On the role(s) of additives in bioinspired silicification

Siddharth V. Patwardhan,*[a] Stephen J. Clarson[b] and Carole C. Perry[a]

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Biological organisms are able to direct the formation of patterned and hierarchical biomineral structures. Extractable organic materials have been found entrapped in diatom, sponge and plant biosilica, some of which have been isolated by selective chemical dissolution methods and their composition and structure studied. Information gained from the bioextracts has inspired materials chemists to design biomimetic analogues and develop bioinspired synthetic schemes for silica formation. The results obtained from bioinspired silicification investigations are hypothesised to arise from specific modes of action of the organic additives, which are described in this review. Specifically, additives in bioinspired silicification act either as catalysts, aggregation promoting agents or structure-directing agents or more typically, exhibit a combination of these behaviours.

1 Background

Silicon, which is the second most abundant element in the earth's crust, in combination with oxygen, forms a variety of silica and silicate minerals. Silica ($SiO_2 \cdot nH_2O$) is one of the most technologically important inorganic compounds. It is extensively used for a wide range of commercial applications as catalyst supports, as separation media, as fillers in polymeric items and in biomedical applications.[1–5] It has been estimated that the global market for silica is around two billion dollars per year[2] thus emphasising its significant role in everyday life. The structure of silica (from molecular to macroscopic levels) governs its properties which in turn determines its end-uses. The preparation of synthetic precipitated silica, which typically

*Siddharth.Patwardhan@ntu.ac.uk

Siddharth V. Patwardhan

Dr Siddharth V. Patwardhan was born in Pune, India (1979). He obtained his first degree in engineering from the University of Pune, India (2000). Under the guidance of Professor S. J. Clarson, Siddharth worked on bioinspired silica synthesis and received Master of Science (2002) and PhD (2003) degrees from the University of Cincinnati, USA. He was appointed as a Visiting Scholar and Postdoctoral Fellow at the University of Delaware where he worked on developing strategies for self-healing glass fibres using biomineralization techniques. Currently, he is a Postdoctoral Research Fellow at Nottingham Trent University, and works on protein–silica interactions under the supervision of Professor C. C. Perry. Dr Patwardhan has published several articles and inventions on bioinspired materials. His research interests include biomineralization, nanostructured materials and polymerisations in supercritical carbon dioxide. Siddharth is a life member of MENSA and a Member of the Royal Society of Chemistry (MRSC).

Stephen J. Clarson

Stephen J. Clarson was born and educated in England but currently resides in Cincinnati. He obtained his doctoral degree in chemistry at the University of York, England in 1985 where he studied with the late Tony Semlyen. He then spent the summer at the Institute of Macromolecular Chemistry, Prague before taking up a postdoctoral appointment with Professor James Mark. In the spring of 1988 he joined the faculty of the Department of Materials Science and Engineering at the University of Cincinnati. During the period 1992–1998 he was the Associate Director and then Director of the Polymer Research Center and from 1995–2000. Dr Clarson served a five year term as Assistant Dean for Educational Development in the College of Engineering. He is currently Professor of Chemical and Materials Engineering at the University of Cincinnati.
In January 2000, Dr Clarson was elected Fellow of the Royal Society of Chemistry (FRSC). He has received numerous awards for his teaching. Dr Clarson has published over one hundred technical articles and co-authored the books 'Siloxane Polymers' (1993), 'Silicones and Silicone-Modified Materials' (2000) and 'Synthesis and Properties of Silicones and Silicone-Modified Materials' (2003). The research carried out in his group has led to a number of inventions and he holds five US patents and two Japanese patents. He has been PI or CO-PI of research awards totalling over $4 M in the past ten years and his current scientific research interests include materials chemistry, polymer synthesis, silicon-based chemistry, biomaterials, opto-electronic materials and surface science.

employs non-aqueous or aqueous sol–gel routes and occurs at relatively low temperatures,[3] generally results in poor control over the structure and processing of the product (see section 2).

Biological organisms, in contrast, are able to uptake, store and process soluble silicon (in an as yet unknown form) and mould it with great sophistication into ornate hierarchical patterned biosilicas.[6–13] As the resulting wide range of biogenic silica structures is highly species-specific, their formation is presumably genetically controlled. Fig. 1 shows some examples of intricately patterned biogenic silica (or biosilica) as observed for a species of diatom, sponge and higher plant respectively. Diatom biosilica forms a cell wall, which offers protection to the cell, while biosilica spicules in sponges provide mechanical support as well as protection from predators.[7] Silicic acid or silica in plants, on the other hand, has been shown to be essential for plant growth and it provides mechanical strength and resistance against attack by fungi and insects.[1,14–16] The process of biosilicification is not described in significant detail in this review and readers are advised to refer to specialised texts.[6,11,17,18] The major differences between biological and bioinspired silica formation can be understood if we compare the following aspects of mineral formation—precursor concentrations, pH at which (bio)silica polymerisation occurs, temperature, time required for (bio)silica deposition, control

