Silicon uptake by sponges: a twist to understanding nutrient cycling on continental margins

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About 75% of extant sponge species use dissolved silicon (DSi) to build a siliceous skeleton. We show that silicon (Si) uptake by sublittoral Axinella demosponges follows an enzymatic kinetics. Interestingly, maximum uptake efficiency occurs at experimental DSi concentrations two orders of magnitude higher than those in the sponge habitats, being unachievable in coastal waters of modern oceans. Such uptake performance appears to be rooted in a former condition suitable to operate at the seemingly high DSi values characterizing the pre-Tertiary (>65 mya) habitats where this sponge lineage diversified. Persistence of ancestral uptake systems causes sponges to be outcompeted by the more efficient uptake of diatoms at the low ambient DSi levels characterizing Recent oceans. Yet, we show that sublittoral sponges consume substantial coastal DSi (0.01–0.90 mmol Si m$^{-2}$ day$^{-1}$) at the expenses of the primary-production circuit. Neglect of that consumption hampers accurate understanding of Si cycling on continental margins.

Silicic acid, a biologically assimilable dissolved form of silicon (DSi), is a key ocean nutrient1–3. It fuels primary production by enhancing growth of diatoms, which polymerize DSi to elaborate their skeletons of biogenic silica (BSi). Increased photosynthesis in diatom populations decreases DSi, nitrate, and phosphate levels in surface waters and facilitates transfer of atmospheric carbon dioxide to the ocean, hence connecting silicon to carbon, phosphate, and nitrogen cycles4–6. Therefore, there is strong interest in predicting the interplay between DSi and BSi budgets and many efforts have been made during the last decades to unravel the route of Si through the oceans. In the current global model describing the Si cycle in the ocean, diatoms are thought to biologically dominate Si cycling, with other Si-consuming organisms, such sponges, radiolarians, choanoflagellates, and silicoflagellates, playing negligible roles7–9. Nevertheless, a concatenation of findings relative to the contribution by siliceous sponges8–11 has alerted us that the notion of a Si cycle exclusively revolving around diatoms incurs an unrealistic oversimplification that also neglects ocean history.

There have always been suspicions that the contribution of sponges to the marine Si cycle, even if never quantified in global terms owing to its complexity, could be of some importance12–13. More recent studies evaluating the contribution of sponges on continental-shelf and slope habitats have revealed that Si standing stocks in sponges may surpass the combined Si stock in living diatoms and ambient DSi of the diverse local systems investigated9–11,12. Moreover, upon death, sponge skeletons are far more refractory to dissolution than diatom frustules, irrespective of potential differences in skeleton surface area12,13–15. Consequently, benthic populations of siliceous sponges appear to function as BSi traps on continental margins, retarding recycling of BSi into DSi. A serious problem limiting our understanding of the magnitude of Si turnover through sponges is the current lack of data regarding uptake kinetics, with only a single available study to date11.

