Biogeochemistry and Geographical Ecology: Embracing All Twenty-Five Elements Required to Build Organisms*

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**Abstract:** Biogeochemistry is a key but relatively neglected part of the abiotic template that underlies ecology. The template has a geography, one that is increasingly being rearranged in this era of global change. Justus von Liebig’s law of the minimum has played a useful role in focusing attention on biogeochemical regulation of populations, but given that ~25+ elements are required to build organisms and that these organisms use and deplete nutrients in aggregates of communities and ecosystems, we make the case that it is time to move on. We review available models that suggest the many different mechanisms that give rise to multiple elements, or colimitation. We then review recent empirical data that show that rates of decomposition and primary productivity may be limited by multiple elements. In that light, given the tropics’ high species diversity and generally more weathered soils, we predict that colimitation at community and ecosystem scales is more prevalent closer to the equator. We conclude with suggestions for how to move forward with experimental studies of colimitation.

**Keywords:** multiple-element limitation, colimitation, law of the minimum, decomposition, substrate age hypothesis, soil nutrient availability.

**Prelude: Ecology as Rational Field Physiology**

Frederic Clements viewed ecology as physiology in service of biogeography (Clements 1905; Hagen 1992). In the nineteenth century, Darwin (1839) and Von Humboldt (Von Humboldt and Bonpland 2010) had begun to reveal patterns of species ranges and the distribution of vegetation. In his *Research Methods in Ecology*, Clements (1905, p. 2) saw these biogeographical patterns as the raw material to develop ecology as “rational field physiology,” simultaneously getting the physiologist out of the lab and into the field while guiding what he saw as a largely descriptive natural history (Hagen 1992). Toward that end, he devoted much of the book to measuring water and temperature, two variables that Krebs (1994, p. 93) would later refer to as ecology’s “master” regulators. Meanwhile, mineralogist Vladimir Vernadsky, summarizing decades of thinking on the subject, was viewing the totality of living organisms as “expressed as in terms of chemical elements . . . as a moving rock formation endowed with free energy” (Vernadsky 1924; Smil 2000). In doing so, the three parts of ecology’s abiotic template—water, temperature, and mineral elements—were in place.

Vernadsky’s work was given a wider audience by G. Evelyn Hutchinson (1948, p. 393), who noted that in addition to the importance of temperature and water, “Looking at man from a strictly geochemical standpoint, his most striking character is that he demands so much—not merely thirty or forty elements for physiological activity, but nearly all the others for cultural activity.” Hutchinson elaborated on the two basic reasons that biogeochemistry was important to ecology: that the recipe for life is elemental and that humans continue to rearrange the geography of those elements in the same way we are rearranging water and temperature.

More than half a century later, despite pioneering work (Sterner and Elser 2002; Simpson et al. 2010), we are still some distance from fully incorporating biogeochemistry into Clements’s vision of a rational physiology. There are challenges to this synthesis. Water and temperature are single quantifiable things; biogeochemistry incorporates at least 25 chemical elements, all required ingredients to build an organism (Marschner 1995; Frausto da Silva and Williams 2001; Wackett et al. 2004). Data on the organismal chemistry of individuals and populations still, arguably, lag behind data on their genomes. Unlike temperature and precipitation, we lack the baseline data describing biogeochemistry at scales from square meters to continents.

But there are also opportunities to synthesize for generating deeply integrative biogeochemical ecology. Elemen-
tal shortfall creates patterns across scales, resulting in pathology and developmental abnormalities in individuals (Marshchner 1995; National Research Council 2000, 2005; Snell-Rood et al. 2014), changing growth rates and behavioral interactions within populations (Jones and Hanson 1985; Elser et al. 1996; McDowell 2003; Simpson et al. 2006) and limiting ecosystem production and respiration (Elser et al. 2007; Kaspari et al. 2008; Townsend et al. 2011; Wieder et al. 2015). Increasingly economical technologies are arising to quantify elements both within individuals and across landscapes (Asner et al. 2015). A large part of global change involves the global rearranging of elements via nitrogen (N) fertilizer manufacture and deposition (Vitousek et al. 1997; Hietz et al. 2011), phosphorus (P) mining (Smil 2000), sodium (Na) pollution through road salt application (Kaspari et al. 2010), heavy metal pollution (Rauch and Pacyna 2009; Nagaiyoti et al. 2010), and increased dust mobility (Field et al. 2010; Mahowald et al. 2010). Understanding the elemental recipes of organisms, the ecological function of those elements, and their geography has considerable potential for understanding our past, present, and future earth.

