We Have Never Been Hosts: Microbial Niche Weaving and the Evolution of Multicellular Life

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Abstract

Microorganisms are commonly understood as associates of multicellular hosts: organisms that first exist as biological individuals and are subsequently colonized by microbial communities. Here, we challenge this temporal and ontological ordering by distinguishing microbial presence from microbial influence. Microbial activity can shape biological processes without persistent physical association, through metabolites, environmental transformations, material effects, and ecological processes whose consequences extend across space, time, and generations. We argue that multicellular life evolved and continues to evolve within this longer history of microbial influence. To capture this relational history, we introduce the concept of microbial niche weaving. Drawing on basket weaving, the metaphor emphasizes how relatively stable biological forms emerge through the interlacing of microbial, macrobial, and abiotic relations, without presupposing a pre-existing host, a single weaver, or a predetermined form. We further propose a principle of recursive microbial influence: biological structures and processes shaped by earlier microbial activity can themselves become conditions through which subsequent microbial influence is organized. Examples spanning multicellularity, development, circulation, reproduction, and terrestrialization illustrate how this perspective reframes major evolutionary transitions as reorganizations of pre-existing ecological relations rather than solely as transitions in individuality. Niche weaving does not replace organisms, individuality, symbiosis, or selection as biological concepts, but it changes their explanatory relationship to ecological processes. Hosthood, on this account, is a derived ecological configuration rather than the starting point of host–microbe relations. Multicellular organisms are relatively stabilized patterns emerging within an evolutionary history of microbial influence: in this sense, we have never been hosts.

"It is unselfconscious privilege that allows us to fantasize – counterfactually – that we each survive alone." (Tsing 2015, *The Mushroom at the End of the World*, p. 29)

Introduction

Just over a decade ago, Gilbert et al. summarized the holobiont perspective with the declaration that "we have never been individuals"[1]. Much of the debate that followed has centered on whether holobionts should count as evolutionary individuals or units of selection[2–12]. Yet proponents and critics have generally retained the same underlying ontology: microbiomes (or microbial communities) on one side and multicellular eukaryotes – their "hosts" – on the other. As a result, the metaphor according to which multicellular organisms (macrobes) are "hosts" is used as a matrix upon which research is structured.

One of the main objectives of this paper is to offer another metaphor according to which multicellular organisms come to be conceived as niches woven (rather than constructed) by complex interaction patterns. Whereas the host metaphor names an entity and then asks how microbes interact with it, niche weaving begins instead with the unbroken processes through which microbial communities, macrobes, and their environments acquire their historically contingent forms.

We invoke weaving here as a process metaphor rather than a design metaphor. There is no single weaver, blueprint, or completed form: biological organization emerges through an ongoing and unbroken history in which each configuration inherits conditions shaped by what came before while altering the conditions from which subsequent configurations emerge. In this messy context, some threads can be followed analytically and help us understand how patterns of interactions, e.g., holobionts, arise. This, as we hope to show, provides a novel and fertile perspective, to be applied

to a wide variety of biological systems, from the biochemical origins of life to the Earth System. Our argument, in this paper, is nonetheless centered on holobionts.

Indeed, we wish to challenge the aptness of the "host" metaphor as a starting point for understanding holobionts. We propose that multicellular eukaryotes are better understood as niches evolutionarily woven by microbial communities rather than as autonomous organisms that first arise and are subsequently colonized.

In the case of holobiosis, this reframing rests on disentangling a common conflation: the absence of microorganisms is often assumed to entail the absence of microbial influence. Historical definitions of symbiosis, as well as the observational traditions of microscopy, culture, and genomics, have privileged physical co-localization. Metabolic and ecological interactions, however, can extend across tissues, developmental stages, generations, and environmental space. Once microbial influence rather than microbial presence becomes the relevant object of inquiry, the host-first narrative becomes less compelling. Microbial communities appear not simply as occupants of preexisting multicellular bodies, but as participants in the weaving of the evolutionary, developmental, physiological, and ecological conditions from which those bodies emerge.

We develop this argument primarily from animal evolution, with particular attention to placental mammals, but extend it to plants and to broader questions of biological individuality and major evolutionary transitions. The resulting framework shifts explanatory emphasis from colonization to niche weaving, from taxonomic presence to ecological relationships, and from increasingly autonomous individuals to increasingly organized and spatially integrated networks of interaction.

Historical contingency and the host-first narrative

The persistence of the host-and-colonization narrative may itself be a consequence of historical contingency. Evolution by natural selection was established before microbiology revealed the ubiquity and functional importance of microorganisms. Its conceptual architecture was therefore built largely around visible multicellular organisms conceived as discrete evolutionary individuals. The microbiome revolution has transformed biology, but its findings have often been assimilated into this pre-existing framework by adding microbial partners to already constituted organisms rather than reconsidering the evolutionary and ecological processes through which those organisms themselves emerged. One result of this rationale is the focus of the major transitions in evolution framework on transitions in individuality, with the assumption being that discrete entities emerge before interacting with their environment.

