

## *Supporting Information*

# Iminoguanidines as Allosteric Inhibitors of the Iron-Regulated Heme Oxygenase (HemO) of *Pseudomonas aeruginosa*

*Geoffrey A. Heinzl, Weiliang Huang, Wenbo Yu, Bennett J. Giardina, Yue Zhou,*

*Alexander D. MacKerell, Jr., Angela Wilks, Fengtian Xue\**

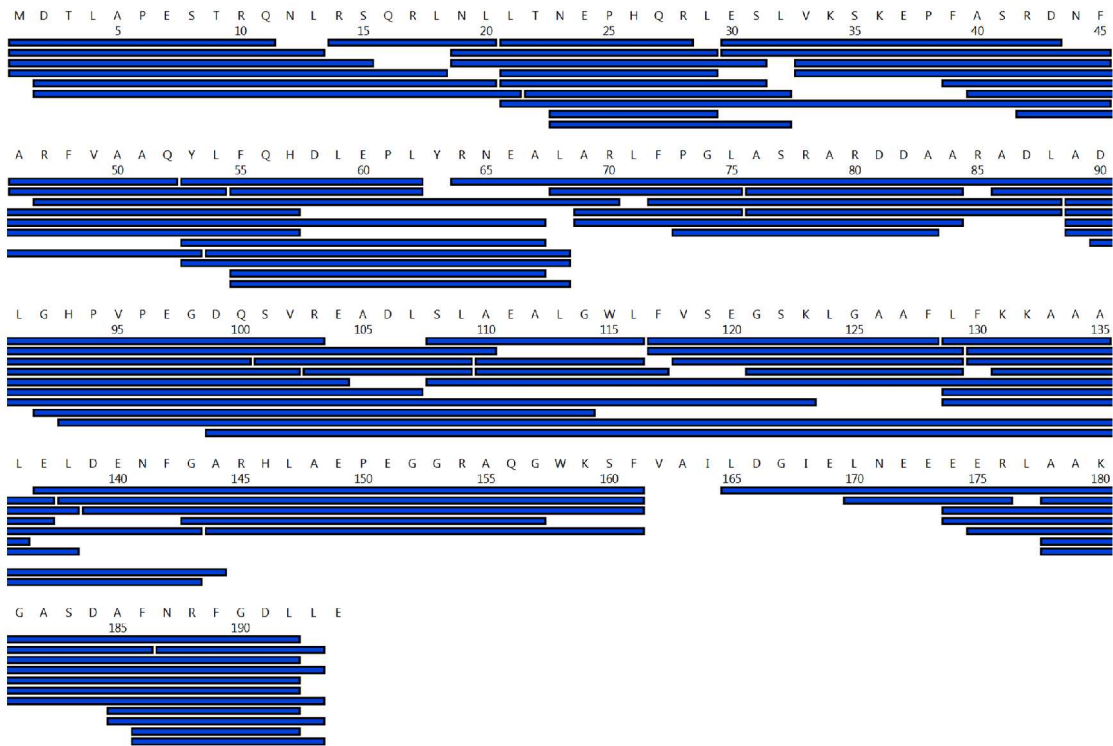
University of Maryland Computer-Aided Drug Design Center

Department of Pharmaceutical Sciences, School of Pharmacy, University of Maryland,