Carole C. Perry

Professor Carole C. Perry obtained her doctoral degree in chemistry at Oxford University (1985) under the supervision of Professor R. J. P. Williams, F. R. S. She spent one month in the laboratory of Professor B. E. Volcani at the Scripps Institute of Oceanography, La Jolla, California and then worked as an E. P. A. Brereton-Sherman Junior Research Fellow at Oxford (1985–1987) before taking up a permanent position at Brunel University as a lecturer in inorganic chemistry. Professor Perry spent six months working with Professor S. Weiner at the Weizmann Institute, Israel (1992). She was appointed as Senior Lecturer at The Nottingham Trent University in 1993. She was Visiting Professor at Universite Pierre et Marie Curie, Paris VI and College de France in the laboratory of Professor J. Livage in 2002. Carole C. Perry is currently Professor of Bioinorganic and Materials Chemistry and Head of the Division of Chemistry at Nottingham Trent University. Carole C. Perry is Member of Royal Society of Chemistry (MRSC) and Fellow of the Royal Microscopical Society (FRMS). She has extensively published articles and reviews on a range of biological materials related topics. Professor Perry is a member of the EPSRC Chemistry College (2003–2005). Her current scientific research interests include in vivo and in vitro silicon–biomolecule interactions, biomaterials, sol–gel derived materials, super-hydrophobic and super-hydrophilic surfaces, developing new materials using specific Al containing clusters and proteins, polymer composites and correlation analysis methods in the study of material structures.

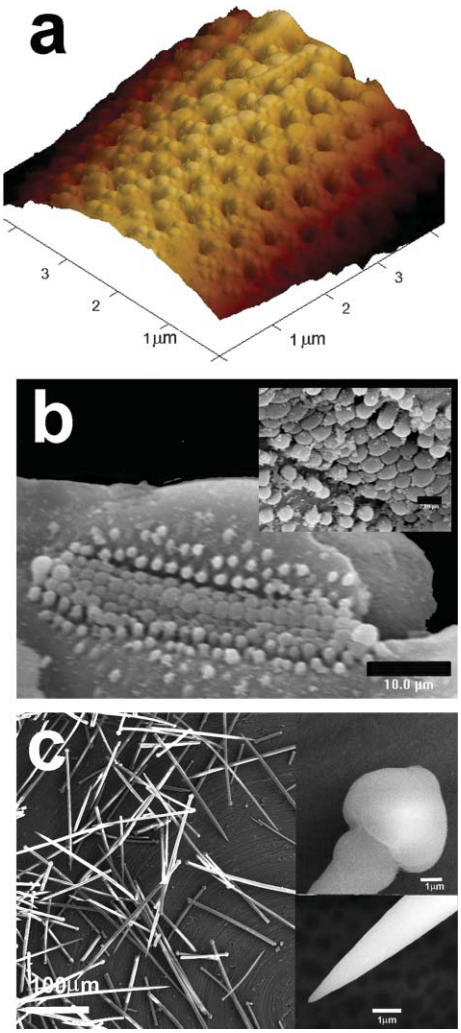


Fig. 1 Micrographs of ornate biosilica patterns. (a) Atomic force microscopy (AFM) image of diatom biosilica (*Aulacoseira granulata*). Diatom kindly provided by Professor Miriam Steinitz-Kannan, image taken by Howard, Franklin and Clarson. Scanning electron microscopy (SEM) images of biosilica from (b) higher plant (*Equisetum telmateia*; images taken by Mr David Belton) and (c) sponge (*Suberites domuncula*) spicule, insets show high magnification images of spicule tip and head (spicules kindly provided by Professor H. C. Shröder and Professor W. E. G. Müller). Bars (a) as shown, (b) 10 μm (2 μm for inset) and (c) 100 μm (1 μm for inset).

over the process and product, involvement of other molecules (ions, organic molecules, membranes, *etc.*) and reproducibility. Biosilicifying organisms typically start with an undersaturated solution of silicic acid from their environments, although it is proposed that these organisms have means of concentrating 'silicon' and storing it.[12] On the other hand the precipitation of silica *in vitro* from an undersaturated solution of silicic acid is yet to be demonstrated. Preparation of silica synthetically typically requires acidic conditions (for the synthesis of gels) or moderately high pH (for the preparation of particulate silicas). Biosilicification in contrast, has been proposed to occur at mildly acidic to neutral pH.[19] As evident from Fig. 1, biological silicification imposes great control over the process of silica formation as well as on the form of the biosilica

generated, while controlled *in vitro* silica synthesis has been only recently realised.[13] Other possible regulating factors present in biological organisms include micro-tubules and filaments as well as cell organelles, although their roles are not yet fully understood.[20] Another major difference between biological and bioinspired silicification is that the former takes place in a dynamic, confined environment (at least for diatoms) and is genetically controlled.

