Self-templated covalent functionalization of graphitic surfaces with a quasi-periodic pattern

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1 | INTRODUCTION

Pattern engineering of surfaces plays a very important role in many applications related to chemical, optical, and electronic properties. Therefore, a variety of strategies have been developed, targeting different dimensional scales, functionalities, and surfaces. Typically, these strategies can be classified according to the templating method used for fabricating the pattern, for example, photolithography, dip-pen lithography, and nanopipette scanning lithography are extensively used for the patterned modification of different substrates. Thereby, graphitic surfaces (GSs) with their sp² hybridized carbon crystalline lattice attract lots of attention due to their unique electronic and interfacial properties.

Among the methods for the covalent functionalization of GS, diazonium salts are well-developed precursors for facilely grafting various organic-addend-groups to GS. Based on diazonium chemistry, the patterned covalent modification (PCF) of GS can be realized through several strategies with different templating methods. These are (1) photolithography-based masks for templating the chemical-functionalization of GS, (2) nanopipette scanning lithography to spatially control the chemical-functionalization of GS, (3) laser scanning lithography to realize the PCF by spatially inducing dediazotization of a diazonium salt on the GS, (4) nanoparticle lithography as template for the chemical-functionalization of GS, and (5) molecular lithography as template for the chemical-functionalization of GS at the molecular scale. In principle, in all of these strategies the chemical-functionalization of GS has to be templated or controlled spatially via external lithography processes, while a one-step PCF of GS without any external template or control remains challenging.

In this context, Palermo et al. reported a self-templated method where a linear pattern was covalently formed on graphene through electrochemical reduction of a diazonium salt with a long alkane chain. Thereby, the produced radicals with the long alkane chain served two different purposes simultaneously, that is, the covalent functionalization of graphene by the radical-end and the physical functionalization by the adsorption of the alkane chain. Further, our group developed an electrochemical reduction method where one-step covalent functionalization of GS forms a PCF layer with so-called fairy circles. In this method, diazonium salts are electrochemically reduced in situ to produce aryl-radicals for the chemical functionalization of the GS. A key point in this method is that the fairy circles were formed...
only when a mixture of two different diazonium salts was used, which was attributed to the adjustment in surface tension of the aqueous solution and electrochemical reduction speed of diazonium salts in combination with the adsorption of nanobubbles of by-produced gas molecules (N₂ and NO), which may physically adsorb on GS and serve as template for the fairy circles in the covalent functionalization layer.

In this work, bypassing the electrochemical reduction, we present a simplified chemical reduction method to realize the self-templated PCF on a GS. Instead of circles, linear vacancies with three-fold symmetry are formed in the PCF layer. Although these vacancies are also attributed to the adsorption of N₂, here the N₂ appears to adsorb in linear structures with a quasi-periodic three-fold symmetry, corresponding to the hexagonally symmetric substrate, without forming nanoscale bubbles. Consequently, a strikingly different nanoscopic pattern (linear with three-fold symmetry vs. round) is produced from the same grafting reaction. Here, a synergistic effect of chemical- and physical-functionalization on GS is proposed to explain the formation of a PCF layer containing striped corrals with three equivalent orientations (Figure 1). The formation of these striped domains is attributed to the self-assembly of N₂ on the GS, a feature that has been reported previously.\[39–44\]

The observed three-fold symmetry corresponds to the underlying graphitic crystalline lattice. Moreover, an evolution of the PCF layer was observed depending on the reactant concentration, that is, the striped corrals were gradually covered during the layer growth. For fabricating the PCF layer, the critical conditions for forming the striped corrals have been established, and the peculiar impact of graphitic steps was revealed, which supports our proposed explanation. All in all, in contrast to PCF strategies via lithography techniques, this self-templated strategy does not require external control to fabricate the PCF layer with a quasi-periodic pattern, providing a new perspective for surface patterning through heterogeneous, simultaneous physi- and chemisorption on crystalline surfaces.

