

Bioenabled Core/Shell Microparticles with Tailored Multimodal Adhesion and Optical Reflectivity

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Supporting Information

ABSTRACT: Nature provides remarkable examples of mass-produced microscale particles with structures and chemistries optimized by evolution for particular functions. Synthetic chemical tailoring of such sustainable biogenic particles may be used to generate new multifunctional materials. Herein, we report a facile method for the development of bioenabled core/shell microparticles consisting of surface-modified ragweed pollen with a magnetic core, for which both multimodal adhesion and optical reflectivity can be tailored. Adhesion of the magnetic-core pollen can be tuned, relative to native pollen, through the combination of tailorable short-range interactions (over ~5 nm, via van der Waals forces and hydrogen bonding), an intermediate-range (over several μ m) capillary force, and long-range (over ~1 mm) magnetic attraction. The magnetic force could be controlled by the amount of iron oxide loaded within the core of the pollen particle, while the short-



range interactions and capillary force can be tuned by coating with polystyrene nanoparticles and/or a layer of viscous pollenkitt on the exine shell surface. Such coatings were also used to tailor the optical reflectance of the magnetic pollen particles; that is, the light-reflectance intensity was enhanced by coating with pollenkitt and significantly reduced by coating with polystyrene nanoparticles. This approach for generating multifunctional core/shell microparticles with tailorable adhesion and optical reflectivity may be extended to other pollen or biological particles or to synthetic biomimetic particles. Such independent control of the core and shell chemistries enabled by this approach also allows for the generation of microparticles with a variety of combination in functions tailorable to other properties.

INTRODUCTION

The control of particle adhesion on surfaces has long been of fundamental interest, as well as being crucial for a variety of technologies, including for paints,^{1,2} adhesives,^{3–5} electronics,^{6,7} photonics,⁸ and medical imaging/treatments.^{9–12} However, most experimental studies and models for studying adhesion are based on smooth spherical particles. There has been far less investigation to understand the adhesion of regular, non-spherical particles possessing surface features with well-controlled patterns. Indeed, the fabrication of microparticles with uniform, tailorable, three-dimensional (nonspherical) shapes, controlled surface topographies, and adjustable chemistries remains a nontrivial challenge.

Nature is a remarkable source of inspiration for the design of microscale particles with complex morphologies tailored for a variety of functions, including adhesion. For example, pollen particles have received increased interest in recent years as abundant, renewable, robust, adhesive biogenic microparticles with unique chemical and structural characteristics.^{13–16} The tough outer shell (exine) of pollen is composed predominantly

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of sporopollenin, a cross-linked polymer consisting of fatty acids and modest levels of oxygenated aromatic and phenylpropanoid residues.^{17,18} Pollen particles range in size from a few microns to hundreds of microns in nominal diameter, with a remarkable variety of exine structural patterns found among different species of pollen, including complex reticulated or echinate morphologies.^{19–21}

The known surface chemistry and available range of complex species-specific (reproducible) morphologies make pollen particles attractive as sustainable microscale templates for further chemical tailoring to affect adhesion or to enable new functions.^{22–29} Indeed, a number of groups have used coating or infiltration techniques to modify the chemistry of pollen for desired (bio)chemical, electrical, structural, or fluid dynamic properties.^{26–33} A recent example is the conformal, layer-by-layer coating of pollen with iron oxide, followed by organic

Received: July 21, 2015 Revised: September 16, 2015 pyrolysis and oxide crystallization, to yield magnetite replica particles with multimodal magnetic (long-range) and van der Waals (short-range) adhesive properties.^{28,29}

Concerns over the effects of airborne pollen particles on atmospheric optical properties and urban visibility, and the desire to develop sensitive and rapid methods for detecting such biological microparticles for public health monitoring, have led to significant interest in the light scattering properties of pollen.^{34–38} Surbek et al. have reported that elastic light scattering of individual pollen particles yielded scattering patterns that could be correlated to species-specific pollen morphologies (pollen grain shape, size, surface topography).^{36,39} However, the chemical tailoring of pollen particle to affect light scattering and reflectivity has not, as far as the present authors are aware, been reported.

The objective of this paper is to demonstrate how pollen particles can be transformed into multifunctional core/shell microparticles with both tailorable multimodal adhesion and optical reflectivity. The approach taken herein has been to selectively replace the pollen core with magnetic material while retaining the morphology and chemistry of the external sporopollenin shell. The external surface of the sporopollenin shell was then coated with other materials (nanoparticles or fluids) to further affect adhesion and/or to tailor optical reflectivity. Demonstrating such independent control of the chemistry of the pollen core and external surface would then provide a highly versatile means of tailoring such sustainable and readily available biological microparticles for a diversity of multiple functions combined.

EXPERIMENTAL SECTION

Pollen Preparation. Nondefatted native ragweed (Ambrosia artemisiifolia, NR) pollen was purchased from Greer Laboratories (Lenoir, NC) and stored at 4 °C. In order to obtain clean hollow pollen particles, a three-step procedure was applied to selectively remove the outer pollen coating (pollenkitt) and the contents of the pollen core. The nondefatted ragweed pollens were treated in an aqueous KOH (6 wt %) solution for 1 day followed by a concentrated H_3PO_4 (85 wt %) solution for 6 days, by using a rotational shaker (Barnstead Thermolyne Lab Quake Shaker Rotisserie). The pollen particles were then washed with DI water three times and each time isolated by centrifugation $(2.5 \times 1,000 \text{ rpm}, 15 \text{ min})$ in a Centra cl2 centrifuge (Thermo Scientific IEC). Finally, these base/acid-treated pollen particles were washed in a mixture of chloroform and methanol (3:1), which is known to be a solvent for external pollenkitt and other residual intracellular soluble organics (but not a solvent for the sporopollenin exine). The resulting hollow ragweed particles (designated as HR) were then dried overnight at 40 °C in a conventional oven.

Preparation of Magnetic Ragweed (MR) Pollen. The magnetic-core pollen particles were prepared via an in situ synthesis procedure reported for generating magnetite (Fe₃O₄) nanoparticles within the core of the hollow sporopollenin shell.⁴⁰ First, a Fe³⁺/Fe²⁺ (2:1) solution was prepared by mixing solutions of 8.11 g (50 mmol) of FeCl₃ (Alfa), 4.97 g (25 mmol) of FeCl₂·4H₂O (Alfa), 2.5 mL of 5 M HCl, 2.5 mL of ethanol, and 20 mL of DI-water in a vial, followed by heating at 40 °C until all iron salts were completely dissolved. Then ~0.2 g of HR particles were dispersed in the above Fe^{3+}/Fe^{2+} (2:1) solution and agitated using a rotational shaker overnight at room temperature. The particles were filtered and quickly washed with DIwater until the filtrate was clear and then transferred immediately into a 5 M ammonia solution to initiate precipitation of magnetite nanoparticles in the pollen core. The DI-water washing step was important to remove excess external Fe³⁺/Fe²⁺ (2:1) solution prior to precipitation of the iron oxide nanoparticles. After 2 h, the particles were isolated from the solution by filtration, washed with DI-water,

and then oven-dried at 60 °C overnight. The resulting dark-yellow magnetic ragweed pollen particles were quickly washed with an 1 M HCl solution and DI-water to remove any large iron oxide debris on the outside surfaces of the sporopollenin shells and oven-dried at 60 °C overnight. The magnetic-core ragweed pollen shells prepared as above were denoted as **MR**. In order to tune the volume of iron oxide in the **MR** pollen core, the same procedures were applied with the Fe³⁺/Fe²⁺ (2:1) solution diluted by a factor of 2 or a factor of 4. These latter magnetic ragweed pollen particles were then denoted as **MR1** or **MR2**, respectively.

