Published in final edited form as:

ACS Appl Mater Interfaces. 2018 July 5; 10(26): 22718–22730. doi:10.1021/acsami.8b05856.

Compressible, Thermally Insulating, and Fire Retardant Aerogels through Self-Assembling Silk Fibroin Biopolymers Inside a Silica Structure—An Approach towards 3D Printing of Aerogels

Hajar Maleki^{*,iD}, Susan Montes, Nastaran Hayati-Roodbari, Florian Putz, and Nicola Huesing^{iD}

Chemistry and Physics of Materials, Paris-Lodron University Salzburg, Jakob-Haringer-Strasse 2a, 5020 Salzburg, Austria

Abstract

Thanks to the exceptional materials properties of silica aerogels, this fascinating highly porous material has found high-performance and real-life applications in various modern industries. However, a requirement for a broadening of these applications is based on the further improvement of the aerogel properties, especially with regard to mechanical strength and postsynthesis processability with minimum compromise to the other physical properties. Here, we report an entirely novel, simple, and aqueous-based synthesis approach to prepare mechanically robust aerogel hybrids by cogelation of silk fibroin (SF) biopolymer extracted from silkworm cocoons. The synthesis is based on sequential processes of acid catalyzed (physical) cross-linking of the SF biopolymer and simultaneous polycondensation of tetramethylorthosilicate (TMOS) in the presence of 5-(trimethoxysilyl)pentanoic acid (TMSPA) as a coupling agent and subsequent solvent exchange and supercritical drying. Extensive characterization by solid-state ^1H NMR, ^{29}Si NMR, and 2D ^1H – ^{29}Si heteronuclear correlation (HETCOR) MAS NMR spectroscopy as well as various microscopic techniques (SEM, TEM) and mechanical assessment confirmed the molecular-level homogeneity of the hybrid nanostructure. The developed silica–SF aerogel hybrids contained an improved set of material properties, such as low density ($\rho_{\text{b,average}} = 0.11\text{--}0.2\text{ g cm}^{-3}$), high porosity ($\sim 90\%$), high specific surface area ($\sim 400\text{--}800\text{ m}^2\text{ g}^{-1}$), and excellent flexibility in compression (up to 80% of strain) with three orders of magnitude improvement in the Young's modulus over that of pristine silica aerogels. In addition, the silica–SF hybrid aerogels are fire retardant and demonstrated excellent thermal insulation performance with thermal conductivities (λ) of $0.033\text{--}0.039\text{ W m}^{-1}\text{ K}^{-1}$. As a further advantage, the formulated hybrid silica–SF aerogel showed an excellent printability in the wet state using a microextrusion-based 3D printing approach. The printed structures had comparable properties to their monolith counterparts, improving postsynthesis processing or shaping of the silica aerogels significantly. Finally, the hybrid silica–SF aerogels reported here represent significant progress for a mechanically

iDORCID

Hajar Maleki: 0000-0002-2813-4700

Nicola Huesing: 0000-0003-2274-9779

^{*}Corresponding Author: hajar.maleki@sbg.ac.at.

Notes

The authors declare no competing financial interest.

customized and robust aerogel for multipurpose applications, namely, as a customized thermal insulation material or as a dual porous open-cell biomaterial used in regenerative medicine.

Keywords

silica; silk fibroin; aerogel; hybrid; 3D printing; thermal insulation

1 Introduction

Aerogels, discovered for the first time by Kistler in 1930,^{1,2} are a striking example for materials with a fascinating combination of properties such as high porosity (90–99%) comprising micro- and mesoporosity ($2 < \text{pore diameter } (\varphi) < 50 \text{ nm}$), low densities ($0.001\text{--}0.2 \text{ g cm}^{-3}$), and high specific surface areas ($500\text{--}1500 \text{ m}^2 \text{ g}^{-1}$), sometimes together with transparency and high heat insulation capabilities. They are typically prepared by sol–gel processes accompanied by an appropriate drying technique (usually supercritical drying),^{3,4} rendering them appropriate candidates for a multitude of applications such as for thermal insulation,⁵ as catalysts and catalyst supports,⁶ as pollutants sorbent for environmental cleaning,^{7,8} or in biomedical and pharmaceutical applications.^{9,10}

Commercially available silica aerogels are being developed in different forms of blankets and granules and commercialized with a global market of ~250 million €. However, research on silica aerogels continues, as the mechanical strength of native silica aerogels is still suboptimal, and a further improvement of the mechanical stability with low or no sacrifice to the other physical properties is necessary.^{4,12–14} The extension of network connectivity in the silica skeleton through cross-linking with various polymeric networks, such as polyurethane, epoxide, and polystyrene, to name only a few, results in mechanically more robust materials that are suitable for most load-bearing applications.^{14–18} However, the bottlenecks with regard to compromises in the set of the physical properties as well as the multistep, non-green tedious processing approaches are still problematic.

Currently, oil-free precursors/biopolymers from natural, renewable resources are drawing tremendous attention regarding their potential in the fabrication of various functional materials with a lower environmental impact.¹¹ In recent years, the aerogel community has devoted large efforts in the preparation of bio/aerogel composites with biopolymers from various sustainable resources or biomasses,¹⁹ e.g., from polysaccharides,^{20–27} proteins,²⁸ and nucleic acids²⁹ either as pure aerogel or as composites with the other components, i.e., silica and/or silsesquioxanes,^{27,30,31} in particular for applications as thermal super-insulation lightweight aerogels.^{11,27,30}

Polysaccharide-based polymers, especially cellulose micro- and nanofibers (CNF), are extensively explored for the hybridization with porous silica structures.¹¹ The utilization of proteins, i.e., silk fibroin (SF), for mechanical reinforcement purposes has not been investigated so far.

Recently, our group has started to exploit SF biopolymers, harvested from *Bombyx mori* silkworm cocoons, as another attractive biopolymer to reinforce the poor mechanical

strength in both silica- and polymethylsilsequioxane-based aerogels. Silk fibroin (SF) has a fibrous structure with numerous advantages like abundance in nature, low cost, versatility in processing, together with a very well-studied surface chemistry.³² In recent years, SF has been widely explored for the fabrication of different resilient bio/materials for a multitude of applications, namely, as cell-based scaffolds used in regenerative medicine or drug delivery research.^{32,33} Silk is advantageous from an economic point of view for the fabrication of functional materials as it is well known from textile industries, and therefore, nearly 1000 tons of silk are produced and processed annually.³⁴ A regenerated aqueous solution of SF can readily be assembled, e.g., by changing the pH, to various stable materials e.g., hydrogels³² with remarkable mechanical strength. However, aside from very few examples in the literature,^{28,35} the preparation of silk fibroin aerogels has not been explored thoroughly, especially for a system in which SF acts as a secondary phase/template to mechanically support the delicate oxide-based gels.

3D printing, which is known as "rapid prototyping", is making a big revolution in the field of materials manufacturing as this process can reduce the time required for producing and processing highly customized materials construct with complex geometry.³⁶ 3D printing of aerogels is not matured but rather very challenging, because the shaping of a well-defined 3D construct with 3D printing requires a printable sol or gel with suitable viscosity and mechanical strength. The 3D printing of pristine silica aerogels could overcome the post-synthesis processing bottlenecks due to the lack of machinability in its fragile dried state. Only a few examples on graphene,³⁷ nano-crystalline cellulose,³⁸ and diacrylate cross-linked silica aerogels³⁹ have been reported so far that all have shown promising features in terms of printing feasibility and customizable design, which motivate devoting effort to research in this direction with expanding aerogel's applicability.

