



Ambient soil cation exchange capacity inversely associates with infectious and parasitic disease risk in regional Australia

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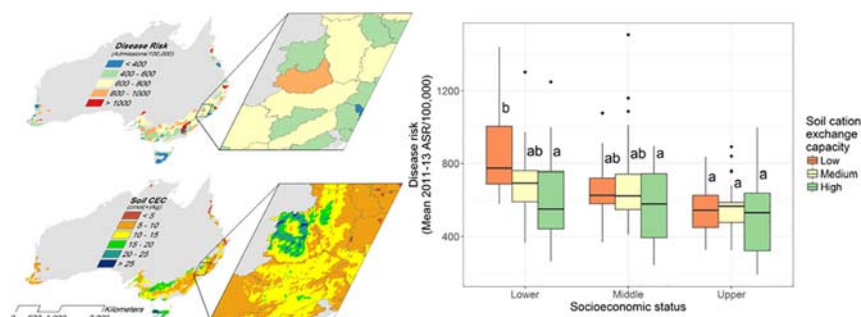
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HIGHLIGHTS

- Soil exposures may benefit human health but effects need to be explored.
- We use soil cation exchange capacity (CEC) as a proxy to test links to human health.
- We compared soils with infectious and parasitic disease risk in regional Australia.
- Effects of ambient soil quality are comparable to increasing socioeconomic status.
- Considering soil significantly improves disease risk prediction in unseen test areas.

GRAPHICAL ABSTRACT



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ABSTRACT

Human contact with soil may be important for building and maintaining normal healthy immune defence mechanisms, however this idea remains untested at the population-level. In this continent-wide, cross-sectional study we examine the possible public health benefit of ambient exposures to soil of high cation exchange capacity (CEC), a surrogate for potential immunomodulatory soil microbial diversity. We compare distributions of normalized mean 2011/12–2012/13 age-standardized public hospital admission rates (cumulative incidence) for infectious and parasitic diseases across regional Australia (representing an average of 29,516 patients/year in 228 local government areas), within tertiles of socioeconomic status and soil exposure. To test the significance of soil CEC, we use probabilistic individual-level environmental exposure data (with or without soil), and group-level variables, in robust non-parametric multilevel modelling to predict disease rates in unseen groups. Our results show that in socioeconomically-deprived areas with high CEC soils, rates of infectious and parasitic disease are significantly lower than areas with low CEC soils. Also, health inequality (relative risk) due to socioeconomic status is significantly lower in areas with high CEC soils compared to low CEC soils (Δ relative risk = 0.47; 95% CI: 0.13, 0.82). Including soil exposure when modelling rates of infectious and parasitic disease significantly improves prediction performance, explaining an additional 7.5% ($\Delta r^2 = 0.075$; 95% CI: 0.05, 0.10) of variation in disease risk, in local government areas that were not used for model building. Our findings suggest that exposure to high CEC soils (typically high soil biodiversity) associates with reduced risk of infectious and parasitic diseases, particularly in lower socioeconomic areas.

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1. Introduction

Increasing urbanisation, declining natural biodiversity, and consequent declines in population contact with environmental sources of microbial diversity, including soils, may compromise normal healthy immune system development and regulation (Rook, 2013; von Hertzen and Hahtela, 2006). Meanwhile, exposure to environmental microbial communities (microbiota) can help shape commensal microbiota in the gut and other sites, and consequently, also the development of immune status and predisposition to both infectious and non-infectious diseases (Ichinohe et al., 2011; Stein et al., 2016).

The notion of natural immunity, or enhanced protection from immune-related disease provided by exposure to environmental microbial diversity, is described by the Biodiversity Hypothesis (Hahtela et al., 2013; von Hertzen et al., 2011). Similarly, the related Old Friends Mechanism (Rook, 2013) suggests a protective immunomodulatory role for key microbial species that is lost or reduced with less exposure to natural and biodiverse environments. Employing beneficial microbiota-mediated immunomodulatory mechanisms from soil biodiversity may represent an underutilized resource for protecting and improving human health (Wall et al., 2015), and possibly a new cost-effective public health intervention (Mills et al., 2017). However, the notion that exposure to microbially-diverse soil may provide benefits to human health remains untested at the population-level.

Soils are a known reservoir of high biological diversity (Coleman et al., 2004) and catalogues of soils and their microbiota are growing due to the use of modern DNA sequencing technology (Bissett et al., 2016). However, to our knowledge, soil microbiota data are not yet available at the scale and coverage needed to compare with human health outcomes, suggesting the need for an intermediary, or proxy measure of soil microbiota with greater spatial coverage suited to epidemiological analysis. Liddicoat et al. (2016) justify the use of proxies as a pragmatic tool for investigating links between environmental microbial diversity and human health. Emerging research is demonstrating connections between soil microbiota, soil properties, and natural and anthropogenic influences including aboveground plant biomass and diversity (Delgado-Baquerizo et al., 2017; Gellie et al., 2017; Yan et al., 2018). Also, recent continent-wide soil mapping for Australia (Grundy et al., 2015) may offer possible candidate proxies, and below we highlight a key measure used in this study.

