Trends in
Ecology & Evolution

CellPress
OPEN ACCESS

Review

Second thoughts about first principles in biology

Dustin Marshall [1,*], Craig White [1], Van Savage [2,3], and Naomi Levine [4]

First-principles approaches, based on physics or chemistry, are key pillars of biological theory. Despite their value, there are recurrent problems with many first-principles approaches in biology: they often include incomplete physics or are driven by unscrutinised biological assumptions. Meanwhile, debates about biological theories based on first principles usually focus on the fit of predictions to data—we argue such a focus confuses prediction with explanation. A good fit of predictions to data is a necessary, but insufficient condition for a theory to be sound. First-principles approaches in biology will only be effective when the physics and biology are both sufficiently complete. Determining what elements are essential to include is an enduring but exciting challenge of science.

Highlights

First-principles approaches underpin many biological theories, many of which claim to be firmly grounded in physics, such that their principles are inviolable.

Many predictions about how global change will affect biological systems are based on biological theories that purport to be based on first principles.

Many first-principles approaches are based on incomplete physics, while others have surprising biological assumptions at their core that are often overlooked.

We argue that first-principles approaches in biology can be powerful and hold tremendous promise, but biologists should increase their scrutiny of these approaches.

Physics as a powerful tool for explaining biology

The richness and complexity of biology demand explanation, but where to start? Biology exists in, and is constrained by, the physical world. Many biological theories reflect this reality by using a '**first-principles approach**' (see Glossary), building upwards from the physics of a biological system. The invocation of first principles (sometimes termed 'mechanistic constraints') as explanations for how, or why, organisms do the things they do has a long, august history in biology [1,2]. First-principles arguments continue to guide biological debates, from the evolutionary drivers of size [3] to theories of biodiversity [4].

The enduring appeal of first-principles approaches in biology has several sources. First-principles approaches feel both more general and more robust. First-principles approaches ideally use established science that has identified simplifying 'rules' at lower levels of biological organisation that can be leveraged to understand more complicated processes. A mechanistic theory, *sensu stricto*, requires no new experimental estimation of parameters [5]; though in practice, most include a mix of established, estimated, and assumed parameters [6]. The predictions of first-principles models should depend less on the vagaries of any one species or condition.

The promise of first-principles approaches is obvious: they start from a strong base (e.g., physical laws) and provide an important starting point for parsing biological complexity into some kind of order. Reducing complex biological phenomena into deterministic engines with clear rules or constraints is necessary for developing predictive models. First-principles approaches are also undeniably successful in solving interesting biological problems. For example, **Stokes' law** successfully predicts pollen dispersal and the sinking of marine snow [7,8]. Finally, first-principles arguments also seem more robust: for example, who can deny that larger cells have less surface area relative to their volume than smaller cells?

First-principles approaches in biology have tremendous potential, but they are also fraught because they necessarily sit at the interface of physics and biology. Consequently, first-principles theories may be inadequately scrutinised because most biologists are not also physicists, and

[1] School of Biological Sciences, Monash University, Melbourne, Victoria, Australia
[2] Department of Biomathematics, University of California-Los Angeles, Los Angeles, CA, USA
[3] Department of Ecology and Evolutionary Biology, University of California-Los Angeles, Los Angeles, CA, USA
[4] Department of Biological Sciences, University of Southern California, Los Angeles, CA, USA

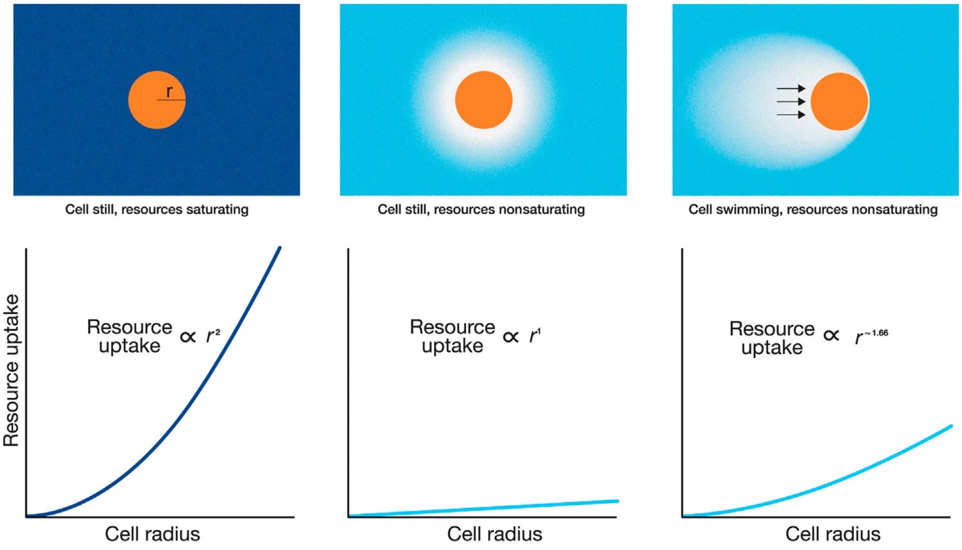
*Correspondence:
dustin.marshall@monash.edu
(D. Marshall).

vice versa. Challenges arise whenever concepts or theories from one field are applied to another without sufficient awareness of their underlying limitations—this applies in science more broadly. Specifically, a biologist might invoke physical ‘laws’ that, as physicists will happily point out, are often less ‘laws’ and more ‘context-dependent rules of thumb that work well up to a very specific limit and fail badly beyond it’. Here, we discuss some of the challenges of first-principles approaches in biology, as well as providing a path forward for what we hope represents best practice in both the interpretation and implementation of such approaches.

