11. Diatoms in temporary rivers: importance in a global climate change context

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Abstract

In this chapter we describe the biological aspects of diatoms in temporary rivers, focusing on their ecology and adaptation mechanisms to the harsh environment in these aquatic ecosystems. Predicted alterations in temporary rivers, according to the climate change scenarios, will most probably affect diatom composition and community structure. Given the importance of diatoms in these ecosystems, changes in diatom taxonomical and functional diversity, physiology, or biomass will be reflected in the whole ecosystem. The adaptation mechanisms to the stressors in temporary rivers can contribute to providing diatoms with an adaptive advantage under the predicted climate change extreme events. Furthermore, the use of diatoms as bioindicators, including the dry phase, contributes to the conservation and management of temporary rivers. However, further studies on diatoms in temporary rivers should be carried out, since their ecology is still poorly known due to the lack of exhaustive and thorough studies on their taxonomy, highlighted by the number of new species recently described from a few sampling sites in Southern Portugal.

11.1 Introduction

Watercourses that are subject to flow cessation, include a wide range of hydrological regimes, namely, temporary or intermittent rivers/streams, that may dry up during some time along the year [11.1], and ephemeral streams, that only flow for a short period, usually after rainfall events [11.1] or after snow melt [11.2]. To describe these watercourses a diverse lexicon is applied, as temporary, intermittent, seasonally intermittent, non-perennial, episodic or ephemeral [11.3], or freezing intermittent rivers and ephemeral streams (FIRES - lotic systems that have the whole water column periodically freezing) [11.4]. In this review, the term "temporary rivers" was selected.

The integration of temporary rivers in riverine ecosystems conceptual models has been critically revised by [11.5], stating that the 18 models jointly describe the main drivers of biogeochemical and ecological patterns and processes along the following four dimensions: (i) *longitudinally* (upstream-downstream); (ii) *laterally* (channel-riparian-floodplain); (iii) *vertically* (surface water-groundwater); and (iv) *temporally* across local and landscape scales. Nevertheless, drying represents a significant hydrological process that structures river ecosystems, contributing to the specificities of temporary watercourses that are not represented in these models, namely:

(i) their longitudinal discontinuity, with the formation of isolated pools or disconnected flowing reaches during the dry phase, with consequences for materials transport and for the connectivity of the populations of riverine organisms [11.6];
(ii) the unidirectional lateral connectivity during part of the year (only terrestrial to aquatic ecosystems);
(iii) the variability in the vertical connectivity between surface and subsurface water throughout the year, since it can be unidirectional during the drying (downwelling of surface water into the hyporheic zone, the saturated subsurface zone beneath the river channel) and rewetting phases (upwelling of subsurface water into the river channel). This variability in the vertical connectivity affects the nutrient and organic matter dynamics [11.7], the vertical exchanges of gases, with litter rewetting contributing to CO_2 emissions [11.8], and the hyporheic zone can act as a refuge for the microorganisms [11.9];
(iv) changes in flow, caused by the drying process should be considered across temporal and spatial scales [11.5]. Thus, these systems are typically characterized by alternation between wet and dry phases, and are archetypal aquatic–terrestrial ecosystems, including flowing, ponded, and dry habitats [11.10], as presented in Figures 11.1 and 11.2.

Temporary rivers are considered the most widespread type of flowing waters, dominating arid, semi-arid, Mediterranean, and dry-subhumid regions, together covering almost half of the land surface [11.2]. Also [11.11] estimate that respectively 69, 56, 49, 42 and 34 % of stream orders 1-5 are ephemeral in COSCATs (coastal segmentation and related catchment) below 60◦ N, therefore, these ecosystems are important even in temperate and humid areas, since headwaters represent more than 70% of the channel length of most river networks [11.12]. Recent estimations indicate that 51-60% of the global river length is non-perennial [11.13], and in Southern Portugal represents >70% of the river network (Agência Portuguesa do Ambiente, I.P.; A.R.H. Alentejo, unpublished data).

[Figure 11.1 here]

Figure 11.1. Example of dry and ponded habitats present in temporary rivers. Lampreia stream, Southern Portugal (left photo); Cuncos stream (right photo), both in Guadiana Basin, Southern Portugal.

[Figure 11.2 here]

Figure 11.2. Temporal dynamics of a temporary river with the presence of wet and dry phases. Valverde stream (Sado Basin), Southern Portugal.

Further, temporary rivers are increasing annually, with more of the land surface drying, due to the reduction and seasonal shifts in precipitation and runoff [11.2]. Temporary rivers are among the aquatic ecosystems more susceptible to being affected by climate change [11.14]. This intermittence increase (both in duration, frequency, and extension, in terms of stretches/areas subject to drying) triggered by climatic drying is aggravated by human activities, such as water abstraction and diversion, with the Anthropocenic intermittence [11.2] creating artificially intermittent reaches.

The importance of naturally temporary rivers for biodiversity support, ecosystem processes and for goods and services provision is already acknowledged [11.2], [11.15], [11.16]. Despite this recent recognition of their value and global increasing spatial and temporal extent, temporary rivers are still largely omitted from legislation and policies, even though the Mediterranean GIG river types recognized by the WFD [11.17], already included "small, lowland, temporary" rivers. Consequently, these rivers are still typically poorly represented in biomonitoring programs, jeopardizing their assessment, and impairing the adoption of adequate management measures to prevent their health degradation [11.18], [11.19]. Temporary rivers are highly vulnerable to multiple anthropogenic pressures, namely agriculture intensification, livestock farming, hydromorphological alteration, and urban wastewater discharge, and these pressures will likely increase under global change [11.20]–[11.22].