2 | RESULTS AND DISCUSSION

2.1 | Protocol for the patterned functionalization of GSs

The chemical-functionalization of GS was realized based on the reduction of 4-nitrobenzenediazonium (NBD) with the reducing reagent potassium iodide (KI).\[45\] in situ at the liquid/solid interface (LSI) in a reaction cell as shown in Figure 1A. Therefore, a cylindrical cell was placed onto freshly cleaved highly oriented pyrolytic graphite (HOPG), creating a closed reaction cell on the HOPG. The closed reaction cell results in a static LSI after the KI and NBD solutions are sequentially dropped into the cell (dimethyl sulfoxide [DMSO] as solvent). Thus, the KI and NBD solutions are mixed in situ at the LSI, and the reduction of NBD immediately takes place, producing N₂ and aryl-radicals near the LSI and in the solution (Figure 1B). Subsequently, the produced aryl-radicals can covalently attach to the GS to realize the chemical functionalization, leading to the appearance of sp³-hybridized defects in the HOPG lattice and forming a grafting layer.

After the functionalization, the HOPG was rinsed, dried, and characterized with atomic force microscopy (AFM) to examine the morphology of the grafted layer. The AFM image in Figure 1C was obtained from the GS functionalized with a KI/NBD concentration of 100 mM and a functionalization time of 5 min. Surprisingly, the AFM images show a covalent functionalization layer patterned with empty striped corrals with three equivalent orientations (see center inset of Figure 1C and corresponding fast Fourier transform [FFT] of the AFM images in Figure 1). The topography of the PCF layer is further visualized by the corresponding 3D representation of the AFM image (Figure 1D). Larger AFM images (Figure 1F, from 4 to 400 µm²) show that the PCF layer has a high homogeneity and similarity in the morphology, with three equivalent orientations of the striped corrals. The corresponding FFT results (upper right insets) further confirm the three orientations and reveal a quasi-periodic pattern with three-fold symmetry.

The observed striped corrals are attributed to a passivation based on a physisorbed template on the GS, which locally prevents the chemical functionalization. This assumption is based on the three-fold symmetry of the striped corrals, which corresponds to the substrate symmetry as well as the absence of the template in the AFM images after the rinsing of the sample. Possible compounds to form the physisorbed layer in the reaction mixture are the educts KI and diazonium salt as well as aryl radicals and N₂ or other more complex side-products, which are formed during the reaction. Of these options, we propose the self-assembly of N₂ to be the origin of the striped corrals as the gas molecules have an affinity to hydrophobic surfaces, in particular at interfaces of two dramatically different phases: the polar liquid phase and the nonpolar hydrophobic solid phase.\[46–50\] Thereby, besides the chemical-functionalization with the aryl radicals, it is reasonable to assume that the in situ produced N₂ preferentially adsorbs on the hydrophobic GS to form physisorbed N₂ domains. The occurrence of the striped domains from the physisorption of N₂ shows that the physisorption prevents the chemisorption, indicating that the nucleation and formation of N₂ are faster compared to the chemisorption. In turn, the chemical functionalization takes place before the entire surface is covered by a physisorbed layer, and thus can encircle and restrict the physisorbed domains. Although the chemisorption would be thermodynamically favored over the physisorption, by blocking the access to the surface for the radicals the physisorption kinetically prevents the chemical functionalization. The AFM images demonstrate that the alignment of the empty striped corrals corresponds to the symmetry of the underlying HOPG surface, indicating that the template of these striped corrals has similar features (Figure 1E). Furthermore, the striped corrals in the PCF layer turn out to be aligned along the armchair direction of the graphitic lattice (vide infra). As discussed previously,\[46,48,50\] the produced N₂ can form a physical functionalization layer at the LSI of GS.

Interestingly, in the past decades, numerous studies have evaluated the interaction between N₂ and GS. Basically, a condensed layer of N₂ adsorbed on GS was a typical consensus based on experimental and simulation studies,\[43,44,51–55\] whereby the formation of the condensed N₂ layer is related to many factors, that is, the concentration of N₂ at the LSI,\[43,51,52\] the temperature of the LSI,\[52\] and the relative
FIGURE 1  (A) Schematic presentation of the typical experimental procedure for the functionalization of graphitic surface (GS) (HOPG); (B) The reaction mechanism of the functionalization of GS through the reduction of a diazonium salt by potassium iodide (KI). (C) Atomic force microscopy (AFM) image of the PCF layer with stripes in three equivalent orientations (indicated by the equilateral triangle) formed on GS and (D) the corresponding 3D representation. (E) Schematic illustration for the striped domains of self-assembled N$_2$ templating the chemical-functionalization on GS, aligned along the graphite armchair direction. (F) The corresponding AFM images with larger scanning sizes. The upper right insets in all AFM images show the fast Fourier transform (FFT) of the corresponding AFM images. The sample was functionalized with 100 mM KI/nitrobenzenediazonium (NBD) solutions and a functionalization time of 5 min.

