Human dissemination of genes and microorganisms

Yong-Guan Zhu[1], Michael Gillings[2], Pascal Simonet[3], Dov Stekel[4], Steve Banwart[5] and Josep Penuelas[6,7]

[1] Key Lab of Urban Environment and Health, Institute of Urban Environment, Chinese Academy of Sciences, Xiamen 361021, China; [2] Department of Biological Sciences, Macquarie University, Sydney, NSW 2109, Australia; [3] Environmental Microbial Genomics Group, Université de Lyon, 69134, France; [4]School of Biosciences, University of Nottingham, Nottingham NG7 2RD, United Kingdom; [5] Department of Geography, The University of Sheffield, Sheffield S10 2TN, United Kingdom; [6] CSIC, Global Ecology Unit, CREAF- CSIC-UAB, Bellaterra, 08193 Barcelona, Catalonia, Spain; [7] CREAF, Cerdanyola del Vallès, 08193 Barcelona, Catalonia, Spain

Corresponding Author:
Y.G. Zhu
Address: Key Lab of Urban Environment and Health, Institute of Urban Environment, Chinese Academy of Sciences, Xiamen 361021, China
Email: ygzhu@iue.ac.cn
M Gillings
Department of Biological Sciences, Macquarie University, Sydney, NSW 2109, Australia
Email: michael.gillings@mq.edu.au

Keywords: Anthropocene; human impacts; resistome; planetary health; pollution; xenogenetic

Abstract

Earth's Critical Zone sustains terrestrial life, and consists of the thin planetary surface layer between unaltered rock and the atmospheric boundary. Within this zone, flows of energy and materials are mediated by physical processes and by the actions of diverse organisms. Human activities significantly influence these physical and biological processes, affecting the atmosphere, shallow lithosphere, hydrosphere and biosphere. The role of organisms includes an additional class of biogeochemical cycling, this being the flow and transformation of genetic information. This is particularly the case for the microorganisms that govern carbon and nitrogen cycling. These biological processes are mediated by expression of functional genes and their translation into enzymes that catalyze geochemical reactions. Understanding human effects on microbial gene activity and microbial distribution is an important component of Critical Zone science, but is highly challenging to investigate across the enormous physical scales of impact ranging from individual organisms to the planet. One arena where this might be tractable is by studying the dynamics and dissemination of genes for antibiotic resistance and the organisms that carry such genes. Here we explore the transport and transformation of microbial genes and cells through Earth's Critical Zone. We do so by examining the origins and rise of antibiotic resistance genes, their subsequent dissemination, and the ongoing colonization of diverse ecosystems by resistant organisms.

Introduction

Earth's Critical Zone is the thin surface layer of the planet upon which terrestrial life depends. It extends from unaltered bedrock, through the land surface, to the vegetation canopy and atmospheric boundary layer. Critical Zone science is complementary to other integrative systems approaches for studying terrestrial, marine and freshwater environments. Crucially, it includes a mechanistic understanding of shallow lithosphere processes and their interactions with the above-ground ecosystems (Mobley 2009)
. It addresses these interactions across wide temporal (sub-second reaction kinetics to geological time spans) and spatial scales (molecular to planetary). The Critical Zone approach recognizes Earth as a physical and geochemical substrate that supports above ground ecological functions, and extends the lower boundary of ecological function to embrace the lithosphere, and its inputs over geological time scales.

This interdisciplinary research area within geobiology links biological and geochemical processes across temporal and spatial scales. However, the distribution, transport and recruitment of functional genes has rarely been investigated via the systems perspective framed by Critical Zone science. Since investigation of Critical Zone biogeochemical processes extends the analysis of flows and transformations of material and energy to explicitly include biodiversity, a tractable approach may be to describe the geospatial dynamics of the genetic information encoded in functional genes, and the microbes that carry these genes. Above–ground human activities generate impacts that are transmitted through the vertical extent of the Critical Zone, via aquifers, and horizontally within water catchments. Analyzing the vertical and horizontal penetration of genetic material should be part of these investigations (Küsel, et al. 2016).

Environmental microbes and genes are traditionally studied in one location, or in one environmental compartment (such as vegetation, the water column, or soil), with little attention paid to the dynamic exchange of microbes and genes across system

boundaries and physical scales. The advent of "omics" tools has facilitated the exploration of Earth's biological 'dark matter', but there remains a substantial conceptual gap between the notion of the Earth's biome and its quantitative manifestation in biogeochemical fluxes. Integrating "omics" data into earth system science should generate better models of biogeochemistry and improve understanding of how environmental changes will impact microorganisms. For instance, incorporating environmental genomics data into biogeochemical models improves predictions about nitrogen cycling (Mock, et al. 2016; Reed, et al. 2014).

Driven by these concepts, there is increasing attention towards system views of the temporal and spatial distribution of microbes and genes in Earth's Critical Zone. Metagenomics has been used to determine the influence of fluvial networks on the co-occurrence of microbes, by examining biofilms in over a hundred streams (Widder, et al. 2014). The distribution and origins of fecal bacteria have been determined in large mixed-use watersheds in Michigan, USA, also using omics technologies (Verhougstraete, et al. 2015). Similar ecosystem wide approaches have been used to demonstrate how below ground microbial diversity might be a primary driver of plant diversity and productivity (Bardgett and van der Putten 2014). Questions are also being asked about how surface activities might influence below ground biota and nutrient cycling, using combinations of omics, biogeochemical, and hydrogeological approaches (Küsel, et al. 2016).

These publications are representative of recent efforts to explore the links between microbial biogeography, biogeochemistry and geological processes. In particular, they reflect a growing interest on the effects that human activities might have on the microbial world (Gillings and Paulsen 2014). Understanding the role that humans might have in changing the distributions of microorganisms, and in generating selective forces that alter adaptive pressures, are essential if we are to predict how global change will affect microbial activity and function. However, many of the most important processes for Critical Zone function are complex, multi-gene and multi-cell

interactions that are difficult to model, due to the complexity and dynamics of genetic and functional diversity within indigenous microbial communities.

There are alternative, simpler systems that we can use to understand the influences that humans have on the transport and transformation of genetic information in the Critical Zone. Antibiotic resistance, for instance, is generally a one-gene, one phenotype character, and has been the subject of considerable research over the last fifty years. Genes conferring resistance, and the cells that host these genes, could be used as a paradigm for assessing the interactions of gene flow with the diversity of microorganisms in the Critical Zone.

Antibiotic resistance might be a good proxy that can inform more general conclusions about alterations in the distribution and activity of the microorganisms that host specific genes within the Critical Zone. The widespread use of antibiotics in agriculture and medicine has increased the abundance of both resistance genes and the bacteria that host them. These genes and microorganisms are then shed into environmental compartments via human and animal waste streams such as manure, sewage sludge, and wastewater (Gillings 2013). As a consequence, antibiotic resistance genes are considered to be emerging environmental contaminants (Pruden, et al. 2013). On the one hand, the spread of resistance determinants within the Critical Zone is caused by human activities, and on the other hand, it also threatens human health worldwide. The history of resistance begins in the 1950s, and is thus co-incident with the 'Great Acceleration' and the rapidly increasing impact of humans activity on the planet since this time point (Steffen, et al. 2015).