Next, we explore some of the common challenges associated with the use of first-principles approaches in biology using specific well-known examples before proposing some solutions. Note that we do not believe that these issues invalidate first-principles approaches in biology in general, nor even in these illustrative examples, but rather we highlight their risks and limitations so that these can be minimised in the future. Indeed, it is our fervent belief that first-principles approaches are powerful biological tools—that is why we seek to refine their deployment in biology.

Unaccounted-for physics or the challenge of *ceteris paribus* in biology

Biologists invoke rules based on established physical principles to explain observed phenomena, but these invocations can be based on physical principles that are oversimplified, incomplete, or inappropriate for that context. To illustrate this issue, we will use a seemingly simple and intuitive example: the relationships among cell size, cell surfaces, and resource uptake (Figure 1). The



Trends in Ecology & Evolution

Figure 1. The relationship between size and resource uptake in cells based on three very different, but equally valid first-principes approaches. For cells (orange circles) of a given size (described by radius, *r*), different physics will apply depending on the availability of resources (shown in blue) and the motion of the cell. The lefthand panel approximates the relationship between cell size and uptake when resources are saturating—a plausible but relatively rare condition—uptake scales with cell surface area (r^2). The centre panel approximates the relationship between cell size and uptake when resources are not saturating, and the cell depletes local resources such that diffusion limits uptake. Importantly, both cells and fluid must be completely still for this condition to apply and uptake scales with cell radius. The righthand panel shows perhaps the most biologically (and therefore physically) realistic condition for free living cells in nonsaturating resource conditions and larger cells swim/sink faster than smaller cells. Here, resource uptake scales with cell size at roughly 1.7. Using the physics represented on the left will usually overestimate resource uptake while using the model represented in the centre would overestimate the constraints larger cells face relative to smaller cells.

Glossary

Akaike Information Criterion (AIC): An information-theoretic metric used for model selection that balances goodness of fit with model complexity. Lower AIC values indicate better-supported models, with penalties applied for additional parameters to avoid overfitting.

Boundary Layers: Regions of fluid adjacent to surfaces where velocity or concentration gradients are steep. In aquatic systems, boundary layers around organisms or their exchange surfaces limit mass and heat transfer, affecting nutrient uptake, gas exchange, and diffusion rates.

Borges' Perfect Map: A conceptual metaphor derived from Jorge Luis Borges' story “On Exactitude in Science” describing a map that is as detailed as the territory it represents such that it is useless. In ecology, it highlights the trade-off between model realism and simplicity, emphasizing that overly detailed models may become impractical or uninformative.

Dynamic Energy Budget (DEB) Theory: A framework for describing how organisms acquire and allocate energy and matter to maintenance, growth, development, and reproduction over their life cycle. DEB models provide a unified description of physiological processes across species and environmental conditions. At the heart of DEB models is the assumption that resource access is limited by the surface areas of ‘blobs’ of reserve that change very specifically with organismal size.

Fick's First Law: A diffusion law stating that flux is proportional to the negative gradient of concentration, implying that substances move from regions of high to low concentration. Commonly used to model passive transport of gases or solutes towards biological surfaces. Fick's First Law is particularly relevant to cells that are not surrounded by saturating resource conditions.

Fick's Second Law: A time-dependent extension of diffusion theory describing how concentration changes through time as diffusion proceeds, predicting the rate at which gradients dissipate and equilibrium is approached. In biology, Fick's second law is often invoked to describe limits of rates of resource flux across biological surfaces.

First Principles: The use of physical theories (often described as ‘laws’) to develop theory and prediction while variation in the size of free-living unicells is astonishing; spanning a 10^{12} size range (for comparison, the smallest shrew to the largest whale spans only 10^8). Myriad theories of cell size begin with discussions of surface area-to-volume ratios (e.g., [3,9–11]): larger cells have less surface area relative to their mass, such that a simple application of **Fick's second law** results in relatively lower rates of resource transport in larger cells [12,13]. A corollary of this supposition is that changing the ratio of resource supply to demand will alter selection on cell size [14] (e.g., higher temperatures should favour smaller cells because warmer temperatures increase demands relative to supply [9,15]). The use of simple geometry and physics seems reasonable at first glance—surface area does scale roughly with cell volume to the two-thirds power [16]. However, the physics of the relationship between cell size and transport rate are more complicated. If resources are completely saturating, then the flux of resources will indeed scale with cell size to the two-thirds power, as rates are limited by transport across the cell membrane [17]. But under the more biologically plausible scenario of scarce resources, fluxes will be limited by diffusion to the cell from the environment. This rate of diffusion scales, according to both **Fick's first** and second laws, with cell radius (or a similar associated length scale) rather than surface area [17,18]. Thus, first principles (geometry and Fickian physics) drive the mechanics of many theories of cell size, but the context determines which physics are relevant (Figure 1).

