Supporting Information

An experimental and steered molecular dynamics simulations approach to histidine assisted liquid-phase exfoliation of graphite into few-layer graphene

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EXPERIMENTAL SECTION

Material. Bulk graphite powder (<20 µm, synthetic) was purchased from Sigma-Aldrich. The pristine graphite powder was used for the interaction of amino acids in water. Glycine (Gly), Alanine (Ala) Histidine (His), tryptophan (Trp), phenylalanine (Phe) were obtained from TCI (Japan). The organic solvents, NMP and DMF were received from TCI (Japan). All these chemicals were used as received without further treatment.

Amino acids-intercalated graphite. 0.4 g amino acids (Gly, Ala, Phe, Trp and His) were prepared in 40 mL of distilled water and the aqueous amino acids solutions are further sonicated in a bath sonicator (Bandelin-Sonorex Digitec, DT 514, 215 W, frequency 60 Hz, Germany) for 10 min for the complete dissolution. A powder of pristine graphite (0.250 g) was added in each amino acid solution and under stirring at 500 rpm for 7 days at room temperature in a dark, after the pre-heated graphite powder at 110°C for overnight. Finally, the resulting amino acids-intercalated

precipitates were obtained by centrifugation at 4,000 rpm for 30 min and washing with distilled water for several times to remove unreacted amino acids.

Exfoliation of amino acids-intercalated graphite samples in water. Amino acids-intercalated graphite samples are dispersed in 50 and 500 mL of water for the batch and large-scale production of graphene nanosheets, respectively (see the Supporting Information, Figure S1). The probe sonication (Labocon LUH-209, Ultrasonic Homogenizer, 2000 W, voltage 220V/50Hz, Probe 25mm, Φ=15 mm) is used for 10 mins (3 repeats) with a pulse rate of 10s on and 10s off. The output intensity of probe sonicator was set to be 50 %. The supernatant of colloidal nanosheets of few-layered graphene was carefully collected by centrifugation at 3,000 rpm for 30 mins for further characterizations. The single layer graphene (SLG) flakes obtained by centrifugation at 10,000 rpm for 60 mins. We also investigate the exfoliation of amino acids-intercalated graphite in various organic solvents (NMP and DMF) by bath sonication for 5 h.

Characterization. All the amino acids-intercalated graphite samples and exfoliated graphite nanosheets were deposited onto the Si substrate and dried at 50°C for 24 h for the X-ray diffraction (Bruker new D8 ADVANCE instrument equipped with a monochromatic Cu Kα radiation with a wavelength of 0.15418 nm which is operated at 40 kV and 40 mA) measurement. The UV-Visible absorption spectra of colloidal solutions of exfoliated graphite (PerkinElmer LAMDA 1050) were recorded by absorbance spectrometer with 1 cm optical path length cell. Fourier-transformed infrared (FT-IR) spectroscopy with Diamond-KRS-5 ATR mode (PerkinElmer Frontier-FTIR, UK) was used for recording spectra in a range of 400 to 4000 cm^{-1} for both, amino acids-intercalated and exfoliated samples with averaging 64 scans at 4 cm^{-1} resolution. The Raman spectra were measured on a RamanScope SENTERRA II (Bruker) with 532 nm excitation wavelength laser. For Raman measurements, the samples were deposited onto the glass substrate and recorded using a thermoelectric-cooled CCD array detector (Andor, DU420A-OE-152). The Raman signal was transferred through a 50 μm slit of a spectrometer and the spectral resolution of the spectrograph at 4 cm^{-1} across the Raman data acquired. The spectral acquisition time was 5s for all the samples with 2 accumulations averaged. The scanning electron micrograph images were obtained by a field emission scanning electron microscopy (FE-SEM, JEOL, JSM-7610F). Prior to the FE-SEM

measurement all the samples were coated with Pt. Transmission electron microscopy (TEM, JEOL JEM-ARM200F, Japan) was used to study the morphology of exfoliated graphite nanosheets. For the TEM sample preparation, all the samples were prepared by drop casting onto a carbon film-coated 300 square mesh copper grid and dried at room temperature in overnight in N_2 atmosphere. The zeta potential (Beckman Coulter, DelsaNano C, USA) analysis of exfoliated 2D nanosheets colloidal solutions was directly used without any further treatment under a constant temperature. High-resolution X-ray photoelectron spectroscopy (XPS) measurements were carried out on a JEOL XPS spectrometer (JPS-9019MC, Japan) for the powdered samples mounted on carbon tape using a monochromatic Mg Kα source (25 W, hυ = 1253.6 eV) and an energy resolution of 1 eV. All the XPS spectra were charge correction aligned on the binding energy scale referenced to the C 1s peak at 284.8 eV and a PF4 software was used for the deconvolution of the narrow-scan XPS spectra.

Computational Method.