Research on these ecosystems has increased over the past decades, as depicted in a search on Scopus in January 2023, where a total of 3 072 manuscripts were published with the terms “temporary river*” OR “intermittent stream*” OR “temporary stream*” OR “intermittent river*” OR “ephemeral stream” OR “non-perennial river*” OR “non-perennial stream*”. The first record of a published paper on this subject was entitled “Intermittent Streams in Berkshire”, written by Rupert Jones in 1885 and published in the Geological Magazine. During the following decades, almost no interest was devoted to this subject, until the 60’s, when one or two papers started to be published each year, increasing in the 70’s and onward, until surpassing the threshold of more than 100 publications per year in 2011. Since this date, more than 100 papers were published every year, with a maximum of 193 publications in 2020, as seen in Figure 11.3.

From these, when the search term “alga*” is included, 95 publications are retrieved, from which only 56 are related to diatoms. The first published record on diatoms in temporary rivers is from 1985, and until 2007 only one paper was published per year. In 2008, 2 papers were published, with this number increasing to 4 in 2014 and reaching a maximum of 7 in 2020 and 2021, revealing a slight increase in interest in this subject, as seen in Figure 11.3. These studies include taxonomical approaches, with the description of 3 new taxa from temporary rivers (*Punctastriata obstinata* E.Morales, M.L.García & Novais; *Planothidium audax* E.Morales, Novais & M.L.García; *Fragilaria odeloucaensis* E.Morales, Novais, C.E.Wetzel, Ector & Morais); community dynamics; monitoring and ecological status assessment, detailed in the following sections. The great majority of the studies are focused in drying rivers, whilst diatom studies in intermittent freezing streams are rare, e.g. [11.23], [11.24].

When comparing these results with those for invertebrates, the latter were present in 369 publications (from 1978 until the end of 2022), with this discrepancy revealing the long way to go in diatom research in these ecosystems. Also [11.3] reviewed the literature on Intermittent Rivers (IR), highlighting the increasing characterization of these systems, considering theoretical aspects, assessment, protection, and habitat restoration. Nevertheless, in their review, only invertebrates and fish were considered among the biota of IR, along with biogeochemistry and hydrological and ecological assessment.

[Figure 11.3 here]

Figure 11.3. Total number of publications on non-perennial watercourses (light grey) and including diatoms (dark grey)

The few publications on algae, and particularly diatoms, do not reflect their essential role in the functioning of temporary rivers, as the basis of the trophic network along with the other primary producers (e.g. cyanobacteria, mosses, and macrophytes in the river channel and the riparian vegetation in the margins). These organisms produce organic matter using upstream and lateral inorganic nutrients and carbon dioxide in the photosynthesis process with solar energy [11.25]. In riverine systems, primary producers differ, with the riparian vegetation dominating the terrestrial-aquatic ecotone, whilst the aquatic environment is colonized by macrophytes and algae [11.25].

The structure of algal assemblages is commonly determined by the type of substratum, the water chemistry, light availability, water temperature, and flow [11.26]. In temporary rivers, these environmental factors are coupled with the challenges associated with the hydrological fluctuations, including the long dry periods and the hydric and thermal stress [11.25].

In this chapter we review 1) the biological aspects of diatoms in temporary rivers, focusing on their adaptation mechanisms to the fluctuating hydrology and associated stressors, such as flow reduction/lentification, increased temperature, desiccation, and UV radiation exposure; 2) diatom community dynamics and the importance of temporary rivers for endangered species conservation; and 3) diatom as bioindicators in temporary rivers.

11.2 Diatoms adaptation mechanisms in temporary rivers

Temporary rivers represent a harsh and rigorous environment for diatoms, due to the hydric stress, related to drying and thermal stress, water flow fluctuations, atmospheric exposure, and desiccation. Globally, water stress directly affects cell structure, biomass, and algal diversity, since water is essential for phototrophs because it is involved in the cells' osmotic equilibrium, in photosynthesis as a reducing agent and in sugar synthesis, and in other metabolic processes [11.25]. Further, vegetative cells of benthic freshwater diatoms are known to be highly sensitive to desiccation, freezing, and abrupt heating, whilst vegetative cells of terrestrial diatoms are more tolerant to temperature extremes [11.27].

Diatoms have developed adaptation mechanisms (either life cycle, morphological or physiological traits) related to the capacity to withstand these harsh conditions and persist in the stream (by staying in, or colonizing potential refuges as pools, leaf litter packs, or subsuperficial sediments) and recover and spread once the flow resumes [11.9]. However, short evidence of diatom adaptation mechanisms is yet available from temporary rivers, as seen in Table 11.1. Thus, in this chapter information about the possible adaptation mechanisms to the stressors diatoms face in temporary rivers is compiled but includes data available from other ecosystems.

[Table 11.1 here]

Morphological and physiological changes in freshwater diatoms exposed to adverse conditions (e.g., temperature, light, desiccation, nutrients, pH, or salinity) have been reported [11.28], namely, the formation of resting stages, defined by [11.29] as cells or group of cells that stop growing and have their metabolic processes at a minimum. Two types have been described [11.27]:

(i) resting spores (hypnospores), that differ in morphology and physiology from vegetative cells, with thicker frustules, often a rounder shape, and less elaborated patterns at the cell surface [11.28];

(ii) resting cells, with cytoplasmatic and physiological alterations but with similar morphology to the vegetative cells, a fact that can contribute to their overlooking.

