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PERSPECTIVE

Conserving the holobiont

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Abstract

1. Organismal biology has undergone a dramatic paradigm shift in the last decade. The realization that host cells and genes are outnumbered by symbiotic microbial cells and their genes has forced us to rethink our focus on 'individuals'. It is also becoming increasingly clear that the ecology and biology of animals and plants are intimately connected with their microbial partners. In the context of conserving functioning species, such revelatory insights beg the question—what exactly should we be trying to conserve?
2. Here, we review how an understanding of host–microbe interactions can benefit conservation biology. We propose a way forward for conservation biologists, to gather evidence of the potential effects of changes to plant and animal microbiomes, and to incorporate the holobiont concept into applied conservation practice.
3. In humans, microbes influence physiology, health, behaviour and psychology. In animals and plants, microbes similarly influence critically important components of health, communication and (in animals) behaviour. Together, the animal or plant and all of its associated micro-organisms are termed the holobiont.
4. At the same time, humans are now the strongest evolutionary force on the planet, causing global change at unprecedented scale. We know that microbial diversity in humans has been compromised in urban societies, with a growing list of consequences for health and function. While we still have limited evidence for similar effects in plants and animals, anthropogenic factors that affect diversity are also likely to affect animal and plant microbiomes, with similar associated effects on host function and health.
5. Microbiome research is still in its relative infancy, particularly in its application to plants and animals, yet the tools are becoming more widely available and affordable. Forward-looking conservation biologists could harness such tools and apply them to the study of plant and animal microbiomes with the goal of understanding which microbiota might be required to ensure future viability of conserved host populations.
6. For now, the precautionary principle applies. We suggest that, to meaningfully and effectively conserve a species, we must also consider how to conserve the bacteria, viruses, fungi and other symbionts intimately associated with that macro-organism.

KEYWORDS
animal behaviour, captive breeding, conservation, genomic diversity, hologenome, microbiome, phyllosphere, seed banking

1 | INTRODUCTION

Humans are the most influential evolutionary force in the world, and our impacts have precipitated a new geological epoch, the Anthropocene (Palumbi, 2001; Waters et al., 2016). Factors that affect the diversity of animals and plants in the Anthropocene, such as pollution, reduced connectivity, habitat fragmentation and domestication, are also likely to impact microbial diversity (Gillings & Paulsen, 2014). Yet, conservation biology as a discipline has generally concentrated on the macro-, rather than the microbial world. We have only recently become aware of the vast diversity of culture-independent microbial life (Turnbaugh et al., 2007). While it is now recognized that all macro-organisms are habitat for microbes, we are still in the earliest stages of understanding the extent of the intricate and critical links between microbial symbionts and the physiology and health of their hosts, with the vast majority of research being focused on the human gut microbiome (Clemente, Ursell, Parfrey, & Knight, 2012; Gilbert et al., 2016; Sonnenburg & Sonnenburg, 2019).

Investigations in humans have shown how the Anthropocene has altered our microbiome, with industrialized microbiomes diverging from ancestral states (Blaser & Falkow, 2009; Sonnenburg & Sonnenburg, 2019). Such changes have been linked to the rise of non-communicable chronic human diseases, such as obesity, insulin resistance, allergies and irritable bowel syndrome (Gilbert et al., 2016; Sonnenburg & Sonnenburg, 2019). We are also now aware that animals and plants possess species-specific microbiomes (e.g. corals: Hernandez-Agreda, Gates, & Ainsworth, 2017; plants: Wasserman, Cernava, Muller, Berg, & Berg, 2019; finches: Engel et al., 2018), which likely play similarly important roles in the physiology and healthy function of their hosts (Delavaux et al., 2019; Inderjit & van der Putten, 2010; Werner, Cornwell, Sprent, Kattge, & Kiers, 2014). Wild animals and plant microbiomes are also being altered through domestication, captivity, antimicrobial use and other forms of disturbance (Table 1; Hird, 2017; Pérez-Jaramillo, Carrión, Hollander, & Raaijmakers, 2018). Given this emerging awareness of the effects of altered microbiomes on health and disease across many well-studied species, we propose that conservation biologists would benefit from greater knowledge and consideration of microbiome composition and dynamics in wild animals and plants. We also propose that conservation biologists widen their scope to conserve both diversity at the level of the individual host genome and at the level of the community of organisms comprising that individual—the holobiont (Roughgarden, 2018).

The holobiont is an animal or plant and all of its associated micro-organisms (Morar & Bohannan, 2019; Zilber-Rosenberg & Rosenberg, 2008). We suggest that there are many potential benefits for conservation biology in working towards conservation of the holobiont. Fortunately, the tools and techniques of microbiome research are becoming readily available and affordable, creating new opportunities to incorporate the microbiome into conservation. We discuss how findings from human and model animal microbiome research apply to wild animals and to plants, and consider potential approaches to research and conservation actions in support of a holobiont-level focus for conservation biology. For simplicity, we focus on micro-organisms, but similar arguments can be made for any symbiotic taxa.

TABLE 1 Examples of human impacts on the natural environment and predicted effects on the microbiome

Human impact	Mechanism of action	Effect on microbes
Antibiotic pollution	Kill bacterial strains, select for antibiotic resistance	Alter community structure of microbiome
Farming, agriculture	Less diverse environmental exposure	Less diverse community
Habitat fragmentation and destruction	Reduced group sizes	Fewer opportunities for horizontal transmission
Urbanization	Altered habitat, altered micro-climates, reduced macro-organism diversity, pollution, rubbish	Altered opportunities for transmission. Altered community structure of microbiomes
Reduced macro-organism biodiversity	Reduced host diversity	Reduced microbial diversity
Domestication	Few species dominate landscape, high antibiotic use	Reduced host diversity and availability
Invasive species	Novel hosts	Increased microbial diversity due to novel additions to local microbiota, OR reduced diversity due to global homogenization of host fauna
Climate change	Alter ambient temperature and other environmental conditions	Affect transmission, external survival outside host, limit host availability where host species that do not adapt to climate change

1.1 | Microbiomes mediate macro-organismal health

The development of new metagenomic techniques to survey microbial diversity has thrown open a window onto the astounding diversity of microbial life consorting with macro-organismal hosts (Thompson et al., 2017; Turnbaugh et al., 2007). In nature, plants and animals are colonized both internally and externally by an array of micro-organisms (bacteria, fungi, archaea and viruses). For instance, the average individual human hosts as many microbial cells as human cells (Sender, Fuchs, & Milo, 2016), and the genes catalogued from the combined human gut microbiota now number almost 10 million (Li et al., 2014). Macro-organismal relationships with micro-organisms are complex and bidirectional, raising new questions about the very definitions of individuals and species (Gissis, Lamm, & Shavit, 2018). Host-associated microbes may or may not have coevolved with the host, and may or may not affect the host's phenotype (Theis et al., 2016; Werner et al., 2014). Many microbes are likely to straddle the line between commensal and pathogen (Gilmore & Ferretti, 2003; Rodriguez et al., 2019). We also now know that some host-associated microbes play fundamental roles in macro-organismal evolution (Alberdi, Aizpurua, Bohmann, Zepeda-Mendoza, & Gilbert, 2016; Brucker & Bordenstein, 2012; Sharpton, 2018; Werner et al., 2014), development and function (Heijtza et al., 2011; Rodriguez et al., 2019; Sampson & Mazmanian, 2015; Vuong, Yano, Fung, & Hsiao, 2017). The evidence is clear that microbes play an important role in the biology and ecology of larger organisms.

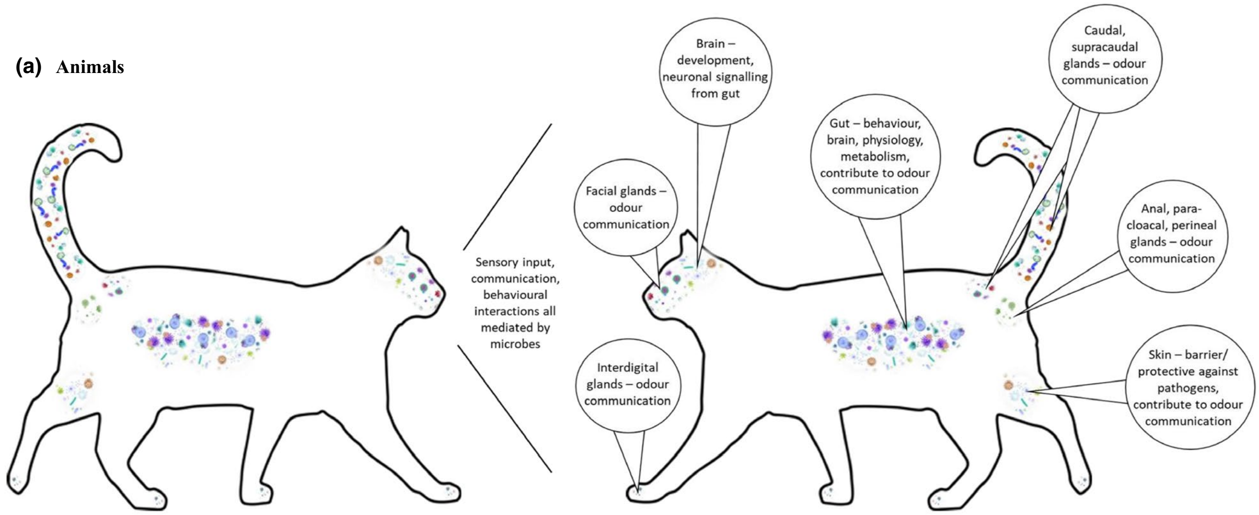
Research to date has largely focused on the importance of the human gut microbiome for health (e.g. Belizário & Napolitano, 2015; Clemente et al., 2012; Shreiner, Kao, & Young, 2015). In humans, the disruption of human microbiota through dietary change, antimicrobial treatments and other cultural practices is directly linked to the rapid increase in complex diseases of previously unknown aetiology (Gillings, Paulsen, & Tetu, 2015; Sonnenburg & Sonnenburg, 2019). These include effects on the autoimmune system (Orr, Kocurek, & Young, 2018), the brain and behaviour (Needham, Tang, & Wu, 2018), and physiology (Alberdi et al., 2016; Kreznar et al., 2017). The implications of such findings are now driving developments in the fields of healthcare (Hadrich, 2018). For plants, microbiome research has similarly focused on health and disease in model species, such as *Arabidopsis*, or agriculturally important crops (Pérez-Jaramillo et al., 2018; Toju et al., 2018); however, attention is now expanding to wild animal and plant microbiomes with the potential to shape conservation practice.