In this article, we discuss efforts so far to come to grips with biogeochemical limitation in ecology, why we need to embrace the diversity of elements required to build organisms and drive ecosystems, and some ways we may do so, all toward achieving Clements’s and Vernadsky’s goal of better understanding the geography of life. We use this framework to speculate on a well-discussed but unresolved issue: the role of biogeochemistry in driving differences between temperate and tropical ecosystems (Walker and Syers 1976). Our primary motivation is to convince more young scientists of a nagging intuition that has grown in the both of us: that much of the pattern in population, community, and ecosystem ecology as one moves from place to place arises from a shortage of one or more of the 25 elements required for life.¹

**Liebig’s Fertilizer and the Origins of Nutrient Limitation**

Nutrient limitation² is a basic concept of integrative biology, beginning inside the cell and scaling up to ecosystems. Consider the Droop equation (Droop 1974), designed to model growth rate of a single cell

\[ \mu = \mu_m(1 - q/Q). \]

Here, \( \mu \) is the biomass growth rate (day⁻¹), \( \mu_m \) is the maximum growth rate at theoretically infinite quota, \( q \) is the minimum quota of a nutrient necessary to just support life (moles per cell at zero growth), and \( Q \) is the observed quota (moles per cell). As the nutrient supply in the cytosol increases, the part of the equation in parentheses converges on 1, giving the maximum growth rate. On the basis of these dynamics, nutrient limitation increases (and growth rate decreases) as nutrient supplies decline. Nutrient limitation thus exists along a range of nutrient availability, where the performance of the system is enhanced with the addition of nutrients or suppressed with their depletion.

In the mid-1800s, Justus von Liebig—a polymath chemist and biologist—devised a simple way to identify limiting elements, one not far distant from the cell scenario of the Droop equation (Brock 2002). Liebig aimed to maximize the yield of crop monocultures—one population, one genome, occupying a large area. His model was a metaphor (Liebig 1855): he likened the problem to an oaken bucket with vertical staves of varying lengths. To increase the capacity of the bucket, it was not necessary to increase the length of each stave; it was only necessary to increase the length of the shortest stave. This metaphor sold lots of fertilizer (Brock 2002).

Liebig’s law of the minimum (LLM) became a foundational model for ecology. Codified by Sterner and Elser (2002), LLM has two assumptions. First, an element’s importance can be ranked by the ratio of biological demand to ecosystem supply (e.g., table 1). The second is that the physiology of organisms and ecosystems is organized such that only one element—the one with the highest demand to supply ratio—limits metabolism. Placed in the context of Droop’s law, LLM has two parts. First, elements are essential: any concentration less than \( \mu_m \) moves performance leftward and downward on the Droop curve. Second, energy and resources are expended to harvest elements, and these costs diminish as availability increases. Measuring the availability of nutrients in the environment and in the tissues of the study system thus informs the working hypothesis for which nutrients are most likely to limit performance (e.g., Sterner 1997).

The application of Liebig logic has since led to a focus on three elements as candidates for most limiting nutrient, in large part on the basis of their considerable fraction in the cells and tissues of life. Optimal foraging theory (Stephens and Krebs 1986) has successfully predicted how consumers—from notonectids to hummingbirds—choose among food and habitats simply by assuming that they are maximizing the intake rate of energy (and ultimately carbon).

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¹. We offer two caveats before proceeding: we are terrestrial ecologists, and not particularly good ones. Forgive us if we ignore aquatic animals. We have also decided to pass when it comes to the dark side of biogeochemistry, the role of out and out toxins like Pb, and when too much of an essential element becomes a decided to pass when it comes to the dark side of biogeochemistry, the role of out and out toxins like Pb, and when too much of an essential element becomes a bad thing.

². At the outset, let us say that nutrient limitation is an imperfect expression that we retain, given its utter ubiquity. To see why, consider opening a bad thing.
Macroeocology uses similar logic to link net primary productivity—in units of carbon/area/time—to increasing nutrient abundance at continental scales (McNaughton et al. 1989; Kaspari et al. 2000). Nitrogen, the next most common element beyond CHO, frequently limits biomass and production in plants and herbivores (White 1993; LeBauer and Treseder 2008). The growth rate hypothesis (Elser et al. 1996) is an elegant integrative hypothesis, extrapolating from P-rich RNA and ribosomes to P-limited growth rates (Elser et al. 2003). Clearly, biogeochemistry has a role to play as a dominant ecological template.