The organisation of this review is as follows. Before a discussion of the roles of additives in bioinspired silicification, a brief summary is provided on silicic acid polymerisation (section 2) and bioinspired silicification (section 3). In section 4, the importance of additives in silica formation is described and their roles described individually in subsequent sections (section 5–7). The review will conclude with a short summary indicating limitations to our current understanding and future scope of this field.

2 Silicic acid polymerisation

Silica can be treated as an inorganic polymer composed of SiO_4 tetrahedra with variable Si–O–Si bond distance and angles.[5] The chemistry of silica and the process of silicic acid polymerisation from aqueous solutions has been reviewed in detail elsewhere.[1,21] Briefly, a variety of precursors can be used to generate orthosilicic acid $Si(OH)_4$ (pK_a ~9.8) in water. Silicic acid at 25 °C is stable at levels below *ca.* 100 ppm. At higher concentrations, polymerisation occurs, which involves three distinct stages:

1. Polymerisation to form stable nuclei.
2. Growth of nuclei leading to fundamental particles.
3. Particle aggregation to form branched networks, larger particles or other structures.

The kinetics of silica polymerisation have been extensively studied using the colorimetric silicomolybdate method.[1] The polymerisation of monosilicic acid in an aqueous phase is somewhat different to classical polymerisation reactions that are used to produce organic polymers.[1] In organic polymerisation reactions, the polymer chains grow by a specific mechanism such as step growth or chain growth. In the case of silicic acid polymerisation, in the early stages, rapid condensation reactions yield a range of oligomers which serve as nuclei. Typically these stable nuclei are of size 1–2 nm, possess an anhydrous SiO_2 core and surface silanol groups (Si–OH). Based on the particle size the particle composition can be estimated as described by Iler.[1]

The addition of small oligomers on to these nuclei and coalescence of these nuclei leads to particle growth. The particles can grow by aggregation, by Ostwald ripening and/or by 'necking' between coalesced particles. Assuming instantaneous coalescence, aggregation of particles is typically a consequence of collision between smaller particles ($v_i + v_j \rightarrow v_k$, where v is volume of a given particle). The rate of aggregation (dv/dt) depends on the collision frequency, which is a function of the transport properties of the medium, pH, temperature and particle concentration.[22,23] As polymerisation proceeds, the pK_a of the particles/polysilicic acids decreases. It is important to note that even at circumneutral pH, the silica particles bear a negative charge in solution. The surface charge thus plays an important role in particle aggregation. On the other hand, in Ostwald ripening, soluble species such as monomers and dimers precipitate on relatively large and insoluble particles contributing to their growth. In the case of necking, due to a negative radius of curvature at the point of contact between two particles, silica from the surface of the bulk dissolves and re-precipitates at the point of contact between the particles.[1]

3 Bioinspired silicification

In the current climate where it is necessary to develop niche markets in order to get a return on investment, it is demanding to look for alternative and often more reliable routes to produce materials with controlled structures. Such high-cost and better-designed materials can be targeted by learning from biological systems that undertake biosilicification, then developing bioinspired methodologies suitable for silica synthesis, perhaps under mild physico-chemical conditions but with increased sophistication.[24]

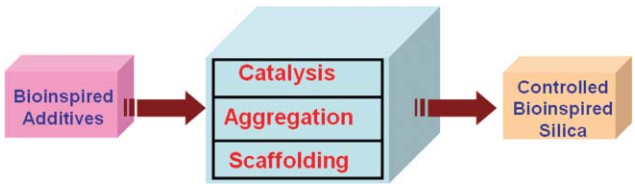
In order to reveal and understand nature's secrets surrounding biosilicification, scientists have been systematically studying biosilica formation in diatoms, sponges and grasses.[6] Several investigations have involved the selective removal of biosilica and isolation of biomolecules (organic material) associated with the biosilicas.[25–30] These isolated biomolecules have been further studied in order to determine their structure and demonstrate their ability to facilitate silicification *in vitro*. Table 1 summarises the information about bioextracts obtained from various biosystems and their proposed role(s) in (bio)silicification.