Spores occurrence has been more common in marine centric diatoms, with few freshwater species as exceptions, mainly belonging to the genera *Urosolenia* and *Acanthoceras* [11.30], *Aulacoseira* and inland *Chaetoceros* [11.28], and being rare amongst pennate diatoms, with few exceptions, such as *Craticula cuspidata* (Kützing) Mann, whose resting-spore formation is detailed in [11.31]. Resting cells are more often formed in freshwater and pennate diatoms [11.28] and their existence explains the persistence of viable cells where spores are not found (for instance, a sample collected in Southern Portugal from cobbles that were dry for a long time, at the end of the summer, revealed a high proportion of live diatoms after rewetting in the laboratory). In diatoms, resting stages are formed vegetatively e.g. [11.32], with a few exceptions of sexual spore formation inside the auxospores in a few marine diatoms, e.g. *Leptocylindrus danicus* Cleve, *L. minimus* Gran [11.28].

Resting stages are reported to withstand unfavourable conditions for years, decades, and even millennia and still be alive, e.g. (i) *Aulacoseira granulata* (Ehrenberg) Simonsen resting cells present in anoxic sediments for twenty years were still able to germinate [11.33]; (ii) ~6600 cal. year BP old *Chaetoceros muelleri* var. *subsalsum* (Lemmermann) Johansen & Rushforth resting spores in hypoxic sediments were still viable (even though the germination rate decreased with increasing age, from three hours to 2 – 3 days) [11.34]. According to [11.28], cell survival in the resting stage depends on several important factors, as light, temperature, and time.

[11.28] listed 16 non-marine taxa as producing resting spores and 17 producing resting cells:

(i) resting spores producing taxa - *Acanthoceras zachariasii* (Brun) Simonsen, *Aulacoseira bellicosa* (Héribaud) Simonsen, *A. italica* (Ehr.) Simonsen, *A. skvortzowii* Edlund, Stoermer et Taylor, *Craticula cuspidata* (Kützing) Mann, *Diatoma anceps* (Ehrenberg) Kirchner, *Eunotia faba* (Ehrenberg) Grunow, *E. pectinalis* (Kützing) Rabenhorst, *E. soleirolii* (Kützing) Rabenhorst, *Melosira charcotii* Peragallo, *M. laevis* (Ehrenberg) Ralfs, *M. semilaevis* Grunow, *M. turgida* Ehrlich, *Meridion circulare* (Greville) C.Agardh, *Urosolenia eriensis* (H.L. Smith) Round et Crawford, and *U. longiseta* (Zacharias) Edlund & Stoermer;

(ii) resting cells producing taxa - *Actinocyclus normanii* (Gregory ex Greville) Hustedt, *Asterionella formosa* Hassall, *Aulacoseira granulata* (Ehrenberg) Simonsen, *A. islandica* (O.Müller) Simonsen, *A. subarctica* (O. Müller) 11.Haworth, *Diatoma tenuis* C.Agardh, *Fragilaria capucina* Desmazières, *Staurosira construens* Ehrenberg, *Fragilaria crotonensis* Kitton, *F. intermedia* Grunow, *F. pinnata* Ehrenberg, *Stephanodiscus alpinus* Hustedt, *S. binderanus* (Kützing) Krieger, *S. medius* Håkansson, *S. niagarae* Ehrenberg, *Tabellaria fenestrata* (Lyngbye) Kützing, and *T. flocculosa* (Roth) Kützing.

Physiological distinctions between resting stages and vegetative cells have been observed and are reported in [11.28], namely: (i) the higher rates of cellular carbon to nitrogen; (ii) higher carbon and chlorophyll content per cell when in the dark; (iii) more condensed organelles; (iv) more or larger vesicles of storage products; (v) lower sugar phosphate content; (vi) decrease in the respiration rate (to less than 20% when compared with actively growing cells); (vii) reduction in the photosynthesis rate (less than 4% to that in vegetative cells), related to the chloroplasts size reduction, loss, or colour change (from green to yellow and orange-red), observed in diatom cells during the non-flow phase by [11.26]. An accumulation of oil in the cells of *Pinnularia viridis* was also observed by [11.29] in experimental drying. According to [11.29], *Pinnularia viridis* (Nitzsch) Ehrenberg, *P. major* (Kützing) Rabenhorst, *Cymbella lanceolata* C. Agardh, and *Nitzschia linearis* W. Smith were species mainly found in the water, but able to survive some exposure and drying, while *Nitzschia palea* (Kützing) W.Smith and *Stauroneis phoenicenteron* (Nitzsch) Ehrenberg were able to survive prolonged and severe drought.

Table 11.1 summarizes the adaptation mechanisms to the stressors in temporary rivers.

11.3 Flow reduction / lentification

In temporary rivers, one of the environmental factors most challenging for biological communities is flow reduction, which can be the flow interruption or even the complete drying of the channel, according to the Aquatic Regime (AR) of the temporary rivers, e.g. temporary-pools, temporary-drying or episodic, following the updated classification proposed by [11.35], that describes the long-term global variability of the hydrological conditions.

In temporary rivers, the duration and predictability of flow and no-flow periods are major factors shaping biological communities [11.35].

