

## Induced polarization applied to biogeophysics: recent advances and future prospects

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### ABSTRACT

This paper provides an update on the fast-evolving field of the induced polarization method applied to biogeophysics. It emphasizes recent advances in the understanding of the induced polarization signals stemming from biological materials and their activity, points out new developments and applications, and identifies existing knowledge gaps. The focus of this review is on the application of induced polarization to study living organisms: soil microorganisms and plants (both roots and stems). We first discuss observed links between the induced polarization signal and microbial cell structure, activity and biofilm formation. We provide an up-to-date conceptual model of the electrical behaviour of the microbial cells and biofilms under the influence of an external electrical field. We also review the latest biogeophysical studies, including work on hydrocarbon biodegradation, contaminant sequestration, soil strengthening and peatland characterization. We then elaborate on the induced polarization signature of the plant-root zone, relying on a conceptual model for the generation of biogeophysical signals from a plant-root cell. First laboratory experiments show that single roots and root system are highly polarizable. They also present encouraging results for imaging root systems embedded in a medium, and gaining information on the mass density distribution, the structure or the physiological characteristics of root systems. In addition, we highlight the application of induced polarization to characterize wood and tree structures through tomography of the stem. Finally, we discuss up- and down-scaling between laboratory and field studies, as well as joint

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interpretation of induced polarization and other environmental data. We emphasize the need for intermediate-scale studies and the benefits of using induced polarization as a time-lapse monitoring method. We conclude with the promising integration of induced polarization in interdisciplinary mechanistic models to better understand and quantify subsurface biogeochemical processes.

**Key words:** Complex conductivity, Induced polarization, IP, Pollution, Hydrogeophysics.

## INTRODUCTION

In the past few decades, there has been an increasing recognition of the pivotal role of biological activity in subsurface processes (e.g. Konhauser 2007). Microorganisms play a crucial part in organic matter (OM) decomposition, precipitation/dissolution of minerals and a multitude of other biogeochemical cycles (e.g. Mitsch and Gosselink 2015). Additionally, plant-root systems can physically and chemically transform geological formations through cracks, water and nutrient uptake and storage (e.g. Tinker and Nye 2000). An improved understanding of the influence of the biosphere on subsurface processes is crucial in the coming years because it is linked with major economical, societal and environmental challenges. For example, microorganisms are vital in the creation of wetlands that are important CO<sub>2</sub> sequestration pools (e.g. Lamers *et al.* 2012; Pester *et al.* 2012; Bodelier and Dedysh 2013); they contribute to soil quality through OM degradation (e.g. Parnas 1975; Fang *et al.* 2005; Lehmann and Kleber 2015) or play an important role in crop growth through their interaction with plant-root systems (e.g. Avis *et al.* 2008; Grayston, Vaughan and Jones 1997; Vacheron *et al.* 2013). Soil microorganisms can even be cultured for engineering applications, such as hydrocarbon degradation, contaminant sequestration, or soil reinforcement (e.g. Atekwana, Sauck and Werkema 2000; Colwell *et al.* 2005; DeJong *et al.* 2010). Apart from agricultural applications, understanding plant systems is also of interest, for example, for pollution monitoring and degradation (e.g. Salt, Smith and Raskin 1998; Pilon-Smits 2005) and soil strengthening (e.g. Swanson and Dyrness 1975; Stokes *et al.* 2009; Ghestem, Sidle and Stokes 2011).

Although the importance of the biosphere for physical and chemical transformations of geological media is widely acknowledged, the complexity of the induced changes remains hard to capture and explain. Soil or plant analyses classically used to study these processes involve sample excavation or trenching, are destructive and typically only provide a sin-

gle snapshot in time (Atekwana and Slater 2009). Geophysical methods offer the advantage of being non-destructive, and have good spatial and temporal resolution. The study of the geophysical signature of biological processes in the subsurface is called biogeophysics. It is an interdisciplinary field at the border between geochemistry, microbiology and geophysics. In its early stages, biogeophysical investigations were mainly focused on the geophysical signature of microbial cells, of dynamic microbial subsurface processes and of their alteration of geological formations (Atekwana and Slater 2009). The first biological interpretation of geophysical signatures in the subsurface was proposed by Sauck, Atekwana and Nash (1998), who observed conductive anomalies in sediments contaminated with petroleum hydrocarbons (also see Bermejo, Sauck and Atekwana 1997). This increase of electrical conductivity was attributed to an increase of ionic strength in groundwater created by weathering of sediments grains due to microbially mediated redox reactions producing CO<sub>2</sub> and organic acids. Besides, living cells themselves are polarizable – in particular in the low-frequency range (below 100 Hz), as it has long been known from dielectric spectroscopy (e.g. Schwan 1957; Fröhlich 1975; Foster and Schwan 1989; Prodan *et al.* 2004). Electrical impedance techniques are also routinely used in medicine and food industry as various cells exhibit characteristic spectral electric signatures (e.g. Holder 2004; Bera 2014; Barsoukov and Macdonald 2018). Recent developments (e.g. Vanderborght *et al.* 2013; Martin 2012, 2015; Mary *et al.* 2017, 2018; Weigand and Kemna 2019) showed that geophysical methods are also sensitive to plant roots and processes associated with the development of plants and trees. We thus extend the definition of biogeophysics to also include the geophysical signature of plant-root systems and trees. Among the existing geophysical methods, induced polarization (IP) is particularly well-suited to study biological processes in the subsurface, as it is sensitive not only to changes in the electrolyte chemistry through electrical conduction (charge transport), but also to changes at the surface

of the mineral grains through electrical polarization (charge storage; e.g. Binley and Kemna 2005; Slater and Atekwana 2009).