From a historical-epistemological perspective, this continuity is unsurprising. Scientific concepts tend to inherit assumptions from the conditions under which they were formed, even when subsequent observations reveal dimensions of nature that those concepts did not originally accommodate. In Gaston Bachelard's terms, such inherited commitments can become epistemological obstacles: progress may require not only the accumulation of new facts, but a reorganization of the conceptual frameworks through which those facts are interpreted.

The conceptual order of discovery can therefore be mistaken for the causal order of evolution. Animals became objects of biological study long before microorganisms, while the pervasive roles of microbial processes in development, physiology, immunity, behaviour, and evolution became apparent only after organism-centred evolutionary narratives were already deeply established. The

historical sequence through which biological knowledge developed—organisms first, microbes later—can consequently be mistaken for the evolutionary sequence itself. The host-and-colonization narrative (i.e., a host pre-exists its interactions with symbionts) is an embodiment of this.

Microbiology now provides grounds for reorganizing that narrative. Microorganisms are increasingly understood not merely as discrete cells or genomes, but as participants in and constructors of extensive metabolic and ecological networks. Central to our argument is the concept of microbial influence, which we define as any causal effect of microbial activity, whether mediated by microbial cells themselves, their products, transformations of the physicochemical environment, interactions with other organisms, or the downstream consequences of previous microbial activity embodied in biological and ecological systems. Microbial influence therefore need not require the immediate physical presence of the microorganisms from which it ultimately derives. Microbial influence can extend beyond the microorganisms themselves in both space and time. This concept emphasizes the reach of microbial activity beyond direct or persistent physical association. For instance, in marine environments, free-swimming larvae of diverse animals use cues produced by environmental bacteria to guide settlement and trigger metamorphosis, allowing microbial activity associated with the seafloor to shape the development and life-history transitions of animals that do not maintain stable associations with those bacteria[13]. The biological reach of a microbial community therefore need not coincide with the spatial boundaries of the community itself.

The distinction between microbial presence and microbial influence changes how the evolutionary sequence itself can be reconstructed. Given the tight integration of microbes in the functioning and

development of multicellular organisms, we should also assume that they were part of the conditions under which multicellular organization emerged. Rather than positing that autonomous multicellular organisms arose first and were subsequently colonized, we propose that they evolved within environments already transformed by microbial activity, becoming themselves components of and contributors to those continuously evolving environments. The relationship is therefore not adequately represented as a linear sequence from organism to host to colonization. It is best represented as the weaving of interrelations, with patterns changing across time.

This reverses the usual explanatory sequence upon which the holobiont rationale rests. Multicellular organisms cannot simply be starting points of "host"–microbe relationships onto which microorganisms were subsequently added. They are themselves evolutionary outcomes of a much older history of ecological interactions and environmental transformations. The chronology of scientific discovery should therefore not be mistaken for the causal chronology of evolution.

Symbiosis, physical association, and observational bias

The requirement for physical association is deeply embedded in the history of symbiosis. Albert Bernhard Frank's 1877 formulation referred to two species living on or in one another, and Anton de Bary's 1878 account emphasized intimate association[14,15]. These definitions emerged from work on lichens and plants, and established a durable framework for thinking about inter-organismal relationships. Nowadays, the concept of symbiosis has become narrowed and blurred. In its broadest biological sense, symbionts could represent parasites, mutualists or commensals. However, most often, they have become synonymous with beneficial microbes present in or on a given host.

As microbiology developed, the concept of the host acquired additional importance through host–pathogen research and was later extended to host–microbe symbioses. Across these contexts, the host is typically treated as the larger, already constituted organism (the “macrobe”), in or on which microorganisms reside.

The same emphasis on physical presence is reflected in the major observational technologies of microbiology. Microscopy revealed microorganisms that could be directly visualized; culture enabled their isolation and classification; genomics vastly expanded the range of organisms that could be detected without cultivation. Yet genomics largely preserved the earlier conceptual emphasis by cataloguing organisms or microbial DNA physically present in a sample. Even when DNA is extracellular or transient, detection is commonly translated into a statement about microbial presence.

Metabolomics and other functional approaches expose a different dimension of microbial life, which emphasizes that symbiotic relationships between microbes and macrobes can occur beyond physical co-occurrence. These approaches reveal interactions that can persist across spatial, temporal, and taxonomic boundaries, including effects exerted far from the cells that generated them (e.g., microbial metabolites produced in the gut influencing the central nervous system). The shift from microscopy and culture to genomics therefore expanded the census of microbial communities without necessarily dissolving the assumption that biologically relevant association requires co-localization. A focus on metabolites, environmental modification, and ecological process makes that assumption harder to sustain, while fostering a more comprehensive view of symbiotic interactions.

Physical association is important, and so is the rationale that ties microscopy, culture, and genomics with symbiosis. These methods reveal essential aspects of the material organization of microbial communities. The window they provide into the realm of biological interactions, however, is limited: physical co-localization is only one form of biological relation. Treating it as the defining criterion of symbiosis obscures microbial influences that operate without direct or continuous contact and thus sustains the host metaphor.

