

Oxygen Requirements for the Cambrian Explosion

Xingliang Zhang*, Linhao Cui

Early Life Institute and State Key Laboratory of Continental Dynamics, Department of Geology, Northwest University, Xi'an 710069, China

ABSTRACT: Hypoxic tolerance experiments may be helpful to constrain the oxygen requirement for animal evolution. Based on literature review, available data demonstrate that fishes are more sensitive to hypoxia than crustaceans and echinoderms, which in turn are more sensitive than annelids, whilst mollusks are the least sensitive. Mortalities occur where O₂ concentrations are below 2.0 mg/L, equivalent to saturation with oxygen content about 25% PAL (present atmospheric level). Therefore, the minimal oxygen requirement for maintaining animal diversity since Cambrian is determined as 25% PAL. The traditional view is that a rise in atmospheric oxygen concentrations led to the oxygenation of the ocean, thus triggering the evolution of animals. Geological and geochemical studies suggest a constant increase of the oxygen level and a contraction of anoxic oceans during Ediacaran–Cambrian transition when the world oceans experienced a rapid diversification of metazoan lineages. However, fossil first appearances of animal phyla are obviously asynchronous and episodic, showing a sequence as: basal metazoans>lophotrochozoans>ecdysozoans and deuterostomes. According to hitherto known data of fossil record and hypoxic sensitivity of animals, the appearance sequence of different animals is broadly consistent with their hypoxic sensitivity: animals like molluscs and annelids that are less sensitive to hypoxia appeared earlier, while animals like echinoderms and fishes that are more sensitive to hypoxia came later. Therefore, it is very likely that the appearance order of animals is corresponding to the increasing oxygen level and/or the contraction of anoxic oceans during Ediacaran–Cambrian transition.

KEY WORDS: oxygenation, Cambrian explosion, metazoan lineages, divergence, fossil first appearance.

0 INTRODUCTION

Abrupt appearances of diverse animal lineages at the beginning of the Cambrian Period, known as the Cambrian explosion (Cloud, 1948), has been puzzling the scientific world for around two centuries. Conscientious and concerted efforts by multidiscipline scientists in recent decades have dramatically increased our knowledge on this unprecedented, unique evolutionary event. On one hand, the Cambrian explosion itself implies that the actual origination and divergence of metazoan phyla are no later than the Cambrian explosion (Zhang and Shu, 2014). On the other hand, molecular clock studies suggest that the origination and diversification of metazoan lineages predate their fossil first appearances, long before the Cambrian explosion (Erwin et al., 2011; Blair, 2009; see Zhang and Shu, 2014 for a summary). Therefore, it is likely that metazoans, at least stem lineages, originated and diverged much earlier than their initial ecological success during the Cambrian explosion, i.e. early divergence and later ecological success in the early history of animals (Erwin and Valentine, 2013; Erwin et al., 2011). However, such earlier divergence

view is challenged by the paucity of unequivocal metazoan fossils before and during the Ediacaran period (Zhang and Shu, 2014; Zhang et al., 2014). No matter metazoan lineages diverged earlier than or during the Cambrian explosion, the metazoan-dominated marine ecosystem was established for the first time at the beginning of the Cambrian Period (Zhang and Shu, 2014; Erwin and Tweedt, 2012). The initial ecological success may have been triggered by environmental perturbation near the Ediacaran-Cambrian boundary and subsequently amplified by ecological interactions within reorganized ecosystems (Wang H Y et al., 2015; Zhang et al., 2014; Sperling et al, 2013a; Knoll and Carroll, 1999). In a word, the Cambrian explosion happened when environmental changes crossed critical thresholds, led to the initial formation of the metazoan-dominated ecosystem through a series of knock-on ecological processes, i.e. “ecological snowball” effects (Zhang and Shu, 2014).

Extant metazoans (multicellular animals) are grouped into 38 phylum-level clades, paraphyletic basal metazoans having six phyla and the remainder 32 phyla comprising the Bilateria that are further classified into three monophyletic supraphyletic clades, i.e. the Lophotrochozoa, the Ecdysozoa, and the Deuterostomia (Nielson, 2012). Thus far, 20 of the living animal phyla have been known to make their first appearances during the Cambrian explosion in an asynchronous manner (Zhang and Shu, 2014; Shu et al., 2014). This manner would raise questions such as: Why some phyla appeared earlier than

*Corresponding author: xzhang69@nwu.edu.cn

© China University of Geosciences and Springer-Verlag Berlin Heidelberg 2016

Manuscript received March 05, 2015.

Manuscript accepted September 12, 2015.

others in fossil record? Is the fossil appearance order of different animal phyla consistent with the order of their originations? What controls the appearance sequence of animal phyla? This paper provides a possible explanation for the questions by discussing three independent lines of evidence: (1) constraining the minimal oxygen requirement for metazoans on the basis of hypoxic sensitivity of different animals; (2) checking the sequence of fossil first appearances of animal phyla and determining whether the appearance order matches the hypoxic sensitivity of animals; (3) constraining the oxygen levels during this Ediacaran–Cambrian transition and determining whether oxygen is sufficient enough for animal diversification during the critical interval.

1 OXYGENATION AND LIFE EVOLUTION

It is generally accepted that fluctuations of atmospheric oxygen levels play a significant role in shaping the evolution of life (Cloud, 1976). For example, the great oxidation event at Early Proterozoic is essentially important for the evolution of early eukaryotes, the later oxidation event at the late Neoproterozoic allowed the metazoan evolution possible (Lyons et al., 2012). The extremely high oxygen level during Late Paleozoic, above 30% of oxygen content in atmosphere, led to gigantism during this geological period. Sea scorpions at the time reached 2.5 m in body length, dragonflies reached 70 cm wingspan, mayflies reached 45 cm wingspan, and millipedes reached 2 m in length (Braddy et al., 2008; Petsch, 2004).

Advantages of oxygen for eukaryote evolution appear in many aspects. The formation of the ozone shelter, protecting eukaryotic organisms from UV radiation, requires the atmospheric oxygen level above 1% PAL (see Kasting, 1993 and references therein). Based on eukaryotic physiology the minimal requirement was determined approximately at this level (Runnegar, 1991). The rise of oxygen level also allows aerobic respiration, multicellularity, and large body size. The aerobic metabolism is many-fold more efficient in extracting energy from foodstuffs.