In a previous review of the field of biogeophysics, Atekwana and Slater (2009) illustrate the use of geophysical methods to detect by-products of microbially mediated redox reactions (biomineralization of metallic iron minerals, calcium carbonate and magnetic minerals) and point at the use of geophysical methods (magnetic and electromagnetic methods) to directly detect microbial cells and biofilms. In this review, we focus on the use of IP (spectral and time domain) in recent biogeophysical studies. We provide an overview of the latest discoveries linking IP signals to subsurface microbial processes; and present new advances towards the characterization of plant-root systems and trees using the IP method, which were not covered in Atekwana and Slater (2009). We also propose conceptual models explaining the polarization processes around a bacterial cell and the IP response of plant-root systems, in accordance with the latest mechanistic descriptions of cells and plant-root polarization and empirical observations. Finally, we discuss challenges associated with the change of scales between laboratory results and field applications, and the joint interpretation of IP measurements with non-geophysical data sets.

## INDUCED POLARIZATION PRINCIPLES

Induced polarization (IP) is a geophysical method used to assess the electrical chargeability of the subsurface (i.e. its capacity to store charges) through the estimation of its complex conductivity  $\sigma^*$  or its reciprocal, the complex resistivity  $\rho^*$  (see Table 1). IP measures the low-frequency polarization (typically between 1 mHz and 10 kHz). It can be measured in the time domain and is then commonly referred to as time-domain IP (TDIP), or in the frequency domain and is then commonly called frequency-domain IP (FDIP) or, when screening a range of frequencies (commonly below 1 kHz), spectral IP (SIP). In TDIP, the secondary decay of the voltage is measured after the current shut-off, typically in form of the integral of the decay curve over predefined time windows (so-called integral chargeability), and the results are usually presented in terms of apparent chargeability. In FDIP, a phase-shifted voltage is measured relative to an injected alternating current. The results are expressed in terms of the amplitude and phase shift of the measured electrical impedance, which can be converted into real and imaginary conductivities,  $\sigma'$  and  $\sigma''$ , respectively, or magnitude and phase of the complex conductivity,  $|\sigma^*|$  and  $\varphi$ , respectively (see Table 1). Assuming a sufficiently high sam-

pling rate of the decay curve of the TDIP measurements, very similar information can be extracted from FDIP and TDIP measurements, and even compared using the Fourier transform. A detailed review of IP methods can be found in, for example, Sumner (1976), Ward (1980) and Binley and Kemna (2005).

To analyse the spectral information contained in the IP data, several phenomenological models are used. The most popular ones are the Cole–Cole model (e.g. Cole and Cole 1941) and the Debye decomposition (e.g. Ustra *et al.* 2016). A comparison between the two approaches can be found, for example, in Weigand and Kemna (2016). The data can then be expressed in terms of: (i) a characteristic relaxation time  $\tau_r$  and an associated chargeability  $M_t$ ; or (ii) a distribution of relaxation times, each associated with a chargeability value. These spectral parameters can in turn be related to different properties of the studied biogeochemical processes.

In the field, the IP effect is generally represented as multi-dimensional maps of apparent or inverted chargeability (usually for TDIP, but also for FDIP measurements), but can also be presented in terms of frequency-dependent resistivity, also called the percent frequency effect (PFE), or as phase shifts in FDIP. These measurements are equivalent, as they describe the ratio between polarizability and conductivity of the studied formation (e.g. Slater and Lesmes 2002). Recent developments of the method attempt to extract even more subsurface information, e.g. from the collection of full-waveform IP data (e.g. Fiandaca *et al.* 2012, 2013), or the acquisition of spectral IP data using a broad range of frequencies (e.g. Kemna *et al.* 2014; Kelter *et al.* 2018).

## NEW INSIGHTS ON MICROBIAL CELL POLARIZATION AND THEIR DETECTION

Early biogeophysical applications of induced polarization (IP) unequivocally highlighted the contribution of microbes to subsurface charge storage (e.g. Atekwana *et al.* 2004b; Atekwana and Slater 2009; Ntarlagiannis and Ferguson 2009; Zhang *et al.* 2014) or microbially mediated biogeochemical transformations (e.g. Slater *et al.* 2007; Personna *et al.* 2008). The role of microbes in modulating subsurface geoelectrical properties was first highlighted at contaminated sites (e.g. Atekwana *et al.* 2004b; Abdel Aal *et al.* 2004). Indeed, the attachment of cells onto mineral grain surfaces was postulated to increase surface roughness, thus enhancing electrochemical polarization, while biofilms were thought to enhance polarization and conduction pathways (Abdel Aal *et al.* 2004, 2009; Davis *et al.* 2006; Slater *et al.* 2007). In parallel, using dielectric