Preparation of Magnetic-Core Pollen Shells Coated with Polystyrene Nanoparticles or Pollenkitt. About 50 mg of MR particles was dispersed in 10 mL of DI-water, to which 0.2 mL of a polystyrene (PS) nanoparticle dispersion (Alfa, 50 nm, 2.5 wt % in water) and 2 mL of tetrahydrofuran (THF) were added, followed by agitation using a rotational shaker for 15 min. The mixture was then washed with DI water and isolated by centrifugation ($5 \times 1,000$ rpm, 5 min, VWR Micro 1207) three times. These pollen particles were denoted as MR@PS.

The pollenkitt (PK) was extracted from nondefatted dandelion pollen (purchased from Greer Laboratories, Lenoir, NC) by exposure to a mixture of chloroform and methanol (3:1). After removal of the solvent, deep orange viscous pollenkitt was obtained. The pollenkitt was redissolved in methanol at three different concentrations of ~20, 40, and 80 mg/mL. MR particles were dispersed into each of the pollenkitt-methanol solutions overnight. A drop of each dispersion was then transferred to a glass slide and held at room temperature to allow for complete methanol evaporation. The samples were denoted as MR@PK, MR@PK1, and MR@PK2, with respect to the three concentrations of PK solutions used: 20, 40, and 80 mg/mL, respectively.

Preparation of Substrates for Adhesion Studies. Polystyrene (PS, M_w = 230,000, Sigma-Aldrich), polyvinyl acetate (PVAc, M_w = 50,000, Alfa Aesar), and poly(vinyl alcohol) (PVOH, $M_w = 89,000-$ 98,000, Sigma-Aldrich) were used in as-received states without further purification. The PS solution was prepared by dissolving 10% by mass in toluene, while 5% by mass PVAc and PVOH solutions were prepared in hexafluoroisopropanol (HFIP, TCI America). Polymeric test surfaces were prepared as thin polymer films on Piranha-etched (30/70 vol %H₂O₂/concentrated H₂SO₄ at 80 °C for 2 h) silicon substrates, by using a knife-edge coating technique described in detail elsewhere.⁴¹ Films were dried at room temperature for 24 h after coating and then annealed at 80 °C under vacuum for 2 h. The resulting films possessed thicknesses, as measured with interferometry, of approximately 1–2 μ m, which far exceeded the range of van der Waals interactions (~20 nm) and negated energetic effects of the underlying silicon substrate on the polymer-pollen interactions. A strongly magnetized ferromagnetic substrate, consisting of an axially poled neodymium-iron-boron alloy (permanent magnet, residual induction = 12,300 G) disk onto which a polished nickel foil (weakly ferromagnetic, 0.15 mm thick) was attached, was utilized in the longrange magnetic force measurements. The mean (R_a) and root-meansquare (rms) surface roughness values of each surface coating were obtained from topography scans of three random $10 \times 10 \ \mu m^2$ areas on each substrate surface using atomic force microscopy (AFM, Veeco Dimension 3100).

Adhesion Force Measurements. The adhesion force was measured using AFM. Tipless rectangular cantilevers with nominal spring constants of 1.2-6.4 N/m (Applied NanoStructures, Inc., Santa Clara, CA) were used. Single pollen particles were glued to the tipless cantilevers with a small amount of epoxy resin using a procedure described in detail elsewhere.¹³ The actual spring constants for the cantilevers with the attached pollen particles were determined directly by the thermal tune methods.^{42,43} A series of 20 force–distance curves were measured for each combination of pollen tip-polymer surface. For each pairing, three separate substrate surfaces were used, with three randomly chosen 1 cm \times 1 cm areas evaluated on each substrate, under a normal air condition (20 °C, humidity 30%). Three separate pollen AFM tips for each type of pollen particle sample were used, and the applied load during force measurements was 2.5 nN.

Scanning Electron Microscopy (SEM), Transmission Electron Microscopy (TEM), and Chemical Analyses. After all force measurements were completed, the pollen probe tips were sputtered with gold and then mounted on metal stubs using carbon tapes. The pollen probes were then characterized using a field emission gun scanning electron microscope (LEO 1530 Gemini FESEM, Carl Zeiss, Inc., Oberkochen, Germany) operating at an accelerating potential of 10.0 kV. The SEM was also equipped with an energy-dispersive X-ray spectroscopy analysis system (X-Max EDS, Oxford instruments, Abingdon, Oxford), which was used for the elemental analysis of the inorganic material within the pollen particle (operated with an 8.5 mm working distance at 10 kV). TEM analyses of magnetic core pollen particles were performed with a TECNAI F30 microscope (FEI, Oregon, USA), operated at 300 kV. TEM samples were prepared by first mixing the MR particles with epoxy (Epoxy Technology Inc., Billerica, MA) and then introducing the mixture into a copper tube (inner diameter = 2.2 mm, height = 2.5 mm). After curing of the epoxy at ~100 °C, the tube was ground into a disk with a thickness of about 100 μ m. The disk was then dimpled with a dimple grinder (Gatan Inc., Pleasanton, CA) so that the central thickness of the disk was about 20 μ m. Finally, the disk was ion milled (PIPS, Gatan Inc., Pleasanton, CA) to perforation. X-ray photoelectron spectroscopy (Thermo K-Alpha, Thermo Fischer Scientific, Inc., Waltham, MA, USA) was conducted using a monochromatic Al k α source (1486.6 eV), a spot size of 400 μ m, a pass energy of 200 eV, a step size of 0.5 eV, and a 50 ms dwell time.

Hyperspectral Analyses. Native pollen, as well as magnetic-core pollen particles, were analyzed for reflectance with or without the addition of pollenkitt or polystyrene nanoparticles. Both the wavelength and the intensity of the reflected spectra were recorded as a function of position (spatial mapping) on a given single pollen sample. The analyses were conducted with the PARISS Hyperspectral Imaging system (LightForm, Inc., Asheville, North Carolina, USA) and performed using a Nikon 80i microscope outfitted with a monochrome Retiga 2000R, CCD camera (QImaging, BC, Canada). Spectra were mapped under specular reflectance mode using a tungsten halogen white light source. Wavelength calibration was performed with a MIDL Hg⁺/Ar⁺ emission lamp (LightForm, Inc., NC, USA), and the accuracy was recorded and verified to be better than 2 nm. For relative comparison of spectral intensities, all acquisition parameters were kept the same except for exposure time. All spectra were then expressed relative to the lowest exposure time used across samples. All spectra from one individual pollen particle showing >99% closeness of fit were identified by one single representative spectrum. In all cases, all analyzed pollen samples showed a mapped area for the representative spectrum that was >90%, indicating strong homogeneity in the reflectance per treatment.