Considering the biological communities' dependence on the changes of the aquatic habitats over time, [11.35] developed a methodology to analyse the aquatic regimes based on Aquatic States (AS), that relate to the sets of mesohabitats occurring in a reach at a determined moment, depending on the hydrological conditions (presented in Figure 11.4):

i) Hyperrheic – high water (flood), that originates the movement of stream bed alluvium and drift of most of the aquatic fauna;

ii) Eurheic – optimal hydraulic connectivity and widest range of discharges due to the succession of riffles and pools, presence and connectivity of all mesohabitats;

iii) Oligorheic – low water discharge, but enough to connect most pools due to water rivulets;

iv) Arheic – surface discharge close to 0, but some disconnected pools are still present in the stream bed;

v) Hyporheic – absence of surface water in most of the stream bed, but the alluvium may be still close to saturation;

vi) Edaphic – no surface water in the entire stream bed, and the dryness of the alluvium does not allow active hyporheic life.

The importance of these aquatic states for the ecological status assessment is also discussed by [11.35], based on macroinvertebrate communities.

[Figure 11.4 here]

Figure 11.4. Aquatic States (AS), depending on the hydrological conditions (based on [11.35]*)*

Further, the hypothesis that also diatom communities respond to the AS was validated by [11.36], based on a one-year study of permanent (as control) and temporary rivers in 2017 in Southern Portugal. 2017 was a very dry year, characterized by severe droughts, with low accumulated precipitation even in winter and resuming only in March 2018. Therefore, the arheic state was distinguished between summer and winter, with temperature being the main environmental factor separating the two types. Diatom communities not only differed between AS (arheic and eurheic), but also reflected the mesohabitats (flowing water and pools). Indicator species of the AS (eurheic in spring 2017, eurheic during the remaining seasons, arheic in summer, arheic in the remaining seasons) were also retrieved and are as follows:

- arheic (except Summer 2017) - *Craticula cuspidata* (Kützing) D.G.Mann, *Fallacia pygmaea* (Kützing) Stickle & D.G. Mann, *Navicula erifuga* Lange-Bertalot, *Nitzschia intermedia* Hantzsch ex Cleve & Grunow, *Nitzschia supralitorea* Lange-Bertalot, *Planothidium pericavum* (J.R.Carter) Lange-Bertalot, *Sellaphora pupula* (Kützing) Mereschkowksy, and *Surirella angusta* Kützing;

- arheic in Summer 2017 - *Achnanthidium eutrophilum* (Lange-Bertalot) Lange-Bertalot, *Cocconeis placentula* Ehrenberg, *Eolimna minima* (Grunow) Lange-Bertalot, *Epithemia turgida* (Ehrenberg) Kützing, *Karayevia clevei* (Grunow) Bukhtiyarova, *Nitzschia amphibia* Grunow, *N. filiformis* var. *conferta* (Richter) Lange-Bertalot, *N. valdestriata* Aleem & Hustedt, *Pseudofallacia tenera* (Hustedt) Liu, Kociolek & Wang, *Pseudostaurosira brevistriata* (Grunow) D.M.Williams & Round, *Pseudostaurosiropsis* sp. 1, and *Staurosirella pinnata* (Ehrenberg) D.M.Williams & Round;

- eurheic - *Denticula subtilis* Grunow, *Encyonema minutiforme* Krammer, *Eunotia implicata* Nörpel, Lange-Bertalot & Alles, *E. minor* (Kützing) Grunow, *E. pectinalis* (Kützing) Rabenhorst, *Fragilaria* aff. *capitellata* (Grunow) J.B.Petersen, and *Nitzschia gracilis* Hantzsch;

- eurheic in Spring 2017 - two Fragilaria species with affinity to the pectinalis group, *Gomphonema pumilum* var. *rigidum* E.Reichardt & Lange-Bertalot, *G. truncatum* Ehrenberg, *Melosira varians* C.Agardh, *Planothidium* cf. *frequentissimum* (Lange-Bertalot) Lange-Bertalot, and *Ulnaria acus* (Kützing) Aboal.

These assemblage shifts were also reported by [11.25], who stated that algal assemblages during the flowing phase were similar to permanent streams. With intermittency, a shift in the communities is observed, with lotic (benthic) algae gradually being replaced by lentic communities, with a dominance of the motile form in the pools, related to the nutrient increase and siltation [11.36]. When complete drying does not occur, these pools play an important role in the recolonization process once the flow resumes [11.37].

11.4 Desiccation stress

Water supports vital biophysical and biochemical processes and influences key cellular functions, such as: protein folding and stability, enzyme catalysis, DNA packaging, molecular recognition, cellular tolerance to freezing, intracellular signalling and transport processes; also biomolecules such as nucleic acids, proteins or membranes are normally in close contact with water solutions [11.38]. Further, alterations in cell organelles morphology and location have been observed, as water leaves the cell during the desiccation process, e.g. the shrinking of the protoplasmic components and distortion of chloroplasts and pyrenoids of *Stauroneis anceps* cells, with dry cells organelles positioned at the ends of the frustule, with the entire protoplast at one end or divided and occupying both ends of the frustule [11.39].