4 Role(s) of additives

The processes of silicification and the resulting product yield, composition and morphology can be controlled by the presence of additives.[1] Additives can affect all aspects of silica deposition from the rates of formation of small oligomers and stable nuclei through to macroscopic properties including

Table 1 Bioextracts from biogenic silicas and their proposed or observed role(s) in (bio)silicification

Biological system	Bioextracts	Proposed role in (bio)silicification	Ref.
Diatoms	Proteins and carbohydrates	Catalysis of siloxane bond formation *via* C–O–Si between Ser/Thr and silanol	25
	Glycoproteins	Stabilisation of silica thus creating local supersaturation of silicic acid	26
	Silaffins and long chain polyamines	Formation of spherical particles and particle networks *in vitro*	30
Sponges	Silicatein proteins	Catalysis of silica polymerisation *in vitro*	28,31,32
Grasses	Proteins and carbohydrates	Increased rates of catalysis of silica polymerisation, control over nucleation and growth, and produced crystalline silica	21,27,29,33



Scheme 1 Schematic presentation of the role(s) of additives in bioinspired silicification. The additives may facilitate the bioinspired silica synthesis by catalysing, by promoting aggregation, by scaffolding or by a combination of these mechanisms.

surface area, pore structure, particle size and aggregation patterns. Thus additives ranging from simple inorganic or organic molecules to complex organic polymers, can be used as powerful tools for the controlled production of silica.[5] The use of organic molecules as templates for the synthesis of inorganic minerals, silicates in particular, having a variety of shapes and morphologies has been demonstrated.[53–57] Iler documented the role of organic additives presented in studies before 1979.[1] During the past several years however, bioinspired synthetic approaches, which enable silica synthesis under ambient conditions, have been developed as a consequence of isolating bioextracts from biosilica-forming organisms. Bioinspired strategies make use of organic biomolecules and their synthetic analogues. Silica syntheses in the presence of such additives obey certain rules, which are discussed in this review. It will be shown here that, based on the investigations of several research groups, the additives control *in vitro* bioinspired/biomimetic silicification by either catalysis, aggregation, structure direction (templating/scaffolding) or a combination thereof (see Scheme 1). Each of these effects will be discussed and representative examples given. A list of peptide-based and synthetic macromolecule-based additives and their proposed effects on bioinspired silicification is presented in Table 2. It should be noted that bioinspired silicification reactions are typically carried out in aqueous media under ambient conditions and circumneutral pH. However, the precursors that are used range from alkoxysilanes, silica complexes and silicate solutions, all of which produce 'silica' containing solutions where the starting point for the subsequent reactions is very different.

5 Additives as catalysts

The source of silica for biological systems in their respective environments is dissolved silicic acid [$Si(OH)_4$] and/or its ionised forms. In the oceans silicic acid exists at *ca.* 70 μM concentration and hence is stable.[58] Organisms take up silicic acid from the environment, store it in an as yet unknown form and then process it into biosilica[6] (which is typically described as hydrated $SiO_2{\cdot}nH_2O$[59]). In contrast, low temperature sol–gel silica synthesis typically starts with a silicate or alkoxysilane precursor, which upon hydrolysis under appropriate conditions (generally acid or base catalysed) liberates silicic acid that undergoes subsequent polymerisation.[1,3] The process of silicic acid polymerisation has been described above. By definition, a catalyst typically increases the rate of a reaction by lowering the activation energy barrier and remains chemically unaltered. In order for the additives to act as catalysts in silicification, they need to promote the condensation of silanol groups (≡Si–OH) leading to the formation of siloxane bonds (≡Si–O–Si≡) with elimination of water.

A computational model has been developed in order to understand the interaction(s) between the organic biomolecules or the organic matrix and biosilica (its precursors and/or intermediates).[60,61] The model assumes that the organic matrix controlling biosilicification in diatoms adopts a beta-sheet like conformation and contains hydroxyl-rich amino acids (serine, threonine and tyrosine).[25] Polycondensation of silanol groups from silicic acid molecules has been modelled. From the calculations which indicated that the overall reaction could result in a net stabilisation of −28.0 kcal mol^{-1} (117.04 kJ mol^{-1}), it was proposed that the organic matrix may be involved in the catalysis of siloxane bond formation.[61] In particular, the formation of C–O–Si bonds between two

Table 2 (Poly)peptides and synthetic macromolecules as additives in bioinspired silicification