RESULTS AND DISCUSSION

Structural Characterization of Hollow and Magnetic Ragweed Pollen Particles. Figure 1a reveals a cross-sectional



Figure 1. Secondary electron images of (a) the hollow core of a hollow ragweed (HR) particle and (b) the solid iron oxide core of a magnetic ragweed (MR) particle (black arrow indicates the shell; red arrow indicates the core). The particles were sectioned with a thin, sharp blade.

view of the hollow core of an HR ragweed pollen particle. Magnetic-core ragweed pollen particles (MR) were obtained through salt diffusion into, followed by precipitation of magnetic iron oxide nanoparticles within, such a hollow pollen interior. The successful loading of magnetic iron oxide inside the hollow ragweed pollen particles was verified using SEM images (Figure 1b), EDS analyses (Table 1), and visual

Table 1. Average Elemental Compositions of the Pollen Shell of Hollow Ragweed (HR) and Magnetic Ragweed (MR) Particles and the Core of MR Particles, by EDS Analysis

	HR		MR			
	shell		shell		core	
element	wt%	at%	wt%	at%	wt%	at%
С	73	80	65	76	21	39
0	21	17	21	18	25	36
Ν	0.65	0.61	1.2	1.2	0.88	1.4
Na	0	0	0	0	1.1	1.1
Si	1.8	0.083	1.4	0.62	1.6	1.3
Cl	0	0	0.72	0.060	0.45	0.060
Fe	0	0	4.9	1.2	49	20
Р	1.6	0.66	3.0	1.4	0.88	0.65
S	1.5	0.60	2.8	1.2	0.66	0.47

observations of the migration of MR particles, when placed within water, toward a permanent magnet (Figure S1). The SEM images shown in Figures 2(a) and (b) reveal clean shell morphologies of both the base/acid-treated ragweed pollen, HR, and as-prepared magnetic pollen, MR. The compositions of the HR shell and the MR shell, obtained by EDS and XPS analyses, are shown in Table 1 and Table S1, respectively.

The HR particle shell consisted mainly of carbon and oxygen with no detectable iron. As expected, EDS analysis of the MR particle core revealed primarily iron and oxygen. However, a small amount of iron was also detected in the MR shell by EDS analyses, even though iron oxide particles were not clearly observed by SEM analyses of the external surface of the MR shell. Iron was also detected by XPS analyses of intact MR particles (Figure S3, Table S1), which indicated that such iron existed at or very near the external shell surface. STEM/EDS analyses were then obtained from an ion-milled cross-section of the MR shell. As shown in Figure 3, iron was detected at various locations through the MR shell, presumably due to the precipitation of iron oxide within the channels running through the shell. This small amount of iron oxide absorbed on/in the shell of MR particles played an important role on the adhesion behavior of MR particles with various substrates, as discussed in detail below.

It has been reported recently that sporopollenin can be coated by metal nanoparticles to allow for surface-enhanced Raman scattering and to generate lightweight materials with good conductivity.^{44,45} However, such *in situ* metal reductions on the sporopollenin biopolymer matrix usually result in irregular and modest metal nanoparticle coverage. In the present work, the **MR** particle surfaces were coated with a high loading of polystyrene (PS) latex nanoparticles (50 nm diameter) by the solvent-controlled heterocoagulation meth-od,⁴⁶ in order to allow for significant tuning of both adhesion and optical reflectance. The monodispersed PS nanoparticles were stabilized with anionic charges from surface sulfate groups,



Figure 2. Scanning electron micrographs of (a) hollow ragweed (**HR**), (b) magnetic ragweed (**MR**), (c) magnetic ragweed pollen coated with PS (**MR@PS**), and (d) magnetic ragweed pollen coated with PK (**MR@PK**). Low and high magnification images are indicated by '1' and '2', respectively.



Figure 3. (a) Transmission electron micrograph of an ion-milled cross-section of a magnetic ragweed (MR) particle placed on a Cu grid and (b)-(e) EDS analyses obtained at various indicated positions within the external shell.

and the shell surfaces of ragweed pollen particles (HR and MR) were also negatively charged according to the zeta-potential measurements (-58 ± 5 mV). Thus, electrostatic attraction was not the driving force for the heterocoagulation of the PS nanoparticles onto the pollen particles. However, THF addition to water swells the pollen particles and simultaneously induces

heteroaggregation of the PS and pollen particles by reducing the dielectric constant, and therefore the Debye length, controlling pollen-PS repulsion.⁴⁶ As shown in Figures 2c and S4, MR pollen particles were heavily coated by several dense layers of PS nanoparticles. However, the thickness and coverage of the PS nanoparticle coating was reduced at locations near the tips of spines (Figure S4a).

Native pollen particles are coated with an oily viscous liquid that resides on/within cavities in the exine wall.⁴⁷ This coating material, named pollenkitt (PK) by Knoll,⁴⁷ is especially prevalent in entomophilous plants and is comprised of a mixture of saturated and unsaturated lipids, and lesser amounts of carotenoids, flavonoids, proteins, and carbohydrates, and is of great importance in pollination ecology.^{48,49} For example, the major components of PK of the sunflower pollens (*Helianthus* annuus) are the triterpene and β -diketones.⁵⁰ The identification of components is quite sensitive to the method used for extraction and the analytical techniques chosen. For the pollenkitt used in this work, it was extracted from nondefatted dandelion pollen, and its major components were identified as polysaccharide (decomposes into butyrolactone and tetrahydro-2-furanol), fatty acids, and saturated hydrocarbon by GC-MS shown in Figures S5 and S6. Although the specific chemical compositions of pollenkitt from different plant pollens are varied, the general classes of molecules represented are relatively conserved, and the measured surface energy and wettabilities of PK from dandelion and sunflower pollens are well comparable.¹⁵

Pollen dispersal has been widely validated to be facilitated by the ability of PK to keep pollen particles together when dispersed and to promote pollen adhesion to animals.¹⁸ Previous work has shown that a significant enhancement in the adhesion of natural pollen to surfaces occurs through capillary interactions enabled by PK.15 Here, a facile solvent dispersion/evaporation method is demonstrated for recoating PK onto the shell surfaces of pollen particles to enable tuning of their capillary forces on different surfaces. The resulting PKcoated magnetic ragweed pollen (MR@PK) particles are displayed in Figures 2d and S7. The PK fully wetted the MR particle surfaces, with varied coating thicknesses that could be adjusted by controlling the concentration of the PK in the methanol solutions. The PK coating thicknesses were estimated as ~100-200 nm, 200-300 nm, and 400-500 nm for MR@ PK, MR@PK1, and MR@PK2 particles, respectively.

