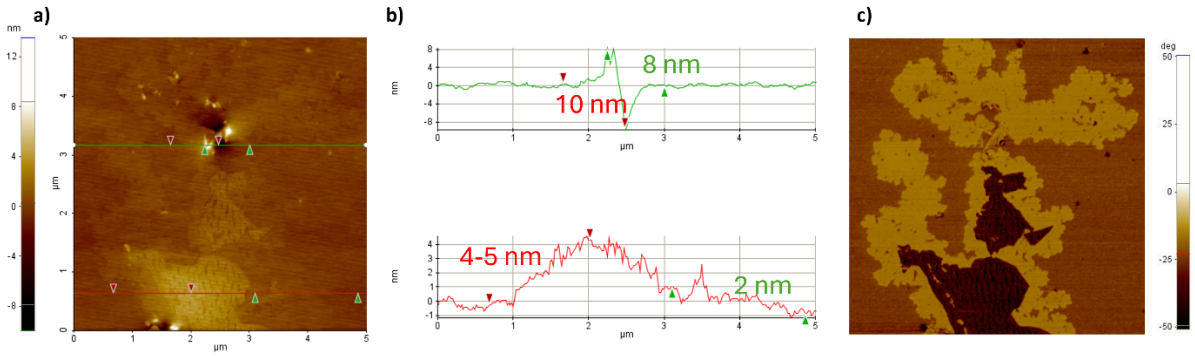

Figure S39. The AFM a) topography and c) phase images depicting the morphologies associated with glucose and HEPES buffer used for GUV suspension, upon drying on the mica substrate. The line scans from a) and corresponds to topographic heights, are shown in b) image.

Supplementary Information for

Investigating Metal-Organic Frameworks Anchors for Giant Unilamellar Vesicle Immobilization

Supplementary VideosTime-lapse videos of POPC:POPG:Chol GUVs anchored by MOF particles were recorded for 5 minutes. We have provided a 30-second excerpt to demonstrate the immobilization.

Supplementary Video SV1 - GUV immobilization by MIL-53(Al)

Supplementary Video SV2 - GUV immobilization by MIL-100(Al)

Supplementary Video SV3 - GUV immobilization by MIL-53(Fe)

Supplementary Video SV4 - GUV immobilization by MIL-100(Fe)

Supplementary Video SV5 - GUV immobilization by CuBDC

Supplementary Video SV6 - GUV immobilization by HKUST-1

Supplementary Video SV7 - GUV immobilization by UiO-66

Supplementary Video SV8 - GUV immobilization by MOF-808

Supplementary Video SV9 - GUV immobilization by MOF-177

Supplementary Video SV10 - GUV immobilization by CaBDC

Supplementary Video SV11 - GUV@MIL-100(Fe) confocal z-stack

Supplementary Video SV12 - GUV@HKUST-1 confocal z-stack

Supplementary Video SV13 - GUV@UiO-66 confocal z-stack

Supplementary Video SV14 - GUV@MOF-808 confocal z-stack

References

(1) Jennings, C. S.; Rossman, J. S.; Hourihan, B. A.; Marshall, R. J.; Forgan, R. S.; Blight, B. A. Immobilising Giant Unilamellar Vesicles with Zirconium Metal–Organic Framework Anchors. *Soft Matter* **2021**, *17* (8), 2024–2027.

(2) Ashling, C. W.; Johnstone, D. N.; Widmer, R. N.; Hou, J.; Collins, S. M.; Sapnik, A. F.; Bumstead, A. M.; Midgley, P. A.; Chater, P. A.; Keen, D. A.; Bennett, T. D. Synthesis and Properties of a Compositional Series of MIL-53(Al) Metal–Organic Framework Crystal-Glass Composites. *J. Am. Chem. Soc.* **2019**, *141* (39), 15641–15648. https://doi.org/10.1021/jacs.9b07557.

(3) Yang, J.; Wang, J.; Deng, S.; Li, J. Improved Synthesis of Trigone Trimer Cluster Metal Organic Framework MIL-100Al by a Later Entry of Methyl Groups. *Chem. Commun.* **2015**, *52* (4), 725–728. https://doi.org/10.1039/C5CC08450D.

(4) Saha, D.; Deng, S. Structural Stability of Metal Organic Framework MOF-177. *J. Phys. Chem. Lett.* **2010**, *1* (1), 73–78. https://doi.org/10.1021/jz900028u.

(5) Bagheri, A. R.; Ghaedi, M. Application of Cu-Based Metal-Organic Framework (Cu-BDC) as a Sorbent for Dispersive Solid-Phase Extraction of Gallic Acid from Orange Juice Samples Using HPLC-UV Method. *Arabian Journal of Chemistry* **2020**, *13* (5), 5218–5228. https://doi.org/10.1016/j.arabjc.2020.02.020.

(6) Chui, S. S.-Y.; Lo, S. M.-F.; Charmant, J. P. H.; Orpen, A. G.; Williams, I. D. A Chemically Functionalizable Nanoporous Material $[Cu_3(TMA)_2(H_2O)_3]_n$. *Science* **1999**, *283* (5405), 1148–1150. https://doi.org/10.1126/science.283.5405.1148.

(7) Simon, M. A.; Anggraeni, E.; Soetaredjo, F. E.; Santoso, S. P.; Irawaty, W.; Thanh, T. C.; Hartono, S. B.; Yuliana, M.; Ismadji, S. Hydrothermal Synthesize of HF-Free MIL-100(Fe) for Isoniazid-Drug Delivery. *Sci Rep* **2019**, *9* (1), 16907. https://doi.org/10.1038/s41598-019-53436-3.

(8) Katz, M. J.; Brown, Z. J.; Colón, Y. J.; Siu, P. W.; Scheidt, K. A.; Snurr, R. Q.; Hupp, J. T.; Farha, O. K. A Facile Synthesis of UiO-66, UiO-67 and Their Derivatives. *Chemical Communications* **2013**, *49* (82), 9449–9451.

(9) Dai, S.; Simms, C.; Dovgaliuk, I.; Patriarche, G.; Tissot, A.; Parac-Vogt, T. N.; Serre, C. Monodispersed MOF-808 Nanocrystals Synthesized via a Scalable Room-Temperature Approach for Efficient Heterogeneous Peptide Bond Hydrolysis. *Chem. Mater.* **2021**, *33* (17), 7057–7066. https://doi.org/10.1021/acs.chemmater.1c02174.

(10) Xiao, F.; Gao, W.; Wang, H.; Wang, Q.; Bao, S.; Xu, M. A New Calcium Metal Organic Frameworks (Ca-MOF) for Sodium Ion Batteries. *Materials Letters* **2021**, *286*, 129264. https://doi.org/10.1016/j.matlet.2020.129264.

(11) Yang, D.-A.; Cho, H.-Y.; Kim, J.; Yang, S.-T.; Ahn, W.-S. CO2 Capture and Conversion Using Mg-MOF-74 Prepared by a Sonochemical Method. *Energy Environ. Sci.* **2012**, *5* (4), 6465–6473. https://doi.org/10.1039/C1EE02234B.