Additive	Role in (bio)silicification	Ref.
Poly-L-lysine	Silica precipitation and formation of novel structures	24,34–39
Poly-L-arginine	Precipitation of gel-like and particulate silica	34,36,37,40,41
Poly-L-histidine	Formation of nearly spherical silica particles	42
R5 peptide (a 19 amino acid sequence derived from the diatom *C. fusiformis*)	Spherical silica particles, fibre-like silica structures and silica patterned polymeric hologram	30,40,43
Mutants of R5 peptide	Precipitation of particulate silica only when peptide contained terminal RRIL motif	44
Poly[(L-alanine)$_{30}$-*b*-(L-lysine)$_{200}$]	Non-ordered silica	45
Poly[(L-glutamine)$_{30}$-*b*-(L-lysine)$_{200}$]	Non-ordered silica	
Poly[(L-serine)$_{30}$-*b*-(L-lysine)$_{200}$]	Non-ordered silica	
Poly[(L-tyrosine)$_{30}$-*b*-(L-lysine)$_{200}$]	Non-ordered silica	
Poly[(L-cystein)$_n$-*b*-(L-lysine)$_m$] n = 10, 30 or 60; m = 200 or 400	Spheres, elongated globules and columnar silica	
Poly[(L-cystein)$_{30}$-*b*-(L-glutamate)$_{200}$]	Non-ordered silica	
Peptides obtained from biopanning	Sphere-like silica	46
Genetically engineered proteins	Spherical, elongated and sheet-like silica particles	47,48
Poly(allylamine hydrochloride)	Spherical and elongated silica particle formation	49,50
Polyallylamine	Precipitation of sphere-like silica	36
Polyethyleneimines	Formation of nearly spherical particles	36,51
Lysine oligomers	Controlled catalysis and aggregation	38
Cellulose	Stabilisation of primary particles	52

adjacent serine functionalities and two molecules of silicic acid (which is still open to debate[62–64]) was proposed. Thus the organic moieties were thought to catalyse the condensation of silanol groups between adjacent silicic acid molecules.[25,60,61]

Proteins, named silicateins, have been extracted from the sponge *Tethya aurantia*, sequenced, and their role in (bio)-silicification studied.[28,31,32] From *in vitro* experiments using tetraethoxysilane (TEOS) as the silica precursor, silicatein protein was proposed to promote the hydrolysis and condensation of TEOS at neutral pH, a process which is typically acid or base catalysed.[1] It was proposed that three active residues, serine, histidine and asparagine, catalyse the hydrolysis of TEOS by a typical acid–base catalysis (see Fig. 2).[31] Further experiments by site-directed mutagenesis of the silicatein protein, wherein either serine-26 or histidine-165 (both proposed to be at the active site) were replaced by an alanine residue, revealed that both of these amino acids were essential for catalysing the hydrolysis of TEOS at neutral pH.[32] However, it should be noted that the method used for the analysis of ''catalysed'' silicification was as follows. After a given time, any precipitatable material was obtained by centrifugation from the reaction mixture (TEOS and protein solution). The amount of precipitated silica was then analysed by first digesting the silica and then treating the digest with molybdic acid solution. Although this method can determine small levels of precipitated silica, it fails to measure any catalytic effect during the course of the reaction. In other words, this protocol fails to differentiate between whether the protein is able to catalyse siloxane bond formation or facilitate aggregation to give large enough particles that can be collected by centrifugation.

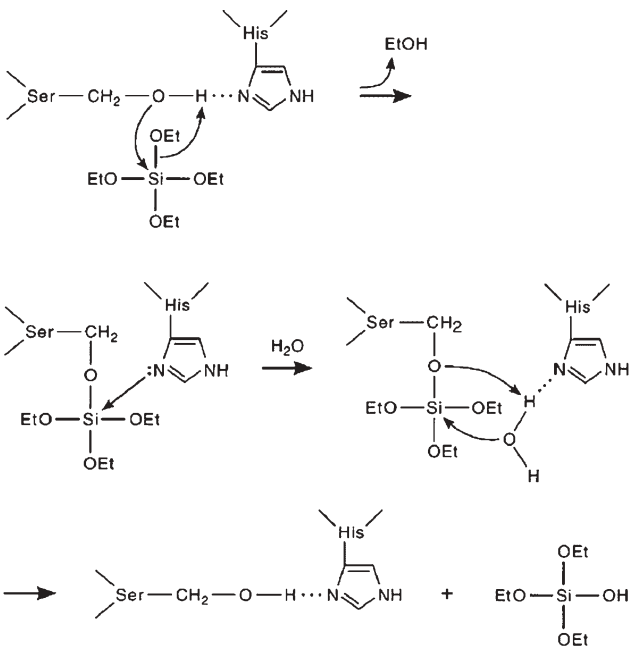


Fig. 2 Schematic presentation of the proposed mechanism for the acid–base catalysis of TEOS *in vitro* in the presence of silicatein proteins (taken from Zhou *et al.*[32] and reproduced with permission from Wiley-VCH).

The kinetics of *in vitro* silica synthesis have been recently studied in detail using the R5 peptide (a nineteen amino acid polypeptide derived from one of the silaffin proteins isolated from the diatom—*Cylindrotheca fusiformis*) and three genetically engineered proteins with known secondary structures (α helix rich, β sheet rich and random coil).[47,48,65] Kinetic experiments carried out using the silicomolybdate assay revealed that these additives were able to increase the rate of trimerisation in the silicic acid polymerisation.