The two graphene sheets of length 2.5X2.5 nm separated by 3Å was prepared using VMD Nano-structure Builder Plugin 1.5. Later the amino acids Glycine (Gly), Alanine (Ala), Tryptophan (Trp), Phenylalanine (Phe), and histidine(His) prepared with N-terminal and C-terminal domains with PH=7 configuration, among the amino acids histidine has different protonation state with respect to the imidazole nitrogen ion. Then the amino acids are placed in equal amounts on both sides of the amino acids using PACKMOL software[1] then the solvation box is built with 55 X 55 X 55 Å^3 sphere box dimension with approximately 6000 water molecules(No of water molecules differ with respect to the type of amino acid). Initial simulation was carried out for different concentrations for the histidine system alone. Later the constant concentration agreeing with experimental studies was used for other amino acids as well. The MD simulation of the system is performed with NAMD software[2] using the CHARMM force field, and the minimization of 10000 steps is done using the conjugate gradient method, followed by five nanosecond equilibration at 300 K with NPT ensemble. During the minimization and equilibration, the carbon atoms of graphene sheets are fixed with a harmonic constraint of 2 kcal/mol/Å^2 with no constraint over amino acids and water molecules. Langevin piston method with a 100 fs piston period was adapted to keep the pressure at 1 atm (damping time

constant was 50 fs, and piston temperature was 300 K). The periodic boundary condition is applied in all three coordinates (X, Y, Z) with grid spacing of 1 Å along the box size dimension. With the particle-mesh Ewald method, full electrostatics was employed with a 1 Å grid. Using a group based cutoff, non-bonded interactions were updated every 10 time steps. We used SHAKE algorithm to hold rigid covalent bonds involving hydrogen.

The steered molecular dynamics (SMD) simulation is run from the last time step from the equilibration process. During this process, only one graphene sheet is kept fixed, and another graphene sheet is pulled along the normalized X, Y, and Z-axis direction between the fixed and the SMD sheet. The pulling was performed at a constant velocity of 0.005 Å/timestep, equivalent to 0.4 Å/ps in the present case where the time step is 2 femtoseconds. The SMD atom will have a spring constant of 10 kcal/mol/Å with output energies printed every ten steps of a total of 20000 steps. The NVT ensemble system setup is used for SMD simulation. Finally, the force of pulling (pN vs. time step) is extracted along the Z-axis direction perpendicular to the graphene sheets.

Graphene parameters:

BONDS

CG2R61 CG2R61 938.00   1.4000 ! PROT benzene, JES 8/25/89

CG2R61 HGR61   340.00   1.0800 ! PROT phe,tyr JES 8/25/89

ANGLES

CG2R61 CG2R61 CG2R61   40.00   120.00   35.00   2.41620 ! PROT JES 8/25/89

CG2R61 CG2R61 HGR61   30.00   120.00   22.00   2.15250 ! PROT JES 8/25/89 benzene

DIHEDRALS

CG2R61 CG2R61 CG2R61 CG2R61   3.1000 2   180.00 ! PROT JES 8/25/89

CG2R61 CG2R61 CG2R61 HGR61   4.2000 2   180.00 ! PROT JES 8/25/89 benzene

HGR61 CG2R61 CG2R61 HGR61   2.4000 2   180.00 ! PROT JES 8/25/89 benzene

NONBONDED nbxmod 5 atom cdiel fshift vatom vdistance vfswitch -

cutnb 14.0 ctofnb 12.0 ctonnb 10.0 eps 1.0 e14fac 1.0 wmin 1.5

CG2R61 0.0   -0.0700   1.9924 !

HGR61   0.0   -0.0300   1.3582 !

CHARGE CENTER PERIPHERAL

CG2R61 0.0   0.15

HGR61   NA   -0.15

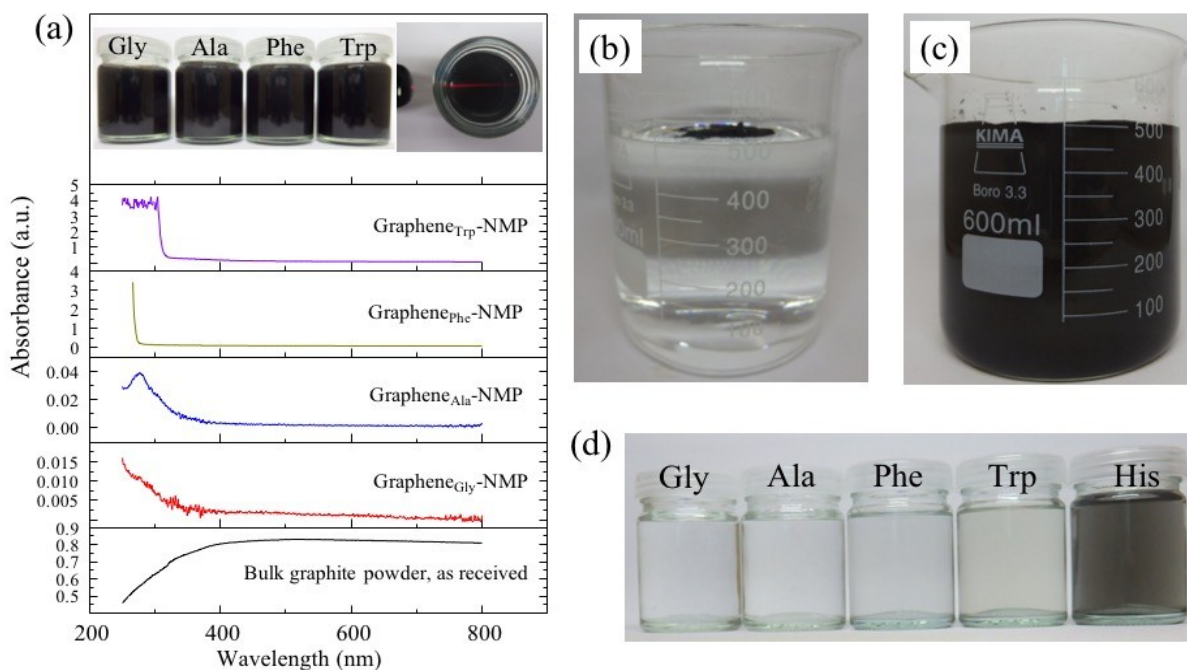For amino acids the standard amino acid parameter file (par_all36m_prot.prm) from CHARMM NAMD website is used.