During the dry phase, biomolecules face air drying/dehydration, which is the process involving the removal of both freezable and non-freezable fractions of water, with extreme dehydration often causing: protein denaturation (a process that may be irreversible); destabilization or disruption of membranes [11.40]; and the accumulation of reactive oxygen species [11.41]. Tolerance to desiccation has been mainly observed in resting cells of terrestrial diatoms by [11.27], with some mechanisms already identified at the cellular level that may contribute to achieving it: (i) the addition of a solute (disaccharides) can contribute to proteins' stabilization, and therefore dehydration to much lower water content. In brief, as dehydration progresses, the solute concentration increases to the point where it penetrates the protein hydration shell (one or two layers of water molecules that are in close interaction with the surface of the proteins [11.38], allowing the preservation of the conformational state of the dry protein [11.40]; (ii) further, sugars (namely the disaccharide trehalose) are effective in the stabilization of phospholipid bilayers under severe dehydration, allowing dry phospholipids to be in liquid crystalline phase at temperatures they would normally be in the gel phase, avoiding the gel to the liquid crystalline phase transition during rehydration [11.40], [11.42]. Further, according to the principles of the excluded solute theory, all compatible solutes (low molecular mass organic solutes) would be expected to provide some protection to cells and intracellular macromolecules, against the reduced intracellular water potential and the resulting increase in intracellular ionic strength caused by the water loss [11.42]. However, under a severe desiccation state, this mechanism is not efficient, since ca. 0.3 – 0.4 g H_2O protein^{-1} are needed to maintain even a single mono-layer of bound water around the proteins [11.42].

Diatom response in temporary rivers to drying includes the prevalence of the life-forms mobile and mucilage-forming [11.25]. The first trait includes species that are able to move ("mobile") and are therefore capable of selecting the most suitable habitat [11.43] and migrating, for instance, to deeper sediments where moisture can remain [11.9], [11.44], or to exploit refugia (as pools). Mucilage-forming species (e.g. *Cymbella* or *Gomphonema* species) form mucilaginous stalks or tubes that can protect the cells. The increase in the production of extracellular substances in stress situations is known to occur when there is an inhibition of cell division but photoassimilation continues [11.45].

These adaptations, either as resting stages or certain life-forms prevalence, allow diatom communities to persist in dry streambeds only when the drying process is recurrent and slow [11.25], resulting from a long evolutionary period during which the multihabitat filter process selects diatoms with a set of traits that allows their adaptation [11.46], as in Mediterranean temporary rivers. This acclimation process has been experimentally tested by [11.27], [11.39]. The first authors acknowledging that *Stauroneis anceps* survival is increased by slowing drying; and the latter proving that gradual heating before desiccation had positive results only on the survival of resting cells of terrestrial diatoms, that have protection mechanisms to survive in soils and temporary water bodies, being regularly exposed to harsh changes in humidity and temperature.

When diatom communities are subjected to rapid alterations in natural flow conditions, i.e. magnitude, frequency, duration, timing, and predictability [11.46], community recovery depends on their capacity to remain in drought refuges (e.g., permanent pools, moss patches, leaf packs, seeps and woody debris, dry biofilm on stones, dry sediments and leaf litter), from where regrow or recolonization may occur [11.37].

Adaptations to desiccation play a major role in shaping diatom communities in view of the increasing magnitude, frequency, occurrence and duration of river drying, driven by climate change and water abstraction [11.5], [11.12], [11.14], [11.47]. An increase in the length of dry rivers with consequent connectivity loss has already been observed in diverse regions due to the streamflow decrease and drought caused by a decrease in precipitation and temperature increase [11.6]. Further, predicted temperature increase will lead to an increase in evapotranspiration pushing systems closer or beyond the balance where water losses to the atmosphere exceed inputs. The probability of river drying is also expected to increase due to the higher probability of dry periods occurrence, i.e., seasonal or multi-year droughts [11.5].

11.5 Thermal stress

In temporary rivers, temperature increase occurs during the lentification process, as observed in Southern Portugal, where a maximum of 33.1 ºC was measured in a pool at the end of the Summer (Ameira stream, 22nd September 2017), with the median for all the summer pools (11 sites) being 22.4 ºC (Interquartile range: 18.35-28.15 ºC), whilst the flowing streams (2 sites) presented lower temperatures (median: 14.60 ºC;

interquartile range: 13.95-15.25 ºC) as depicted in [11.36]. Thus, diatoms are also exposed to thermal stress during this dry phase, an important issue considering that temperature is a relevant environmental factor that regulates algal growth and species distribution, either seasonally, altitudinally or latitudinally [11.48]–[11.50].

Diatom specific growth in response to temperature depends on both the ranges and temperature-specific dependency [11.50], which is more evident at low (< 10 ºC) and high temperatures (> 20 ºC), but can be modified by light flux density and photoperiod [11.48].

Species growth rate is known to increase up to a temperature maximum, followed by an abrupt decrease until reaching the upper limit of the growth range, therefore, the maximum rate occurs near the upper temperature range [11.50]. Hence, according to the optimum conditions (with a higher relative growth rate) tendentially occur at the temperature each species predominates in the field [11.50]. In the absence of data from temporary rivers, [11.50] study with planktonic diatoms in tem11.perate climate is presented as an example. In this case, temperature dependency was higher in high-temperature species and lethal heating was 5 ºC above the higher limit of the growth range of the species, with the highest value of 40 ºC for *Chaetoceros pseudocurvisetus* Mang., *Stephanodiscus hantzschii* Grun. and *Skeletonema costatum* (Grev.) Cleve, allowing most of the species to grow under the annual habitat temperature range. Several freshwater planktonic algae, including diatoms, were not able to withstand temperatures above 30 ºC even though they were actively growing at 25ºC (e.g., *Asterionella formosa* Hassall, *Tabellaria flocculosa* var. *asterionelloides* (Grunow) Knudson, *Aulacoseira subarctica* (O.Müller) E.Y.Haworth and *Discostella pseudostelligera* (Hustedt) Houk & Klee), except for *Fragilaria crotonensis* Kitton that was still actively growing at this temperature, but for which 35ºC were lethal [11.48]. Furthermore, in the experiments carried out by [11.51], abrupt heating up to 40 ºC was lethal for 40% of the 34 studied species, whilst more species were able to withstand gradual heating.