Microbial influence on a macro-organism's phenotype and ecology is potentially extremely far-reaching (Figure 1a,b). There is now good evidence that across numerous animal species gut microbiomes play a role in moderating development, physiology and behaviour, and protecting against disease (Archie & Theis, 2011; Archie & Tung, 2015; Ezenwa, Gerardo, Inouye, Medina, & Xavier, 2012; Hanning & Diaz-Sanchez, 2015; Heijtza et al., 2011; Lizé, Mckay, & Lewis, 2013; Round & Mazmanian, 2009). Micro-organisms associated with the skin of amphibians (Harris, James, Lauer, Simon, & Patel, 2006) and marine organisms (Wahl, Goecke, Labes, Dobretsov, & Weinberger, 2012) protect against pathogens and promote wound healing (Ross, Rodrigues Hoffmann, & Neufeld, 2019). In plants, microbial community structure on the phyllosphere is correlated with key plant growth characteristics, including wood density, leaf mass per area and mortality (Kembel et al., 2014). Similarly, soil microbial diversity and structure have been shown to strongly impact plant biomass production, plant–plant interactions, and plays a role in maintaining species diversity (Compant, Samad, Faist, & Sessitsch, 2019; Yang, Wagg, Veresoglou, Hempel, & Rillig, 2018). The contribution of micro-organisms to plant growth through mycorrhizal fungi, nitrogen fixing bacteria, and the wider above- and below-ground microbiome, is well known, and there is a growing appreciation for additional roles in resistance to biotic and abiotic stressors (Vandenkoornhuyse, Quaiser, Duhamel, Van, & Dufresne, 2015).

While we are still learning what a 'healthy' or fully functional microbiome comprises in humans, let alone in plants and animals, a diverse microbiome is generally considered desirable and indicative of health (Huttenhower et al., 2012). A diverse microbiome is more likely to have functional redundancy for important metabolic processes or other critical services provided by the microbiota to the host (Louca et al., 2018; Moya & Ferrer, 2016; Valdes, Walter, Segal, & Spector, 2018). Yet, there is currently little other concrete evidence linking diversity to an 'optimal' or 'healthy' microbiome. This is partly because the identification of functional roles played by individual, groups of, or interactions between microbes within a host microbiome, remains a pressing goal for much current human (let alone plant and animal) microbiome research (Heintz-Buschart & Wilmes, 2018).

One approach to identifying a healthy microbiome in humans is to compare ancestral with industrialized human microbiomes, with the assumption that the ancestral state is an optimal state to which modern microbiomes might aspire (e.g. Blaser & Falkow, 2009; Sonnenburg & Sonnenburg, 2019). However, this approach does not account for the possibility that ancestral microbiomes might no longer be 'optimal' in the modern world. For example, a gut microbiome fine-tuned for the digestion of microbiota-accessible carbohydrates will not result in health if the host is limited to a standard Western diet (Sonnenburg & Sonnenburg, 2019). Another approach is to map microbial gene sequences to databases of known metabolic functions, or functional profiling, with subsequent correlation to disease or health states (e.g. Armour, Nayfach, Pollard, & Sharpton, 2019). Such approaches are useful insofar as the presence of individual metabolic functions link directly to health or disease in the host, but this may not often be the case. Other studies are now profiling both microbiome taxonomic and functional composition to uncover patterns that may be important for health and disease in humans (Franzosa et al., 2018; Manor & Borenstein, 2017). Such methods have not yet been applied widely to plant and animal microbiomes, but most likely can and will be in the near future. Conservation biologists have an emerging opportunity to engage with this research in a way that informs and directs the application of these methods to best understand the needs of threatened and vulnerable species.

(a) Animals



(b) Plants

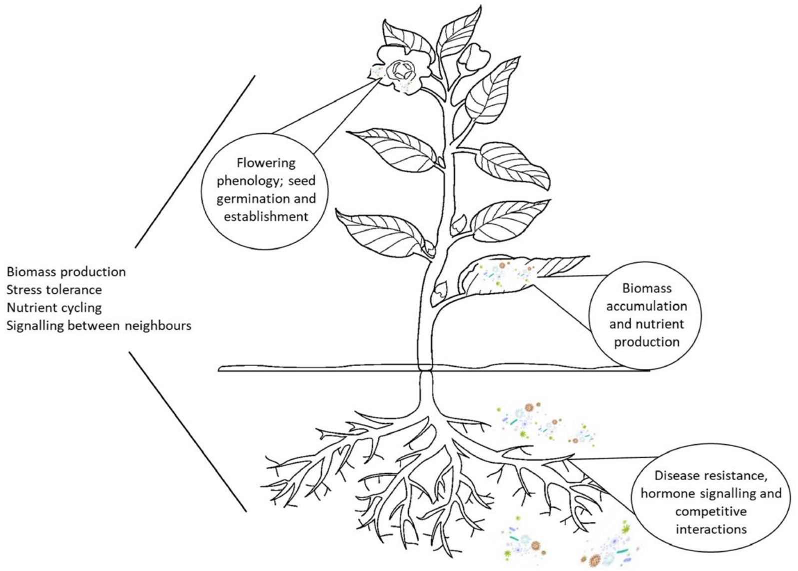


FIGURE 1 The different microbes on and within different areas of an (a) animal and (b) plant, and some ways in which they influence phenotype, function and intraspecific interactions. Microbes may be expected to directly influence population and community structure of host populations via their influence on intraspecific interactions among hosts

1.2 | Impacts of the Anthropocene on animal and plant microbiomes

Human activities impact the natural environment in many ways that potentially affect animal and plant microbiomes, much as they have affected human gut microbiomes (Blaser & Falkow, 2009; Sonnenburg & Sonnenburg, 2019). It seems likely that any event or action that reduces opportunities for the transmission or maintenance of functionally important microbes will also have a negative impact on animal microbiome diversity and community structure with potential flow on effects for the host. The advent of farming, widespread antimicrobial pollution, habitat fragmentation and urbanization are all potential examples (Table 1).

Anthropogenic disturbance can reduce microbiome diversity, and in some cases this reduced diversity has been associated with harm. For example, early-life reduction of gut microbial diversity in tadpoles is associated with increased parasite establishment in adult frogs (Knutie, Wilkinson, Kohl, & Rohr, 2017). However, in some cases, anthropogenic disturbances may increase gut microbiome diversity. This might be due to the introduction of new pathogenic microbes, or the displacement of key, functionally important microbial species by other, less functional microbes. For example, in one study, phyllosphere (leaf-based) microbiomes across an urban land use gradient had the highest diversity in the most urbanized environments, yet the typically dominant Alphaproteobacteria were reduced in abundance (Laforest-Lapointe, Messier, & Kembel, 2017). Of two studies on urbanized sparrows, one study (on house sparrows, *Passer domesticus*) found reduced gut microbial diversity at more urbanized locations (Teyssier et al., 2018), whereas the other (on white-crowned sparrows *Zonotrichia leucophrys*) found gut microbiome diversity to be higher in urban compared to rural populations (Phillips, Berlow, & Derryberry, 2018). Human impacts may therefore alter different species' microbiomes in different ways. Phillips et al. (2018) speculated that 'urban adapted' species may actually benefit from novel urban microbiota, although again, we do not yet have evidence to support these proposed benefits.