Figure S1. (a) UV-vis spectra pristine graphite and graphene dispersion in NMP solvent. Note that before the sonication in NMP, the graphite powder was intercalated with different amino acids (AAs) where amino acids are being used as intercalant agent and it is denoted as a subscript. The AAs-intercalated graphite samples were collected by centrifugation at 4000 rpm at 30 mins and dried over night at 50C. The inset is digital photographs of colloidal graphene in NMP obtained by different AAs-intercalated graphite being used as starting materials and colloidal nature of graphene was also confirmed by the observation of the Tyndall scattering effect by passing red laser beam. The photographs graphene dispersion in water before (b), after sonication probe sonication (c) and an aqueous solution of graphene obtained by 0.1 g of histidine intercalated-graphite powder added in 500 mL of water and applied probe sonication of 30 min. (d) A digital photograph of graphene dispersion in water by different AAs-intercalated graphite as a starting material.
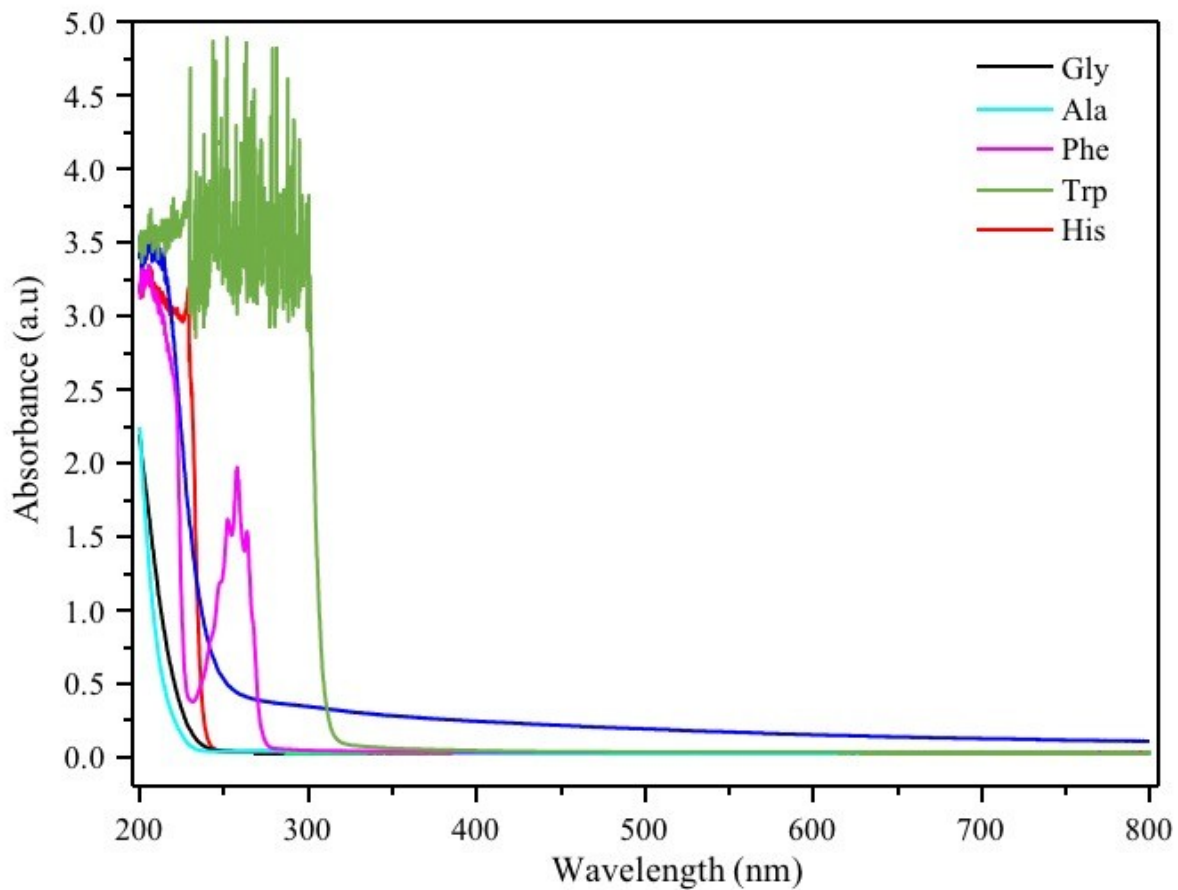
Figure S2. UV-vis spectra of aqueous amino acids solution. Note, 0.4g of each amino acids was dissolved in 40 mL of distilled water, the bath sonication was applied for 10 mins before the UV-vis measurement.
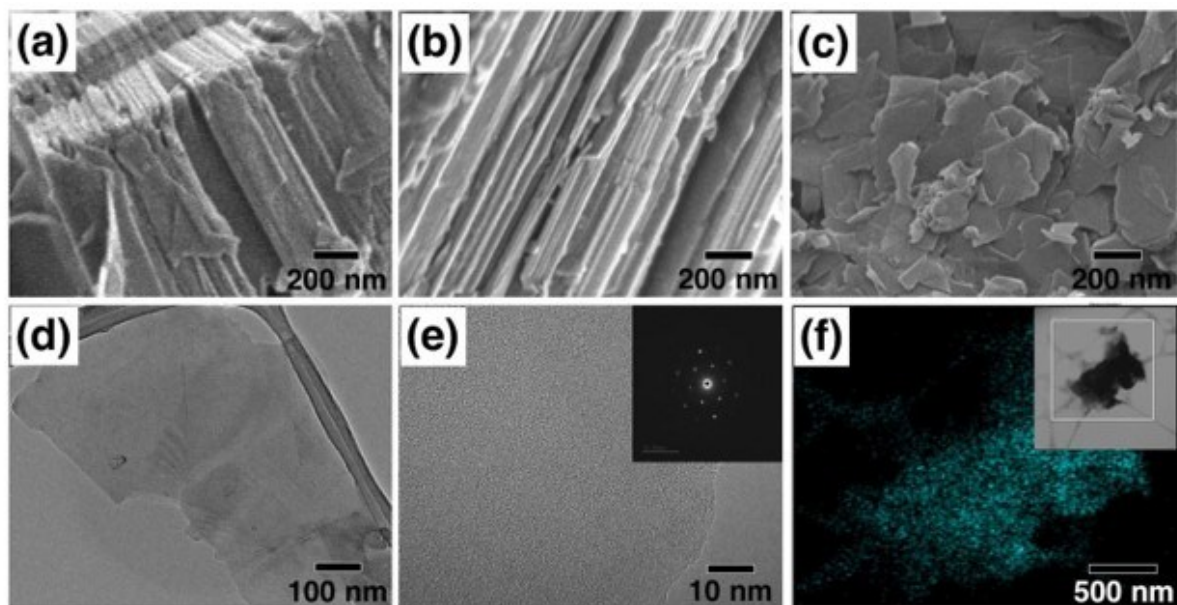