**Table 1** List of induced polarization parameters used to understand the link between the polarization effects and the biogeochemical transformations in the subsurface

List of induced polarization parameters	
$\rho^*$ ( $\Omega\cdot\text{m}$ ) = complex electrical resistivity	<b>Cole–Cole model</b> (Cole and Cole 1941):
$\sigma^*$ (S/m) = complex electrical conductivity	$\sigma^* = \sigma_\infty + \frac{\sigma_0 - \sigma_\infty}{1 + (i\omega\tau_r)^c} = \sigma_\infty \left(1 - \frac{M_t}{1 + (i\omega\tau_r)^c}\right)$
$\sigma^* = 1/\rho^*$	$c$ [] = Cole–Cole model coefficient
$\sigma^* = \sigma' + i\sigma'' =  \sigma^*  e^{i\varphi}$	$\sigma_0$ (S/m) = asymptotic low-frequency conductivity
$\sigma'$ (S/m) = real part of electrical conductivity	$\sigma_\infty$ (S/m) = asymptotic high-frequency conductivity
$\sigma''$ (S/m) = imaginary part of electrical conductivity	$M_t$ [] = total chargeability
$ \sigma^* $ (S/m) = magnitude/modulus of the complex electrical conductivity	$\tau_r$ [s] = characteristic relaxation time
$\varphi$ (rad) = phase shift or phase lag of the complex electrical conductivity	$f_r$ (Hz) = characteristic relaxation frequency ( $f_r = 1/2\pi\tau_r$ )

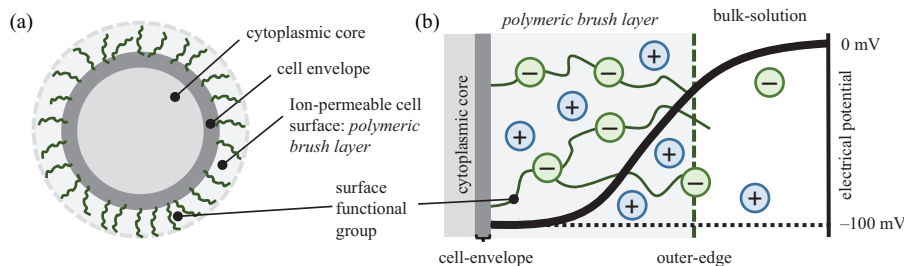
spectroscopy methods, Prodan *et al.* (2004) detected low-frequency (10 Hz) dielectric permittivity variations in *Escherichia coli* suspensions proportional to differences in their abundance, thus highlighting the polarization effect of the microorganisms themselves. In biogeophysics, it is only more recently that the contribution of the charged properties of cell membranes and biofilms themselves to polarization was studied and even quantified (e.g. Revil *et al.* 2012 and references therein). In the following subsections, we first describe the electrical behaviour and polarization of a microbial cell, before looking at the IP signature of biofilms and extracellular electron transfer. We also discuss current state and future research direction to directly detect and characterize microbial cells and biofilms using IP.

### The bacterial cell structure and its electrical behaviour

Bacterial cells consist of a cytoplasm (a strong aqueous electrolyte exhibiting conductivities of up to  $1 \text{ S}\cdot\text{m}^{-1}$ ) encapsulated by a cell membrane (also called plasma membrane or cytoplasmic membrane), surrounded by a cell wall (Willey, Sherwood and Woolverton 2014; Parker *et al.* 2017). The outermost edge of the cell wall can be conceptualized as a three-dimensional ion-permeable shell made up of a network of polymers (Carstensen and Marquis 1968; Poortinga *et al.* 2002; Sanchis *et al.* 2007; Revil *et al.* 2012). Thus, the cell-surface resembles a polymeric brush layer which harbours carboxyl, phosphate and amino surface groups and controls the cell-surface charge (Revil *et al.* 2012). The enclosing cell membrane (Fig. 1) prevents the flux of ions into the cell interior and can be considered an electrical insulator. Hence, as an approximation, the bacterial cell can be considered as a non-conductive inorganic particle (van der Wal *et al.* 1997b). Two major groupings of bacteria emerge as function of the composition of their cell wall: gram-positive and gram-negative.

They are detected via the selective gram staining procedure (Bartholomew and Mittwer 1952). Specifically, gram-positive cell walls consist of a thick (from 20 to 80 nm) and homogeneous layer of peptidoglycan, surrounding the cytoplasmic membrane (Willey *et al.* 2014). Molecules of teichoic and lipoteichoic acids are embedded into the peptidoglycan and extend to the surface of the cell wall. Gram-negative cell walls have a much thinner peptidoglycan layer (from 2 to 7 nm), surrounded by a second lipid bilayer called the outer membrane (Willey *et al.* 2014). The outer leaflet of the lipid bilayer contains lipopolysaccharides extending outward of the surface (Willey *et al.* 2014; Parker *et al.* 2017). Despite these differences, the zeta potential of gram-positive and gram-negative bacteria has been shown not to vary systematically (Baszkin and Norde 1999; Poortinga *et al.* 2002). Thus, within the context of this review, we have generalized the cell-membrane model for both types (Fig. 1).