Absence of microbes is different from absence of microbial influence

The distinction between the absence of microbes and the absence of microbial influence is especially clear in animals whose life cycles have traditionally been divided into symbiotic and aposymbiotic phases. Some animals maintain intimate microbial associations throughout development, including vertically transmitted endosymbioses. Other macrobes, by contrast, have often been described as passing through an aposymbiotic phase because microorganisms are assumed to be physically absent in fetal tissues[16,17]. The use of the term "aposymbiotic" to characterize the fetal phase rests on equating the absence of microbes with the absence of symbiosis (the prefix "apo" meaning "without"). Yet microbial influence persists throughout this phase, as it does in other phases or spaces considered "sterile," calling into question whether the absence of microorganisms can meaningfully be described as the absence of symbiosis.

Evidence from placental development challenges the equivalence between absence of microbes and absence of microbial influence. In placental mammals, the maternal microbiome influences fetal and placental development and offspring phenotype before birth. Because the placenta is a fetal

197 organ, microbial effects on placental angiogenesis challenge a simple description of fetal
198 development as aposymbiotic[17]. The extensive use of the vascular system during mammalian
199 pregnancy is particularly important in this respect: maternal microbial metabolites enter the
200 maternal circulation and influence placental and fetal development without requiring
201 microorganisms themselves to inhabit fetal tissues. Whether microorganisms are physically present
202 in a particular fetal compartment is therefore distinct from whether development in that
203 compartment occurs under microbial influence. Given the placenta's central role in fetal
204 development, microbial contributions to its development constitute a biologically consequential
205 relationship that extends symbiosis beyond the physical presence of microorganisms, rendering the
206 fetal phase fundamentally symbiotic.

207

208 Placental mammals, however, represent only one reproductive architecture. Rather than expecting
209 microbial influence to operate through the same mechanism across animals, variation in
210 reproductive mode can itself be understood as variation in the spatial and physiological routes
211 through which microbial influence reaches developing organisms. Oviparity provides a particularly
212 instructive contrast because embryos generally develop from resources provisioned within the egg
213 and lack the continuous maternal vascular connection characteristic of placental gestation. Yet
214 microbial influence persists despite this physical separation from maternal tissues.

215

216 In zebrafish, for example, the chorion largely restricts direct contact between microbial cells and the
217 embryo. Nonetheless, experimental manipulation of the surrounding aquatic microbiome alters
218 embryonic transcriptomic, proteomic and metabolomic programs before hatching, including
219 pathways involved in energy metabolism and neurodevelopment[18]. The authors conclude that,

220 although the chorion provides a physical barrier to microorganisms, it is permeable to microbial
221 communication from the surrounding environment. More broadly, egg-associated microorganisms
222 are known across diverse animal taxa and can contribute to vertical symbiont transmission,
223 protection against pathogens and biofouling, and, in some systems, successful development[19].
224
225 These observations suggest that animals as diverse as oviparae and mammals rely on microbial
226 influence throughout their life cycles, including during phases commonly assumed to be
227 aposymbiotic. The most economical evolutionary explanation is therefore to assume that
228 differences in their reproductive architectures result from a diversification through which
229 longstanding microbial influence is spatially reorganized. In placental mammals, microbial products
230 are conveyed across maternal physiological and vascular networks; in externally developing aquatic
231 eggs, environmental microbial signals act across embryonic barriers; in other oviparous systems,
232 egg-associated microorganisms or their products may provide additional routes of interaction. Some
233 of these mechanisms are experimentally established, whereas others will require further testing.
234
235 The diversity of reproductive architectures and respective pathways for microbial influence suggests
236 that the evolutionary question is not whether microbial influence is switched on or off by a particular
237 mode of reproduction, but how that influence is redistributed and mediated across changing
238 reproductive architectures. Comparative studies across reproductive strategies will be required to
239 establish how broadly particular mechanisms operate. The available evidence already cautions,
240 however, against assuming that developmental stages lacking resident microorganisms are
241 genuinely aposymbiotic. More fundamentally, it raises the question of whether any animal
242 developmental stage can be considered independent of microbial influence.

Microbial metabolites are thus adequately described as multi-kingdom intermediates[20]: molecules produced by particular cells whose biological effects extend beyond those cells, across tissues and often across kingdoms. This reframes the proposition that "not all animals need a microbiome"[21]. If "microbiome" is defined strictly as a physically resident community, the statement may be defensible for some species or life stages. If, however, the relevant question is whether animal evolution and development have unfolded, and continue to proceed, independently of microbial influence, the statement no longer addresses the relevant biological question.

From hosts to niches: microbial niche weaving

All of the above suggests that the alleged guests of macrobes do much more than enter an already constituted biological space. They actively contribute to their constitution. In this section, we want to bridge the developmental perspective described above with an evolutionary reading of the symbiotic relationship between microbes and macrobes, which extends beyond direct physical co-occurrence. From that evolutionary perspective, the spaces, interfaces, transport systems, and developmental processes through which microbial influence now operates are recast as products of microbial niche weaving.