Fossil records and molecular clocks arguably suggest that sponges were already present in the Precambrian17–19, but the oldest unequivocal fossil spicules date back to the Lower Cambrian20, about 542 my ago. It means that the Si-consuming activity by sponges evolved before that of the two others major Si consumers characterizing modern oceans, namely radiolarians and diatoms. Radiolarians are also quite an old group, with arguable fossils from as early as the Lower Cambrian21 and the earliest unequivocal fossils dating from the Lower Ordovician, about 488 my ago22. Compared to sponges and radiolarians, diatoms are newcomers, with controversial remains reported from Jurassic sediments23 and unequivocal fossils from the Early Cretaceous24, about 140 my ago. Geochemical modeling25 and analysis of marine chert formations26–28 support the notion that the Precambrian and Cambrian oceans in which sponges and radiolarians thrived had average DSi concentrations ranging from 1 to 2.2 mM, that is, at least two orders of magnitude higher than the average (10 μM) in the Recent world ocean.
Likewise, from the Cambrian to the Jurassic, ocean average DSI concentrations have been estimated not lower than 650 μM \( \text{S}^{4,6,29} \). Those paleo-estimates along with an abundant fossil record of highly silicified spicules and tests \(^{27}\) support that the Si-uptake systems of sponges and radiolarians operated with notable success in waters with very high DSI concentrations for about 400 to 500 million years, until the evolutionary emergence and the subsequent ecological expansion of diatoms. There is a congruent body of evidence supporting that the expansion of diatoms during the Late Cretaceous and the Lower Tertiary drastically decreased DSI concentrations in surface waters, leading to the low values characterizing modern oceans, which appear to have experienced only minor variations for the last 60 my \(^{9}\). Such a decrease in DSI availability put a selective pressure on both sponges \(^{11,28}\) and radiolarians \(^{29–31}\) to evolve skeletons that required less silica. This process is well illustrated by the extant shallow-water demosponge *Crambe crambe* (Family Crambeidae, Order Poecilosclerida). Long-term exposure of this sponge to DSI concentrations much higher than those in its natural habitats induces secretion of not only thicker and larger spicules, but also additional types that are never produced in wild populations \(^{11}\). Such a response suggests that the genetic systems controlling Si-uptake and silicification are up-regulated by threshold DSI concentrations higher than those naturally available to the sponge. This opens the intriguing possibility that extant sponge species belonging to lineages that diversified before the expansion of diatoms may still silicify through uptake systems originally suitable to deal with the high DSI concentrations that seemingly characterized pre-Tertiary oceans. A better knowledge of the performance of these uptake systems could help us to understand the evolution of nutrient concentrations as well as that of the biosilicification process characterizing modern sponges. Additionally, by improving our knowledge on sponge Si uptake, we will be able to approach more realistically the role of these organisms on Si cycling in Recent marine ecosystems. To these aims, we have investigated Si-uptake kinetics in axinellid demosponges, members of a lineage with a fossil record thought to pre-date the Cretaceous-Tertiary boundary, as rhabdostyles and styles characterizing some axinellid sub-lineages have been reported from the middle Triassic (245 to 228 my ago) \(^{32}\).

**Results**

**Experimental DSI uptake.** Silicon uptake was investigated in individuals of three Atlantic-Mediterranean demosponges of the genus *Axinella* (fam. Axinellidae; order Halichondrida) collected from sublittoral north-western Mediterranean populations (see SI 1). The bulk of uptake data was obtained from individuals of *A. damicornis*, a species scattered in moderate density on sublittoral rocky walls. Additional information was obtained from individuals of both *A. verrucosa* and *A. polypoides*, which are rarer, to-be-protected species. One experiment (Exp. I) examined uptake responses of 10 individuals of *A. damicornis* and 3 of *A. verrucosa* to DSI concentrations that were increased, in 48h steps, from field values (1.6 μM) to 10, 20 30, 40, 50, 100, 150, and 200 μM, using sodium hexafluorosilicate (SFS) as DSI source. In response, average Si uptake by the set of assayed sponges progressively increased with increasing DSI, significantly fitting a linear regression (Fig. 1; \( n = 13, r = 0.978, P < 0.001 \)). After the 200 μM DSI step, sponges were transferred back to the natural concentration (1.6 μM) for a 5-day resting period, then exposed again to 20, 70, and 100 μM DSI experimental steps for 48h periods each. Sponges responded to these treatments after resting with readjustments in uptake rate that mirrored the marked shifts in DSI availability, showing at each concentration step an average uptake rate that fell within the 95% prediction interval of the previously calculated linear regression (Fig. 1). These results indicated that the Si uptake system of the sponges is able to react quickly and, more importantly, predictably to rapid, abrupt changes in ambient DSI.

![Figure 1](https://www.nature.com/scientificreports)  Relationship between DSI uptake and DSI availability during initiation of experiment I. The uptake-rate response by the sponges (μmol Si per h and ml of sponge) was linearly related to DSI concentration in the experimental bottles, whenever DSI availability ranged from natural values (1.6 μM) to 200 μM. Crosses are 3 further treatment steps (20, 70 and 100 μM DSI) conducted as a test after allowing sponges to rest for 5 days at natural DSI concentration. Note that all 3 test responses fell within the 95% prediction interval of the previously calculated regression equation.

