Advancing the case for microbial conservation

CHARLES S. COCKELL and HARRIET L. JONES

Abstract The majority of the biomass and biodiversity of life on the Earth is accounted for by microbes. They play pivotal roles in biogeochemical cycles and harbour novel metabolites that have industrial uses. For these reasons the conservation of microbial ecosystems, communities and even specific taxa should be a high priority. We review the reasons for including microorganisms in conservation agenda. We discuss some of the complications in this endeavour, including the unresolved argument about whether microorganisms have intrinsic value, which influences some of the non-instrumental motivations for their conservation and, from a more pragmatic perspective, exactly what it is that we seek to conserve (microorganisms, their habitats or their gene pools). Despite complications, priorities can be defined for microbial conservation and we provide practical examples of such priorities.

Keywords Biodiversity, biosphere, corals, ex situ conservation, microbial conservation, microorganisms

Introduction

Microorganisms are rarely considered by conservation biologists and yet they form the base of most food chains and accomplish biogeochemical transformations of critical importance to the biosphere (Hawksworth, 1997). For example, each year c. 120 million tonnes of nitrogen gas are removed from the atmosphere by microbial nitrogen fixation and made available to the rest of the biosphere (Freiberg et al., 1997). More than 40% of the carbon dioxide drawn down from the atmosphere is accomplished by microorganisms in the marine environment, giving them an important role in climate regulation (Behrenfeld et al., 2006). Microorganisms take part in a range of processes that are essential for nutrient flux in the biosphere, including rock weathering, organic decomposition and reductive and oxidative transformations of a range of essential elements, changing their mobility and biological availability (Colwell, 1992). Marine microorganisms are responsible for producing and degrading dimethyl sulphide (DMS), a major climate-cooling trace gas. The health of marine microbial

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ecosystems is therefore directly linked to the sulphur cycle and the climate system of the Earth (Bates et al., 1987; Gondwe et al., 2003).

From a biodiversity perspective, microorganisms dominate life on Earth and it is estimated that < 10% of the Earth's microbial diversity has been characterized (Budhiraja et al., 2002). In the deep subsurface, where multicellular organisms cannot persist, microorganisms are the only biological entities (Amy & Halderman, 1997; Whitman et al., 1998). Here, we consider microorganisms to cover fungi, bacteria, archaea and protists (unicellular eukaryotes) and viruses, although the examples we discuss are necessarily limited to just some of these groups.

The impact of human activity and pollution on microbial ecosystems is not well-known because it has not received the attention afforded to animals and plants. Weinbauer & Rassoulzadegan (2007) discussed concerns about microbial extinction and suggested that habitat fragmentation, a pervasive problem for animals and plants, is unlikely to cause microbial extinctions. They highlighted microbial communities that may be at risk from extinction, such as those closely associated with host animals and plants that themselves are at risk. A possible example is the high microbial diversity associated with marine sponges (Hentschel et al., 2003) or microorganisms endemic to particular environments such as endemic *Sulfolobus* species, heat-loving microorganisms that inhabit hot springs (Whitaker et al., 2003). However, as with animals and plants, extinction is not necessarily required to justify conservation efforts. Regional extinctions, or even merely a reduction in microbial community diversity, can be sufficient to have important knock-on effects on other organisms warranting conservation.

Despite overwhelming facts about the role of microorganisms in the biosphere and examples of where they might be threatened, formulating a consistent environmental ethic for the conservation of microorganisms is notoriously difficult (Cockell, 2004, 2005, 2008). On what basis should they be protected?

Why protect microorganisms?

Although it is clear that microorganisms play an important role in ecosystem and biosphere health and that they are diverse, the basis of any conservation policy must be a consistent environmental ethic that seeks to understand why the organism(s) under consideration merit protection in the first place. These arguments were explored by Cockell (2005, 2008). A summary of the essential points that bear on practical policy is valuable, from which we will then suggest steps that could be taken in microbial conservation.