In this work, we have shown for the first time that silk fibroin can be used as an appropriate biopolymer to improve the compressive strength and flexibility of pristine silica aerogels. A homogeneous SF polymer/silica hybrid structure has been synthesized by a simple, one-step acid catalyzed sol–gel reaction in the presence of a phase separation suppressing agent. In order to form stable covalent linkages between silica and the secondary silk fibroin structure at a molecular scale, a carboxylic acid functionalized silane coupling agent as coprecursor has been used. To be clear, the synthesis of silica–SF aerogel hybrids is shown in Figure 1a. The developed procedure leads to a series of silica–silk fibroin hybrid aerogel monoliths with an encouraging set of materials properties, such as low density, high porosity, and high flexibility for both compression and bending. The promising mechanical performance of the resulting silica–SF aerogel hybrids let us apply the processed wet silica–SF gels as a printable ink to deposit a unique aerogel construct with defined overall morphology and customizable inner pore architecture, paving the way for the fabrication of multifunctional aerogels (Figure 1b).

2 Methods

2.1 Materials

B. mori silkworm cocoons were purchased from Wild Fibers, UK. Tetramethylorthosilicate (98% purity, TMOS), hexadecyltrimethylammonium bromide (98% purity, CTAB),

methanol (99.8%, MeOH), trimethoxysilane (95% purity), 4-pentanoic acid (98% purity), lithium bromide anhydrous, (99.99% purity, LiBr), ammonium hydroxide (28-30%, NH_4OH), and sodium carbonate (Na_2CO_3) were procured from Sigma-Aldrich. All chemicals were used without purification. Slide-A-Lyzer Dialysis Cassettes with molecular weight cutoffs (MWCOs) of 3.5KD and a volume capacity of 3.5–5 mL were purchased from ThermFisher Scientific.

2.2 Silk Fibroin Extraction

The SF aqueous solution was extracted from silkworm cocoons through a slightly modified procedure reported in *Nature Protocols* by Kaplan et al.³² First, silk cocoons (5 g) were cut into dime-sized pieces and boiled for 30 min in a 2 L solution of Na_2CO_3 (0.02M); then fibers were rinsed with plenty of ultrapure water and dried overnight. The dry silk fibers were dissolved in LiBr (12–15 M) solution at 60 °C for 4 h and then dialyzed against ultrapure water for 48 h. The dialyzed SF solution was centrifuged at 9000 rpm twice, and the supernatant was stored at 4 °C for later use.

2.3 Synthesis of 5-(Trimethoxysilyl)pentanoic Acid (TMSPA)

The synthesis of TMSPA has been previously reported in our group.⁴⁰ Trimethoxysilane (0.05 mol) was added dropwise to a suspension of 4-pentenoic acid (0.05 mol) and platinum(IV) oxide (0.05 mmol) at 0 °C under argon atmosphere. After stirring the mixture for 6 h at 0 °C and 12 h at room temperature (23 °C) and filtration over a polytetrafluoroethylene syringe filter, the product was obtained quantitatively as a light brown-colored liquid. The color results from colloidal Pt(0) particles, which can be removed by addition of charcoal and filtration by Whatman Puradisc syringe.⁴⁰

2.4 Synthesis of Silica–SF Aerogel Hybrids

To obtain silica–SF composites, we adopted a one-step/one-pot acid-catalyzed sol-gel approach (cf. Figure 1) in which a sol of organosilanes (TMOS and TMSPA), with a given silicon molar content of $[\text{Si}] = 3.37$ mmol and a varied TMSPA concentration of 0, 10, and 20 mol % of the total silicon, and a SF biopolymer is prepared in an aqueous acetic acid solvent in the presence of CTAB (0.25 g). In the total sol mixture, the SF/silica mass fractions were 15:100 and 30:100. The samples are labeled as ET-SF- y and ETT- x -SF- y , where x and y represent the TMSPA and SF percentage, respectively. For example, for ET-SF-30 which contains SF/Silica = 30/100, a total sol (5 mL) containing silanes (9.39 w/v%, 0.5 mL) and SF (2.8 w/v%, 4 mL) was prepared. The concentration of acetic acid (0.5 mL) is varied between 100 and 120 mM for the system with the SF/silica mass fraction of 15:100 and 30:100, respectively. Gelation occurred concomitant with an increase in sol viscosity (after 10 min). Depending on the TMSPA concentration, the overall gelation time varied from ~1 to 10 h for ET- x -SF- y and ETT- x -SF- y , respectively. The homogeneous hybrid silica-SF gels were aged in an oven (40 °C, 2 days). Byproducts were extracted by solvent exchange with methanol, followed by drying of the filigree wet gels with supercritical CO_2 .

2.5 3D Printing of Silica–SF Aerogel Hybrids

The printed grid-like construct has been prepared using one selected representative formulation of the silica–SF hybrid preset gels, ET-SF-15, as a printable ink and applying a microextrusion-based 3D printer (Engine E3, Hyrel 3D, 5 μm (x,y), and 1 μm (z)). First, the computer-aided 3D structure was designed using a CAD design software (MicroStation V8i, Bentley Systems), resulting in a file which was exported to STL format and then transferred to G code for each layer in the 3D structure. The G code determined the nozzle (y) and stage movement (x,z) to generate the 3D architecture through a layer by layer deposition. Next, the sol mixture of ET-SF-15 is loaded to a 1 mL syringe with an 18-gauge needle equipped with a trapped-type nozzle prior to gelation. Then the syringe was transferred to the oven (40 °C) to start gelation in both silica and SF phases to obtain the proper viscosity of printing. After the syringe was loaded into the printer, the integrated stepping motor generated a pressure on the piston which was used to deposit the ET-SF-15 gel with a concomitant motion of the stage and nozzle (linear feed rate of 10 mm/s) in 3D to print a simple grid-like structure (ca. 2 cm X 2 cm X 0.5 cm) with grid mesh sizes of ca. 1 mm on a silicone-type substrate. The as-printed construct was covered in a Petri dish or an appropriate chamber with controlled humidity and transferred to the oven (40 °C) for continuing the remaining sol–gel reaction/gelation and aging steps (~3 days). After aging, the construct was submerged in methanol, washed, solvent exchanged, and dried supercritically.

2.6 Characterization Techniques

The bulk density was calculated from the corresponding mass and volume of the cylindrical aerogel. The skeletal density was measured using a helium pycnometer (*AccuPyc II 1340*, Micromeritics, USA). TEM images were recorded with a JEOL 200F cold field-emission (W) filament, 200 keV operated at an accelerating voltage of 300 kV. Scanning electron microscopy (SEM) images were taken with a scanning electron microscope (Zeiss ULTRA Plus) running at 5–10 kV with an in-lens detector and a working distance around 3 mm. Fourier transform infrared spectroscopy (ATR-FTIR) was obtained on a Bruker Vertex 70 spectrometer with a 4 cm^{-1} resolution (scan range from 400 to 4000 cm^{-1}). Nitrogen adsorption-desorption measurements were carried out at 77 K using a Micromeritics ASAP 2420. Before analysis, the sample was outgassed at 60 °C in vacuum (10^{-5} bar) for 24 h to remove adsorbed species. The specific surface area was calculated with the Brunauer, Emmett, and Teller 5-point method in the relative pressure range of 0.05–0.3. From the skeleton and bulk densities values, the porosity ϵ (%), pore volume (V_p), and pore diameter (D_p) of the samples were calculated through eqs 1, 2, and 3, respectively.