Natural transport and biogeography of bacteria

We live in a world where organismal abundance and gene frequencies have been significantly shaped by human activities. Nevertheless, it is worth reflecting on the historical dynamics of microbial organisms and ecosystems, before the rise of human influence. This allows comparisons with the modern world.

It has been known for some time that microorganisms exhibit the same taxa-area relationships and turnover in species assemblages with distance that are characteristic of larger organisms (Green, et al. 2004; Horner-Devine, et al. 2004). Taxa are distributed non-randomly in environments such as soil, fresh water and groundwater, at scales from meters to many thousands of kilometers (Martiny, et al. 2006). These patterns are driven by a combination of factors, including: the ability to disperse over distance; selection at the destination; and stochastic processes such as drift and mutation (Hanson, et al. 2012). Teasing apart the relative contributions of the processes that generate patterns of microbial biogeography is difficult, and is further complicated by the diversity and complexity of microbial communities themselves (Evans, et al. 2017; Haggerty and Dinsdale 2016). The impact of human migration as a transport vector on structuring prokaryotic communities is still poorly understood. Some authors have argued that stochastic events could be more important than deterministic factors such as competition and niche differentiation (Sloan, et al. 2006).

At the largest possible temporal and spatial scales, bacteria are the best candidates to survive interplanetary transfer inside rock. Such lithopanspermia is a potential means that life could be transferred between planetary bodies within and outside our solar system (Nicholson 2009). On Earth, but still across large spatial scales, microorganisms are capable of long-distance dispersal, being ubiquitous and abundant, even in the upper atmosphere (Barberán, et al. 2015). Thousands of distinct bacterial taxa, accompanied by other microorganisms, are carried within dust plumes in long-range intercontinental transport events. For instance, Asian aerosols contribute to microbial species richness in North American air (Smith, et al. 2013), and dust storms generated in the African Sahara-Sahel transport microorganisms that eventually contribute to bacterial assemblages in European mountain lakes (Perfumo and Marchant 2010; Peter, et al. 2014).

Natural release and survival of DNA

Microbial biogeography is further complicated by the ability of microorganisms to acquire foreign DNA, and consequently movement of genes through the Critical Zone can occur independently of organismal movement. DNA released from organisms can transfer to unrelated species either through close contact, or at a distance, when DNA can survive in the environment for extended time periods (Gillings 2017b).

Extracellular DNA can be readily detected in environmental samples, and can originate from dead bacterial, animal or plant cells. All soils contain significant quantities of extracellular DNA (Frostegård, et al. 1999). This DNA can persist in the environment and can be transported away from cell debris. Because DNA can resist physical and biological degradation under some conditions, it has even been proposed as a potential signature of life during interplanetary exploration (Lyon, et al. 2010).

Under natural conditions, DNA released via cell lysis is in contact with other cellular components (wall debris, proteins, lipids, RNA, etc.). The presence of both organic compounds and inorganic molecules in soil particles strongly influences the adsorption of DNA (Pietramellara, et al. 2009). Consequently, DNA can be protected from enzymatic degradation in soil by adsorption onto soil minerals and humic substances (Levy-Booth, et al. 2007). Protection against degradation by DNases of microbial origin is aided by the concomitant adsorption of nucleases (Demanèche, et al. 2001). Many studies on survival of DNA in the environment have been conducted using plasmids and antibiotic resistance genes as markers.

The DNA persisting in soil is only a tiny fraction of the total DNA being released at any one time from decaying plants, animals and microorganisms. This DNA usually undergoes rapid degradation (Ceccherini, et al. 2007; Pontiroli, et al. 2007; Poté, et al. 2010). Degradation is biological and enzymatic, since DNA can survive in autoclaved treatments (Zhu 2006). Nevertheless, a proportion of extracellular DNA does persist in natural environments, either bound to soil particles, or inside biofilms, where it is an important structural component (Pietramellara, et al. 2009; Whitchurch, et al.

2002). In the long term, persistence eventually requires being taken up by a recipient cell, and incorporated into that cell's genome. The likelihood of this occurring improves with increasing phylogenetic and ecological similarity of donor and recipient (Beiko, et al. 2005), and also improves markedly if the donor DNA can confer an adaptive phenotype. This is one reason why genes that confer antibiotic resistance are a good marker for these processes in natural environments.

Movement and transport of extracellular DNA.

DNA is able to be transported vertically in unsaturated soils, to eventually penetrate groundwater and aquifers, where it can be immobilized through adsorption onto mineral surface or be transported with groundwater flow (Poté, et al. 2009). Forced pumping of groundwater for drinking can thus induce rapid flow and associated transport of DNA over considerable distances. DNA can also move upwards in the soil column via capillary action (Ceccherini, et al. 2007), potentially allowing subsequent long distance movement via erosion and run-off.

The presence of extracellular DNA in environmental samples is increasingly being used to perform multi-taxa surveys, or to detect rare and elusive species (Zinger, et al. 2016). However, the parameters that affect transport and survival of extracellular DNA are not well understood, and may compromise some of these experiments (Jerde, et al. 2016). Given the problems of differential survival and transport of extracellular DNA, guidelines for the design and interpretation of environmental DNA methods are required (Goldberg, et al. 2016).

Experiments to address this problem have used a variety of indicator DNAs. Antibiotic resistance genes known to be associated with humans are a good choice. They have been used to show survival and dissemination of DNA into freshwater sediments in an aquatic environment used for drinking water supply (Thevenon, et al. 2012). Similarly, plasmids (Poté, et al. 2003) and bacteriophages (Chetochine, et al. 2006) have been used to demonstrate transport over considerable distances in water

saturated soil and groundwater. However, the dynamic relationships between DNA transport, immobilization, survival, and the limits of detection are not well established (Hunter, et al. 2016).

One way to track and understand dissemination of DNA through the environment, and indeed, throughout Earth's Critical Zone is to use a model system that is tractable and reflects the history of human impacts. Antibiotic resistance genes, their plasmid vectors, and the bacteria that host them are a good candidate for use as a proxy for anthropogenic influences (Gillings, et al. 2015).

The evolutionary history of antibiotic resistance

The genes that we regard as antibiotic resistance genes are, by and large, recently descended from genes whose original functions were *not* to confer resistance to clinical concentrations of antibiotic compounds. Two kinds of event are responsible for the genesis of modern antibiotic resistance genes: mutation of a pre-existing gene within a cell lineage; and co-option of a gene acquired by lateral gene transfer from an unrelated lineage (Gillings, et al. 2017). In the latter case, it has been suggested that many of these laterally transferred genes originally functioned in defensive responses to small signaling molecules arising from antagonistic biota, including those molecules we now use as antimicrobial agents (Davies and Davies 2010; Davies, et al. 2006; Linares, et al. 2006).