In ecology and evolutionary biology, a niche denotes the subset of environmental conditions and interactions relevant to the persistence and dynamics of a focal organism, population, or species[22,23]. Niche construction emphasizes that organisms are more than passive recipients of environmental conditions: they modify those conditions, thereby altering ecological interactions

and selective pressures acting on themselves and others[24–27]. Organism and environment are linked by feedback rather than by a one-way relation. We suggest emphasizing niche weaving instead of niche construction because the latter may suggest that the construct can stand on its own once construction is complete. In contrast, the microbial–macrobial interactions examined here suggest a structure whose form and stability emerge from the interlacing of its components and may unravel when constituent strands are removed.

This has implications for the metaphor of the "host." Hosthood is better understood as a derived ecological configuration, the outcome of a much older recursive history of interactions (see next section), rather than as the ontological starting point of the relationship. Multicellular organisms can thus be understood as historically layered ecological weavings, in which past microbial influence has contributed to structural spokes, such as the circulatory system, that can in turn channel subsequent microbial influence, for example by carrying microbial metabolites across the organism. We therefore propose a different metaphor: multicellular organisms are patterns woven through interactions among microbial, macrobial, and abiotic components. The metaphors of "host" and "construction" obscure both the relational complexity and the temporal and spatial continuity of these reciprocal interactions, which are necessary to understand how complex patterns such as holobionts emerge and persist.

The niche perspective on holobionts is not new, but it has only partially permeated discussions of host–microbe symbiosis. Chiu and collaborators have described host–microbe relationships in terms of reciprocal developmental scaffolding and mutual niche construction, emphasizing how microbes and macrobes contribute to one another's developmental, ecological, and evolutionary

conditions[28,29]. Our approach builds on this work but deliberately foregrounds microbial influence while taking the relational argument a step further. Reciprocity can still imply entities that first exist and subsequently act upon one another. Niche weaving instead questions this point of departure: rather than asking only how already constituted microbes and macrobes modify one another and their respective niches, it asks how their biological forms and ecological roles themselves emerge through histories of microbial, macrobial, and abiotic relations. It therefore does not begin with a macrobe conceived as an already constituted niche that is subsequently colonized by microorganisms, but considers how the forms we recognize as multicellular organisms have themselves emerged through these relations. Importantly, it also extends microbial influence beyond the physical presence of microorganisms by considering how such influence can persist and act across space and time.

This shift should not be taken to mean that microbes produce multicellular organisms or that macrobes are passive products of microbial activity. Niche weaving does not simply invert the direction of causation; it challenges the premise that the entities involved must be fully constituted before their relations begin. Developing multicellular bodies generate habitats, interfaces, gradients, and selective environments for microorganisms, while microbial activities contribute biochemical and physical conditions that shape development, physiology, immunity, and selection. Through these ongoing relations, relatively stable biological forms emerge and, once established, become conditions through which subsequent relations unfold. Neither microbes nor macrobes should therefore be treated as fully constituted entities that precede the relationships through which they acquire their ecological forms. In this respect, niche weaving resonates with Barad's

concept of intra-action, in which relata emerge through, rather than simply precede, their relations[30].

An important distinction follows between microbial metabolites and microbial niche weaving. Metabolites are central mechanisms through which microbial communities modify their surroundings, but they are not the evolutionary process itself. Beaver dams matter in niche construction theory because of how they transform ecological conditions and selective pressures, not because the dam itself is the process. Likewise, microbial metabolites matter because their production, consumption, transformation, and degradation continually reshape biochemical environments experienced by microbial and multicellular organisms.

The explanatory shift is therefore from molecules to processes. Microbial communities generate recurring biochemical environments whose composition, abundance, and spatial distribution are shaped by ecological interaction and evolutionary change. Such environments can be reconstructed despite turnover in the identities of the taxa producing them [ITSNTS]. Metabolites are best understood as a biochemical currency of microbial niche weaving; the more enduring phenomenon is the repeated production (development) and constant reorganization (evolution) of ecological conditions.

Metabolomics makes this process experimentally tractable by revealing biochemical states and transformations that taxonomic surveys alone cannot capture. The conceptual shift from hosts to niches therefore follows not only from theory but also from changing observational capacities within

331 microbiology: from identifying which organisms are present toward asking which ecological
332 processes are being produced and maintained.
333
334 **The Principle of Recursive Microbial Influence**
335 We propose a principle of recursive microbial influence: biological patterns and processes shaped
336 by microbial activity can themselves become the conditions, substrates, or spokes through which
337 subsequent microbial influence shapes the lattice. Microbial effects are therefore neither merely
338 repeated across evolutionary time, nor limited to increasing the microbes' fitness (as in niche
339 construction). They can be incorporated into biological organization in ways that alter the spatial,
340 temporal, and physiological routes through which later microbial effects are expressed.
341

342 Microbial influence extends from maternal environments to offspring, from zygote to death, from
343 local sites to distant tissues, and from surrounding ecosystems into developing bodies. More
344 fundamentally, however, this influence is recursive across evolutionary time. The biological systems
345 through which microbes exert their effects are themselves products of earlier rounds of ecological
346 interaction and niche weaving. Microbial influence is ongoing and predates the origin of animals,
347 plants, or even eukaryotes. Microbial niche weaving can itself be understood as emerging from an
348 earlier history of niche weaving by chemical species[31], after which successive forms of biological
349 organization evolved within environments already transformed by microbial activity through
350 nutrient cycling, redox chemistry, atmospheric modification, mineral transformations, metabolite
351 production, and the restructuring of physical habitats. Later organisms did not encounter microbial
352 influence as a novel external force; they emerged as new patterns within ecological conditions that

353 earlier microbial communities had already helped weave, including patterns that themselves
354 became conditions for subsequent biological organization.