In addition, the specific mechanisms by which such changes might translate to either benefit or harm remain unclear. The simplest mechanism would be via effects of microbiome composition on dietary niche—possessing bacteria that can metabolize novel dietary components might allow hosts to rapidly adapt to urban or otherwise modified environments (Alberdi et al., 2016). However, evidence from humans suggests that while our microbes may be rapidly adapting to the Anthropocene, our own genomes have not kept pace—resulting in a mismatch between the composition and function of the modern human gut microbiome and the metabolic and physiological requirements of the host. Purported consequences include the rise of chronic non-communicable modern diseases such as obesity (Sonnenburg & Sonnenburg, 2019). The key challenge for microbiome research remains, in both human and non-human organisms, to uncover specific functional roles required for health, and to determine whether particular microbial species, multiple microbial species or interactions between sets of microbial species are responsible for providing these functions (Heintz-Buschart & Wilmes, 2018).

Importantly, the loss of microbial diversity can be irreversible. Host-associated microbial communities are generally species-specific (Kohl, Dearing, & Bordenstein, 2018; Nishida & Ochman, 2018), and are in large part passed vertically from parents to offspring (Ferretti et al., 2018). Many microbial species do not exist independently of their hosts in the environment (Thompson et al., 2017), meaning that they cannot be horizontally acquired from the environment by subsequent generations if they are lost from a host due to dietary changes, antibiotic exposure, captivity, founder effects or other disturbance (Bello, Knight, Gilbert, & Blaser, 2018; Sonnenburg & Sonnenburg, 2019). Thus, the loss of bacterial species from the microbiome can compound over subsequent generations (Sonnenburg & Sonnenburg, 2019), resulting in a stepwise reduction in diversity akin to an extinction vortex (see fig. 1 in Blaser & Falkow, 2009). This loss of diversity should trouble us, as there is intrinsic conservation value in microbial genomic diversity, regardless of potential effects on host organisms. Of course, it is also possible that other microbes could fulfil lost functions for hosts, through functional redundancy (Louca et al., 2018; Moya & Ferrer, 2016). At this time, however, we do not have much evidence for whether functional redundancy is common in human, plant or wild animal microbiomes. As our understanding of the functional contributions of different microbiome members develops, we will increasingly be able to identify the sets of micro-organisms likely contributing to host health, development and behaviour, to better determine which microbiome components should be prioritized in conservation decision-making.

1.3 | Captivity for conservation

Last-ditch efforts to save endangered animals and plants may be made through captive breeding and ex situ seed banking (hereafter 'captivity')—usually with great difficulty, and at great cost (Balmford, Mace, & Leader-Williams, 1996). In other cases, captivity is a prerequisite for conservation. For example, in Australia, feral predators such as cats *Felis catus* and foxes *Vulpes vulpes* decimate native wildlife, meaning that many native animals persist only within predator-proof exclosures (Moseby, Carthey, & Schroeder, 2015). In each of these cases, we are well aware of the need to maintain genetic diversity and avoid inbreeding in captivity (Reed & Frankham, 2003), though a microbial perspective on captivity also offers many chances to improve conservation outcomes. Given the new research technologies available, and what we are learning about the likely critical role of the microbiome in host health and functioning, now is an opportune time for conservation scientists and practitioners to consider how best to conserve host-associated microbial diversity when planning captivity or other high-risk conservation actions.

Captivity differs from the wild in important ways that are likely to impact microbial diversity (McKenzie et al., 2017; Trevelline, Fontaine, Hartup, & Kohl, 2019; Tsukayama et al., 2018; West et al., 2019; Table 2). Captive animals are fed less varied, more processed (e.g. pelleted foods), and more hygienic diets than they would consume in the wild. Exposure to harmful microbes is limited through hygienic practices and sanitization procedures, inadvertently also vastly reducing the opportunity for captive animals to acquire new microbes. Animal feed is regularly mixed with antibiotics, and veterinary treatment for captive animals includes a similar array of antibiotics as those used in human medicine (with its documented ill-effects on the diversity of the human microbiota—Gillings et al., 2015). Upon arrival into captivity, animals may be quarantined and treated with antibiotics to ensure they will not introduce new diseases into the established populations. Finally, animals in captivity may be kept in smaller groups than they naturally form in the wild, leading to fewer intraspecific interactions and a potential consequent decline in the microbial diversity available for transmission between individuals.

Each of these facets of captivity, while effective in minimizing disease risks, may have unintended consequences for captive animal microbiomes (Table 2). These impacts might be worse for captive-born and reared animals, which would be subject to multiple reductions in opportunities to acquire a diverse set of microbes, resulting in a stepwise loss of diversity (Blaser & Falkow, 2009). For example, the human gut microbiome is established within the first 2–3 years of life, and then remains relatively stable, unless antibiotics or other destabilizing events occur (Gillings et al., 2015; Yatsunenko et al., 2012). Captive-born and reared animals might not experience the opportunity to build a natural and functionally diverse microbiome. The current evidence confirms that captivity alters the community structure, and in many cases, the diversity of animal gut microbiota (e.g. Borbon-Garcia, Reyes, Vives-Florez, & Caballero, 2017; Cheng et al., 2015; Delport, Power, Harcourt, Webster, & Tetu, 2016; McKenzie et al., 2017; Wasimuddin et al., 2017), although there is little evidence to date equating these changes with harm.

The arguments set out above for management of captive animal populations likely apply equally to plants. The potential to

TABLE 2 Differences between the wild and captivity that might affect opportunities for microbial exposure and maintenance of a microbiome, once acquired

	Wild	Captivity	Predicted effect of captivity on microbial diversity
Water	Waterhole—soil, carcasses, faecal matter, bacteria from con- and hetero-specifics	Clean water (filtered?), regularly disinfected vessel	Eliminate or drastically reduce opportunities for acquisition
Food	Diverse	More prescriptive and restricted	Drastically reduce opportunities for acquisition
Predators and scavengers	Prey gut contents often consumed. Other prey microbiota consumed. Communal consumption of carcass—opportunity for transmission from con and hetero-specifics	Clean, hygienic pieces of muscle (i.e. less guts, entire animals, offal?)	
Herbivores	Environmental bacteria on plants	Clean and hygienic plant and vegetable matter, pellet foods	
Environment	Environmental pool of bacterial diversity can be acquired, for example, through soil, contact with other organisms, etc.	Limited opportunity for contact with soil rich in microbes, and with other organisms	
Group size	Freely determined	Could be smaller or larger than the wild, most often probably smaller	Smaller group size: reduced opportunity for acquisition. Larger group size: greater opportunity for acquisition
Interactions with other species	Freely determined	Drastically reduced in most cases, occasionally increased	Where reduced: reduced opportunity for acquisition, where increased: increased opportunity for acquisition
Antibiotics	None	Potentially in feed, accompanying medical procedures, prior to release for reintroductions	Drastically reduce existing diversity
Quarantine	None	Potentially whenever sick, upon initial introduction to a captive population	Reduce existing diversity

harness particular microbial species for plant improvement has been known for a long time, but the complexities of manipulating whole microbial populations for plant health and sustainable agriculture are only now beginning to be explored (Busby et al., 2017; Toju et al., 2018; Wallenstein, 2017). Beyond this practical application of plant holobiont studies for agriculture and food production, the principles underlying assembly of plant/microbial associations are receiving increasing attention (Fitzpatrick et al., 2018; Sasse, Martinoia, & Northen, 2018). While it is clear that the microbiome is as important for plants as it is for animals (Vandenkoornhuyse et al., 2015), whether microbial diversity is being lost as a consequence of our management of cropping and grazing lands is less certain.

Like animals in captivity, plants are routinely conserved using strategies of last resort such as ex situ seed banks and living collections in botanic gardens. The Millennium Seed Bank (MSB) stores frozen seed samples from almost 40,000 plant species and is designed to provide long-term insurance against extinction in the wild (https://www.kew.org/science/collections/seed-collection). Seed banks, by definition, store only the essential reproductive material needed to resurrect plant species, yet evidence continues to emerge about the importance of micro-organisms in maintaining seed viability, and increasing germination and establishment success (Sarmiento et al., 2017; Shade, Jacques, & Barret, 2017; Torres-Cortés et al., 2018). Seed banks are already conserving seed and symbionts in parallel for some select taxa, such as orchids, which are well known to require obligate fungal symbionts for germination and development (Arditti, 1967; Batty, Dixon, Brundrett, & Sivasithamparam, 2001; Rasmussen, Dixon, Jersáková, & Těšitelová, 2015). Given the MSB seeks to effectively bank seed from 25% of all flora by 2020, there is an urgent need to accelerate our knowledge about the role of micro-organisms across a far wider breadth of plant families. For instance, what role do vertically transmitted microbiota (mother to offspring) contained within internal seed tissues play in shaping seed viability? Or, how do routine practices such as seed surface sterilization impact the seed microbiome? Managing the antagonism between the need to conserve beneficial microbiota while also excluding harmful pathogens when banking seed for long-term storage is a key challenge.