Protonation and deprotonation of functional groups within the polymeric brush layer result in the development of charges at bacterial surfaces (Carstensen and Marquis 1968; Poortinga *et al.* 2002; Claessens *et al.* 2004). Within the near-neutral pH range (pH 5–7), most gram-positive and gram-negative bacteria are net-negatively charged due to the higher proportion of carboxyl and phosphate groups with respect to amine groups, and their respective lower point of zero charge (Poortinga *et al.* 2002). The net-negative surface charge is neutralized by the electrostatic attraction of counterions, and their accumulation near the cell-membrane surface, resulting in an electrical double layer (EDL). Unlike the EDL of mineral grains (very thin in comparison to the radius of curvature of the particle; van der Wal *et al.* 1997a), the EDL of cells originates at, and within, the polymeric brush layer (Poortinga *et al.* 2002). The fixed negative charges are spatially distributed over the complete thickness of the polymeric brush layer, on the order of 10 nm (Fig. 1b). High fixed-charge



**Figure 1** (a) Conceptual diagram of a generalized EDL model of a bacterial cell. The cell envelope encapsulates the conductive cytoplasmic core and is made up of a plasma membrane and a cell wall. The cell wall consists of (i) either a thin layer of peptidoglycan, and an outer membrane for gram-negative bacteria, or a thick peptidoglycan layer for gram-positive bacteria (Poortinga *et al.* 2002; Willey *et al.* 2014); and (ii) a charged, ion-permeable cell surface, denoted here as a *polymeric brush layer* (figure modified from Revil *et al.* 2012).

(b) Potential distribution in the ion-permeable cell surface (diagram content is modified from Poortinga *et al.* 2002). (Note that the cell envelop structure presented here is meant to depict a generalized membrane for both gram-positive and -negative bacteria. The contribution to the IP effect is thought to be dominated by the charged functional groups in the ion-permeable cell surface.)

densities within the ion-penetrable cell surface, in conjunction with a high mobility of the corresponding counterions, yield a high surface conductivity of most bacterial cells.

### Polarization of bacterial cells

Revil *et al.* (2012) developed a quantitative model to describe the IP of bacteria, where Stern layer polarization and surface conduction within the polymeric brush (acting as a fixed-charge membrane at constant pH) drive significant charge storage (increase in  $\sigma''$ ) at the outer edge of cells. The Cole–Cole model (Cole and Cole 1941) was used to explain the low-frequency relaxation (1–100 Hz) associated with bacterial polarization, as a function of the ionic mobility within the cell EDL and the size of the polarizing cells (Revil *et al.* 2012). The modelling results of Revil *et al.* (2012) were supported by Zhang *et al.* (2014), who measured increasing frequency-dependent  $\sigma''$  signals with increasing cell density of both, cell suspensions and sand-cell mixtures of *Zymomonas mobilis*. Consistent, low-frequency  $\sigma''$  peaks were measured around 0.1 and 10 Hz.

Furthermore, Mellage *et al.* (2018b) monitored the  $\sigma''$  response of *Shewanella oneidensis* MR1 under growth conditions, in saturated quartz sand and iron-coated sand packed column reactors. The proportional increase in  $\sigma''$  with increasing biomass density further confirmed the direct dependence of  $\sigma''$  on cell density. Mellage *et al.* (2018b) also extracted relaxation times ( $\tau$ ) from their measured spectra, using the Cole–Cole model ( $\sigma''$  spectral peaks between 5 and 40 Hz). Their relaxation times  $\tau$  showed distinct variations as a function of the available electron accepting pathway in the column reactors: (1) reduction of aqueous nitrate and nitrite and (2) reduction of solid-phase ferric iron. The authors argue that  $\tau$

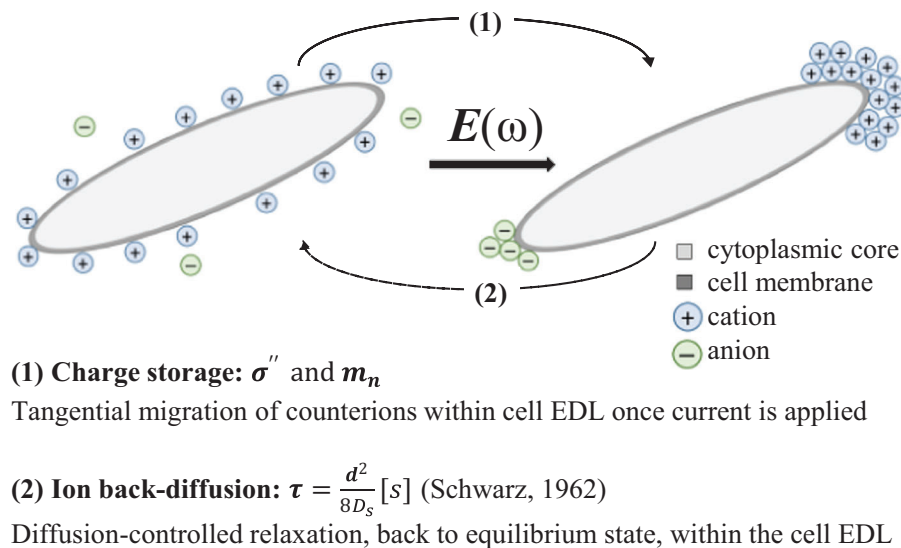
can provide qualitative information relating to the size, activity and dynamic surface charging properties of the polarizing bacterial cells for spectra with well-defined peaks. The conceptualized Stern layer polarization of *S. oneidensis* is shown in Fig. 2.