355

356 Recursiveness follows from this historical continuity. Microbial activity modifies environmental
357 conditions; those altered conditions influence the evolution and development of biological
358 structures and physiological systems; and those structures and systems can in turn constitute new
359 habitats, transport networks, gradients, interfaces, and substrates through which microbial
360 influence operates. The consequence is an iterative process in which the products of earlier
361 microbial influence become infrastructures for later microbial influence. Evolutionary novelty
362 therefore need not interrupt microbial causation. It can reorganize and extend it.

363

364 Circulatory systems provide an illustrative example. Microbial metabolites influence vascular
365 development, while vascular networks subsequently expand the distances over which microbial
366 products generated at localized sites can affect distant and otherwise inaccessible tissues. A
367 microbial effect on the development of a transport system can thus contribute to the realization of
368 a new route through which later microbial effects are propagated (e.g., impact of metabolites on
369 the development of the blood-brain barrier). If multispecies assemblages such as holobionts are
370 products of interweaving encounters between microbes and macrobes, then the circulatory system
371 is a spoke that holds the lattice together, that structures further workings and evolution of the
372 system. By contributing to the evolution of the circulatory system[32], microbes recursively shape their
373 future evolution too.

Similar recursive relationships may occur, with different levels of empirical support, in the evolution and development of mucosal surfaces, internal microbial habitats, placentation, and other physiological interfaces. In other words, structures emerging within microbially modified environments can subsequently alter the scope and organization of microbial influence.

From this perspective, the emergence of eukaryotic cells, multicellularity, developmental programs, internal microbial habitats, vascular systems, and reproductive architectures can be viewed as successive reorganizations of a pre-existing ecological relationship rather than as points at which microbial influence begins anew. Each organizational innovation changes the conditions under which interactions occur and thereby creates new possibilities for microbial effects to be localized, transmitted, amplified, filtered, or integrated. Recursion therefore links evolutionary history to present-day physiology: contemporary routes of microbial influence may themselves bear the consequences of much older microbial–environmental interactions.

The relevant continuity is therefore compatible with turnover in microbial taxa, communities, metabolites, and symbiotic associations. It is a continuity of microbial processes within the causal history of biological evolution. Particular taxa may disappear, communities may turn over, and mechanisms may change, while the broader recursive pattern persists. What is inherited across evolutionary time is therefore not a specific microbiome, but an evolving pattern of causal interactions in which earlier patterns become conditions for those that follow. We have never been hosts. We are patterns emerging through an ongoing process of niche weaving, shaped by—and in turn shaping—the biological and abiotic relations through which we persist.

From microbial signals to expanded ecological organization

Our framework suggests a different way of thinking about the origin of multicellularity. There was never a stage in the evolution of multicellularity in which cells first existed as ecologically isolated units and only later began interacting. Cellular life has always unfolded through interactions across space and time. Among microbes, these interactions can be diffuse and relatively weak, as among free-living cells connected through shared metabolites and environmental modification, or highly localized and intense, as in aggregates and biofilms. The evolutionary perspective we adopt therefore moves beyond asking whether biological entities interact to consider the spatial, temporal, and organizational regimes through which their interactions unfold. From this perspective, the evolutionary distinctiveness of the holobiont lies not in the existence of interaction itself, but in the particular regimes of interaction through which holobiont forms are constituted.

The same logic applies to the evolutionary history of eukaryotes. Eukaryotic cells did not emerge into an interaction-free world and subsequently become social or multicellular. They evolved within environments already structured by microbial activity and in continual exchange with other cells. From this perspective, the origin of multicellularity is better understood as the emergence of new ways of organizing, stabilizing, coordinating, and reproducing cellular interactions rather than as the origin of those interactions. Multicellular organisms introduced qualitatively new forms of adhesion, developmental integration, differentiation, division of labour, conflict regulation, and reproduction. The point is instead that these innovations were built upon a much older ecological history of cell–cell and cell–environment interaction.

The host metaphor obscures this continuity by encouraging a contrast between supposedly autonomous organisms and microbial communities that later enter into association with them. Once that framing is relaxed, a more continuous picture emerges: interactions extending across space and time and repeatedly reorganized into new ecological and developmental configurations. Multicellularity then appears not as a transition from isolated cells to interacting cells, but as one major evolutionary transformation in how cellular interactions are structured and maintained.