Selection and Preparation of Substrates and AFM Probes. Five different substrates were used to examine the effect of surface chemistry on the adhesion forces of each pollen particle. PS was chosen as a model apolar hydrocarbon substrate. PVOH and PVAc were chosen to represent organic materials with protic and basic characteristics, respectively. The polymer films used herein exhibited a range of water contact angles (θ_w) of 46°, 61°, and 101° for PVOH, PVAc, and PS, respectively. Piranha-etched silicon (Si) is a model inorganic, hydrophilic, and protic surface with $\theta_w \approx 14^\circ$. Piranha-treated Si is known to bear a thin oxide layer (2-10 nm thick)consisting of both oxygenated (SiO_x) and hydroxylated (SiOH)silicon. To provide a model metallic substrate, as well as to explore the magnetic force of attraction, a nickel-coated magnetic substrate (Ni–Nd) was also selected ($\theta_w \approx 80^\circ$). The rms surface roughnesses of all substrate surfaces (Table S2) fall within a range of 0.3 to 2.8 nm. Since the variation in roughness was orders of magnitude smaller than the scale of pollen surface microtopography (Figure 2), the effect of these

small differences in roughness was likely to be negligible relative to other effects observed herein.

The cantilevers with pollen particles attached are shown in Figure S8, taken after all force measurements were completed. The high magnification secondary electron images in this figure reveal the surface ornamentations of each cleaned pollen particle (HR and MR), MR@PS, and MR@PK. No obvious damage to pollen particle surfaces was observed after typical applied forces (2.5 nN, approximately the weight of a single pollen particle). As demonstrated previously, the pollen particles were not subjected to measurable deformation and did not exhibit compliance under the small loading forces used here.¹⁵

Adhesion of Magnetic-Core Ragweed Pollen. Figure 4(a) reveals the adhesion forces of HR, MR, and MR@PS



Figure 4. (a) Total adhesion forces between the hollow ragweed (HR), magnetic ragweed (MR), and magnetic ragweed pollen coated with PS (MR@PS) AFM probes and various substrates. Error bars are 95% confidence intervals. (b) Force-distance curves between magnetic ragweed particle (MR) and various substrate surfaces.

particles by contact-mode AFM measurements with five different substrates. The adhesion forces between **HR** and various substrate surfaces were independent of the wettabilities of the substrate surfaces. This is consistent with previous demonstrations that clean pollen adhesion is governed by short-range (<5 nm) VDW forces.^{13,15} In contrast, an interesting variation of the adhesion forces of **MR** particles with the five substrate surfaces was observed, in which the short-range forces decreased as the water wettabilities of the substrate decreased from Si to PS (Figure 4a). The representative force–distance curves are displayed in Figure

4b (for tip-to-surface distances below 2 μ m). The correlation of the short-range adhesion forces with the substrate wettability suggested that the VDW attraction was not the only shortrange interaction between the pollen spines and substrate surfaces; that is, other specific interactions, such as hydrogenbonding, may also be important. Hydrogen bonding may result from native -OH or iron oxide or $-NH_2$ groups on the pollen shell introduced during exposure to the ammonia solution. Control measurements of the adhesion forces between HR A (HR particles directly treated by 5 M ammonia solution) showed no apparent dependence of adhesion forces of HR A with water wettability of the surfaces (Figure S9). These results suggest the dependence of adhesion of MR particles with various surfaces could be due to the effect of iron oxide present on the external surface of the pollen shell. As noted earlier, a small amount of iron oxide was detected on/in the MR shell by XPS, SEM/EDS, and TEM/EDS (Figures 3 and S3, Table 1).

By using van Oss and Good's Lifshitz-van der Waals acidbase theory for the solid (**MR** surface, P)-solid (substrate surface, S) interface, the relationship of the short-range adhesion force with the surface energies and molecular structures of the polymers can be expressed as⁵¹

$$F_{ad} \propto \alpha \sqrt{\gamma_P^{\nu w} \gamma_S^{\nu w}} + b \sqrt{\gamma_P^+ \gamma_S^-} + c \sqrt{\gamma_P^- \gamma_S^+}$$
(1)

where F_{ad} is the experimentally determined adhesion force, and *a*, *b*, and *c* are coefficients scaling the VDW and acidic-basic contributions, respectively. Since the VDW components of the substrates $(6.3 < (\gamma_S^{\nu W})^{1/2} < 6.5)$ are similar and, in this model, the magnitude of the acid contribution from the substrates (γ_S^+) is negligible (Table S3), eq 1 can be simplified as follows:

$$F_{ad} \propto a + b \sqrt{\gamma_{\rm S}^{-}} \tag{2}$$

The adhesion force data was fitted to the form of eq 2, as shown in Figure 5. Fitting all five substrate surfaces led to a



Figure 5. Linear fitting of eq 2 to the short-range adhesion force data for magnetic ragweed (MR) particles on various substrates.

correlation coefficient, $R^2 \sim 0.96$, suggesting that the differences of total adhesion forces on the varied surfaces resulted from the basicity of the substrate surfaces. The effect of the basicity of the substrate surfaces was consistent with the polar interactions of iron oxides with substrate surfaces, where the oxygen groups (Si–O) on the substrate surfaces can act as the major electron donor components and form hydrogen

Ε

bonds and dipole–dipole interaction with the hydroxyl group of Fe–OH and the Fe ions of iron oxides on the pollen exine shell (after HCl rinse), respectively.

The short-range adhesion force associated with a given substrate surface was also significantly larger for MR@PS particles than for MR particles which, in turn, was larger than for HR particles (Figure 4a). For example, the adhesion forces between these three chemically treated pollen particles and PS surfaces were 63 ± 9 nN for MR@PS particles, 32 ± 3 nN for MR particles, and 18 ± 4 nN for HR particles. For interaction with a PS surface, the adhesion force was primarily governed by VDW interactions. Differences in the short-range adhesion force magnitudes for these three types of pollen particles interacting with the same substrate surface are likely to be due to differences in surface chemistry and structure. Differences in the surface chemistries of these three pollen particles should have altered their VDW components (γ_p^{VW}) , since the nonretarded Hamaker constants for the pollen shell (for HR particles), iron oxide (for MR particles), and PS (for MR@PS particles) are ~8.5 × 10^{-20} J, 21×10^{-20} J, and 6.58×10^{-20} J, respectively.^{15,52} The contact radii, R, estimated as the pollen ornamentation spike radius from SEM images, were found to be 45 ± 5 , 70 ± 5 , and 120 ± 10 nm for HR, MR, and MR@PS particles, respectively. The increased spine radius of MR was attributed to the presence of iron oxide on the shell surface (Table 1 and S1, Figures 3 and S3), while MR@PS particles displayed even larger effective spine radii due to the presence of PS nanoparticles on the end of spike tips (Figures S4a and S8c2).

