



SYMPOSIUM INTRODUCTION

The Temporal and Environmental Context of Early Animal Evolution: Considering All the Ingredients of an “Explosion”

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Synopsis Animals originated and evolved during a unique time in Earth history—the Neoproterozoic Era. This paper aims to discuss (1) when landmark events in early animal evolution occurred, and (2) the environmental context of these evolutionary milestones, and how such factors may have affected ecosystems and body plans. With respect to timing, molecular clock studies—utilizing a diversity of methodologies—agree that animal multicellularity had arisen by ~800 million years ago (Ma) (Tonian period), the bilaterian body plan by ~650 Ma (Cryogenian), and divergences between sister phyla occurred ~560–540 Ma (late Ediacaran). Most purported Tonian and Cryogenian animal body fossils are unlikely to be correctly identified, but independent support for the presence of pre-Ediacaran animals is recorded by organic geochemical biomarkers produced by demosponges. This view of animal origins contrasts with data from the fossil record, and the taphonomic question of why animals were not preserved (if present) remains unresolved. Neoproterozoic environments demanding small, thin, body plans, and lower abundance/rarity in populations may have played a role. Considering environmental conditions, geochemical data suggest that animals evolved in a relatively low-oxygen ocean. Here, we present new analyses of sedimentary total organic carbon contents in shales suggesting that the Neoproterozoic ocean may also have had lower primary productivity—or at least lower quantities of organic carbon reaching the seafloor—compared with the Phanerozoic. Indeed, recent modeling efforts suggest that low primary productivity is an expected corollary of a low-O₂ world. Combined with an inability to inhabit productive regions in a low-O₂ ocean, earliest animal communities would likely have been more food limited than generally appreciated, impacting both ecosystem structure and organismal behavior. In light of this, we propose the “fire triangle” metaphor for environmental influences on early animal evolution. Moving toward consideration of all environmental aspects of the Cambrian radiation (fuel, heat, and oxidant) will ultimately lead to a more holistic view of the event.

Introduction

Questions regarding early animal evolution are a hot research topic—from determining the sister group to all other animals (compare Whelan et al. [2015] and Simion et al. [2017]), to understanding the genetic and developmental changes driving body plan evolution (Fernandez-Valverde et al. 2018; Paps 2018; Paps and Holland 2018), to determining how best to analyze the exponential increase in new genomic sequence data being generated (Laumer 2018). However, these debates can often take place in a sterile setting, between nodes on a cladogram. We gain a much deeper understanding of metazoan

evolution by rooting our discussions in time and place. Here, we aim to review and synthesize recent advances in understanding the temporal and environmental context of early animal evolution.

Some of the most famous early animal fossils are exceptionally preserved specimens from the Cambrian Burgess Shale and Chengjiang lagerstätte. Their canonization as “weird wonders” (Gould 1990) stimulated decades of debate on the nature of early animal evolution, and they continue to yield remarkable insights into the nature of Cambrian animals and ecosystems (see for instance Sheppard et al. [2018]). However, phylogenetic research on these

Cambrian faunas has demonstrated that almost all of the “weird wonders” can convincingly be placed into the stem-groups of extant phyla, if not within the crown group (reviewed by Erwin and Valentine 2013; Briggs 2015). In other words, Cambrian faunas document evolution along the stem-lineages of individual phyla and classes. Thus these fossils represent the end of the first chapter in animal evolution. To start at the beginning of this chapter and the early “landmark” events (the origin of multicellularity, the nervous system, gut, mesoderm, and bilaterality) we must turn earlier in the geological record. But how much earlier? Darwin (1859) famously suggested animals had an extremely long pre-history, and this early history of animals was lost through geological processes of erosion and metamorphism. This explained why animals appeared so suddenly on the scene at the base of the Cambrian: it was an artifact of an imperfect geological record. In the last three decades there has been a tremendous research effort focused on the time period preceding the Cambrian (the Neoproterozoic Era—comprised of the Tonian, Cryogenian, and Ediacaran periods; Fig. 1). Contrary to Darwin’s hypothesis, this interval is indeed present in the rock record and does record the roots of animal evolution. Geologists have established a robust chronology and stratigraphic framework for the interval (Narbonne et al. 2012; Shields-Zhou et al. 2016), and targeted geochronology has firmly established ages and durations for the Neoproterozoic “Snowball Earth” glaciations (Fig. 1) (Macdonald et al. 2010; Rooney et al. 2015; Prave et al. 2016; Pu et al. 2016). Meanwhile, paleontologists have discovered new fossil localities, re-investigated known deposits with advanced imaging techniques, and harnessed information from new approaches such as molecular clocks and the organic geochemical (biomarker) record.

The first goal of this paper is to review these new advances regarding the timing of early animal evolution. Aside from putting ages on the evolutionary trees considered by neontologists, the question of timing is central to evaluating causal hypotheses for the Cambrian radiation. It is well agreed that animals are monophyletic—an evolutionary singularity—and thus the origins of animals and multicellularity likely have more to do with genetic challenges than an environmental “trigger” (Butterfield 2018). If crown-group animals (as a marker for complex multicellularity) evolved in the mid-to-late Ediacaran, just before the Cambrian, as has been proposed by some researchers (Antcliffe et al. 2014; Budd and Jensen 2017), the Cambrian radiation could simply be read as an adaptive

radiation. Alternatively, if animals originated far earlier in the Proterozoic, with traits like large body size, skeletonization, and the range of new morphological features seen in Cambrian fossils appearing convergently and simultaneously, it is more likely an external ecological or environmental trigger was involved.

The nature of this trigger has been widely debated in the literature. Comprehensive review of all hypotheses is not possible here, but several recent papers have covered this topic (Erwin et al. 2011; Zhang et al. 2014; Schiffbauer et al. 2016). Proposed ecological triggers for the Cambrian radiation, or certain aspects of it, range from ecosystem engineering (Butterfield 2011, 2018; Erwin and Tweedt 2011), direct organism–organism interactions including predation and expansion of food webs (Peterson et al. 2005; Marshall 2006; Butterfield 2007; Schiffbauer et al. 2016), and changes in resource patchiness (Budd and Jensen 2017). Proposed environmental triggers include true polar wander (Mitchell et al. 2015), changes in the major ion composition of seawater (Peters and Gaines 2012), and most prominently in the geological literature, a substantial increase in oxygen levels in the ocean/atmosphere system (reviewed by Mills and Canfield 2014; Xiao 2014). The exact mechanistic relationship between increasing oxygen levels and the Cambrian radiation in the literature is varied. Many geochemical studies simply state that animals require oxygen, and assert that more oxygen must be better. Other studies have linked oxygen changes to ecological (Sperling et al. 2013a), developmental (Hammarlund et al. 2018), or evolutionary (Wood and Erwin 2018) mechanisms that were themselves proposed as the proximate cause of the Cambrian radiation. Indeed, as recognized in a recent paper by Schiffbauer et al. (2016), the search for a sole trigger may be simplistic given the myriad interconnected links between the biosphere and geosphere.

In this paper, we build on published geochemical modeling and present a new investigation of shale organic carbon to suggest that both oxygen and food supply to the benthos likely changed through the Neoproterozoic–Paleozoic transition. Looking across natural environmental gradients in the modern ocean, we then demonstrate that the ecological effects of increasing food or increasing oxygen are directionally similar. While it is evident that the Cambrian radiation was a multi-faceted event, we emphasize that directly assigning causal significance to individual forcing factors remains a challenging task, and demands consideration of a cascade of coupled genomic, environmental, and ecological changes.

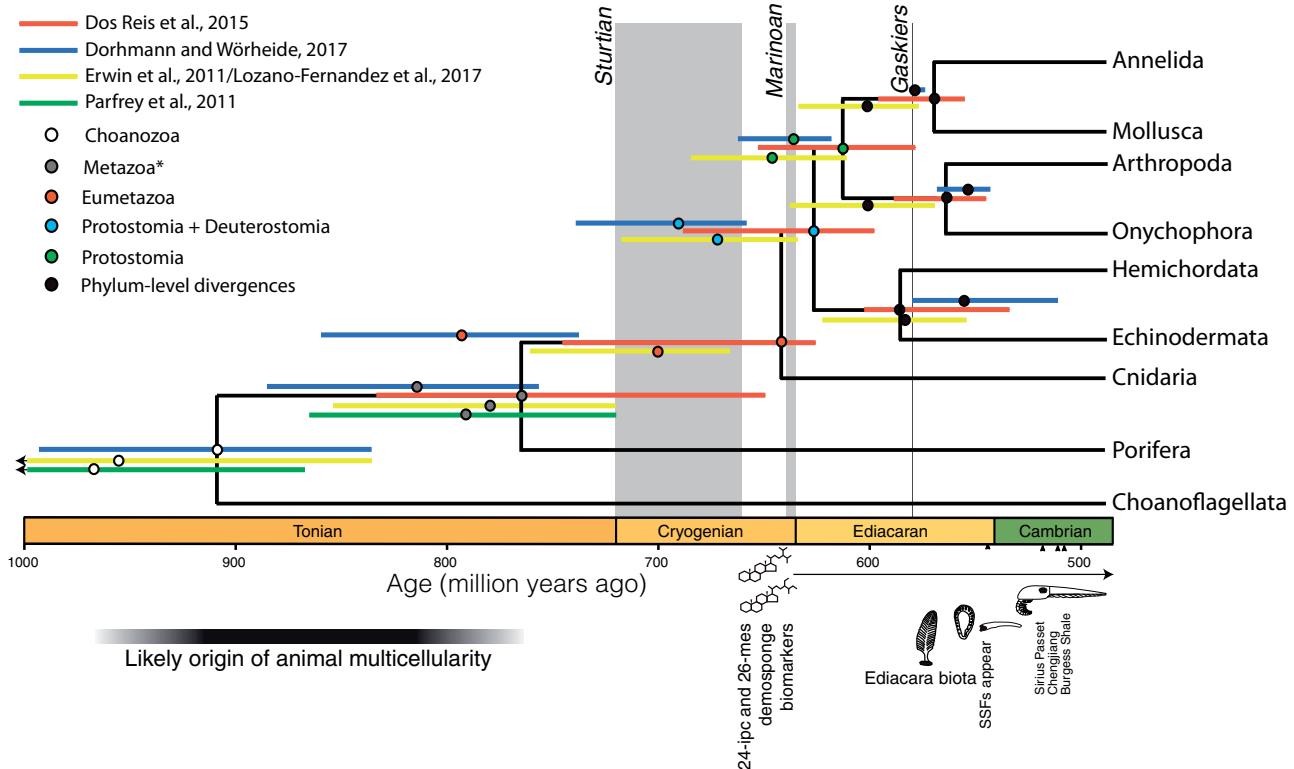


Fig. 1 Paleontological, organic geochemical, and molecular clock evidence regarding the timing of early animal evolution. Major Neoproterozoic glaciations are shown as vertical gray bars. Based on molecular clock evidence, animal multicellularity evolved between white circles (Choanoflagellata/Metazoa split; Choanozoa of Brunet and King [2017]) and gray circles (crown Metazoa). Colored bars represent 95% confidence intervals from four recent studies. Metazoa* marked with asterisk to recognize uncertainty in metazoan phylogeny; however, sensitivity analyses in which ctenophores are the sister group of all other animals reached consistent results (Dohrmann and Wörheide 2017). Organic geochemical evidence (24-ipc and 26-mes biomarkers) suggests the sponge body plan was present at least between the Sturtian and Marinoan glaciations in the Cryogenian. The first unequivocal fossil evidence for animals is found in the late Ediacaran in the form of bilaterian trace fossils and Small Shelly Fossils (SSFs), prior to the “explosion” of new body plans and large, active, biomineralized organisms in the Cambrian.