In a recent investigation, Belton *et al.* studied a series of amino acids and lysine oligomers (monomer to pentamer) and polymer for their role(s) in bioinspired silica synthesis.[38] Lysine oligomers were found to increase the rate of the trimerisation reaction, but the additives did not contribute significantly in the later stages of the silica polymerisation. Moreover, the increase in this rate was proportional to the lysine oligomer chain-length.[38] This might indicate that these biomolecules exhibit selective control over the process of silicification *in vitro* and this may also be applicable to the case of biosilicification wherein certain biomolecules may only regulate a part of the process of biosilica formation.

It is thus clear that some biomolecules are able to catalyse silica formation at neutral pH under ambient conditions, which would otherwise not be possible. Although there may be a number of examples of biomimetic additives that can catalyse silica polymerisation, either partly or entirely, there may be some (bio)molecules that may not catalyse but facilitate silica synthesis by some other means (see below).

6 Aggregation promoting additives

Oligomers that are formed during the silica polymerisation process are negatively charged at neutral pH; and also possess free hydroxyl groups.[1] It is thus likely that the presence of an additive may alter the stability of such intermediates and/or the silicic acid polymerisation process due to electrostatic and/or chemical interactions (other than catalysis). Specifically, the attraction between positively charged (bio)molecules and negatively charged silica species, and hydrogen bond formation between silanol groups and proton donors (such as free amines present in the additives) may drive silicification. Information to support these possibilities has been obtained from *in vitro* studies of biosilica associated macromolecules and from model studies.

Biochemical studies of the diatom *C. fusiformis* have isolated phosphorylated native silaffin proteins that have been found to be active for *in vitro* silica formation. Based on time-dependent microscopy studies and phosphorus nuclear magnetic resonance spectroscopy (NMR), it was proposed that the silaffin proteins form supramolecular structures which in turn promote the aggregation of silica.[66] Further analysis of the R5 peptide (derived from silaffin proteins) and its mutants revealed that this peptide, due to its particular terminal amino acid sequence, forms aggregates in solution which promote the precipitation of particulate silica.[44] It is clear that the R5 peptide plays a dual role in silicification *in vitro* in that it not only catalyses the formation of Si–O–Si linkage but also facilitates the aggregation of silica.

Cationic polyelectrolytes are also candidates for aggregating silica species with subsequent phase separation. It is thought that the aggregation of silica species in the presence of cationic additives could enhance the condensation of silanol groups thus driving silica polymerisation forward. An elegant model has been proposed by Coradin and Lopez based on this hypothesis (see Fig. 3).[67] In particular, a range of cationic macromolecules have been investigated individually which include lysine polymers of varying chain length, poly-L-arginine, poly(allylamine hydrochloride) and polyethyleneimine. These cationic polymers were found to dramatically promote the aggregation of silica (for a detailed list of cationic additives used in bioinspired silica synthesis see ref. 13 and Table 1 in ref. 39).[13,24,34–41] Some of these cationic additives, poly(allylamine hydrochloride) in particular, were found to only facilitate aggregation and did not contribute significantly towards catalysis of silicic acid condensation, for example no effect was seen on the trimerisation reaction.[65] This later observation again demonstrates the specific interaction between additives and silica, and perhaps their importance in only selective stages of silicic acid polymerisation.

7 Additives as structure-directing agents

A structure directing agent should be able to direct the organisation of matter. Structure direction, which can be on any length scale, can be either three dimensional (scaffolding) or two dimensional (templating). The use of organic templates such as surfactants in the synthesis of porous silica has been demonstrated and is not described herein.[53–55] This section of the review will deal with the role of bioinspired additives (analogous to or inspired from bioextracts isolated from biosilicas) which act as structure-directing agents in silica synthesis. Structure direction in bioinspired silicification has usually been associated with either catalysis or aggregation (see above). (Bio)molecules may self-assemble into a certain structure and then catalyse silica formation. As an example, silicatein proteins extracted from the sponge *T. aurantia* were assembled into fibrous structures that were used as supports to generate silica structures by the hydrolysis and condensation of tetraethoxysilane (TEOS) on their surfaces. The denatured protein, which loses its spatial structure, was unable to facilitate silica formation in the presence of TEOS.[31] *In vivo*, silicatein proteins assemble into fibres that act as scaffolds for biosilica deposition leading to the formation of needle-like glass spicules with an occluded central silicatein filament.[6,68,69]