The total adhesion force acting between MR particles and a Ni-Nd magnetic substrate is also plotted versus probe-distance from the substrate surface in Figure 4b (orange line). The total adhesion force between these particles and this substrate includes both the short-range VDW interaction and long-range magnetic interaction. The longer-range magnetic forces acting between particle/cantilever probes and the axially magnetized permanent magnet were measured with the probes located at two radial positions over the Ni-Nd substrate: at the center, and near the outer perimeter (edge), of the disk-shaped Ni-Nd substrate. As shown in Figure 4b (green diamonds labels), no appreciable magnetic attraction was detected between the MR particle and the Ni-Nd disk when the particle was positioned above the center location of the Ni-Nd disk, where the magnetic field was undetectable. However, a strong magnetic attraction was detected between the MR particle and Ni-Nd disk when the particle was located above the outer perimeter of disk, which is the position where the magnetic field intensity associated with this magnetized Ni-Nd substrate was the highest.²⁸ The magnetic force was detected at a vertical distance out to ~ 1 mm, with a decrease in this separation distance resulting in an increase in the magnitude of the magnetic force (Figure 3b, orange line). The total adhesion force between the MR particle and the Ni–Nd disk substrate (at the perimeter of the disk) was ~61 nN, with the short-range VDW and longerrange magnetic forces contributing ~33 nN and ~28 nN, respectively. In other words, the total adhesion force acting between the MR pollen particle and the magnetic edge of the Ni-Nd disk substrate was much larger than the total adhesion force experienced with the other four (nonmagnetic) substrates (~54 nN, ~47 nN, ~40 nN, and ~32 nN for Si, PVOH, PVAc, and PS, respectively).

The force acting on a magnetic particle inside a magnetic field can be expressed as 53

$$F = \frac{V \cdot \Delta \chi}{\mu_o} (B \cdot \nabla) B \tag{3}$$

where V is the volume of the magnetic material in the particle (m³), $\Delta \chi$ is the difference in magnetic susceptibilities between the particle and the surrounding medium (dimensionless), μ_{a} is the permeability of vacuum $(T \cdot m \cdot A^{-1})$, and B is the applied magnetic field (T). According to eq 3, the magnetic force depends on the volume of magnetic material present in the particle, V. Here we applied a facile method to adjust the volume of magnetic material in the core of the MR pollen particle, in order to tune the magnetic attraction of the MR pollen particle with the Ni-Nd disk; that is, the volume of the deposited iron oxide in the MR core was controlled by simply adjusting the concentration of the Fe^{3+}/Fe^{2+} solution (with the magnetic cores of the MR1 and MR2 particles generated with 1/2 and 1/4 of the iron concentration, respectively, as for the MR particles). The varied amounts of iron oxide deposited in the particle cores were estimated and confirmed by TGA analysis in Figure S10. As shown in Figure 6, the magnetic force decreased monotonically with the amount of iron oxide deposited in the particle cores.

Enhanced Adhesion of Magnetic and PK-Bearing Pollen. The adhesion mechanisms of natural pollen to manmade or natural surfaces are not fully understood, with the possible roles of a variety of surface characteristics (i.e.,



Figure 6. Long range magnetic forces between the magnetic perimeter of the Ni–Nd substrate and three different magnetic ragweed (MR) particles: (a) long-range magnetic force–distance curves and (b) magnetic force values obtained with varied volumes of iron oxide in the MR cores.

varied influences of surface structural and chemical factors) still requiring further experimental exploration. Nonetheless, the capillary force attributed to a viscous pollenkitt coating has proven to play a significant role in pollen adhesion.^{15,16} In this work, the combined influence of PK/capillary-based adhesion with short-range (i.e., VDW, hydrogen bonding) and longerrange magnetic adhesion has also been examined. Such multimodally adhesive particles were prepared by applying controlled amounts of PK to the shells of the magnetic ragweed particles (MR@PK; Figures 2d and S8d). The total adhesion forces between MR@PK particles with varied surfaces are displayed in Figure 7a, with typical force–distance curves for



Figure 7. (a) Total adhesion forces of hollow ragweed (HR), nondefatted (ND), magnetic ragweed (MR), and MR@PK with varied PK coating thicknesses on various substrate surfaces. Error bars are 95% confidence intervals. (b) Force-distance curves between MR@PK1 and magnetic surfaces.

MR@**PK** particles with the magnetic Ni–Nd substrate (at the center or perimeter locations) plotted in Figure 7b (The small undulation of the AFM forces is due to an optical effect during the probe approach and retraction.). The total adhesion forces of the **MR**@**PK** particles were greater by a factor of ~2–30 relative to **HR** and **MR** particles, depending on the thickness of the PK coating. This enhancement was caused by the capillary interaction of PK between **MR**@**PK** particle and substrate, which operated over the intermediate range of a few microns, as seen in Figures 7b and S11. The PK-enabled capillary force was also found to decrease in the substrate order Si > PVOH >

PVAc > PS, which was consistent with a monotonic decrease in the contact angle of PK in this substrate order.

To evaluate the role of capillary meniscus forces on the pulloff force for each substrate, a capillary force model was developed to describe the adhesive force of a liquid bridge between a sphere and a flat surface. Upon retraction, the adhesive force of a liquid bridge can have both a capillary contribution (due to the surface tension and LaPlace pressure across the curved meniscus), as well as an adhesive force upon retraction due to the viscous resistance upon capillary thinning. For a liquid bridge with constant volume, V, the capillary meniscus force at constant volume can be expressed as⁵⁴

$$F_{c} = 4\pi\gamma cR \left(1 - \frac{D}{\sqrt{\frac{V}{\pi R} + D^{2}}}\right)$$
(4)

$$c = \frac{\cos(\theta_1 + \beta) + \cos\theta_2}{2} \tag{5}$$

where γ is the surface tension of the liquid (0.045 N m⁻¹ for PK at 30% RH¹⁵), *R* is the sphere radius, β is the filling angle, θ_1 and θ_2 are the contact angles of liquid to sphere and to the plane, and *D* is the distance between the sphere and the planar surface. *V* was assumed to be constant, since drops of PK do not evaporate in air. The filling angle β was estimated to be $\sim 10-40^{\circ}$, which was dependent on the amount of PK.

Using both c and V as fitting parameters, the green dashed fitting lines were generated in Figures 7 and S11. These fitted lines agreed well with the experimental retraction results. The fitting results for the values of c and V are listed in Table S4. From the fitting, the volumes of the PK bridges between MR@ PK1 and various substrates decreased from Si to PVOH to PVAc to PS to Ni-Nd, which was consistent with decreased PK wetting in the same substrate order (as determined by contact angle measurements of these substrates with PK, Table S2). This trend in fitted capillary volume values was also consistent with the longer observed range of capillary forcebased adhesion on the Si surface (from $\sim 100 \text{ nm to} \sim 800 \text{ nm}$) relative to the Ni–Nd surface (from ~ 100 nm to ~ 400 nm), as shown in Figures 7 and S11. In the fitting procedure above, the contribution of the viscous resistance (hydrodynamic force) to capillary thinning was neglected, since the magnitude of this force contribution was estimated to be several orders of magnitude smaller than the capillary contributions resulting from surface tension and LaPlace pressure (as discussed in the Supporting Information).