However, oxygen molecule is not always beneficial to organisms. First, oxygen is toxic to anaerobic biological world. Many types of prokaryotic microbes like sulfate reducers and methanogens live strictly in anoxic conditions. Second, accumulation of oxygen may lead to the formation of Reactive Oxygen Species (ROS), e.g. superoxide, hydrogen peroxide. They are dangerous to all organisms. To cope with ROS, organisms have evolved a number of mechanisms which they use for defense of ROS, e.g. scavenger molecules, detoxicating enzymes, antioxidants, cell aggregation for defense, oxygen transport proteins, and so on. Among these the cell aggregation is particularly interesting in understanding the origin of multicellularity. In cell aggregations, surface cells protect interior cells from reactive oxygen species, which means cell differentiation. Therefore it is very likely that protection against ROS is an important evolutionary driving force to multicellularity (Decker and van Holde, 2011).

2 OXYGEN REQUIREMENTS FOR METAZOAN LINEAGES

Oxygen has long been proposed as a prerequisite to early animal evolution. The minimal oxygen requirement has been estimated based on different theoretical models but the results are quite variable (Sperling et al., 2013a, b; Catling et al., 2005; Runnegar, 1982; Cloud, 1976; Rhoads and Morse, 1971; Berkner and Marshall, 1965) (Table 1). For example, the last common ancestor of bilaterians, supposed to be sub-millimeter in size, has a very low oxygen requirement ranging from 0.14% to 0.36% PAL (Sperling et al., 2013b), while the oxygen requirement for centimeter-sized metazoans with circulatory physiology, is loosely constrained between 10%–100% PAL (Catling et al., 2005). These estimates suggest that early animals, in general, may have had relatively low oxygen requirements. However, these theoretical efforts are based on highly simplified, very small organisms that do not reflect the complexity of living animals or Cambrian animals either. In living animals the minimal oxygen requirement varies from lineages to lineages, which can be determined by empirical and experimental evidence for hypoxic sensitivity of different animals.

Empirical evidence indicates that non-chemosymbiotic benthic macrofaunal (retained on 0.3 mm sieves) bilaterians can and do live in modern low-oxygen environments (Sperling et al., 2013b; Rhoads and Morse, 1971). Moreover, there is evidence that a few metazoans can live under permanently anoxic and sulfidic conditions. Three species of the animal phylum Loricifera have been known in the sediments of the deep anoxic, sulfidic and hypersaline L'Atalante basin (Mediterranean Sea) (Danovaro et al., 2010). It is very interesting to find that anaerobic biochemistry and anaerobic mitochondria are widespread across metazoan lineages and all eukaryotic groups, which suggest that metazoan ancestors may have evolved in a more anoxic past (Mental and Martin, 2010).

A recent experimental study shows that demosponges, serving as analogs for early animals, are sensitive to anoxia and stagnant seawater, but can survive, and even thrive, under oxygen levels as low as 0.5%–4% of present-day levels (Mills et al., 2014). The experiments suggest that some basal metazoans can live at very low oxygen levels, a condition likely to have characterized surface oceans long before the Cambrian explosion. However, it might be too soon to conclude that the origin and earliest evolution of animal life on Earth was not triggered by a rise of atmospheric and marine oxygen in the Neoproterozoic Era (Mills et al., 2014). Just as one swallow does not make a summer, a sponge species neither represents the entire sponge clade nor does the diverse metazoans. Equally important, at the minimum oxygen levels capable of supporting sponges, most familiar animals would die, so there must be more to the story (Knoll and Sperling, 2014). Experimental studies on hypoxic and/or anoxic tolerance of bilateral lineages are more helpful to determine the minimum oxygen requirements for maintaining the metazoan biodiversity since the Cambrian explosion, although data are limited to some lineages. Effects of reduced oxygen concentrations on marine organisms were previously reviewed (Gray et al., 2002; Diaz and Rosenberg, 1995) but not discussed in the context of

animal evolution.

Crabs, lobsters, and shrimps, that are all crustacean arthropods, show hypoxic effects on feeding at oxygen concentrations between 2 and 3.5 mg/L (see Gray et al., 2002 and references therein). For the marine amphipod *Melitta longidactyla*, oxygen concentrations below 3.5 mg/L cause a decrease in growth rates. When oxygen concentrations below 1 mg/L, abundances are reduced and mortalities occur in the sediment-living isopod *Saduria entomon*.

Growth effect experiments show that when oxygen concentration is below 2.7 mg/L, the growth of ophiuroid echinoderm *Amphiura filiformis* is reduced; when below 2.1 mg/L, the growth of polychaetes *Capitella capitata* is reduced; and when below 1.5 mg/L, the growth of bivalve molluscs *Crassostrea virginica* and *Mytilus edulis* is affected (Rosenberg, 1972). It appears that echinoderms are more sensitive than polychaetes and mollusks in growth effects.

Marine vertebrates like fishes are more sensitive to hypoxia (see Gray et al., 2002 and references therein). When oxygen concentration below 4.5 mg/L it will impair production, larval growth and food intake of early-life non-salmonid species, and therefore the growth will be reduced. The actively swimming cod *Gadus morhua* shows reductions in growth at 6.5 mg O₂/L, a high oxygen concentration, whereas bottom-living fishes show metabolic effects at concentrations of 4 mg O₂/L, and mudskippers at around 1 mg O₂/L. Thus the hypoxic tolerance level of a species is also related with its behavioral habits. Oxygen concentration below 2 mg/L would cause mortalities of most fishes.