Soil microbes are variously involved in, and depend on, the development, turnover and stabilisation of soil organic matter (Kallenbach et al., 2016). Soil clay content supports water and nutrient retention, and together with plant interactions, moisture inputs, and organic matter, clay also supports soil aggregation and the resulting diversity of microbial habitats (Jastrow and Miller, 1998; Young and Crawford, 2004). As such, high soil clay content and increasing quantity and diversity of organic matter content generally contribute to greater soil microbial abundance and diversity (Torsvik and Øvreås, 2002). Soil organic matter and clay content also dominate measures of soil cation exchange capacity (CEC) (Peverill et al., 1999), since cations associate with the negatively charged sites in clay minerals and organic matter. CEC indicates the soil's storage capacity for nutrients and is estimated from the sum of exchangeable major cations (Ca^{2+} , Mg^{2+} , K^{+} , Na^{+}) expressed in centimoles of positive charge per kilogram of soil ($\text{cmol}(+)/\text{kg}$). Consequently, soil CEC should provide a useful proxy for soil microbial diversity, which we explore here using soil microbiome data relevant to our study area (refer to Methods and Fig. 2). Furthermore, Docherty et al. (2015) identified CEC as a key factor explaining variation in soil microbial community structure.

We can anticipate exposure pathways between ambient soils and people, through direct contact and diffuse airborne microbiota. In the latter case, soil microbiota form part of the so-called aerobiology (Polymenakou, 2012) that derives from the surrounding environment. In areas containing similar soils, and through diffuse aerobiology, it is plausible that soils may contribute to immunomodulatory effects

(discussed later) in individuals and potentially in local populations, including possibly through naturally-acquired herd immunocompetence.

In epidemiology, health influences and outcomes are typically studied at the level of individuals or populations (groups), or in combination in multilevel scenarios (i.e. with individual and group-level influences and outcomes) (Susser, 1994). At this early research stage, it is impractical and cost-prohibitive to undertake a large-area study of soil exposures and infectious disease outcomes in individuals. Instead, we maximise value from existing nationwide health and social context reporting data through analysis of group-level (i.e. area-based or ecological) datasets.

We also perform more advanced analyses to explore multilevel models of disease risk and the significance of soil CEC by combining group-level predictor and outcome data with pseudo individual-level probabilistic sampling of environmental exposures. Such multilevel analyses are recognized in public health (Koopman and Longini Jr, 1994; Susser, 1994), and here we add to the rare examples in the literature of so-called micro-macro studies (Croon and van Veldhoven, 2007) by offering a robust non-parametric modelling approach. Our motivation for this work is to better understand possible connections between soils and human health, and explore the previously untested influence of natural ambient soil exposures on population health.

In this study we present a cross-sectional analysis of infectious and parasitic disease and ambient environmental exposures—including soil CEC, a proxy of environmental microbial diversity—for regional Australia in 2011–13, to explore possible beneficial soil-associated influences on immune-related human health. We specifically test the hypotheses that (a) soil CEC may provide an indicative proxy of soil microbial diversity; (b) cumulative incidence of infectious and parasitic disease (hereafter termed disease risk) and the relative risk of disease or health inequality (defined below) differ with ambient soil CEC, and; (c) soil CEC is a significant predictor of infectious and parasitic disease risk.

2. Methods

2.1. Soil CEC and microbial diversity

We extracted representative soil sample data, relevant to our study, from the Biomes of Australian Soil Environments (BASE; Bissett et al., 2016; <https://data.bioplatforms.com/organization/bpa-base>) by matching available: (a) sample locations within our study area (defined below), plus a 20 km buffer zone; (b) data for exchangeable major cations (Ca^{2+} , Mg^{2+} , Na^{+} , K^{+}), which were summed to estimate soil CEC; and (c) community composition (operational taxonomic unit, OTU) data for bacteria, fungi and eukaryotes identified from soil environmental DNA (eDNA). Respective sample locations used are shown in Fig. S1 B–D. Bissett et al. (2016) and Delgado-Baquerizo et al. (2017) describe the BASE methods used for sample collection; soil chemical analyses; eDNA extraction; sequencing of the bacterial 16S rRNA gene, eukaryotic 18S rRNA gene, and fungal internal transcribed spacer (ITS) region; and the bioinformatic analyses to derive OTU abundance tables.

We filtered out samples with extreme, outlying soil conditions due to their potential to exert undue influence on our assessment of soil CEC-microbial diversity relationships. Samples were filtered out that were strongly acid ($\text{pH}_{\text{H}_2\text{O}} < 4.5$), strongly alkaline ($\text{pH}_{\text{H}_2\text{O}} > 9$), highly saline (electrical conductivity $> 8 \text{ dS/m}$), very high ($> 50\%$) clay content, or deep (900 cm soil depth). These samples were considered unrepresentative of population-soil exposures in the study area as they are uncommon in rural and regional settings (Grundy et al., 2015; McKenzie et al., 2004; Peverill et al., 1999), and represent known or potential limitations to biological activity, microbial food webs, or soil structure, and therefore may excessively impact the activity and diversity of certain microbial taxa (Barrett et al., 2004; Fierer and Jackson, 2006; Zahran, 1997). Samples that lacked major cation data required to estimate CEC were also excluded.

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