### Why It Is Time to Retire Liebig and Embrace Colimitation across the Periodic Table

One’s success as a scientist can be measured more by the number of people he or she puts to work on new problems than by the correctness of specific research results. (Raup 1999)

The Droop equation uses a single cell as its starting point. Nutrient limitation also expresses itself in tissues and organisms as aggregates of cells and in populations and ecosystems as aggregates of individuals (Arrigo 2005; Sperfeld et al. 2015). In these collections of individuals and populations in time and space, the seeds of colimitation are planted.

Liebig’s law has had a good run in introducing ecologists to the power of biogeochemistry. LLM has the power of parsimony; a single limiting element is the simplest starting point, but at the same time it has an epistemological Achilles’s heel: it is difficult (and, to our knowledge, unheard of) to rigorously evaluate LLM’s prediction that one element, and one alone, contributes to an ecological process. Another reason to retire LLM lies in its demonstrated application in nutritional ecology. Considerable time and energy has been invested in formulating optimal diets precisely because they maximize the health of model organisms (Smith 1987; Wertz 1987; Cohen Silva and Williams 2001). Moreover, while trace metals comprise 0.1% of animal mass—Mn, Fe, Co, Ni, Cu, Zn, Mo, B, Si, and Se—they are functional parts of up to one-third of 4,000 known enzymes that catalyze life (Bairoch 2000; Wadron et al. 2009). Each of these elements is essential; shortfalls in any cause pathology. And all are patchily distributed in space (Jones and Hanson 1985; John et al. 2007; Smith et al. 2014).

Perhaps the single biggest case for ubiquitous colimitation lies in its demonstrated application in nutritional ecology. Considerable time and energy has been invested in formulating optimal diets precisely because they maximize the health of model organisms (Smith 1987; Wertz 1987; Cohen 2004). Likewise, mineral licks—supplementing structural, electrolytic, and trace elements—are placed in the field to enhance the health and biomass of wildlife and domesticated animals (Jones and Hanson 1985). Geometric optimal diet models are consistently describing how consumers in the lab and field balance their diets by aiming at mixed nutritional targets (Simpson et al. 2010).

### Limitation Cascades

In metabolic pathways, what happens in one part of the network often ramifications downstream. Limitation cascades arise
when nutrient limitation in one component influences the intensity of nutrient limitation in another. They can result in a ranking of nutrient effects, not toward identifying a single limiting factor sensu Liebig but in distinguishing, in Peter Vitousek’s (2010) parlance, proximate limitation (where adding a nutrient influences a process but does not ramify) from ultimate limitation (where the added nutrient ramifies and restructures the system, changing both process dynamics and standing stocks).

Limitation cascades were powerfully documented when whole lake P fertilization transformed oligotrophic lakes into eutrophic ones (Schindler 1990). In the process, N inputs increased through biological fixation, as did CO₂ diffusion. Likewise, because shortages of N commonly limit population growth (Vitousek et al. 1993; White 1993), ecosystem respiration (Horner et al. 1988), and productivity (LeBauer and Treseder 2008), any process increasing ecosystem N has the potential to generate a limitation cascade. For example, the fixation of atmospheric N₂ to organic forms useful for life requires ample carbohydrates as well as Fe and Mo to build the catalyzing enzyme, nitrogenase. In the dark, low-carbohydrate understory of a Panamanian rain forest, fertilization with the trace metal Mo generated two- to threefold increases in the production of organic N by heterotrophic microbes (Barron et al. 2008). Moreover, in an era of increasing CO₂, the resulting increase in carbohydrate production density should increase plant demand for N—while simultaneously providing the energy for N-fixing microbes—and the demand for Mo and Fe (Hungate et al. 2003).

For plant consumers (i.e., herbivores and detritivores), Na is a prime candidate as an ultimate nutrient that can initiate limitation cascades. Sodium exists as a trace element in most plant tissue (Marchner 1995). Herbivores and detritivores—particularly fungi and animals—maintain Na tissue levels 100–1,000-fold higher than plants (Gromack et al. 1977; National Research Council 2005). Consumers confronting Na shortfall often conserve Na by decreasing activity (Kaspari et al. 2014). If Na shortfall decreases the physiological performance of plant consumers, one corollary is that the positive benefits of N and P to herbivores and detritivores will vary along a Na gradient. If so, gradients of Na availability may account for the demonstrated variation in consumer responses to N and P fertilization (Haddad et al. 2000; Ritchie 2000; Milton and Kaspari 2007; Kaspari et al. 2008b, 2009; Bishop et al. 2010; Loaiza et al. 2011; Joern et al. 2012; Lind et al. 2014).