This idea is supported by the observation that natural environments and environmental bacteria contain large numbers of genes that *could* confer resistance to antibiotics if they were present in clinical contexts. These genes are collectively termed the resistome. The resistome is far larger and far older than the small subset of problematic resistome elements that have recently made their way into human and animal bacteria of clinical importance (Allen, et al. 2010). For example, gene families that can confer resistance to particular antibiotic classes are plausibly related to defense mechanisms selected in response to naturally-occurring compounds which

induce chemical stress. These gene families date back hundreds of millions of years, and can be recovered from ancient environments such as caves and permafrost (Baltz 2008; Bhullar, et al. 2012; D'Costa, et al. 2011).

The widespread use of antibiotics in health care and intensive animal farming since the 1950s has exerted strong selection for rare, individual cells that had recently acquired a mutation or resistome element. As a result of continuing antibiotic use resistant organisms have rapidly increased in both abundance and distribution (Gillings 2017b). Under this selection pressure, resistant organisms and their genetic cargo have spread between individuals, species and continents (Bengtsson-Palme, et al. 2015; Hu, et al. 2016). These resistance genes are readily identifiable because their recent expansion means they have highly conserved DNA sequences. Carriage of such resistance genes is now a universal feature of gut bacteria in humans and agricultural animals (Pal, et al. 2016).

As a consequence of their universal carriage, resistant bacteria are continually discharged into the environment via waste water, sewage treatment plants and animal manure, thus spreading both resistant organisms and resistance genes. These same waste streams also release antibiotics (Grenni, et al. 2017; Liu, et al. 2017), which have significant effects, and trigger chemical stress responses even at sub-inhibitory concentrations (Chow, et al. 2015). Waste waters then become giant reactors where complex interactions occur between chemical compounds, molecular responses, cells, resistance genes, and genetic transformation driven by lateral transfer and mutation (Gillings and Stokes 2012).

The broad-scale dissemination of bacterial genes, including resistance genes, is mediated by a number of factors. This transport and transformation is controlled at various nested levels. Firstly, DNA can be released from cells and persist in the environment. From here it can be taken up and incorporated into environmental bacteria. Secondly, genes can be transported within their host bacteria. Where such

bacteria are dispersed by water or wind, their cargo genes are carried with them. Finally, the bacteria themselves can be carried inside animal hosts via mass migration, or in the case of humans, by travel and tourism.

Tracking the movement of resistance genes in Earth's Critical Zone

Interest in the dispersal of antibiotic resistance genes and their host bacteria is growing rapidly as the environmental consequences of this dissemination become more apparent. Partly, this is because resistance genes themselves have unique properties. On the one hand, they behave like pollutants which exhibit environmental exposure routes, and on the other hand, they can replicate, making them more akin to an invasive species with multiple cellular hosts (Gillings 2017a).

Human activities directly promote the invasion and spread of resistance determinants. Waste water treatment plants occupy a position between human waste streams and the aquatic environment, but do not effectively remove resistance genes, thus distributing them in effluent (Aubertheau, et al. 2016; Ben, et al. 2017; Karkman, et al. 2016). Effluents also contain significant concentrations of selective agents, thus promoting the survival of resistant organisms, potentially at the expense of endemic species (Borruso, et al. 2016; Caucci, et al. 2016; Koczura, et al. 2016; Lehmann, et al. 2016). Application of sewage sludge, or antibiotics alone, increases the abundance of resistance genes, and changes the microbial community in soils (Chen, et al. 2016; Cleary, et al. 2016).

Agricultural activities also strongly promote the environmental spread of resistance through disposal of wastes and application of manure (Heuer, et al. 2011; Sandberg and LaPara 2016). Similarly, aquaculture is increasingly being recognized as a focal point for enhancing and dispersing resistance in the environment (Muziasari, et al. 2016). In both of these cases, the simultaneous release of antibiotics and other selective agents promotes selection of organisms containing resistance genes (He, et al. 2016; Liu, et al. 2017; Wang, et al. 2016). This generates opportunities for co-

selection and fixation of chemical (toxic metals) and resistance determinants in species, and within individual DNA molecules (Johnson, et al. 2016; Zhou, et al. 2016).

A combination of phenomena, including the volume of human and agricultural waste streams, and the concomitant release of selective agents, means that resistance genes and resistant organisms can become extraordinarily widespread and abundant over very short time frames. A single multidrug resistant clone of *E. coli* has become globally disseminated since its origin as recently as the year 2000 (Petty, et al. 2014).

Antimicrobial resistance in Earth's Critical Zone is thus dependent on human activities, the action of selection in natural environments, and upon natural transport mechanisms, such as rivers, groundwater and soil movement. At landscape scale, antibiotic resistance genes can move with soil erosion and drainage from top soil to groundwater.

Modeling of the dynamics of resistance genes in the Critical Zone

Effective modelling of the spread of antimicrobial resistance is essential for making predictions that can inform policy, practice and environmental surveillance. Policy makers are interested in models for two reasons. First, they support general policies that can inform handling of antimicrobials in the environment, during production, agricultural use or waste water treatment. Second, they inform possible interventions in the face of a specific outbreak of an antibiotic resistant human or animal pathogen. Models need to be flexible, realistic, and able to be used in different contexts.

However, developing realistic and flexible models that operate on an environmental scale is a significant challenge. AMR encompasses a broad range of organisms, genes and antimicrobial agents, and mobile genetic elements. Sensitive and resistant organisms live in complex, heterogeneous communities. The processes that drive fixation of resistance occur at microscopic scales. Selection and spread within the

Critical Zone can involve slurry tanks (Baker, et al. 2016), the animal gut (Volkova, et al. 2012), wastewater treatment plants (Sharifi, et al. 2014) and industrial effluents, while broader dissemination might be driven by soil movement, water percolation, rivers, domestic animals and wildlife.

Mathematical modelling of resistance spread has been applied at a range of scales. Models for laboratory-scale experiments have been valuable for establishing rates of mutation, selection and the spread of resistance (Bootsma, et al. 2012; De Gelder, et al. 2004). However, while these models are useful for characterizing key processes, they do not scale up to the required complexity for whole environments. Consideration of the spatial structure of microbial communities, for example biofilms, gives a more accurate representation of the spread resistance in a community (Lardon, et al. 2011). Models of farms or sewage treatment plants have shown that it is possible for resistant organisms or pathogens to persist even in the absence of antibiotic treatment (Sharifi, et al. 2014), and can also make predictions about the duration of persistence (Volkova, et al. 2013). However, these models have been limited to considering a single type of bacterium or antimicrobial agent.