$$\epsilon(\%) = \frac{1/\rho_b - 1/\rho_s}{1/\rho_b} \times 100 \quad (1)$$

$$V_{\text{pore}}(\text{cm}^3/\text{g}) = 1/\rho_b - 1/\rho_s \quad (2)$$

$$D_{\text{pore}}(\text{nm}) = 4V_p/S_{\text{BET}} \quad (3)$$

Solid-state ^{29}Si , ^{13}C , and ^1H NMR spectra of the aerogels were obtained by using an Inova 500 spectrometer using a 4 mm solids probe with cross-polarization and magic angle spinning at 11 kHz. A solid-state ^1H – ^{29}Si heteronuclear correlation spectrum was collected using 4 mm zirconia rotors with a spinning rate of 11 kHz and applying homo-nuclear decoupling using continuous phase modulation (DUMBO) to increase the ^1H spectral resolution. Mechanical characterization of the composites was carried out on the monolithic cylindrical samples using a universal mechanical testing equipment (Zwick/Z010, Zwick/Roell, Germany) equipped with a 1 kN force transducer (KAP-S, AST Gruppe GmbH, Germany) in a controlled environment (23 °C, 50% relative humidity). Stress–strain curves were plotted in compression mode, and elastic moduli were calculated from the linear range of the curves which was typically found at $3 \pm 1\%$ strain. A constant deformation rate of 0.5 mm/min was used, and final strength was taken at the first signs of buckling which occurred typically at $>30\%$ strain. The three-point bending (flexural) test on the 3D-printed sample was also carried out with the same equipment, but with a custom build load cell and support span and the deflection velocity of 0.05 mm/min. The thermal conductivity of the silica–SF aerogels was measured using a transient method (Thermal constants analyzer TPS 2500 S, Hot Disk). The sensor is clamped between two identical disc-shaped pieces of the sample, which have a diameter and thickness of 1 cm (adequately cut from the cylindrical aerogel samples). This analysis was carried out at 20 °C, and the equipment presents a reproducibility and accuracy over 1% and 5%.

3 Results and Discussions

Silk fibroin is a protein which is produced by silkworm but also by other species, such as spiders, mites, butterflies, and bees to name only a few.³⁴ It typically comprises both crystalline and amorphous domains. Compared to the other natural biopolymers, silk fibroin is very promising for different applications due to its unique physicochemical and excellent mechanical properties as well as the possibility of large-scale production.⁴¹ Silk fibroin has been investigated in diverse applications, mainly as biomaterial,^{33,34} but also for forming various 2D and 3D materials via self-assembly processes, resulting in structures ranging from macro-, micro-, to nanoscale with unique functionalities. A summary and more comprehensive information with regard to the possibilities in structural design, surface chemistry, possibility of processing, and its diverse biorelated applications can be found in various recent review papers.^{34,41–43}

As shown in Figure 2, similar to the other living organisms in nature, silk fibroin extracted from *B. mori* silkworm cocoon shows a hierarchical/multilevel organization, which allows the fabrication of multifunctional hierarchical materials for various applications.

The protein chain of SF from *B. mori* mainly consists of the amino acids, Gly (43%), Ala (30%), and Ser (12%), which are arranged in different repetitive sequences to form amorphous elastic chains that are connected to the stable hydrophobic crystalline antiparallel

β -sheet regions to form silk I and silk II structures.⁴¹ The β -sheet regions are connected through stable hydrogen bonds and provide strength and stiffness.

The extracted silk fibroin solution can be processed in a combination of a sol–gel approach and supercritical drying. This results in an ultralight (bulk density = 0.02 g cm⁻³), soft and superflexible (compressibility up to 80% of strain, cf. Table 1), and micro-/macroporous SF aerogel with the relatively high surface area (~412 m² g⁻¹). Figure 3a–d presents some of the resulting unique properties of these SF aerogels, which will be named AeroSF in the following. This is the first report on an aerogel monolith hitherto reported from silk fibroin biopolymer with better materials properties than the other biopolymer-based aerogels^{44,45} but also to the majority of the reported silica-based aerogels.¹¹ Assembly of the silk fibroin to a gel structure via sol–gel processing is based on aqueous acid catalysis, which results in physical-cross-linking/gelation (hydrogen bonding) between the SF polymer chains but also in an increase in β -sheet structures upon processing with further postsynthesis solvent exchange and supercritical drying.³⁵ The β -sheet secondary structure in the SF polymer is known as the mechanically more stable conformation; therefore, it gives rise to resilient and structurally stable functional aerogel-based materials. Keeping this in mind, the synergy between silk fibroin and silica could result in tailored hybrid aerogels with a competitive and unique set of physicochemical and mechanical properties.

Table 1 details the starting compositions, gelation times, as well as some essential physical properties of the developed silica–SF aerogel hybrids. The simplicity and the advantage of the proposed synthesis approach relies on the fact that surface modification of the silica network and SF incorporation into the underlying silica are all performed by a single-step sol–gel process. In this single-step approach, the sequential reactions of (1) co-condensation of TMOS with an organofunctional silane carrying a carboxylic acid functionality, TMSPA, that acts as silane coupling agent to link to the silk fibroin (cf. Figure 1) and (2) in situ assembling the silk fibroin fibers/polymers inside the preformed silica network occur.

The in situ gelation of the SF biopolymer with the (organo)-silanes is occurring in two steps. First, gelation is induced by assembling the SF in diluted acidic media. The silk fibroin proteins become physically cross-linked through intra- and interchain hydrogen bonds followed by simultaneous hydrolysis and polycondensation of the (organo)silanes in a second step (cf. Figure 1). For an efficient mixing of the (organo)silanes in the aqueous medium and to inhibit macroscopic phase separation during the sol–gel transition, the cationic surfactant (CTAB) is added to the mixture. The silane coupling agent, TMSPA, carrying carboxylic acid moieties, also increases the acidity of the sol mixture and therefore allows one to perform the sol–gel reaction in a one-step acid-catalyzed reaction in which the gelation kinetics of the silanes are matched with that of SF, controlling the concentration of the acid catalyst. During gelation, a network of particles forms which is "glued" together by the secondary phase of the silk fibroin network. The silk fibroin network interpenetrates and supports the silica skeleton by acting as a mechanically stable support or scaffold. The interactions between the two phases (silica and silk) are mediated by various covalent and noncovalent linkages between the carboxylic acid moieties of TMSPA and/or surface silanols with the targeted amino acids in SF.

A substantial effect of TMSPA is recognized in the gelation process. While in the formulations without or with lower contents of TMSPA (<10%) gelation of the organosilane occurs within the first hour (~1 h) after mixing, addition of TMSPA significantly slows gelation down. Higher contents of TMSPA require a longer gelation time (7 h) to yield stable monoliths. A possible explanation might be the different reaction kinetics (strong dependency on pH value and the reduced number of cross-linking possibilities). The pristine silica aerogel (produced from TMOS only) is transparent, while upon addition of TMSPA and SF the gels turned opaque, probably due to phase separation of the hydrophobic organosilane species (hydrocarbon chain of TMSPA) and the hydrophobic β -sheet SF secondary structure from the aqueous phase.

Figure 4a shows the solid-state ^{29}Si CP-MAS NMR spectra of silica and carboxylic acid-modified silica aerogels proving the successful linkage/co-condensation of TMSPA with TMOS during the *in situ* gelation. The characteristic peaks for TMOS (ET) are indexed as Q_n : Q_4 , (δ –106 ppm), Q_3 , (δ –98 ppm), and Q_2 (δ –88 ppm), where n is the number of $\equiv\text{Si}-\text{O}-\text{Si}\equiv$ bridges, while for carboxylic acid-modified samples, ETT-20, the extra peaks of TMSPA are indexed as T_n : T_{3t} (δ –62 ppm), T_{2t} (δ –53 ppm), implying successful linkage of the coupling agent to the underlying silica structure.