1.4 | Translocation, assisted colonization and reintroduction

The consequences of ignoring plant (and animal) symbionts may also inhibit the success of higher risk conservation measures such as translocation or assisted colonization under climate change. Intentional introduction of populations to novel locations (translocation), sometimes in anticipation of more suitable future climatic habitat (assisted colonization), will be an essential part of conservation in the Anthropocene (Seddon, 2010). Yet insights about how the plant holobiont (e.g., phyllosphere, rhizosphere microbes) influences plant growth and survivorship are just beginning to be incorporated into approaches to translocation. For instance, the transplantation of soil-associated microbes may increase translocation success in newly established insurance populations of the rare and threatened Wollemi pine *Wollemia nobilis* (Rigg et al., 2017). Plants have formed associations with micro-organisms throughout their evolution and are thought to be able to modulate their microbiota to improve fitness in different environments (Vandenkoornhuyse et al., 2015). Plants can modify their exudate profiles, root morphology and immune system activities in ways that are thought to assist with selecting a beneficial microbiome (Rodriguez et al., 2019; Sasse et al., 2018), and there is evidence to suggest that response to abiotic stress is in part mediated by adaptive changes to the host–microbiome (Fitzpatrick et al., 2018). However, recruitment of a beneficial microbiome relies on having sufficient microbial diversity to select from in the surrounding environment and plant transplantations as part of conservation efforts may therefore benefit from greater consideration of soil microbiota.

For animals, reintroductions of captive-bred animals often fail due to high mortality from predation (Short, Bradshaw, Giles, Prince, & Wilson, 1992). In some cases, this is due to relaxed selection after some generations without predation risk (e.g. Jolly, Webb, & Phillips, 2018), and in others it may be due to evolutionary naivety towards alien predators (Carthey & Blumstein, 2018). However, while speculative, we must consider how a gut microbiome altered by captivity may affect behaviour relevant to predator avoidance. There is substantial evidence that the gut microbiota affect stress-related behaviours such as freezing and exploration (reviewed by Vuong et al., 2017). For example, germ-free laboratory mice (Heijtza et al., 2011; Neufeld, Kang, Bienenstock, & Foster, 2011; Vuong et al., 2017) are more willing to explore open space than laboratory mice with normal gut microbiota. Abnormalities in exploratory behaviours were re-established by restoring a normal gut microbiota—but only in juvenile, not adult mice (Heijtza et al., 2011; Neufeld et al., 2011). While this evidence comes from highly controlled laboratory experiments rather than the field, it is worth considering the implications of altered gut microbiota in captive animals intended for release. Could an altered microbiome influence risk-taking and fear-based behaviours in released animals? These simple but powerful ideas are yet to be systematically explored in the context of translocation, assisted colonization and reintroduction.

1.5 | Microbially mediated communication

In animals, microbes that colonize body regions other than the gut and skin may also have important influences on phenotype. For example, evidence is mounting in support of the fermentation hypothesis of olfactory communication (Ezenwa & Williams, 2014; Leclaire, Nielsen, & Drea, 2014; Theis et al., 2013; Whittaker et al., 2016), which proposes that resident microbes act upon animal excretions and secretions to create a diverse array of chemical compounds that animals use for olfactory communication (Albone & Shirley, 1984; Gorman, Nedwell, & Smith, 1974), effectively extending the host animal's phenotype and genotype (Carthey, Gillings, & Blumstein, 2018).

Culture-independent bacterial surveys of animal scent glands suggest that bacterial community structure in the host can affect volatile fatty acid profiles, and thus inter-individual communication. The bacterial communities and fatty acids of wild spotted and striped hyena scent marks covary with species, social group membership, and with sex and reproductive status within a social group. These bacterial species belong to fermentative bacterial families—strongly suggesting that microbes play a role in the production of hyena scent used in communication (Theis et al., 2013). Similarly, the microbiota found in meerkat anal scent secretions vary with age, sex and group membership—all characteristics that meerkats communicate through scent (Leclaire et al., 2014). In another example, bacteria have been shown to produce a key component of desert locust *Schistocerca gregaria* aggregating pheromone, guaiacol (Dillon, Vennard, & Charnley, 2002). If microbes mediate olfactory communication in many animals, and the right skin and/or scent gland microbiome is key to communication and the behaviour it governs, we must expect that disrupted or depauperate microbiomes could interfere with communication and hence, the performance of natural behaviours.

Olfactory communication is extremely widespread among animals, and facilitates complex social interactions among many vertebrates (Bienenstock, Kunze, & Forsythe, 2018; Wyatt, 2014). The revelation that microbes could be essential to this process forces a rethink of many aspects of behavioural and sensory ecology (Archie & Theis, 2011; Archie & Tung, 2015; Bienenstock et al., 2018). The recognition that microbially mediated olfactory communication is an important driver of vertebrate behaviour has led to a recent call to incorporate olfaction into the microbiome–gut–brain axis (Bienenstock et al., 2018). Microbes may influence behaviour via direct neuronal connections between the gut and the brain, via their effect on physiology (Bienenstock et al., 2018), and via their effect on animal communication, via either the gut or through scent glands (Carthey et al., 2018). The microbial role in olfactory communication suggests we must reconsider much of what we know about signalling and information theory as it applies to this mode of animal interactions.

We know that captivity will influence the gut microbiota (McKenzie et al., 2017), and it seems likely that these changes will consequently will also affect any microbiota used in olfactory signalling and communication. If animals cannot acquire or maintain the microbial diversity required for communication and hence natural behaviours, captive breeding is likely to remain difficult. Animals that rely on olfactory cues and complex courtship behaviours are most likely to be affected by an altered scent-producing microbiome. Where captive breeding is currently failing, changes to microbially mediated signals associated with captivity could be investigated.

1.6 | Towards a new era of holobiont-focused conservation

This is an opportune time for conservation scientists to work collaboratively with microbiologists to improve conservation outcomes for threatened species. The field of microbiome research is in its relative infancy, even for the human gut microbiome. Therefore, a large amount of experimental research is still required before we can conclude that observed changes to plant and animal microbiomes in the Anthropocene cause harm. Yet, the evidence from the human research is clear—disturbed human gut microbiomes are closely linked to poor health outcomes and to chronic non-communicable diseases that have become prevalent in the modern age. In recognition of the potential for great harms from disappearing microbes in the Anthropocene, multiple global consortia have been launched with the intention of cataloguing and preserving microbial diversity. For example, the Human Microbiome Project (Turnbaugh et al., 2007), the Earth Microbiome Project (Thompson et al., 2017), the Global Microbiome Conservancy (http://microbiomeconservancy.org/) and the Microbiome Vault (Bello et al., 2018) are each concerned with ensuring that the required microbial diversity will be available to future generations seeking to repopulate and regenerate depauperate microbiomes. There is a great opportunity here for conservation biologists to be involved in such projects, and lobby for the preservation of not just human but wild animal- and plant-associated microbiota. However, the vast majority of microbes currently remain uncultured, meaning it may be difficult or impossible to effectively preserve them.

Some conservation plans for threatened species take into account the need to conserve associated microbiota along with a host of interest—for example, the Save the Tasmanian Devil Project in Tasmania, Australia (Cheng et al., 2015). Though a rapid search of 30 Species Recovery Plans for a suite of plants, mammals, birds, reptiles and invertebrates listed as threatened on the Australian *Environmental Protection and Biodiversity Conservation Act 1995* (Appendix S1) finds no reference to various terms associated with micro-organisms (i.e. 'microbiome', 'micro-organism', 'micro-organism', 'bacteria' or 'microbe'). These recovery plans did refer to 'pathogens' and 'parasites' but only in reference to threatening processes that may affect species long-term viability.

A precautionary approach would assume that disturbed microbiomes are likely to be harmful to plants and animals, until proven otherwise. At the same time, we acknowledge that all conservation management decisions are made under constraints, such that actions to conserve microbial diversity must be weighed against other actions to reduce potential harms. In the absence of hard evidence linking disturbed microbiomes to harm, we propose that conservation management decisions at least consider how the transmission of helpful microbes among plants and animals might be facilitated, while limiting the impact of pathogenic microbes. For captive animals, this might be as simple as providing dietary offal including prey stomachs, providing access to as many natural environmental elements such as soil and vegetation as possible, and interactions with con- and hetero-specifics that mirror natural conditions as closely as possible. From a research perspective, if such an approach were designed to allow comparison with procedural controls, evidence for the potential benefits of encouraging microbial transmission could be gathered.

As a potential solution to depauperate or disturbed animal microbiomes, microbial transplants from wild to captive animals are beginning to be considered. Faecal transplants have shown remarkable success in restoring healthy microbiomes in human patients afflicted with gut disorders such as recurrent *Clostridium difficile* infection, presumably by restoring a healthy microbial community (Pamer, 2014). Faecal transplants are currently being explored as a means of expanding vulnerable species' diets (Miller, Dale, & Dearing, 2017; Miller, Oakeson, Dale, & Dearing, 2016). For example, Australian koalas *Phascolarctos cinereus* are an extremely specialized herbivore, with individual koalas having gut microbiomes that are finely attuned to the digestion and metabolism of particular species of food tree (*Eucalyptus* spp.). Researchers recently showed that koalas were unable to switch between food tree species without the help of a faecal transplant pellet, containing a live microbiome sample from koalas that regularly ate the target tree species. Koalas that received the transplant established a gut microbiome more similar to koalas that ate the target species regularly, and were able to transition their diet to consume more of the target tree species (Blyton et al., 2019). This is one of the first studies to experimentally investigate the use of faecal transplants for conservation purposes—in this case, allowing dietary expansion in a highly specialized wild herbivore.