Anoxic tolerance experiments at 17 psu, 10 °C and <0.1 mg O₂/L show that a number of bivalve mollusc species (*Astarte borealis*, *A. elliptica*, *A. monatguyi*, *Arctica islandica* and *Mytilus edulis*) survived more than 1 000 h, whereas the polychaetes (*Nephtys ciliata* and *Nereis diversicolor*) survived for ca. 200 h and the crustaceans (*Carcinus maenas*, *Gammarus oceanicus*, *Idotea balthica* and *Crangon crangon*) survived for less than 100 h (Dries and Theede, 1974). Similar results have also been reported: the ranking of anoxic tolerance from greatest to least as: bivalves (*Macoma balthica* > *Cardium lamarkii*) > polychaetes (*N. diversicolor* > *Scoloplos armiger* > *Ampharete grubei* > *Terebellides stroemi*) (Henriksson, 1969). It seems evident that molluscs are much more tolerant to anoxic conditions than polychaetes and arthropods.

Available data demonstrate that fishes are more sensitive to hypoxia than crustaceans and echinoderms, which in turn are more sensitive than annelids, whilst molluscs are the least sensitive (Gray et al., 2002). Larval fishes and crustaceans are more sensitive than the adults (Miller et al., 1995). In general, prolonged exposure to 4 mg O₂/L causes acute mortality in many invertebrates and non-salmonid fish embryos; while 3 mg O₂/L causes acute mortality in most non-salmonid fishes. Overall, growth is affected at oxygen concentrations below 6.0 mg/L and mortalities occur in all bilateral lineages where concentrations are below 2.0 to 0.5 mg O₂/L (Gray et al., 2002), equivalent to a saturation state with oxygen content about 25% PAL. Therefore, the minimal O₂ requirement for the animal diversity in the Cambrian is determined as 25% PAL.

3 FOSSIL APPEARANCE SEQUENCES OF METAZOAN LINEAGES

Molecular dating suggests a very early divergence of animals, down to the Cryogenian (e.g. Erwin et al., 2011). Theoretically two sister groups like protostomes and deuterostomes, branched from a single common ancestor and therefore originated at the same time. But in the fossil record first appearances of phyla are mostly concentrated in the first three stages of the Cambrian System, significantly later than the dates suggested by molecular dating. By the Cambrian Stage 3, 20 of 38 living phyla were present. Their appearances are obviously asynchronous and episodic, broadly can be recognized as three phases. The first phase involving basal metazoans and a few lophotrochozoans like molluscs and annelids, took place during the late Ediacaran. The second phase is the explosive appearances of lophotrochozoans and calcified basal metazoans during the Terreneuvian. The third phase, also the largest phase, is marked by abrupt appearances of ecdysozoans and deuterostomes during Cambrian Stage 3 (Shu et al., 2014; Zhang and Shu, 2014).

Particular attention will be paid to fossil first appearances of fishes, echinoderms, annelids, molluscs and arthropods because hypoxic sensitivity data are available for these five groups (Fig. 1). And the time scales are resolved to geological stages and in some cases to biozones.

The earliest putative mollusc is *Kimberella* from the Late Ediacaran White Sea biota, Russia, dated at 555 million years in age (Fedonkin and Waggoner, 1997). Molluscs with complete hard shell (not composed of sclerites), as well as shelly fossils showing mollusk affinities (e.g. hyoliths, halwaxiids and cancelloriids), first occurred in the mid-Fortunian at the transition between *Anabarites trisulcatus* and *Purella squamulosa* zones (Kouchinsky et al., 2012).

The tubular fossil *Cloudina* that have been reported from the latest Ediacaran of several paleo-continent, about 548 Ma in age, was morphologically compared with annelid tubes (Hua et al., 2005). However it was alternatively considered as cnidariomorphs (Kouchinsky et al., 2012). *Hyolithellus* and similar tubular fossils from the lower Cambrian were considered to be most likely secreted by annelid-grade animals (Skovsted and Peel, 2011), which first appeared at the base of the Cambrian within the *Anabarites trisulcatus*-*Protohertzina anabarica* Assemblage Zone (Yang et al., 2014). The earliest unequivocal annelid fossil appears to be *Phragmochaeta* from the Sirius Passet fauna (Conway Morris and Peel, 2008), correlated to the upper part of the Cambrian Stage 3.

Trace fossils of non-mineralized arthropods (e.g. *Rusophycus*) have been known since the Fortunian (Mángano and Buatois, 2014). The earliest known arthropods with biomineralized exoskeletons, i.e. trilobites, appeared at the base of Stage 3, succeeded by the first appearances of bradoriids (carapaces probably consisting of calcium phosphate) and many other lineages of lightly sclerotized arthropods in the middle and upper parts of Stage 3. The appearance of the earliest trilobites coincides with the onset of explosive diversification of the phylum Arthropoda, and more or less with the superphyla Ecdysozoa.

The earliest biomineralizing representatives of

echinoderms appeared in the middle part of Stage 3, within the third trilobite Zone of Siberia, the *Pagetiellus anabarus* Zone (Kouchinsky et al., 2015). Among them, the eocrinoids, members of the stem-group Echinodermata, can be recognized (Kouchinsky et al., 2015; Kouchinsky et al., 2012).

The earliest vertebrates represented by small, soft-bodied, jawless fishes have been found in the Chengjiang fauna (Shu et al., 1999), which is correlated to the upper part of the Cambrian Stage 3. The larger, armored jawless fishes appeared much later, up to the Middle Ordovician.

When we combine data of fossil record with hypoxic sensitivity of animals so far as we know, we find that the appearance sequence of different animals is broadly consistent with hypoxic sensitivity of animals. Animals like molluscs and annelids that are less sensitive to hypoxia appeared earlier, while those animals like echinoderms and fishes that are more sensitive to hypoxia came later. The case of sponges should be mentioned here! Direct experimental evidence indicates that sponges are able to tolerate lower oxygen levels (Mills et al., 2014), and thus they might have diverged much earlier than bilateral lineages as molecular and biomarker data suggested (Erwin et al., 2011; Love et al., 2009). However, according to the reanalysis of sponge fossils previously reported from the Precambrian, the earliest convincing fossil sponge remains appeared at around the time of the Precambrian-Cambrian boundary, associated with the great radiation events of that interval (Antcliffe et al., 2014). The microfossil *Eocyathispongia qiania* from the Weng'an biota (ca. 600 Ma) was interpreted as a sponge grade organism (Yin et al., 2015). However, it has not yet suffered from critical evaluation.