Microorganisms have high instrumental value, where instrumental refers to their uses to the biosphere and to humans. They carry out important biogeochemical transformations in most environments on which multicellular organisms depend. From an instrumental point of view, one reason to conserve and protect microorganisms is that we depend on them. They are not merely useful, our survival depends on them. From a more pragmatic perspective, microbial biological diversity also harbours huge resources of value for the pharmaceutical, biotechnology and food industries (Bull, 2004; Challis, 2008). The loss of microbial diversity implies a loss of potentially valuable resources analogous to existing concerns about loss of potential medicinal compounds associated with plant biodiversity.

The conservation of microorganisms on account of their instrumental value is not a controversial motive. The importance of microorganisms in biogeochemical processes and human industries is understood by most conservationists, who agree that from an instrumental perspective, their protection is merited (Colwell, 1992; Hawksworth, 1997). However, we still know little about which microorganisms carry out important biogeochemical functions. The relatively recent discovery of the anammox process (anaerobic ammonium oxidation; Strous & Jetten, 2004), a part of the nitrogen cycle, is evidence for our continued ignorance of many microbial transformations; this ignorance argues for maximizing microbial conservation on instrumental arguments alone.

A more controversial question, and one that is more difficult to answer, is whether microorganisms have intrinsic value, a phrase used to mean a value independent of uses. In other words, should we protect microbial communities because we think they have some sort of right to exist (where right is used here in a non-legal and loose fashion to denote the possession of some type of value independent of whether they are useful to someone or something)? For many people, this discussion is philosophical but its resolution is practically important because it determines whether a conservation policy for microorganisms should be driven purely by the conservation of microorganisms useful to ecosystems and humans. Apart from conservation policy, a resolution to this question is also important for determining whether it is ethically acceptable to drive a microorganism to extinction intentionally, such as a disease-causing microorganism (Cockell, 2005), a matter raised by Dixon (1976) in relation to the smallpox virus. As microorganisms cannot reason and use language and do not exhibit responses to pleasure and pain that we associate with some animals, they usually fall outside the purview of most theories of intrinsic value.

However, some people, for example, if confronted by an exquisitely layered microbial mat while taking a walk, would walk around it to avoid destroying it. This may be seen as evidence that some microbial ecosystems do elicit a more general respect from people that is not directly linked to their instrumental use and suggests that conservation efforts should be driven by more than just the protection of useful resources.

Clarifying the notion of microbial conservation

A challenge for microbial conservation is determining what should be conserved. It is usually impractical to protect individual species, let alone strains, in the natural environment. Species and strains can be conserved by isolation and culturing ex situ, in which they are maintained as a specific type strain in a culture collection (Supardiyono & Smith, 1997; Arora et al., 2005), but this species-specific conservation is difficult to achieve in the natural environment and may be one reason for the apparent impracticability of including microorganisms in conservation agenda (Cockell, 2008). The generally large population size of most species and their ubiquity further reduces the perceived need to protect them and focuses attention instead on their habitats (Gerhardson & Wright, 2002). The small scale of the organisms contributes to other reasons for their omission from conservation goals, including their general lack of macroscopically visible qualities that can be appreciated by the general public, which provides a powerful impetus for the conservation of many animals and plants. Many microbial mats are obvious exceptions to this generalization. Giant tufa (calcium carbonate) mounds produced by microbial biomineralization are artefacts associated with microbial ecosystems that can be appreciated on a macroscopic scale. However, lack of macroscopically visible features is not a robust reason for the exclusion of microorganisms from conservation.

In the case of microorganisms, their instrumental value is of highest priority. Without the vital functions they perform there would be few microbial ecosystems worth preserving (or even left to preserve) for other reasons. From this perspective, it is the processes carried out by microorganisms rather than specific taxa per se that are of highest priority. If, for example, nitrogen fixation is disrupted in an ecosystem, this will have knock-on effects on other parts of the ecosystem. In this case, it does not matter which organisms are carrying out the transformation, provided it is carried out so that all other organisms (microorganisms, animals and plants) continue to receive adequate nitrogen. This may be expressed as the protection of the ecosystem gene pool, the genetic material that codes for vital functions that exists both within microorganisms and outside them (including mobile genetic material such as plasmids that code for antibiotic and heavy metal resistance and can exist as naked DNA outside organisms).