Therefore, three developments are needed to move forward with environmental scale models that can be effective in understanding and predicting spread or reduction in resistance in the Critical Zone: inclusion of heterogeneity; multi-scaling in space and time; and effective global data sharing.

First, models will need to consider a fuller range of organisms, resistance genes, mobile genetic elements and antimicrobials, that reflect the complexity of the observed system (Chen, et al. 2016; Perron, et al. 2015) and the importance of co-selection of antibiotic and metal resistance genes (Gullberg, et al. 2014; Pal, et al. 2015). Importantly, different organisms, genes and mobile genetic elements will behave differently, leading to heterogeneity in growth, transmission and selection. However, their inclusion will be essential to determine the pace and range of spread or

elimination of resistance, and the relative contributions of resistance genes to the emergence of potentially resistant pathogens. This is a considerable modeling challenge, because the number of possible genetic and resistance combinations increases exponentially with the degree of biological complexity to be included. For example, even within a mass action ordinary differential equation framework, to model populations of a single bacterial species in an environment with two different antimicrobials, two respective resistance genes, that each might be carried on one of two different mobile genetic elements, requires many differential equations, and such models are difficult to parameterize or analyze.

Second, models will need to operate on multiple scales. While the best representation of spread of AMR on a microscopic scale is through individual-based models, such models do not extend to an environmental scale. Therefore, it will be necessary to coarse-grain predictive outcomes of small-scale models into larger scale, multi-compartment models that can consider populations of humans, farm animals and wildlife in their respective geographical compartments. It may also be necessary to use models that combine deterministic with stochastic elements. Deterministic models are capable of simulating large populations of bacteria, while stochastic models can capture rare and random events, for example the spread of a particular resistance determinant from one species to another. A further feature of such models will be the need to embed geospatial data (Pruden, et al. 2012), to include factors such as topography, land use and water flows.

Third, such models will require considerable calibration against real data. Researchers carrying out environmental and field studies will need to share data in a way that is useful for embedding into predictive models. To do this, we will require agreed standards for data capture and sharing, and the development of an international database for resistance in the critical zone. Such data could include observations from a wide range of experimental techniques, and data on taxa, species, phenotypes, genomes, resistance genes, mobile genetic elements, antibiotics, heavy metals and

412 other antimicrobials. Ideally, the data would also include geospatial coordinates so
413 that they can be used in geospatially explicit models. While this challenge alone is
414 considerable, there is considerable precedent for agreed data standards in other areas
415 of high throughput biology, which this development can draw upon.
416
417 **Dispersal of resistance genes in the Critical Zone – A planetary view**
418 Understanding movement of antibiotic resistance through the Critical Zone is
419 complex, and difficult to model (Figure XX). Quantifying ARG movement requires
420 the coupling between the transport of bacterial cells (and resistance genes they carry)
421 and materials (and associated selective agents) and their interactions within the
422 Critical Zone. We can then infer more general principles about the movement and
423 transformation of genes and microorganisms. These principles might then be tested
424 and applied to even more complex, multi-gene phenotypes of central importance to
425 global biogeochemistry.
426
427 Before humans had a major influence on the planet, movement of microorganisms
428 and the genes they carry was mainly driven by physical phenomena, such as air
429 currents and water flow. Without human influence, a relatively small number of
430 microbial cells would be transported to any specific location, therefore chance played
431 a large role in dispersal of bacterial cells/genes. This dispersal did not necessarily
432 result in survival or recruitment, since locally adapted cells were already present, and
433 filled existing niches. With the advent of the Anthropocene, human activities now
434 have large effects on the dispersal of microorganisms and the genes they carry (Table
435 1). Movement of humans around the globe transports our internal microbiota to new
436 locations at an unprecedented scale. Human migration changes the abundance of
437 resistance genes, and successfully transports resistance genes between continents
438 (Bengtsson-Palme, et al. 2015; Sun, et al. 2016).
439
440 The fact that biomass of humans and domestic animals now comprise 35 times that of
441 wild terrestrial mammals (Smil 2011) may have consequences for the microbial

world. Firstly, humans, domestic and agricultural animals all carry resistance genes in their gut microbiota, thus vastly increasing the abundance and distribution of these genes on the planet. Secondly, on a global scale the fecal microbiota are now mainly represented by the gut microbiota of six species: humans, cattle, sheep, goats, pigs and chickens. Thus, the overall diversity of bacteria being shed in feces has consequently declined. At the same time, the quantity of fecal microbiota has increased as the biomass of humans and their domesticates approaches five times the global carrying capacity for terrestrial vertebrates (Smil 2011). Therefore , disposal of both human and animal manures has a significant impact on the dissemination of both microbial organisms and genes (Chen, et al. 2016; Jechalke, et al. 2013). These cells and genes can contaminate agricultural produce (Bengtsson-Palme 2017; Jones-Dias, et al. 2016), which is then transported between countries.

Humans disperse microorganisms by mass movement of materials (Table 1). Transport of ballast water in ships is estimated to move 10^{19} bacteria each day (Endresen, et al. 2004; Ruiz, et al. 2000), spreading diverse microorganisms around the globe and thus reshaping microbial biogeography (Brinkmeyer 2016; Lohan, et al. 2016). It has been suggested that anthropogenic movement of soil, sand and rock now surpasses all natural processes combined (Wilkinson and McElroy 2007), incidentally transporting huge numbers of microbial cells. Wastewater also transports microorganisms and their cargo genes into the environment. With increasing human populations, the volume of wastewater is increasing, but global data on the treatment, reuse, or volumes of waste water is difficult to assemble (Sato, et al. 2013). As an example, antibiotic resistance genes now pollute over 4,000 kilometers of the Chinese coastline at levels up to 100 million genes per gram of sediment (Zhu, et al. 2017b). None of these genes would have been present in this sediment 50 years ago.

Human activities increase the numbers of microorganisms being transported within the Critical Zone and around the Earth ecosystem, thus increasing the chances for successful recruitment (Table 1). Furthermore, during transport, microorganisms are

often exposed to pollutants, particularly during discharge of manure and waste water. Exposure to antibiotics and other co-selective agents, even at low does, can enhance the rate at which bacteria generate diversity via mutation (Kohanski, et al. 2010), recombination (Guerin, et al. 2009) and lateral gene transfer (Prudhomme, et al. 2006). The simultaneous dispersal of microorganisms and various selective agents increases the genetic variation being generated in those microbial populations, enhancing their potential to evolve (Gillings and Stokes 2012). Consequently a subset of the cells dispersed to new locations are adapted to the co-dispersed pollutants, increasing their probability of recruitment at these new locations. Further, because genes for metal, disinfectant and antibiotic resistance are often closely linked (Johnson, et al. 2016), exposure to any one selective agent drives their co-selection, and maintains mosaic clusters of resistance determinants (Di Cesare, et al. 2016; Gaze, et al. 2005; Skurnik, et al. 2010). Possession of diverse resistance determinants significantly increases the probability of recruitment at novel destinations by providing a selective advantage over endemic microorganisms (Table 1).