However, strict Stern layer polarization is not expected based on the structure of the outer cell membrane. The poly-disperse, permeable volume of the brush layer is in fact a three-dimensional distribution of charge (Poortinga *et al.* 2002; Revil *et al.* 2012). This is similar to the polydisperse polymeric coating on engineered nanoparticles, which deviates from the standard EDL model for non-permeable surfaces and exerts a strong control on electrochemical polarization (Mellage *et al.* 2018a). Polarization of coated nanoparticles deviates to a larger extent from the classical Stern layer model due to the much larger polydisperse volume in relation to the particle itself (Mellage *et al.* 2018a). Although a relatively thin layer in comparison to coated nanoparticles, polarization in the brush layer of bacterial cell membranes could, to some extent, also be controlled by charge accumulation within that layer. Incorporating elements from the NICA–Donnan model (Marinsky, Lin and Chung 1983; Koopal *et al.* 1994; Benedetti *et al.* 1996; Kinniburgh *et al.* 1996), which accounts for the three-dimensional structure of soft polymeric substances, can be useful in improving our understanding and conceptualization of ionic migration along the surface of a bacterial cell, as has been postulated in Mellage *et al.* (2018a).

### Biofilms

Biofilms are structured communities of cells enclosed in complex, hydrated (>80% water) organic structures attached





**Figure 2** Conceptual diagram depicting Stern layer polarization tangential to a negatively charged bacterial cell (modified from Mella *et al.* 2018b). Polarization of cells induces charge storage, quantified as imaginary conductivity ( $\sigma''$ ) or normalized chargeability ( $m_n$ ). The time scale for ion back diffusion (i.e. the relaxation time,  $\tau$  [s]) is governed by the effective length scale or diameter ( $d$ ) of the polarizing particles, after Schwarz (1962). Surface diffusion, dependent on ionic mobility within the cell EDL, is quantified by the effective-surface diffusion coefficient,  $D_s$  ( $\mu\text{m}^2 \text{s}^{-1}$ ).

to mineral and organic surfaces (Herbert-Guillou *et al.* 1999; Whitchurch *et al.* 2002; Kanematsu and Barry 2015; Berlanga and Guerrero 2016). The enclosing matrix is composed of diverse extracellular polymeric substances (EPS) such as polysaccharides, glycoproteins, lipopolysaccharides, uronic acids and extracellular DNA excreted by the bacteria (Poortinga *et al.* 2002; Whitchurch *et al.* 2002). The ions and nutrients transported in the aqueous biofilm phase make biofilms excellent ionic conductors (Malvankar *et al.* 2012; Beyenal and Babauta 2015). Potassium ion-channel-based electrical signalling within biofilms can attract distant cells (Humphries *et al.* 2017), and biofilm matrices can also include noncellular materials, such as minerals, clay or silt particles, depending on their external environment (Donlan 2002), which contribute to enhanced surface conduction pathways. Furthermore, EPS might display semi-conductive properties once hydrated or ionized and behave like polymer electrolytes or conductive polymers (Borole *et al.* 2011). However, the electrical conductivity of biofilms has never been measured *in situ* (i.e. under natural conditions, while still attached to surfaces; Borole *et al.* 2011).

The role of biofilms in modulating the IP response of biologically active subsurface systems was suggested by Davis *et al.* (2006), who measured increasing charge storage ( $\sigma''$ ) during biofilm growth in biostimulated column experiments.

Pore structure-induced changes, due to processes such as bio-clogging, were postulated as the major drivers of biofilm-mediated SIP responses (Davis *et al.* 2006, 2010; Ntarlagianis and Ferguson 2009; Wu *et al.* 2014). While bioclogging unequivocally mediates pore geometry and therefore affects current flow and polarization, electromigration and charge storage within the biofilm matrix as well as within the bacterial surfaces concurrently contribute to the measured SIP responses (Abdel Aal *et al.* 2010). Albrecht *et al.* (2011) measured a strong correlation between phase shift (as large as 50 mrad, at 4 Hz) and biofilm volume growth in *Burkholderia sp.* (NAH1)-mediated phenanthrene degradation experiments. Identical experiments performed with a non-biofilm producing strain, *Stenotrophomonas maltophilia* (MATE10), showed similar growth kinetics, but 10 times lower phase shift magnitude. Rosier *et al.* (2019) measured increasing  $\sigma''$  signals (characteristic peak frequency between 1 and 10 Hz) with increasing cell density of *Pseudomonas aeruginosa* suspensions in experimental columns. The authors reported a direct dependence of measured SIP signals in response to artificially added biofilm components (alginate, phenazine and DNA), both from signal contributions of elevated concentrations of the components themselves, and when added to *P. aeruginosa* suspensions. Furthermore, they suggest that SIP can be developed to assess specific biofilm developmental changes.