Figure S3. The SEM micrographs of pristine graphite, His intercalated graphite and exfoliated graphite nanosheets (a-c) by His. The low (d) and high-resolution TEM images of exfoliated graphite (e). The high-resolution TEM image of exfoliated flakes showing the mono-/or -a few layered graphene nanosheets. The inset (panel e) is the SAED diffraction pattern for the selected area of graphene. The inset (panel f) TEM image of the exfoliated graphite used for the EDX mapping analysis, showing the homogeneous distribution of carbon.
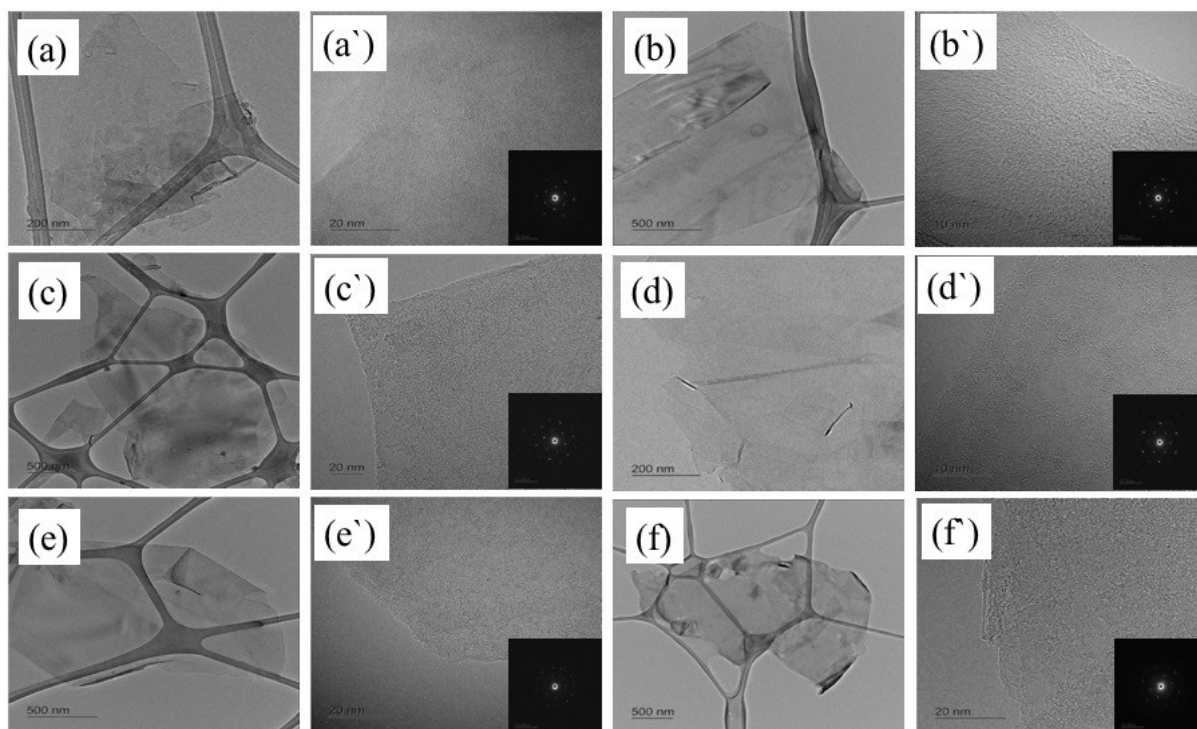
Figure S4. TEM images of exfoliated graphite in NMP solvent. The low resolution TEM images of exfoliated graphite (graphene) obtained by