The stress response to high temperatures is considered to be a universal and highly conservative mechanism, that is typically characterized by a decrease in the production of constitutive proteins coupled with an increase in stress-inducible proteins (heat shock proteins, HSP). This response can be observed, for instance in the diatoms *Chaetoceros muelleri* [11.52], *Nitzschia alba* [11.53], and *Skeletonema costatum* [11.54], with a faster response to temperature increase detected in thermo-tolerant species (e.g. *Chaetoceros muelleri*) possessing higher amounts of constitutively expressed HSPs.

Additionally, changes in diatom fatty acid composition have been reported in response to temperature increases above the optimal growth temperature level, triggered by a particular temperature and not by the stress duration [11.55].

The exposure of *Skeletonema costatum* to heat stress (25ºC – 40 min.) has caused a rapid increase in free polyamine content (putrescine and spermidine titers), which can be explained by their release from the bound form (as a "first aid" against the initial shock), followed by a new synthesis of putrescine after 30ºC – 40 min [11.54]. The polycationic nature of polyamines seems to provide the cells with the ability to regulate the intracellular environment, to have a protective effect on membrane integrity by putrescine binding to phospholipids or the stabilization of other cell molecules by inhibition of degradative enzymes [11.54]. Also, conjugated polyamines are hypothesized to be involved in the cellular response to heat stress (e.g. TCA-soluble bound spermine in *Skeletonema costatum*) [11.54].

Thermotolerance of enzymes can also be enhanced by the presence of compatible solutes, since they are preferentially excluded from the immediate hydration shell of proteins, consequently, their presence in high concentrations would tend to inhibit the thermal denaturation of proteins [11.42].

Diatom's dependency on temperature and sensitivity to abrupt temperature changes make them a highly sensitive group to climate change, with the predicted temperature increase, alteration in precipitation patterns, and increase in the frequency and duration of extreme events such as droughts and floods. These extreme climate events can compromise the conditions beyond which many species are adapted [11.21] [11.56]. Climate change impacts on freshwater ecosystems have already been detected at a global scale, including shifts in species range and in timing. Also, at the 1.5°C global warming scenario, species and ecosystems (mainly in polar and already-warm areas, where most of the non-perennial watercourses occur), will face temperatures beyond their historical values [11.56], that can severely alter freshwater diatom distribution, favouring species already adapted to more harsh habitats (e.g. terrestrial diatoms) and even impairing their survival, with consequences for non-perennial watercourses functioning.

11.6 UV Radiation exposure

Solar radiation is the electromagnetic radiation emitted by the sun and comprises a wide range of wavelengths, known as the electromagnetic spectrum, from radiowaves through the infrared, visible and ultraviolet to X-rays and gamma rays. However, 99% of the energy of solar radiation is comprised in the wavelength band from 150 to 4 000 nm (near ultraviolet, visible, and near-infrared regions of the solar spectrum).

According to the UCAR COMET Program, ca. 43% of radiant energy from the sun is in the visible part of the spectrum (between 400 – 700 nm), roughly 49% is infrared (between 700 nm -1 mm), about 7% is from ultra-violet (between 100 – 400 nm), and less than 1% of solar radiation is emitted as x-rays, gamma rays and radio waves (https://sos.noaa.gov/catalog/datasets/climatebits-solar-radiation/).

Considering that shorter wavelength radiation is more energetic, ultraviolet is the range that rises more concern, due to the possible damage it may cause in living organisms. It is separated into three ranges: UV-C (100-280 nm), which makes up only 0.5% of all solar radiation, and very little reaches the surface because it is absorbed by ozone; UV-B (280-320 nm) is an energetic, photoactivating band of radiation, only partially absorbed in the stratosphere, that can penetrate the water reaching 20 m depth in the ocean; UV-A (320-400 nm) is not absorbed by ozone, though it can be blocked by cloud cover and can penetrate deeper in the water than UV-B (Fondriest Environmental, 2014, https://www.fondriest.com/environmental-measurements/parameters/weather/solar-radiation/).

Under climate change scenarios, drying increase in temporary rivers, both in length, extension, frequency, and duration, may affect the exposure of diatoms to radiation, including photosynthetically active radiation (PAR) and ultraviolet (UV). Thus, organisms in these exposed areas should possess efficient adaptation mechanisms [11.51].

UV-B irradiance can be hazardous to living beings, as it affects biomolecules directly or by inducing the formation of reactive oxygen species (ROS, as hydrogen peroxide and superoxide radicals) inside the cell. ROS can further affect the cellular structures, block enzymatic reactions, interfere with photosynthesis (through changes to the main photosynthetic enzyme, ribulose bis-phosphate carboxylase), the nutrient uptake, DNA replication and physiological responses as motility and orientation [11.57]–[11.61]. Thus, at the cellular level, UV radiation toxic effect can be triggered in two photochemical pathways: (i) direct or primary mechanisms - biomolecules as proteins and nucleic acids have chromophores that absorb in the UV region of the radiation spectrum and are photochemically degraded or transformed under high UV flux, impairing or losing their biological function; (ii) indirect mechanisms – intermediate compounds absorb UV (photosentising agents) either inside or outside the cell, and produce reactive oxygen species (ROS). These high energy oxidants as hydrogen peroxide, superoxide or hydroxyl radicals can further react with other cellular components [57]. DNA and RNA are especially sensitive to UV radiation because of their highest absorbance coefficients for short wavelength UV [11.58], making genetic one of the most reported damages, also for diatoms [11.57].

