# Supporting Information

## Size-Selective Biocatalysis of Myoglobin Immobilized into a Mesoporous Metal-Organic Framework with Hierarchical Pore Sizes

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**Materials Syntheses**

Crystalline samples of the mesoporous MOF, Tb-mesoMOF and the mesoporous silica material, SBA-15 were prepared according to the procedures reported in ref. S1 and ref. S2 respectively.

**Myoglobin uptake experiments**

Typically 2.5 mg/mL of met-myoglobin from equine skeletal muscle (Sigma) was prepared in 2.0 mL 4-(2-hydroxyethyl)-1-piperazineethanesulfonic acid (HEPES) buffer added into 5.0 mg of Tb-mesoMOF and incubated at 37°C for 94 hours. The protein concentration in the supernatant was determined at different time points using the BCA method of protein determination (Bicinchoninic Acid protein assay using bovine serum albumin as the standard) in order to establish the uptake of Mb.[S3] For the SBA-15 (5.0 mg) the same procedure and conditions were followed. For each preparation, a sample of the Mb solution was also incubated under the same conditions and was used to determine the reference concentrations.

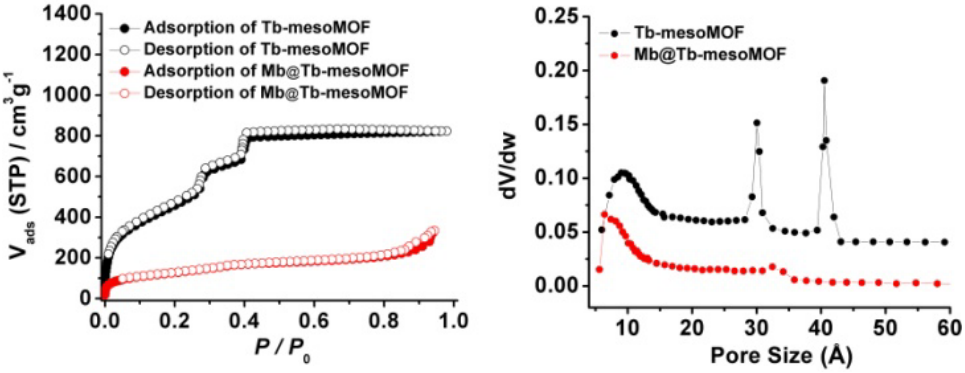
**Examination of the catalytic activities**

The initial rates for $ABTS^{+\bullet}$ formation were monitored using 0.5 mM 2,2′ azinodi(3-ethylbenzthiazoline)-6-sulfonate (ABTS) in presence of 10 mM $H_2O_2$ in HEPES buffer by various catalysts discussed in this work (~5mg used for each solid catalyst and 0.5μM in HEPES buffer for free Mb) on a Varian CARY50 spectrophotometer. The oxidation of the substrate to the corresponding oxidized product $ABTS^{+\bullet}$ was directly monitored at 660 nm ($\varepsilon$ = 12 $M^{-1}cm^{-1}$)[S4] by taking the absorption spectra of the supernatant solution at various time points over the reaction.

The initial rates of trihydroxybenzene (THB) oxidation were monitored using 0.5 mM THB in presence of 10 mM $H_2O_2$ in HEPES buffer by various catalysts discussed in this work (~4mg and ~5mg used for the solid catalysts of Mb@Tb-mesoMOF and Mb@SBA-15 respectively, and 0.5 μM in HEPES buffer for free Mb) also on a Varian CARY50 spectrophotometer. The oxidation of the substrate to the corresponding oxidized product *purpurogallin* dimer were directly monitored at 320 nm ($\varepsilon$ = 31,250 $M^{-1}cm^{-1}$)[S5] by taking the absorption spectra of the supernatant solution at various time points over the reaction.

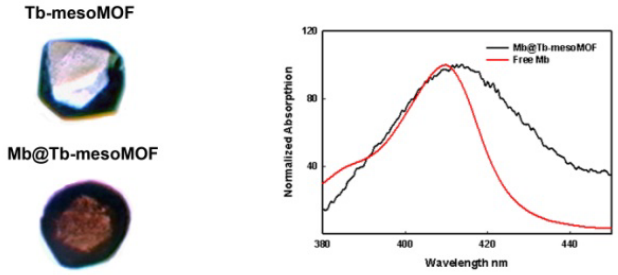
**$N_2$ sorption measurements**

$N_2$ sorption isotherms of Tb-mesoMOF and Mb@Tb-mesMOF were collected using the surface area analyzer ASAP-2020. Before the measurements, the freshly prepared samples were thoroughly solvent-exchanged with methanol, and activated under dynamic vacuum first at room temperature overnight and then at 120 $^oC$ for two hours. The surface area of Tb-mesoMOF is comparable with that reported in ref. 10 for the sample activated at 80 $^oC$.