different treatments, such as without AAs (a), Gly (b), Ala (c), Phe (d), Trp (e) and His (f) intercalated graphite and corresponding high resolution TEM images (a`, b`, c`, d`, e` and f`), respectively. Inset images are SAED diffraction pattern of graphene by various amino acids.
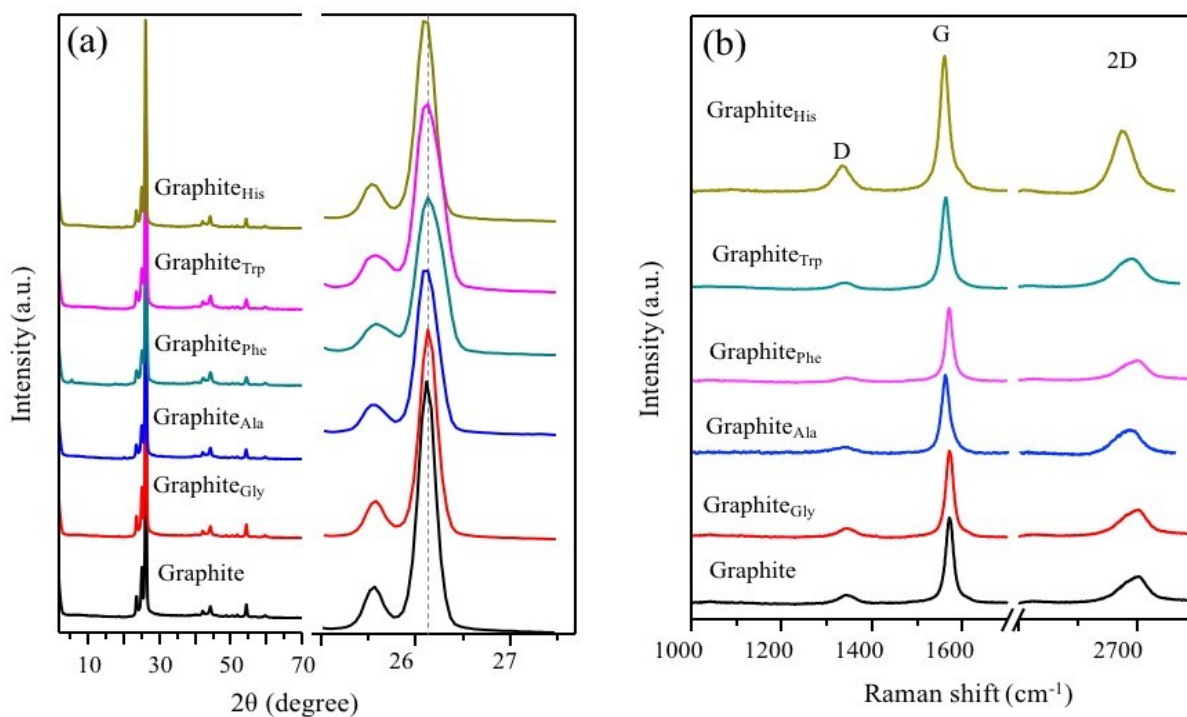
Figure S5. (a) The powder XRD patterns of different amino acids intercalated graphite, before the exfoliation in water. Note that, amino acids are being used as intercalant agent and it is denoted as a subscript in each sample. (b) Raman spectra of amino acids intercalated graphite. For Raman analysis, the samples are deposited onto glass substrate and excitation