Temporal context

Fossil record

The most obvious path to studying the early history of animals is by direct investigation of the fossil record. Moving back in time from the Cambrian, the Ediacara biota represents the first large, complex fossils in the fossil record (reviewed by Boag et al. 2016; Droser et al. 2017; Tarhan et al. 2018). The first Ediacaran organisms appear in the “Avalon” assemblage of Newfoundland and UK after ~571 million years ago (Ma) (Noble et al. 2015; Pu et al. 2016). These generally represent enigmatic sessile frond-like organisms with a “fractal” organization that are unlikely to be animals (Sperling et al. 2007)—or at least show no obvious characters relating them to animals. However, trace fossils possibly representing diploblastic organisms are also present (Liu et al. 2010), and some body fossils may represent cnidarians (Liu et al. 2014) or sponges (Sperling et al. 2011). The

succeeding “White Sea” and “Nama” assemblages record a considerable increase in the diversity of form, movement, reproduction strategies, and biomineralization (Tarhan et al. 2018). The species making up these assemblages flourished only briefly, as possible Cambrian survivors are controversial, and certainly very rare even if genuine survivors (Laflamme et al. 2013; Budd and Jensen 2017). Whether these Ediacaran organisms suddenly became extinct at the end of the Ediacaran due to an environmental perturbation (Smith et al. 2016), or gradually disappeared due to biological interactions with evolving bilaterians (Darroch et al. 2015), remains a critical question in Ediacaran paleobiology (reviewed by Tarhan et al. [2018]). In any case, as noted by the title of a recent review paper on the Ediacara biota (Droser et al. 2017), these fossils in the latter half of the Ediacaran likely represent the “rise of animals” rather than their origin and early evolution, and consequently we must look earlier.

The early Ediacaran, Cryogenian, and Tonian fossil records have been relatively silent (or at least controversial) with respect to the origin of animals. As discussed below, a number of paleontologists believe this silence to be an accurate record. During these time periods a number of putative sponge fossils have been reported, but none can be fully accepted as representing evidence of early animals. A single phosphatized fossil named *Eocyathispongia qiania* from the Doushantuo Formation (~600 Ma) has sponge-like features and a body plan possibly consistent with a water-canal system (Yin et al. 2015). However, only a single specimen has been discovered, from a geological formation with a large variety of intriguing but clearly abiogenic phosphatic structures. For instance, *Vernanimalcula* from the Doushantuo was initially described as a bilaterian fossil (Chen et al. 2004), based on a few specimens discovered during an initial search of over 50,000 specimens (Bengtson et al. 2012). *Vernanimalcula* has now been convincingly demonstrated to be a pseudo-fossil, and examination of the full range of morphologies in the Doushantuo shows how *Vernanimalcula* is an end-member artifact of phosphatization (Bengtson et al. 2012). Putative siliceous demosponge spicules from the Doushantuo have also recently been invalidated (Muscente et al. 2015). Considering *Eocyathispongia* then, with only one specimen, and the possibility that its body plan is not consistent with a functioning water-canal system (Cavalier-Smith 2017), its status as a fossil sponge must remain uncertain (as acknowledged by the authors). Phosphatized embryos from the Doushantuo Formation (Xiao et al. 1998) still have promise for understanding evolutionary trends during the Ediacaran, but more plausibly represent single-celled organisms than animals (Huldtgren et al. 2011).

Moving farther back, sponge fossils have also been described from the Cryogenian of South Australia, but again they are not without issue. Maloof et al. (2010) applied a serial grinding technique to image specimens from the Trezona Formation, and described a population of millimeter- to centimeter-sized non-symmetric organisms with a network of interior canals leading to openings on the fossil surface. However, these openings are recessed into the surface of the fossil, rather than elevated like most sponge oscula, and would lead to recycling of the feeding current. It is also unclear if the postulated fossils match expectations for flow in biological fluid transport systems (LaBarbera 1990), and it is possible these clasts represent ripped up microbial mats (Antcliffe et al. 2014). Other possible Neoproterozoic sponges,

including even older reported fossils, were reviewed by Antcliffe et al. (2014) and found unconvincing.

In short, while many fossils have not been conclusively falsified as sponges—including the two fossils discussed above—neither can there be considered strong fossil evidence for sponges (or any other animal group) in the Cryogenian or Tonian. The key question is whether the fossil record, with an absence of Tonian to mid-Ediacaran animals, can be read literally with respect to the timing of animal origins. Many paleontologists believe it can (e.g., Budd and Jensen 2017). Alternative sources of evidence, such as the organic geochemical record and molecular clocks, suggest otherwise; below we discuss both of these records in turn.

Organic geochemistry

While most paleontologists search for body fossils, an alternative approach to interrogate the early history of clades is to search for molecular fossils: diagnostic organic molecules produced by organisms (Briggs and Summons 2014). These “biomarkers” are the hydrocarbon remains of lipids such as sterols in eukaryotes and hopanoids in bacteria. Compared with microbes, the biomarker complement of animals is relatively “boring” (i.e., not taxonomically specific)—with the exception of demosponges. One sterol in particular (a C₃₀ sterol, 24-isopropylcholesterol, with its fossilized sterane complement known as 24-isopropylcholestane, or 24-ipc) is exclusively made as a major sterol by living demosponges. Love et al. (2009) published a record of 24-ipc steranes from the South Oman Salt Basin, where it was found in Cryogenian strata—beneath the Marinoan glaciation—and throughout the Ediacaran into the Cambrian (Fig. 1). Critically, 24-ipc was found in both conventional solvent extracts and in the insoluble kerogen phase, helping rule out secondary contamination (which has been a recent concern in previous ancient biomarker studies; see French et al. [2015]). 24-ipc has also been found in “relatively high” abundance in other rocks worldwide of this age (McCaffrey 1994; Kelly et al. 2011). Note that “high” really is relative, as total C₃₀ steranes (of which 24-ipc is just one component) represent a minor component of total steranes in these Neoproterozoic rocks—typically only 1–4% of the total C₂₇–C₃₀ steranes (Love et al. 2009). Based on the phylogenetic pattern of 24-isopropylcholesterol synthesis in living organisms, Love et al. (2009) proposed that these Cryogenian and Ediacaran 24-ipc steranes were likely produced by total-group demosponges.

The inference that 24-ipc in Neoproterozoic rocks represent the preserved organic remains of Neoproterozoic demosponges has been controversial. Brocks and Butterfield (2009) suggested that unicellular relatives of animals could have produced these ancient biomarkers. Antcliffe (2013) suggested that these steranes could have been produced by pelagophyte algae (some of which produce trace quantities of 24-isopropylcholesterol), could have been a contaminant introduced by drilling fluid, or could have been produced by diagenetic alteration of other sterol skeletons. These latter two points are based on fundamental misunderstandings of organic geochemistry (Love and Summons 2015), but concerns about 24-ipc as a phylogenetically unique marker for demosponges are valid. To address this, Gold et al. (2016) conducted sterol assays of the living unicellular relatives to animals, and demonstrated they do not make 24-ipc sterol precursors (or indeed any C₃₀ sterols at all; see also Kodner et al. [2008]). Gold and colleagues then investigated the molecular underpinnings of 24-isopropylcholesterol synthesis, and demonstrated that demosponges and pelagophyte algae independently evolved the ability to methylate C₃₀ sterols. Given the timing of gene duplications for the known extant sterol producers, only demosponges, and not pelagophytes, could have been the source of Neoproterozoic 24-ipc. Most recently, Zumberge et al. (2017) discovered a new C₃₀ sterane biomarker, named 26-methylstigmastane (26-mes), whose sterol precursors are only produced by demosponges among the living biota. Thus, 26-mes does not suffer from some ambiguities present in interpreting the 24-ipc record. 26-mes was found in the same Cryogenian through Cambrian rock and oil samples as 24-ipc, providing convincing evidence of the pattern described by Love et al. (2009). Cryostane, a C₂₈ sterane, appears in even earlier pre-Sturtian rocks of the Tonian period and may have also been made by demosponges, but this is speculative because unlike 24-ipc or 26-mes the natural sterol precursors have not been identified (Brocks et al. 2016).

A subtle but important distinction is that, as far as currently known, 24-ipc and 26-mes are not sponge biomarkers but rather *demosponge* biomarkers. Certainly, more sterol assays and genomic studies are needed on other sponge groups and unicellular relatives of animals, but no other sponge or holozoon protist group has been found to have any unconventional steroid structure with respect to side-chain chemistry. Only conventional sterols with common straight-chain alkylated side-chains are found. The fact that these two unconventional

biomarkers are only made as a major sterol by demosponges (and not all sponges) indicates the presence of a derived group of sponges in the Cryogenian. Consequently, the Porifera crown group must have originated earlier. In other words, no matter the phylogenetic relationship of sponges and ctenophores to other animals, the biomarkers provide a minimum age constraint for crown-group sponges and suggest multicellular animals with a water-canal system were living prior to 635 Ma.

Molecular clocks

Molecular clocks calculate the rate of molecular evolution through time in groups with a presumably robust fossil record, and then in simplest form apply these rates across the phylogenetic tree to infer divergence times. This practice as applied to animal evolution has a somewhat sordid history (Graur and Martin 2004). However, beginning with the use of multiple calibration points spread phylogenetically across invertebrates (Peterson et al. 2004), and most importantly through the introduction of “relaxed clock” methods, analyses have reached broad agreement on the timing of animal origins (Fig. 1) (Aris-Brosou and Yang 2003; Peterson et al. 2008; Erwin et al. 2011; Parfrey et al. 2011; dos Reis et al. 2015; Dohrmann and Wörheide 2017; Lozano-Fernandez et al. 2017). Significant uncertainties do still remain regarding the validity of specific paleontological calibration points, how uncertainty around calibration points should be modeled, and whether uncorrelated or autocorrelated rate models should be used. In light of these uncertainties both dos Reis et al. (2015) and Cunningham et al. (2017) have provided useful and timely discussion of the precision afforded by molecular clocks. To address this uncertainty, the full range of possible parameter space in terms of gene sequences, partitions, possible paleontological calibration points, soft- versus hard-bounds on calibration points, calibration probability densities, root priors, phylogenetic topologies, rate models, and software choices have now been explored in sensitivity analyses—filling pages of supplementary tables and figures (see references above for details). Two robust messages have emerged.

First, and consistent with demosponge biomarker data, animal multicellularity emerged prior to the Ediacaran, possibly in the Cryogenian but more likely in the Tonian. Although there is considerable uncertainty on these deeper nodes, the mean divergence between sponges and all other animals is in the Tonian in all “most likely” analyses (e.g., the

main summary figure of a paper). Only in one analysis does the 95% confidence interval (CI) for crown-group animals extend into the Cryogenian (dos Reis et al. [2015]; note that we use updated definitions for the Cryogenian: 720–635 Ma, see Shields-Zhou et al. [2016]). Molecular divergence estimations using topologies where ctenophores rather than sponges are the sister group of all other animals have not been extensively examined, but analyses by Dohrmann and Wörheide (2017) demonstrated that the 95% CI for crown-group animals does not extend into the Cryogenian using this topology. Indeed, across the dozens of different sensitivity analyses conducted by multiple research groups, the mean estimate for crown group animals is never in the Cryogenian, and 95% CIs generally exclude this period. Likewise, in all “most likely” analyses the 95% CIs for crown-group Metazoa exclude the Mesoproterozoic (>1000 Ma), and only rarely do 95% CIs extend to the Mesoproterozoic in sensitivity analyses. The animal crown node of course represents the minimum time for the origin of animal multicellularity, which must have evolved along the metazoan stem-lineage. Most estimates place the animal–choanoflagellate divergence at ~900–1000 Ma (Fig. 1), suggesting that the dawn of animal multicellularity was likely in the Tonian even accounting for all sources of uncertainty (specifically between the white and gray nodes in Fig. 1).