Empirical work on choanoflagellates makes this alternative sequence more concrete. Choanoflagellates, the closest known unicellular relatives of animals, are themselves embedded in microbial environments in which bacterial products can regulate major life-history transitions. In *Salpingoeca rosetta*, bacterial sulfonolipids induce multicellular rosette development, while a bacterial chondroitinase can trigger mating[33–35]. These findings do not demonstrate that bacteria caused the origin of animal multicellularity. They do, however, show that developmental decisions closely related to multicellular organization could be responsive to microbial signals near the base of the animal lineage. Early multicellularity therefore evolved within interaction patterns already structured by microbial activity rather than outside microbial ecology.

Niche weaving provides a natural framework for understanding this continuity. Through metabolism, spatial organization, collective growth, and modification of local physicochemical conditions, microbial communities actively reshape the settings in which subsequent interactions occur. These altered environments can in turn favour new forms of proximity, persistence, cooperation, and coordination. Multicellularity must therefore be viewed as a novel form of biological organization that builds on a much older evolutionary pattern in which unicellular

organisms modify their surroundings and thereby change the conditions under which cellular interactions unfold.

The evolution of circulatory systems is a case of niche weaving that exemplifies how structurally important this reciprocal process can be. Vascular systems are conventionally understood as adaptations that overcome diffusion constraints imposed by increasing body size and metabolic demand. That account remains essential, but circulation also transformed the ecological geometry of animal–microbe interactions by allowing microbial products to influence tissues far from sites of microbial residence[13]. It expanded the spatial scale over which microbial niche weaving could affect development, physiology, immunity, and homeostasis. As discussed below, it also increased the carrying capacity of macrobes, by making them bigger, such that this process can also be said to be a process of niche weaving through which microbes could ultimately increase their reproductive output.

Anatomical innovations can therefore be interpreted not only as solutions to organismal constraints but also as spokes that stabilize, coordinate, and amplify persistent ecological interactions. Choanoflagellate developmental responses show that microbial signals could regulate major biological decisions before the emergence of animals; circulatory systems illustrate how later anatomical innovations could extend the reach of such interactions across larger bodies and longer developmental timescales.

Our objective here is to show that the evolution of structural spokes such as the vascular system can be understood as a reorganization of pre-existing relations between microbes and their

environments, as those environments came to include increasingly complex multicellular forms. This perspective complements evolutionary accounts centered on macrobes and their fitness by broadening the ecological consequences and selective contexts considered relevant to their evolution. Structures that enable larger bodies, differentiated tissues, and long-distance transport simultaneously generate new microbial habitats and new routes through which microbial products can influence the organism. The same anatomical innovation can therefore constitute both an adaptation of the multicellular organism and part of an evolving ecological infrastructure for microbe–macrobe relations. From the perspective of niche weaving, these are not competing interpretations but different aspects of an evolutionary process in which multicellular organization and its ecological relations emerge together.

From external to internalized microbial niche weaving

A further implication is that the microbial niche weaving that shaped macrobes may initially have operated largely through the surrounding environment before becoming progressively internalized. In marine ecosystems, seawater provides a shared biochemical medium through which products released by free-living microbes can diffuse, circulate, and repeatedly encounter eukaryotic cells and early animals. The responses of *S. rosetta* to bacterial products, as discussed in the previous section, demonstrate that biologically important microbial effects do not require stable physical colonization. Early animal evolution thus unfolded within environments already shaped by microbial niche weaving, where unicellular ancestors participated in microbial relations that cannot readily be captured by the host metaphor.

As bodies grew larger and their cells and tissues became increasingly differentiated, reliance on diffusion alone—whether through seawater or across tissues—would have become progressively less effective for coordinating metabolite-mediated interactions, particularly as deeper tissues came to lie beyond effective diffusion distances. More intimate associations with microorganisms, together with epithelial and mucosal interfaces and specialized microbial habitats, could increase the reliability, persistence, and spatial organization of those interactions. Circulatory systems could then extend the reach of locally generated microbial products throughout the body. Resident microbiomes and vascular transport can therefore be viewed as complementary components in a progressive spatial reorganization of microbial niche weaving that led to the internalization of microbes. The pattern in the weave evolved.

This perspective may also help explain divergent forms of animal–microbe association across habitats. Terrestrialization disrupted much of the environmental continuity afforded by marine environments. This restricts the spatial scales over which microbial metabolites could affect other entities. Different architectures evolved through which microbial influence could be expressed. In many cases, stable internal microbial ecosystems brought microbial communities and their metabolic activities into persistent proximity with animal tissues. Other macrobes may lack substantial resident gut microbiomes yet remain embedded in microbial processes through intracellular symbionts (e.g., *Buchnera* in aphids), transient environmentally acquired microbes (e.g., microorganisms ingested by aquatic filter feeders), or environmentally derived microbial products (e.g., bacterial cues inducing settlement and metamorphosis in marine invertebrate larvae). Microbial influence is therefore a stable fact across evolutionary divergence, but its

ecological, spatial, and biological organization has changed across time, in part to adapt to different environments.