Variations in biofilm constituents are a function of community development and microbial metabolic state and function. For example, the accumulation of phenazines can enhance extracellular electron transfer, potentially yielding by-products that affect polarization (Wang *et al.* 2014). Biofilm development requires extracellular DNA (eDNA), which can be produced by certain types of bacteria (Whitchurch *et al.* 2002). Cell lysis can also result in the release of large quantities of eDNA, which could suppress polarization, for example, by eDNA-cation chelation (Mulcahy *et al.* 2008). The suppression of polarization (decrease in  $\sigma''$ ) during cell lysis was previously reported by Davis *et al.* (2006). The complex nature of biofilm-mediated polarization mechanisms is clearly a function of the transient nature of biofilm development and the changes experienced or induced by the microbial community.

While efforts have already been made to isolate SIP signal contributions stemming from various biofilm components (e.g. Rosier *et al.* 2019), the determination of the sensitivity of SIP to specific biofilm development changes requires an experimental shift from suspension measurements to experiments under growth conditions. Time-lapse monitoring of both changes in biofilm composition and structure and concurrent SIP signals would allow us to develop direct linkages between a process of interest and variations to SIP-derived parameters (e.g.  $\sigma''$ ,  $\tau$ ,  $M_n$ ).

### Extracellular electron transfer

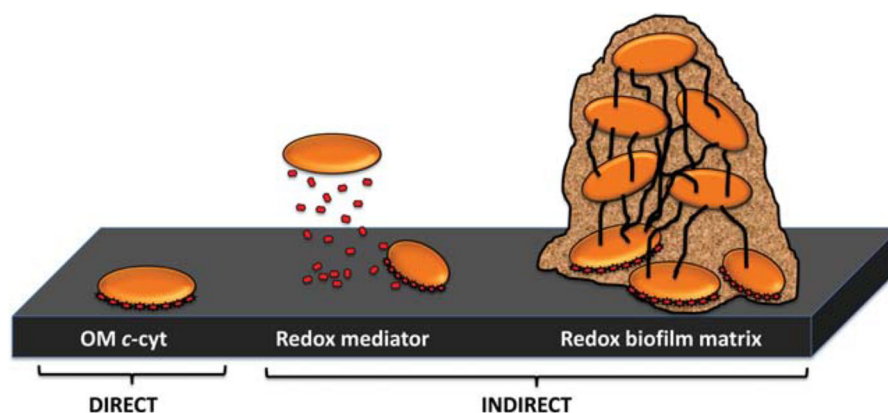
The vast majority of bacteria take up soluble electron acceptors and donors. Some bacteria, however, have shown the ability to remotely access electrons either within adjacent solid (mineral) electron donors or at a distance (e.g. *Geobacter* spp. and *Shewanella* spp.; Babauta *et al.* 2012; Nealson 2017). These electrochemically active microorganisms perform extracellular electron transfer (EET). They can form electroactive biofilms that have been extensively studied for the past two decades for their ability to generate electrical energy from biochemical energy in microbial fuel cells (Logan and Regan 2006). Both direct and indirect mechanisms have been proposed to explain the electron transfer from the cell membrane of the bacteria located inside biofilms (sometimes more than 20-cell lengths away) to an external solid surface (summarized in Fig. 3). The exact mechanisms, however, remain intensively debated (Borole *et al.* 2011; Babauta *et al.* 2012; Bond *et al.* 2012; Dominguez-Benetton *et al.* 2012; Beyenal and Babauta 2015). Due to the inherent sensitivity of IP signals to electrochemical processes within biofilms, and at the

cell-membrane–fluid interface, IP can be applied to shed light on the mechanisms aiding EET.

### Direct detection of microbial cells and biofilms using induced polarization

Geomicrobiological quantification techniques are destructive, and sample preparation often alters the microbial metabolic state, when subjecting samples to detection-enhancing treatments (Vives-Rego, Lebaron and Nebe-von Caron 2000; Wardman, Nevin and Lovley 2014). Furthermore, extracting *in situ* information of microbially mediated reactions, at both the field and laboratory scales, requires destructive sampling methods and provides only spatially and temporally scattered sampling points (e.g. Anneser *et al.* 2008). High spatiotemporal resolution IP measurements can fill the methodological gap highlighted within the geomicrobiological literature (e.g. Kappler *et al.* 2005; Wardman *et al.* 2014) and provide non-destructive information that is able to capture the adaptability of microbial communities at the field and laboratory scales. The challenge in the adoption of SIP as a geomicrobiological measurement tool lies in the inherent ambiguity of signal interpretation, due to the contribution of multiple properties and processes. Thus, a better mechanistic understanding of the polarization processes involved not only by the presence of the microbial cells themselves but also by their interactions with their environment is necessary. To solve this ambiguity, time-lapse IP measurements can also be part of the solution. They record dynamic processes that change more rapidly than most petrophysical soil properties, effectively normalizing the effect of the background signal contributions.