Another way to view this challenge is the protection of ecosystem diversity. Because microorganisms tend to evolve rapidly and have a propensity to exchange genetic information, species definitions are often difficult to achieve.

Conserving microbial diversity will often, in a practical sense, equate to the conservation of the ecosystem microbial gene pool. From a pragmatic point of view, the conservation of the gene pool and microbial diversity itself equates to the conservation of the physical and chemical conditions within an environment that best support the indigenous microbiota. Gerrath et al. (2000) have suggested that the microbial species diversity of the cliffs of the Niagara Escarpment, Canada, should be a priority in management plans in that region. The cliffs host a diverse microbial biota that is not uniformly distributed, suggesting that protection of large areas of the cliffs is required to maximize the conservation of the microbial diversity.

However, it is clear that conservation may be required in some environments long before clear shifts in microbial community structure are recorded. Recent research suggests that microbial function can be altered without changing the genetic profile of a community. The cattle production hormone, trenbolone, was found to reduce the enzyme activity of the microbial community in a lake in southern Germany but not community structure (Radl et al., 2005). In some cases, therefore, clean-up or conservation efforts may be merited based on observations of reduced microbial functions even when there is no clear evidence of loss of genetic diversity. Thus, conservation should focus on microbial health as well as microbial diversity and the gene pool because, as with animals and plants, compromised health may well lead to the loss of diversity.

Conservation efforts for microorganisms could be achieved by focusing on the protection of habitats, although the protection of habitats is itself motivated by the fact that they contain microbial transformations of importance, so a more direct conservation ethic would draw attention to habitats and their microorganisms. It makes no sense to discuss the conservation of a habitat alone (Pearson, 2007) because few people would be interested in preserving a sterile habitat, the habitat is only of interest because it contains microorganisms. This same argument already applies to the conservation of plants and animals whose habitats, as well as the organisms themselves, are the focus of conservation efforts.

Once ecosystem diversity and health have been secured, it would then be possible to contemplate conservation of microorganisms, from specific species to phyla, where such specific identification can actually be achieved. The motivations for such policies would be the same as those for animals and plants. One such motivation for conservation of specific taxa is the preservation of potentially important products of importance to industry, including novel secondary metabolites with pharmaceutical applications and enzymes with novel physical and chemical tolerances. The iconic example of such a motive is the microorganism *Thermus aquaticus*, a thermophile from which heat-resistant polymerase is obtained for the polymerase chain reaction,

now used in standard molecular biological analysis and applied applications, including forensics. Where such taxa can be identified, ex situ conservation can be used to isolate and conserve strains in the laboratory. Microbial conservation efforts would also focus, however, on preserving these taxa in the wild, in particular because the same environments may harbour similar but as yet undiscovered products.

Within the diverse reasons for conservation there is a dilemma for the microbial conservation agenda that concerns toxic environments. Some polluted environments harbour microorganisms that can adapt to the extreme conditions. Sites polluted by heavy metals such as arsenic, chromium or uranium host organisms with unique genes and biochemical pathways (Stierle et al., 2007). Investigating these organisms is not only beneficial for understanding the effects of pollution, it can also yield insights into natural toxic environments. The natural oxidation of pyrites, for example, creates acidic waters containing sulphuric acid, which bear many similarities to acid mine tailings generated near copper and iron ore mines (Joeckel et al., 2005). Plants and even some animals can survive in some polluted sites, but the rate of evolution and adaptation of microorganisms makes them of special interest. In some cases, a polluted site may be worth conserving because it possesses a novel or scientifically important microbial ecosystem that has become established in extreme conditions.

Conservation priorities and examples

Having established both the reasons for protecting microorganisms and clarified some of the complications in formulating a conservation approach for microorganisms, i.e. functions and/or specific taxa, we suggest here four priority categories for conservation. None of these are mutually exclusive or even exhaustive. For example, microorganisms involved in important ocean biogeochemical cycles (Category 1) also harbour useful genes and bioactive compounds (Category 3). However, these categories broadly define the major priorities in descending order for a microbial conservation effort and they can be used to order priorities in these efforts in the light of the usual limitation of available practical and financial resources.