4 OXYGENATION DURING THE CAMBRIAN EXPLOSION

In the air saturated shallow water of modern marine

environments at temperature of 15 degree, the oxygen concentration is about 7.9 mg/L. Except for the OMZ (oxygen minimum zone), oxygen concentrations in both shallow sea and deep sea are well above 2 mg/L, sufficient for animal metabolism. In the geological past time, particularly during the Ediacaran-Cambrian transition, straightforward evidence for oxygen concentrations is not available in ancient sea or in the atmosphere. Persistence of animal diversity suggests oxygen level is above 25% PAL since the Cambrian.

Geological evidence suggested that oxygen levels were increasing during Ediacaran-Cambrian transition. The assembly of Gondwana was a polyphase process that was accomplished during the Pan-Africa Orogeny. The Brasilina-Damara Orogeny (630–520 Ma) and the Kuungan Orogeny (570–530 Ma) were nearly synchronous with the Cambrian explosion (Meert, 2011; Campbell and Squire, 2010; Rogers and Santosh, 2004; Meert, 2003; Li and Powell, 2001). Continental collisions at this time led to trans-Gondwana mountain ranges extending over several thousand kilometers. The uplift and erosion of the huge mountains would bring nutrients to the ocean regime, and thus increase the biomass and the burial rate of organic carbon that is believed to have triggered a critical rise in the availability of atmospheric oxygen (Campbell and Squire, 2010; Campbell and Allen, 2008; Knoll and Walter, 1992). The aftermath of Snowball Earth is similar to that of tectonics. Again it is nutrient and oxygenation that were linked to evolution. During the end of Snowball Earth, ice melting brought nutrient P from continents to the ocean regimes, and thus led to increase of biomass and organic burial, and further oxygenation of atmosphere (Papineau, 2010; Planavsky et al., 2010).

Overwhelming geochemical data suggested a stratified ocean throughout the most time of the Proterozoic Eon (Kendall et al., 2012; Lyons et al., 2012). The surface oceans

Table 1 Theoretical estimates on minimal oxygen requirements for metazoan evolution

Atmospheric oxygen levels	Requirements	Sources	Remarks
1% PAL	Formation of ozone shelter, protecting metazoans from UV radiation.	Berkner and Marshall, 1965	Lethal quantities of UV penetrate to 5 or 10 m depth in water. The ozone shelter permits widespread extension of life to the surface ocean. Such level may have been reached since the initial rise of atmospheric O ₂ .
10% PAL	A small, soft-bodied fauna	Rhoads and Morse, 1971	Based analogues to benthic invertebrates living in low oxygen regions, O ₂ concentration ≤ 0.1 ml/L corresponds to an azoic region, 0.3–1.0 ml/L to a low diversity, small, soft-bodied infauna, and ≥ 1.0 ml/L to a diverse calcareous fauna.
6.2% PAL	Primitive metazoans that rely on diffusion for their O ₂ supply	Cloud, 1976	Early metazoans were assumed to be thin (~1 mm thick) and soft-bodied but some had large surface areas. Acquisition of hard parts required a level above 10% PAL.
1%–3% PAL	Sheet-like organism <i>Dickinsonia</i>	Runnegar, 1982	Assumed that the sheet-like geometry was an adaptation to low oxygen levels.
10%–100% PAL	Millimeter- to centimeter-sized animals	Catling et al., 2005	An O ₂ level 10% PAL is needed to grow as large as approximately millimeter scale on the basis of diffusion and an O ₂ level 10%–100% PAL is a prerequisite to cross the threshold for organisms of size ~1 cm with closed, pulsatile circulatory systems.
0.14%–0.36% PAL	Ancestral bilaterians millimeter to sub-millimeter in size	Sperling et al., 2013b	The most likely minimal oxygen requirement for a 3 mm-long \times 67 μ m-wide worm with a circulatory system is ~0.14% PAL. The most likely minimal oxygen requirements for a 600 μ m long \times 25 μ m wide diameter worm limited by pure diffusion is ~0.36% PAL.