Different possible covalent and noncovalent interactions of SF to the silica network are evident from the solid-state MAS ^1H NMR and ATR-FTIR spectra, Figure 4b and 4c. SF possesses various functional groups due to the presence of different amino acids⁴⁶ that can interact with silica and/or carboxylic acid-modified silica networks in different ways. Typical examples might be based on covalent bonds, hydrogen bonding, or/and dipole–dipole interactions (see also Figure 1).³³ For example, ET-SF-15, consisting of TMOS-based silica and the SF protein with no organofunctional silane, is likely to show an interaction via the highly abundant surface silanol groups ($-\text{Si}-\text{OH}$) and the hydroxyl side chains of SF (e.g., with serine amino acids). This interaction is substantiated by the presence of characteristic bands $\delta(-\text{SiOCH}_2-\text{Ser, Thr})$ 4.1 ppm in the ^1H MAS NMR spectrum (see Figure 4b) and $\nu_{\text{as}}(\text{Si}-\text{O}-\text{C})$ 1145 cm^{-1} in the ATR-FTIR spectrum (see Figure 4c), supporting the presence of covalent linkages between SF and the silica phase.

For all hybrid samples, the incorporation of the SF biopolymer into the silica aerogel network is also supported by the presence of SF characteristic peaks in the ATR-FTIR spectra: the amide I ($\nu_{\text{as}}(\text{C}=\text{O})$ 1618–1640 cm^{-1}), amide II ($\delta_s(\text{N}-\text{H})$ deformation/bending 1512–1544 cm^{-1}), and amide III ($\nu_{\text{as}}(\text{C}-\text{N})$ 1230 cm^{-1}) vibrations. In addition, the ATR-FTIR spectra show that upon SF gel formation and *in situ* gelation of silanes and SF solution, a β -sheet conformational state in SF biopolymers is favored, as the amide I band, the most sensitive band for the conformational changes from the α -helix to the α -sheet structure, at ~1640 cm^{-1} (for neat SF film with high random coil and α -helix structures) shows a shift to 1618 cm^{-1} (for AeroSF) and 1627 cm^{-1} (for ET-SF-15 and ETT-20-SF-15 aerogels).⁴⁷ During the *in situ* sol–gel process, the randomly oriented silk fibroin biopolymers undergo a physical cross-linking or gelation procedure which results in a soft SF gel with low amounts of β -sheet structures. However, the *in situ* sol–gel reaction of SF with organosilanes exposes the SF polymer to methanol released from the hydrolysis of TMOS and TMSPA, which together with critical point drying result in the β -sheet

conformations in the final SF network are induced. However, the conformational changes to the β -sheet structure of SF are expected to provoke structurally/mechanically more stable silica–SF aerogel monoliths. Consequently, all hybrid samples have undergone a methanol treatment and extraction before supercritically drying. Thermogravimetric analysis as shown in Figure 4d indicates that the silica–SF aerogel hybrids are thermally stable up to ~ 260 °C. SF and the alkyl moiety in the coupling agent (TMSPA) decompose around 253 and 380 °C, respectively. Also, the different weight losses (wt %) and overall rest masses of the hybrids confirm the presence of SF and TMSPA at the various loading extents.

The microstructure and properties of silica–SF aerogel hybrids can be regulated by varying the TMSPA and SF concentrations. Scanning electron microscopy (SEM) micrographs of all hybrid series, Figure 5a, exhibit a 3D bicontinuous network structure in which the macroporosity becomes evident in the silica network with the addition of SF and coupling agent resulting in more pronounced phase separation. Evidently, the pristine silica aerogels are composed of aggregates of colloidal particles or clusters interlinked together in a pearl-necklace-type morphology.⁴ AeroSF displays a network of polymer “strands” or “nanofibers” with diameters of a few tens of nanometers with mesopores and small macropores (see Figure 3d). Although in the microstructures of all silica–SF hybrid aerogels no evidence of SF fibers is visible, it is expected that the SF and silica phase with their fine network structures and similar network feature sizes are intertwined together creating an interpenetrated continuous network. Meanwhile, the addition of coupling agents renders the network particles to become coarser. The hierarchical porous structure is another characteristic of the hybrid samples with coupling agents demonstrating a macropore morphology with an average pore size of ~ 2 μm which are interconnected through mesopores. Although both TMSPA and SF loading to silica leads to phase separation and macropore development, for the samples with the same silica structure, the macroporosity upon increasing the SF loading in the hybrid is lost.

The surface area and the average pore diameter of aerogel hybrids are calculated from the nitrogen sorption data. As it is evident from the nitrogen sorption isotherms of the hybrid samples shown in Figure 5b, all of the aerogel hybrids have a type IV isotherm with capillary condensation occurring at $p/p_0 > 0.5$, reflecting the mesoporous characteristic of almost all hybrid aerogels rather than microporous aerogels. The hybrid samples with coupling agent show very narrow hysteresis loops, which is indicative of a relatively low mechanical deformation of the samples upon desorption of the liquid nitrogen condensed in the capillaries and, therefore confirming the stiffness of the hybrid materials. However, due to the possible mechanical deformation, during the desorption/drying of liquid N_2 , experienced by the hybrid aerogel in the desorption branch of the capillary condensation range, the pore volume (V_{pore}) and average pore diameter (D_{pore}) determined by Barrett–Joyner–Halenda (BJH) or density functional theory (DFT) is not entirely reliable. Therefore, we reported V_{pore} and D_{pore} calculated by eqs 2 and 3 for entirety, see Figure 5c.

As seen in the data in Figure 5c, the ET-SF-15 aerogels exhibit a high $S_{\text{BET}} > 750$ $\text{m}^2 \text{g}^{-1}$ which apparently decreases with increasing the SF loading and TMSPA content. Meanwhile, the mean pore diameter increases from 37 nm for ET-SF-15 to 79 nm for ETT-20-SF-30. As also seen from SEM micrographs, the aerogel with higher SF and TMSPA loadings exhibits

larger pores and aggregated particles which results from the macroscopic phase separation of the hydrophobic condensates of silane species and the formation of SF β -sheet structure during the in situ sol–gel reaction. Except for the samples with high loadings of SF, ETT-10-SF-30, and ETT-20-SF-30, the average pore diameter (D_{pore}) of all hybrids is below the mean free path of the air under ambient conditions (70 nm, STP),⁴⁸ which together with their low density suggests that the materials should display very low gas and solid thermal conductivity.⁴⁹

The TEM micrographs in Figure 6 indicate that compared to a pristine silica structure (acid catalyzed), Figure 6a, in which the quasi-spherical nanoparticles (size $\approx$ 5–10 nm) are connected through a pearl-necklace-like morphology, co-gelation of SF with silica under acidic conditions leads to the morphological changes toward “cluster–cluster”-type growth with thicker struts, Figure 6b. The SF and TMSPA incorporation to the silica network increased the packing density of the clusters with somewhat smaller pores, Figure 6c.

In order to obtain more evidence for a homogeneous and fine dispersion/mixing of SF in the silica structure at a molecular and microstructural level, we investigated the structure of ET-SF-15 with EFTEM elemental mapping, Figure 7a, and energy-dispersive X-ray spectroscopy (EDS)-based elemental analysis combined with EDS spectroscopy, Figure 7b. The mapping micrographs of selected samples indicate the presence of all four main network constituents of silicon, oxygen, carbon, and nitrogen in the hybrid aerogels. The nitrogen content in the SF biopolymer is very low; therefore, the nitrogen signals are not very prominent; however, the theoretical content of nitrogen is still beyond the detection limit of TEM mapping. Also, the elemental maps authenticate the homogeneous chemical or physical mixing of silica with the SF phase instead of forming isolated silica or SF-rich domains/aggregates in the structure.