The origin of eumetazoan and bilaterian body plans generally falls in the Cryogenian but there is considerable uncertainty. In contrast to the relative cross-study consensus for the animal crown node, for Eumetazoa and Bilateria the 95% CIs of some studies exclude the mean estimate of other analyses. The disparate ages between studies for Eumetazoa and Bilateria remain an area for investigation. In any case, the second robust finding from molecular clock studies is that the divergences between individual sister phyla likely occurred in the late Ediacaran (Fig. 1, black nodes). This pattern is best illustrated by the taxon-rich study of Erwin et al. (2011), where many more sister phyla were analyzed than could be visually included in Fig. 1. Molecular clock evidence often places the crown nodes of these phyla within the Cambrian when uncertainty is included, and/or identifiable fossil representatives of these phyla occur in the early–middle Cambrian (even if not having all the characteristics of the crown group; Budd and Jensen [2000]). Consequently, it is clear that the pulse of body plan innovation, skeletonization, and size increase that characterizes the Cambrian radiation took place as a polyphyletic radiation across

multiple lineages around the Precambrian–Cambrian boundary (Peterson et al. 2005). In essence, while animals have a cryptic Tonian origin according to molecular clocks and biomarker data, the explosion of new body plans recorded in the Cambrian fossil record is a real evolutionary event. As discussed in the “Introduction” section, this polyphyletic radiation implies an external forcing.

We recognize that this reading of the animal evolutionary record leans heavily on molecular clock and biomarker data over a literal reading of the fossil record. Most persuasive to us, in this regard, is that there is now strong concordance between molecular clocks and the fossil record in the Phanerozoic (Erwin et al. 2011; Cunningham et al. 2017; Daley et al. 2018). Put simply, if clades such as crown-group Arthropoda or Brachiopoda diverged in the early Cambrian, as indicated by both clocks and rocks, there is no way to telescope all the molecular evolution between these divergences and the last common ancestor of animals into the ~30 million years demanded by proponents of a literal reading of the fossil record (Antcliffe et al. 2014; Budd and Jensen 2017). This is true even if accelerated (but plausible) rates of molecular evolution existed in some lineages (Lee et al. 2013).

The view presented here then leads to the eternal question of why early animals are not fossilized if they arose ~800 Ma. Some of this tension relates to time of clade origin (reflected by molecular clocks) versus time of ecological dominance (recorded by the fossil record), and also to some mis-understandings of various records. For instance, the presence of Cryogenian demosponges as supported by biomarkers evokes a seafloor covered in sponges. But as discussed above, the 24-ipc biomarker only occurs at the ppm level in sterol extracts: only minute inputs are required. A long preservational gap is also not an anomaly in the deep-time fossil record. As an example, oxygenic photosynthesis (the major source of oxygen on Earth) must have appeared prior to or coincident with the Great Oxygenation Event at ~2300–2400 Ma, but the first cyanobacterial fossils do not appear until hundreds of millions of years later, at ~2000 Ma (Fischer et al. 2016). Below we discuss some factors (mainly rarity and small body size) that may have played into low preservational potential for early Neoproterozoic animals. However, like most paleontologists, do not find them fully convincing. We view this taphonomic debate, as well as the basic question of temporal origins, as still-open scientific questions. For the purposes of discussing causes, though, we must take this current best-

guess understanding of timing in order to move forward and examine the type of environments early animals evolved in, how the marine environment may have changed during their early evolutionary history, and how these changes may have played into the Cambrian radiation.

Environmental context

Animals evolved in a low-oxygen ocean

It has long been hypothesized that animals evolved in a less oxygenated ocean than exists today (e.g., [Nursall 1959](#)). Some of this initial reasoning was circular—for instance some evidence for lower oxygen levels hinged on the fact that there were no animals (reviewed by [Mills and Canfield 2014](#))—but it was also clear from straightforward clues in the rock record, like widespread Archean and Proterozoic Banded Iron Formations, that the early Earth was less oxygenated than the modern Earth. In the past decade, more sophisticated geochemical redox studies have generally supported this narrative, providing both local and global evidence for widespread Proterozoic subsurface anoxia ([Partin et al. 2013](#); [Reinhard et al. 2013](#); [Liu et al. 2016](#); [Sperling et al. 2015b](#); [Hardisty et al. 2017](#)). The presumption is that widespread Proterozoic marine anoxia was a direct result of lower atmospheric oxygen concentrations, although there are arguments against this ([Butterfield 2018](#)). Oxygen levels are generally believed to have increased substantially at some point between the mid-Ediacaran to early Cambrian (the hypothesized Neoproterozoic Oxygenation Event; [Shields-Zhou and Och 2011](#)). It is this rise in oxygen that is commonly invoked as the geologic trigger for the Cambrian “explosion.” Whether atmospheric oxygen levels rose to modern in the Ediacaran–Cambrian, or the increase was smaller and oxygen levels remained relatively muted through the early Paleozoic, is a current topic of debate ([Dahl et al. 2010](#); [Chen et al. 2015](#); [Sperling et al. 2015b](#); [Wallace et al. 2017](#); [Stolper and Keller 2018](#)), as are the implications of a muted rise for the Cambrian explosion ([Sperling et al. 2015a](#)). However, the prevalence of subsurface marine anoxia is a clear message of the Neoproterozoic geochemical record.

The implications for early animals of a poorly oxygenated ocean have been extensively explored in the literature. Based on theoretical calculations, animals would be limited to relatively small and thin body plans ([Raff and Raff 1970](#); [Runnegar 1982a](#); [Payne et al. 2010](#)). This is matched by empirical evidence from transects across modern oxygen minimum zones (OMZs), which show that animals living in

naturally very low-O₂ settings are small, thin, generally unmineralized, and have low-energy (non-carnivorous) feeding strategies ([Rhoads and Morse 1971](#); [Levin 2003](#); [Gooday et al. 2009](#); [Sperling et al. 2013b, 2015a](#)). At the very lowest oxygen levels, macrofaunal abundance (organisms retained on a 300 μm sieve) drops dramatically, and the dominant animals are meiofauna such as nematode worms ([Gooday et al. 2009](#)). This has important implications for fossil taphonomy and reconciliation between the fossil record and molecular/biomarker data. As noted by [Sperling \(2013\)](#), nematodes occur by the 100–1000s in every square meter of modern marine seafloor, and probably have for quite some time (the nematode crown group is likely at least 500 million years old; [Erwin et al. 2011](#)). Yet, paleontologists have only rarely, if ever, found a marine nematode body fossil (most putative fossil marine nematodes are at least an order of magnitude larger than living nematodes and show few characteristic features). Nematodes are preserved under exceptional conditions such as in terrestrial amber, but in the marine realm such small, thin animals simply do not preserve (reviewed by [Poinar 2011](#)). [Budd and Jensen \(2000\)](#) argued that ancestral bilaterians must have been larger than nematodes, at least several millimeters long, but such organisms also lack an extensive fossil record. Consequently, if oxygen levels were low enough to keep marine “worms” small and thin, there is little discrepancy between molecular clock ages for bilaterians in the Cryogenian and the absence of an unequivocal bilaterian fossil record until the late Ediacaran.

Animals evolved in a food-limited ocean

While most studies of the Neoproterozoic environment have focused on oxygen and redox, emerging evidence suggests there were also important differences in primary productivity and food delivery to the seafloor compared with the Phanerozoic. Analysis of a large compilation of sedimentary phosphorous data supports modeling results indicating that anoxic, ferruginous Proterozoic ocean waters may have effectively scavenged bioavailable P, and that oceans during that time could have been considerably P-limited ([Derry 2015](#); [Reinhard et al. 2017](#)). Phosphorous limitation appears to have eased at some point in the Neoproterozoic, although the exact global timing is obscured by a dearth of compiled samples from outside South China ([Reinhard et al. 2017](#)). Whether linked to these changes in nutrients or not, the Neoproterozoic also witnessed a change in the dominant primary producers, with

organic geochemical data demonstrating that algae broke the incumbency of cyanobacteria between the Sturtian and Marinoan “Snowball Earth” glaciations (Brocks et al. 2017). Most discussion in these and other papers have related changes in primary productivity back to oxygen production in the Earth system, and a more detailed examination of how changes in food supply might have affected early animal ecosystems is required.

To further understand how food supply to the seafloor changed through time, we investigate here the direct record of organic carbon in sedimentary rocks. Specifically, we have conducted the largest compilation of total organic carbon (TOC) contents in shale (fine-grained clastic rocks) to date. This lithology was chosen because (1) it provides a consistent and geographically widespread lithology through time, (2) there are abundant shale TOC measurements in the literature, and (3) shale depositional environments represent the greatest sink for organic carbon in the geological record (Berner 1982). Our analyses combine previous compilations of TOC assembled from US Geological Survey (USGS) data (compiled by Husson and Peters [2017]; 3345 samples), and geochemical compilations focused on other elemental systems but with associated TOC data: specifically studies by Reinhard et al. (2017; 5207 samples) and Partin et al. (2013; 711 samples). To these existing compilations we added new data from the literature (3437 samples). Finally, 992 new shale TOC analyses conducted at Stanford University are reported here. The complete dataset consisted of 13,650 shale TOC measurements through Earth history. Full analytical methods, details of the compilation, and additional caveats are located in the [Supplementary Information](#).

Our results show a distinct change in shale TOC contents at the base of the Phanerozoic (Fig. 2). Specifically, Neoproterozoic shales have a mean organic carbon content of 0.8 wt% (median, 0.2%), compared with 4.3 wt% (median, 2.6%) for the Phanerozoic. These differences are highly significantly different (e.g., $P < 2.2 \times 10^{-16}$ for a two-group Mann–Whitney U -test), with our statistical subdivisions supported by a large TOC increase evident from the first time bin fully in the Cambrian (525–500 Ma). When considering these results, it is worth noting there are a large number of depositional and post-depositional factors that influence the TOC value of a rock. A non-exhaustive list includes sedimentation rate, bottom-water O_2 content, and type of organic carbon (Canfield 1994), organic carbon loss during thermal maturation (and eventually, metamorphism) (Raiswell and

Berner 1987), and oxidative weathering in outcrop versus core (Petsch et al. 2000). We have not attempted to correct for these factors, or for spatial-temporal sampling density, and thus these results should be viewed as a “first pass.” However, we do not anticipate secular changes in either biology or geology that would inhibit interpretation of these data as a first-order summary of benthic food supply.