Theories should not seek to capture every possible nuance of the phenomenon they describe; otherwise they would be as useless as **Borges's perfect map** [19]. Rather, they seek to simplify and capture first-order responses, which requires assumptions, the most common being *ceteris paribus*—that all other things will be equal. Sometimes these assumptions are explicitly stated, but more often they are not. Assumptions of *ceteris paribus* are innocuous when the factors that are assumed to be equal show no covariance with our factors of interest. Even if these factors strongly influence the component we seek to predict, ignoring these factors will only reduce the fit between theoretical predictions and observations. However, when there are unincluded covariates of our trait of interest, assuming *ceteris paribus* will disconnect the theory from the phenomenon in question.

To return to our example of cell size, factors that covary with cell size and affect resource fluxes into cells, but are unaccounted for, will violate the strict assumptions of the associated physics. In their simplest form, cell size theories predict that resource fluxes scale with cell volume at an exponent of 0.33, but this is likely to be an underestimate. More complete theories recognise that cell swimming speeds and sinking rates covary with cell size, such that larger cells sink and swim typically faster than smaller cells [14,17]. Because **boundary layers** surrounding faster-moving cells are thinner than those surrounding slower cells, diffusion barriers are also diminished. For example, the resource fluxes into a 1000-μm diameter cell are 300 times greater when the cells are swimming relative to when they are immobile [17]. Thus, cell size theories that exclude the covariance between cell size and swimming/sinking speed will systematically overestimate boundary layers around larger cells and underestimate their resource fluxes [17,20]. In other words, some theories of cell size inadvertently disadvantage larger cells in terms of resource fluxes because they exclude some of the relevant biology and, thus, exclude the relevant physics (Figure 1).

Using an incomplete physical model can alter its biological implications: for example, models that exclude covariances between swimming speed and size predict that larger cells will be disfavoured under climate warming, mostly because of resource transport constraints [9,15]. Meanwhile, theories using more complex physics come to the opposite conclusion [21,22]. Overall, we would argue that, even for a seemingly simple trait like cell size, some of the theories we use to describe the physics governing that trait (and the traits that covary with it) are

diminishing reliance on empirically estimated parameters. In biology, first-principles approaches often invoke conservation laws (e.g. mass, energy) to explain organismal or ecosystem patterns or processes from a mechanistic perspective.

Gill Oxygen Limitation Theory (GOLT): A hypothesis proposing that oxygen supply, constrained by the scaling of gill surface area to body size, limits growth and maximum body size in water-breathing ectotherms. It predicts that as organisms grow, oxygen demand increases at a greater rate than supply, particularly under warming conditions.

Metabolic Theory of Ecology (MTE): A theoretical framework linking metabolic rate—constrained by body size and temperature—to ecological patterns and processes across multiple levels of organization. Different elements of MTE predict scaling relationships of metabolic rate, metazoan growth, and population dynamics.

Sensitivity Analyses: Procedures used to quantify how variation in model inputs or parameters influences outputs. They identify key drivers of model behaviour, assess robustness of predictions, and highlight parameters requiring precise estimation.

Stokes' Law: A physical law describing the drag force on a spherical particle moving through a viscous fluid at low Reynolds number, predicting that settling velocity scales with the square of particle radius and inversely with fluid viscosity. Widely applied to understand sedimentation, wind-driven dispersal, and plankton sinking rates.

incomplete. Thus, though first principles can simplify a biological problem, some simplifications may create too much dissonance between theory and reality.

Using incomplete physics in biology is not unique to theories of cell size. For example, some theories on how warming affects the size of water-breathing ectotherms seem similarly incomplete (Box 1). Our goal here is not to critique specific studies; rather, we want to illustrate that first-principles approaches do not guarantee the revelation of universal biological truths. Unaccounted-for covariances between factors can mar predictions to such an extent as to render them irrelevant. We acknowledge that parsimoniously describing the mechanics of complex biological systems will often require the assumption of *ceteris paribus*, but this assumption should be verified rigorously. All else is rarely equal in biology; instead, organisms have evolved in response to, and to compensate for, the physics they experience. *Ceteris paribus* might be a reasonable place to start in biology but may not be where to end. Ensuring that both the biology and physics are sufficiently complete remains an enduring challenge.