As discussed in the previous section, the evolution of the vascular system is a particularly illustrative case of reorganization. Whereas internalization could concentrate microbial communities and their metabolic activities at particular sites within an animal, circulation provided a mechanism through which products generated at these localized interfaces could reach tissues throughout an increasingly large and differentiated body. Microbial influence could therefore become simultaneously more spatially localized at its source and more widely distributed in its effects. In this sense, internal microbial ecosystems and vascular transport represent complementary, although evolutionarily distinct, transformations in the spatial architecture of microbial influence: one concentrates microbial activity within specific anatomical space, while the other expands the distances over which microbial metabolites and other products can act.

In summary, through terrestrialization, interactions that could operate through a continuous external aqueous environment became increasingly partitioned among localized microbial habitats, intracellular associations, recurrent environmental exposure, and transport systems capable of distributing microbial products between otherwise distant sites. Internal microbiomes and vascular systems can therefore be understood as components of a broader evolutionary reorganization through which microbial influence became differently localized, transmitted, concentrated, and physiologically integrated. The underlying continuity lies not in any particular microbiome or mode of association, but in microbial influence itself; what changed through evolution were the patterns through which that influence could operate.

Beyond metabolism: the physical niche

Microbial niche weaving should not be reduced to *metabolic* niche weaving. Microorganisms modify their environments through their physical presence and collective material properties as well as through the molecules they produce and consume. The physical ecology of microbiomes is structured by variables such as osmolality, flow, pH, mucus mechanics, spatial confinement, and mechanical perturbation. Transient changes in these conditions can have persistent ecological effects, and spatially explicit studies show that flow and bacterial growth jointly shape community organization in ways that cannot be inferred from taxonomy or metabolic exchange alone.

The reciprocal effect is equally important: microbes also reshape physical conditions. Growing populations occupy volume, displace fluids and neighboring cells, alter porosity and diffusion distances, generate local stresses, and change the rheological properties of their surroundings. Biofilms and extracellular polymeric matrices can transform relatively fluid habitats into heterogeneous viscoelastic materials, while dense growth can exert mechanical pressure on neighboring populations and macrobial surfaces. Microbial activity can also couple to changes in density and pressure that alter macroscopic fluid behavior.

Microorganisms should therefore be treated simultaneously as chemical transformers and physical constituents of their environments. A woven microbial niche is not adequately represented by metabolite concentrations alone, but by a multidimensional physicochemical state that may include pressure, microbial volume fraction, viscosity, elasticity, flow, spatial architecture, diffusion, pH, redox state, and metabolite concentrations. This broader formulation allows microbial biomass and spatial organization to modify environmental states directly rather than only through metabolite-

production terms. It also matches traditional definitions of the niche that emphasize its multidimensionality.

A general principle across multicellular life

Although the argument developed here focuses primarily on animal evolution, it extends naturally to plants. The rhizosphere, phyllosphere, and endosphere are active ecological interfaces in which microbial communities influence nutrient availability, hormone production, stress tolerance, pathogen resistance, and development. Roots expanded the spatial interface between plants and soil microorganisms, while mycorrhizal symbioses created integrated metabolic networks linking plants, fungi, and surrounding microbial communities. Root exudates reshape microbial community composition and metabolism, while microbial products influence root architecture, nutrient acquisition, and immunity.

The shared trait (i.e., presence of structures that facilitate microbial interactions across space) between plants and animals is significant because multicellularity evolved independently in these lineages. Despite different anatomical solutions—roots and vascular tissues in plants; circulatory systems, epithelial barriers, and mucosal interfaces in animals—both repeatedly evolved structures that stabilize and expand the spatial and temporal scales over which microbial niche weaving can influence development and physiology. This suggests that microbial niche weaving may be a necessary feature of multicellular evolution rather than a peculiarity of animal microbiomes. Indeed, while plant and animal microbiomes differ significantly and while all multicellular lineages depend on resident microbial communities in different ways, the central insight of this paper is that

multicellular evolution necessarily unfolds within ecological systems continuously modified by microorganisms. As a result, multicellular anatomies themselves become parts of those systems.

Biological individuality and major evolutionary transitions

Reconceptualizing multicellular organisms as microbial niches has consequences for how biological individuality is understood. Conventional accounts often begin with bounded organisms and ask how microbial associates modify, extend, or destabilize their boundaries. Niche weaving reverses this point of departure: it asks how relatively stable individuals emerge through ongoing ecological relations, and what conditions allow such forms to arise, persist, and repeatedly re-form. Individuality, from this perspective, is not prior to ecological interaction but one of its possible outcomes.