affinity of gas molecules to GS.$^{[51,56]}$ Thereby, the condensed N$_2$ layer on GS has been clearly visualized by AFM imaging and shown to consist of striped domains, which have three equivalent orientations due to the alignment effect of the hexagonal graphitic lattice.$^{[39-44]}$

The reported self-assembly behavior of N$_2$ on GS is consistent with the behavior of the physical functionalization, that is, the template of the striped corrals in our functionalization system, thus supporting our proposed explanation that the striped domains formed by the self-assembly of N$_2$ templates the chemical functionalization of aryl radicals on the GS, fabricating the PCF layer.

2.2 | Concentration dependent functionalization

According to the discussed explanation of the self-templated PCF on GS, the concentration of the produced N$_2$ and aryl-radicals are expected to affect the efficiency and outcome of the functionalization of GS. To investigate the concentration dependence of the functionalization process, we systematically varied the concentration of KI/NBD from 5 to 500 mM, while keeping the functionalization time (5 min) and the molar ratio of KI/NBD (1:1) constant. Thereby, the experimental procedure described in Figure 1A was followed,
The concentration-dependent functionalization of graphitic surface (GS). The reaction time (5 min) as well as the molar ratio of nitrobenzenediazonium (NBD) and potassium iodide (KI) (1:1) was kept constant. (A) Representative atomic force microscopy (AFM) images of the concentration-dependent functionalized GS. (B) Representative AFM images with higher resolution and the corresponding line profiles and layer thicknesses. (C and D) Quantification of the area, number, length, and width of the striped corrals obtained at 20 mM and 100 mM. The area of striped corrals was classified into three types depending on their relative orientations. The three different orientations are marked with three different colors, red (orientation-1), green (orientation-2), and blue (orientation-3), as shown in B2 and B3. The average corral densities, that is, number of corrals per square micrometer, were determined and compared for each orientation. (E) I(D)/I(G) ratio as a function of the reactant concentration. The average value and error bar (the standard deviation) are based on the 25 values from the Raman mapping.

Representative AFM images reveal a significant change in layer morphology with increasing concentration (Figure 2A). At a low concentration (5 mM, Figure 2A1), the GS was functionalized with a low-density, regular layer. The striped corrals appeared when increasing the concentration to 20 and 100 mM (Figure 2A2, A3) but vanished again upon increasing the concentration above 300 mM (Figure 2A4, A5). Additionally, AFM images with higher resolution unveil an increase in the densities of the grafted molecules with increasing concentration (Figure 2B). Further, line profiles of these AFM images show an increase in layer height from monolayer (around 1 nm) to multilayer (around 1.8 nm) upon increasing the concentration to 300 mM. This increase in layer height is attributed to the dendritic growth of aryl-radicals on GS, as reported in previous studies.\cite{22,24,57,58}

To further evaluate the PCF layer, the number, length, width, and area of the striped corrals of the 20 and 100 mM samples were quantified. Thereby, the striped corrals were classified into three types based on their relative orientation and marked red (orientation-1), green (orientation-2), and blue (orientation-3), as shown in Figures 2B2, B3 (see Figures S1 and S2 for more details). The statistical analysis revealed a homogenous distribution of the three different orientations, confirming that the orientations are equivalent and result from the substrate symmetry, and similar corral density (around 5 corrals/µm²) of the 20 mM and 100 mM samples (Figure 2C, D and Figure S3). The main difference between the 20 and 100 mM samples is the relative grafted area, which increases from 79.8% to 87.2% upon increasing concentration. As the corral density is very similar, this difference is due to the shrinking in average dimension (width/length/area) of the striped corrals at the higher concentration of 100 mM (Figure S3). Additionally, the shrinking in dimension of the...
striped corrals improves the uniformity of the striped corrals, which is reflected in the scatter plots of corral width versus length (Figure S3A and B).