Organisms Deplete Multiple Elements, Equalizing Utility: Availability

Another process promoting colimitation results from life’s long interaction with geochemistry (Frausto da Silva and Williams 2001; Vernadsky 2012). If we plot the ubiquity of elements in a common vertebrate (us) against the availability of elements in seawater or soil, we find a generally positive relationship (fig. 1). It is hard, seeing this robust pattern, not to conclude that natural selection favors the use of common elements when building ubiquitous biological orga-
tures. One consequence is that the ratio of demand to supply tends to equalize across the range of essential elements. Put another way, if the ratios of demand:availability converge toward a common value, there will never be the clear, enduring maximum. Instead, the elements most in demand relative to supply may alternate at a timescale proportional to the physiology of the smallest organisms, among a number of potential candidates (Appling and Heffernan 2014). Any experiment that sums over those cycles will observe colimitation.

Scale: Aggregating Populations over a Matrix of Biogeochemical Availability

A final way of achieving biogeochemical colimitation that complements all the rest is relevant when ecologists measure processes and standing stocks over larger areas, longer timespans, and more populations (Levin 1992). About half of the globally observed variability in NP stoichiometry exists among species in a given community (Hillebrand et al. 2009; Elser et al. 2010). The elemental recipes of taxa differ in ways directly relatable to their niche. One of us has found, for example, that across 26 forest stands in Peru and Panama, soils richer in calcium supported more Ca-rich isopods; those richer in nitrogen supported more organisms that use N-rich silk (Kaspari and Yanoviak 2009). Any study examining the abundance and diversity of soil invertebrates collectively would conclude that Ca and N colimit this community.

The role of nutrients in limiting decomposition is key to understanding half the carbon cycle (primary production being the other half). In a square meter of forest floor, thousands of species and billions of individual bacteria, fungi, and invertebrates break down leaf litter (Swift et al. 1979). Scattered within the plot may be numerous kinds of substrate in various states of decay (fig. 2). Suppose that two are a freshly fallen leaf and a decayed leaf that is little more than lignin and cellulose. A third substrate is a dead grasshopper fallen from the forest canopy. If the bacteria and fungal decomposers exploiting these substrates use different enzymes to do the job, and if a third of these enzymes requires metal

Figure 2: Conceptual model of the processes contributing to colimitation at increasingly coarse spatial scales. Photo of the forest canopy by R. Montgomery.
cofactors, it is easy to surmise that different portions of that square meter plot will vary in their demand for mineral elements.

Tests for colimitation and importance of elements other than N and P in decomposition have thus, not surprisingly, found it. Eleven-week lab incubations of Costa Rican leaf litter with added N, P, Zn, K, Mg, or Ni yielded higher mass loss and CO₂ efflux with P and Zn (Powers and Salute 2011). In a Panamanian forest, at least three elements—P, K, and at least one micronutrient (B, Ca, Cu, Fe, Mg, Mn, Mo, S, and/or Zn) when applied to 40 × 40-m plots—enhanced decomposition of cellulose wood and/or leaf litter (Kaspari et al. 2008a). NaCl in inland forests, applied at dosages mimicking urine and coastal rainfall, enhanced decomposition as much as P and K (Kaspari et al. 2009, 2014). Ecosystems that combine the activity of 10⁻¹⁷-g bacteria through 10⁻³-g trees will invariably sum over the activities of a multitude of populations, each with their own footprint (Waring et al. 2015), most not directly interacting. When we ecologists quantify nutrient limitation, colimitation increasingly seems inevitable.

Is There a Geography of Colimitation?