The homogeneous mixing of organic and inorganic phases in both ET-SF-15 and ETT-10-SF-15 hybrid aerogels is also confirmed by ^1H – ^{29}Si heteronuclear correlation (HETCOR) MAS NMR spectroscopy (see Figure 8a and 8b, respectively).

This technique further confirms the molecular proximity of network constituents, SF, and silica in the hybrid structure through the coupling of SF protons with Si atoms at the silica network, as it was also reported for other hybrid aerogel systems like silica–pectin³⁰ and silica–chitosan²⁶ structures. The probable covalent interactions between two phases are also investigated and assigned in this study. For both ET-SF-15 (Figure 8a) and ETT-10-SF-15 (Figure 8b), the surface $\equiv\text{Si-OH}$ groups interact with dangling hydroxyl functionalities of the SF network (cf. Figure 4b) through covalent bonding. Therefore, the correlation of methylene protons in the side chain of SF (serine and threonine, $-\text{CH}_2\text{OH}$, ^1H NMR, δ 4.1–4.9 ppm) with silicon atoms (Q_4 , Q_3 , ^{29}Si MAS NMR, δ –106 and –98 ppm, respectively) of the silica network shows cross-peaks that reflect the mixing of both phases through covalent linkages (e.g., formation of $\equiv\text{Si-O-CH}_2\text{-(Ser, Thr)}$). In general, this technique presents a good understanding with regard to the type of interactions in the network and provides a broad view regarding the molecular proximity of the SF and silica phases.³⁰

Benefiting from their molecular and nanoscale structures, silica–SF aerogel hybrids exhibit improved mechanical properties. SF incorporation into the silica network can dramatically overcome the brittleness of silica aerogels provided that the TMSPA coprecursor concentration and an optimal SF loading has been used. As shown in the stress–strain curves and photographs of the compression tests, the optimized aerogel, ETT-20-SF-15, combines high compression flexibility and strength as well as excellent bending flexibility by hand, Figure 9a. Also, as seen from the surface plots in Figure 9b and 9c, respectively, there are mainly two factors that are involved in optimal mechanical strength with regard to the maximum compression strength and compressibility. First, the elastic siloxane network in the skeleton contains long and flexible aliphatic (pentanoic acid) chains of TMSPA, which improve the strength against bending and compression. Second, the silica network is further linked to the flexible hydrocarbon chains of the SF biopolymer, which leads to a high deformability without fracture of the skeleton on both compression and bending. Evidently, bulk density (ρ_b) in the silica–SF aerogel hybrids can also be influenced by varying the SF loading and TMSPA concentration (Figure 9d, Table 1). At constant SF loading, increasing the TMSPA concentration up to 10% results in a slight increase in density, indicating enhanced incorporation of SF to the silica network upon increasing the amount of coupling agent. Under the same conditions, a hybrid with 30% of SF with a TMSPA content of 20% results in a lower incorporated amount of SF, probably due to steric hindrances, etc., which subsequently gives a lower density and decreased compressibility. This is also confirmed by a comparison of the thermogravimetric analysis of ETT-20-SF-30 and ETT-20-SF-15. For the first one, the ceramic yield is lower (46%) than for the latter one (39%), indicative of a lower amount of organic moieties in the network.

3.1 3D-Printed Silica–SF Aerogel Hybrid Constructs

Due to structural and mechanical restrictions, machinability or postsynthesis shaping of silica aerogels by scalable and controllable strategies remains a significant challenge.³⁹ 3D printing of aerogels can pave the way to develop genuine 3D architectures, e.g., with customized morphology and microstructure, and with pore sizes in a broad range from micro- to macropores with well-defined shapes/architecture.³⁷ To realize this goal, we aimed to demonstrate the printability of the developed silica–SF hybrid gel in order to allow for shaping it to defined complex constructs. To this aim, as illustrated in Figure 1b and Video S1 we developed a novel extrusion-based 3D printing approach and printed the silica–SF preset gels as ink with an optimized viscosity in order to be quickly deposited through a layer by layer deposition method. The viscosity of the gel is a very determinant factor for the printing feasibility of gel, as high viscosity in the gel impedes the homogeneous deposition while gels with a low viscosity are reluctant to reshape and flow after deposition. The optimized viscosity for printing of a silica–SF hybrid gel was found to be optimal at ~2 h after the onset of gelation, while the polycondensation in the silica phase is still incomplete. Each layer is deposited by controlling the deposition rate and nozzle diameter, based on the predesigned CAD model (see Video S1). As shown in Figure 1b, processing includes postprinting aging for 2 days, immersion of the 3D-printed architectures in methanol to induce β -sheet conformation to increase the mechanical durability in the printed constructs, solvent extraction, and finally supercritical drying, which results in a robust architecture that

can easily be handled for multipurpose applications. Figure 10a shows a representative 3D-printed grid-like construct of ET-SF-15 along with its microstructure which was characterized by SEM from the top and side views, Figure 10b–c. Viewed from the top in Figure 10b, an individual grid, in each printed construct, possessing a side length of $\sim 450\ \mu\text{m}$ (about 50% of initially printed grids) and a wall thickness of $\sim 350\ \mu\text{m}$ is seen.⁵⁰

The magnified view of the wall microstructure (Figure 10c) indicates that except for a slight decrease in the extent of the mesoporosity compared to the cylindrical monolith (ET-SF-15) in Figure 5, probably due to the shear stress applied to the gel structure during the microextrusion process, no significant visible structural changes in the network build-up is visible. The printed sample exhibited mesoporous characteristics as confirmed by N_2 adsorption–desorption isotherm (Figure 10d); also most of the physical and microstructural properties have been hardly influenced by the extrusion and concurrent shear forces on the structure during printing.

Due to the flat geometry of the printed grid construct, we have only been able to study its flexural properties using three-point bending tests. Figure 11 displays the stress–strain curve of the printed sample during deflection and its failure. Although the flexural strength (3.2 Pa) and modulus of the printed construct (38.74 Pa) are relatively low, it is still easy to handle and expected to be usable for most of the intended applications.

In summary, the dual pore silica–SF aerogel hybrids are successfully printed into a customizable 3D structure. With that the silica–SF-based scaffold designs become suitable for specific cell integration requirements but may also be beneficial for other high-performance applications.

3.2 Thermal Insulation and Fire Retardancy of Silica–SF Aerogel Hybrids

Due to the low density and great extent of porosity comprising micro-, meso-, and macropores, aerogels are increasingly drawing attention as high-performance thermal insulation materials in various domains.^{51,52} The thermal conductivity of pure silk fibroin aerogel, AeroSF, of this study is as low as $0.026\ \text{W m}^{-1}\ \text{K}^{-1}$, which is the first superinsulator aerogel from silk fibroin biopolymer hitherto reported.⁵³ The thermal insulation performance of AeroSF is also comparable with that of pectin aerogels, Aeropectin ($\lambda = 0.020\ \text{W m}^{-1}\ \text{K}^{-1}$).²⁰ Figure 12a presents the total thermal conductivity of two different representative silica–SF aerogel hybrids. It is seen that ET-SF-15 has a thermal conductivity (λ) of $0.033\ \text{W m}^{-1}\ \text{K}^{-1}$, which by replacing 20 mol % of total silicon with coupling agents in the overall hybrid, ETT-20-SF-15, is raised to $0.039\ \text{W m}^{-1}\ \text{K}^{-1}$. Even though ETT-20-SF-15 shows a slight increase in the bulk density and subsequently in the solid thermal conductivity, the hybrid aerogel is three times stronger with a lower brittleness and excellent compressibility. In addition, the pore size of almost all silica–SF hybrid aerogels is mainly in the mesopore regime (see Figure 5c), thus favoring the Knudsen effect, which implies that the air circulation is confined inside the pores. This finally results in a lower gaseous thermal transfer. The thermal insulation properties of the silica–SF aerogel hybrids are superior to those of previously reported polymer-reinforced silica aerogels ($\lambda =$