(1) Microbial communities/ecosystems involved in important global-scale biogeochemical cycles

Microorganisms that play a role in global biogeochemical cycles are a high priority for conservation because of the importance of these cycles to the rest of the biosphere. These communities include ocean phytoplankton and ocean sediment communities, many of which are threatened by anthropogenic activities. One example of this globally important role of microorganisms is in the production and decomposition of DMS (Bates et al., 1987; Gondwe et al., 2003). This gas plays a role in the sulphur cycle and it is

a climate-cooling gas. The diversity and health of global marine microbial ecosystems may therefore influence the DMS cycle and thus the long-term climate cycle. As the influence of global marine pollution on the organisms responsible for this chemical transformation is not fully understood, it shows the need both for a greater research effort to understand these organisms and for conservation efforts directed at better ensuring the health and diversity of organisms that take part in this cycle.

The pivotal role of microorganisms in global geochemical cycles shows the necessity of microbial research in assessing deliberate intervention in these processes. Large-scale projects, such as the iron fertilization of the oceans that has been shown to stimulate carbon sequestration (Blain et al., 2007; Boyd et al., 2007), have been discussed as a means to increase carbon sequestration artificially. These projects could have unintended consequences on microbial ecosystems. The assessment of these projects and their effects on the biosphere requires both a better understanding of the complexity and interconnectedness of microbial ecosystems and their links to the rest of the biosphere and a conservation policy that requires an adequate assessment of these links before such schemes are implemented.

It is worthwhile to note that viruses have an influence on microbial ecosystems, particularly in regulating microbial populations (Suttle, 2005). Their role in some global marine ecosystems has been elucidated. These studies show that viruses must also be included within a microbial conservation effort, despite the often negative association that they have in the medical context.

(2) Microbial communities/ecosystems involved in regional- and local-scale cycles

Pollution can cause declines or changes in microbial populations at both regional and local scales. Areas of concern include large areas of soil, forest ecosystems or marine and freshwater ecosystems. An important example is coral reefs, which harbour great microbial diversity. Bourne et al. (2008) studied microbial changes, focusing on bacterial populations, associated with the bleaching of corals in the Great Barrier Reef, Australia. Changes in the bacterial populations were apparent even before the bleaching event occurred, with a marked shift in the bacterial populations towards *Vibrio* species. Although the bacterial populations returned to similar profiles observed before the bleaching event, the data highlight the fact that bacterial populations are influenced by environmental and climate changes. Dramatic changes in coral health and other marine ecosystems will affect their microbial populations, which could detrimentally influence other communities of organisms.

A major regional influence is the effect of diverse types of pollution on soil microbial populations. These problems highlight the need for ongoing efforts to develop microbial

indicators to study soil health (Machulla, 2003) and, ultimately, to monitor and allow conservation efforts focused on soil microbiology. Pennanen et al. (1996) reported on microbial community shifts caused by the heavy metal contamination of soils. Their study is one of many that have investigated the influence of pollution on soil microbial ecosystems and showed that lowered diversity is caused by heavy metal pollution. Dobler et al. (2000) investigated changes in the metabolic diversity and activity of microorganisms in soil polluted with hydrocarbons and showed that functional changes occurred in the community. This type of work underscores the points made earlier that genetic changes in microbial populations may not be necessary to vindicate conservation efforts. Effects on microbial health, manifested as changes in functional capabilities of the microbial communities, which themselves will affect other organisms in the system, can be sufficient to cause declines in microbial diversity.

(3) Microbial communities/ecosystems that have immediate or potential uses (e.g. medical or industrial uses)

Certain microbial taxa harbour novel metabolites with important industrial uses. We place the conservation of these organisms as a lower priority than conserving those that take part in important biogeochemical cycles and other processes because the survival of industrially significant taxa is itself dependent on the health and functioning of the biogeochemical cycles in which they are embedded. There exists many examples of this microbial conservation motive. *T. aquaticus*, for example, was isolated from Yellowstone National Park. In recognition of these resources the Yellowstone Thermophiles Conservation Project was established to conserve the heat-loving microorganisms' association with the Park's hot springs (Varley & Scott, 1998). Its motivation was primarily instrumental, which was to conserve the organisms for their potential biotechnological uses.