Baltimore, Baltimore, Maryland 21201, United States

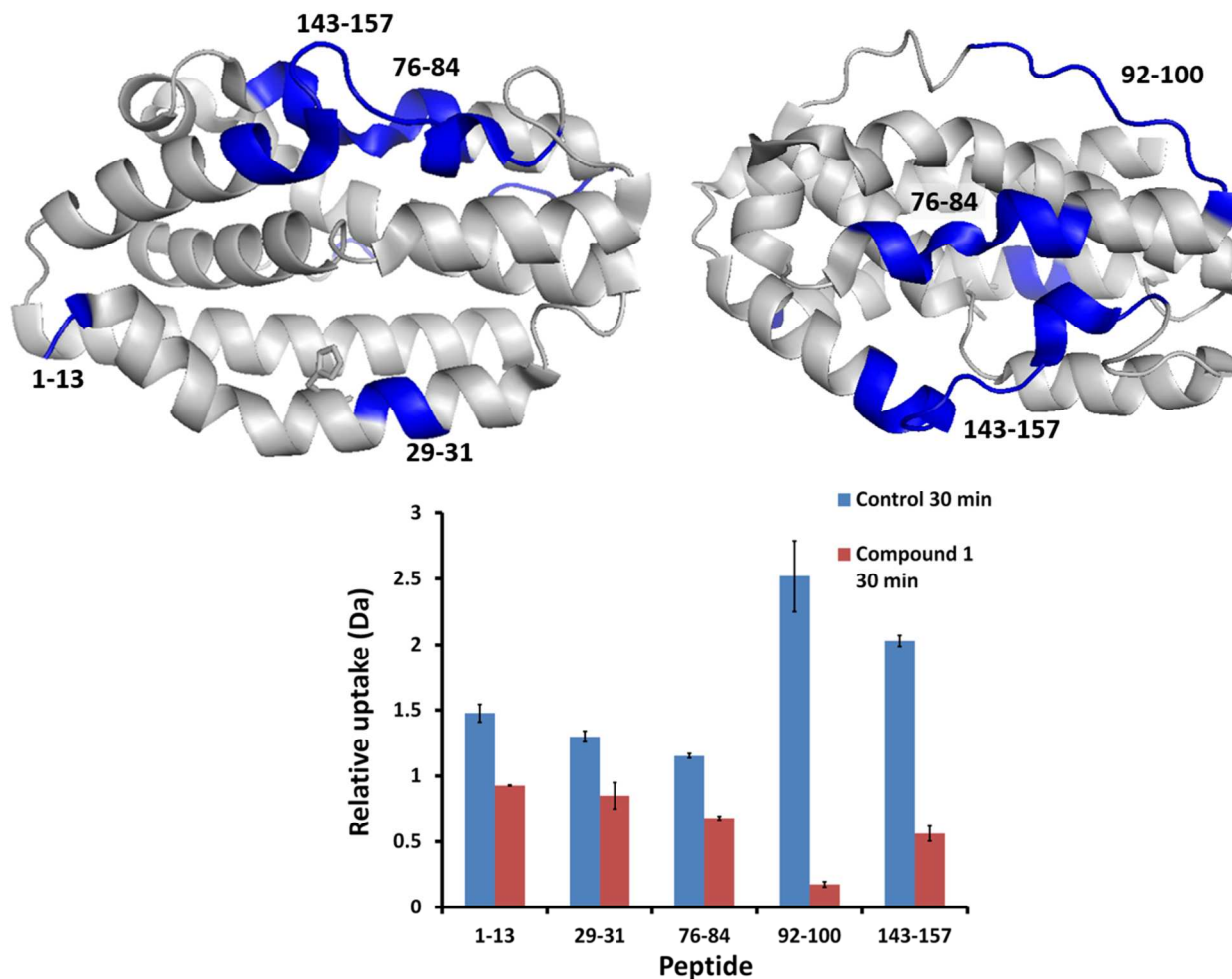
### **Table of Contents**

Figure S1. HXMS peptide coverage map for HemO following pepsin digestion.	S-2
Figure S2. Color coded protein structure of apo-HemO representing deuterium uptake change upon binding of compound 1.	S-3
Figure S3. Fluorescence quenching as a function of HemO inhibition.	S-4

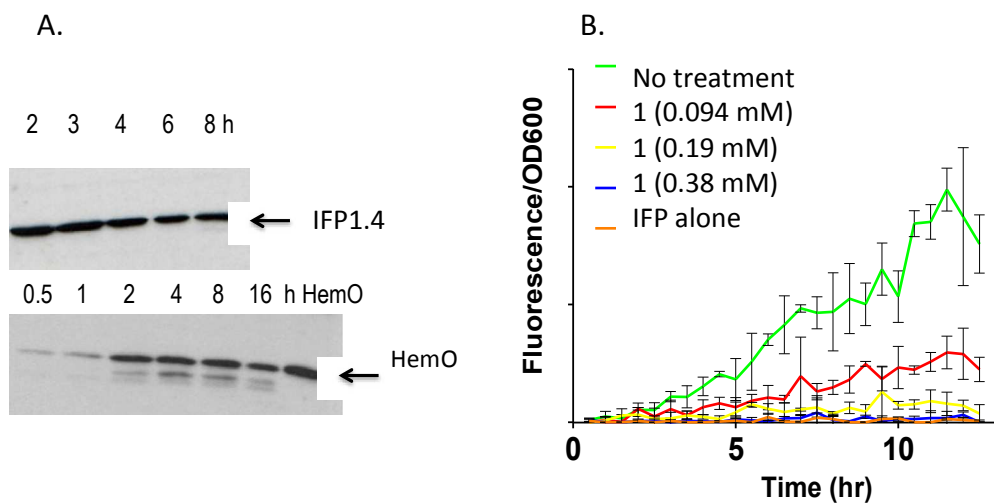


Total: 97.9% Coverage, 6.71 Redundancy

**Figure S1. HXMS peptide coverage map for HemO following pepsin digestion.** Protease treatment following hydrogen exchange and peptide analysis as described in “Experimental Section”.



**Figure S2. Color coded protein structure of apo-HemO representing deuterium uptake change upon binding of compound 1.** Deuterium incorporation was mapped onto the HemO crystal structure (PDB 1SK7) as viewed from front (heme-binding pocket, upper left) and from above (upper right). Peptide regions of apo-HemO which were more protected from deuterium exchange in the presence of **1** are colored in blue, whereas regions which became more prone to deuterium exchange are colored in red. Relative deuterium uptake of peptides showed differences between apo-HemO and apo-HemO in the presence of **1** at 30 min (bottom). All five peptides were significantly more protected from deuterium exchange upon binding of **1** (Student's *t*-test,  $p < 0.05$ ,  $n=2$ ). Error bar, standard deviation.



**Figure S3. Fluorescence quenching as a function of HemO inhibition.** A. Western blot over the time course of the experiment following induction of HemO with 0.02% arabinose. All experiments were performed in triplicate and averaged. IFP1.4 is induced at time 0 with 1 mM IPTG. B. Fluorescence as a function of growth corrected for OD<sub>600</sub> and concentration dependent quenching with compound **1**. Experiments were performed as described in “Experimental Section”.