After the 48h step in the 100 μM DSI treatment, sponges were exposed to 300 μM DSI and, surprisingly, their average uptake rate decreased (Fig. 2). Furthermore, average uptake rates at subsequent 400 and 600 μM DSI treatments were even lower (Fig. 2). When sponges were transferred from 600 down to 185 μM DSI, a concentration that had elicited nearly maximum uptake during the first part of the experiment, they showed no perceptible Si uptake (Fig. 2: dashed line). Such a physiological inactivity suggested that the sponges had somehow been damaged during treatments at 300, 400 and/or 600 μM DSI. Although no sponge died either partially or entirely during those treatments, slow progressive darkening of the bright yellow-orange skin (ectosome) was noticed, supporting our concern of potential sponge damage. We suspected that the use of SFS as DSI source could proof harmful when aiming for high DSI concentrations. When using SFS to increase DSI concentration by a factor of 1 in the experimental bottles, fluorine concentration (as fluoride) was concomitantly increased by a factor of 6. For instance, concentrations as well as that of the biosilicification process characterizing modern sponges...
Interestingly, the set of new sponges exposed to the 200 μM DSI treatment during Exp. II (Fig. 2) showed an average uptake rate (0.11±0.06 μmol Si h⁻¹ ml⁻¹ sponge) nearly identical to that measured for the previous set of individuals during the “healthy” 200 μM DSI step of Exp. I (0.12±0.09 μmol Si h⁻¹ ml⁻¹), confirming repeatability in the sponge responses. Experiment II also revealed that subsequent increases in DSI to 300, 600, and 800 μM neither stimulated nor decreased uptake rates, relative to the 200 μM DSI treatment (Fig. 2). We even run a final 72 h step at 850 μM DSI to examine the possibility that the uptake system could need several days to get adapted to those high concentrations. Nevertheless, such an extended treatment—resulting in a total of five days of exposure to 800–850 μM DSI—did not stimulate any significant shift in uptake relative to values achieved at 200 μM DSI. Therefore, the results strongly suggest that the uptake system becomes saturated somewhere around 200 μM DSI.

When the uptake responses of each sponge in the various DSI concentrations in both experiments were compared, considerable inter-individual variability was found (Fig. 3 a–b). It is worth noting that the few individuals of *A. verrucosa* and *A. polypoides* that we assayed for exploratory purposes did not show uptake rates substantially different from the bulk of individuals of *A. damicornis*, suggesting that interspecific differences in uptake kinetics, if any, should be minimal (Fig. 3 a–b).

Interspecific variability showed a consistent pattern, as individuals taking up faster at a given DSI concentration were also good performers at most other concentrations (Fig. 3 a–b). Maximum uptake rate (average±s.d.; recorded in both experiments at 200 μM DSI) was 0.12±0.07 μmol Si h⁻¹ ml⁻¹ (sponges of both experiments pooled; n = 26), but again it was affected by a large inter-individual variability, ranging from 0.002 μmol Si h⁻¹ ml⁻¹ in individual 10 to 0.387 in individual 6 of Exp. I.

At least some of the detected inter-individual variability can be attributed to size differences. Volume of assayed sponges (ranging from 4 to 37 ml) appeared to have a moderate effect on uptake rate, with smaller individuals (<9 ml) showing slightly higher rates on average than larger individuals (>9 ml). Such relationship between size and uptake can be perceived (even if diffuse) on Fig. 3, where green lines (large sponges) often run above blue lines (smaller sponges). Likewise, the relationship between saturated uptake rate (i.e., that at 200 μM DSI) and sponge volume grossly fitted an inverted, first order polynomial regression (sponges from both experiments pooled: n = 26, r = 0.600, P = 0.001; Fig. 4), confirming a subtle negative relationship between both variables. Interestingly, in nearly all DSI concentrations assayed (Fig. 3a), the highest uptake rates were achieved by individual 6, which was the second smallest (4.4 ml) sponge out of the 26 assayed (Fig. 4). In this individual, silica spicules largely protruded from the epithelium, making a velvety body surface visible by the naked eye and indicating that the sponge was involved in a more intense process of skeletal production than the other individuals.