Given available data, then, the Neoproterozoic is a clear outlier. In fact, the main difference in shale TOC contents through time is not between the Proterozoic and Phanerozoic, but rather between the Neoproterozoic and all other time periods (see Summary Distributions, Fig. 2). Considering the relative lack of data from the Paleoproterozoic and Mesoproterozoic, and the sampling dominance of a few formations, we are agnostic as to whether these Era-level differences are real. The post-850 Ma interval of the Neoproterozoic, though, is much more consistently sampled in space and time. The substantial difference between this interval—when cryptic animals evolved—and the Phanerozoic is likely to be real. It is also pertinent that the most obvious geochemical signals are in high TOC shales, and consequently, geochemists tend to preferentially sample the most TOC-rich horizons. If abundant, high TOC Neoproterozoic shale formations existed, they would have been sampled. These data contrast with the relatively enriched background carbonate carbon isotope record of the Neoproterozoic (Macdonald et al. 2010), which is often taken to represent high fractional rates of organic carbon burial. Aspects of the Neoproterozoic carbon isotope record in general are difficult to explain using conventional frameworks (discussed in Schrag et al. 2013), and these shale TOC data are another line of evidence that must be reconciled.

Other anecdotal lines of evidence can be found to support the shale TOC record. For instance, the rate of organic carbon supply to investigated Neoproterozoic manganese deposits was lower (Johnson et al. 2016), and paleontological evidence suggests Avalonian Ediacaran ecosystems may have been more food-limited than the Cambrian (Vinther et al. 2014; Hoyal-Cuthill and Conway Morris 2017). Many Neoproterozoic carbonates also have low TOC (Johnston et al. 2012a). Finally, given the feedback between organic carbon burial and oxygen production, recent modeling results suggest that a low- O_2 ocean/atmosphere system is difficult (or impossible) to achieve without low primary productivity (Laakso and Schrag 2014, 2017; Derry 2015). Accepting a low O_2 Neoproterozoic world

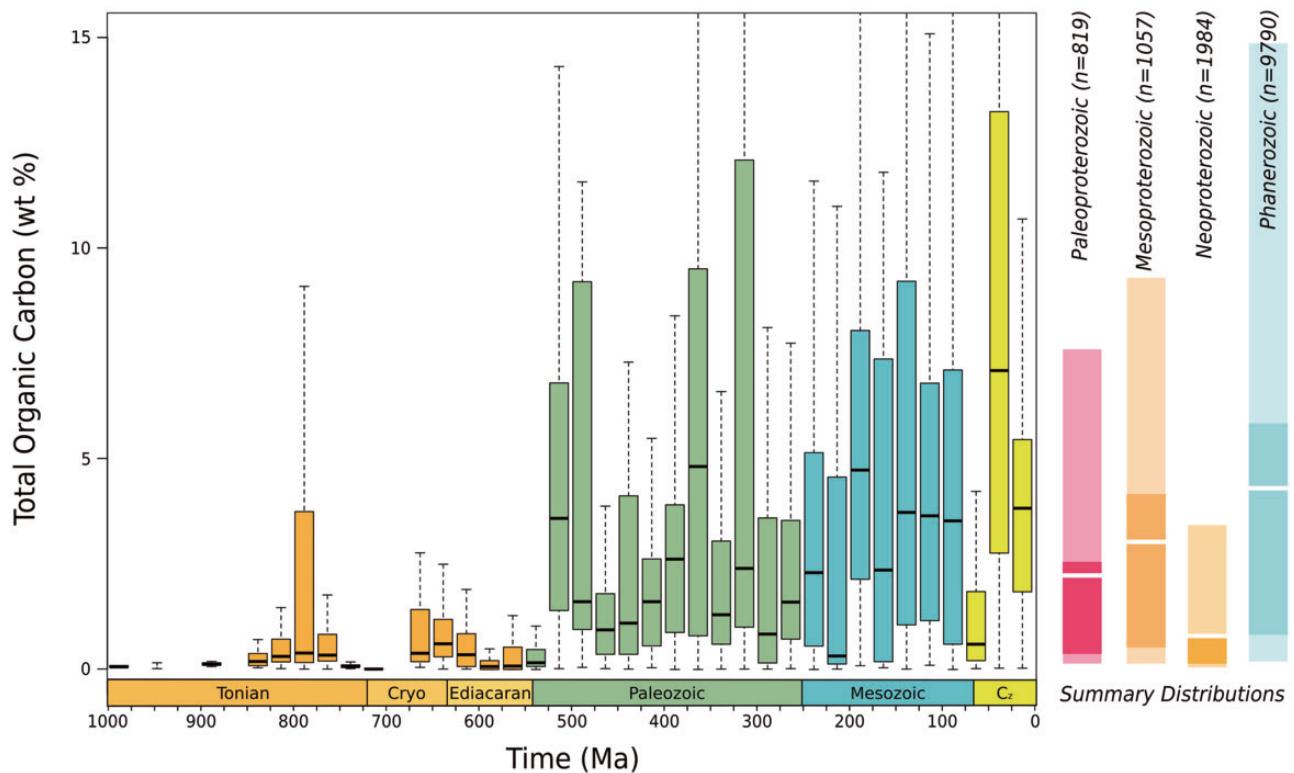


Fig. 2 Boxplot analysis of 13,650 shale total organic carbon (TOC) weight percent measurements through Earth history. Left panel depicts the last 1000 million years of Earth history with data binned into 25 million year intervals; the Cambrian “explosion” is at ~525 Ma, the base of the Paleozoic. In boxplots, the thick horizontal lines represent median values, boxes represent first quartile and third quartile, and dashed lines extend to 1.5 times inter-quartile range. Outliers removed for figure clarity. Right panel depicts summary distributions for the Paleoproterozoic (2500–1600 Ma), Mesoproterozoic (1600–1000 Ma), Neoproterozoic (1000–541 Ma), and Phanerozoic (541–0 Ma) eras. In summary boxes, thick white lines represent means, dark colored boxes represent first and third quartiles, and light colored boxes represent 5th and 95th percentiles.

likely requires accepting a low-food Neoproterozoic world.

The shale TOC results in Fig. 2 do not necessarily speak to overall rates of primary productivity, oceanic nutrient inventories, or the dominance of primary producer types, which likely began shifting earlier in the Neoproterozoic (Brocks et al. 2017; Reinhard et al. 2017). Higher primary productivity may have instead been balanced by more respiration of sinking organic matter. However, what the TOC results do estimate is the flux of organic carbon to seafloor ecosystems (export productivity), which is what would have mattered to early benthic animals. And this is essentially all early animals, as aside from possibly medusozoan cnidarians, Neoproterozoic animal evolution was a benthic affair (reviewed by Gold 2018). Further, if oxygen levels were low in the Proterozoic, O₂ would have interacted synergistically with low organic carbon supply in two ways to limit early animals’ access to food. First, some early animals such as sponges (which are constrained to be present by the Cryogenian, see above) are likely to have had very high energetic costs of feeding (Leys

and Kahn 2018). High relative metabolic cost of feeding combined with low food supply would obviously limit scope for growth, and perhaps the geographic extent of metabolically viable habitat. Second, in a low-O₂ world, any area of higher primary productivity (such as upwelling zones) would have quickly lost all oxygen in the water column due to aerobic remineralization of relatively abundant organic carbon. In the modern ocean animals can survive and even thrive in low oxygen, but not in the complete absence of oxygen (Sperling et al. 2013b), and early skeletal metazoans were unable to inhabit low-oxygen areas (Tostevin et al. 2016). This is not to argue that Neoproterozoic animals would have been excluded from all possible food-rich areas, but basic biogeochemistry does suggest they would have been pushed toward primary productivity “deserts.” Hammarlund et al. (2017) noted that exclusion from higher productivity areas may have played a role in structuring Cambrian ecosystems. We argue this effect likely played an even *bigger* role in the Neoproterozoic, when oxygen levels are canonically believed to be far lower.

Implications of a food-limited Neoproterozoic ocean

If Neoproterozoic food supply was lower, and synergistic environmental interactions kept animals from productive regions, how did this affect early animals? In terms of body plan evolution, many morphological characteristics of early animals may have been adaptations to food-poor environments (Leys and Kahn 2018). To answer such questions at the ecosystem level, paleontologists have long looked to ecological patterns along natural environmental gradients in the modern ocean (e.g., Rhoads and Morse [1971] for oxygen). The sharpest natural gradients for almost every environmental parameter occur along continental margins (Levin and Sibuet 2012), with the change from very low to higher oxygen levels (re-capitulating the presumed Neoproterozoic–Paleozoic environmental transition) occurring at relatively shallow bathyal depths in OMZs. In contrast, the transition from very low to higher food supply (again re-capitulating the presumed ancient trend) occurs in the deep abyss, toward the center of ocean gyres. This is due to low primary productivity in the overlying waters, and very long transport distance to the seafloor, across which organic carbon rain is attenuated by progressive remineralization.

Ecological changes across these abyssal food supply gradients are interesting to consider. The frigid, stenothermal, continuously dark, and high-pressure environments of the abyss inhibit a perfect analogy for shelfal/bathyal evolution in the Neoproterozoic and Cambrian. However, the abyss is attractive as a model because so many environmental factors are invariant, and variations in chemical energy (food) can therefore be studied as a relatively isolated system (in contrast to OMZs, where many environmental parameters co-vary, Sperling et al. [2016]). Recent mega-analyses of the deep-sea have established firm links between organismal abundance, body size, animal biomass, and food supply (Smith et al. 2008; Wei et al. 2010; McClain et al. 2012). The most recent large-scale analysis of food supply gradients in the global ocean also demonstrated a positive relationship between species-level diversity in the deep-sea and chemical energy (Woolley et al. 2016). This contrasts with shallow-water environments, where kinetic energy (temperature) is a better predictor of diversity (Tittensor et al. 2010; Woolley et al. 2016). The overall pattern in the deep-sea seems clear, but note questions remain regarding the exact relationship between diversity and chemical energy (for instance, linear versus quadratic [McClain et al. 2012]), and whether the relationship exists in all areas and

for all taxonomic groups, e.g., Glover et al. [2002]). The sediment mixed layer depth (bioturbation) is also generally shallower in areas with lower food supply or nutritional quality (Smith and Rabouille 2002; Smith et al. 2008), although more study is clearly needed (Teal et al. 2008). Unfortunately for consideration of how feeding strategies change with food supply, quantitative analysis of feeding strategies, or indeed even direct evidence of feeding mode for any given species, is relatively rare for deep-sea organisms (Gage and Tyler 1991). Qualitative analyses, though, suggest that low-food abyssal regions are (1) dominated by deposit feeders, (2) have higher occurrences of omnivory, to better utilize any available food source, and (3) have relatively few specialist carnivores, likely because prey simply is not available (Gage and Tyler 1991).

A final and intriguing lesson regarding food supply in the modern ocean concerns organismal responses to exogenous environmental stresses. Recent experimental studies have demonstrated that animals can offset the negative effects of stressors such as ocean acidification through increased food supply (Melzner et al. 2011; Hettinger et al. 2013; Sanders et al. 2013). It is more energetically costly to calcify under acidified conditions, but increased food provides energy to pump ions more vigorously and maintain the correct carbonate chemistry at the site of calcification. Growth will even increase with more food in spite of adverse conditions: over a range of external carbonate chemistry conditions, this is simply an energetics problem. Likewise, heat-shock proteins and other molecular chaperones involved in stress responses require energy for protein synthesis. Environmental instability in Neoproterozoic oceans at a variety of temporal and spatial scales is increasingly being recognized (Johnston et al. 2012b; Reinhard et al. 2016; Lu et al. 2017; Wood and Erwin 2018), and given the relationship between food supply and stress tolerance seen in modern experiments, it can be predicted that food-limited Neoproterozoic animals would have been relatively more susceptible to short-term environmental fluctuations.