The coated thickness of PK was controlled by simply adjusting the PK concentration in the coating solution. The thicknesses of PK coatings on the magnetic pollen particles were estimated by AFM to be ~100–200 nm, 200–300 nm, and 400–500 nm for MR@PK, MR@PK1, and MR@PK2 particles, respectively. The magnitude of the adhesion forces of MR@PK particles on the Si surface increased with the thickness of PK coating, from ~110 nN for 100–200 nm coatings to ~750 nN for 400–500 nm coatings (Figure 7a). The increased thickness of the PK coating resulted in the formation of PK capillary bridges with larger volumes. The resulting higher filling angle β of the PK bridge also led to a higher capillary force.¹⁵ The volumes of the PK bridges, obtained through fitting of the force–distance curves (Figure S12) with eq 4, for each MR@PK sample are listed in Table S7.

For the magnetic Ni-Nd surface, the additive effects of both the capillary force and magnetic force contributions were also observed. The magnetic force was, of course, independent of the application of a PK coating and was the same for all the magnetic pollen particles (~30 nN). With no PK or at low PK coating thicknesses, the magnetic force was an important component of the total adhesion force. For example, for MR@ PK particles, the total adhesion force at the perimeter location of the Ni–Nd substrate (~100 nN) acting over a short distance $(\sim 10 \text{ nm})$ consisted predominantly of the sum of the capillary force (\sim 70 nN) and the magnetic force (\sim 30 nN). It was comparable with the total adhesion of MR@PK particles on Si $(\sim 110 \text{ nN})$, consisting of the sum of short-range and capillary force, and was larger than the total adhesion forces on PVOH (~95 nN), PVAc (~86 nN), and PS (~75 nN). However, with PK coatings of increasing thickness, the magnetic force contribution became much less important (e.g., the capillary force increased to \sim 500 nN for MR@PK2 particles).

Tuning Optical Properties via Tailored Coatings. The light scattering behavior of pollen has been found to be correlated with morphological characteristics (shape, size, surface topography) of such biogenic particles.^{36,39} In the present work, hyperspectral reflectance imaging was used to evaluate the reflectivities of ragweed pollen particles with varied surface treatments (Figure 8). It is notable that the light



Figure 8. Hyperspectral reflectance spectra of ragweed pollen particles: native nondefatted (NR), treated hollow (HR), and magnetic-core (MR, MR@PS, and MR@PK) pollen.

reflectance intensities of native ragweed pollen (NR) and PKbearing magnetic-core pollen (MR@PK) were much higher than for the hollow cleaned HR pollens lacking PK on the shell surface. In addition, magnetite cores alone with no surface treatment (MR) increased optical reflectance to a value in between NR and MR@PK. On the other hand, the magnetitecore pollen coated with PS nanoparticles (MR@PS) displayed significantly reduced light reflection intensity, lower than all others. SEM images indicated that there were no appreciable differences in the sizes and shapes of the different base pollen particles. In the absence of such morphological differences, the influence of the refractive indices of materials on the shell surface and within the particle core should be considered. The refractive indices of PS and magnetite are ~1.60 and 2.42, respectively.^{55,56} According to the standard Cargille Refractive Index Liquids (Cargille Laboratories, Inc.), the refractive indices of ragweed pollen and PK used in this work were $\sim 1.75 \pm 0.50$ and $\sim 1.90 \pm 0.10$, respectively. The observed relative values in reflectance intensities are consistent with these refractive indices if one takes into consideration the surface and internal microstructure of the sporopollenin shell. For example, using NR as a reference, we note that the highest reflectances occurred for samples containing high index magnetite cores, MR and MR@PK. These cores provide internal reflective interfaces with air or sporopollenin, due to the large with large differences in refractive index. The PK coating likely enhances reflectance of MR@PK above MR by filling outer surface defects on the pollen shell (reducing scattering) and allowing more light transmission to and from the high index magnetite core. Without the PK coating that NR possesses, acid-cleaned HR particles show significantly reduced reflectance. The acid cleaning process also likely opens up internal pore spaces within the sporopollenin, leading to diffuse scattering within the shell microstructure. Not only does PS have the lowest refractive index among these materials, but the roughened surface of MR@PS may decrease transmission to and from the internal magnetite core interface.

CONCLUSIONS

A facile strategy for transforming native pollen into a multifunctional adhesive particle with tunable short-, medium-, and long-range adhesion modes, as well as tailored optical reflectivity, is demonstrated. The external pollen shape and size were preserved after base/acid treatment to selectively remove the internal pollen core, followed by the infiltration and precipitation of magnetic iron oxide within the pollen core. While the adhesion of cleaned ragweed pollen on all substrates examined was governed by short-range VDW forces (~20-30 nN), the adhesion of magnetic-core pollen particles to a magnetic substrate was significantly enhanced with the addition of a long-range magnetic force (\sim 30 nN, out to \sim 1 mm). The addition of an exterior pollenkitt coating contributed an additional significant medium-range capillary force (up to ~800 nN, 5–2,000 nm), and the addition of a PS nanoparticle coating increased the contact area that directly affected the short-range VDW forces. The light reflective properties of pollens or related particles can be adjusted by decoratively coating a layer of low or high refractive index material, such as nanoparticle-based film or a liquid film, respectively. The pollenkitt coatings, which possessed an enhanced refractive index relative to sporopollenin, yielded a strong enhancement in light reflectivity. On the other hand, the coating of pollen with polystyrene nanoparticles led to a significant reduction in reflectivity. The present work demonstrates how chemical tailoring of the core and shell of pollen can yield particles with tailorable multimodal adhesion and optical reflectivity. Such independent control of the chemistry of the pollen core and external surface provides a highly versatile means of tailoring the properties of such sustainable and readily available biological microparticles. Indeed, this general approach may be extended to generate pollen-derived (or other biologically derived) microparticles with a wide variety of other chemistries and functions (i.e., beyond tailored adhesion and optical reflectivity).

ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.chemma-ter.5b02782.

XPS spectra and elemental compositions of the pollen shells of HR and MR particles, surface tension values of each substrate, GC-MS profiles of compositions of PK, fitting parameters of Figures S10 and S11, photos revealing the attraction of MR particles toward a permanent magnet, high magnification SEM images of MR@PS and of MR@PK, adhesion force data for HR and HR_A, TGA curves of each samples, typical AFM adhesion and calculated force-distance curves of MR@ PK particles on each substrate, and the experimental and calculated force-curves of MR@PK particles on Si with different PK coatings (PDF)

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Notes

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■ ABBREVIATIONS

VDW, van der Waals; NR, nondefatted native ragweed pollen; HR, hollow clean ragweed pollen; MR, magnetic-core ragweed pollen; Si, piranha-treated silicon wafer; PS, polystyrene; PVOH, poly(vinyl alcohol); PVAc, polyvinyl acetate; Ni–Nd, permanent magnet neodymium–iron–boron alloy; PK, pollenkitt; AFM, atomic force microscopy; SEM, scanning electron microscopy; TEM, transmission electron microscopy; EDS, energy-dispersive X-ray spectroscopy; XPS, X-ray photoelectron spectroscopy; TGA, thermal-gravity analysis

REFERENCES

(1) Zimon, A. D. Adhesion of Dust and Powder; Springer US: 1982; pp 241–270.

(2) Tadros, T. F. Colloids in Paints: Colloids and Interface Science; Wiley-VCH: 2010.