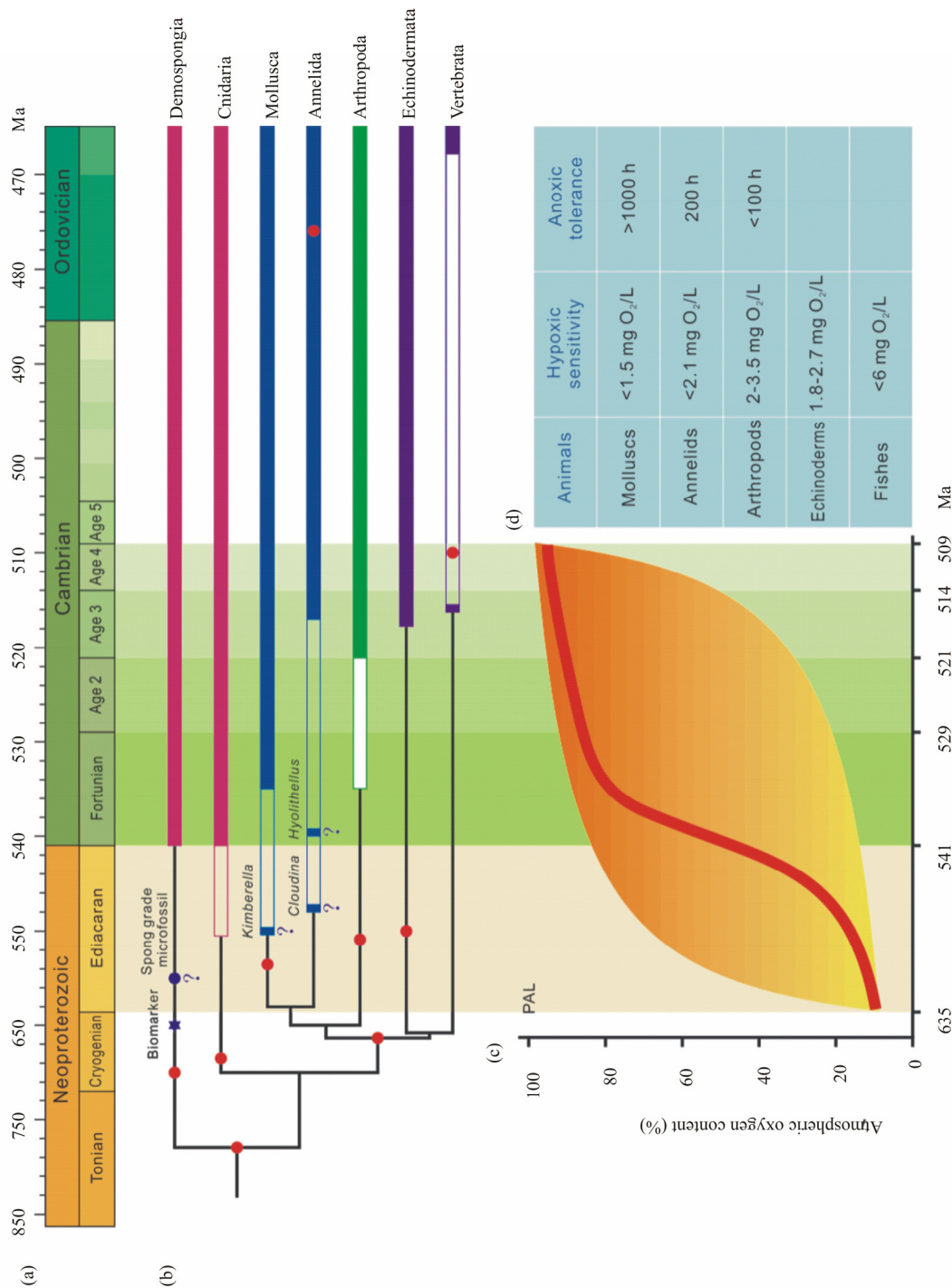


Figure 1. Temporal origins of animal lineages with hypoxic sensitivity data. (a) Geological time scale for Tonian–Ordovician; (b) fossil record of animal lineages (colored bars, white bars representing possible range extension) and their timing of originations (red dots) inferred from molecular clock studies (Erwin et al., 2011). In general fossil first appearance of a lineage tends to be younger than its evolutionary origin. The appearance order is decoupled from that of origination but broadly corresponds to the hypoxic sensitivity. The appearance sequence of different animals is broadly consistent with hypoxic sensitivity of animals, these animals like molluscs and annelids that are less sensitive to hypoxia appeared earlier, while those animals like echinoderms and fishes that are more sensitive to hypoxia came later; (c) range of atmospheric oxygen levels during the Cambrian explosion and possible increasing mode (the red curve) based on the Uranium record (Partin et al., 2013); (d) hypoxic sensitivity and anoxic tolerance of different animal lineages (see text for specific effects).

were mildly oxygenated after the initial rise of atmospheric O₂ at the beginning of the Proterozoic, but the deep oceans were anoxic (ferruginous or euxinic). When ferruginous condition prevailed in the open ocean, euxinic water bodies rimed the global ocean as “dynamic wedges” at mid-water depths (Li et al., 2010) or were limited to restricted, marginal marine basins. These possibilities of the marine redox structure likely existed at different times and also at the same time in different places of the ocean (Lyons et al., 2012). However, the timing of the deep ocean oxygenation is somewhat controversial. Geochemical data from the Avalonia suggest the deep ocean was anoxic and ferruginous before and during the Gaskiers glaciation some 580 million years ago and became oxic afterward (Canfield et al., 2007). More recent data are tend to suggest that the stratified ocean, at least locally, persisted to the Early Cambrian, in which euxinic water-bodies dynamically wedged at the mid-water depths (Feng et al., 2014; Jin et al., 2014; Wen et al., 2014; Wang et al., 2012).

It is evident that the surface oceans were oxygenated long before the Cambrian explosion. The problem is the lack of precise values of oxygen level before and during initial appearances of diverse metazoan fossil. Therefore, it is difficult to determine when oxygen concentrations in shallow water rose above the critical requirement and was capable of supporting the metazoan diversity. Under this predicament, the ocean redox models have been used to estimate the atmospheric oxygen levels. The oxygenation of deep sea requires atmospheric oxygen levels above 15% of PAL (Canfield et al., 2007), while the persistence of stratified oceans requires atmospheric oxygen level below 40% PAL (Kump, 2008; Canfield, 2005). If the deep sea was periodically anoxic during Late Ediacaran to the Early Cambrian, the oxygen level would be varying between 15% and 40% of PAL. However, such estimation based on uniformitarian principles was questioned by Butterfield (2009). He argued that Precambrian oceans (stratified, turbid anoxic water column dominated by cyanobacteria) were quite unlike the Phanerozoic counterparts (well-mixed, clear-water system dominated by eukaryotic algae), and thus deep-sea geochemical signatures were unable to be used as a direct proxy for atmospheric oxygen levels under such non uniformitarian conditions (Butterfield, 2009). On the other hand, the rise of atmospheric oxygen level can cause the immediate oxygenation of shallow seas. However, the evolution of deep ocean oxygenation and atmosphere oxygenation may have not been fully coupled. This again undermines the estimate of atmospheric oxygen level based on the stratified ocean redox model.

It is worth mentioning that cerium anomaly recognized in South China suggested a stepwise rise of oxygen levels in the shallow marine environments during the Ediacaran Period (Ling et al., 2013). This study also implied that shallow oceans remained anoxic or suboxic before 551 Ma, and became well oxygenated afterward. The record of U in shales also implied that the atmospheric oxygen content was constantly rising during the Ediacaran-Cambrian transition (Partin et al, 2013). Molybdenum isotope data from South China suggest the areal extent of oxygenated bottom waters increased in the Early Cambrian and modern-like oxygen levels characterized the

ocean at ~521Ma for the first time in Earth history (Chen et al., 2015). These results are fairly consistent with the fossil record that metazoans suddenly occurred in abundance in the Early Cambrian. Moreover, earliest benthic faunas almost exclusively colonized in shallow water environments and dispersed to deep water environments since Cambrian Stage 3. Intriguingly, iron speciation and sulfur isotopic data suggested the deep ocean was rapidly oxygenated since the Cambrian stage 3 (Wang et al., 2012), coinciding with the onset of the major episode of the Cambrian explosion. Whether the invasion of benthic faunas caused the oxygenation of deep water environments or vice versa is still a matter of debate.