Raman measurements were performed to quantify the chemical functionalization, that is, grafting density, of the GS. In the Raman spectra, the D band represents defects such as covalent modifications. Typically, the ratio of D band to G band intensity (I(D)/I(G) ratio) is used to quantify the grafting density of chemically functionalized GS. The I(D)/I(G) ratio of the functionalized samples gradually increases with a higher concentration as shown in Figure 2E (see details in Figure S4), indicating a gradual increase in grafting density when using higher concentrations, which is consistent with the trend of the layer density observed in AFM imaging.

Interestingly, in some areas of the 500 mM samples, the striped corrals were not empty and instead covered by grafted molecules with an increased height, that is, appearing as white stripes in the AFM images (Figure S5). These covered striped most likely result from the dendritic multilayer growth, which not only leads to an increased layer height but can also result in covering the striped corrals by growing sideways as illustrated schematically in Figure S5. Similarly, on the 100 mM sample, apart from the typical PCF layer described above, several other layer morphologies were observed occasionally. Based on the morphology and thickness of the PCF layer, five different functionalization states were identified by AFM imaging (Figure S6). The first state of the functionalization layer (state 1) is the typical PCF monolayer with quasi-periodic pattern (Figure S6A), which has been described above (Figures 1 and 2). In some areas, however, a thicker functionalization layer (up to 1.5 nm) with smaller (state 2 and 3), covered (state 4) or even disappeared (state 5) striped corrals was observed. These differences are also attributed to dendritic layer, resulting from local concentration variations during the functionalization. That is, after the addition of the reactant solutions, turbulences or convective effects can result in an increased mass transport and thus locally increased radical concentrations.

Additionally, the impact of the functionalization time on the layer formation was investigated. Therefore, the GS was exposed to a reaction mixture with a concentration of 20 mM for different times ranging from 30 s to 5 min. AFM imaging of these samples shows that the PCF layer is formed already after a functionalization time of only 30 s and that the morphology of the grafting layer does not change significantly with longer reaction times (Figure S7A,B). Additionally, Raman measurements show an I(D)/I(G) ratio above 0.025 already after 30 s, which increases only slightly to 0.035 after 3 min and does not increase further after 5 min (Figure S7C). Similarly, the average thicknesses of the PCF layer slightly increased from 1.3 nm (30 s) and saturated at 1.5 nm after 3 min (Figure S7D). Overall, the time-dependent measurements show that the grafting reaction occurs on a time scale of several seconds after the second reactant solution is added to the reaction cell, which is followed by the formation of an equilibrium at the LSI. In the formed equilibrium, the local concentration of radicals near the surface is reduced, and thus the layer does not grow significantly with increasing reaction time. Additionally, the reaction of the formed radicals with each other in the solution results in a decrease in radical concentration, thereby reducing further layer growth after longer reaction times.

2.3 The critical conditions for forming the PCF layer

To unveil the critical conditions required for the self-templated functionalization and evaluate the proposed mechanism, univariate experiments with a modified experimental protocol were carried out by changing single steps of the protocol shown in Figure 1A, while keeping the concentration (20 mM) and functionalization time (3 min) constant. The AFM images in Figure 3A show the PCF layer obtained with the typical experimental protocol, which was used as reference. The first control experiment (control-1) was performed without the rinsing step, that is, after the functionalization reaction the solution was simply removed without the rinsing steps. The PCF layer morphology and I(D)/I(G) ratios obtained with the control-1 experiment (Figure 3B) are very similar to the reference PCF layer (Figure 3A), except for some residues on the PCF layer without rinsing. Also the layer thickness (Figure S8) was in the range of the layer thickness obtained with the reference protocol, indicating that the template responsible for the striped corrals is volatile and easily removed to leave empty corrals. Thus, the empty corrals observed without rinsing support the hypothesis of physisorbed N2 gas as template for the striped corrals.