By broadening our conservation toolkit, we can generate novel solutions to formerly intractable problems. For instance, until recently southern white rhinoceroses *Ceratotherium simum simum* were very difficult to breed in captivity. This was in stark contrast to the ease with which greater one-horned rhinoceros *Rhinoceros unicornis* were bred. Researchers discovered that the different gut microbiota of the two species were digesting dietary plant phytoestrogens differently, such that these hormone-mimicking phytoestrogens were being excreted by female southern white rhinoceroses and interfering with fertility/mating behaviours. Altering the diet to exclude plants high in phytoestrogens rapidly led to breeding success in southern white rhinoceroses (Williams, Ybarra, Meredith, Durrant, & Tubbs, 2019). Further research (Gibson et al., 2019) found that captive black rhinoceros gut microbiomes were distinctly different to those of wild black rhinoceroses, and that captive microbiomes were populated with bacterial species commonly found in domesticated animals. Functional profiling of the microbiomes indicates changes in metabolic capabilities from breakdown of fibrous material to one of greater carbohydrate metabolism. While such changes may reflect adaptation of the captive rhinoceros gut microbiomes to dietary content, the question remains as to whether such changes may be beneficial or harmful to the host, reflecting a mismatch between microbiome and host as suggested by Sonnenburg and Sonnenburg (2019) for humans. Clearly, a focus on microbes has had and will continue to have important implications for rhinoceros management.

In plant agricultural systems, soil microbiota have been manipulated for centuries to improve yield, though practices to support healthy translocated populations of threatened plants have not been systematically investigated. For example, the transfer of leaf litter from healthy conspecifics of the crop plant cacao *Theobroma cacao* introduced beneficial fungal endophytes which reduced pathogen damage (Christian, Herre, Mejia, & Clay, 2017). Similarly, transfer of the foliar microbiome from wild relatives to the critically endangered plant *Phyllostegia kaalaensis* (Lamiaceae) significantly reduced disease rates and decreased dependence on fungicidal applications which otherwise maintain populations (Zahn & Amend, 2017). Finally, community cultivation initiatives designed to keep plant populations in natural habitats rather than in ex situ collections (Willis, 2017) are likely to maintain a greater diversity of plant-associated microbiota, conserving the plant holobiont rather than just the plant species.

1.7 | Key future foci for holobiont-focused conservation

Our review has revealed six key unanswered questions that we put forward as key future foci for conservation that incorporates the holobiont.

1. What are our options for conserving microbial diversity?

In reality, we do not yet know enough about the intricacies of host–microbiome interactions to rely on microbiome vault projects. The most practical, immediate approach is to take measures that improve the conservation of host-associated microbiota in vivo. That is, conserving the host species but also its microbiota, through microbiota-friendly practices such as reduced use of antibiotics and excessive hygiene.

2. Is functional redundancy common in plant and/or wild animal microbiomes?

Answering this question with targeted research is important because if functional redundancy is common, the loss of individual microbial taxa from a host species' microbiome may be less cause for concern.

3. Do disturbed microbiomes easily revert to a healthy state once disturbance ceases?

For example, a key question is if captive-type animal gut microbiomes revert to a wild type upon release of the host animal? We might predict that reversion to wild type is only likely if wild conspecifics with healthy microbiomes persist in the environment.

4. How can we best determine potential harms and benefits from altered microbiomes?

Linking microbial taxonomic diversity to functional capacity and related effects on host health is a critical next step towards understanding how best to conserve the holobiont.

5. How can we use the holobiont concept to improve seed banking, translocation, reintroduction and captive breeding successes?

We can immediately implement low-cost actions aimed at conserving host-associated microbiota, for example by reducing routine exposure to antimicrobials.

6. Could an altered microbiome influence risk-taking and fear-based behaviours in released animals, with implications for vulnerability to predation?

This question could be answered by pre-release microbiome profiling and fear-based behavioural assays, which could then be linked to post-release survival.

2 | CONCLUSIONS

Conservation science aims to protect threatened species from extinction yet has so far not widely considered the critical role of host-associated microbiota in healthy development and function. Through effects on physiology, competitive interactions, behaviour and communication, the microbiome of a species targeted for conservation is likely to be critical for maintaining healthy populations. We recognize that conservation of genetic diversity is important for maintaining animal and plant populations (Frankham, 1995, 2005). However, the genetic diversity embodied in microbial symbionts can be orders of magnitude larger than that of the nuclear genome, with the consequent potential for even greater loss of function. While this microbial diversity is not always reliably transmitted from generation to generation, we nevertheless need to consider how conservation practices might reduce its potential for transmission, particularly in artificial environments.

The holobiont concept recognizes that animals and plants comprise a host genome and an associated microbial genome, both of which must be protected if our goal is effective conservation. The spatial and temporal diversity of microbial symbionts is poorly known for the majority of organisms, but it is clear that contractions in ranges and population sizes of macro-organisms are likely to be accompanied by parallel contractions in their microbial diversity. We can no longer attempt to conserve only the macro-organism, but must also conserve its attendant microbiome. The target of our conservation strategies should be the holobiont.

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AUTHORS' CONTRIBUTIONS

A.J.R.C., D.T.B. and M.R.G. conceived the manuscript, and A.J.R.C. wrote the initial draft. R.V.G. and S.G.T. contributed the material on plants and their microbiota. All authors edited and reviewed the manuscript prior to submission.

DATA AVAILABILITY STATEMENT

There is no data repository associated with this review.

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REFERENCES

Alberdi, A., Aizpurua, O., Bohmann, K., Zepeda-Mendoza, M. L., & Gilbert, M. T. P. (2016). Do vertebrate gut metagenomes confer rapid ecological adaptation? *Trends in Ecology & Evolution*, *31*, 689–699. https://doi.org/10.1016/j.tree.2016.06.008

Albone, E. S., & Shirley, S. G. (1984). *Mammalian semiochemistry: The investigation of chemical signals between mammals*. Chichester, UK: John Wiley & Sons.

Archie, E. A., & Theis, K. R. (2011). Animal behaviour meets microbial ecology. *Animal Behaviour*, *82*, 425–436. https://doi.org/10.1016/j.anbehav.2011.05.029

Archie, E. A., & Tung, J. (2015). Social behavior and the microbiome. *Current Opinion in Behavioral Sciences*, *6*, 28–34. https://doi.org/10.1016/j.cobeha.2015.07.008

Arditti, J. (1967). Factors affecting the germination of orchid seeds. *The Botanical Review*, *33*, 1–97. https://doi.org/10.1007/BF02858656

Armour, C. R., Nayfach, S., Pollard, K. S., & Sharpton, T. J. (2019). A metagenomic meta-analysis reveals functional signatures of health and disease in the human gut microbiome. *mSystems*, *4*, e00332-18. https://doi.org/10.1128/mSystems.00332-18

Balmford, A., Mace, G. M., & Leader-Williams, N. (1996). Designing the ark: Setting priorities for captive breeding. *Conservation Biology*, *10*, 719–727. https://doi.org/10.1046/j.1523-1739.1996.10030719.x

Batty, A., Dixon, K., Brundrett, M., & Sivasithamparam, K. (2001). Long-term storage of mycorrhizal fungi and seed as a tool for the conservation of endangered Western Australian terrestrial orchids. *Australian Journal of Botany*, *49*, 619–628. https://doi.org/10.1071/BT01029

Belizário, J. E., & Napolitano, M. (2015). Human microbiomes and their roles in dysbiosis, common diseases, and novel therapeutic approaches. *Frontiers in Microbiology*, *6*, 1050. https://doi.org/10.3389/fmicb.2015.01050

Bello, M. G. D., Knight, R., Gilbert, J. A., & Blaser, M. J. (2018). Preserving microbial diversity. *Science*, *362*, 33–34. https://doi.org/10.1126/science.aau8816

Bienenstock, J., Kunze, W. A., & Forsythe, P. (2018). Disruptive physiology: Olfaction and the microbiome–gut–brain axis. *Biological Reviews*, *93*, 390–403. https://doi.org/10.1111/brv.12348

Blaser, M. J., & Falkow, S. (2009). What are the consequences of the disappearing human microbiota? *Nature Reviews Microbiology*, *7*, 887. https://doi.org/10.1038/nrmicro2245

Blyton, M. D. J., Soo, R. M., Whisson, D., Marsh, K. J., Pascoe, J., Le Pla, M., … Moore, B. D. (2019). Faecal inoculations alter the gastrointestinal microbiome and allow dietary expansion in a wild specialist herbivore, the koala. *Animal Microbiome*, *1*, 6. https://doi.org/10.1186/s42523-019-0008-0

Borbon-Garcia, A., Reyes, A., Vives-Florez, M., & Caballero, S. (2017). Captivity shapes the gut microbiota of Andean bears: Insights into health surveillance. *Frontiers in Microbiology*, *8*, 1316. https://doi.org/10.3389/fmicb.2017.01316