We envision that the production of further IP data sets, monitoring microbial dynamics, will help calibrate IP tools directly applicable in biogeochemistry and geomicrobiology experimental set-ups. For a specific porous medium, SIP signal changes can be used to directly compute microbial growth rates, and, subsequently, rates of substrate turnover, simply by integrating electrode arrays into conventional flow-through column and/or tank experiments. The availability of real-time data can aid in experimental design, specifically when timing invasive sampling events. Conceivably, technological and theoretical advances will eventually lead to similar quantitative applications at the lysimeter and field scales. Improving our understanding of microbially mediated IP responses, laying the foundations for a unified theoretical framework for polarization of bacteria and biofilms in porous media and developing IP tools directly integrated in biogeochemical or geomicrobiological experimental set-ups can help position IP



**Figure 3** Electron transfer by (left) direct contact between the insoluble electron donor/acceptor (black surface) and a redox-active component associated with the cell surface such as c-type cytochromes (OM c-cyt), (middle) the diffusion of soluble redox mediators (e.g. flavins, phenazines and quinones), also known as electron shuttles, and (right) the production of elongated conductive pili or nanowires and redox cofactors (c-type cytochromes) within biofilms (sequential electron-transfer self-exchange reactions; Borole *et al.* 2011).

as a bridge between our understanding of microbial dynamics and our ability to measure it.

## MICROBIALY MEDIATED PROCESSES AND APPLICATIONS

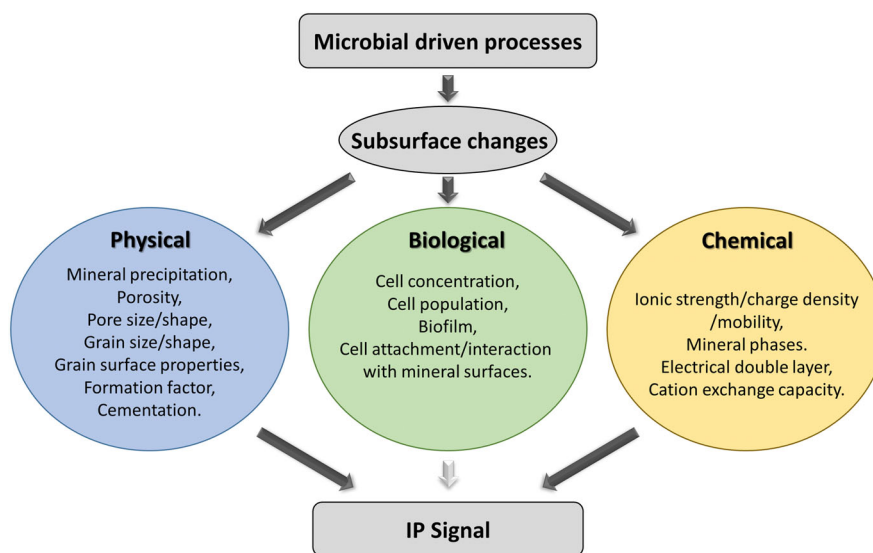
Before detecting the polarization signature of subsurface microorganisms themselves, the first biogeophysical studies recognized the role of microbially mediated processes in the measured IP signal. Indeed, early observations showed ‘anomalous’ conductive signals at hydrocarbon-contaminated sites (e.g. Bermejo *et al.* 1997; Atekwana *et al.* 2000, 2004a, b), which were attributed to microbially driven hydrocarbon degradation and the associated release of metabolic by-products (Atekwana and Slater 2009; Atekwana and Atekwana 2010 and references therein). Most of the current biogeophysical work regarding microbial activity in the subsurface is still focused on microbially mediated processes. The main reasons for this are twofold: (i) there is an urgent need for characterization and monitoring of microbially mediated processes in contaminated environments, and (ii) the geophysical signals generated by the by-products of microbial processes are significantly larger than the signal from the microbial presence or activity *per se*. Indeed, microbially mediated processes trigger physical changes (e.g. mineral precipitation, bioclogging at the grains surface and change in grain roughness) and chemical changes (e.g. mineral etching, mineral dissolution and redox reactions catalysed by microorganisms) that directly impact the resulting IP signal (Fig. 4; e.g. Atekwana and Slater 2009; Heenan *et al.* 2013; Kimak *et al.* 2019). Common applications of the IP method

in biogeophysics linked with microbially mediated processes include hydrocarbon spill characterization and monitoring (e.g. Flores-Orozco *et al.* 2012; Cassiani *et al.* 2014; Heenan *et al.* 2015; Ntarlagiannis *et al.* 2016, 2018; Kimak *et al.* 2019), contaminant sequestration and/or immobilization and mineral precipitation. In the last decade, new areas have also emerged, such as calcite biomineralization or OM characterization in peatland studies. In this section, we will present recent insights related to these applications before discussing direct detection of microbially mediated processes using IP.