Further, in the control-2 experiment the reactant solutions were premixing in a vial and then transferred into the reaction cell. The morphology of the functionalized layer obtained with the control-2 protocol is dramatically different from the typical PCF layer, indicating a different layer formation mechanism (Figure 3C). That is, the layer consists of sparse irregular island-like domains with a low grafting density (see details in Figure S9), consistent with the low I(D)/I(G) ratio of 0.004 (Figure 3C), which is about one order of magnitude lower compared to the reference layer. The premixing implies that the reduction of NBD has started before the solution is in contact with the GS, thus producing aryl-radicals and N2 before the mixture arrives at the GS and can form stable conditions at the LSI in the reaction cell. Thus, in the premixed solution, the produced aryl-radicals inevitably can react with each other to form complex side-products, while the produced N2 can also aggregate to form nanobubbles in the solution.45,59–61 Both, the side products and nanobubbles can adsorb on the GS and prevent the reaction of the remaining radicals on the GS. Further, the concentration of the radicals in solution may be lower due to the side reactions taking place before the solution is in contact with the GS. Overall, the control-2 experiment shows that the in situ production of aryl-radicals and N2 at the LSI are key factors for the formation of the reference PCF layer.

The control-3 experiment was performed without the circular cell, that is, the KI and NBD solutions were sequentially drop-cast onto the bare GS (Figure 3D). As a result, the KI and NBD solutions were mixed in situ but also flowing over the edges of the GS. The obtained grafting layer showed a high density with bright stripes, which have been observed previously as shown in Figures S5 and S6. However, a few striped corrals remained empty, allowing the determination of the layer thickness to be around 2 nm, which was further confirmed by scratching of the grafted layer (Figure S10). Additionally, the Raman map (Figure 3D) of control-3 shows a higher I(D)/I(G) ratio with a more uniform distribution compared to the reference and other control experiments.
Both observations can be explained by the stronger disturbance in the mass transport at the LSI due to the missing circular cell. That is, the uncontrollable mass transport over the whole GS leads to a faster and more random mixing of reactants, thus a higher radical concentration and higher grafting density. Note that a different dropcasting order, that is, dropcasting the NBD solution before the KI solution, also results in a different functionalization layer that has very low grafting density and thin layer thickness as shown in Figure S11, which may be attributed to the differences in diffusion coefficient and affinity to GS between KI and NBD.

Overall, these control experiments show the importance of the in situ generation of the aryl-radicals and N₂ due to the sequential addition of the KI and NBD solutions and the static conditions at the LSI resulting from the circular reaction cell for the formation of the grafting layer with striped corrals. The empty corrals without rinsing further support the hypothesis of self-assembled N₂ as template for the striped corrals.

### 2.4 Graphitic step-edge induced striped corrals

As mentioned above, the striped corrals in the typical layer have three equivalent orientations. However, additional patterns were observed locally at graphitic step edges, originating from the influence of the GS itself. In these patterns, the striped corrals are parallel with respect to each other and perpendicular to the adjacent step edge (Figure 4 and Figure S12). Due to the alignment effect of the underlying crystalline substrate and the resulting three equivalent orientations of the striped corrals, these parallel striped corrals appear predominantly when the graphitic step edges are perpendicular to one of the three corral orientations (green dotted lines in Figure 4 and Figure S12). In contrast thereto, the step edges marked with red dotted lines in Figure 4B and Figure S12 have different orientations and no adjacent parallel striped corrals. Furthermore, the corresponding 3D representations in Figure 4 clearly show the topographical details in the graphitic step-induced PCF layer.

Graphitic step edges have been shown to act as preferential adsorption sites for various materials, induce the growth of metal nanorods, and show different chemical reactivities. Similarly, assuming that the templates of the striped corrals are aligned based on the symmetry of the underlying graphitic lattice, a graphitic step edge can induce the formation of parallel striped corrals perpendicular to the graphitic step if the edge is aligned perpendicular to the preferential orientation of the physisorbed template for the striped corrals. When the aryl-radicals and N₂ are produced in situ near the graphitic step, the corner of the...
Graphitic step-induced patterns in the functionalized layer. (A₁–D₁) Representative atomic force microscopy (AFM) images showing graphitic step-induced parallel striped corrals in the functionalized layers. (A₂–D₂) Corresponding 3D representations of the AFM images. (E) Schematic illustration for the formation of the graphitic step induced self-templated patterning functionalization: (E₁) Graphitic step-induced nucleation and self-assembly of N₂. (E₂) The striped domains formed perpendicular to the graphitic step due to the self-assembly of N₂ templating the covalent functionalization. (E₃) Step-induced striped corrals formed in the covalent functionalization (CF) layer. (A and B) Obtained from a sample functionalized with 20 mM potassium iodide/nitrobenzenediazonium (KI/NBD) for 0.5 min. (C and D) Obtained from a sample functionalized with 20 mM KI/NBD for 3 min. More additional AFM images are shown in Figure S11.