Physics as an accidental trojan horse for unscrutinised biological assumptions

Invoking first principles as drivers of biological phenomena can seem straightforward but often requires ancillary assumptions. We will illustrate the problem with a simple example from some of our own work. One of us was interested in how the size of photosynthesising cells affects their capacity to take up inorganic carbon [37]. We hypothesised that larger cells would be more transport-limited and invoked Fick's second law. We tested our hypothesis by evolving a 10-fold range of cell volumes and estimating the ratio of supply to demand for inorganic carbon across that size range. We were surprised to find that larger cells met their increased inorganic carbon demands despite their lower surface area-to-volume ratios [37]. These larger cells had 'evaded' the constraints of Fick's law by upregulating the density of transporters on their surface. In this simple example, we invoked (seemingly) robust first principles (surface area-to-volume, Fick's Law) to generate a prediction, but we unconsciously made additional, unstated assumptions. We had assumed the density of inorganic carbon transporters on the surface of cells was size-independent (despite theory to the contrary [14,38]), and we assumed that only surface transport rates limit photosynthesis (see [14] for an elegant counter example). In other words, we had inadvertently made cryptic assumptions when building our putatively first-principles model. Cryptic assumptions such as ours pervade first-principles approaches.

The **Metabolic Theory of Ecology (MTE)** has been influential in biology, touching on many facets of ecology, evolution, and biogeography [39]. MTE uses first-principles approaches to conclude that fractally branching distribution networks drive metabolism to scale with body mass with an exponent of 0.75 because that is the most efficient scaling for transporting resources [39]. Efficient scaling makes sense at first glance—minimising transport costs maximises the remaining amount of energy that can be allocated to fitness-enhancing processes such as reproduction. However, this theory implicitly assumes that the total energy intake will be unaffected by the efficiency of the transport network. In other words, an organism with an efficient transport network is assumed to have equivalent energy intake to an organism with an inefficient transport network, such that the former organism will have more energy to allocate to fitness-enhancing processes than the latter. But, as MTE makes clear, metabolism affects every aspect of the ecology and evolution of organisms. Consequently, substantive changes in the transport network, with its concomitant impacts on metabolism, could change an organism's entire biology, from energy intake to expenditure on fitness-enhancing functions [40,41]. In essence, MTE assumes that the scaling of life histories, the rate of energy use, and the pace of life have all evolved to maximise the efficiency of the transport network alone. Thus, a nominally first-principles argument about transport efficiencies makes a strong assumption about the

Box 1. Fick's laws and fish laws

Why do fish grow the way they do? Biologists have long recognised that the surface area of gills affects the rate of gas exchange and might therefore limit the maximum body sizes in fish. Attempts to couple painstaking estimates of gill surface area [23] to formal applications of Fick's law to gas exchange began at least 50 years ago. Most models focus on the relationship between maximum possible oxygen uptake rate of fish via their gills and their body size [24–27]. A prominent modern example is the **Gill Oxygen Limitation Theory (GOLT)** [28], which argues that the area of respiratory surfaces of water-breathing ectotherms scale with body size sublinearly, so as organisms grow, their metabolic demands will approach the limit of supply via these surfaces and growth will cease. Advocates of GOLT have argued '…GOLT presents a unifying theory—based solely upon first principles and their corollaries' [25]. GOLT and related theories [25,26] argue that oxygen limitation should occur at relatively smaller sizes when waters are warmer because of the effect of temperature on oxygen consumption. While these are putatively first-principles arguments, here we show that not all of the relevant first principles have always been applied, with major biological implications. To explore these issues, we will focus on perhaps the most prominent modern example, GOLT using the version described in [25].

GOLT is based on Fick's second law and uses a specific version of it to describe gas exchange rates, Q, according to the equation:

$$Q = \mathrm{d}P \cdot U \cdot G \cdot \mathrm{WBD}^{-1}$$

where dP represents the difference in partial pressure of oxygen between the water and blood, U is Krogh's diffusion 'constant', G is the surface area of the gills, and WBD is 'water capillary distance' [25]. GOLT assumes gas exchange in water is diffusion-limited, and according to this equation, the surface area of the gills limits the rate of oxygen intake [25]. Crucially, GOLT holds [28] that only G changes greatly with fish body size (which we have emphasised in the above equation in blue). Other theories of gas exchange in water breathers similarly focus on the surface area of gills alone [29].

Given these theories model how oxygen availability covaries with body size (and often, temperature), it is therefore important that first-principles approaches include all of the relevant factors that covary with body size (and temperature). Under the standard model of GOLT, only G varies with body size but the distance between the respiratory medium (i.e., water) and bloodstream also covaries with body size in two ways. First, the thickness of the gill tissue decreases with body mass such that larger water breathers have thinner gill membranes [30]. Second, the thickness of the boundary layer, which acts as the major barrier to oxygen transfer in water breathers [31], also covaries with fish body size [24,32,33]. Larger fish tend to swim faster than smaller fish [33] such that, on average, ventilation rates are greater through the gills of larger fish relative to smaller [31]. Consequently, boundary layers will be thinner in larger fish relative to smaller fish (and thinner in active fish relative to inactive fish) [32,34]. Thus, the WBD term (now divided into its two components, the gill tissue and the water boundary layer) also depends on body size such that:

$$Q = \mathrm{d}P \cdot U \cdot G \cdot (WBD_{Gills} + WBD_{Water})^{-1}$$

If the effects of body size on the water-capillary distance are excluded from calculations of oxygen uptake rates (such as they are in GOLT), then the capacity for larger fish to take up oxygen will be systematically underestimated. Indeed those models that allow WBD to change with body size come to opposite conclusion to GOLT: they predict that larger fish are at 'less' risk of oxygen limitation than smaller fish [31,32].