6 DISCUSSION AND CONCLUSIONS

Direct experiments of hypoxic sensitivity have been known in a limited number of metazoan lineages, in which metabolisms of different animals are affected by different levels of oxygen content. In general, vertebrate fishes are more sensitive to hypoxia than crustaceans and echinoderms, which in turn are more sensitive than annelids, whilst molluscs are the least sensitive (Gray et al., 2002). Available data indicates most macroscopic metazoans could not survive when oxygen concentrations are below 2 mg/L. Therefore, the minimal oxygen requirement for maintaining metazoan diversity since the Cambrian explosion is constrained to this level, a concentration corresponding to the air saturated shallow water in atmospheric oxygen level about 25% PAL.

A robust phylogeny of the metazoa in combination with molecular dating results is useful to infer the timing and sequence of originations of different lineages. Theoretically two sister groups branched from a single common ancestor and therefore originated at the same time. However, there is currently a time lag between the phylogenetic diversification of metazoan lineages and their first appearances in fossil record (Wang Y et al., 2015; Wray, 2015; Zhang and Shu, 2014; Zhang et al., 2014). In general fossil first appearance of a species in a given place will tend to be younger than its evolutionary origin for a number of reasons, e.g. evolutionary dispersal, non-preservation, hiatus, inappropriate biofacies, and/or artifact of non-discovery (see Landing et al., 2013 for discussion), but the crux is its rarity. It is, therefore, very likely that later appearance of a lineage in fossil record is due to its insignificance in ecology, which was constrained by certain environmental threshold. Earlier ancestors might be ecologically insignificant for small size, soft body, and/or rarity, and have little chance to be found. They became ecologically abundant when environmental changes, most likely the oxygen content, crossed the critical demand. Therefore, abrupt appearances of metazoan lineages in abundance reflected their initial ecological success (occurring in abundance) during the Cambrian explosion. On the other hand, fossil first appearances of metazoan lineages are asynchronous in spite of concentrating in a relatively short time interval. The appearance order is decoupled from that of origination but broadly corresponds to the hypoxic sensitivity (Fig. 1). This again suggests a stepwise rise of oxygen during the Ediacaran–Cambrian transition.

Both geological and geochemical data suggested that

atmospheric oxygen levels were increasing during the Cambrian explosion. Shallow waters were well-oxygenated at least since Late Ediacaran, whereas the deep oceans may have remained anoxic or sulfidic up to the Early Cambrian in some areas. The direct evidence for constraining the oxygen level is currently not available. Deduced from the ocean redox structure oxygen levels varied between 15% and 40% PAL at the time of Cambrian explosion, embracing the critical demand of animals. Recent isotopic data suggest an oxygen increase to modern-like level by Cambrian Age 3 (Chen et al., 2015).

With the increase of oxygen level, metazoan lineages abruptly appeared in an order of hypoxic sensitivity during the Ediacaran–Cambrian transition. The least sensitive animals (molluscs and annelids) appeared earlier, followed by more sensitive animals (arthropods and echinoderms), and the most sensitive animals (fishes) appeared much later. Therefore it is very likely that the increasing oxygen level controls the sequence of animal appearances in fossil record. Alternatively, if the dissolved oxygen concentration of surface oceans was sufficient for metazoan lineages well before the Cambrian explosion, the frequent upwelling of anoxic deep water may suppress the animal diversification. It is also likely that contraction of anoxic area causes the rapid diversification of metazoans during the Cambrian explosion because the areal extent of oxygenated bottom waters increased in the Early Cambrian (e.g. Chen et al., 2015).

ACKNOWLEDGMENTS

This work is supported by National Basic Research Program of China (No. 2013CB835002) and National Natural Science Foundation of China (No. 41272036).