2.5 Orientation of the striped corrals

One question that remains to be addressed is the orientation of the striped corrals. In order to decode the orientation of the striped corrals with respect to the crystalline GS, the self-assembly of pentacontane (n-C₅₀H₁₀₂) on the PCF layer was investigated, as an internal ruler. It is known that long alkane chains align along the zigzag direction of the graphitic lattice [70,71] and are stacked in rows. These rows run parallel to the armchair direction of the graphitic lattice (as shown in Figure S13B). Amid the striped corrals in the PCF layer, an additional square corral (1 by 1 µm²) was made via a nanolithography approach, that is, nanoshaving, based on contact mode AFM [71] as shown in Figure S13A. Based on the orientation of the rows of stacked n-C₅₀H₁₀₂ molecules in the square nanocorral, and their orientation with respect to the striped corrals surrounding the square corral, it can be concluded that the striped corrals in the PCF layer are aligned along the armchair direction of the graphitic lattice. This is confirmed by the self-assembly of n-C₅₀H₁₀₂ in the striped corrals (Figure S14). Additionally, based on the armchair orientation of the striped corrals, we can conclude that the graphitic step edges have a zigzag morphology (Figure 4).

3 CONCLUSIONS

In summary, a facile and fast self-templated approach without the requirement of an additional template or control has been developed for the PCF of GS with in situ produced aryl radicals. A quasi-periodic pattern is formed in the PCF layer in the form of striped corrals with three equivalent orientations aligned to the armchair direction of the underlying substrate. The formation of the striped corrals is attributed to a local passivation of the substrate by a physisorbed layer, which prevents the chemical functionalization. The physisorbed layer is proposed to consist of self-assembled N₂, which is produced in situ by the reduction of a diazonium salt by KI as reducing reagent. The advantage of the presented approach lies in its simplicity and convenience to obtain patterned covalent functionalization of GSs, showing the potential of physisorbed layers as templates for patterned functionalization of graphitic materials, providing a new perspective for the development of heterogeneous quasi-periodic modification of crystalline surfaces.
4  |  EXPERIMENTAL SECTION

4.1  |  Materials

4-NBD tetrafluoroborate (98%) from TCI EUROPE, DMSO (99.9%), phenyloctane (PO) (≥ 99%) and pentacontane (n-C_{50}H_{102}) (98%) from Sigma-Aldrich. Ethanol (Spectroscopy purity) from EMD Millipore. All chemicals were used without further purification. High purity water (Milli-Q, Millipore, 18.2 MΩ) was used for all the experiments. The highly ordered pyrolytic graphite (HOPG, grade ZYB, Advanced Ceramics Inc., Cleveland, USA) substrates were freshly cleaved before use. The circular cell is home-made out of Teflon.

4.2  |  Self-templated covalent functionalization of GSs

To prepare the reaction solutions, 4-NBD and KI were dissolved in DMSO to prepare the NBD and KI solutions with the desired molar concentrations. Before performing the functionalization experiment, the HOPG was freshly cleaved to prepare a clean surface and then clamped in a home-made circular cell to form a reaction cell for the functionalization. The material of the home-made circular cell is teflon (polytetrafluoroethylene). The inside diameter of the cylindrical cell is about 0.7 cm. An O-ring was put on the bottom of the cylindrical cell. The reaction cell was built by placing the cylindrical cell onto the center area of freshly cleaved HOPG. The O-ring surrounds the bottom of the reaction cell to avoid leakage. Subsequently, 50 µL KI solution was firstly drop-casted into the reaction cell, followed by 50 µL NBD solution. In all experiments, to minimize the influence of differences in the dropcasting speed, the reactant solutions (50 µL) were always added in one go using the pipette adjusted to a volume of 50 µL. After a given time, the reaction was stopped by pouring out the reaction solution. The functionalized surface was immediately rinsed three times with ethanol and water and dried under a stream of argon.