When average uptake data from the "healthy" phase of Exp. I (i.e., DSI treatments <300μM) and Exp. II were pooled together and analyzed by non-linear regression, the equation better fitting the data was a hyperbolic function (Fig. 5; n = 16, r = 0.948, P < 0.001), similar to that of a Michaelis-Menten ligand-binding kinetic with one site saturation. Consequently, DSI uptake (V, μmol Si per hour and per sponge ml) by *Axinella* spp. can be regarded as an enzyme-mediated transport, with a half-saturation constant (Kₘ) of 74.47 μM and a saturated uptake rate (Vₘₐₓ) of either 0.13 μmol per hour and sponge ml or 1.74 μmol h⁻¹ g⁻¹, if expressed as ash-free dry weight (AFDW).

### Estimates of field DSI demands

Conservative field surveys along 100 km of the relatively sponge-poor and oligotrophic Mediterranean rocky sublittoral where *Axinella* spp. grew (see Methods and Supplementary Information: Section I) revealed that siliceous sponges average 0.34±0.52 L m⁻² (n = 100 quadrats) and ambient DSI 0.73±0.44 μM (n = 240 water samples over a year cycle). At that ambient DSI concentration, sponge uptake rate is...
predicted to average $1.31 \pm 0.79 \times 10^{-3}$ μmol Si per h and sponge ml (according to equation in Fig. 2). It means that the sponge fauna per m² of rocky habitat at this Mediterranean coast use yearly about $3.9 \pm 5.9$ mmol DSi. Such consumption represents yearly about $21.4 \pm 32.7\%$ of the average DSi available in a 30m overlying sublittoral water column and about $10.7 \pm 16.3\%$ of that in a 50 deep water column. Similarly, we have conservatively estimated (see Methods) on 21 km² of a Mesoamerican continental shelf (Belize) that the abundance of siliceous sponges averages $2.6 \pm 14.3$ L per m² of bottom. Mean yearly DSi concentration in the 25m-deep water column of such continental-shelf system is about $3.6 \pm 0.6$ μM13, being the sponge communities predicted to consume DSi at an average rate of about $14.5 \pm 0.35 \times 10^{-3}$ μmol per h and sponge ml (according to the equation in Fig. 2). It means that yearly sponge uptake is about $332 \pm 1826$ mmol DSi m⁻², and that it would virtually deplete that shallow Belizean shelf of DSi once every $98.9 \pm 83.7$ days, if there is no DSi replenishment. The estimated Si consumption rate by the poor (in volume) sponge fauna of the oligotrophic Mediterranean sublittoral is about $0.01 \pm 0.01$ mmol Si m⁻² day⁻¹, while that of the

Figure 3 | Summary of individual uptake data. Some variability was noticed in the individual uptake responses during the “healthy” phase of experiment I (a) and through experiment II (b). Green and blue lines indicate small (< 9ml) and large (> 9 ml) individuals of Axinella damicornis, respectively. Red lines indicate individuals of Axinella verrucosa or Axinella polypoides. Note that the sponge individuals used for experiments plotted in “a” and “b” graphs are different.

Figure 4 | Relationship between sponge size and uptake. A weak negative relationship between uptake rate (μmol Si h⁻¹ ml⁻¹) at 200 μM DSi and sponge size (ml) was detected, grossly fitting an inverted, first order, polynomial regression. The highest uptake rate corresponded to individual 6 of Exp. I.

Figure 5 | DSi uptake model for Axinella. Michaelis-Menten kinetics function modeling the relationship between Axinella spp. average uptake rate and ambient DSi in the experimental bottles, obtained after pooling data of experiments I and II.
Discussion