Oxygen-exchange theories focused on water breathers often make predictions about how oxygen limitation might change with temperature [35], so it is important to incorporate the physical effects of temperature in these theories: some do, others do not. For example, although GOLT precludes the effect of temperature on U: within the range of temperatures typically experienced by water-breathers, Krogh's diffusion coefficient increases such that diffusion rates increase with warmer temperatures [35,36]. Similarly, warmer fluids are less viscous with thinner boundary layers such that excluding temperature effects will overestimate diffusive barriers in warmer water. Accordingly, we have color-coded these terms (Krogh's coefficient in red; the thickness of the boundary layer in purple) to show that both should be allowed to change with temperature.

$$Q = \mathrm{d}P \cdot U \cdot G \cdot (WBD_{Gills} + WBD_{Water})^{-1}$$

Standard GOLT precludes these temperature effects on gas exchange; instead, it assumes that metabolic demands increase without any changes to the physics of oxygen supply. Unsurprisingly then, GOLT underestimates rates of oxygen transport in warmer waters and overestimates supply constraints on larger fish. The above-mentioned example emphasises the importance of including all the relevant physics when seeking to apply first-principles approaches.

ultimate evolutionary drivers of whole-organism metabolism. To the credit of MTE's progenitors, they clearly stated their assumption from the outset [39], but it never received adequate attention, partly because it is challenging to test empirically. Some have delineated approaches such as MTE as 'Newtonian' while describing other approaches as 'Darwinian' [42]; ironically, we would argue that MTE has a very strong Darwinian assumption at its core. Much of the debate around MTE has focused on the precise value of the metabolic exponent [43–46], rather than testing its assumptions (but see [47]). We argue later that this focus on model fit to predictions rather than on model assumptions pervades first-principles approaches in biology.

The **Dynamic Energy Budget theory (DEB)** also makes strong biological assumptions at its core that are untested [48]. DEB seeks to explain trajectories of growth and physiological traits and is routinely described as a first-principles approach [49]. DEB assumes that an organism's access to nutritive material (assimilation) is limited by the surface area of 'blobs' of reserves, and that these blobs increase in size with body size so that access to the reserve scales sublinearly with the volume of the organism [48,49]. This sublinear scaling is critical as it drives decreases in growth rates as organisms increase in size. Importantly, were the shape or number of reserves to change with organismal size, then sublinear scaling may no longer apply. For example, if the blobs were cylindrical and only elongated with increasing size, or if the number of blobs increased with body size, the scaling of reserve access would be 1. Instead, DEB typically assumes that blob shape and number remain constant with size, which they describe as 'isomorphy', arguing that '…isomorphy is strongly selected at the cellular level to minimise regulatory complexity as size changes' [49]. This conjecture—that individuals with more isomorphous blobs have a fitness advantage—is untested and may be untestable [50]. Thus, even though DEB is argued to have a strong mechanistic underpinning, it actually rests on a decidedly nonfirst-principles premise. Again, most discussion of DEB has focused not on this assumption but instead on the capacity of the theory to predict observations—its proponents focus on the strong fit between DEB predictions and observations as evidence of its explanatory capacity [49]. Next, we explore the issue with predictions-as-evidence more generally.

The prediction-explanation fallacy or the 'Clever Hans' problem

Clever Hans was a horse that tapped his hoof to correctly answer complex mathematical problems posed by humans. In reality, Hans was not doing complex calculations but instead reacting to the inadvertent prompts of his interlocutors. The Clever Hans effect is a good example of confusing a process that 'resembles' reality with one that 'explains' reality. Because Hans got the right answer, some took this as proof that Hans understood the answer. Concluding that a theory explains a biological phenomenon just because its predictions match observations is committing the same logical error.

Across many of the examples we have discussed here, predictions from first-principles theories matched patterns observed in nature. For example, organisms tend to be smaller in warmer temperatures, matching predictions based on surface-area constraints; growth slows down as organisms increase in size, as predicted by numerous theories of growth [6], and metabolic scaling is around 0.75, as predicted by several theories of scaling [11,51,52]. The close matching of data to theory is often invoked as powerful evidence that this theory is sound and, by extension, its assumptions are supported. Some of us are guilty of this type of reasoning—we recently argued that a life-history optimisation model of metabolic scaling does a good job of recovering metabolic scaling observed in nature [52].

The fit between the theory and observations leads many to conclude that a theory has robust explanatory power. To do so is to succumb to the prediction-explanation fallacy [53], which assumes that because a theory's predictions 'match' the data, the theory 'explains' the data. The prediction-explanation fallacy is a growing problem in Machine Learning [53], but it can manifest anywhere, particularly when a complex model is used to explore nonlinear relationships [6,54]. For a theory to have explanatory power, it is necessary but insufficient that it fit observed patterns.