If colimitation should be the working hypothesis in ecology, are we equally likely to observe it everywhere, or should we expect geographic variation in which suites of nutrients are most important? The Walker-Syers model (1976), also referred to as the substrate age hypothesis (Zechmeister-Boltenstern et al. 2015), is a popular paradigm in terrestrial ecosystems ecology, and biogeochemistry that provides a model for where to expect N versus P limitation. In brief, the model differentiates between elements whose ultimate source is from rocks (e.g., P, cations) versus the atmosphere (primarily N). During primary succession (the classic thought experiment imagines a freshly formed lava flow condensing and cooling before being colonized by organisms), a terrestrial ecosystem has the maximum amount of rock-derived nutrient capital like P, with negligible quantities of elements like N derived from the atmosphere. Over geological time as soils and ecosystems develop, P availability decreases through processes such as leaching and occlusion to soil minerals, which render it unavailable. Concurrently, ecosystem N capital and availability increase through processes including free-living and symbiotic nitrogen fixation. Thus, over long periods of time, nutrient limitation to primary producers is expected to shift from N to P, with perhaps an intermediate phase of N and P colimitation.

Tests of the Walker-Syers hypothesis demonstrate the variety of ways ecologists can document the geography of nutrient limitation. Studies of elemental composition of trees show higher tissue P and higher P-induced growth rates at higher latitudes (Reich and Oleksyn 2004; Kerkhoff et al. 2005; Lovelock et al. 2007). Chronosequence studies allow one to the test how local patterns of substrate age and weathering generate the same transition from N to P limitation (Wardle et al. 2004), often overlaying fertilization experiments with N and P for good measure (Vitousek et al. 1993, 1997; Herbert et al. 2003). Collectively, these data support the importance of soil weathering at multiple scales in generating transitions from soil-derived nutrients to air-derived nutrients (Reich and Oleksyn 2004; Huston 2012).

But can Walker-Syers’s logic for a latitudinal gradient of single-element limitation be expanded to predict patterns of colimitation? We suggest two reasons why and then add a big caveat. First, multiple elements like Zn, Cu, and Mg are leached from rock. As one moves in time and space from rock newly emerged from under a glacier that has been exposed to millions of years of weathering, it should result in the simultaneous decline in a variety of essential elements. This is one way to move into a zone of potential colimitation (Saito et al. 2008; Appling and Heffernan 2014). Second, tropical diversity may be more likely to generate colimitation if individuals and species differ in their nutrient use and uptake, as has been shown in tropical forest trees (Baribault et al. 2012; Waring et al. 2015). The increase in diversity of many tropical clades combined with the depletion of multiple essential elements predicts an increase in the incidence of colimitation toward the equator.

Now the caveat. The more biogeochemists map out the distribution of elements, the more intriguing variation we find at multiple spatial scales, including continental ones. For example, geomorphic processes of erosion and deposition and/or nutrient inputs via sea salt inputs or dust deposition may disrupt substrate age hypothesis predictions of which nutrients limit productivity (Chadwick et al. 1999; Porder et al. 2007; Weintraub et al. 2015) and, by extension, colimitation, by adding nutrients or exposing unweathered substrate. Consider how a recent interpolated map of zinc concentrations in the top 5 cm of soil across the United States (Smith et al. 2014) demonstrates striking landscape and regional variability, with concentrations up to three orders of magnitude lower in the highly weathered soils of the Southeast (fig. 3). Contemplating this and other maps in Smith et al.’s (2014) opus highlights that biogeochemical gradients occur at every spatial scale, arise from a variety of causes (including anthropogenic), and show just how much potential lies in a research program that exploits these gradients.

That said, we need to build on studies of N and P limitation to reveal the extent of colimitation from populations to ecosystems. One model toward this end is NutNet (Fay et al. 2015), a distributed fertilization experiment in 42 grasslands, where 5 × 5-m plots were fertilized with factorial combinations of N, P, or K combined with micronutrients. Across the 42 sites spanning five continents, evidence for colimitation of aboveground primary production was pervasive, with NP limitation at 60% of the sites. Colimitation was observed
on all continents, suggesting that the presence and magnitude of colimitation depends locally on climate and background edaphic conditions. Nevertheless, N limitation to aboveground primary production was highest in cool, temperate latitudes, although low latitudes were not well represented in the data set. Clearly, much more work is needed to resolve the where, when, and what elements questions related to colimitation.

**Going Forward**

We understand that the parsimonious simplicity of LLM has an attraction, and the first half of Liebig logic—the element with the largest ratio of demand to availability is most limiting—has gotten us this far. At the same time, as the integration of biogeochemistry into evolutionary and global change ecology goes forward, there are ample opportunities for transformative insights. Here we identify a few, but first . . .