Many microorganisms offer an opportunity to gather scientific information about microorganisms in general, their habitats, and physiological and genetic capabilities (ultimately, this knowledge may have industrial applications). In theory, any microorganism offers the potential for new knowledge but organisms in extreme environments, or that exhibit unusual physiological traits, may merit priority because their loss potentially represents the loss of specific and unusual information that is not taxonomically widespread. Examples of such communities may be the highly stratified cryptoendolithic (hidden within rocks) communities of the Antarctic Dry Valleys, which inhabit sandstones and offer an opportunity to understand microbial community development in extreme polar environments (Friedmann, 1980, 1982). As they contribute to primary production in this region of the Antarctic (Vestal, 1988) they

yield insights into how carbon is sequestered by microorganisms under extreme environmental conditions, with implications for the global carbon cycle, and the responses of microbial communities to extreme climatic conditions, now and in the past.

Another example may be giant sulphur bacteria, such as those found in the sediments of the Namibian Shelf, Africa. As well as offering insights into why microorganisms achieve certain specific sizes (in this case, large sizes up to 600 μm long), these microorganisms can provide insights into the sulphur cycle (Head et al., 2000).

(4) Microorganisms with intrinsic value (communities, ecosystems and individual species)

The fourth category we suggest for conservation is microorganisms that merit protection on account of their intrinsic value. As discussed earlier this is a controversial area of conservation, particularly when applied to microorganisms, because the decision that they have intrinsic value could potentially lead to protection for all microorganisms, which would impose considerable restrictions on many activities. This category may cover any microbial ecosystems and communities that are practical to protect because their preservation places a manageable constraint on people's activities. An example of such communities may include cyanobacteria and algae growing on public buildings, which in some cases may be doing little damage (although this is not always the case) but their destruction through cleaning efforts is implemented merely to satisfy a public perception that buildings should be clean.

Practical steps in microbial conservation

The conservation priorities outlined above require practical steps and changes in policy to achieve their effective implementation. There are a number of new activities and existing efforts that could be strengthened, by both individuals and institutions, and would improve the incorporation of microbial communities and ecosystems into conservation agenda and the culture that would allow a more positive view of the value of microbial conservation. Below, we suggest seven major categories of these efforts.

(1) Microbial ecology research

The conservation priorities discussed here can only be assessed with improved knowledge of microbial ecosystems, their diversity, their functions and their response to human impacts. This information is required to determine how conservation efforts in any region that is deemed to be threatened by human impacts should be focused. Therefore, a continuing effort that should be strengthened is the gathering of information on microorganisms in an ever greater number of environments.

(2) Incorporation of microorganisms into conservation efforts

A logical consequence of the previous discussions is that microbial communities and ecosystems should be incorporated into plans for habitat conservation and protection. Where microorganisms have been suggested for inclusion in conservation strategies it has usually been because of their fortuitous inclusion within the general definition of plants, such as the inclusion of lichens (a microbial symbiosis between a fungus and a cyanobacterium or an alga) within concerns about the effects of rock climbing on cliff biota (McMillan & Larson, 2002; Kuntz & Larson, 2006). It is not obviously the case that by securing habitats for plants and animals microorganisms are automatically protected. Some microbial ecosystems require specific physical and chemical conditions that need attention. For example, the protection of a lake may conserve the plants and animals that live near or in it but subtle changes in water geochemistry can dramatically affect microbiota (Allgaier et al., 2007) without this being obvious in the plant and animal populations. The conservation of microorganisms may require more thorough water quality assessments and the implementation of long-term studies to examine the microbial populations over diurnal, seasonal and annual timescales (Hahn, 2006). In summary, a better balance in considering competing demands on resources that cover the whole phylogenetic tree of life should be attempted.