Concluding remarks

It is becoming more and more important to understand how human activities cause systematic changes in ecosystems (Alberti, et al. 2017), and especially the effects on the emergence and spread of ARGs in urbanizing Earth's Critical Zone (Zhu, et al. 2017a). To better understand the dynamics of ARGs in the Critical Zone, future studies should emphasize linkages between biogeochemical cycling of nutrients and contaminants with the movement of microorganisms. Under the framework of Critical Zone science, tracking the dynamics of ARGs should give us insights into the interconnections between multiple environmental compartments within the entire Critical Zone. Due to the extreme heterogeneity of the Critical Zone, we should also focus on hot spots for ARG dissemination such as locations receiving high loads of wastewater or manure. Understanding the complex feedbacks between the dynamics of ARGs and interactions with physical, chemical and biological processes in the Critical Zone is a grand challenge. Progress can only be made by forging

interdisciplinary research teams that can manage and interpret the enormous datasets of genomics and biogeochemistry, and by developing predictive models based on these datasets.

Acknowledgements

YGZ is supported by Natural Science Foundation of China (41571130063), Chinese Academy of Sciences (XDB15020302 and XDB15020402), MRG is supported by the Australian Research Council, JP is supported by the European Research Council Synergy grant SyG-2013-610028 IMBALANCE-P. The authors declare no conflicts of interest.

References

Alberti M, Marzluff J, Hunt VM. 2017. Urban driven phenotypic changes: empirical observations and theoretical implications for eco-evolutionary feedback. Phil. Trans. R. Soc. B 372(1712):20160029.

Allen HK, Donato J, Wang HH, Cloud-Hansen KA, Davies J, Handelsman J. 2010. Call of the wild: antibiotic resistance genes in natural environments. Nature Reviews Microbiology 8(4):251-259.

Aubertheau E, Stalder T, Mondamert L, Ploy M-C, Dagot C, Labanowski J. 2016. Impact of wastewater treatment plant discharge on the contamination of river biofilms by pharmaceuticals and antibiotic resistance. Science of The Total Environment 579:1387-1398.

Baker M, Hobman JL, Dodd CE, Ramsden SJ, Stekel DJ. 2016. Mathematical modelling of antimicrobial resistance in agricultural waste highlights importance of gene transfer rate. FEMS microbiology ecology 92(4):fiw040.

Baltz RH. 2008. Renaissance in antibacterial discovery from actinomycetes. Current Opinion in Pharmacology 8(5):557-563.

Barberán A, Ladau J, Leff JW, Pollard KS, Menninger HL, Dunn RR, Fierer N. 2015. Continental-scale distributions of dust-associated bacteria and fungi. Proceedings of the National Academy of Sciences 112(18):5756-5761.

Bardgett RD, van der Putten WH. 2014. Belowground biodiversity and ecosystem functioning. Nature 515(7528):505-511.

Beiko RG, Harlow TJ, Ragan MA. 2005. Highways of gene sharing in prokaryotes. PNAS 102(40):14332-14337.

Ben W, Wang J, Cao R, Yang M, Zhang Y, Qiang Z. 2017. Distribution of antibiotic resistance in the effluents of ten municipal wastewater treatment plants in China and the effect of treatment processes. Chemosphere 172:392-398.

Bengtsson-Palme J. 2017. Antibiotic resistance in the food supply chain: Where can sequencing and metagenomics aid risk assessment? Current Opinion in Food Science 14:66-71.

Bengtsson-Palme J, Angelin M, Huss M, Kjellqvist S, Kristiansson E, Palmgren H, Larsson DJ, Johansson A. 2015. The human gut microbiome as a transporter of antibiotic resistance genes between continents. Antimicrobial Agents and Chemotherapy 59(10):6551-6560.

Bhullar K, Waglechner N, Pawlowski A, Koteva K, Banks ED, Johnston MD, Barton HA, Wright GD. 2012. Antibiotic resistance is prevalent in an isolated cave microbiome. PLoS ONE 7(4):e34953.

Bootsma M, van der Horst M, Guryeva T, Ter Kuile B, Diekmann O. 2012. Modeling non-inherited antibiotic resistance. Bulletin of mathematical biology 74(8):1691-1705.

Borruso L, Harms K, Johnsen PJ, Nielsen KM, Brusetti L. 2016. Distribution of class 1 integrons in a highly impacted catchment. Science of The Total Environment 566:1588-1594.

Brinkmeyer R. 2016. Diversity of bacteria in ships ballast water as revealed by next

560 generation DNA sequencing. Marine pollution bulletin 107(1):277-285.
561 Caucci S, Karkman A, Cacace D, Rybicki M, Timpel P, Voolaid V, Gurke R, Virta M,
562 Berendonk TU. 2016. Seasonality of antibiotic prescriptions for outpatients
563 and resistance genes in sewers and wastewater treatment plant outflow. FEMS
564 Microbiology Ecology 92(5):fiw060.
565 Ceccherini MT, Ascher J, Pietramellara G, Vogel TM, Nannipieri P. 2007. Vertical
566 advection of extracellular DNA by water capillarity in soil columns. Soil
567 Biology and Biochemistry 39(1):158-163.
568 Chen Q, An X, Li H, Su J, Ma Y, Zhu Y-G. 2016. Long-term field application of
569 sewage sludge increases the abundance of antibiotic resistance genes in soil.
570 Environment international 92:1-10.
571 Chetochine AS, Brusseau ML, Gerba CP, Pepper IL. 2006. Leaching of phage from
572 class B biosolids and potential transport through soil. Applied and
573 environmental microbiology 72(1):665-671.
574 Chow L, Waldron L, Gillings M. 2015. Potential impacts of aquatic pollutants: sub-
575 clinical antibiotic concentrations induce genome changes and promote
576 antibiotic resistance. Frontiers in microbiology 6:803.
577 Cleary DW, Bishop AH, Zhang L, Topp E, Wellington EM, Gaze WH. 2016. Long-
578 term antibiotic exposure in soil is associated with changes in microbial
579 community structure and prevalence of class 1 integrons. FEMS Microbiology
580 Ecology 92(10):fiw159.
581 D'Costa VM, King CE, Kalan L, Morar M, Sung WWL, Schwarz C, Froese D, Zazula
582 G, Calmels F, Debruyne R and others. 2011. Antibiotic resistance is ancient.
583 Nature 477:457-461.
584 Davies J, Davies D. 2010. Origins and evolution of antibiotic resistance.
585 Microbiology and Molecular Biology Reviews 74(3):417-433.
586 Davies J, Spiegelman GB, Yim G. 2006. The world of subinhibitory antibiotic
587 concentrations. Curr. Opinion Microbiol. 9(5):445-453.
588 De Gelder L, Ponciano JM, Abdo Z, Joyce P, Forney LJ, Top EM. 2004. Combining
589 mathematical models and statistical methods to understand and predict the
590 dynamics of antibiotic-sensitive mutants in a population of resistant bacteria
591 during experimental evolution. Genetics 168(3):1131-1144.
592 Demanèche S, Jocteur-Monrozier L, Quiquampoix H, Simonet P. 2001. Evaluation of
593 biological and physical protection against nuclease degradation of clay-bound
594 plasmid DNA. Applied and Environmental Microbiology 67(1):293-299.
595 Di Cesare A, Eckert E, Corno G. 2016. Co-selection of antibiotic and heavy metal
596 resistance in freshwater bacteria. Journal of Limnology 75(s2).
597 Endresen Ø, Behrens HL, Brynestad S, Andersen AB, Skjong R. 2004. Challenges in
598 global ballast water management. Marine Pollution Bulletin 48(7):615-623.
599 Evans S, Martiny JB, Allison SD. 2017. Effects of dispersal and selection on
600 stochastic assembly in microbial communities. The ISME Journal 11:176-185.
601 Frostegård Å, Courtois S, Ramisse V, Clerc S, Bernillon D, Le Gall F, Jeannin P,
602 Nesme X, Simonet P. 1999. Quantification of bias related to the extraction of
603 DNA directly from soils. Applied and environmental microbiology