wavelength of 532 nm laser used. Before Raman analysis, the wavenumber was calibrated using the Si peak at 520 cm−1 as a reference.
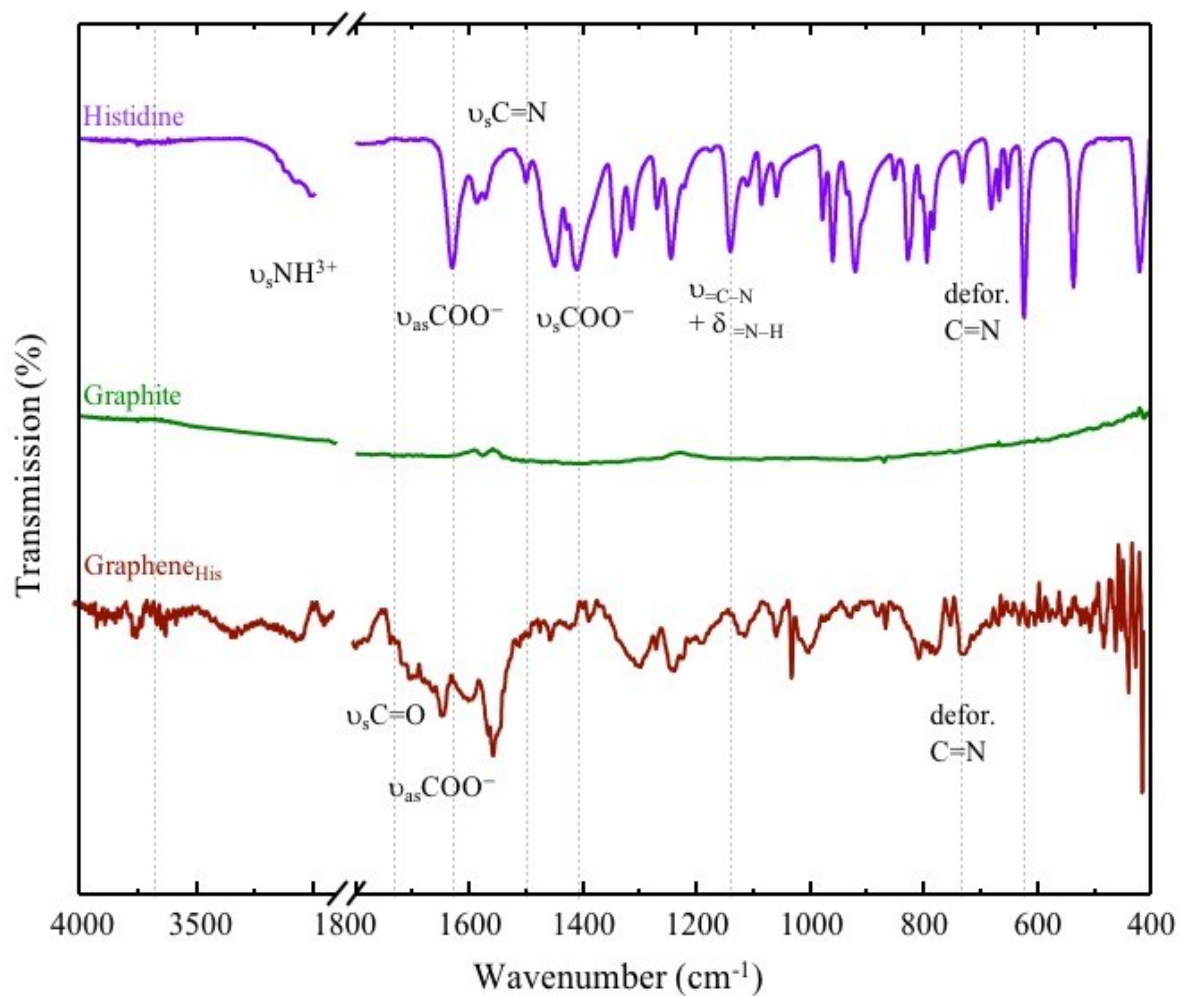
Figure S6. ATR-FTIR spectra of histidine (His) powder, pristine graphite and exfoliated graphite nanosheets by His.
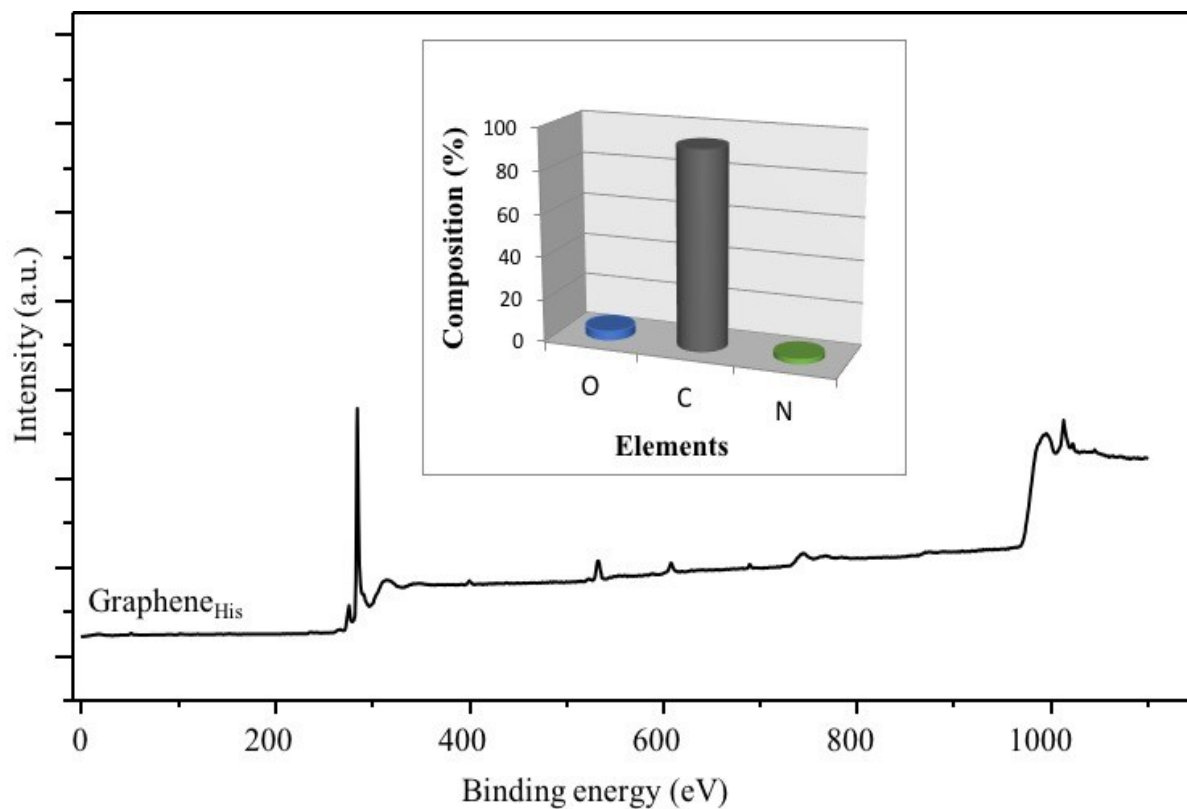
Figure S7. The XPS survey scan spectra of exfoliated graphite nanosheets by His. The inset bar graph showing the elemental composition of exfoliated graphite nanosheets based on XPS measurements.
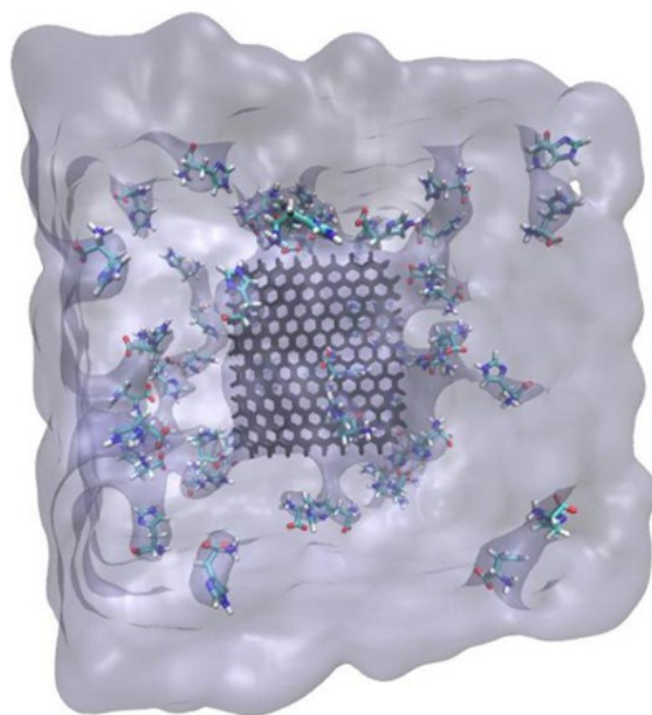
Figure S8. The initial simulation setup of water box size 55 X 55 X 55 Å3 with histidine amino acids and graphene sheet.