The distinction between prediction and explanation in biology is important. Biological theories are meant to provide insights into why a biological process occurs; they are most useful when they explain these processes. Complex biological patterns often require complicated, nonlinear models—so, unfortunately, multiple approaches or theories can arrive at the same predictions for different reasons [51]. For example, multiple theories predict metazoan growth trajectories equally well [6,52]. These good fits are not coincidental; each theory was constructed with the goal of fitting observations, and none would have progressed if they had failed at the first hurdle of prediction. In other words, each of these models is potentially a Clever Hans—the product of the humans that made them.

Competing theories cannot be compared based on predictive success alone if one wishes to avoid the prediction-explanation fallacy. Yet many evaluations of theories tend to focus on fit, rather than assumptions. For example, intense debates followed the publication of many of the theories we have discussed here; common to all these debates was whether the model in question predicted observations. Meanwhile, as we have shown, some crucial assumptions underpinning these theories went relatively unscrutinised. We would argue that the veracity and testability of these assumptions are the most reliable ways to compare competing theories with analogous predictive power. We suspect that first-principles approaches are less scrutinised because they present as physical truths rather than testable assumptions that require scrutiny.

Concluding remarks and proposed guidelines for using first-principles approaches in biology

We have highlighted critiques of first-principles approaches to biology, not to dismiss first-principles approaches as universally unhelpful, but rather to help the field progress in the application of these approaches (see Outstanding questions). We acknowledge that physical constraints play a fundamental role in shaping many aspects of biology and that we need theories and mathematical frameworks capable of robust predictions. We advocate for the application of first-principles approaches that carefully consider the underlying (often unstated) assumptions. Next, we suggest potential ways to avoid or minimise recurrent problems with first-principles approaches (and also recommend [54]).

Completeness (including the relevant covariances)

Although models should be partial abstractions, theoreticians should ensure that the relevant processes are included. Specifically, those elements that covary with the biological process or trait of interest should be either explicitly included or simplified accurately. For example, some theories of gas exchange in fishes only consider the role of gill surface area and conclude that bigger fish are more constrained in their oxygen uptake [23,25]. Meanwhile, more complete theories consider gill surface area as well as gill tissue thickness, swimming speed, and ventilation frequencies because not only do these factors matter, but they also covary with body size—the biological trait of interest [24,31]. One could also include the resistance of gills to water flow, counter-current exchange ratios, and the synchronisation of pulsatile flow to make the physics more realistic. But we would argue that these components are unnecessary complications if they do not covary with the trait of interest. Theories should seek to strike the right balance between complexity and necessity—whether processes covary with the traits/functions of interest seems to us to be a good criterion for inclusion.

Outstanding questions

While each of the theories we have discussed earlier requires further refinement and raises as many questions as they have answered, rather than exploring them, we would like to use this section to highlight the questions biologists should ask themselves whenever they encounter or build their own first-principles approaches.

Do the physics invoked for a biological phenomenon robustly apply to the context, or are they merely intuitive? This is perhaps the most difficult question for biologists to answer, especially if they lack training in the relevant physics. In our experience, however, literature searches routinely reveal a rich theory base for any number of biological processes. For example, with a little reading, it swiftly becomes apparent that at small scales, surface areas alone rarely limit resource exchange; instead, the boundary layers around the surfaces represent far more formidable barriers.

What drives the theory? Are the associated biological assumptions reasonable and well-accepted? Some theories (e.g., Dynamic Energy Budget theory) have esoteric assumptions about the underlying biology that would seem incongruous outside of a first-principles context. Similarly, theories with lots of tunable parameters will inevitably fit better than those with fewer parameters—again, fit alone may not be an adequate criterion, and information-theoretic approaches such as the Akaike Information Criterion might help qualify or contextualise relative fits.

Are the assumptions (biological or physical) that are made testable? If not, how much do they matter? We propose that sensitivity analyses are key here. Specifically, it is often possible to mathematically test the sensitivity of a theory to an underlying assumption. Some assumptions may not (yet) be testable—that is not necessarily disqualifying for a theory, but as a rule of thumb, if the theory entirely depends on an untestable assumption, it is probably less robust.

Clarity (what is assumed to matter, what is assumed not to matter)

Models invoking first principles are typically focused on one particular aspect of the system, such that many ancillary assumptions are implicit rather than explicit. We suggest that, in addition to listing assumptions about what is 'included' in a theory, it might also be useful to list assumptions about what is 'excluded'. For example, in our own work [37], it would have been better to explicitly state that we assumed resource uptake was transport-limited and that we assumed transporter density does not change with cell size. **Sensitivity analyses** are a useful tool for determining whether such exclusions are likely to matter (for some exemplars of this approach, see [17,22]).

Congruence (the right physics for the right scale)

The physics that are appropriate for one scale (be it spatial, temporal, or a biological scale of organisation) may be inappropriate for another scale. For example, life at low Reynolds numbers can be deeply nonintuitive, such that simple assumptions about how resources move between organisms and their environment can be wrong. Researchers should think carefully about the scale at which the processes of interest work. There are excellent examples showing how to robustly apply different physics across different scales, with varying degrees of complexity that could be emulated in a range of contexts (e.g., [8,17,20]). For example, Kiørboe [17] provides an outstanding demonstration of how different physics dominate diffusion rates depending on size and speed.