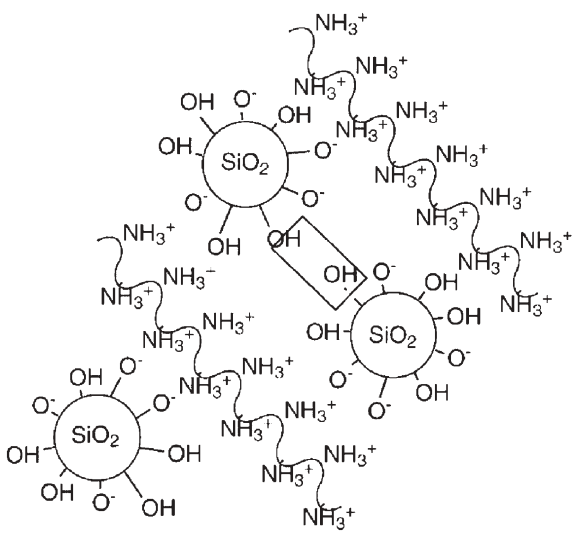


Fig. 3 Schematic representation of the role of cationic polyelectrolytes as aggregation promoting agents in bioinspired silica synthesis proposed by Coradin and Lopez[67] (reproduced with permission from Wiley-VCH).

Another example for a templating effect can be given when considering a bioextract from the higher plant *Equisetum telmateia*.[29,33] The bioextracts, when present in the silicic acid system, were able to increase the rate of condensation of silicic acid to form small oligomers such as trimers. In addition, the bioextracts were able to control the primary particle sizes and produced crystalline silica under ambient conditions. It was proposed that these bioextracts, which were rich in β-sheet/turn forming amino acid residues, formed a particular assembly that templated the formation of the crystalline silica structures. These crystalline silicas were similar to quartz, a mineral that is usually produced by high temperature/pressure procedures.

Synthetic functional macromolecules may also self-assemble into three-dimensional structures with defined 'shapes' in solutions that may direct silica formation *in vitro*. As an example, polyelectrolytes behave in unusual ways in solution due to their charged nature and sometimes generate specific assemblies.[70] Studies of the behaviour of poly-L-lysine and poly(allylamine hydrochloride) in solution using small angle light scattering (SALS) have revealed that these (bio)macromolecules adopt a particular conformation, which is dependent on the salts present and the concentration of the (bio)macromolecule.[40,71] This self-assembly has been exploited in the design of new silica structures.[24] In particular, poly-L-lysine was found to direct the formation of spherical, petal-like and sheet-like hexagonal silica. It was proposed that poly-L-lysine self-assembles in a particular fashion that guides the formation of such novel structures (see Fig. 4).

8 Concluding remarks and future outlook

Bioinspired/biomimetic chemistry has now become a separate branch of materials chemistry, wherein lessons learnt from biological systems are implemented into *in vitro* syntheses. In the quest for novel chemistries for the preparation of silica synthetically, a *ca.* two billion dollar annual market, researchers have focused their attention on biological organisms that process and organise soluble 'silicon' into highly intricate biosilica structures with great control and reproducibility. Recent investigations have now started to unveil the mechanisms underpinning biosilicification. Organic biomolecules such as proteins and carbohydrates play important role(s) in biosilicification. In order to understand the interactions between these biomolecules and the biosilicas generated in various organisms and also to make use of any understanding so gained, *in vitro* experiments have been designed. Bioinspired analogues of biosilica bioextracts have been used for such bioinspired studies.

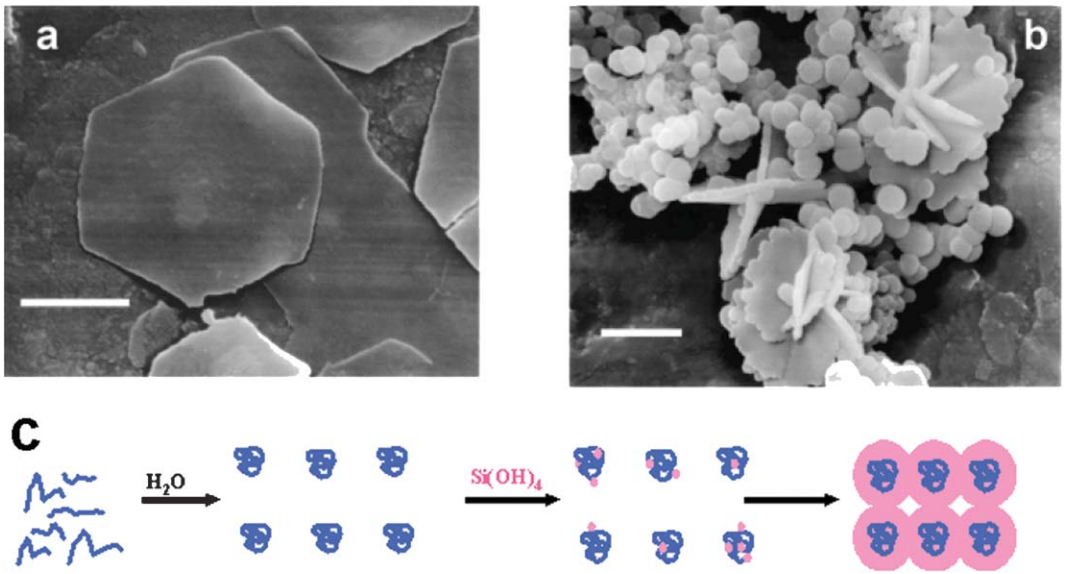


Fig. 4 Scanning electron micrographs of (a) hexagonal and (b) petal-like silica structures synthesised by poly-L-lysine which acts as a scaffold (bar = 1μm). (c) Schematic presentation of the role of polyelectrolytes in bioinspired silicification, wherein they self-assemble and template silica formation[24] (reproduced and adapted by permission of the Royal Society of Chemistry).