**Why Assuming Liebig Single-Element Limitation Can Be Very, Very Bad**

There are compelling reasons for embracing colimitation and its corollary: that we investigate the periodic table beyond C, N, and P. In global change biology, interactions between carbon and other elements are critical for accurately modeling the carbon cycle (Hungate et al. 2004; Townsend et al. 2011; Wieder et al. 2015). In studies of insect development, monarch butterflies reared on milkweeds enriched with road salt runoff showed altered muscle mass and neural development compared with those on plants from pristine environments (Snell-Rood et al. 2014). Anthropogenic changes in just one element’s distributions3 have the potential to transform our understanding of such diverse fields of inquiry as life-history evolution (Snell-Rood et al. 2015) and carbon cycling (Kaspari et al. 2009, 2014).

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3. An element that is, with apologies to T. C. R. White, not nitrogen.
Colimitation makes the question of whether nitrogen limits X contingent on the availability of others. Thus, understanding the covariance structure of nutrients in soils is useful for two reasons. First, it allows us to refine our answer (i.e., in P-poor soils, N is more limiting than in P-rich soils). Second, it provides us with ways of exploring the interaction of N with other nutrients, using the comparative method (for inspiration, see fig. 3).

Where to begin? Plots of an organism’s elemental composition versus environmental availability can catalyze a lot of working hypotheses. Figure 1, comparing the composition of a vertebrate endotherm to seawater, suggests that N, P, Fe, and Zn would be worth a look as limiting elements. Comparing that same stoichiometry to the forest soils of Peru, we see that P, Na, and Zn show LLM’s requisite high ratio of use to availability. But to make such plots, we need the raw data at scales applicable to the organisms and systems in question. Toward that end, Karasov and Martinez del Rio’s (2007) *Physiological Ecology* is a productive mix of literature and methodology toward evaluating dietary requirements.

*Describing the Chemical Recipe of Life and Its Consequences*

We suspect that at the time of this writing, it is easier to find a population’s genome than its elemental recipe. Documenting stoichiometric variation within and between populations is a critical first step for generating hypotheses of nutrient limitation (Saito et al. 2008; Sterner 2008; Kaspari and Yanoviak 2009; Kaspari et al. 2009). Such studies are revealing that about half of the globally observed variability in stoichiometry exists among species in a given community (Kraft et al. 2008; Hillebrand et al. 2009; Elser et al. 2010). Some of these differences reflect niche differences linked to metabolic subunits: high densities of ribosomes (and high titers of P) are found in bacterial taxa characterized by rapid growth (Stevenson and Schmidt 2004). To what extent do gradients of biogeochemistry undergird coexistence in communities (Tilman 1982), and what are the functional traits that provide a mechanism for these population interactions (McGill et al. 2006)?

*Mapping Availability at Multiple Scales*

For ecological stoichiometry to be predictive, we need better maps of biogeochemistry at scales relevant to our study organisms (e.g., fig. 3 is more likely to describe the Zn availability experienced by a population of trees, less so a population of mites). New technologies and approaches have the potential to transform our understanding of biogeochemical distributions and their consequences. Maps of mineral licks—deposits rich in elements like Na, Mg, and Ca (Jones and Hanson 1985)—allow for the study of mineral deficiency using the comparative approach. Networks of in situ solute sensors that collect high-frequency data on nutrient availability may provide information on the temporal uncoupling of nutrient supply and demand (Appling and Heffernan 2014). Last, recent advances in airborne imaging and spectralomics are providing regional-scale snapshots of foliar canopy chemistry and revealing strong coupling between canopy chemistry and underlying geologic and geomorphological landscape templates (Asner and Martin 2009; Asner et al. 2015).

*Testing for Colimitation*

Testing for colimitation can seem daunting. How does one sensibly and systematically test which biogeochemicals drive biology and ecology? As we document above, field factorial experiments have been key in recognizing the ubiquity of colimitation (see also Sperfeld et al. 2015). However, at the same time, such experiments blow up when more than four elements—and their interactions—are simultaneously explored. We suggest two expedient routes to effectively explore colimitation. As a test case, we use the hypothesis that an ecosystem’s decomposition rate is limited by biogeochemistry. The kitchen sink experiment aims at documenting the presence of colimitation by fertilization with multiple elements (i.e., everything but the kitchen sink), while the drug discovery experiments aims at rapid screening of candidate elements across the periodic table.