Oxygen, food, and the fossil record

Studies of natural environment gradients in the modern ocean suggest that under very low oxygen levels animal communities are dominated by small size classes, are comprised of unskeletonized organisms with low-energy, non-carnivorous feeding strategies, have shallower (but often intense) bioturbation levels, and have low species-level diversity (Levin

2003; Sperling et al. 2013b, 2016). As discussed above, animal communities with very low food supply are also dominated by smaller size classes, are comprised of organisms with low-energy, non-carnivorous feeding strategies, have shallow bioturbation levels, and have low organismal abundance and species-level diversity. These are listed back-to-back to emphasize the point: there are strong first-order ecological similarities between low-oxygen and low-food areas of the modern ocean (Fig. 3). This has important implications regarding the lack of obvious animal fossils in the ~800–550 Ma interval, when animals were apparently present based on molecular clock and biomarker data but not preserved in the fossil record (Fig. 1). This is often attributed to low- O_2 oceans (for instance, Sperling [2013]). The results of Fig. 2, combined with ecological patterns in modern abyssal ocean regions, suggest that food supply may have been equally important in explaining the scarce fossil record of early animals. Specifically, a crucial difference between modern OMZs and the abyss is the relationship between animal abundance and body size. In both low-food and low-oxygen regimes animal body sizes are depressed, but abundances in OMZs—for the small, thin organisms that can tolerate very low O_2 —can be incredibly high due to abundant food and lack of predators (Fig. 3). For instance, Levin et al. (2002) found ~15,000 nematodes and ~60,000 foraminifera per square meter of seafloor at a station off Peru with bottom-water oxygen levels of 0.02 ml/l ($<1\ \mu\text{M}$) and TOC contents of ~17 wt%. In the modern ocean, there are simply no appropriate analogs for combined low food supply and low oxygen, as OMZs are directly caused by high organic carbon delivery. We therefore have to examine environments with low oxygen and organic carbon delivery separately, and infer how observed biotic responses might interact. Simple logic would suggest the ecology of this postulated low- O_2 /low food Neoproterozoic ocean would combine aspects of modern ocean OMZs and abyssal plains: early animals would be small, thin, and rare. This hinges of course on the veracity of the geochemical record; it has been proposed that geochemists have fundamentally mis-understood the record (Butterfield 2018). However, if true, this environmental landscape would provide an even more plausible explanation for the discrepancy between molecular clock evidence for bilaterians in the early Ediacaran or before and the first fossil evidence for bilaterians at the end of the Ediacaran (Droser et al. 2017; Parry et al. 2017) than oxygen-related hypotheses alone.

From this (likely) small, thin, rare, and unfossilized Neoproterozoic animal biota, the end of the Ediacaran and early Cambrian record increases in body size, diversity, skeletonization, abundance, biomass, and depth of bioturbation. The early Cambrian also witnessed the rise of bilaterian predators (Sperling et al. 2013a). When environmental factors are considered as possible drivers of these Cambrian biological changes, oxygen is most often invoked. But again, when using the modern ocean space-for-time translation most of these biological changes could alternatively be explained through changing food supply (Fig. 3). Looking across natural gradients in the modern ocean, perhaps the only Ediacaran–Cambrian biological change that appears influenced by oxygen gradients but not food gradients is the appearance of biomineralized forms, as calcified echinoderms and molluscs are relatively common in the abyss (Smith et al. 2008). Note though that we are not aware of any actual studies of biomineralization in the deep sea, and the relative lack of calcified forms in the cores of OMZs could alternatively be because modern O_2 minima are CO_2 maxima (i.e., acidic) due to the remineralization of organic carbon (Paulmier et al. 2011).

An increase in nutrients and food has been proposed as a driver of marine ecosystem change in the Phanerozoic (Bambach 1999), and during the 1980s and 90s was considered as a possible player in the Cambrian radiation (Cook and Shergold 1984; Brasier 1992; Cook 1992). But these Cambrian studies were mainly concerned with trends in phosphatic skeletonization and preservation, and in any case, such hypotheses have fallen from favor. Given new observations on food supply through the Neoproterozoic–Paleozoic, and the inherent linkage between oxygen and primary productivity (Laakso and Schrag 2014; Derry 2015), they should clearly be revisited. Discussion of the ultimate cause(s) of such oxygen and/or primary productivity changes—from disparate geological triggers to ecological feedbacks—are beyond the scope of this article (see recent papers by Lenton et al. 2014; Husson and Peters 2017; Laakso and Schrag 2017; Reinhard et al. 2017; Butterfield 2018). We note though that specific causal forcings of oxygen and organic carbon dynamics are irrelevant to early animal ecosystems, aside from any additional environmental side effects. Rather, here we simply raise the alternative hypothesis that if oxygen and food supply changed in tandem through the Ediacaran–Cambrian interval, many of the biological responses commonly ascribed to oxygen changes could equally well have been driven by food.

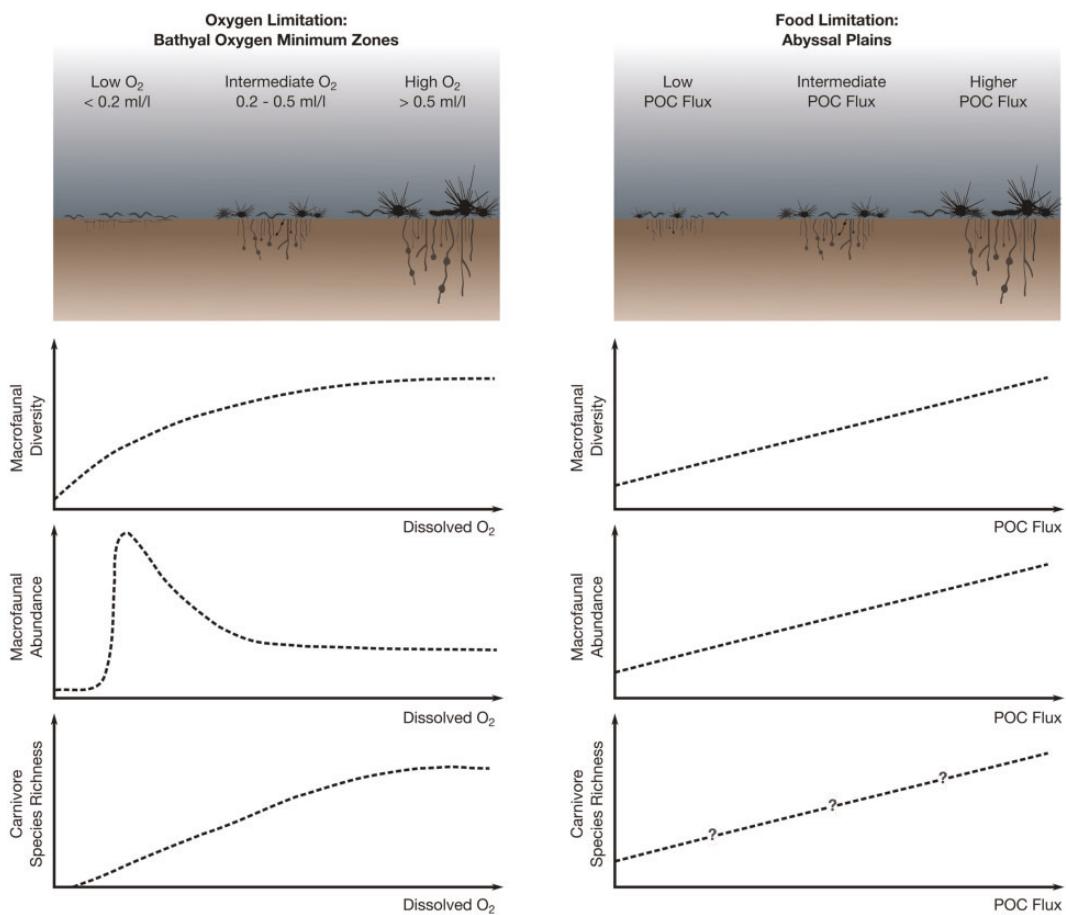


Fig. 3 Comparison of animal ecology across natural gradients in oxygen (left panel; modern bathyal oxygen minimum zones) and food supply (right panel; modern abyssal plains). Figures are schematic and scales are unitless so as to not imply equivalency of responses between the right and left panel. The illustrative point is that body size, depth of bioturbation, species diversity, abundance, and carnivore species richness increase across natural gradients of both oxygen and food supply. Note that neither panel considers the full spectrum of food supply/oxygen in the modern ocean. For oxygen, many ecological responses exhibit “threshold” like effects (Sperling et al. 2015a, 2016). For food supply, the figure only considers changes from very low food supply to “higher” food supply on abyssal plains. On continental margins at very high levels of food supply (i.e., organic carbon loading), ecological factors such as diversity drop again due to the physiological stress from developing low oxygen conditions (Levin et al. 2001). Figure based on data in Levin (2003), Smith et al. (2008), Gage and Tyler (1991), and Sperling et al. (2015a, 2016).

Considering all ingredients of an explosion

In an insightful paper on early animal evolution and the Cambrian “explosion,” Runnegar (1982b) evocatively closed by writing “the other ingredient, as in most explosives, may have been a strong oxidizing agent.” The idea that oxygen change was the key factor in the geologically sudden appearance of large, complex animals in the fossil record has increasingly dominated geological thinking and research in the subsequent decades. We do not disagree with Runnegar’s conclusion or this general line of research. Animals are clearly limited by low oxygen, and if oxygen levels rose past critical thresholds, this is an obvious trigger to explain many aspects of the Cambrian radiation. [Note such thresholds

are lower than previously believed, and also that animal ecosystems respond non-linearly to oxygen changes (Sperling et al. 2015a).] But we also believe it is worthwhile to step back and consider Runnegar’s closing metaphor more broadly. Students of (literal) explosions know that oxygen is one key ingredient of an explosion, but insufficient on its own. For instance, discussing explosive forest fire events, Maclean (1992) wrote “An old-timer knows that, when a ground fire explodes into a crown fire with nothing he can see to cause it, he has not witnessed spontaneous combustion but the outer appearance of the invisible pressure of a ‘fire triangle’ suddenly in proper proportions for an explosion.”

For future study of environmental influences on early animal evolution and particularly the Cambrian

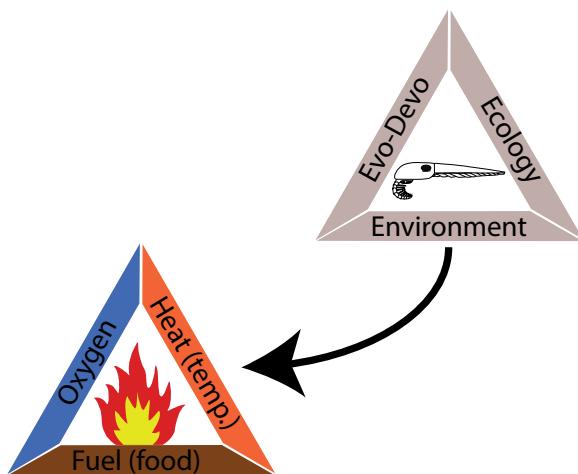


Fig. 4 Causal triangles for the Cambrian “explosion.” This evolutionary event likely involved multi-faceted drivers including genomic and developmental innovations (Evo-Devo), new ecological interactions, including the advent of widespread predation and ecosystem engineering, and environmental change. Environmental drivers were themselves likely multi-causal and involved multiple linked changes to the Earth system. This is illustrated by the “fire triangle”: an explosion requires oxygen, fuel (food supply to ecosystems), and heat (regional and global temperature dynamics) in proper proportion.