65(12):5409-5420.
Gaze WH, Abdouslam N, Hawkey PM, Wellington EMH. 2005. Incidence of class 1 integrons in a quaternary ammonium compound-polluted environment. Antimicrobial Agents and Chemotherapy 49(5):1802-1807.
Gillings MR. 2013. Evolutionary consequences of antibiotic use for the resistome, mobilome and microbial pangenome. Frontiers in Microbiology 4:4.
Gillings MR. 2017a. Class 1 integrons as invasive species. Current Opinion in Microbiology.
Gillings MR. 2017b. Lateral gene transfer, bacterial genome evolution, and the Anthropocene. Annals of the New York Academy of Sciences 1389:20-36.
Gillings MR, Gaze WH, Pruden A, Smalla K, Tiedje JM, Zhu Y-G. 2015. Using the class 1 integron-integrase gene as a proxy for anthropogenic pollution. The ISME Journal 9(6):1269-1279.
Gillings MR, Paulsen IT. 2014. Microbiology of the Anthropocene. Anthropocene.
Gillings MR, Paulsen IT, Tetu SG. 2017. Genomics and the evolution of antibiotic resistance. Annals of the New York Academy of Sciences 1388:92-107.
Gillings MR, Stokes HW. 2012. Are humans increasing bacterial evolvability? Trends in Ecology and Evolution 27(6):346-352.
Goldberg CS, Turner CR, Deiner K, Klymus KE, Thomsen PF, Murphy MA, Spear SF, McKee A, Oyler‐McCance SJ, Cornman RS. 2016. Critical considerations for the application of environmental DNA methods to detect aquatic species. Methods in Ecology and Evolution 7(11):1299-1307.
Green JL, Holmes AJ, Westoby M, Oliver I, Briscoe D, Dangerfield M, Gillings M, Beattie AJ. 2004. Spatial scaling of microbial eukaryote diversity. Nature 432(7018):747-750.
Grenni P, Ancona V, Caracciolo AB. 2017. Ecological effects of antibiotics on natural ecosystems: A review. Microchemical Journal.
Guerin E, Cambray G, Sanchez-Alberola N, Campoy S, Erill I, Da Re S, Gonzalez-Zorn B, Barbe J, Ploy M-C, Mazel D. 2009. The SOS Response Controls Integron Recombination. Science 324(5930):1034.
Gullberg E, Albrecht LM, Karlsson C, Sandegren L, Andersson DI. 2014. Selection of a multidrug resistance plasmid by sublethal levels of antibiotics and heavy metals. MBio 5(5):e01918-14.
Haggerty JM, Dinsdale EA. 2016. Distinct biogeographical patterns of marine bacterial taxonomy and functional genes. Global Ecology and Biogeography.
Hanson CA, Fuhrman JA, Horner-Devine MC, Martiny JB. 2012. Beyond biogeographic patterns: processes shaping the microbial landscape. Nature Reviews Microbiology 10(7):497-506.
He L-Y, Ying G-G, Liu Y-S, Su H-C, Chen J, Liu S-S, Zhao J-L. 2016. Discharge of swine wastes risks water quality and food safety: Antibiotics and antibiotic resistance genes from swine sources to the receiving environments. Environment international 92:210-219.
Heuer H, Schmitt H, Smalla K. 2011. Antibiotic resistance gene spread due to manure application on agricultural fields. Current Opinion in Microbiology 14:236-

243.
Horner-Devine MC, Lage M, Hughes JB, Bohannan BJ. 2004. A taxa–area relationship for bacteria. Nature 432(7018):750-753.
Hu Y, Yang X, Li J, Lv N, Liu F, Wu J, Lin IY, Wu N, Weimer BC, Gao GF. 2016. The transfer network of bacterial mobile resistome connecting animal and human microbiome. Applied and Environmental Microbiology:AEM. 01802-16.
Hunter ME, Dorazio RM, Butterfield JS, Meigs‐Friend G, Nico LG, Ferrante JA. 2016. Detection limits of quantitative and digital PCR assays and their influence in presence–absence surveys of environmental DNA. Molecular Ecology Resources.
Jechalke S, Schreiter S, Wolters B, Dealtry S, Heuer H, Smalla K. 2013. Widespread dissemination of class 1 integron components in soils and related ecosystems as revealed by cultivation-independent analysis. Frontiers in Microbiology 4:420.
Jerde CL, Olds BP, Shogren AJ, Andruszkiewicz EA, Mahon AR, Bolster D, Tank JL. 2016. Influence of Stream Bottom Substrate on Retention and Transport of Vertebrate Environmental DNA. Environmental Science & Technology 50(16):8770-8779.
Johnson TA, Stedtfeld RD, Wang Q, Cole JR, Hashsham SA, Looft T, Zhu Y-G, Tiedje JM. 2016. Clusters of Antibiotic Resistance Genes Enriched Together Stay Together in Swine Agriculture. mBio 7(2):e02214-15.
Jones-Dias D, Manageiro V, Ferreira E, Barreiro P, Vieira L, Moura IB, Caniça M. 2016. Architecture of Class 1, 2, and 3 Integrons from Gram Negative Bacteria Recovered among Fruits and Vegetables. Frontiers in Microbiology 7:1400.
Karkman A, Johnson TA, Lyra C, Stedtfeld RD, Tamminen M, Tiedje JM, Virta M. 2016. High-throughput quantification of antibiotic resistance genes from an urban wastewater treatment plant. FEMS microbiology ecology 92(3):fiw014.
Koczura R, Mokracka J, Taraszewska A, Łopacinska N. 2016. Abundance of Class 1 Integron-Integrase and Sulfonamide Resistance Genes in River Water and Sediment Is Affected by Anthropogenic Pressure and Environmental Factors. Microbial Ecology 72(4):909-916.
Kohanski MA, DePristo MA, Collins JJ. 2010. Sublethal antibiotic treatment leads to multidrug resistance via radical-induced mutagenesis. Molec. Cell 37(3):311-320.
Küsel K, Totsche KU, Trumbore SE, Lehmann R, Steinhäuser C, Herrmann M. 2016. How deep can surface signals be traced in the critical zone? Merging biodiversity with biogeochemistry research in a central German Muschelkalk landscape. Frontiers in Earth Science 4:32.
Lardon LA, Merkey BV, Martins S, Dötsch A, Picioreanu C, Kreft JU, Smets BF. 2011. iDynoMiCS: next‐generation individual‐based modelling of biofilms. Environmental Microbiology 13(9):2416-2434.
Lehmann K, Bell T, Bowes MJ, Amos GC, Gaze WH, Wellington EM, Singer AC. 2016. Trace levels of sewage effluent are sufficient to increase class 1 integron