Moving beyond fit—increasing scrutiny on assumptions

Just because a theory purports to be based on first principles or invokes impressive-sounding physical laws does not make it sacrosanct. Oftentimes, the invocation of a physical principle involves a series of assumptions and choices, and biologists should recognise and scrutinise these just as they would in any other theoretical approach. Where competing theories successfully predict observations with equal success, hopefully, the assumptions underlying them differ sufficiently such that they might provide a distinguishing test. Ideally, competing theories should have very different, testable assumptions or predictions so that the field can distinguish among them. Ultimately, theories are most informative when they provide an expansion of knowledge outside the range of current observations.

Acknowledgments

The authors wish to thank the editor and reviewers for their very helpful comments, which improved the manuscript. The authors are also grateful for conversations with Hayley Cameron, Tim Coulson, Troy Day, and Marc Mangel, which have helped shape their thinking on these issues. Thanks to Nick Stone and Sarah Prendergast for their assistance with manuscript preparation.

Declaration of interests

The authors declare no competing interests.

References

1. Thompson, D.W. (1917) *On Growth and Form*, Cambridge University Press
2. Sarrus, F. (1839) Rapport sur une mémoire adressé á l'Académic Royale de Médecine. *Bull. Mem. Acad. R. Med. Paris* 3, 177
3. Czarnoleski, M. and Verberk, W.C.E.P. (2025) Cell size matters: a unifying theory across the tree of life. *Trends Ecol. Evol.* 40, 1113–1125
4. Behrenfeld, M.J. and Bisson, K.M. (2024) Neutral theory and plankton biodiversity. *Annu. Rev. Mar. Sci.* 16, 283–305
5. Otto, S.P. and Day, T. (2007) *A Biologist's Guide to Mathematical Modelling in Ecology and Evolution*, Princeton University Press
6. Marshall, D.J. and White, C.R. (2019) Have we outgrown the existing models of growth? *Trends Ecol. Evol.* 34, 102–111
7. van Hout, R. and Katz, J. (2004) A method for measuring the density of irregularly shaped biological aerosols such as pollen. *J. Aerosol Sci.* 35, 1369–1384
8. Laurenceau-Cornec, E.C. *et al.* (2020) New guidelines for the application of Stokes' models to the sinking velocity of marine aggregates. *Limnol. Oceanogr.* 65, 1264–1285
9. Leles, S.G. and Levine, N.M. (2023) Mechanistic constraints on the trade-off between photosynthesis and respiration in response to warming. *Sci. Adv.* 9, eadh8043