In the absence of specific information about diatoms in temporary rivers, the results of [11.57] using cultured marine diatom species in laboratory experiments are addressed. Marine diatom species (*Cyclotella* sp., *Nitzschia closterium* and *Thalassiosira nordenskioldii*) exposed to an increasing range of daily doses of UV-B (280-320 nm) radiation revealed that, at the lowest dose (< 2000 J m^{-2} d^{-1}, DNA weighted biologically effective dose, normalized at 300 nm: daily $BED_{DNA\ 300\ nm}$), division rates decreased, volume enlargement occurred and high cellular protein and pigment content levels were detected, including the photoprotective carotenoids (diadinoxanthin + diatoxanthin), or both, in the *Cyclotella* species. The highest UV-B treatments (between 2000 and 3800 J m^{-2} d^{-1} daily $BED_{DNA\ 300\ nm}$) resulted in complete growth inhibition, with minor changes in protein, pigments and cell volume [11.57]. Further, high UV-B radiation levels induced plasmolysis and disorientation of cell organelles in *Cyclotella* sp., whilst lower levels seemed to increase chloroplasts in number and volume [11.57]. Thus, these results support the hypothesis that UV-B radiation causes growth without cell division, due to DNA damage and the arrest in the S or G2 phase of the cell cycle [11.57]. Most microalgae developed strategies to minimize the UV-induced damage. According to [11.62], these include:

(i) avoidance mechanisms, by controlling their movements towards light or water surface, as is the case of motile diatoms, that can migrate to less exposed areas [11.61];

(ii) screening - reduction of the effective UV radiation that penetrates the cell, by synthesising compounds that absorb the damaging wavelengths (screening agents), either intracellular, as the mycosporine-like amino acids (MAAs), also reported by [11.63] as being involved in the defence strategy in response to UV-induced damage in the diatom *Porosira glacialis*) or extracellular (at the cell walls where they block incoming UV radiation);

(iii) repair of UV-induced damages to the nuclear material (the most important damages, for which cells have developed repair mechanisms generally involving biosynthetic pathways mediated by stress proteins), proteins and enzymes, pigments and photosynthetic reaction centres, lipids and membranes. In the case of DNA repair, several mechanisms have been identified as photoreactivation (light-induced repair), nucleotide and base excision repair and recombinational repair; and the activation of heat shock proteins for protein damage repair. For the UV damage caused by ROS, detoxifying mechanisms have been developed, by means of antioxidant enzymes, lipid-soluble antioxidants in cellular membranes (e.g. carotenoids as diadinoxanthin and beta-carotene), and water-soluble reductants in the cytosol;

(iv) acclimation – the process by organisms adapt to changes in the environment, either by physiological changes or by alterations in the species composition at the community level. Recent research also verified the hypothesis that the optical properties of centric diatoms frustule can protect the cell from harmful radiation (UVB and UVC), and three main mechanisms involved in cell protection were identified, using *Coscinodiscus wailesii* and *C.* cf. *radiatus* as a model [11.61], [11.64]. These are: absorption by silica and trace organic compounds incorporated in the frustule; diffraction (due to the geometry of the valve and refractive index contrast with respect to the surrounding environment); and photoluminescence (mainly due to chemical surface defects of hydrated amorphous porous silica). This hypothesis arose from the fact that diatoms evolved under low concentrations of atmospheric oxygen and ozone and are characterized by high tolerance to UV irradiation, even with low concentrations of screening agents such as MAAs [11.61], [11.64]. Nonetheless, it could be interesting to verify it in pennate diatoms, since the test with *Navicula perminuta* was not conclusive due to its small dimensions and high curvature [11.64]. Thus, this distribution of UV radiation due to their SiO_2 frustules can be an important evolutionary aspect to be considered for the presence and evolution of frustules in diatoms, contributing to a decrease in the rate of UV radiation-induced damage in the cells [11.64]. Further, in the

> review made by [11.60] it is concluded that research into the role of diatom frustules in UV protection shows great potential, both in relation to diatom biology and potential technological applications, in spite of the few number of diatom species analysed so far.

The development of these strategies, is related with the previous light history (e.g. for the development of UV screening) and cell size, for instance in the production of MAAs [11.62], and a differential response to UV radiation was observed for two diatoms from contrasting niches, planktic (collected offshore) and benthic (collected from intertidal sediments. Benthic species presented higher tolerance to extreme temperature and UV radiation) and a better response was observed under elevated temperature [11.65]. Given these environmental-specific responses, it would be interesting to carry out detailed studies on the adaptation mechanisms to UV exposure from diatoms in temporary rivers.

Thus, as an overview, UV exposure net stress reflects the balance between damage, repair and the energetic costs of protection, and can be presented in terms of an increase in energy demand, changes in cell composition, and a decrease in growth and survival rates, as depicted in the scheme in [11.58]. Given that the physiological changes observed in response to the increase in UV-B radiation do not confer full protection to the organisms, and a number of organisms have already reached their UV-B tolerance limits [11.62], the diatom frustule UV protection detailed in [60] may confer them an additional advantage in temporary rivers, more susceptible to drying.

11.7 Communities dynamics in temporary rivers

Community response to perturbations depends on the environment where it is developed, with low-stress environments supporting complex communities, but fragile, when compared to those subjected to environmental stress, e.g. pollutants exposure, light, current or desiccation, as reported in the diverse studies on diatom communities cited by [11.45].