0.045–0.07 W m⁻¹ K⁻¹)¹⁸ and even better than those of currently used insulators such as polystyrene foam ($\lambda = 0.03\text{--}0.06$ W m⁻¹ K⁻¹).⁵⁴

Traditional fossil-fuel insulating materials are easily ignitable and therefore require the addition of the flame retardants.^{55,56} Most of the flame-retardant materials, such as halogenated and phosphorus compounds, have a negative impact on the health and the environment.⁵⁷ Silica aerogels are known as fire-retardant materials.⁵⁶ Thus, another advantage of silica–SF aerogel hybrids is their fire-retardant behavior due to the homogeneous mixing of silk fibroin biopolymer and the silica network in the overall aerogel composite. Figure 12b demonstrates the vertical burning of ET-SF-15 which displayed excellent fire retardancy without self-propagation of the flame and resulted in a carbonized residue with a similar shape and dimension as the original aerogel, while in comparison the pure AeroSF displayed only some fire retardancy and shrunk upon burning.

4 Conclusions

In summary, mechanically durable and lightweight silica–SF aerogel hybrids have been prepared from cogelation of two silica precursors—tetramethoxysilane (TMOS) and 5-(trimethoxysilyl)-pentanoic acid (TMSPA)—and a silk fibroin (SF) biopolymer—extracted from silkworm cocoon—through a simple, aqueous-based, and straightforward one-step acid-catalyzed sol–gel reaction. The reaction follows a stepwise sol–gel transition in which first a network is formed from the SF biopolymer and subsequently the (organo)silanes hydrolyze and condense to yield an organic–inorganic hybrid network. In the entirely aqueous-based system a surfactant, cetyltrimethylammonium bromide (CTAB), is used to homogenize the alkoxy silane and SF phases and to suppress phase separation between the various species and water. Variation in the molar ratios of the different components, such as TMOS, TMSPA, and SF, has a strong influence on the final physical, microstructural, as well as mechanical (compressive) properties of the synthesized aerogel hybrids.

The coupling agent, TMSPA, serves various functions during processing: On one hand, the acidic nature of TMSPA catalyzes the sol–gel transition and facilitates a homogeneous mixing of the alkoxy silanes with the secondary SF phase to allow for processing everything in a one-pot reaction. On the other hand, the carboxylic acid moieties efficiently mediate the interactions with the functional moieties, e.g., amino groups, in the silk fibroin polymer chains. Moreover, the flexible hydrocarbon chain of the coupling agent provides the underlying brittle silica structure with some flexibility and resiliency.

We have shown by a number of characterization techniques that the resulting silica–silk fibroin aerogel hybrids show a molecular level homogeneous interpenetrated nanostructure with a combination of unique properties such as low density (0.11–0.19 g cm⁻³), high surface area (311–798 m² g⁻¹), and flexibility in compression (endure up to 80% of compression strain, with a maximum strength of ~8 MPa).

Therefore, the silica–SF monoliths of this study, from various perspectives comprise several advantages. (1) A green and naturally occurring biopolymer of SF has been used for the first time to mechanically reinforce silica aerogels. (2) The hybridization reaction is simple and

leads to a considerable reduction in the time that is basically required to hybridize silica with fossil-fuel-based polymers along with a considerable reduction in the consumption of the hazardous organic solvents during gel processing. (3) An excellent improvement in the mechanical strength (3 orders of magnitude in Young's modulus) compared to a pristine silica aerogel has been achieved with minimal or almost no compromise on the density and thermal insulation performance. (4) The developed silica–SF aerogel shows a fire retardancy.

Last but not least, 3D printing of the silica–SF aerogel hybrid architecture is a novel strategy in order to enable tailoring the micro- and macrostructure. To the best of our understanding, 3D printing of silica-based hybrid aerogels could solve the shortcomings of low postsynthesis machinability of aerogel thanks to the excellent controllability in achieved mechanical strength upon hybridization and simultaneously allow one to design the micro–macrostructure in the gel by controlling the printing method. The 3D-printed hybrid aerogel construct in this study offers a high potential for several applications, in particular, for designing anisotropic thermal insulation materials, and promotes future application of dual porous aerogels, with the simultaneous presence of micrometer and nanosized pores, as scaffolds for bone-based regenerative medicine.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

H.M. acknowledges financial support by the Austrian FWF for the Lise Meitner fellowship (project no. M2086-N34). S.M. acknowledges financial support from Interreg Österreich–Bayern 2014–2010 Project AB29 *Synthese, Charakterisierung und technologische Fertigungsansätze für den Leichtbau “n2m” (nano-to-macro)* and F.P. acknowledges financial support from the Austrian Science Foundation FWF (Project I 1605-N20). H.M. acknowledges Dr. Mohsen Bandar Abadi for providing the logistics services during the project.

References

- (1). Kistler SS. Coherent Expanded Aerogels and Jellies. *Nature*. 1931; 127:741–741.
- (2). Kistler SS. Coherent Expanded-Aerogels. *J Phys Chem*. 1931; 36:52–64.
- (3). Hüsing N, Schubert U. Aerogels—Airy Materials: Chemistry, Structure, and Properties. *Angew Chem Int Ed*. 1998; 37:22–45.
- (4). Maleki H, Durães L, Portugal A. An overview on silica aerogels synthesis and different mechanical reinforcing strategies. *J Non-Cryst Solids*. 2014; 385:55–74.
- (5). Koebel, MM, Rigacci, A, Achard, P. Aerogels for Super-insulation: A Synoptic View. *Aerogels Handbook*. Aegerter, MA, Leventis, N, Koebel, MM, editors. Springer; 2011. 607–633.
- (6). Maleki H, Hüsing N. Current status, opportunities and challenges in catalytic and photocatalytic applications of aerogels: Environmental protection aspects. *Appl Catal B*. 2018; 221:530–555.
- (7). Maleki H. Recent advances in aerogels for environmental remediation applications: A review. *Chem Eng J*. 2016; 300:98–118.
- (8). Lamy-Mendes A, Silva RF, Durães L. Advances in carbon nanostructure—silica aerogel composites: a review. *J Mater Chem A*. 2018; 6:1340–1369.
- (9). Stergar J, Maver U. Review of aerogel-based materials in biomedical applications. *J Sol-Gel Sci Technol*. 2016; 77:738–752.
- (10). Maleki H, Durães L, García-González CA, del Gaudio P, Portugal A, Mahmoudi M. Synthesis and biomedical applications of aerogels: Possibilities and challenges. *Adv Colloid Interface Sci*. 2016; 236:1–27. [PubMed: 27321857]