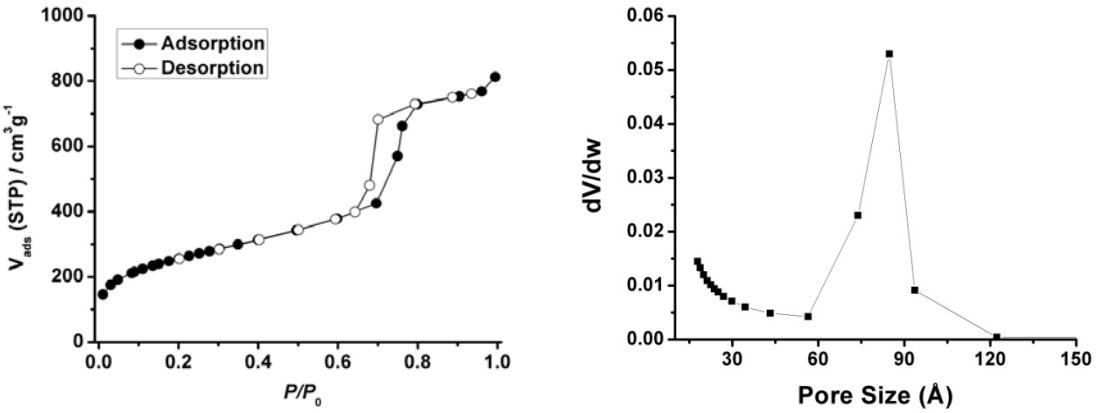

(a) (b)

**Figure S1.** (a) $N_2$ sorption isotherms and (b) Pore size distributions of Tb-mesoMOF and Mb@Tb-mesoMOF.




(a) (b)

**Figure S2.** (a) Optical images of Tb-mesoMOF and Mb@Tb-mesoMOF; (b) UV-Vis absorption spectra for Mb@Tb-mesoMOF (black) and free Mb in buffer solution (red).




(a) (b)

**Figure S3.** (a) $N_2$ sorption isotherms of SBA-15 at 77 K, which reveals a BET surface area of ~900 $m^2/g$; (b) pore size distribution of SBA-15.

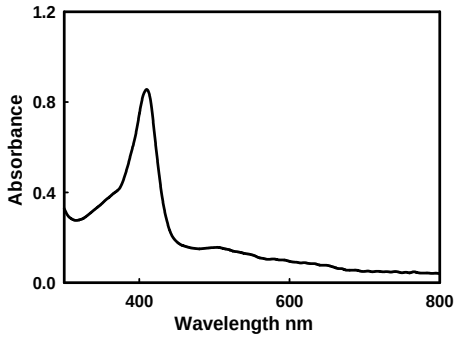



**Figure S4.** UV-Vis spectrum of the superanant for Mb@SBA-15 assay, revealing a leaching of free Mb in HEPES buffer.

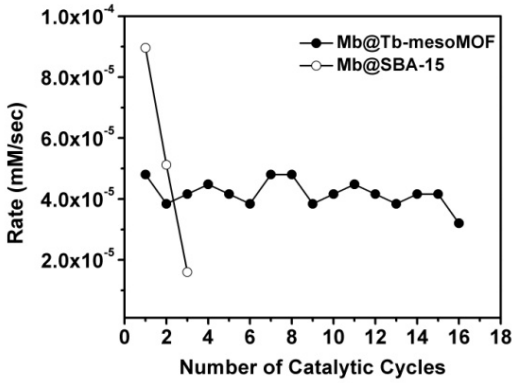



**Figure S5.** Reaction rates of THB oxidation for Mb@Tb-mesoMOFand Mb@SBA-15 at different cycles.

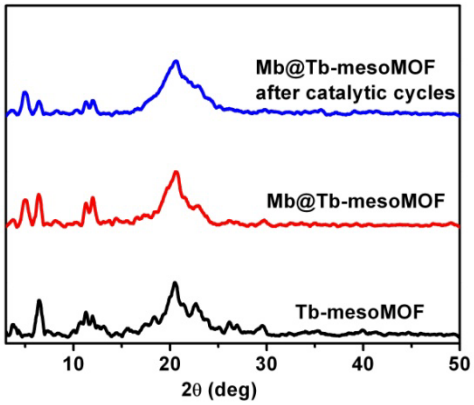



**Figure S6.** PXRD patterns for Tb-mesoMOF, Mb@Tb-mesoMOF, and Mb@Tb-mesoMOF after catalytic cycles.

**Table S1.** Summary of catalysis results of oxidizing ABTS and THB in the presence of 10 mM $H_2O_2$ in HEPES buffer.

| | Tb-mesoMOF |
|---|---|
| Initial rate for ABTS+• formation (mM/sec)[a] | $9.38\times 10^{-7}$ |
| Initial rate for THB oxidation[b] (mM/sec) | $2.24\times 10^{-6}$ |
| Average Rate for THB oxidation [c] (mM/sec) | $1.07\times 10^{-6}$ |

[a] Initial rate calculated in the first four minutes; [b] initial rate calculated in the first five minutes; [c] average rate over one hour.

**Table S2.** Summary of reaction rates at different catalytic cycles.

| **Cycle** | Mb@TbmesoMOF (mM/sec) | Mb@SBA-15 (mM/sec) |
|---|---|---|
| 1 | $4.80\times 10^{-5}$ | $8.96\times 10^{-5}$ |
| 2 | $3.84\times 10^{-5}$ | $5.12\times 10^{-5}$ |
| 3 | $4.16\times 10^{-5}$ | $1.60\times 10^{-5}$ |
| 4 | $4.48\times 10^{-5}$ | |
| 5 | $4.16\times 10^{-5}$ | |
| 6 | $3.84\times 10^{-5}$ | |
| 7 | $4.84\times 10^{-5}$ | |
| 8 | $4.83\times 10^{-5}$ | |
| 9 | $3.84\times 10^{-5}$ | |
| 10 | $4.16\times 10^{-5}$ | |
| 11 | $4.48\times 10^{-5}$ | |
| 12 | $4.16\times 10^{-5}$ | |
| 13 | $3.84\times 10^{-5}$ | |
| 14 | $4.16\times 10^{-5}$ | |
| 15 | $4.16\times 10^{-5}$ | |
| 16 | $3.20\times 10^{-5}$ | |

## References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