*Manipulating Fertility: The Kitchen Sink*

There are a variety of foundational working hypotheses of nutrient limitation, which is a fundamentally bottom-up process (Power 1992). But beyond the null hypothesis of no nutrient limitation in the system, a second relatively simple hypothesis is that increasing the availability of all elements equally serves to maximize decomposition rates. Such experiments could relatively quickly describe what is biogeochemically possible in the nutrient limitation of decomposition. At the same time, if performed across multiple sites, the magnitude of observed increase (or lack thereof) would be informative. If those ecosystems lie along a biogeochemical, thermal, precipitation, or biodiversity gradient, kitchen sink experiments could shed light on the relative contribution of these different abiotic versus biotic, bottom-up versus top-down drivers.

We envision two basic types of kitchen sink experiments. The first would compare decomposition on control plots with those that double the soil availability of N through Zn. The second would raise the nutrient availability of all sites to that of the richest site. Obviously, the devil is in the details (i.e., what form of nutrients do you add, what concentrations,
for how long). That said, it is interesting that a micronutrient kitchen sink in Panama generated higher rates of decomposition than P or K fertilization plots (Kaspari et al. 2008a).

Drug Discovery as a Model for Decomposing the Biogeochemistry of Decomposition

Drug prospectors search for the compounds, dosages, and contexts that control disease (Sterner and Elser 2002; Harvey 2008; Kerns and Di 2008). We suggest that Big Pharma’s progression from in vitro to in vivo—from low-investment, high-throughput tests to study direct effects to long-term studies with the most promising candidates to study indirect effects—is another promising approach for studies of colimitation.

We envision a three-step screening process to identify which elements affect decomposition, similar to the drug discovery model. Much of drug discovery still relies on open (or grind and find) screening: broadly collecting organisms and testing for the efficacy of their constituent compounds (see Adriamycin and Taxol; Subramanian et al. 2006). Candidate compounds are isolated and tested in vitro over hours to days in simplified assays against laboratory cell cultures (phase 1). Such tests occur at a massive scale. Consistent success (e.g., killing cancer cells) moves a candidate drug to the next phase of the trial. In the case of decomposition, a parallel experiment would consist of adding a battery of single nutrient element additions (e.g., N, Ca, P, S, Na, K, Cl, Mg, Si, Fe, Zn, Cu, I, Mn, F, Cr, Se, Mo, Co) to decomposing microcosms in vitro for several weeks (following Powers and Salute 2011). Enhanced CO₂ fluxes or mass loss rates would move the candidate element to the next phase.

During phase 2, the fraction of candidate drugs that function as predicted in tissue culture is next tested in model organisms (e.g., mice). The focus is on the direct effect (e.g., apoptosis, killing cancer cells) in a functioning system. If the compound fails muster, further testing is discontinued. Such tests take days to months. In decomposition experiments, candidate elements that increase CO₂ fluxes in soil should be tested in mesocosms stocked with soil, litter, and macroinvertebrates (e.g., isopods) and monitored for several months.

The <0.1% of candidate compounds that reach therapeutic trials are administered in the target organism (typically us) under realistic conditions. The goal of phase 3 trials is to evaluate the drug’s function, the chains of ramifying indirect interactions (side effects), and host acclimation (loss of drug function). Candidate elements advancing to phase 3 in decomposition experiments would then be tested in the field—in large-scale, longer-term (3–5 years) fertilization experiments alone and in factorial combinations—to look for the presence of interactions and track effects throughout trophic levels.

Conclusions

Humans are rearranging the geography of biogeochemistry and element availability, with uncertain consequences for ecosystem function and evolution. Addressing these challenges requires us to be equipped with robust theories and conceptual models that inform our data collection. We have argued that it is time to retire Justus von Liebig’s law of the minimum and embrace a more complex view of element limitation that acknowledges the interactions of 25+ elements both inside and outside the cell. We propose the untested hypothesis that colimitation is more pervasive in the tropics than in the temperate or boreal zone. Although moving from one square on the periodic table to a much larger number of squares—and from there to the biosphere—may seem daunting, we think that the ground for such studies is fertile, and there is much to be learned.

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Literature Cited


The Case for Multiple-Element Limitation


Symposium Editor: Marlene Zuk

Tropical forest of Barro Colorado Island, Panama. The thousands of species that live here, each with a subtly different chemical recipe, are increasingly seen to respond positively to additions of a variety of elements. Photo credit: Michael Kaspari.