REFERENCES CITED

- Antcliffe, J. B., Callow, R. H. T., Brasier, M. D., 2014. Giving the Early Fossil Record of Sponges A Squeeze. *Biological Reviews*, 89: 972–1004
- Berkner, L. V., Marshall, L. C., 1965. On the Origin and Rise of Oxygen Concentration in the Earth's Atmosphere. *Journal of Atmospheric Sciences*, 22: 225–261
- Blair, J. E., 2009. Animals (Metazoa). In: Hedges, S. B., Kumar, S., eds., *The Timetree of Life*. Oxford University Press, Oxford. 223–230
- Braddy, S. J., Poschmann, M., Tetlie, O. E., 2008. Giant Claw Reveals the Largest Ever Arthropod. *Biology Letter*, 4: 106–109
- Butterfield, N. J., 2009. Oxygen, Animals and Oceanic Ventilation: An Alternative View. *Geobiology*, 7: 1–7
- Campbell, I. H., Allen, C. M., 2008. Formation of Supercontinents Linked to Increases in Atmospheric Oxygen. *Nature Geoscience*, 1: 554–558
- Campbell, I. H., Squire, R. J., 2010. The Mountains that Triggered the Late Neoproterozoic Increase in Oxygen: the Second Great Oxidation Event. *Geochimica et Cosmochimica Acta*, 74: 4187–4206
- Canfield, D. E., 2005. The Early History of Atmospheric Oxygen: Homage to Robert M. Garrels. *Annual Review of Earth and Planetary Science*, 33: 1–36
- Canfield, D. E., Poulton, S. W., Narbonne, G. M., 2007. Late-Neoproterozoic Deep-Ocean Oxygenation and the Rise of Animal Life. *Science*, 315: 92–95
- Catling, D. C., Glein, C. R., Zahnle, K. J., et al., 2005. Why O₂ Is Required by Complex Life on Habitable Planets and the Concept of Planetary “Oxygenation Time”. *Astrobiology*, 5: 415–438
- Chen, X., Ling, H. F., Vance, D., et al., 2015. Rise to Modern Levels of Ocean Oxygenation Coincided with the Cambrian Radiation of Animals. *Nature Communications*, 6: 7142 (DOI: 10.1038/ncomms8142)
- Cloud, P. E Jr., 1948. Some Problems and Patterns of Evolution Exemplified by Fossil Invertebrates. *Evolution*, 2: 322–350
- Cloud, P. E., 1976. Beginnings of Biospheric Evolution and Their Biogeochemical Consequences. *Paleobiology*, 2: 351–387
- Conway M. S., Peel, J. S., 2008. The Earliest Annelids: Lower Cambrian Polychaetes from the Sirius Passet Lagerstätte, Peary Land, North Greenland. *Acta Palaeontologica Polonica*, 53: 137–148
- Danovaro, R., Dell’Anno, A., Pusceddu, A., Gambi, C., Heiner, I., Kristensen, R.M., 2010. The First Metazoa Living in Permanently Anoxic Conditions. *BMC Biology*, 8: 30.
- Decker, H., van Holde, K. E., 2011. *Oxygen and the Evolution of Life*. Springer, Berlin. 172
- Diaz, R. J., Rosenberg, R., 1995. Marine Benthic Hypoxia: A Review of Its Ecological Effects and the Behavioural Responses of Benthic Macrofauna. *Oceanography and Marine Biology: An Annual Review*, 33: 245–303
- Dries, R. R., Theede, H., 1974. Sauerstoffmangelresistenz Mariner Bodenvertebraten aus der Westlichen Ostsee. *Marine Biology*, 25: 327–333
- Erwin, D. H., Laflamme, M., Tweedt, S. M., et al. 2011. The Cambrian Conundrum: Early Divergence and Later Ecological Success in the Early History of Animals. *Science*, 334: 1901–1907
- Erwin, D. H., Tweedt, S., 2012. Ecological Drivers of the Ediacaran-Cambrian Diversification of Metazoa. *Evolutionary Ecology*, 26: 417–433
- Erwin, D. H., Valentine, J. W., 2013. *The Cambrian Explosion: the Construction of Animal Biodiversity*. Roberts and Company Publishers, Inc., Greenwood Village. 406
- Fedonkin, M. A., Waggoner, B. M., 1997. The Late Precambrian Fossil *Kimberella* Is a Mollusc-Like Bilaterian Organism. *Nature*, 388: 868–871
- Feng, L. J., Li, C., Huang, J., et al., 2014. A sulfate Control on Marine Mid-Depth Euxinia on the Early Cambrian (Ca. 529–521 Ma) Yangtze Platform, South China. *Precambrian Research*, 246: 123–133
- Gray, J. S., Wu, R. S., Or, Y. Y., 2002. Effects of Hypoxia and Organic Enrichment on the Coastal Marine Environment. *Marine Ecology Progress Series*, 238: 249–270
- Henriksson, R., 1969. Influence of Pollution on the Bottom Fauna of the Sound (Öresund). *Oikos*, 20: 507–523
- Hua, H., Chen, Z., Yuan, X. L., et al., 2005. Skeletogenesis and Asexual Reproduction in the Earliest Biomineralizing Animal *Cloudina*. *Geology*, 33: 277–280
- Jin, C. S., Li, C., Peng, X. F., et al., 2014. Spatiotemporal