- (11). Zhao S, Malfait WJ, Guerrero-Albuquerque N, Koebel MM, Nyström G. Biopolymer Aerogels and Foams: Chemistry, Properties and Applications. *Angew Chem Int Ed*. 2018; doi: 10.1002/anie.201709014
- (12). Leventis N, Sotiriou-Leventis C, Zhang G, Rawashdeh A-MM. Nanoengineering Strong Silica Aerogels. *Nano Lett*. 2002; 2:957–960.
- (13). Leventis N. Three-Dimensional Core-Shell Superstructures: Mechanically Strong Aerogels. *Acc Chem Res*. 2007; 40:874–884. [PubMed: 17488055]
- (14). Maleki H, Durães L, Portugal A. Synthesis of lightweight polymer-reinforced silica aerogels with improved mechanical and thermal insulation properties for space applications. *Microporous Mesoporous Mater*. 2014; 197:116–129.
- (15). Katti A, Shimpi N, Roy S, Lu H, Fabrizio EF, Dass A, Capadona LA, Leventis N. Chemical, Physical, and Mechanical Characterization of Isocyanate Cross-linked Amine-Modified Silica Aerogels. *Chem Mater*. 2006; 18:285–296.
- (16). Meador MAB, Fabrizio EF, Ilhan F, Dass A, Zhang G, Vassilaras P, Johnston JC, Leventis N. Cross-linking Amine-Modified Silica Aerogels with Epoxies: Mechanically Strong Lightweight Porous Materials. *Chem Mater*. 2005; 17:1085–1098.
- (17). Maleki H, Durães L, Portugal A. A New Trend for Development of Mechanically Robust Hybrid Silica Aerogels. *Mater Lett*. 2016; 179:206–209.
- (18). Maleki H, Durães L, Portugal A. Synthesis of mechanically reinforced silica aerogels via Surface-Initiated Reversible Addition-Fragmentation Chain Transfer (RAFT) Polymerization. *J Mater Chem A*. 2015; 3:1594–1600.
- (19). Gandini A, Lacerda TM, Carvalho AJ, Trovatti E. Progress of Polymers from Renewable Resources: Furans, Vegetable Oils, and Polysaccharides. *Chem Rev*. 2016; 116:1637–1669. [PubMed: 26291381]
- (20). Rudaz C, Courson R, Bonnet L, Calas-Etienne S, Sallée H, Budtova T. Aeropectin: Fully Biomass-Based Mechanically Strong and Thermal Superinsulating Aerogel. *Biomacromolecules*. 2014; 15:2188–2195. [PubMed: 24773153]
- (21). White RJ, Budarin VL, Clark JH. Pectin-Derived Porous Materials. *Chem - Eur J*. 2010; 16:1326–1335. [PubMed: 19998430]
- (22). Fischer F, Rigacci A, Pirard R, Berthon-Fabry S, Achard P. Cellulose-based aerogels. *Polymer*. 2006; 47:7636–7645.
- (23). Innerlohinger J, Weber HK, Kraft G. Aerocellulose: Aerogels and Aerogel-like Materials made from Cellulose. *Macromol Symp*. 2006; 244:126–135.
- (24). Wan J, Zhang J, Yu J, Zhang J, Zhang Jun. Cellulose Aerogel Membranes with a Tunable Nanoporous Network as a Matrix of Gel Polymer Electrolytes for Safer Lithium-Ion Batteries. *ACS Appl Mater Interfaces*. 2017; 9:24591–24599. [PubMed: 28654232]
- (25). Liebner F, Haimer E, Wendland M, Neouze M-A, Schluffer K, Miethe P, Heinze T, Potthast A, Rosenau T. Aerogels from Unaltered Bacterial Cellulose: Application of scCO₂ Drying for the Preparation of Shaped, Ultra-Lightweight Cellulosic Aerogels. *Macromol Biosci*. 2010; 10:349–352. [PubMed: 20166232]
- (26). Zhao S, Malfait WJ, Jeong E, Fischer B, Zhang Y, Xu H, Angelica E, Risen JWM, Suggs JW, Koebel MM. Facile One-Pot Synthesis of Mechanically Robust Biopolymer—Silica Nanocomposite Aerogel by Cogelation of Silicic Acid with Chitosan in Aqueous Media. *ACS Sustainable Chem Eng*. 2016; 4:5674–5683.
- (27). Takeshita S, Yoda S. Translucent, hydrophobic, and mechanically tough aerogels constructed from trimethylsilylated chitosan nanofibers. *Nanoscale*. 2017; 9:12311–12315. [PubMed: 28825069]
- (28). Mallepally RR, Marin MA, Surampudi V, Subia B, Rao RR, Kundu SC, McHugh MA. Silk fibroin aerogels: potential scaffolds for tissue engineering applications. *Biomed Mater*. 2015; 10
- (29). Schuster, P, Wolschann, P. Hydrogen Bonding: From Small Clusters to Biopolymers. Springer; 1999. 3–16.
- (30). Zhao S, Malfait W, Demilecamps A, Zhang Y, Brunner S, Huber L, Tingaut P, Rigacci A, Budtova T, Koebel MM. Strong. Thermally Superinsulating Biopolymer-Silica Aerogel Hybrids by Cogelation of Silicic Acid with Pectin. *Angew Chem Int Ed*. 2015; 54:14282–14286.

- (31). Demilecamps A, Reichenauer G, Rigacci A, Budtova T. Cellulose–silica composite aerogels from “one-pot” synthesis. *Cellulose*. 2014; 21:2625–2636.
- (32). Rockwood DN, Preda RC, Yücel T, Wang X, Lovett ML, Kaplan DL. Materials fabrication from *Bombyx mori* silk fibroin. *Nat Protoc*. 2011; 6:1612–1631. [PubMed: 21959241]
- (33). Murphy AR, Kaplan DL. Biomedical applications of chemically-modified silk fibroin. *J Mater Chem*. 2009; 19:6443–6450. [PubMed: 20161439]
- (34). Kundu B, Rajkhowa R, Kundu SC, Wang X. Silk fibroin biomaterials for tissue regenerations. *Adv Drug Delivery Rev*. 2013; 65:457–470.
- (35). Tseng P, Napier B, Zhao S, Mitropoulos AN, Applegate MB, Marelli B, Kaplan DL, Omenetto FG. Directed assembly of bio-inspired hierarchical materials with controlled nanofibrillar architectures. *Nat Nanotechnol*. 2017; 12:474–480. [PubMed: 28250472]
- (36). Zhang YS, Yue K, Aleman J, Mollazadeh-Moghaddam K, Bakht SM, Yang J, Jia W, Dell'Erba V, Assawes P, Shin SR, Dokmeci SR, et al. 3D Bioprinting for Tissue and Organ Fabrication. *Ann Biomed Eng*. 2017; 45:148–163. [PubMed: 27126775]
- (37). Zhang Q, Zhang F, Medarametla SP, Li H, Zhou C, Lin D. 3D Printing of Graphene Aerogels. *Small*. 2016; 12:1702–1708. [PubMed: 26861680]
- (38). Li VC-F, Dunn CK, Zhang Z, Deng Y, Qi HJ. Direct Ink Write (DIW) 3D Printed Cellulose Nanocrystal Aerogel Structures. *Sci Rep*. 2017; 7
- (39). Saeed S, Al-Sobaihi RM, Bertino MF, White LS, Saoud KM. Laser induced instantaneous gelation: aerogels for 3D printing. *J Mater Chem A*. 2015; 3:17606–17611.
- (40). Feinle A, Flaig S, Puchberger M, Schubert U, Hüsing N. Stable carboxylic acid derivatized alkoxy silanes. *Chem Commun*. 2015; 51:2339–2341.
- (41). Qi Y, Wang H, Wei K, Yang Y, Zheng R-Y, Kim IS, Zhang K-Q. A Review of Structure Construction of Silk Fibroin Biomaterials from Single Structures to Multi-Level Structures. *Int J Mol Sci*. 2017; 18:237.
- (42). Kapoor S, Kundu SC. Silk protein-based hydrogels: Promising advanced materials for biomedical application. *Acta Biomater*. 2016; 31:17–32. [PubMed: 26602821]
- (43). Melke J, Midha S, Ghosh S, Ito K, Hofmann S. Silk fibroin as biomaterial for bone tissue engineering. *Acta Biomater*. 2016; 31:1–16. [PubMed: 26360593]
- (44). Rudaz C, Courson R, Bonnet L, Calas-Etienne S, Sallee H, Budtova T. Aeropectin: fully biomass-based mechanically strong and thermal superinsulating aerogel. *Biomacromolecules*. 2014; 15:2188–2195. [PubMed: 24773153]
- (45). Druel L, Bardl R, Vorwerk W, Budtova T. Starch Aerogels: A Member of the Family of Thermal Superinsulating Materials. *Biomacromolecules*. 2017; 18:4232–4239. [PubMed: 29068674]
- (46). Zhou CZ, Confalonieri F, Jacquet M, Perasso R, Li ZG, Janin J. Silk fibroin: structural implications of a remarkable amino acid sequence. *Proteins: Struct,Funct Genet*. 2001; 44:119–122. [PubMed: 11391774]
- (47). Lu Q, Zhang X, Hu X, Kaplan DL. Green process to prepare silk fibroin/gelatin biomaterial scaffolds. *Macromol Biosci*. 2010; 10:289–298. [PubMed: 19924684]
- (48). Jennings SG. The mean free path in air. *J Aerosol Sci*. 1988; 19:159–166.
- (49). Koh, YK. Heat transport by phonons in crystalline materials and nanostructures. Ph.D. Dissertation; University of Illinois at Urbana-Champaign: 2010.
- (50). Putz F, Scherer S, Ober M, Morak R, Paris O, Hüsing N. 3D Printing of Hierarchical Porous Silica and α -Quartz. *Adv Mater Technol*. 2018
- (51). Koebel M, Rigacci A, Achard P. Aerogel-based thermal superinsulation: an overview. *J Sol-Gel Sci Technol*. 2012; 63:315–339.
- (52). Cuce E, Cuce PM, Wood CJ, Riffat SB. Toward aerogel based thermal superinsulation in buildings: A comprehensive review. *Renewable Sustainable Energy Rev*. 2014; 34:273–299.
- (53). Maleki H, Whitmore L, Huesing N. *J Mater Chem A*. 2018; doi: 10.1039/C8TA02821D
- (54). Wang X, Zhang H, Jana SC. Sulfonated syndiotactic polystyrene aerogels: properties and applications. *J Mater Chem A*. 2013; 1:13989–13999.