“explosion,” we suggest this “fire triangle” might be an appropriate metaphor. The classic fire triangle relates heat (flame or spark), an oxidizing agent (oxygen) and fuel (buildings, plants, trees, etc.), in the correct mixture (Fig. 4). For environmental influences on the Cambrian radiation, we might consider the first leg of the triangle to be global and regional temperature dynamics (heat). Temperature interacts synergistically with oxygen (through effects on metabolic rate) to limit aerobic habitability (Pörtner 2010; Deutsch et al. 2015), plays important and sometimes non-intuitive roles in oxygen solubility and diffusion (Verberk et al. 2011), and is the source of kinetic energy for ecosystems (Tittensor et al. 2010). The second leg would be oxygen, the focus of extensive geobiological research. The third leg, fuel, would represent primary productivity and labile organic carbon delivery to the seafloor. We note the “fire triangle” metaphor is not a perfect one—fires/explosions are destructive, whereas the Cambrian radiation is about the construction of animal body plans and ecosystems. Nonetheless, we believe it has conceptual utility as a current reading of the geological literature might suggest it was only the second leg (oxygen) that was important. Considering there were very large temperature changes in the Neoproterozoic (Hoffman et al. 2017), and clear changes to organic carbon abundance, type, and patchiness (Fig. 2; Brocks et al. 2017; Budd and

Jensen 2017; Reinhard et al. 2017), this is unlikely to be the case. In other words, even if oxygen is ultimately demonstrated to be the most important environmental factor, we propose that consideration of a complete “fire triangle” of metabolically important environmental factors for the Cambrian “explosion” will help prevent tunnel vision that may be creeping in regarding the role of oxygen. Recognizing this broader biogeochemical framework is critical as environmental factors themselves are just one leg of an even broader causal triangle composed of the evolution of genomic/developmental factors, ecological, and environmental influences (Fig. 4). Given the fundamental linkages between biology and geology within the Earth system, the Cambrian radiation increasingly appears driven by a cascade of factors rather than a single, isolated trigger.

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Supplementary data

Supplementary data available at *ICB* online.

References

Antcliffe JB. 2013. Questioning the evidence of organic compounds called sponge biomarkers. *Palaeontology* 56:917–25.

Antcliffe JB, Callow RHT, Brasier MD. 2014. Giving the early fossil record of sponges a squeeze. *Biol Rev* 89:972–1004.

Aris-Brosou S, Yang Z. 2003. Bayesian models of episodic evolution support a late Precambrian explosive diversification of the Metazoa. *Mol Biol Evol* 20:1947–54.

Bambach RK. 1999. Energetics in the global marine fauna: a connection between terrestrial diversification and change in the marine biosphere. *Geobios* 32:131–44.

Bengtson S, Cunningham JA, Yin C, Donoghue PCJ. 2012. A merciful death for the earliest bilaterian, *Vernanimalcula*. *Evol Dev* 14:421–7.

Berner RA. 1982. Burial of organic carbon and pyrite sulfur in the modern ocean: its geochemical and environmental significance. *Am J Sci* 282:451–73.

Boag TH, Darroch SAF, Laflamme M. 2016. Ediacaran distributions in space and time: testing assemblage concepts of earliest macroscopic body fossils. *Paleobiology* 42:574–94.

Brasier MD. 1992. Nutrient-enriched waters and the early skeletal fossil record. *J Geol Soc* 149:621–9.

Briggs DEG. 2015. Extraordinary fossils reveal the nature of Cambrian life: a commentary on Whittington (1975) 'The enigmatic animal *Opabinia regalis*, Middle Cambrian, Burgess Shale, British Columbia.'. *Philos Trans R Soc B* 370:20140313.

Briggs DEG, Summons RE. 2014. Ancient biomolecules: their origins, fossilization, and role in revealing the history of life. *BioEssays* 36:482–90.

Brocks JJ, Butterfield NJ. 2009. Early animals out in the cold. *Nature* 457:672–3.

Brocks JJ, Jarrett AJM, Sirantoin E, Hallmann C, Hoshino Y, Liyanage T. 2017. The rise of algae in Cryogenian oceans and the emergence of animals. *Nature* 548:578–81.

Brocks JJ, Jarrett AJM, Sirantoin E, Kenig F, Moczydłowska M, Porter S, Hope J. 2016. Early sponges and toxic protists: possible sources of cryostane, an age diagnostic biomarker antedating Sturtian Snowball Earth. *Geobiology* 14:129–49.

Brunet T, King N. 2017. The origin of animal multicellularity and cell differentiation. *Dev Cell* 43:124–40.

Budd GE, Jensen S. 2000. A critical reappraisal of the fossil record of the bilaterian phyla. *Biol Rev* 75:253–95.

Budd GE, Jensen S. 2017. The origin of the animals and a 'Savannah' hypothesis for early bilaterian evolution. *Biol Rev* 92:446–73.

Butterfield NJ. 2007. Macroevolution and macroecology through deep time. *Palaeontology* 50:41–55.

Butterfield NJ. 2011. Animals and the invention of the Phanerozoic Earth system. *Trends Ecol Evol* 26:81–7.

Butterfield NJ. 2018. Oxygen, animals and aquatic bioturbation: an updated account. *Geobiology* 16:3–16.

Canfield DE. 1994. Factors influencing organic carbon preservation in marine sediments. *Chem Geol* 114:315–29.

Cavalier-Smith T. 2017. Origin of animal multicellularity: precursors, causes, consequences—the choanoflagellate/sponge transition, neurogenesis and the Cambrian explosion. *Philos Trans R Soc Lond B Biol Sci* 372:20150476.

Chen J-Y, Bottjer DJ, Oliveri P, Dornbos SQ, Gao F, Ruffins S, Chi H, Li C-W, Davidson EH. 2004. Small bilaterian fossils from 40 to 55 million years before the Cambrian. *Science* 305:218–22.

Chen X, Ling H-F, Vance D, Shields-Zhou GA, Zhu M, Poulton SW, Och LM, Jiang S-Y, Li D, Cremonese L, et al. 2015. Rise to modern levels of ocean oxygenation coincided with the Cambrian radiation of animals. *Nat Commun* 6:7142.

Cook PJ. 1992. Phosphogenesis around the Proterozoic–Phanerozoic transition. *J Geol Soc Lond* 149:615–20.

Cook PJ, Shergold JH. 1984. Phosphorus, phosphorites and the skeletal evolution at the Precambrian–Cambrian boundary. *Nature* 308:231–6.

Cunningham JA, Liu AG, Bengtson S, Donoghue PCJ. 2017. The origin of animals: can molecular clocks and the fossil record be reconciled? *BioEssays* 39:1–12.

Dahl TW, Hammarlund EU, Anbar AD, Bond DPG, Gill BC, Gordon GW, Knoll AH, Nielsen AT, Schovsbo NH, Canfield DE. 2010. Devonian rise in atmospheric oxygen correlated to the radiation of terrestrial plants and large predatory fish. *Proc Natl Acad Sci U S A* 107:17911–5.

Daley AC, Antcliffe JB, Drage HB, Pates S. 2018. Early fossil record of Euarthropoda and the Cambrian Explosion. *Proc Natl Acad Sci U S A* published online (doi: 10.1073/pnas.1719962115).

Darroch SAF, Sperling EA, Boag TH, Racicot RA, Mason SJ, Morgan AS, Tweedt S, Myrow P, Johnston DT, Erwin DH, et al. 2015. Biotic replacement and mass extinction of the Ediacara biota. *Proc R Soc B* 282:20151003.

Darwin C. 1859. *On the origin of species by means of natural selection, or preservation of favoured races in the struggle for life*. London: John Murray.

Derry LA. 2015. Causes and consequences of mid-Proterozoic anoxia. *Geophys Res Lett* 42:8538–46.

Deutsch C, Ferrel A, Seibel B, Pörtner H-O, Huey RB. 2015. Climate change tightens a metabolic constraint on marine habitats. *Science* 348:1132–5.

Dohrmann M, Wörheide G. 2017. Dating early animal evolution using phylogenomic data. *Sci Rep* 7:3599.

dos Reis M, Thawornwattana Y, Angelis K, Telford MJ, Donoghue PCJ, Yang Z. 2015. Uncertainty in the timing of origin of animals and the limits of precision in molecular timescales. *Curr Biol* 25:2939–50.

Droser ML, Tarhan LG, Gehling JG. 2017. The rise of animals in a changing environment: global ecological innovation in the late Ediacaran. *Annu Rev Earth Planet Sci* 45:593–617.

Erwin D, Valentine J. 2013. *The Cambrian Explosion: the construction of animal biodiversity*. Greenwood Village (CO): Roberts and Company Publishers.

Erwin DH, Laflamme M, Tweedt SM, Sperling EA, Pisani D, Peterson KJ. 2011. The Cambrian conundrum: early divergence and later ecological success in the early history of animals. *Science* 334:1091–7.

Erwin DH, Tweedt S. 2011. Ecological drivers of the Ediacaran–Cambrian diversification of the Metazoa. *Evol Ecol* 26:417–33.

Fernandez-Valverde S, Aguilera F, Ramos-Díaz R. 2018. Inference of developmental gene regulatory networks beyond classical model systems: new approaches in the post-genomic era. *Integr Comp Biol* published online (https://doi.org/10.1093/icb/icy061).

Fischer WW, Hemp J, Johnson JE. 2016. Evolution of oxygenic photosynthesis. *Annu Rev Earth Planet Sci* 44: 647–83.

French KL, Hallmann C, Hope JM, Schoon PL, Zumberge JA, Hoshino Y, Peters CA, George SC, Love GD, Brocks JJ, et al. 2015. Reappraisal of hydrocarbon biomarkers in Archean rocks. *Proc Natl Acad Sci U S A* 112:5915–20.

Gage JD, Tyler PA. 1991. Deep-sea biology. Cambridge: Cambridge University Press.

Glover AG, Smith CR, Paterson GLJ, Wilson GDF, Hawkins L, Sheader M. 2002. Polychaete species diversity in the central Pacific abyss: local and regional patterns and relationships with productivity. *Mar Ecol Prog Ser* 240: 157–70.

Gold DA. 2018. Life in changing fluids: a critical appraisal of swimming animals before the Cambrian. *Integr Comp Biol* published online (doi: 10.1093/icb/icy015).

Gold DA, Grabenstatter J, de Mendoza A, Riesgo A, Ruiz-Trillo I, Summons RE. 2016. Sterol and genomic analyses validate the sponge biomarker hypothesis. *Proc Natl Acad Sci U S A* 113:2684–9.

Gooday AJ, Levin LA, da Silva A, Bett BJ, Cowie G, Dissard D, Gage JD, Hughes DJ, Jeffreys R, Lamont PA, et al. 2009. Faunal responses to oxygen gradients on the Pakistan margin: a comparison of foraminiferans, macrofauna and megafauna. *Deep Sea Res Part 2 Top Stud Oceanogr* 56:488–502.