Our perspective thus shares a central objective with the major transitions in individuality research program: explaining how biological individuality arises. It differs, however, in both its point of departure and the processes it foregrounds. Major-transitions accounts typically focus on how previously distinct entities become integrated into higher-level evolutionary individuals through changes in reproductive alignment, cooperation, conflict mediation, and the distribution of fitness. Niche weaving instead foregrounds the reorganization of ecological relations through which the entities themselves acquire their biological and ecological forms. From this perspective, transitions in individuality were embedded within broader transformations in ecological, metabolic, and interkingdom organization rather than constituting isolated transitions among pre-existing biological units. New evolutionary individuals can therefore be understood as one outcome of changes in connectivity, spatial organization, metabolic exchange, and conflict mediation—not

596 necessarily as the privileged events around which evolutionary history must be organized. The
597 evolution of circulatory systems provides a particularly instructive example: circulation contributed
598 to the integration and coordination of increasingly large multicellular bodies, but simultaneously
599 reorganized pre-existing microbe–macrobe relations by creating new spatial and physiological
600 routes through which microbial metabolites and other products could act across the organism. What
601 appears from the perspective of individuality as an innovation integrating the multicellular organism
602 can thus also be understood as a reorganization of the ecological relations through which that
603 organism emerged and continues to function.
604
605 Microbial cells have always existed in networks ranging from diffuse, low-rate interactions to dense
606 and highly structured communities such as biofilms. Hence, there was no evolutionary world devoid
607 of interaction among cells and organisms in which "hosts" could have emerged *before* being
608 colonized. Eukaryotic evolution unfolded within this continuity of ecological relations with other
609 cells and organisms. What changed across evolutionary time was not the existence of interaction
610 but its organization, intensity, spatial scale, persistence, and degree of internalization: individuality
611 can thus be understood as the relative stabilization of a pattern of interactions within a broader
612 relational system.
613
614 Evolutionary continuity, in this view, resides in recurring ecological relationships and processes
615 reconstructed despite turnover in their constituent taxa. This resonates with process-oriented
616 approaches such as 'It's the Song, Not the Singer' and related formulations, in which persistence lies
617 in recurring organization rather than uninterrupted presence of particular lineages[3,4]. One such

evolutionary ‘song’ may be the network of microbial–macrobial interactions that repeatedly reconstructs metabolically and physically integrated ecological systems. Yet we prefer the weaving metaphor to the musical one, as it better emphasizes the relationships among components: a strand acquires its structural significance through its interlacing with others and the patterns that emerge from those relations.

Meta-organisms or holobionts can therefore be viewed as relatively stable realizations of persistent ecological interactions rather than the point from which those interactions begin.

Major evolutionary transitions may thus be understood not only as transitions in individuality, but as spatial and functional reorganizations of pre-existing ecological processes through which new forms of individuality emerge. In this broader view, recurrent organized processes spanning multiple kinds of biological entities and their relationships – also called evosystems[36] – become legitimate objects of evolutionary explanation, whether they be units of selection or not.

Conclusion: We have never been hosts

Our argument rests on a central distinction between microbial presence and microbial influence. Physical association is an important form of interaction and has long been central to microbiology and ecology, but microbial causation extends beyond direct or persistent physical contact. Metabolites, environmental transformations, material effects, and recurring ecological processes allow microbial activity to influence biological organization across space, time, and generations. Once these processes are placed at the center of analysis, multicellular organisms appear less as autonomous hosts subsequently colonized by microorganisms and more as evolving ecological forms that emerged within, and continue to participate in, microbial niche weaving.

Recognizing microbial influence beyond the physical presence of microorganisms challenges the metaphor of multicellular organisms as “hosts.” The main limitation of this metaphor is the temporal and ontological asymmetry it implies: the host is conceived as existing first, with microbial occupants arriving later. This sequence obscures a much longer relational history. Microbial activity preceded multicellular life and has continued to shape the biochemical, physical, and ecological conditions within which multicellular forms emerged, evolved, and develop. Hosthood is therefore better understood as a derived ecological configuration within this history than as its starting point.

We propose niche weaving as an alternative metaphor for understanding this process. The metaphor emphasizes how relatively stable forms emerge through the interlacing of components whose significance depends partly on their relations with others. Holobionts can accordingly be understood as patterns emerging through particular organizations of microbial, macrobial, and abiotic relations rather than as pre-existing hosts plus their microbial associates. Such patterns need not depend on fixed constituent taxa: similar forms and functions can emerge through different microbial communities, mechanisms, and associations. Moreover, patterns produced through earlier interactions can themselves become conditions or structural spokes around which subsequent interactions are organized. Niche weaving therefore captures not only relational organization but its historical and recursive character: biological forms emerge through ecological relations while simultaneously reorganizing the conditions through which later relations unfold.

This perspective does not require abandoning concepts such as organism, lineage, individuality, symbiosis, or selection; it changes their explanatory relationship to ecological processes. Rather than treating relations as interactions among fully constituted entities, niche weaving asks how

relatively stable biological forms themselves emerge through, and remain embedded within, broader ecological relations. Individuality can consequently be understood as the relative stabilization of particular patterns of interaction rather than as the ontological condition from which interaction begins. This shift also redirects microbiome research from asking primarily “Which microbes inhabit the host?” toward a broader evolutionary and developmental question: “Which microbial processes have contributed, and continue to contribute, to the emergence and persistence of the biological forms we call holobionts?” Microorganisms have changed, communities have turned over, and the spatial organization of their associations has repeatedly been transformed, but microbial influence has remained woven into the processes through which multicellular life takes form. In this sense, we have never been hosts.

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