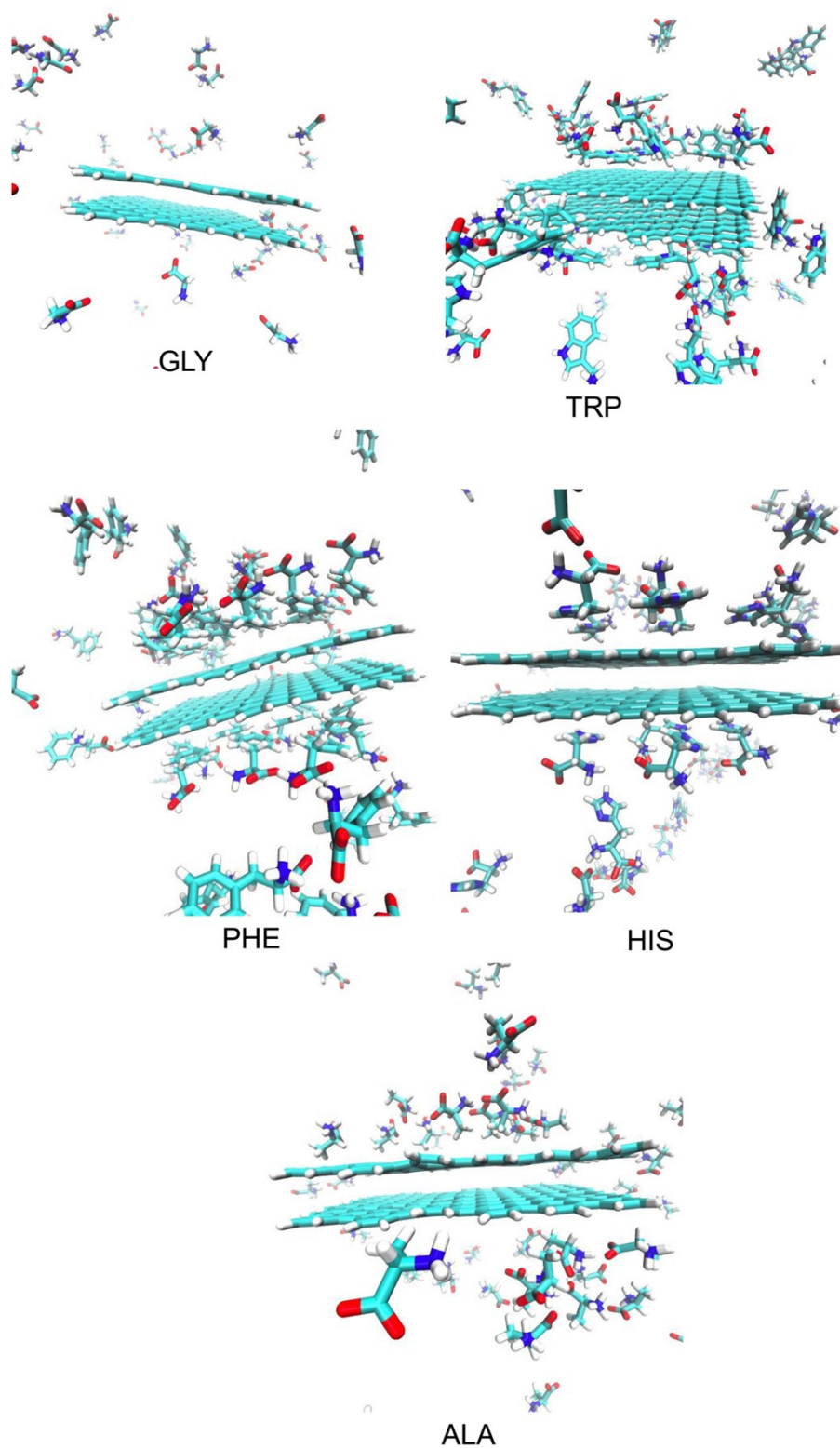
Figure S9: Cross sectional view of last frame from 5ns MD equilibration system.
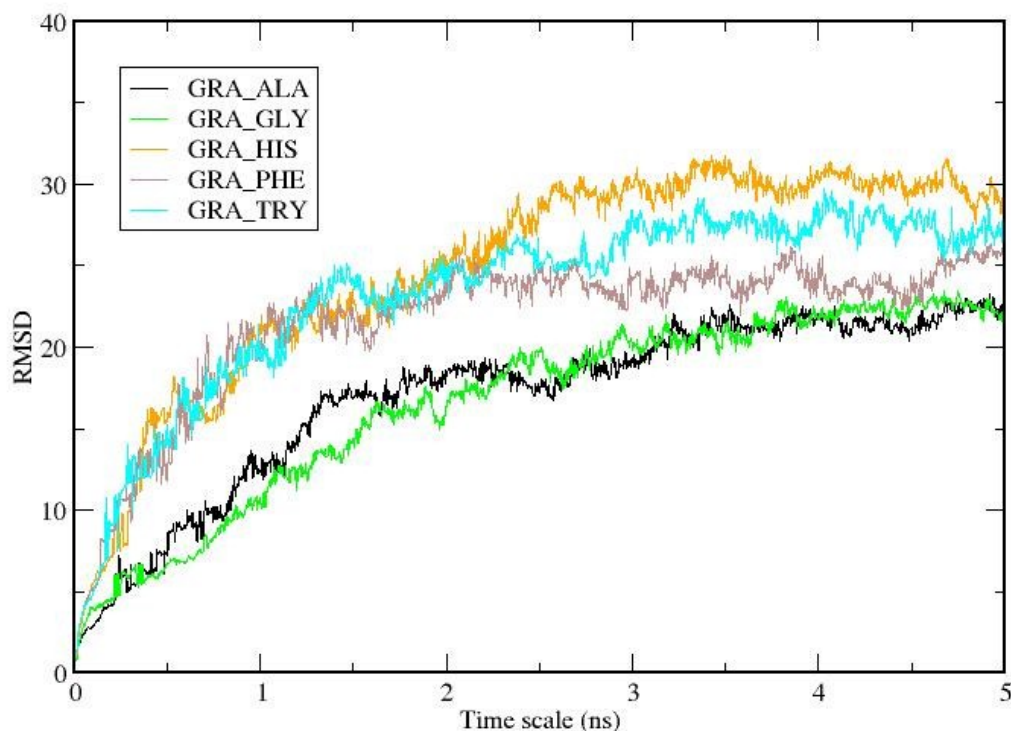
Figure S10. Root mean square deviation of the equilibration run of 5ns.

References for the supporting information:

1.   Martínez, L., Andrade, R., Birgin E. G., Martínez J. M. (2009). PACKMOL: A Package for Building Initial Configurations for Molecular Dynamics Simulations. J. Comput. Chem. *13*, 2157-2164.

2.   Phillips, J. C., Braun, R., Wang, W., Gumbart, J., Tajkhorshid, E., Villa, E., Chipot, C., Skeel, R. D., Kalé, L., Schulten, K. (2005). Scalable Molecular Dynamics with NAMD. J. Comput. Chem. *16*, 1781-1802.