692 prevalence in freshwater biofilms without changing the core community.
693 Water Research 106:163-170.
694 Levy-Booth DJ, Campbell RG, Gulden RH, Hart MM, Powell JR, Klironomos JN,
695 Pauls KP, Swanton CJ, Trevors JT, Dunfield KE. 2007. Cycling of
696 extracellular DNA in the soil environment. Soil Biology and Biochemistry
697 39(12):2977-2991.
698 Linares JF, Gustafsson I, Baquero F, Martinez JL. 2006. Antibiotics as intermicrobial
699 signaling agents instead of weapons. PNAS 103(51):19484-19489.
700 Liu X, Steele JC, Meng X-Z. 2017. Usage, residue, and human health risk of
701 antibiotics in Chinese aquaculture: A review. Environmental Pollution
702 223:161-169.
703 Lohan KP, Fleischer R, Carney K, Holzer K, Ruiz G. 2016. Amplicon-Based
704 Pyrosequencing Reveals High Diversity of Protistan Parasites in Ships' Ballast
705 Water: Implications for Biogeography and Infectious Diseases. Microbial
706 ecology 71(3):530-542.
707 Lyon DY, Monier J-M, Dupraz S, Freissinet C, Simonet P, Vogel TM. 2010. Integrity
708 and biological activity of DNA after UV exposure. Astrobiology 10(3):285-
709 292.
710 Martiny JBH, Bohannan BJ, Brown JH, Colwell RK, Fuhrman JA, Green JL, Horner-
711 Devine MC, Kane M, Krumins JA, Kuske CR. 2006. Microbial biogeography:
712 putting microorganisms on the map. Nature Reviews Microbiology 4(2):102-
713 112.
714 Mobley ML. 2009. Monitoring Earth's critical zone. Science 326(5956):1067-1068.
715 Mock T, Daines SJ, Geider R, Collins S, Metodiev M, Millar AJ, Moulton V, Lenton
716 TM. 2016. Bridging the gap between omics and earth system science to better
717 understand how environmental change impacts marine microbes. Global
718 change biology 22(1):61-75.
719 Muziasari WI, Pärnänen K, Johnson TA, Lyra C, Karkman A, Stedtfeld RD,
720 Tamminen M, Tiedje JM, Virta M. 2016. Aquaculture changes the profile of
721 antibiotic resistance and mobile genetic element associated genes in Baltic Sea
722 sediments. FEMS Microbiology Ecology 92(4):fiw052.
723 Nicholson WL. 2009. Ancient micronauts: interplanetary transport of microbes by
724 cosmic impacts. Trends in microbiology 17(6):243-250.
725 Pal C, Bengtsson-Palme J, Kristiansson E, Larsson DJ. 2015. Co-occurrence of
726 resistance genes to antibiotics, biocides and metals reveals novel insights into
727 their co-selection potential. BMC Genomics 16(1):964.
728 Pal C, Bengtsson-Palme J, Kristiansson E, Larsson DJ. 2016. The structure and
729 diversity of human, animal and environmental resistomes. Microbiome
730 4(1):54.
731 Perfumo A, Marchant R. 2010. Global transport of thermophilic bacteria in
732 atmospheric dust. Environmental microbiology reports 2(2):333-339.
733 Perron GG, Inglis RF, Pennings PS, Cobey S. 2015. Fighting microbial drug
734 resistance: a primer on the role of evolutionary biology in public health.
735 Evolutionary applications 8(3):211-222.

736 Peter H, Hörtnagl P, Reche I, Sommaruga R. 2014. Bacterial diversity and
737 composition during rain events with and without Saharan dust influence
738 reaching a high mountain lake in the Alps. Environmental microbiology
739 reports 6(6):618-624.
740 Petty NK, Zakour NLB, Stanton-Cook M, Skippington E, Totsika M, Forde BM, Phan
741 M-D, Moriel DG, Peters KM, Davies M. 2014. Global dissemination of a
742 multidrug resistant Escherichia coli clone. Proceedings of the National
743 Academy of Sciences 111(15):5694-5699.
744 Pietramellara G, Ascher J, Borgogni F, Ceccherini M, Guerri G, Nannipieri P. 2009.
745 Extracellular DNA in soil and sediment: fate and ecological relevance.
746 Biology and Fertility of Soils 45(3):219-235.
747 Pontiroli A, Simonet P, Frostegard A, Vogel TM, Monier J-M. 2007. Fate of
748 transgenic plant DNA in the environment. Environmental biosafety research
749 6(1-2):15-35.
750 Poté J, Ceccherini MT, Rosselli W, Wildi W, Simonet P, Vogel TM. 2003. Fate and
751 transport of antibiotic resistance genes in saturated soil columns. European
752 Journal of Soil Biology 39(2):65-71.
753 Poté J, Ceccherini MT, Rosselli W, Wildi W, Simonet P, Vogel TM. 2010. Leaching
754 and transformability of transgenic DNA in unsaturated soil columns.
755 Ecotoxicology and environmental safety 73(1):67-72.
756 Poté J, Mavingui P, Navarro E, Rosselli W, Wildi W, Simonet P, Vogel TM. 2009.
757 Extracellular plant DNA in Geneva groundwater and traditional artesian
758 drinking water fountains. Chemosphere 75(4):498-504.
759 Pruden A, Arabi M, Storteboom HN. 2012. Correlation between upstream human
760 activities and riverine antibiotic resistance genes. Environmental Science &
761 Technology 46(21):11541-11549.
762 Pruden A, Larsson DJ, Amézquita A, Collignon P, Brandt KK, Graham DW,
763 Lazorchak JM, Suzuki S, Silley P, Snape JR. 2013. Management options for
764 reducing the release of antibiotics and antibiotic resistance genes to the
765 environment. Environmental Health Perspectives 121:878-885.
766 Prudhomme M, Attaiech L, Sanchez G, Martin B, Claverys J-P. 2006. Antibiotic stress
767 induces genetic transformability in the human pathogen *Streptococcus*
768 *pneumoniae*. Science 313(5783):89-92.
769 Reed DC, Algar CK, Huber JA, Dick GJ. 2014. Gene-centric approach to integrating
770 environmental genomics and biogeochemical models. Proceedings of the
771 National Academy of Sciences 111(5):1879-1884.
772 Ruiz GM, Rawlings TK, Dobbs FC, Drake LA, Mullady T, Huq A, Colwell RR. 2000.
773 Global spread of microorganisms by ships. Nature 408(6808):49-50.
774 Sandberg KD, LaPara TM. 2016. The fate of antibiotic resistance genes and class 1
775 integrons following the application of swine and dairy manure to soils. FEMS
776 Microbiology Ecology 92(2):fiw001.
777 Sato T, Qadir M, Yamamoto S, Endo T, Zahoor A. 2013. Global, regional, and country
778 level need for data on wastewater generation, treatment, and use. Agricultural
779 Water Management 130:1-13.