Gould SJ. 1990. Wonderful life: the Burgess Shale and the nature of history. New York (NY): W. W. Norton & Company.

Graur D, Martin W. 2004. Reading the entrails of chickens: molecular timescales of evolution and the illusion of precision. *Trends Genet* 20:80–6.

Hammarlund EU, Gaines RR, Prokopenko MG, Qi C, Hou X-G, Canfield DE. 2017. Early Cambrian oxygen minimum zone-like conditions at Chengjiang. *Earth Planet Sci Lett* 475:160–8.

Hammarlund EU, Stedingk K, von Pählsman S. 2018. Refined control of cell stemness allowed animal evolution in the oxic realm. *Nat Ecol Evol* 2:220–8.

Hardisty DS, Lu Z, Bekker A, Diamond C, Gill B, Jiang G, Kah L, Knoll A, Loyd S, Osburn MR, et al. 2017. Perspectives on Proterozoic surface ocean redox from iodine contents in ancient and recent carbonate. *Earth Planet Sci Lett* 463:159–70.

Hettinger A, Sanford E, Hill TM, Hosfelt JD, Russell AD, Gaylord B. 2013. The influence of food supply on the response of Olympia oyster larvae to ocean acidification. *Biogeosciences* 10:6629–38.

Hoffman PF, Abbot DS, Ashkenazy Y, Benn DI, Brocks JJ, Cohen PA, Cox GM, Creveling JR, Donnadieu Y, Erwin DH, et al. 2017. Snowball Earth climate dynamics and Cryogenian geology–geobiology. *Sci Adv* 3:e1600983.

Hoyal-Cuthill JFH, Conway Morris S. 2017. Nutrient-dependent growth underpinned the Ediacaran transition to large body size. *Nat Ecol Evol* 1:1201–4.

Huldtgren T, Cunningham JA, Yin C, Stampanoni M, Marone F, Donoghue PCJ, Bengtson S. 2011. Fossilized nuclei and germination structures identify Ediacaran “animal embryos” as encysting protists. *Science* 334: 1696–9.

Husson JM, Peters SE. 2017. Atmospheric oxygenation driven by unsteady growth of the continental sedimentary reservoir. *Earth Planet Sci Lett* 460:68–75.

Johnson JE, Webb SM, Ma C, Fischer WW. 2016. Manganese mineralogy and diagenesis in the sedimentary rock record. *Geochim Cosmochim Acta* 173:210–31.

Johnston DT, Macdonald FA, Gill BC, Hoffman PF, Schrag DP. 2012a. Uncovering the Neoproterozoic carbon cycle. *Nature* 483:320–3.

Johnston DT, Poulton SW, Goldberg T, Sergeev VN, Podkorytov V, Vorob'eva NG, Bekker A, Knoll AH. 2012b. Late Ediacaran redox stability and metazoan evolution. *Earth Planet Sci Lett* 335–336:25–35.

Kelly AE, Love GD, Zumberge JE, Summons RE. 2011. Hydrocarbon biomarkers of Neoproterozoic to Lower Cambrian oils from eastern Siberia. *Org Geochem* 42:640–54.

Kodner RB, Summons RE, Pearson A, King N, Knoll AH. 2008. Sterols in a unicellular relative of the metazoans. *Proc Natl Acad Sci U S A* 105:9897–902.

Laakso TA, Schrag DP. 2014. Regulation of atmospheric oxygen during the Proterozoic. *Earth Planet Sci Lett* 388:81–91.

Laakso TA, Schrag DP. 2017. A theory of atmospheric oxygen. *Geobiology* 15:366–84.

LaBarbera M. 1990. Principles of design of fluid transport systems in zoology. *Science* 249:992–1000.

Laflamme M, Darroch SA, Tweedt SM, Peterson KJ, Erwin DH. 2013. The end of the Ediacara biota: extinction, biotic replacement, or Cheshire Cat? *Gondwana Res* 23:558–73.

Laumer CE. 2018. Inferring ancient relationships with genomic data: a review of best practices. *Integr Comp Biol* published online (https://doi.org/10.1093/icb/icy075).

Lee MSY, Soubrier J, Edgecombe GD. 2013. Rates of phenotypic and genomic evolution during the Cambrian explosion. *Curr Biol* 23:1889–95.

Lenton TM, Boyle RA, Poulton SW, Shields-Zhou GA, Butterfield NJ. 2014. Co-evolution of eukaryotes and ocean oxygenation in the Neoproterozoic era. *Nat Geosci* 7:257–65.

Levin L, Gutierrez D, Rathburn A, Neira C, Sellanes J, Munoz P, Gallardo V, Salamanca M. 2002. Benthic processes on the Peru margin: a transect across the oxygen minimum zone during the 1997–98 El Niño. *Prog Oceanogr* 53:1–27.

Levin LA. 2003. Oxygen minimum zone benthos: adaptation and community response to hypoxia. *Oceanogr Mar Biol Annu Rev* 41:1–45.

Levin LA, Etter RJ, Rex MA, Gooday AJ, Smith CR, Pineda J, Stuart CT, Hessler RR, Pawson D. 2001. Environmental influences on regional deep-sea species diversity. *Annu Rev Ecol Syst* 32:51–93.

Levin LA, Sibuet M. 2012. Understanding continental margin biodiversity: a new imperative. *Annu Rev Mar Sci* 4:79–112.

Leys SP, Kahn A. 2018. Oxygen and the energetic requirements of the first multicellular animals. *Integr Comp Biol* published online (doi: 10.1093/icb/icy051).

Liu AG, Matthews JJ, Menon LR, McIlroy D, Brasier MD. 2014. *Haootia quadriformis* n. gen., n. sp., interpreted as a muscular cnidarian impression from the Late Ediacaran period (approx. 560 Ma). *Proc R Soc B Biol Sci* 281:20141202.

Liu AG, McIlroy D, Brasier MD. 2010. First evidence for locomotion in the Ediacara biota from the 565 Ma Mistaken Point formation, Newfoundland. *Geology* 38:123–6.

Liu X-M, Kah LC, Knoll AH, Cui H, Kaufman AJ, Shahar A, Hazen RM. 2016. Tracing Earth's O₂ evolution using Zn/Fe ratios in marine carbonates. *Geochem Perspect Lett* 2:24–34.

Love GD, Grosjean E, Stalvies C, Fike DA, Grotzinger JP, Bradley AS, Kelly AE, Bhatia M, Meredith W, Snape CE, et al. 2009. Fossil steroids record the appearance of Demospongiae during the Cryogenian period. *Nature* 457:718–21.

Love GD, Summons RE. 2015. The molecular record of Cryogenian sponges—a response to Antcliffe (2013). *Palaeontology* 58:1131–6.

Lozano-Fernandez J, dos Reis M, Donoghue PCJ, Pisani D. 2017. RelTime rates collapse to a strict clock when estimating the timeline of animal diversification. *Genome Biol Evol* 9:1320–8.

Lu W, Wörndl S, Halverson GP, Zhou X, Bekker A, Rainbird RH, Hardisty DS, Lyons TW, Lu Z. 2017. Iodine proxy evidence for increased ocean oxygenation during the Bitter Springs Anomaly. *Geochem Perspect Lett* 5:53–7.

Macdonald FA, Schmitz MD, Crowley JL, Roots CF, Jones DS, Maloof AC, Strauss JV, Cohen PA, Johnston DT, Schrag DP. 2010. Calibrating the Cryogenian. *Science* 327:1241–3.

Maclean N. 1992. *Young men and fire*. Chicago (IL): University of Chicago Press.

Maloof AC, Rose CV, Beach R, Samuels BM, Calmet CC, Erwin DH, Poirier GR, Yao N, Simons FJ. 2010. Possible animal-body fossils in pre-Marinoan limestones from South Australia. *Nat Geosci* 3:653–9.

Marshall CR. 2006. Explaining the Cambrian “Explosion” of animals. *Annu Rev Earth Planet Sci* 34:355–84.

McCaffrey MA, Moldowan MJ, Lipton PA, Summons RE, Peters KE, Jeganathan A, Watt DS. 1994. Paleoenvironmental implications of novel C₃₀ steranes in Precambrian to Cenozoic age petroleum and bitumen. *Geochim Cosmochim Acta* 58:529–32.

McClain CR, Allen AP, Tittensor DP, Rex MA. 2012. Energetics of life on the deep seafloor. *Proc Natl Acad Sci U S A* 109:15366–71.

Melzner F, Stange P, Trübenbach K, Thomsen J, Casties I, Panknin U, Gorb SN, Gutowska MA. 2011. Food supply and seawater pCO₂ impact calcification and internal shell dissolution in the blue mussel *Mytilus edulis*. *PLoS One* 6:e24223.

Mills DB, Canfield DE. 2014. Oxygen and animal evolution: did a rise of atmospheric oxygen “trigger” the origin of animals?. *BioEssays* 36:1145–55.

Mitchell RN, Raub TD, Silva SC, Kirschvink JL. 2015. Was the Cambrian explosion both an effect and an artifact of true polar wander? *Am J Sci* 315:945–57.

Muscente AD, Michel FM, Dale JG, Xiao S. 2015. Assessing the veracity of Precambrian ‘sponge’ fossils using in situ nanoscale analytical techniques. *Precambrian Res* 263:142–56.

Narbonne GM, Xiao S, Shields GA, Gehling JG. 2012. The Ediacaran period. In: Gradstein FM, Ogg JG, Schmitz M, Ogg G, editors. *The geologic time scale*. Boston: Elsevier. p. 413–35.

Noble SR, Condon DJ, Carney JN, Wilby PR, Pharaoh TC, Ford TD. 2015. U-Pb geochronology and global context of the Charnian Supergroup, UK: constraints on the age of key Ediacaran fossil assemblages. *Geol Soc Am Bull* 127:250–65.

Nursall JR. 1959. Oxygen as a prerequisite to the origin of the Metazoa. *Nature* 183:1170–2.

Paps J. 2018. What makes an animal? The molecular quest for the origin of the Animal Kingdom. *Integr Comp Biol* published online (doi: 10.1093/icb/icy036).

Paps J, Holland PWH. 2018. Reconstruction of the ancestral metazoan genome reveals an increase in genomic novelty. *Nat Commun* 9:1730.

Parfrey LW, Lahr DJG, Knoll AH, Katz LA. 2011. Estimating the timing of early eukaryotic diversification with multi-gene molecular clocks. *Proc Natl Acad Sci U S A* 108:13624–9.

Parry LA, Boggiani PC, Condon DJ, Garwood RJ, Leme JM, McIlroy D, Brasier MD, Trindade R, Campanha GAC, Pacheco MLAF, et al. 2017. Ichnological evidence for meiofaunal bilaterians from the terminal Ediacaran and earliest Cambrian of Brazil. *Nat Ecol Evol* 1:1455–64.

Partin CA, Bekker A, Planavsky NJ, Scott CT, Gill BC, Li C, Podkovyrov V, Maslov A, Konhauser KO, Lalonde SV, et al. 2013. Large-scale fluctuations in Precambrian atmospheric and oceanic oxygen levels from the record of U in shales. *Earth Planet Sci Lett* 369–370:284–93.

Paulmier A, Ruiz-Pino D, Garçon V. 2011. CO₂ maximum in the oxygen minimum zone (OMZ). *Biogeosciences* 8:239–52.