10. Hillebrand, H. *et al.* (2022) Cell size as driver and sentinel of phytoplankton community structure and functioning. *Funct. Ecol.* 36, 276–293
11. Glazier, D.S. (2010) A unifying explanation for diverse metabolic scaling in animals and plants. *Biol. Rev. Camb. Philos. Soc.* 85, 111–138
12. Westfall, C.S. and Levin, P.A. (2017) Bacterial cell size: multifactorial and multifaceted. *Ann. Rev. Microbiol.* 71, 499–517
13. Marañón, E. (2015) Cell size as a key determinant of phytoplankton metabolism and community structure. *Annu. Rev. Mar. Sci.* 7, 241–264
14. Yoshiyama, K. and Klausmeier, C.A. (2008) Optimal cell size for resource uptake in fluids: a new facet of resource competition. *Am. Nat.* 171, 59–70
15. Deutsch, C. *et al.* (2022) Impact of warming on aquatic body sizes explained by metabolic scaling from microbes to macrofauna. *Proc. Natl. Acad. Sci. U. S. A.* 119, e2201345119
16. Secaira-Morocho, H. *et al.* (2024) An evolutionary optimum amid moderate heritability in prokaryotic cell size. *Cell Rep.* 43, 114268
17. Kiørboe, T. (1993) Turbulence, phytoplankton cell size, and the structure of pelagic food webs. In *Advances in Marine Biology* (29) (Blaxter, J.H.S. and Southward, A.J., eds), pp. 1–72, Academic Press
18. Berg, H.C. and Purcell, E.M. (1977) Physics of chemoreception. *Biophys. J.* 20, 193–219
19. Borges, J.L. (1935) *A Universal History of Infamy*, Penguin Modern Classics
20. Karp-Boss, L. *et al.* (1996) Nutrient fluxes to planktonic osmotrophs in the presence of fluid motion. *Oceanogr. Mar. Biol.* 34, 71–107
21. Landy, J. (2014) Shape-dependent bounds on cell growth rates. *Europhys. Lett.* 105, 68002
22. Perna, A. *et al.* (2025) Energy allocation explains how protozoan phenotypic traits adapt to temperature and nutrient supply. *bioRxiv* https://doi.org/10.1101/2023.07.25.550219
23. Rubalcaba, J.G. *et al.* (2020) Oxygen limitation may affect the temperature and size dependence of metabolism in aquatic ectotherms. *Proc. Natl. Acad. Sci. U. S. A.* 117, 31963–31968
24. Hills, B.A. and Hughes, G.M. (1970) A dimensional analysis of oxygen transfer in the fish gill. *Respir. Physiol.* 9, 126–140
25. Pauly, D. (2021) The gill-oxygen limitation theory (GOLT) and its critics. *Sci. Adv.* 7, eabc6050
26. 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, 881–893
27. Atkinson, D. *et al.* (2006) From cells to colonies: at what levels of body organization does the 'temperature-size rule' apply? *Evol. Dev.* 8, 202–214
28. Pauly, D. (2010) *Gasping Fish and Panting Squids: Oxygen, Temperature and the Growth of Water-breathing Animals*, International Ecology Institute
29. Jacoby, D.M.P. *et al.* (2015) Is the scaling of swim speed in sharks driven by metabolism? *Biol. Lett.* 11, 20150781
30. Gillooly, J.F. *et al.* (2016) Body mass scaling of passive oxygen diffusion in endotherms and ectotherms. *Proc. Natl. Acad. Sci. U. S. A.* 113, 5340–5345
31. Hughes, G.M. (1966) The dimensions of fish gills in relation to their function. *J. Exp. Biol.* 45, 177–195
32. Luo, Y. *et al.* (2020) Ventilation frequency reveals the roles of exchange surface areas in metabolic scaling. *Physiol. Biochem. Zool.* 93, 13–22
33. Hirt, M.R. *et al.* (2017) A general scaling law reveals why the largest animals are not the fastest. *Nat. Ecol. Evol.* 1, 1116–1122
34. Piiper, J. and Scheid, P. (1984) Model analysis of gas transfer in fish gills. In *Fish Physiology* (10) (Hoar, W.S. and Randall, D.J., eds), pp. 229–262, Academic Press
35. Cheung, W.W.L. *et al.* (2013) Shrinking of fishes exacerbates impacts of global ocean changes on marine ecosystems. *Nat. Clim. Chang.* 3, 254–258
36. Steyn-Ross, D.A. *et al.* (2023) Determination of Krogh coefficient for oxygen consumption measurement from thin slices of rodent cortical tissue using a Fick's law model of diffusion. *Int. J. Mol. Sci.* 24, 6450
37. Malerba, M.E. *et al.* (2020) Cell size influences inorganic carbon acquisition in artificially selected phytoplankton. *New Phytol.* 229, 2647–2659
38. Andersen, K.H. and Visser, A.W. (2023) From cell size and first principles to structure and function of unicellular plankton communities. *Prog. Oceanogr.* 213, 102995
39. West, G.B. *et al.* (1997) A general model for the origin of allometric scaling laws in biology. *Science* 276, 122–126
40. Houston, A.I. *et al.* (1993) General results concerning the trade-off between gaining energy and avoiding predation. *Philos. Trans. R Soc. Lond. B Biol. Sci.* 341, 375–397
41. Schuster, L. *et al.* (2021) Metabolism drives demography in an experimental field test. *Proc. Natl. Acad. Sci. U. S. A.* 118, e2104942118
42. Glazier, D.S. (2022) Variable metabolic scaling breaks the law: from 'Newtonian' to 'Darwinian' approaches. *Proc. Biol. Sci.* 289, 20221605
43. White, C.R. and Marshall, D.J. (2023) How and why does metabolism scale with body mass? *Physiology* 38, 266–274
44. Dodds, P.S. *et al.* (2001) Re-examination of the "3/4-law" of metabolism. *J. Theor. Biol.* 209, 9–27
45. Savage, V.M. *et al.* (2004) Effects of body size and temperature on population growth. *Am. Nat.* 163, 429–441
46. Glazier, D.S. (2005) Beyond the '3/4-power law': variation in the intra-and interspecific scaling of metabolic rate in animals. *Biol. Rev. Camb. Philos. Soc.* 80, 611–662
47. Price, C.A. *et al.* (2012) Testing the metabolic theory of ecology. *Ecol. Lett.* 15, 1465–1474
48. Kooijman, S.A.L.M. (2010) *Dynamic Energy Budget Theory for Metabolic Organisation*, Cambridge University Press
49. Kearney, M.R. (2020) What is the status of metabolic theory one century after Pütter invented the von Bertalanffy growth curve? *Biol. Rev. Camb. Philos. Soc.* 96, 557–575
50. Kearney, M.R. and White, C.R. (2012) Testing metabolic theories. *Am. Nat.* 180, 546–565
51. White, C.R. and Marshall, D.J. (2023) Optimisation and constraint: explaining metabolic patterns in biology. *J. Exp. Biol.* 226, jeb245426
52. White, C.R. *et al.* (2022) Metabolic scaling is the product of life-history optimization. *Science* 377, 834–839
53. Del Giudice, M. (2024) The prediction-explanation fallacy: a pervasive problem in scientific applications of machine learning. *Methodology (Gott)* 20, 22–46
54. Berro, J. (2018) "Essentially, all models are wrong, but some are useful"—a cross-disciplinary agenda for building useful models in cell biology and biophysics. *Biophys. Rev.* 10, 1637–1647