Our experimental approach revealed that sponges react to ambient DSi availability with a rapid and predictable response of their uptake rate. Interestingly, the assayed sponges achieved their highest uptake rates at concentrations around 200 μM DSi, that is, nutrient values that are not available to them in shallow waters of modern oceans and that are more than 100 times the yearly DSi average experienced by these animals in their natural habitats (see Supplementary Information: Section 1). Consequently, these sponges are suffering a severe, chronic limitation by DSi (<2μM) in their natural habitats. The idea that low DSi concentration in Recent shallow waters might limit sponges was originally disregarded35–37, as their Si-uptake system’s ability could have resulted from evolvement of the original uptake and transport process of direct polymerization of DSi into BSi inside the sclerocyte cells having a demonstrated role in the internalization of DSi from ambient seawater. Suggestions have been made that a sodium-bicarbonate co-transporting system could somehow be involved in taking up silicic acid from seawater46, which would be consistent with the detected enzymatic kinetics. Although active DSi uptake in diatoms has been shown to be supplemented by some passive diffusion across the cell membrane44, which is in direct contact with the ambient water, a similar diffusion process is unlikely in sponges. Silica secreting cells of sponges (sclerocytes) occur typically at the inner mesohyl regions of the sponge body, isolated from ambient seawater by epithelial cell layers and dense intercellular deposits of collagen and other macromolecules. Furthermore, the possibility that DSi can passively diffuse into the sponges has been discarded experimentally by demonstrating that DSi uptake rates are abated following sponge starvation, what indicates that most DSi internalization results from an active process that requires energy45. Likewise, when the epithelial cells of the Axinella individuals were sub-lethally poisoned with fluorine during our experiment I, rates of DSi uptake decreased inversely to DSi concentrations, corroborating that DSi transport is not a passive diffusion process (Fig. 2). The finding that smaller Axinella individuals incorporate DSi at higher rates than larger individuals (Fig. 4) also supports active DSi uptake, because the opposite pattern should occur under a passive diffusion model, particularly in these branching sponges (Fig. S1), in which surface area increases almost exponentially with increasing individual volume. Further evidence of non-diffusive uptake is provided by investigations on the relationship between the Si isotopic composition of ambient DSi and that of sponge spicules. Sponges are known to fractionate Si isotopes during their BSi production process46, with ΔSi (i.e., δSi_{sponge} - δSi_{seawater}) fractionation increasing (i.e., lower values) with increasing ambient DSi concentration59. More importantly, Si isotopic data in sponges show good agreement with a Michaelis-Menten function for Si uptake, what again supports uptake and δSi fractionation being biologically -rather than diffusively- controlled46. Likewise, isotopic analyses strongly suggest that DSi transport from the ambient water to the specific silicifying sites within the sponge body is well differentiated from the process of DSi polymerizing as BSi around the organic template, because the fractionation factor is constant during uptake transport, but it appears to decrease in value (i.e., more intense fractionation) with increasing DSi concentration during strictly the polymerization process59.

Ample between-individual variability in DSi uptake rate has been found in our study and it was also reported in H. panicea50,54. The physiological reasons behind such an inter-individual variability

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Table 1 | Average rates of Si use by communities of planktonic diatoms and sublittoral sponge communities in various marine systems. Diatom Si demands were originally measured as BSi production rates13, while sponge demands have been derived from Si uptake rates.

| HABITAT/ SYSTEM                  | Si DEMAND (mmol Si m⁻² day⁻¹) |
|----------------------------------|-------------------------------|
| **Planktonic diatoms**           |                               |
| Coastal upwelling                | 90                            |
| Other coastal conditions          | 15                            |
| Southern ocean                    | 15                            |
| Deep ocean                        | 2.3                           |
| World ocean average               | 1.6 – 2.1                     |
| **Sublittoral sponges**           |                               |
| Outer shelf of Belize            | 0.90                          |
| Baltic sublittoral bottoms        | 0.44                          |
| Mediterranean rocky bottoms       | 0.01                          |

richer Caribbean sponge assemblages would be 0.90±5.00 mmol Si m⁻² day⁻¹. Both sponge figures are lower than the demand average estimated for diatoms in diverse marine systems (Table 1).
remain unclear. Nevertheless, the level of variability detected in laboratory studies is consistent with many field studies reporting that neighboring conspecific sponges subjected to nearly identical environmental conditions in terms of food supply, oxygen, DSI, and others, exhibit puzzling differences in body growth rates—and implicitly in BSI production to skeletonize the new soft tissues—over months or years.