Brucker, R. M., & Bordenstein, S. R. (2012). Speciation by symbiosis. *Trends in Ecology & Evolution*, *27*, 443–451. https://doi.org/10.1016/j.tree.2012.03.011

Busby, P. E., Soman, C., Wagner, M. R., Friesen, M. L., Kremer, J., Bennett, A., … Dangl, J. L. (2017). Research priorities for harnessing plant microbiomes in sustainable agriculture. *PLoS Biology*, *15*, e2001793. https://doi.org/10.1371/journal.pbio.2001793

Carthey, A. J. R., & Blumstein, D. T. (2018). Predicting predator recognition in a changing world. *Trends in Ecology & Evolution*, *33*, 106–115. https://doi.org/10.1016/j.tree.2017.10.009

Carthey, A. J. R., Gillings, M. R., & Blumstein, D. T. (2018). The extended genotype: Microbially mediated olfactory communication. *Trends in Ecology & Evolution*, *33*, 885–894. https://doi.org/10.1016/j.tree.2018.08.010

Cheng, Y. Y., Fox, S., Pemberton, D., Hogg, C., Papenfuss, A. T., & Belov, K. (2015). The Tasmanian devil microbiome—Implications for conservation and management. *Microbiome*, *3*, 76. https://doi.org/10.1186/s40168-015-0143-0

Christian, N., Herre, E. A., Mejia, L. C., & Clay, K. (2017). Exposure to the leaf litter microbiome of healthy adults protects seedlings from pathogen damage. *Proceedings of the Royal Society B: Biological Sciences*, *284*, 20170641.

Clemente, J. C., Ursell, L. K., Parfrey, L. W., & Knight, R. (2012). The impact of the gut microbiota on human health: An integrative view. *Cell*, *148*, 1258–1270. https://doi.org/10.1016/j.cell.2012.01.035

Compant, S., Samad, A., Faist, H., & Sessitsch, A. (2019). A review on the plant microbiome: Ecology, functions, and emerging trends in microbial application. *Journal of Advanced Research*, *19*, 29–37. https://doi.org/10.1016/j.jare.2019.03.004

Delavaux, C. S., Weigelt, P., Dawson, W., Duchicela, J., Essl, F., van Kleunen, M., … Bever, J. D. (2019). Mycorrhizal fungi influence global plant biogeography. *Nature Ecology & Evolution*, *3*, 424–429. https://doi.org/10.1038/s41559-019-0823-4

Delport, T. C., Power, M. L., Harcourt, R. G., Webster, K. N., & Tetu, S. G. (2016). Colony location and captivity influence the gut microbial community composition of the Australian sea lion (*Neophoca cinerea*). *Applied and Environmental Microbiology*, *82*, 3440–3449. https://doi.org/10.1128/AEM.00192-16

Dillon, R. J., Vennard, C. T., & Charnley, A. K. (2002). A note: Gut bacteria produce components of a locust cohesion pheromone. *Journal of Applied Microbiology*, *92*, 759–763. https://doi.org/10.1046/j.1365-2672.2002.01581.x

Engel, K., Sauer, J., Jünemann, S., Winkler, A., Wibberg, D., Kalinowski, J., … Caspers, B. A. (2018). Individual- and species-specific skin microbiomes in three different estrildid finch species revealed by 16S amplicon sequencing. *Microbial Ecology*, *76*, 518–529. https://doi.org/10.1007/s00248-017-1130-8

Ezenwa, V. O., Gerardo, N. M., Inouye, D. W., Medina, M., & Xavier, J. B. (2012). Animal behavior and the microbiome. *Science*, *338*, 198–199. https://doi.org/10.1126/science.1227412

Ezenwa, V. O., & Williams, A. E. (2014). Microbes and animal olfactory communication: Where do we go from here? *BioEssays*, *36*, 847–854. https://doi.org/10.1002/bies.201400016

Ferretti, P., Pasolli, E., Tett, A., Asnicar, F., Gorfer, V., Fedi, S., … Segata, N. (2018). Mother-to-infant microbial transmission from different body sites shapes the developing infant gut microbiome. *Cell Host & Microbe*, *24*, 133–145.e135. https://doi.org/10.1016/j.chom.2018.06.005

Fitzpatrick, C. R., Copeland, J., Wang, P. W., Guttman, D. S., Kotanen, P. M., & Johnson, M. T. J. (2018). Assembly and ecological function of the root microbiome across angiosperm plant species. *Proceedings of the National Academy of Sciences of the United States of America*, *115*, E1157–E1165. 201717617.

Frankham, R. (1995). Conservation genetics. *Annual Review of Genetics*, *29*, 305–327. https://doi.org/10.1146/annurev.ge.29.120195.001513

Frankham, R. (2005). Genetics and extinction. *Biological Conservation*, *126*, 131–140. https://doi.org/10.1016/j.biocon.2005.05.002

Franzosa, E. A., McIver, L. J., Rahnavard, G., Thompson, L. R., Schirmer, M., Weingart, G., … Huttenhower, C. (2018). Species-level functional profiling of metagenomes and metatranscriptomes. *Nature Methods*, *15*, 962–968. https://doi.org/10.1038/s41592-018-0176-y

Gibson, K. M., Nguyen, B. N., Neumann, L. M., Miller, M., Buss, P., Daniels, S., … Pukazhenthi, B. (2019). Gut microbiome differences between wild and captive black rhinoceros – Implications for rhino health. *Scientific Reports*, *9*, 7570. https://doi.org/10.1038/s41598-019-43875-3

Gilbert, J. A., Quinn, R. A., Debelius, J., Xu, Z. J. Z., Morton, J., Garg, N., … Knight, R. (2016). Microbiome-wide association studies link dynamic microbial consortia to disease. *Nature*, *535*, 94–103. https://doi.org/10.1038/nature18850

Gillings, M. R., & Paulsen, I. T. (2014). Microbiology of the Anthropocene. *Anthropocene*, *5*, 1–8. https://doi.org/10.1016/j.ancene.2014.06.004

Gillings, M. R., Paulsen, I. T., & Tetu, S. G. (2015). Ecology and evolution of the human microbiota: Fire, farming and antibiotics. *Genes*, *6*, 841–857. https://doi.org/10.3390/genes6030841

Gilmore, M. S., & Ferretti, J. J. (2003). Microbiology: The thin line between gut commensal and pathogen. *Science*, *299*, 1999–2002. https://doi.org/10.1126/science.1083534

Gissis, S. B., Lamm, E., & Shavit, A. (2018). *Landscapes of collectivity in the life sciences*. Cambridge, UK: MIT Press.

Gorman, M., Nedwell, D. B., & Smith, R. M. (1974). An analysis of the contents of the anal scent pockets of *Herpestes auropunctatus* (Carnivora: Viverridae). *Journal of Zoology*, *172*, 389–399. https://doi.org/10.1111/j.1469-7998.1974.tb04115.x

Hadrich, D. (2018). Microbiome research is becoming the key to better understanding health and nutrition. *Frontiers in Genetics*, *9*, 212. https://doi.org/10.3389/fgene.2018.00212

Hanning, I., & Diaz-Sanchez, S. (2015). The functionality of the gastrointestinal microbiome in non-human animals. *Microbiome*, *3*, 51. https://doi.org/10.1186/s40168-015-0113-6

Harris, R. N., James, T. Y., Lauer, A., Simon, M. A., & Patel, A. (2006). Amphibian pathogen *Batrachochytrium dendrobatidis* is inhibited by the cutaneous bacteria of amphibian species. *EcoHealth*, *3*, 53. https://doi.org/10.1007/s10393-005-0009-1

Heijtza, R. D., Wang, S. G., Anuar, F., Qian, Y., Bjorkholm, B., Samuelsson, A., … Pettersson, S. (2011). Normal gut microbiota modulates brain development and behavior. *Proceedings of the National Academy of Sciences of the United States of America*, *108*, 3047–3052. https://doi.org/10.1073/pnas.1010529108

Heintz-Buschart, A., & Wilmes, P. (2018). Human gut microbiome: Function matters. *Trends in Microbiology*, *26*, 563–574. https://doi.org/10.1016/j.tim.2017.11.002

Hernandez-Agreda, A., Gates, R. D., & Ainsworth, T. D. (2017). Defining the core microbiome in corals' microbial soup. *Trends in Microbiology*, *25*, 125–140. https://doi.org/10.1016/j.tim.2016.11.003

Hird, S. M. (2017). Evolutionary biology needs wild microbiomes. *Frontiers in Microbiology*, *8*, 725. https://doi.org/10.3389/fmicb.2017.00725

Huttenhower, C., Gevers, D., Knight, R., Abubucker, S., Badger, J. H., Chinwalla, A. T. … White, O.; The Human Microbiome Project Consortium. (2012). Structure, function and diversity of the healthy human microbiome. *Nature*, *486*, 207.