According to [11.25], flow intermittence is a selective force on algal community composition, usually causing a decrease in alpha diversity. Indeed, a long drying phase can eliminate most of the sensitive species, few species are able to resist drying and there are few pioneer species able to recolonize when flow resumes. Thus, flow intermittency also alters diatom composition throughout the year, with shifts from lotic to species adapted to lentic conditions, even up to aerophilous species during the lentification process and drying up of the riverbed [11.66], [11.67]. Nevertheless, when drying is not complete, beta diversity is higher than expected, since lotic, lentic, and terrestrial habitats cooccur sequentially in a longitudinal direction [11.25].

Further, according to [11.68], diatom communities in temporary rivers have less species and a lower proportion of specialist taxa than perennial streams. Also [11.66] detected taxonomic and functional differences between temporary and permanent reaches in Liguria (NW-Italy), both at local and regional scales, with the temporary reaches being characterized by less heterogeneous communities, with higher percentages of small, mainly stalked and pioneer taxa belonging to the low profile guild, which are able to tolerate physical disturbance. These results are in accordance with [11.69], in their study of intermittent and permanent lowland rivers in Carpathian basin. The authors observed that intermittent rivers were characterized by lower taxonomical diversity, higher Berger-Parker index (a metric to measure dominance, defined by the proportional abundance of the dominant species in the assemblage), and lower functional richness and diversity. Disturbed sections were dominated by small-sized generalists (including pioneers and small low-profile taxa) and aerophilic taxa, the latter considered good indicators of drought, since they are able to survive the drying up of the riverbed and colonise rapidly the habitats once flow resumes. The importance of hydrology-related variables in determining diatom species distribution and community ecological attributes in Cyprus was also acknowledged by [11.71]. Further, even in large rivers, a continuous decrease in precipitation (drying period) significantly decreased benthic diatoms, in the number of taxa, Shannon diversity, and functional richness [11.70], addressing the implications of the longer periods with low precipitation, as predicted in the climate change scenarios.

Temporary rivers' importance for species conservation is another issue that should be highlighted, thus reinforcing the need for the adoption of appropriate conservation and management measures. This is supported by the relevance and frequency of species that fulfil the descriptions of Red List threat categories and that are presented in the diatom Red List for Central Europe (2018) in intermittent streams in Cyprus [11.71]. This is in accordance with the findings of [11.36] and [11.72], who highlighted the fact that flow instability, lentification and habitat fragmentation represented the major threats for endangered species, whilst water stability and river habitat heterogeneity favour their presence. These results highlighted the importance of the isolated pools as refugia for benthic diatoms during the dry phase, especially under the climate change scenarios, with increasing drought causing the persistence of isolated pools beyond the summer season (e.g. in 2007 Southern Portugal was subjected to a severe drought, with isolated pools permanence during the winter until flow resuming only in the next spring).

11.8 Diatoms as bioindicators in temporary rivers

Diatoms have been largely used as bioindicators in the framework of the monitoring in accordance with the WFD [11.17], being commonly used as proxies for the Biological Quality Element - Phytobenthos [11.73]. However, for the ecological status assessment diatom sampling should be carried out in stable flow conditions [11.74], avoiding zones of very slow current [11.75]. The fact that the dry phase is excluded gives an incomplete idea of the habitat quality in temporary rivers [11.76], thus, complementary methods for the ecological status assessment should be designed and tested. Further, an adaptation or development of novel metrics to assess the ecological status in temporary rivers is envisaged, since metrics developed for permanent streams may underestimate their ecological status since sensitivity to intermittency and to environmental degradation often covary [11.76]. Diatoms have the potential to fill this lacune, as presented in the preliminary results depicted in [11.76] and further tested by [11.36], with the Specific pollution sensitive index (SPI) negatively correlated with the percentage of dry riverbed and presenting a good response over time, independently of the aquatic state The Ecological Quality Ratio (EQR) for dry biofilm samples collected in Summer was comparable with the values from the previous spring, thus dry biofilm can be considered a viable alternative for the ecological status assessment during the dry phase, instead of the isolated pools, whose communities are not considered by [11.35] to be representative of the ecological status of the stream. Also, [11.77] observed that the Biological Diatom Index (BDI) was slightly affected by drying, but these effects had no significant influence on the ecological quality assessment. [11.78] also validated this hypothesis, in a study conducted in small hilly streams in the Pannonian ecoregion (Hungary), with drying only affecting negatively the Rott’s trophic index (TID), whilst the Specific pollution sensitivity index (IPS), Rott’s saprobic index (SID) and the Hungarian phytobenthos metric (IPSITI) were not impacted.

11.9 Final remarks

Diatoms have a recognized importance in temporary rivers as the main algal component of biofilms, with any changes in their taxonomical and functional diversity, physiology or biomass being repercussed in the whole ecosystem. Furthermore, adaptation mechanisms of diatoms have already been detected, and contribute to enhance their response to main environmental stressors (e.g. flow reduction, temperature increase, increased exposure to UV radiation and desiccation), and reveal that diatoms can have an adaptative advantage under the predicted climate change extreme conditions. However, some of these strategies were tested with diatoms adapted to other aquatic ecosystems. Therefore, detailed studies should be carried out with diatoms adapted to naturally intermittent rivers to validate their effectiveness to the particular conditions characterizing these ecosystems. Further, diatoms can also play an important role in the management of temporary rivers, since they can be used as bioindicators even during the dry phase. Appropriate management of naturally temporary rivers is a must since, apart from their value and global importance, they also host fragile diatoms (still poorly explored) and include threatened species. Further studies on diatoms in temporary rivers should be addressed, since the ecology of most species inhabiting these ecosystems is still poorly studied.

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