Si uptake by diatoms shows some fundamental differences with that of sponges. Planktonic diatoms arguably follow a Michelsen-Menten kinetics, saturating at 10 μM DSI on average and with a half saturation constant that ranges from 0.3 to 5 × 10⁻⁰⁵, except for some Antarctic species that have shown a Kₘ from 12 to 22 μM in laboratory cultures. Beside saturating at relatively low concentrations, planktonic diatoms are able to achieve high, diffusion-mediated uptake rates with non-saturable kinetics during short periods, which allows them to transiently store much DSI if high ambient concentrations are sporadically encountered. Diatom uptake uses an electronegative, sodium/silicic acid symporter, being also able to transport the ionized form SiO₄⁻². Consequently, Recent planktonic diatoms are finely tuned to work with maximum efficiency under relatively low DSI values. This tuning probably resulted from their genetic uptake system being more "plastic" than that characterizing sponges, so that it was able to evolve in an Early Tertiary ocean where ambient DSI concentrations were progressively decreasing owing to their own ecological expansion. The available evidence to date indicates that the genetic systems controlling biosilicification in sponges, diatoms, and Si-using plants are completely unrelated, corresponding to independent evolutionary acquisitions. Interestingly, by maintaining ambient DSI values low in the long run, planktonic diatoms favor proliferation of Si users with similar uptake kinetics (i.e., more planktonic diatoms), while limiting any other Si user characterized by a higher saturation constant, hence becoming strong competitors for sponges, radiolarians, and probably benthic diatoms (see Supplementary Information: Section 3).

Despite chronic DSI limitation of siliceous sponges owing to diatom overcompetition, measured uptake rates (which ranged from V₁μM = 0.001 to V₂300μM = 0.098 μmol Si per h and sponge mL) suggest that DSI consumption by sponges on coastal systems is of some relevance. We conservatively estimated average Si consumption rate by the poor (in volume) sponge fauna of the oligotrophic Mediterranean sublittoral in about 0.01 ± 0.01 mmol Si m⁻² day⁻¹, while that of the richer Caribbean sponge assemblages is about 0.90 ± 5.00 mmol Si m⁻² day⁻¹ (Table 1). Note that these calculations are not maxima, but conservative averages. They come from estimates of sponge biomass with enforced mechanisms to prevent overestimation (see Methods) and are based on highly-replicated field measurements scattered over large continental shelf areas, realistically including large extensions of habitats that are not favorable to sponges, as indicated by the large standard deviation values associated to mean sponge volumes per bottom area unit (i.e., 0.34 ± 0.52 and 2.6 ± 14.3 L m⁻² for the investigated Mediterranean and Mesoamerican continental shelf, respectively). Similarly, by using available uptake data for the seasonal Baltic populations of the sponge *H. panicea* and the mean biomass (20 ml m⁻²) in the less favorable sponge habitat, a conservative average consumption of 0.44 mmol Si m⁻² day⁻¹ during summer months may be arrived at. These Si consumption rates by sponges (Table 1) are somewhat smaller than the average BSI production by diatoms in the global ocean, estimated at 1.6–2.1 mmol Si m⁻² day⁻¹.

At present, it is impossible to estimate with any accuracy Si consumption by sponges in the world oceans—as will be the case for decades to come owing to the extremely variable distribution of sponge biomass on the continental margins at depths that prohibit extensive measurements of individual Si contents per bottom area. However, if the Si consumption rates herein estimated for sponges per unit area of bottom at the Mediterranean, the Baltic, and the Caribbean sea are extrapolated over the entire continental shelf of the earth (22 × 10⁶ Km²), a first, very tentative estimate of the global Si consumption by sponges could be suggested, falling somewhere between 8.6 × 10⁵ and 7.3 × 10⁶ mol Si year⁻¹. This figure would still be about two to four orders of magnitude smaller than the 2.0 to 2.8 × 10⁹ mol Si year⁻¹ estimated for diatoms. By incorporating into the calculations the large sponge populations that are being discovered at bathyal depths by the advent of ROVs and manned oceanographic submersibles, a generous— but unlikely to be ever globally quantified— increase of the yearly sponge Si demands herein estimated for only continental shelves might be arrived at (see Supplementary Information: Section 4 and Figs. S5).