Recent efforts have begun to reveal the role(s) of various additives in bioinspired silicification. These roles can be largely categorised as catalysing additives, aggregation promoters and structure-directing additives (or molecules exhibiting a combination of these abilities). Some additives catalyse a distinct stage of the silicic acid polymerisation process, *e.g.* silicateins have been proposed to catalyse the hydrolysis and condensation of TEOS.[31] On the other hand, cationic polyelectrolytes such as poly(allylamine hydrochloride) promote the aggregation of the growing silica species while they appear to contribute only marginally towards catalysis.[50,65] Scaffolding silica on assembled structures of (bio)molecules constitutes a third mechanism. Poly-L-lysine, for example was found to assemble into three dimensional structures, the structures concerned leading to the formation of silicas with novel form.[24]

In order to clearly identify the role(s) that a given additive plays in both biological and bioinspired silicification, it is necessary to standardise assessment techniques and protocols. For example, we need to be able to clearly monitor catalysis and distinguish it from aggregation. As discussed above, the use of precipitated silica in studying the catalytic effect of an additive may be misleading and hence may not be suitable. In addition, as the additives in most cases become occluded into the final structures, it is necessary to answer questions such as—although the additive cannot be regained, is it a catalyst by definition?; does the additive retain its chemical structure so that it can be termed a catalyst? Furthermore, bioinspired silicification is viewed as a process that occurs at circumneutral pH. However, it can be argued that additives such as polyamines form local domains of basic pH wherein silica polymerisation occurs. In such cases, one must ask the question—is this process correctly termed as a process at neutral pH or should one be considering micro- or nanoscale local environments? It is hoped that as more research is performed, these issues will be carefully addressed.

As we have started to understand the molecular basis of the underlying mechanisms regulating (bio)silicification, it is now important to apply this knowledge in the development of novel materials. Future directions for this field can be categorised into molecular chemistry, theoretical chemistry, and engineering/technological applications.

Advances in molecular chemistry may include synthesis and use of designer biomolecules in generating bioinspired materials and the control of biomolecular architectures by self-assembly. Tailored (bio)molecules that will generate silicas with desirable properties can be designed. For example, peptides possessing particular primary and secondary structures, known chain lengths, and desired chemical properties such as pIs can be prepared.[72,73] Organising such functional molecules into novel assemblies will be the next challenge. The organisation can be in one-, two- or three-dimensions. In particular, materials can be synthesised to form a variety of architectures such as microvesicles and self-assembled monolayers (SAMs)[74–77] and larger scale structures such as those generated by lithographic techniques.[78] In principle, one could envisage combining three dimensional architectures with solution species to further fine-tune silica structures. As mentioned above, biological organisms that deposit biosilica are typically able to use a significantly undersaturated solution of silicic acid from their environments to prepare amorphous silica. To the author's knowledge, none of the additives described in this article have been used to induce the precipitation of silica from an undersaturated solution of silicic acid (<1 mM). Molecular modelling of the interactions between bioextracts and 'model' additives with silicic acid, polysilicic acids and particulate silicas may reveal more insights into the role(s) that these and such additives play in (bio)silicification.[62–64] In order to realise the technological impact of bioinspired methodologies, 'scale-up' studies of laboratory syntheses to pilot-plant and then plant scale

production will be highly fruitful. It will also be important to implement the knowledge gained from silica-based systems for the preparation of other oxides, salt and hybrid systems such as aluminium-based, calcium-based, germanium-based, titanium-based, gallium-based, zinc-based, lead-based compounds and other inorganic compounds thereby expanding our arsenal of materials with controlled structure and form.[13,24,36,40,79–87]

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Siddharth V. Patwardhan,*[a] Stephen J. Clarson[b] and Carole C. Perry[a]
[a]Interdisciplinary Biomedical Research Centre, School of Biomedical and Natural Sciences, Nottingham Trent University, Nottingham, UK NG11 8NS. E-mail: Siddharth.Patwardhan@ntu.ac.uk; Fax: +44 (0)115 8483384 (Fax); Tel: +44 (0)115 8486375
[b]Department of Chemical and Materials Engineering, University of Cincinnati, Cincinnati, OH 45221, USA

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