(3) Rimai, D. S.; Sharpe, L. H. Advances in Particle Adhesion; Gordon and Breach: Adhesion Society Meeting, 1996.

(4) Shu, S.; Husain, S.; Koros, W. J. A general strategy for adhesion enhancement in polymeric composites by formation of nanostructured particle surfaces. *J. Phys. Chem. C* **2007**, *111*, 652–657.

(5) Huang, Y.; Liu, M. J.; Wang, J. X.; Zhou, J. M.; Wang, L. B.; Song, Y. L.; Jiang, L. Controllable Underwater Oil-Adhesion-Interface Films Assembled from Nonspherical Particles. *Adv. Funct. Mater.* **2011**, *21*, 4436–4441.

(6) Xia, Y.; Zhang, W. K.; Xiao, Z.; Huang, H.; Zeng, H. J.; Chen, X. R.; Chen, F.; Gan, Y. P.; Tao, X. Y. Biotemplated fabrication of hierarchically porous NiO/C composite from lotus pollen grains for lithium-ion batteries. *J. Mater. Chem.* **2012**, *22*, 9209–9215.

(7) Stassi, S.; Canavese, G. Spiky nanostructured metal particles as filler of polymeric composites showing tunable electrical conductivity. *J. Polym. Sci., Part B: Polym. Phys.* **2012**, *50*, 984–992.

(8) Deleuze, C.; Delville, M. H.; Pellerin, V.; Derail, C.; Billon, L. Hybrid Core@Soft Shell Particles as Adhesive Elementary Building Blocks for Colloidal Crystals. *Macromolecules* **2009**, *42*, 5303–5309. (9) Lorch, M.; Thomasson, M. J.; Diego-Taboada, A.; Barrier, S.; Atkin, S. L.; Mackenzie, G.; Archibald, S. J. MRI contrast agent delivery using spore capsules: controlled release in blood plasma. *Chem. Commun.* **2009**, 6442–6444.

(10) Barrier, S.; Diego-Taboada, A.; Thomasson, M. J.; Madden, L.; Pointon, J. C.; Wadhawan, J. D.; Beckett, S. T.; Atkin, S. L.; Mackenzie, G. Viability of plant spore exine capsules for microencapsulation. J. Mater. Chem. **2011**, 21, 975–981.

(11) Dong, W.; Li, Y.; Niu, D.; Ma, Z.; Gu, J.; Chen, Y.; Zhao, W.; Liu, X.; Liu, C.; Shi, J. Facile Synthesis of Monodisperse Superparamagnetic Fe3O4 Core@hybrid@Au Shell Nanocomposite for Bimodal Imaging and Photothermal Therapy. *Adv. Mater.* **2011**, *23*, 5392–5397.

(12) Smolensky, E. D.; Neary, M. C.; Zhou, Y.; Berquo, T. S.; Pierre, V. C. Fe3O4@organic@Au: core-shell nanocomposites with high saturation magnetisation as magnetoplasmonic MRI contrast agents. *Chem. Commun.* **2011**, *47*, 2149–2151.

(13) Thio, B. J. R.; Lee, J. H.; Meredith, J. C. Characterization of Ragweed Pollen Adhesion to Polyamides and Polystyrene Using Atomic Force Microscopy. *Environ. Sci. Technol.* **2009**, *43*, 4308–4313. (14) Lee, J.-H.; Suttle, B. M.; Kim, H.-J.; Meredith, J. C. Pollen: A Novel, Biorenewable Filler for Polymer Composites. *Macromol. Mater. Eng.* **2011**, *296*, 1055–1062.

(15) Lin, H.; Gomez, I.; Meredith, J. C. Pollenkitt Wetting Mechanism Enables Species-Specific Tunable Pollen Adhesion. *Langmuir* **2013**, *29*, 3012–3023.

(16) Lin, H.; Lizarraga, L.; Bottomley, L. A.; Meredith, J. C. Effect of water absorption on pollen adhesion. *J. Colloid Interface Sci.* **2015**, *442*, 133–139.

(17) Blackmore, S.; Wortley, A. H.; Skvarla, J. J.; Rowley, J. R. Pollen wall development in flowering plants. *New Phytol.* **200**7, *174*, 483–498.

(18) Pacini, E.; Hesse, M. Pollenkitt – its composition, forms and functions. *Flora* **2005**, *200*, 399–415.

(19) Nowicke, J. W.; Takahashi, M.; Webster, G. L. Pollen morphology, exine structure and systematics of Acalyphoideae (Euphorbiaceae) - Part 1. Tribes Clutieae (Clutia), Pogonophoreae (Pogonophora), Chaetocarpeae (Chaetocarpus, Trigonopleura), Pereae (Pera), Cheiloseae (Cheilosa, Neoscortechinia), Erismantheae pro parte (Erismanthus, Moultonianthus), Dicoelieae (Dicoelia), Galearieae (Galearia, Microdesmis, Panda) and Ampereae (Amperea, Monotaxis). *Rev. Palaeobot.Palynol.* **1998**, *102*, 115–152.

(20) Nowicke, J. W.; Takahashi, M.; Webster, G. L. Pollen morphology, exine structure and systematics of Acalyphoideae (Euphorbiaceae) - Part 2. Tribes Agrostistachydeae (Agrostistachys, Pseudagrostistachys, Cyttaranthus, Chondrostylis), Chrozophoreae (Speranskia, Caperonia, Philyra, Ditaxis, Argythamnia, Chiropetalum, Doryxylon, Sumbaviopsis, Thyrsanthera, Melanolepis, Chrozophora), Caryodendreae (Caryodendron, Discoglypremna, Alchorneopsis), Bernardieae (Bernardia, Necepsia, Paranecepsia, Discocleidion, Adenophaedra) and Pycnocomeae (Pycnocoma, Drocelonica, Argomuellera, Blumeodendron, Podadenia, Ptychopyxis, Botryophora). *Rev. Palaeobot. Palynol.* **1999**, *105*, 1–62.

(21) Scott, R. J. Pollen Exine - the Sporopollenin Enigma and the Physics of Pattern. Soc. Exp. Biol. Semin. S. 1994, 55, 49–81.

(22) Wang, Y.; Liu, Z.; Han, B.; Huang, Y.; Yang, G. Carbon Microspheres with Supported Silver Nanoparticles Prepared from Pollen Grains. *Langmuir* **2005**, *21*, 10846–10849.

(23) Murphy, D. J. The extracellular pollen coat in members of the Brassicaceae: composition, biosynthesis, and functions in pollination. *Protoplasma* **2006**, *228*, 31–39.

(24) Hesse, M. Biological Adhesive Systems: Chap 1. Bonding Single Pollen Grains Together: How and Why?; Springer Wien New York: Austria, 2010.

(25) Hall, S. R.; Swinerd, V. M.; Newby, F. N.; Collins, A. M.; Mann, S. Fabrication of porous titania (brookite) microparticles with complex morphology by sol-gel replication of pollen grains. *Chem. Mater.* **2006**, *18*, 598–600.