4.3  |  Univariate experiments compared to the reference protocol

Based on the reference protocol of the self-templated covalent functionalization of GS, a series of univariate experiments were performed. The first univariate experiment is without the rinsing step of the reference protocol, that is, all other experimental steps in this control experiment were the same as the reference protocol apart from the step of rinsing. The second univariate experiment was performed with premixing the NBD and KI solutions before dropcasting them into the reaction cell. That is, instead of sequentially dropcasting the KI and NBD solutions, 50-µL KI and 50-µL NBD solutions were mixed together, and then the 100-µL mixture was dropcasted into the reaction cell. In the third univariate experiment, the KI and NBD solutions were sequentially dropcasted onto the HOPG without the circular cell.

4.4  |  Self-assembly of n-C_{50}H_{102} in corrals

The 10^{-3} M pentacontane (n-C_{50}H_{102}) solution was prepared using the solvent 1-PO for dissolving the solid powder of n-C_{50}H_{102}. The experiment of n-C_{50}H_{102} self-assembly was based on the sample functionalized with 20 mM KI/NBD for 3 min. Firstly, an additional square corral (1 by 1 µm²) was made on the patterned covalent functionalization (PCF) layer by nanoshaving based on the contact mode of AFM. Subsequently, 5 µL 10^{-3} M n-C_{50}H_{102} was drop-casted on the PCF layer with the shaved square corral. The sample was kept at ambient condition overnight and dried naturally. The dried sample was further characterized using AFM for visualizing the self-assembly of n-C_{50}H_{102} on the surface.

4.5  |  Raman measurements

Raman characterizations were carried out at room temperature using a Raman microscope (Monovista CRS+, S&I GmbH). The 632.8 nm He–Ne laser beam was directed and focused onto the HOPG surface with an objective (OLYMPUS, BX43 100x, N.A. 0.7). The power density of the laser point focused at the functionalized surface was about 590 kW/cm². The Raman scattering signal was collected with the same objective and directed to a Raman spectrograph (Horiba JY, iHR-320) equipped with a cooled charge-coupled device camera operating at −100°C (Andor, DU920P) through a dichroic mirror, a pinhole, and a long pass filter (Chroma, HQ645LP). The accumulation time for a single Raman spectrum was 10 s. Raman mapping was performed based on array scanning in an area of 50 × 50 µm with 25 Raman spectra (5 by 5 arrays).

4.6  |  AFM measurements

AFM characterizations were performed using a Cypher ES (Asylum Research) system at 32°C in tapping mode at the air/solid interface. OMCL-AC160TS-R3 probes (spring constant ∼26 N/m) with a resonance frequency around 100 kHz were used for the AFM imaging. OMCL-AC240TS-R3 probes (spring constant ∼2 N/m) with a resonance frequency around 70 kHz were used in contact mode for scratching the functionalized layer.

4.7  |  Results analysis

The AFM images and the corresponding 3D representations were analyzed using the SPIP 6.3.5 software. In detail, the functionalized layer thicknesses were measured by lines profiles over the functionalized layer and empty areas (striped corrals). As comparison, the functionalization layer was artificially scratched away with the AFM tip in contact mode to confirm the layer thickness. To characterize the pattern formed in the functionalized layer, statistics regarding several intrinsic parameters of the striped corrals, such as relative orientation, length (nm), width (nm), and area (nm²) of the striped corrals were analyzed. Therefore, the striped corrals in an AFM image were marked along the edges of the cor-
The marked striped corrals were then classified into three types, according to their three relative orientations, and marked blue, red, or green. Based on the marked striped circles, the length (nm), width (nm), and area (nm$^2$) of the striped corrals measured. The uniformity of the three different relative orientations of the striped corrals was evaluated by FFT. For the Raman results, we focused on the D band (related to the presence of sp$^3$ covalent defects) and the G band (intrinsinc band to GS) in the Raman spectrum and further calculated the intensity ratio of D band to G band, namely I(D)/I(G). Based on 25 values for every Raman map, we further did the statistical analysis regarding the ratio of I(D)/I(G), obtaining the average value, and error bar (the standard deviation).

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CONFLICT OF INTEREST
The authors declare no conflict of interest.

DATA AVAILABILITY STATEMENT
The data that support the findings of this study are available from the corresponding author upon reasonable request.

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