- Variability of Ocean Chemistry in the Early Cambrian, South China. *Science China: Earth Science*, 57: 579–591
- Kasting, J. F., 1993. Earth's Early Atmosphere. *Science*, 259: 920–926
- Kendall, B., Anbar, A. D., Kappler, A., et al., 2012. The Global Iron Cycle. In: Knoll, A. H., Canfield, D. E., Konhauser, K. O., eds., *Fundamentals of Geobiology*. Wiley-Blackwell, Oxford. 65–92
- Knoll, A. H., Sperling, E. A., 2014. Oxygen and Animals in Earth History. *Proceedings of the National Academy of Sciences of the United States of America*, 111: 3907–3908
- Knoll, A.H., Carroll, S.B., 1999. Early Animal Evolution: Emerging Views from Comparative Biology and Geology. *Science*, 284: 2129–2137
- Knoll, A. H., Walter, M. R., 1992. Latest Proterozoic Stratigraphy and Earth History. *Nature*, 356: 673–678
- Kouchinsky, A., Bengtson, S., Clausen, S., et al., 2015. A Lower Cambrian Fauna of Skeletal Fossils from the Emyaksin Formation, Northern Siberia. *Acta Palaeontologica Polonica* (in press).
- Kouchinsky, A., Bengtson, S., Runnegar, B., et al., 2012. Chronology of Early Cambrian Biomineralization. *Geological Magazine*, 149: 221–251
- Kump, L. R., 2008. The Rise of Atmospheric Oxygen. *Nature*, 451: 277–278
- Landing, E., Geyer, G., Brasier, M. D., et al., 2013. Cambrian Evolutionary Radiation: Context, Correlation, and Chronostratigraphy—Overcoming Deficiencies of the First Appearance Datum (FAD) Concept. *Earth-Science Reviews*, 123: 133–172
- Li, C., Love, G. D., Lyons, T. W., et al., 2010. A Stratified Redox Model for the Ediacaran Ocean. *Science*, 328: 80–83
- Li, Z. X., Powell, C. M., 2001. An Outline of the Palaeogeographic Evolution of the Australasian Region since the Beginning of the Neoproterozoic. *Earth-Science Review*, 53: 237–277
- Ling, H. F., Chen, X., Li, D., et al., 2013. Cerium Anomaly Variations in Ediacaran–Earliest Cambrian Carbonates from the Yangtze Gorges Area, South China: Implications for Oxygenation of Coeval Shallow Seawater. *Precambrian Research*, 225: 110–127
- Love, G. D., Grosjean, E., Fike, D. A., et al., 2009. Fossil Steroid Record the Appearance of Demospongiae during the Cryogenian Period. *Nature*, 457: 718–721
- Lyons, T. W., Reinhard, C. T., Love, G. D., et al., 2012. Geobiology of the Proterozoic Eon. In: Knoll, A. H., Canfield, D. E., Konhauser, K. O., eds., *Fundamentals of Geobiology*. Wiley-Blackwell, Oxford. 371–402
- Mángano, M. G., Buatois, L. A., 2014. Decoupling of Body-Plan Diversification and Ecological Structuring during the Ediacaran–Cambrian Transition: Evolutionary and Geobiological Feedbacks. *Proceedings of the Royal Society B* 281, 20140038.
- Meert, J. G., 2003. Proterozoic East Gondwana: Supercontinent Assembly and Breakup. Special Publication 206, Eos, Transactions American Geophysical Union, 84: 372
- Meert, J. G., 2011. Gondwanaland, Formation. In: Reitner, J., Thiel, V., eds., *Encyclopedia of Geobiology*, Springer, Berlin. 434–436
- Mentel, M., Martin, W., 2010. Anaerobic Animals from an Ancient, Anoxic Ecological Niche. *BMC Biology*, 8: 32
- Miller D. C., Pouchet SL., Coiro L., et al., 1995. Effects of Hypoxia on Growth and Survival of Crustaceans and Fishes of Long Island Sound. In: McElroy A., Zeidner J., eds., *Proceedings of the Long Island Sound Research Conference: Is the Sound Getting Better or Worse*. New York Sea Grant Institute, Stony Brook, NY, p1–92
- Mills, D. B., Ward, L. M., Jones, C. A., et al., 2014. Oxygen Requirements of the Earliest Animals. *Proceedings of the National Academy of Sciences of the United States of America*, 111: 4168–4172
- Nielsen, C., 2012 (3rd edition). *Animal Evolution: Interrelationships of the Living Phyla*. Oxford University Press, Oxford. 402
- Papineau, D., 2010. Global Biogeochemical Changes at Both Ends of the Proterozoic: Insights from Phosphorites. *Astrobiology*, 10: 165–181
- Partin, C. A., Bekker, A., Planavsky, N. J., et al., 2013. Large-Scale Fluctuations in Precambrian Atmospheric and Oceanic Oxygen Levels from the Record of U in Shales. *Earth and Planetary Science Letter*, 369–370: 284–293
- Petsch, S. T., 2004. The Global Oxygen Cycle. In: Schlesinger, W. H., ed., *Biogeochemistry. Treatise on Geochemistry*, 8: 515–555
- Planavsky, N. J., Rouxel, O. J., Bekker, A., et al., 2010. The Evolution of the Marine Phosphate Reservoir. *Nature*, 467: 1088–1090
- Rhoads, D. C., Morse, J. W., 1971. Evolutionary and Ecological Significance of Oxygen-Deficient Marine Basins. *Lethaia*, 4: 413–428
- Rogers, J. J. W., Santosh, M., 2004. *Continents and Supercontinents*. Oxford University Press, Oxford. 289
- Rosenberg, R., 1972. Benthic Faunal Recovery in a Swedish Fjord Following the Closure of a Sulphite Pulp Mill. *Oikos*, 23: 92–108
- Runnegar, B., 1982. Oxygen Requirements, Biology and Phylogenetic Significance of the Late Precambrian Worm *Dickinsonia*, and the Evolution of the Burrowing Habit. *Alcheringa*, 6: 223–239
- Runnegar, B., 1991. Precambrian Oxygen Levels Estimated from the Biochemistry and Physiology of Early Eukaryotes. *Global and Planetary Change*, 97: 97–111
- Shu, D. G., Luo, H. L., Conway Morris, S., et al., 1999. Lower Cambrian Vertebrates from South China. *Nature*, 402: 42–46
- Shu, D. G., Isozaki, Y., Zhang, X. L., et al., 2014. Birth and Early Evolution of Metazoans. *Gondwana Research*, 25: 884–895
- Skovsted, C., B., Peel, J. S., 2011. *Hyolithellus* in life position from the Lower Cambrian of North Greenland. *Journal of Paleontology*, 85: 37–47
- Sperling, E. A., Frieder, C. A., Raman, A. V., 2013a. Oxygen, Ecology, and the Cambrian Radiation of Animals. *Proceedings of the National Academy of Sciences of the United States of America*, 110: 13446–13451

- Sperling, E. A., Halverson, G. P., Knoll, A. H., et al., 2013b. A Basin Redox Transect at the Dawn of Animal Life. *Earth and Planetary Science Letter*, 371–372: 143–155
- Wang, J. G., Chen, D. Z., Yan, D. T., et al., 2012. Evolution from An Anoxic to Oxidic Deep Ocean during the Ediacaran–Cambrian Transition and Implications for Bioradiation. *Chemical Geology*, 306: 129–138
- Wang, H., Li, C., Hu, C., et al., 2015. Spurious Thermoluminescence Characteristics of the Ediacaran Doushantuo Formation (Ca. 635–551 Ma) and Its Implications for Marine Dissolved Organic Carbon Reservoir. *Journal of Earth Science*, 26(6): 883–892
- Wen, H. J., Carignan, J., Chu, X. L., et al., 2014. Selenium Isotopes Trace Anoxic and Ferruginous Seawater Conditions in the Early Cambrian. *Chemical Geology*, 390: 164–172
- Wang, Y., Wang, X. L., Wang, Y., 2015. Cambrian Ichnofossils from the Zhoujieshan Formation (Quanjia Group) Overlying Tillites in the Northern Margin of the Qaidam Basin, NW China. *Journal of Earth Science*, 26(2): 203–210
- Wray, G. A., 2015. Molecular Clocks and the Early Evolution of Metazoan Nervous Systems. *Philosophical Transactions of the Royal Society Series B*, 370 (150046), 1–11
- Yang, B., Steiner, M., Li, G. X., et al., 2014. Terreneuvian Small Shelly Faunas of East Yunnan (South China) and Their Biostratigraphic Implications. *Palaeogeography, Palaeoclimatology, Palaeoecology*, 398: 28–58
- Yin, Z. J., Zhu, M. Y., Davidson, E. H., et al., 2015. Sponge Grade Body Fossil with Cellular Resolution Dating 60 Myr before the Cambrian. *Proceedings of the National Academy of Sciences of the United States of America*, 112: E1453–E1460
- Zhang, X. L., Shu, D. G., 2014. Causes and Consequences of the Cambrian Explosion. *Science China—Earth Sciences*, 57: 930–942
- Zhang, X., Shu, D., Han, J., et al., 2014. Triggers for the Cambrian Explosion: Hypotheses and Problems. *Gondwana Research*, 25: 896–909