Payne JL, McClain CR, Boyer AG, Brown JH, Finnegan S, Kowalewski M, Krause JRA, Lyons SK, McShea DW, Novack-Gottshall PM, et al. 2011. The evolutionary consequences of oxygenic photosynthesis: a body size perspective. *Photosynth Res* 107:37–57.

Peters SE, Gaines RR. 2012. Formation of the “Great Unconformity” as a trigger for the Cambrian explosion. *Nature* 484:363–6.

Peterson KJ, Cotton JA, Gehling JG, Pisani D. 2008. The Ediacaran emergence of bilaterians: congruence between the genetic and geological fossil records. *Philos Trans R Soc Lond Ser B* 363:1435–43.

Peterson KJ, Lyons JB, Nowak KS, Takacs CM, Wargo MJ, McPeek MA. 2004. Estimating metazoan divergence times with a molecular clock. *Proc Natl Acad Sci U S A* 101:6536–41.

Peterson KJ, McPeek MA, Evans DAD. 2005. Tempo and mode of early animal evolution: inferences from rocks, Hox, and molecular clocks. *Paleobiology* 31:36–55.

Petsch ST, Berner RA, Eglinton TI. 2000. A field study of the chemical weathering of ancient sedimentary organic matter. *Org Geochem* 31:475–87.

Poinar GO. 2011. The evolutionary history of nematodes: as revealed in stone, amber and mummies. In: Hunt D, Perry RN, editors. *Nematology Monographs and Perspectives*. Vol. 9. Leiden, The Netherlands: Koninklijke Brill NV.

Pörtner H-O. 2010. Oxygen- and capacity-limitation of thermal tolerance: a matrix for integrating climate-related

stressor effects in marine ecosystems. *J Exp Biol* 213:881–93.

Prave AR, Condon DJ, Hoffmann KH, Tapster S, Fallick AE. 2016. Duration and nature of the end-Cryogenian (Marinoan) glaciation. *Geology* 44:631–4.

Pu JP, Bowring SA, Ramezani J, Myrow P, Raub TD, Landing E, Mills A, Hodgin E, Macdonald FA. 2016. Dodging snowballs: geochronology of the Gaskiers glaciation and the first appearance of the Ediacaran biota. *Geology* 44:955–8.

Raff RA, Raff EC. 1970. Respiratory mechanisms and the metazoan fossil record. *Nature* 228:1003–5.

Raiswell R, Berner RA. 1987. Organic carbon losses during burial and thermal maturation of normal marine shales. *Geology* 15:853–6.

Reinhard CT, Planavsky NJ, Gill BC, Ozaki K, Robbins LJ, Lyons TW, Fischer WW, Wang C, Cole DB, Konhauser KO. 2017. Evolution of the global phosphorus cycle. *Nature* 541:386–9.

Reinhard CT, Planavsky NJ, Olson SL, Lyons TW, Erwin DH. 2016. Earth's oxygen cycle and the evolution of animal life. *Proc Natl Acad Sci U S A* 113:8933–8.

Reinhard CT, Planavsky NJ, Robbins LJ, Partin CA, Gill BC, Lalonde SV, Bekker A, Konhauser KO, Lyons TW. 2013. Proterozoic ocean redox and biogeochemical stasis. *Proc Natl Acad Sci U S A* 110:5357–62.

Rhoads DC, Morse JW. 1971. Evolutionary and ecologic significance of oxygen-deficient marine basins. *Lethaia* 4:413–28.

Rooney AD, Strauss JV, Brandon AD, Macdonald FA. 2015. A Cryogenian chronology: two long-lasting synchronous Neoproterozoic glaciations. *Geology* 43:459–62.

Runnegar B. 1982a. Oxygen requirements, biology and phylogenetic significance of the late Precambrian worm Dickinsonia, and the evolution of the burrowing habit. *Alcheringa* 6:223–39.

Runnegar B. 1982b. The Cambrian explosion: animals or fossils? *J Geol Assoc Aust* 29:395–411.

Sanders MB, Bean TP, Hutchinson TH, Le Quesne WJF. 2013. Juvenile King Scallop, *Pecten maximus*, is potentially tolerant to low levels of ocean acidification when food is unrestricted. *PLoS One* 8:e74118.

Schiffbauer J, Huntley JR, O'Neil G, Darroch S, Laflamme M, Cai Y. 2016. The latest Ediacaran Wormworld Fauna: setting the ecological stage for the Cambrian Explosion. *GSA Today* 26:4–11.

Schrag DP, Higgins JA, Macdonald FA, Johnston DT. 2013. Authigenic carbonate and the history of the global carbon cycle. *Science* 339:540–3.

Sheppard KA, Rival DE, Caron J-B. 2018. On the hydrodynamics of *Anomalocaris* tail fins. *Integr Comp Biol* published online (doi: 10.1093/icb/icy014).

Shields-Zhou G, Och L. 2011. The case for a Neoproterozoic oxygenation event: geochemical evidence and biological consequences. *GSA Today* 21:4–11.

Shields-Zhou GA, Porter S, Halverson GP. 2016. A new rock-based definition for the Cryogenian Period (circa 720–635 Ma). *Episodes* 39:3–8.

Simion P, Philippe H, Baurain D, Jager M, Richter DJ, Franco AD, Roure B, Satoh N, Quéinnec É, Ereskovsky A, et al. 2017. A large and consistent phylogenomic dataset supports sponges as the sister group to all other animals. *Curr Biol* 27:958–67.

Smith CR, De Leo FC, Bernardino AF, Sweetman AK, Arbizu PM. 2008. Abyssal food limitation, ecosystem structure and climate change. *Trends Ecol Evol* 23:518–28.

Smith CR, Rabouille C. 2002. What controls the mixed-layer depth in deep-sea sediments? The importance of POC flux. *Limnol Oceanogr* 47:418–26.

Smith EF, Nelson LL, Strange MA, Eyster AE, Rowland SM, Schrag DP, Macdonald FA. 2016. The end of the Ediacaran: two new exceptionally preserved body fossil assemblages from Mount Dunfee, Nevada, USA. *Geology* 44:911–4.

Sperling EA. 2013. Tackling the 99%: can we begin to understand the paleoecology of the small and soft-bodied animal majority? In: Bush AM, Pruss SB, Payne JL, editors. *Ecosystem paleobiology and geobiology*. New Haven (CT): Yale University Printing and Publishing Services. p. 77–86.

Sperling EA, Frieder CA, Levin LA. 2016. Biodiversity response to natural gradients of multiple stressors on continental margins. *Proc R Soc B* 283:20160637.

Sperling EA, Frieder CA, Raman AV, Girguis PR, Levin LA, Knoll AH. 2013a. Oxygen, ecology, and the Cambrian radiation of animals. *Proc Natl Acad Sci U S A* 110:13446–51.

Sperling EA, Halverson GP, Knoll AH, Macdonald FA, Johnston DT. 2013b. A basin redox transect at the dawn of animal life. *Earth Planet Sci Lett* 371–372:143–55.

Sperling EA, Knoll AH, Girguis PR. 2015a. The ecological physiology of Earth's second oxygen revolution. *Annu Rev Ecol Evol Syst* 46:215–35.

Sperling EA, Peterson KJ, Laflamme M. 2011. Rangeomorphs, Thectardis (Porifera?) and dissolved organic carbon in the Ediacaran oceans. *Geobiology* 9:24–33.

Sperling EA, Pisani D, Peterson KJ. 2007. Poriferan paraphyly and its implications for Precambrian palaeobiology. In: Vickers-Rich P, Komarower P, editors. *The rise and fall of the Ediacaran Biota*. London: Geological Society, London, Special Publications. p. 355–68.

Sperling EA, Wolock CJ, Morgan AS, Gill BC, Kunzmann M, Halverson GP, Macdonald FA, Knoll AH, Johnston DT. 2015b. Statistical analysis of iron geochemical data suggests limited late Proterozoic oxygenation. *Nature* 523:451–4.

Stolper DA, Keller CB. 2018. A record of deep-ocean dissolved O₂ from the oxidation state of iron in submarine basalts. *Nature* 553:323–7.

Tarhan LG, Droser ML, Cole DB, Gehling JG. 2018. Ecological and environmental diversification in the Late Ediacaran. *Integr Comp Biol* published online (https://doi.org/10.1093/icb/icy020).

Teal L, Bulling M, Parker E, Solan M. 2008. Global patterns of bioturbation intensity and mixed depth of marine soft sediments. *Aquat Biol* 2:207–18.

Tittensor DP, Mora C, Jetz W, Lotze HK, Ricard D, Berghe EV, Worm B. 2010. Global patterns and predictors of marine biodiversity across taxa. *Nature* 466:1098–101.

Tostevin R, Wood RA, Shields GA, Poulton SW, Guilbaud R, Bowyer F, Penny AM, He T, Curtis A, Hoffmann KH, et al. 2016. Low-oxygen waters limited habitable space for early animals. *Nat Commun* 7:12818.

Verberk WCEP, Bilton DT, Piero C, Spicer JI. 2011. Oxygen supply in aquatic ectotherms: partial pressure and solubility together explain biodiversity and size patterns. *Ecology* 92:1565–72.

Vinther J, Stein M, Longrich NR, Harper DAT. 2014. A suspension-feeding anomalocarid from the Early Cambrian. *Nature* 507:496–9.

Wallace MW, Hood A, Shuster A, Greig A, Planavsky NJ, Reed C. 2017. Oxygenation history of the Neoproterozoic to early Phanerozoic and the rise of land plants. *Earth Planet Sci Lett* 466:12–9.

Wei C-L, Rowe GT, Escobar-Briones E, Boetius A, Soltwedel T, Caley MJ, Soliman Y, Huettmann F, Qu F, Yu Z, et al. 2010. Global patterns and predictions of seafloor biomass using random forests. *PLoS One* 5:e15323.

Whelan NV, Kocot KM, Moroz LL, Halanych KM. 2015. Error, signal, and the placement of Ctenophora sister to all other animals. *Proc Natl Acad Sci U S A* 112:5773–8.

Wood R, Erwin DH. 2018. Innovation not recovery: dynamic redox promotes metazoan radiations. *Biol Rev* 93:863–73.

Woolley SNC, Tittensor DP, Dunstan PK, Guillera-Arroita G, Lahoz-Monfort JJ, Wintle BA, Worm B, O'Hara TD. 2016. Deep-sea diversity patterns are shaped by energy availability. *Nature* 533:393–6.

Xiao S. 2014. Oxygen and early animal evolution. In: Holland HD, Turekian KK, editors. *Treatise on geochemistry*. Oxford, United Kingdom: Elsevier. p. 231–50.

Xiao S, Zhang Y, Knoll AH. 1998. Three-dimensional preservation of algae and animal embryos in a Neoproterozoic phosphorite. *Nature* 391:553–8.

Yin Z, Zhu M, Davidson EH, Bottjer DJ, Zhao F, Tafforeau P. 2015. Sponge grade body fossil with cellular resolution dating 60 Myr before the Cambrian. *Proc Natl Acad Sci U S A* 112:E1453–60.

Zhang X, Shu D, Han J, Zhang Z, Liu J, Fu D. 2014. Triggers for the Cambrian explosion: hypotheses and problems. *Gondwana Res* 25:896–909.

Zumberge JA, Love GD, Sperling E, Cardenas P. 2017. Novel sterane biomarkers sourced from sponges supports the evidence for Neoproterozoic animals. 28th International Meeting on Organic Geochemistry, Abstracts with Program (<http://imog2017.org/wp-content/uploads/2017/04/084.pdf>).