Nevertheless, the ecological importance of sponge DSI demands does not derive only from the magnitude of its uptake rate, but also from the fact that it largely concentrates on continental margins, where substantial amounts of coastal DSI are progressively accumulated in long-lived (often centennial) sponges under the form of BSI skeletal pieces that are extremely reluctant to dissolution following sponge death. Therefore, even when sponge DSI uptake rates are clearly lower on average that those of diatoms, populations of siliceous sponges may still operate as relevant biological traps, slowing down Si cycling and favoring Si sinking on continental margins. To improve the current knowledge of Si fluxes on continental margins is one of the most urgent needs in order to refine modeling of both the Si cycle and its connections to the Carbon cycle. Unfortunately, most on-going research efforts towards these aims are guided by the extended notion attributing the preponderance of DSI removal on continental margins to formation and burial of BSI primarily by diatoms and radiolarians, disregarding the contribution by the large sponge populations characterizing most continental margins (see also Figs. S5). For instance, after the recent realization that early estimates of BSI accumulation in the Southern Ocean and Antarctic deep sea were about 35% overestimated, the widely accepted model of steady-state balance for the marine Si cycle became unbalanced, because a Si sink equivalent to approximately one quarter of the global BSI burial is now missing. Because in order to bring back the cycle into its assumed balance an additional BSI sink should be identified, it has been proposed that BSI accumulation by diatoms on continental margins should account for most of the "missing" BSI burial. Admittedly, reliable direct estimates of diatom Si retention on continental margins are still lacking. Our current data on sponge DSI demands, along with those already available on BSI standing stocks in sponge populations, suggest that much of the "missing" BSI could correspond to that in the sponge populations of continental margins. Therefore, if we are to refine realistically our current understanding of continental margins as transitory and permanent Si sinks, the role played by sponges has to be incorporated into regional budget calculations. Additionally, the fact that all DSI used by sponges on continental shelves is at the expense of the stock available for diatoms deserves careful consideration, as the DSI slowly— but progressively— accumulated into sponge BSI on continental shelves (and slopes) is taken away from the primary-production circuit for a long time, given the longevity of most sponges and the low levels of dissolution characterizing sponge BSI. By disregarding this sponge DSI-sequestering process while considering exclusively diatom-related DSI and BSI stocks, we are currently overestimating the real levels of connection between Si and C cycles on continental shelves.

**Methods**

**Uptake laboratory experiments.** Silicon uptake was investigated in the laboratory using erect, branching demosponges belonging to the genus *Axinella* (Family Axinellidae; Order Halichondrida; see Fig. S1). Sponges for the uptake experiment were collected from the rocky sublittoral bottoms of the Southern Mediterranean Sea, between the sites 41° 42′ 21″ N -2° 48′ 17″ E and 41° 42′ 25″ N -2° 54′ 51″ E, at depths ranging from 12 to 25 m.

There are three major reasons why species of *Axinella* were selected as a laboratory model: 1) Unlike encrusting species, they can be detached from natural bottoms with...
Field data on DSI availability and sponge abundance. To estimate the annual average of DSI available to the Mediterranean sublittoral sponges in the study area, we sampled seawater from 1 cm above the rocky substrata where sponges grew using syringes during scuba dives; we also sampled the open water column above the continental shelf (about 0.5 km off the coastline) using Niskin bottles (see Supplementary Information: Section 1). These nutrient analyses (n = 240 water samples) extended over a year cycle. To estimate field DSI demands by sponges, we measured average volume uptake rates for field sponges measured by our team on sponge abundance (n = 409, 1×1 m quadrats) and DSI availability (n = 48 water samples) obtained from 21.7 km of Belizean continental shelf (Caribbean Sea), including reefs, mangroves, seagrass beds, and sandy non-vegetated bottoms, to comparatively infer the magnitude of the annual DSI demand by the sponge populations in that ecosystem. It is worth noting that to prevent overestimation, we applied a one-fourth reduction to volume values calculated for each individual in both the Mediterranean and the Caribbean field studies.

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M.M. designed research, analyzed data and wrote the paper. M.M., L.A. A.Gr., A.Go, and I.V. performed research. I.V. performed research.

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