(26) Song, F.; Su, H. L.; Chen, J. J.; Moon, W. J.; Lau, W. M.; Zhang, D. 3D hierarchical porous SnO2 derived from self-assembled biological systems for superior gas sensing application. *J. Mater. Chem.* **2012**, *22*, 1121–1126.

(27) Song, F.; Su, H. L.; Han, J.; Lau, W. M.; Moon, W. J.; Zhang, D. Bioinspired Hierarchical Tin Oxide Scaffolds for Enhanced Gas Sensing Properties. J. Phys. Chem. C 2012, 116, 10274–10281.

(28) Goodwin, W. B.; Gomez, I. J.; Fang, Y.; Meredith, J. C.; Sandhage, K. H. Conversion of Pollen Particles into Three-Dimensional Ceramic Replicas Tailored for Multimodal Adhesion. *Chem. Mater.* **2013**, *25*, 4529–4536.

(29) Gomez, I. J.; Goodwin, W. B.; Sabo, D.; Zhang, Z. J.; Sandhage, K. H.; Meredith, J. C. Three-dimensional magnetite replicas of pollen particles with tailorable and predictable multimodal adhesion. *J. Mater. Chem. C* 2015, *3*, 632–643.

(30) Hall, S. R.; Bolger, H.; Mann, S. Morphosynthesis of complex inorganic forms using pollen grain templates. *Chem. Commun.* 2003, 2784–2785.

(31) Cao, F.; Li, D. X. Morphology-controlled synthesis of SiO2 hollow microspheres using pollen grain as a biotemplate. *Biomed. Mater.* **2009**, *4*, 025009.

(32) Wang, Y.; Liu, Z. M.; Han, B. X.; Sun, Z. Y.; Du, J. M.; Zhang, J. L.; Jiang, T.; Wu, W. Z.; Miao, Z. J. Replication of biological organizations through a supercritical fluid route. *Chem. Commun.* **2005**, 2948–2950.

(33) Thio, B. J. R.; Clark, K. K.; Keller, A. A. Magnetic pollen grains as sorbents for facile removal of organic pollutants in aqueous media. *J. Hazard. Mater.* **2011**, *194*, 53–61.

(34) Hinds, W. C. Aerosol Technology: Properties, Behavior, and Measurement of Airborne Particles; Wiley-Interscience: New York, 1999.

(35) Taylor, P. E.; Flagan, R. C.; Valenta, R.; Glovsky, M. M. Release of allergens as respirable aerosols: A link between grass pollen and asthma. *J. Allergy Clin. Immunol.* **2002**, *109*, 51–56.

(36) Surbek, M.; Esen, C.; Schweiger, G. Elastic Light Scattering on Single Pollen: Scattering in a Large Space Angle. *Aerosol Sci. Technol.* **2009**, 43, 679–684.

(37) Hill, S. C.; Pinnick, R. G.; Niles, S.; Fell, N. F.; Pan, Y. L.; Bottiger, J.; Bronk, B. V.; Holler, S.; Chang, R. K. Fluorescence from airborne microparticles: dependence on size, concentration of fluorophores, and illumination intensity. *Appl. Opt.* **2001**, *40*, 3005–3013.

(38) Pan, Y. L.; Aptowicz, K. B.; Chang, R. K.; Hart, M.; Eversole, J. D. Characterizing and monitoring respiratory aerosols by light scattering. *Opt. Lett.* **2003**, *28*, 589–591.

(39) Surbek, M.; Esen, C.; Schweiger, G.; Ostendorf, A. Pollen characterization and identification by elastically scattered light. *J. Biophotonics* **2011**, *4*, 49–56.

(40) Paunov, V. N.; Mackenzie, G.; Stoyanov, S. D. Sporopollenin micro-reactors for in-situ preparation, encapsulation and targeted delivery of active components. *J. Mater. Chem.* **2007**, *17*, 609–612.

(41) Meredith, J. C.; Karim, A.; Amis, E. J. High-throughput measurement of polymer blend phase behavior. *Macromolecules* **2000**, 33, 5760–5762.

(42) Burnham, N. A.; Chen, X.; Hodges, C. S.; Matei, G. A.; Thoreson, E. J.; Roberts, C. J.; Davies, M. C.; Tendler, S. J. B. Comparison of calibration methods for atomic-force microscopy cantilevers. *Nanotechnology* **2003**, *14*, 1–6.

(43) Hutter, J. L.; Bechhoefer, J. Calibration of Atomic-Force Microscope Tips. *Rev. Sci. Instrum.* **1993**, *64*, 1868–1873.

(44) Joseph, V.; Schulte, F.; Rooch, H.; Feldmann, I.; Dorfel, I.; Osterle, W.; Panne, U.; Kneipp, J. Surface-enhanced Raman scattering with silver nanostructures generated in situ in a sporopollenin biopolymer matrix. *Chem. Commun.* **2011**, 47, 3236–3238.

(45) Xu, L. N.; Zhou, K. C.; Xu, H. F.; Zhang, H. Q.; Huang, L.; Liao, J. H.; Xu, A. Q.; Gu, N.; Shen, H. Y.; Liu, J. Z. Copper thin coating deposition on natural pollen particles. *Appl. Surf. Sci.* **2001**, *183*, 58–61.

(46) Lee, J. H.; Mahmoud, M. A.; Sitterle, V.; Sitterle, J.; Meredith, J. C. Facile Preparation of Highly-Scattering Metal Nanoparticle-Coated

Polymer Microbeads and Their Surface Plasmon Resonance. J. Am. Chem. Soc. 2009, 131, 5048-5049.

(47) Knoll, F. R. Pollen-cement and method of pollination. Zeit Schr Bot [Festschr Oltmanns] 1930, 23.

(48) Howlett, B. J.; Knox, R. B.; Heslop-Harrison, J. Pollen-wall proteins: release of the allergen Antigen E from intine and exine sites in pollen grains of ragweed and Cosmos. *J. Cell Sci.* **1973**, *13*, 603–619.

(49) Dominguez, E.; Mercado, J. A.; Quesada, M. A.; Heredia, A. Pollen sporopollenin: degradation and structural elucidation. *Sex. Plant Reprod.* **1999**, *12*, 171–178.

(50) Schulz, S.; Arsene, C.; Tauber, M.; McNeil, J. N. Composition of lipids from sun-flower pollen (Helianthus annuus). *Phytochemistry* **2000**, *54*, 325–336.

(51) Good, R. J.; van Oss, C. J. Modern Approach of Wettability: Theory and Applications; Plenum Press: New York, 1991.

(52) Pallab, G. Colloid And Interface Science; Asoke K. Ghosh, PHI Learing Private Limited: New Delhi, 2009.

(53) Pamme, N. Magnetism and microfluidics. *Lab Chip* 2006, 6, 24–38.

(54) Butt, H. J.; Kappl, M. Normal capillary forces. Adv. Colloid Interface Sci. 2009, 146, 48–60.

(55) Marx, E.; Mulholland, G. W. Size and Refractive-Index Determination of Single Polystyrene Spheres. J. Res. Natl. Bur. Stand. **1983**, 88, 321–338.

(56) Patnaik, P. Handbook of Inorganic Chemicals; McGraw-Hill: NewYork, 2002.