Supporting Information

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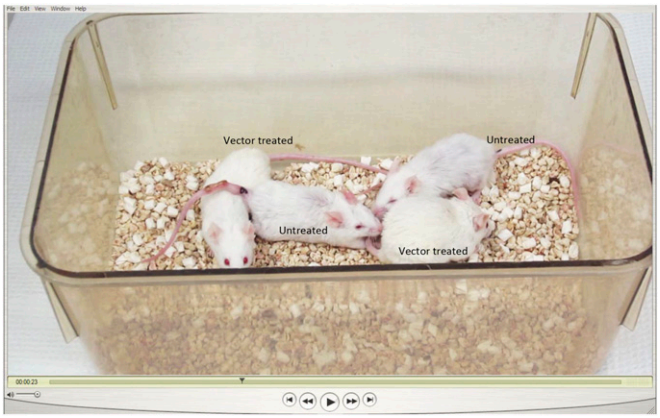
SI Materials and Methods

The initial model of GSS^{octanoyl}FL·hBChE was generated by manually docking ghrelin residues 1–5, with *N*-methyl substituted at the C terminus in an extended backbone conformation, into the hBChE active site taken from the crystal structure of full-length recombinant hBChE (Protein Data Bank ID: 3O9M). This docking placed ghrelin's ammonium group atop Trp82^{BChE} and Phe4^{Ghrelin} close to Phe329^{BChE} and Tyr332^{BChE}. Force-field parameters for Ser with *n*-octanoylated hydroxyl ($S^{octanoyl}$) were generated by a published procedure (1, 2).

The energy-minimized complex was neutralized with 11 chlorides, solvated with 13,050 TIP3P water molecules (3) containing 36 NaCl molecules, and energy-minimized for 100 cycles as above followed by 100 cycles of conjugate-gradient minimization to remove close van der Waals contacts using FF12MC. The resulting system was then heated from 0 to 300 K at a rate of 10 K/ps under constant temperature and constant volume and finally simulated with 100 2-ns low-mass molecular dynamics simulations (4), each of which used a unique seed number for initial velocities, using the PMEMD module of the AMBER 11 program. All simulations used (*i*) a dielectric constant of 1.0; (*ii*) the Berendsen coupling algorithm; (*iii*) a periodic boundary condition at a constant temperature of 300 K and a constant pressure of 1 atm with isotropic molecule-based scaling; (*iv*) the Particle Mesh Ewald method to calculate long-range electrostatic interactions; (*v*) a time step of 1.0 fs; (*vi*) SHAKE bond-length constraints applied to all bonds involving the H atom; (*vii*) a protocol to save the image closest to the middle of the "primary box" to the restart and trajectory files; (*viii*) formatted restart file; (*ix*) the FF12MC forcefield; and (*x*) default values of all other inputs of the PMEMD module. A total of 1,000 trajectories saved at 100-ps intervals during the last 1-ns period of 100 simulations were subjected to a cluster analysis using the PTRAJ module of AmberTools 13 with the average linkage algorithm (epsilon = 2.5 Å; RMS on :532–537@C*, :67@CA, :79@CA, :112–117@CA, :125@CA, :194–195@CA, :228@CA, :283–285@CA, :326@CA, :329@CA, :395@CA, :435–436@CA; wherein residue 1 corresponds to residue 4 of the 3O9M crystal structure). This analysis identified six clusters of GSS^{octanoyl}FL·hBChE conformations with respective populations of 90.9, 4.1, 2.0, 1.0, 1.0, and 1.0%. The distances shown in Fig. 3 were derived from the time-averaged conformation of the most populated cluster. The coordinates of this conformation, residue 1 of which corresponds to residue 4 of the 3O9M crystal structure, are provided in Dataset S1.

1. Cornell WD, et al. (1995) A second generation force field for the simulation of proteins, nucleic acids, and organic molecules. *J Am Chem Soc* 117(19):5179–5197.
2. Cieplak P, Cornell WD, Bayly C, Kollman PA (1995) Application of the multimolecule and multiconformational RESP methodology to biopolymers: Charge derivation for DNA, RNA, and proteins. *J Comput Chem* 16(11):1357–1377.
3. Jorgensen WL, Chandreskhar J, Madura JD, Impey RW, Klein ML (1983) Comparison of simple potential functions for simulating liquid water. *J Chem Phys* 79(2):926–935.
4. Pang Y-P (2014) Low-mass molecular dynamics simulation: a simple and generic technique to enhance configurational sampling. *Biochem Biophys Res Commun* 452(3): 588–592.



Movie S1. Behavior video showing the appearance of untreated 16-mo-old male BALB/c and AAV-mBChE mut vector-treated, same-aged mice. The untreated mice (n = 23) exhibited ragged coats and bite wounds whereas the vector-treated mice (n = 29) retained clean and glossy fur. Two representative mice from each group are shown.

Movie S1

Other Supporting Information Files

Dataset S1 (TXT)