- (55). Kashiwagi T, Du F, Douglas JF, Winey KI, Harris RH Jr, Shields JR. Nanoparticle networks reduce the flammability of polymer nanocomposites. *Nat Mater.* 2005; 4:928–933. [PubMed: 16267575]
- (56). Kashiwagi T, Gilman JW, Butler KM, Harris RH Jr, Shields JR, Asano A. Flame retardant mechanism of silica gel/silica. *Fire Mater.* 2000; 24:277–289.
- (57). Hale RC, La Guardia MJ, Harvey EP, Gaylor MO, Mainor TM, Duff WH. Flame retardants: Persistent pollutants in land-applied sludges. *Nature.* 2001; 412:140–141. [PubMed: 11449259]

Figure 1.

Synthesis route and resulting silica–SF aerogel hybrids. (a) Schematic description of the facile synthesis of pure SF and silica–SF aerogels from extracted SF solution and a mixture of TMOS and TMSPA through one-step sol–gel reaction followed by supercritical drying (SCD) with a photograph of representative ET-SF-15 cylindrical monoliths. (b) 3D printing process of silica–SF aerogels, ET-SF-15 being the used ink along with the stepwise processing of the 3D-printed hybrid grid-like architecture.

Figure 2.

Schematic presentation of the hierarchical structure of raw silkworm filament along with its different conformations and structures.

Figure 3.

Some characteristics of pure silk fibroin aerogel monoliths (AeroSF) of this study.

Mechanical properties (stress–strain curve for compression) (a), with AeroSF behavior upon compression with hand (b), N_2 adsorption–desorption isotherm (c), and SEM micrograph of typical AeroSF of this study (d).

Figure 4. (a) Solid-state MAS ^{29}Si NMR spectra, (b) MAS ^1H NMR spectra, (c) ATR-FTIR spectra, and (d) TG curves of silica-SF and SF aerogels.

Figure 5.

(a) SEM micrographs, (b) N₂ adsorption–desorption isotherms, and (c) BET specific surface area (S_{BET}), pore volume (V_{pore}), and average pore diameter (D_{pore}) data for silica–SF hybrid aerogels.

Figure 6.

TEM micrographs of (a) the reference silica aerogel, (b) ET-SF-15 aerogel monolith, and (c) ETT-20-SF-15 aerogels.

Figure 7.

(a) EFTEM micrographs and elemental maps, and (b) SEM micrographs with SEM-EDS elemental maps and spectrum of ET-SF-15.

Figure 8.
 ^1H - ^{29}Si heteronuclear correlation MAS NMR spectroscopy of (a) ET-SF-15 and (b) ETT-10-SF-15.

Figure 9.

(a) Stress–strain curves for a uniaxial compression test on silica–SF hybrid aerogels along with photographs of compression behavior of an optimized aerogel (ETT-20-SF-15) before and after applying the maximum load as well as its bendable flexibility by hand. Surface plot of (b) maximum compression strength, (c) maximum compressibility, and (d) bulk density variation against TMSPA and SF concentrations.

$$\rho_{\text{bulk}} = 0.19 \text{ g cm}^{-3}, S_{\text{BET}} = 660 \text{ m}^2 \text{ g}^{-1}, \varepsilon = 90 \%, D_{\text{pore, BJH}} = 10.7 \text{ nm},$$

$$V_{\text{pore}} = 4.8 \text{ cm}^3 \text{ g}^{-1}$$

Figure 10.

Microstructure of 3D-printed silica-SF aerogel hybrids (ET-SF-15) with selected physical properties. (a) 3D-printed ET-SF-15 grid-like architecture. (b) Microstructure of the overall 3D-printed construct indicating the grid size and wall thickness. (c) Top view of the grid wall region in b. (d) N₂ adsorption-desorption isotherm for 3D-printed ET-SF-15.

Figure 11. Stress-strain curve of three-point bending (flexural) tests on the printed silica-SF aerogel hybrid.

Figure 12.

(a) Thermal conductivity of some representative silica–SF aerogel hybrids; (b) ET-SF-15 burning behavior over time.

Table 1
Composition and Properties of Synthesized Silica–SF Aerogel Hybrids in the Present Study

aerogels	SiO ₂ in the sol [wt/v%]	SF ^a	ρ_{bulk} [g cm ⁻³] ^b	ρ_{skeleton} [g cm ⁻³] ^c	gelation time [h]	ϵ (%) eq 1 ^d	compressive strength, δ_{max} [MPa]	E modulus [MPa]
ET-SF-15	9.7	15	0.130 ± 0.03	2.44 ± 0.1	1	94.7	2.17	11.57
ET-SF-30	9.7	30	0.115 ± 0.01	2.43 ± 0.3	1	95.3	3.15	38.46
ETT-10-SF-15	9.7	15	0.190 ± 0.03	2.34 ± 0.3	7	90.5	2.34	28.61
ETT-10-SF-30	9.7	30	0.150 ± 0.01	2.14 ± 0.4	7	91.4	1.22	6.4
ETT-20-SF-15	9.7	15	0.170 ± 0.02	2.26 ± 0.2	10	90.7	7.45	13.55
ETT-20-SF-30	9.7	30	0.140 ± 0.03	2.32 ± 0.3	10	92.3	1.19	4.7
AeroSF	0	30	0.020 ± 0.01	3.59 ± 0.1	30	99	0.33	0.09

^a SF content is reported with respect to the silane [wt/v]% in the sol mixture.

^b Bulk density (ρ_{bulk}); the values are the average of three replicas.

^c Skeletal density (ρ_{skeleton}); the values are the average of five measurements of one replica.

^d Porosity (ϵ %, calculated by eq 1) of the synthesized aerogels.