780 Sharifi S, Murthy S, Takács I, Massoudieh A. 2014. Probabilistic parameter
781 estimation of activated sludge processes using Markov Chain Monte Carlo.
782 Water research 50:254-266.
783 Skurnik D, Ruimy R, Ready D, Ruppe E, Bernede-Bauduin C, Djossou F, Guillemot
784 D, Pier GB, Andremont A. 2010. Is exposure to mercury a driving force for the
785 carriage of antibiotic resistance genes? Journal of medical microbiology
786 59(7):804-807.
787 Sloan WT, Lunn M, Woodcock S, Head IM, Nee S, Curtis TP. 2006. Quantifying the
788 roles of immigration and chance in shaping prokaryote community structure.
789 Environmental microbiology 8(4):732-740.
790 Smil V. 2011. Harvesting the biosphere: The human impact. Population and
791 Development Review 37(4):613-636.
792 Smith DJ, Timonen HJ, Jaffe DA, Griffin DW, Birmele MN, Perry KD, Ward PD,
793 Roberts MS. 2013. Intercontinental dispersal of bacteria and archaea by
794 transpacific winds. Applied and environmental microbiology 79(4):1134-1139.
795 Steffen W, Broadgate W, Deutsch L, Gaffney O, Ludwig C. 2015. The trajectory of
796 the Anthropocene: the great acceleration. The Anthropocene Review 2(1):81-
797 98.
798 Sun M, Ye M, Schwab AP, Li X, Wan J, Wei Z, Wu J, Friman V-P, Liu K, Tian D.
799 2016. Human migration activities drive the fluctuation of ARGs: Case study of
800 landfills in Nanjing, eastern China. Journal of Hazardous Materials 315:93-
801 101.
802 Thevenon F, Adatte T, Wildi W, Poté J. 2012. Antibiotic resistant bacteria/genes
803 dissemination in lacustrine sediments highly increased following cultural
804 eutrophication of Lake Geneva (Switzerland). Chemosphere 86(5):468-476.
805 Verhougstraete MP, Martin SL, Kendall AD, Hyndman DW, Rose JB. 2015. Linking
806 fecal bacteria in rivers to landscape, geochemical, and hydrologic factors and
807 sources at the basin scale. Proceedings of the National Academy of Sciences
808 112(33):10419-10424.
809 Volkova VV, Lanzas C, Lu Z, Gröhn YT. 2012. Mathematical model of plasmid-
810 mediated resistance to ceftiofur in commensal enteric Escherichia coli of
811 cattle. PLoS One 7(5):e36738.
812 Volkova VV, Lu Z, Lanzas C, Scott HM, Gröhn YT. 2013. Modelling dynamics of
813 plasmid-gene mediated antimicrobial resistance in enteric bacteria using
814 stochastic differential equations. Scientific reports 3.
815 Wang J, Ben W, Yang M, Zhang Y, Qiang Z. 2016. Dissemination of veterinary
816 antibiotics and corresponding resistance genes from a concentrated swine
817 feedlot along the waste treatment paths. Environment international 92:317-
818 323.
819 Whitchurch CB, Tolker-Nielsen T, Ragas PC, Mattick JS. 2002. Extracellular DNA
820 required for bacterial biofilm formation. Science 295(5559):1487-1487.
821 Widder S, Besemer K, Singer GA, Ceola S, Bertuzzo E, Quince C, Sloan WT,
822 Rinaldo A, Battin TJ. 2014. Fluvial network organization imprints on
823 microbial co-occurrence networks. Proceedings of the National Academy of

Sciences 111(35):12799-12804.
Wilkinson BH, McElroy BJ. 2007. The impact of humans on continental erosion and sedimentation. Geological Society of America Bulletin 119(1-2):140-156.
Zhou B, Wang C, Zhao Q, Wang Y, Huo M, Wang J, Wang S. 2016. Prevalence and dissemination of antibiotic resistance genes and coselection of heavy metals in Chinese dairy farms. Journal of Hazardous Materials 320:10-17.
Zhu B. 2006. Degradation of plasmid and plant DNA in water microcosms monitored by natural transformation and real-time polymerase chain reaction (PCR). Water research 40(17):3231-3238.
Zhu Y-G, Reid BJ, Meharg AA, Banwart SA, Fu B-J. 2017a. Optimizing Peri-URban Ecosystems (PURE) to re-couple urban-rural symbiosis. Science of The Total Environment 586:1085-1090.
Zhu Y-G, Zhao Y, Li B, Huang C-L, Zhang S-Y, Yu S, Chen Y-S, Zhang T, Gillings MR, Su J-Q. 2017b. Continental-scale pollution of estuaries with antibiotic resistance genes. Nature Microbiology 2:16270.
Zinger L, Chave J, Coissac E, Iribar A, Louisanna E, Manzi S, Schilling V, Schimann H, Sommeria-Klein G, Taberlet P. 2016. Extracellular DNA extraction is a fast, cheap and reliable alternative for multi-taxa surveys based on soil DNA. Soil Biology and Biochemistry 96:16-19.

847 **Table 1: Dissemination of genes and microorganisms in Earth's Critical Zone.**

848 Three phenomena, or drivers, affect microbial/gene spread. These are: opportunity for
849 dispersal; stochastics (the number of foreign cells landing at a particular location,
850 processes that generate local variation such as mutation and drift); and recruitment
851 (the persistence of cells at the new location, often driven by local selection).
852 Historically, these forces generate biogeographic patterns for microorganisms that are
853 similar to those of animals and plants. Human impacts have changed the dynamics of
854 these phenomena, and are altering microbial biogeography in the process.
855