(3) Protection of microbial communities in their own right

Microbial conservation efforts should lead to the protection and conservation of regions in which there are no plants or animals to protect, such as extreme environments, but where there are microorganisms. Gerrath et al. (2000) suggested the conservation of rock-dwelling communities of the Niagara Escarpment, Canada, based purely on their observation of remarkable microbial diversity. The protection of Antarctica already represents the practical implementation of a microbial conservation policy because in many areas of the continent there are no plants and animals. In this specific case, the historical motivations for the International Antarctic Treaty were not brought about by a concern for microbial conservation, although microbial conservation in Antarctica to protect biodiversity and biotechnological potential has been recognized as an important priority (Vincent, 2000). This category of conservation could be motivated by arguments of intrinsic value. An example of microorganisms that may receive special protection in this category are endoliths that inhabit the interior of rocks in extreme environments, such as those in the Antarctica Dry Valleys or similar communities in other deserts around the world.

(4) Change of legislation to protect microorganisms

Conservation legislation needs to be broadened to include microbial communities and ecosystems while recognizing the potentially serious impediments to progress that could result if microorganisms were afforded sweeping protection in all environments. Some laws show overt discrimination against microorganisms and should be reversed. An example is the Rhode Island State Law for the protection of animals and plants, which states that 'animal and plant means any living or dead organism or organisms other than bacteria or viruses . . .' (Rhode Island General Law, 1956). The motivation for this exclusion is unclear. Perhaps it was regarded as impractical to include microorganisms in such a practical conservation definition but it nevertheless illustrates the legislative barriers to the conservation of microorganisms. Laws should seek to provide special protection for large areas that include functionally important microbial ecosystems (e.g. coral reefs), regions or locations that harbour rare or outstanding examples of microbial ecosystems (e.g. some extreme environments such as polar environments) and regions that contain microbial ecosystems of potential industrial importance.

(5) Improved public outreach and education in schools and colleges

Improving education can lead to significant developments in the protection of microbial communities and ecosystems. At present many of the public view microorganisms as the cause of disease and little more. The view of bacteria as germs is strongly embedded in public information and TV advertisements. Microbiologists are improving the balance of information on the vital roles of microorganisms in biogeochemical cycles and the health of the biosphere. They are doing this from primary school to graduate level. Societies such as the Society for General Microbiology and the American Society for Microbiology are working to improve public understanding of microorganisms. More could be done by the media to promote these efforts and to bridge the divide between microbiology and conservation by improving public knowledge of the beneficial role of microorganisms. Greater efforts to provide microbiology information to the media and educators by societies and microbiology institutes could improve the balance of information entering the public domain.

(6) Ex situ conservation

The storage and culture of microorganisms ex situ, in culture collections, have made huge contributions to existing microbial conservation efforts. However, the collection of these organisms can be haphazard. A more systematic link between field microbial conservation efforts and the preservation of important microorganisms from those environments would increase the value of culture collections and strengthen a system of well-organized microbial conservation in the field. The expansion of culture collections must be accompanied with field approaches discussed earlier because, as with zoos, they have the weakness that they cannot conserve the full diversity of microorganisms found in the natural environment. The development and expansion of collections of archival DNA extracted from natural environments would also be valuable as a means to monitor changes in microbial diversity in environments over time or to assess short-term impacts on microbial community diversity.

(7) Institutional changes

Institutions, including environmental and conservation organizations, centres for diversity and university conservation biology departments should attempt to take a more active role in microbial conservation. Where resources permit, they should hire individuals with responsibility for implementing policy in microbial conservation in the same way as individuals are hired to protect plant and animal diversity.

Conclusions

In summary, microorganisms have been largely ignored by conservation efforts. Yet their role in biogeochemical processes, their diversity and abundance, and their potential as repositories of valuable genetic information and metabolic products make them as important as animals and plants to the biosphere and human welfare. However, the arguments for their conservation, as we have shown here, require careful examination and are not merely an extrapolation of the arguments for animal and plant conservation. We have suggested practical means by which microbial conservation efforts could be advanced.

Acknowledgements

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