Inderjit, & van der Putten, W. H. (2010). Impacts of soil microbial communities on exotic plant invasions. *Trends in Ecology & Evolution*, *25*, 512–519. https://doi.org/10.1016/j.tree.2010.06.006

Jolly, C. J., Webb, J. K., & Phillips, B. L. (2018). The perils of paradise: An endangered species conserved on an island loses antipredator behaviours within 13 generations. *Biology Letters*, *14*, 20180222. https://doi.org/10.1098/rsbl.2018.0222

Kembel, S. W., O'Connor, T. K., Arnold, H. K., Hubbell, S. P., Wright, S. J.,& Green, J. L. (2014). Relationships between phyllosphere bacterial communities and plant functional traits in a neotropical forest. *Proceedings of the National Academy of Sciences of the United States of America*, *111*, 13715–13720. https://doi.org/10.1073/pnas.1216057111

Knutie, S. A., Wilkinson, C. L., Kohl, K. D., & Rohr, J. R. (2017). Early-life disruption of amphibian microbiota decreases later-life resistance to parasites. *Nature Communications*, *8*, 86. https://doi.org/10.1038/s41467-017-00119-0

Kohl, K. D., Dearing, M. D., & Bordenstein, S. R. (2018). Microbial communities exhibit host species distinguishability and phylosymbiosis along the length of the gastrointestinal tract. *Molecular Ecology*, *27*, 1874–1883.

Kreznar, J. H., Keller, M. P., Traeger, L. L., Rabaglia, M. E., Schueler, K. L., Stapleton, D. S., … Rey, F. E. (2017). Host genotype and gut microbiome modulate insulin secretion and diet-induced metabolic phenotypes. *Cell Reports*, *18*, 1739–1750. https://doi.org/10.1016/j.celrep.2017.01.062

Laforest-Lapointe, I., Messier, C., & Kembel, S. W. (2017). Tree leaf bacterial community structure and diversity differ along a gradient of urban intensity. *mSystems*, *2*, e00087-e17. https://doi.org/10.1128/mSystems.00087-17

Leclaire, S., Nielsen, J. F., & Drea, C. M. (2014). Bacterial communities in meerkat anal scent secretions vary with host sex, age, and group membership. *Behavioral Ecology*, *25*, 996–1004. https://doi.org/10.1093/beheco/aru074

Li, J. H., Jia, H. J., Cai, X. H., Zhong, H. Z., Feng, Q., Sunagawa, S., … Meta, H. I. T. C. (2014). An integrated catalog of reference genes in the human gut microbiome. *Nature Biotechnology*, *32*, 834–841. https://doi.org/10.1038/nbt.2942

Lizé, A., Mckay, R., & Lewis, Z. (2013). Gut microbiota and kin recognition. *Trends in Ecology & Evolution*, *28*, 325–326. https://doi.org/10.1016/j.tree.2012.10.013

Louca, S., Polz, M. F., Mazel, F., Albright, M. B. N., Huber, J. A., O'Connor, M. I., … Parfrey, L. W. (2018). Function and functional redundancy in microbial systems. *Nature Ecology & Evolution*, *2*, 936–943. https://doi.org/10.1038/s41559-018-0519-1

Manor, O., & Borenstein, E. (2017). Systematic characterization and analysis of the taxonomic drivers of functional shifts in the human microbiome. *Cell Host & Microbe*, *21*, 254–267. https://doi.org/10.1016/j.chom.2016.12.014

McKenzie, V. J., Song, S. J., Delsuc, F., Prest, T. L., Oliverio, A. M., Korpita, T. M., … Knight, R. (2017). The effects of captivity on the mammalian gut microbiome. *Integrative and Comparative Biology*, *57*, 690–704. https://doi.org/10.1093/icb/icx090

Miller, A. W., Dale, C., & Dearing, M. D. (2017). The induction of oxalate metabolism in vivo is more effective with functional microbial communities than with functional microbial species. *mSystems*, *2*, e00088-e117.

Miller, A. W., Oakeson, K. F., Dale, C., & Dearing, M. D. (2016). Microbial community transplant results in increased and long-term oxalate degradation. *Microbial Ecology*, *72*, 470–478. https://doi.org/10.1007/s00248-016-0800-2

Morar, N., & Bohannan, B. J. M. (2019). The conceptual ecology of the human microbiome. *Quarterly Review of Biology*, *94*, 149–175. https://doi.org/10.1086/703582

Moseby, K., Carthey, A., & Schroeder, T. (2015). The influence of predators and prey naivety on reintroduction success: Current and future directions. In *Advances in reintroduction biology of Australian and New Zealand fauna* (pp. 29–42). Clayton South: CSIRO.

Moya, A., & Ferrer, M. (2016). Functional redundancy-induced stability of gut microbiota subjected to disturbance. *Trends in Microbiology*, *24*, 402–413. https://doi.org/10.1016/j.tim.2016.02.002

Needham, B. D., Tang, W., & Wu, W. L. (2018). Searching for the gut microbial contributing factors to social behavior in rodent models of autism spectrum disorder. *Developmental Neurobiology*, *78*, 474–499.

Neufeld, K.-A.-M., Kang, N., Bienenstock, J., & Foster, J. A. (2011). Effects of intestinal microbiota on anxiety-like behavior. *Communicative & Integrative Biology*, *4*, 492–494. https://doi.org/10.4161/cib.15702

Nishida, A. H., & Ochman, H. (2018). Rates of gut microbiome divergence in mammals. *Molecular Ecology*, *27*, 1884–1897.

Orr, M. R., Kocurek, K. M., & Young, D. L. (2018). Gut microbiota and human health: Insights from ecological restoration. *The Quarterly Review of Biology*, *93*, 73–90. https://doi.org/10.1086/698021

Palumbi, S. R. (2001). Humans as the world's greatest evolutionary force. *Science*, *293*, 1786–1790. https://doi.org/10.1126/science.293.5536.1786

Pamer, E. G. (2014). Fecal microbiota transplantation: Effectiveness, complexities, and lingering concerns. *Mucosal Immunology*, *7*, 210. https://doi.org/10.1038/mi.2013.117

Pérez-Jaramillo, J. E., Carrión, V. J., de Hollander, M., & Raaijmakers, J. M. (2018). The wild side of plant microbiomes. *Microbiome*, *6*, 143. https://doi.org/10.1186/s40168-018-0519-z

Phillips, J. N., Berlow, M., & Derryberry, E. P. (2018). The effects of landscape urbanization on the gut microbiome: An exploration into the gut of urban and rural white-crowned sparrows. *Frontiers in Ecology and Evolution*, *6*, 148. https://doi.org/10.3389/fevo.2018.00148

Rasmussen, H. N., Dixon, K. W., Jersáková, J., & Těšitelová, T. (2015). Germination and seedling establishment in orchids: A complex of requirements. *Annals of Botany*, *116*, 391–402. https://doi.org/10.1093/aob/mcv087

Reed, D. H., & Frankham, R. (2003). Correlation between fitness and genetic diversity. *Conservation Biology*, *17*, 230–237. https://doi.org/10.1046/j.1523-1739.2003.01236.x

Rigg, J. L., Offord, C. A., Zimmer, H., Anderson, I. C., Singh, B. K., & Powell, J. R. (2017). Conservation by translocation: Establishment of Wollemi pine and associated microbial communities in novel environments. *Plant and Soil*, *411*, 209–225. https://doi.org/10.1007/s11104-016-3010-2

Rodriguez, P. A., Rothballer, M., Chowdhury, S. P., Nussbaumer, T., Gutjahr, C., & Falter-Braun, P. (2019). Systems biology of plant microbiome interactions. *Molecular Plant*, *12*, 804–821. https://doi.org/10.1016/j.molp.2019.05.006

Ross, A. A., Rodrigues Hoffmann, A., & Neufeld, J. D. (2019). The skin microbiome of vertebrates. *Microbiome*, *7*, 79. https://doi.org/10.1186/s40168-019-0694-6

Roughgarden, J. (2018). Model of holobiont population dynamics and evolution: A preliminary sketch. In S. Gissis, E. Lamm, & A. Shavit (Eds.), *Landscapes of collectivity in the life sciences* (pp. 325–349). Cambridge, MA: MIT Press.

Round, J. L., & Mazmanian, S. K. (2009). The gut microbiota shapes intestinal immune responses during health and disease. *Nature Reviews Immunology*, *9*, 313.

Sampson, T. R., & Mazmanian, S. K. (2015). Control of brain development, function, and behavior by the microbiome. *Cell Host & Microbe*, *17*, 565–576. https://doi.org/10.1016/j.chom.2015.04.011

Sarmiento, C., Zalamea, P.-C., Dalling, J. W., Davis, A. S., Stump, S. M., U'Ren, J. M., & Arnold, A. E. (2017). Soilborne fungi have host affinity and host-specific effects on seed germination and survival in a lowland tropical forest. *Proceedings of the National Academy of Sciences of the United States of America*, *114*, 11458–11463. https://doi.org/10.1073/pnas.1706324114

Sasse, J., Martinoia, E., & Northen, T. (2018). Feed your friends: Do plant exudates shape the root microbiome? *Trends in Plant Science*, *23*, 25–41. https://doi.org/10.1016/j.tplants.2017.09.003

Seddon, P. J. (2010). From reintroduction to assisted colonization: Moving along the conservation translocation spectrum. *Restoration Ecology*, *18*, 796–802. https://doi.org/10.1111/j.1526-100X.2010.00724.x

Sender, R., Fuchs, S., & Milo, R. (2016). Revised estimates for the number of human and bacteria cells in the body. *PLoS Biology*, *14*, e1002533. https://doi.org/10.1371/journal.pbio.1002533

Shade, A., Jacques, M.-A., & Barret, M. (2017). Ecological patterns of seed microbiome diversity, transmission, and assembly. *Current Opinion in Microbiology*, *37*, 15–22. https://doi.org/10.1016/j.mib.2017.03.010

Sharpton, T. J. (2018). Role of the gut microbiome in vertebrate evolution. *mSystems*, *3*(2), e00174-17. https://doi.org/10.1128/mSystems.00174-17

Short, J., Bradshaw, S. D., Giles, J., Prince, R. I. T., & Wilson, G. R. (1992). Reintroduction of macropods (Marsupialia: Macropodoidea) in Australia: A review. *Biological Conservation*, *62*, 189–204. https://doi.org/10.1016/0006-3207(92)91047-V

Shreiner, A. B., Kao, J. Y., & Young, V. B. (2015). The gut microbiome in health and in disease. *Current Opinion in Gastroenterology*, *31*, 69–75. https://doi.org/10.1097/MOG.0000000000000139

Sonnenburg, E. D., & Sonnenburg, J. L. (2019). The ancestral and industrialized gut microbiota and implications for human health. *Nature Reviews Microbiology*, *17*, 383. https://doi.org/10.1038/s41579-019-0191-8

Teyssier, A., Rouffaer, L. O., Saleh Hudin, N., Strubbe, D., Matthysen, E., Lens, L., & White, J. (2018). Inside the guts of the city: Urban-induced alterations of the gut microbiota in a wild passerine. *Science of the Total Environment*, *612*, 1276–1286. https://doi.org/10.1016/j.scitotenv.2017.09.035

Theis, K. R., Dheilly, N. M., Klassen, J. L., Brucker, R. M., Baines, J. F., Bosch, T. C. G., … Bordenstein, S. R. (2016). Getting the hologenome concept right: An eco-evolutionary framework for hosts and their microbiomes. *mSystems*, *1*, e00028-e16. https://doi.org/10.1128/mSystems.00028-16

Theis, K. R., Venkataraman, A., Dycus, J. A., Koonter, K. D., Schmitt-Matzen, E. N., Wagner, A. P., … Schmidt, T. M. (2013). Symbiotic bacteria appear to mediate hyena social odors. *Proceedings of the National Academy of Sciences of the United States of America*, *110*, 19832–19837. https://doi.org/10.1073/pnas.1306477110

Thompson, L. R., Sanders, J. G., McDonald, D., Amir, A., Ladau, J., Locey, K. J., … Knight, R. (2017). A communal catalogue reveals Earth's multiscale microbial diversity. *Nature*, *551*, 457. https://doi.org/10.1038/nature24621

Toju, H., Peay, K. G., Yamamichi, M., Narisawa, K., Hiruma, K., Naito, K., … Kiers, E. T. (2018). Core microbiomes for sustainable agroecosystems. *Nature Plants*, *4*, 247–257. https://doi.org/10.1038/s41477-018-0139-4

Torres-Cortés, G., Bonneau, S., Bouchez, O., Genthon, C., Briand, M., Jacques, M.-A., & Barret, M. (2018). Functional microbial features driving community assembly during seed germination and emergence. *Frontiers in Plant Science*, *9*, 902. https://doi.org/10.3389/fpls.2018.00902

Trevelline, B. K., Fontaine, S. S., Hartup, B. K., & Kohl, K. D. (2019). Conservation biology needs a microbial renaissance: A call for the consideration of host-associated microbiota in wildlife management practices. *Proceedings of the Royal Society B*, *286*, 20182448. https://doi.org/10.1098/rspb.2018.2448

Tsukayama, P., Boolchandani, M., Patel, S., Pehrsson, E. C., Gibson, M. K., Chiou, K. L., … Dantas, G. (2018). Characterization of wild and captive baboon gut microbiota and their antibiotic resistomes. *mSystems*, *3*, e00016-18. https://doi.org/10.1128/mSystems.00016-18

Turnbaugh, P. J., Ley, R. E., Hamady, M., Fraser-Liggett, C. M., Knight, R., & Gordon, J. I. (2007). The human microbiome project. *Nature*, *449*, 804. https://doi.org/10.1038/nature06244

Valdes, A. M., Walter, J., Segal, E., & Spector, T. D. (2018). Role of the gut microbiota in nutrition and health. *BMJ*, *361*, k2179. https://doi.org/10.1136/bmj.k2179

Vandenkoornhuyse, P., Quaiser, A., Duhamel, M., Le Van, A., & Dufresne, A. (2015). The importance of the microbiome of the plant holobiont. *New Phytologist*, *206*, 1196–1206. https://doi.org/10.1111/nph.13312

Vuong, H. E., Yano, J. M., Fung, T. C., & Hsiao, E. Y. (2017). The microbiome and host behavior. *Annual Review of Neuroscience*, *40*, 21–49. https://doi.org/10.1146/annurev-neuro-072116-031347

Wahl, M., Goecke, F., Labes, A., Dobretsov, S., & Weinberger, F. (2012). The second skin: Ecological role of epibiotic biofilms on marine organisms. *Frontiers in Microbiology*, *3*, 292. https://doi.org/10.3389/fmicb.2012.00292

Wallenstein, M. D. (2017). Managing and manipulating the rhizosphere microbiome for plant health: A systems approach. *Rhizosphere*, *3*, 230–232. https://doi.org/10.1016/j.rhisph.2017.04.004

Wasimuddin, Menke, S., Melzheimer, J., Thalwitzer, S., Heinrich, S., Wachter, B., & Sommer, S. (2017). Gut microbiomes of free-ranging and captive Namibian cheetahs: Diversity, putative functions and occurrence of potential pathogens. *Molecular Ecology*, *26*, 5515–5527. https://doi.org/10.1111/mec.14278

Wassermann, B., Cernava, T., Muller, H., Berg, C., & Berg, G. (2019). Seeds of native alpine plants host unique microbial communities embedded in cross-kingdom networks. *Microbiome*, *7*, 108. https://doi.org/10.1186/s40168-019-0723-5

Waters, C. N., Zalasiewicz, J., Summerhayes, C., Barnosky, A. D., Poirier, C., Gałuszka, A. ... Wolfe, A. P. (2016). The Anthropocene is functionally and stratigraphically distinct from the Holocene. *Science*, *351*, aad2622. https://doi.org/10.1126/science.aad2622

Werner, G. D., Cornwell, W. K., Sprent, J. I., Kattge, J., & Kiers, E. T. (2014). A single evolutionary innovation drives the deep evolution of symbiotic N2-fixation in angiosperms. *Nature Communications*, *5*, 4087. https://doi.org/10.1038/ncomms5087

West, A. G., Waite, D. W., Deines, P., Bourne, D. G., Digby, A., McKenzie, V. J., & Taylor, M. W. (2019). The microbiome in threatened species conservation. *Biological Conservation*, *229*, 85–98. https://doi.org/10.1016/j.biocon.2018.11.016

Whittaker, D. J., Gerlach, N. M., Slowinski, S. P., Corcoran, K. P., Winters, A. D., Soini, H. A., ... Theis, K. R. (2016). Social environment has a primary influence on the microbial and odor profiles of a chemically signaling songbird. *Frontiers in Ecology and Evolution*, *4*, 90. https://doi.org/10.3389/fevo.2016.00090

Williams, C. L., Ybarra, A. R., Meredith, A. N., Durrant, B. S., & Tubbs, C. W. (2019). Gut microbiota and phytoestrogen-associated infertility in Southern White Rhinoceros. *Mbio*, *10*(2), https://doi.org/10.1128/mBio.00311-19

Willis, K. (2017). *State of the world's plants 2017*. Report. Royal Botanic Gardens, Kew.

Wyatt, T. D. (2014). *Pheromones and animal behavior: Chemical signals and signatures*. Cambridge, UK: Cambridge University Press.

Yang, G., Wagg, C., Veresoglou, S. D., Hempel, S., & Rillig, M. C. (2018). How soil biota drive ecosystem stability. *Trends in Plant Science*, *23*, 1057–1067. https://doi.org/10.1016/j.tplants.2018.09.007

Yatsunenko, T., Rey, F. E., Manary, M. J., Trehan, I., Dominguez-Bello, M. G., Contreras, M., ... Gordon, J. I. (2012). Human gut microbiome viewed across age and geography. *Nature*, *486*, 222. https://doi.org/10.1038/nature11053

Zahn, G., & Amend, A. S. (2017). Foliar microbiome transplants confer disease resistance in a critically-endangered plant. *PeerJ*, *5*, e4020. https://doi.org/10.7717/peerj.4020

Zilber-Rosenberg, I., & Rosenberg, E. (2008). Role of microorganisms in the evolution of animals and plants: The hologenome theory of evolution. *FEMS Microbiology Reviews*, *32*, 723–735. https://doi.org/10.1111/j.1574-6976.2008.00123.x

SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section.

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