### Hydrocarbon degradation

Hydrocarbon-impacted porous media appear to have a complex, but clearly measurable, IP signature that provides information on the hydrocarbon contaminant and on the biogeochemical processes associated with the hydrocarbon degradation (Davis *et al.* 2006; Schmutz *et al.* 2010, 2012; Ustra *et al.* 2012; Heenan *et al.* 2013; Personna *et al.* 2013b; Kessouri *et al.* 2016; Fernandez *et al.* 2018; Kimak *et al.* 2019). One of the main reasons for the complex behaviour of the signature lies with the nature and the properties of hydrocarbons. Non-polar (i.e. non-wetting) hydrocarbons tend to increase the IP signature of the subsurface (Revil, Schmutz and Batzle 2011; Flores-Orozco *et al.* 2015; Johansson, Fiandaca and Dahlin 2015), while polar (i.e. wetting) ones have shown to either increase (Vanhala 1997; Kemna, Binley and Slater 2004; Deceuster and Kaufmann 2012) or dampen the IP effect (Ustra *et al.* 2012; Personna *et al.* 2013b). In addition, environmental conditions (e.g. geology) and biodegradation





**Figure 4** Microbially driven processes that lead to enhanced IP signals through physical, chemical and biological subsurface changes. The lighter grey arrow between ‘biological’ and ‘IP signal’ represent the weaker influence of the direct biological markers compared with biologically mediated physical and chemical transformations. Modified from Atekwana and Slater (2009).

of hydrocarbons through redox reactions lead to additional physical and chemical changes in the subsurface with measurable IP signatures (Fig. 4), which introduces ambiguity in data interpretation (Personna *et al.* 2013a; Ntarlagiannis *et al.* 2016, 2018; Fernandez *et al.* 2018; Kimak *et al.* 2019).

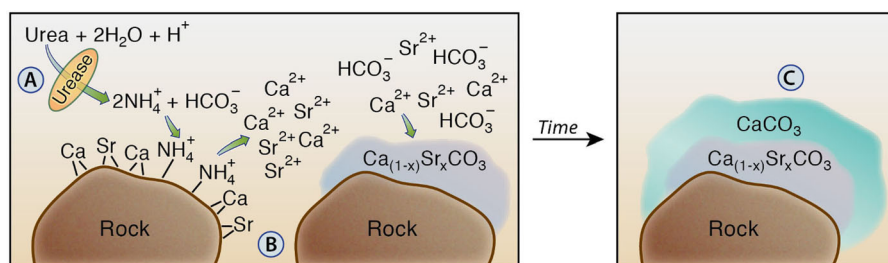
IP applications at the field scale are becoming increasingly common (Deceuster and Kaufmann 2012; Flores-Orozco *et al.* 2012, 2018; Ntarlagiannis *et al.* 2016, 2018), yet the interpretation of imaging results is still open to debate. Studies have shown that the IP response tends to increase with subsurface hydrocarbon concentration (Flores-Orozco *et al.* 2015; Johansson *et al.* 2015) and this is often attributed to microbially mediated degradation of hydrocarbons. On the contrary, there are observations with negligible polarization in the case of oversaturation of hydrocarbons and/or the presence free-phase non-aqueous phase liquids (NAPL; Flores-Orozco *et al.* 2012). Complex subsurface conditions, including infrastructure and groundwater level changes, can also contribute to non-trivial IP interpretation (Flores-Orozco *et al.* 2018).

Laboratory research is paving the way for quantitative interpretation of IP signals for hydrocarbon biodegradation. The links of IP signals to the type of hydrocarbon (e.g. Schmutz *et al.* 2010; Revil *et al.* 2011; Ustra *et al.* 2012; Personna *et al.* 2013b) and to bioremediation processes (e.g. Heenan *et al.* 2013; Kimak *et al.* 2019) have been identified. The next step should be to quantify these signals and try to deconvolute the spectral response of remediation processes in complex environments.

Advances are also being made in modelling. For example, Bückner *et al.* (2017) proposed an extension of the analytical model for membrane polarization to study the IP effect associated with the presence of both ‘wetting’ and ‘non-wetting’ hydrocarbon compounds in the pore space. Their model is consistent with field and laboratory observations, describing a decrease of the IP response with hydrocarbon saturation in the case of wetting hydrocarbons and in high saturation cases for non-wetting ones. Depending on the surface properties of the particular hydrocarbon contaminant, the extended model (Bückner *et al.* 2017) also explains increased IP responses at intermediate hydrocarbon concentrations. Biodegradation of hydrocarbon, as well as stimulated microbial activity, still needs to be taken into account in the modelling of the polarization mechanisms.

### Mineral precipitation

In subsurface remediation strategies, typically, the desired result is complete removal or degradation of the contaminant. For some forms of contaminants that cannot be degraded (e.g. radionuclides and toxic metals), alternative remediation approaches can consist in immobilization or sequestration *in situ*. A common example is urea hydrolysis based co-precipitation of radionuclides with calcite, as shown in Fig. 5 (Colwell *et al.* 2005; Fujita *et al.* 2008, 2010; Wu *et al.* 2011; Smith *et al.* 2012). Urea hydrolysis is catalysed by the urease enzyme, expressed



**Figure 5** Conceptual diagram of ureolytically driven calcium carbonate precipitation approach for remediation of  $^{90}\text{Sr}$  contaminated geologic media. (a) Microbially catalysed hydrolysis of urea. (b) Cation exchange and calcium carbonate precipitation. (c) Continued precipitation of calcium carbonate (from Wu *et al.* 2011).

by many microorganisms in the subsurface to harvest nitrogen.

Monitoring tools are needed to assess the efficiency of immobilization or sequestration remediation approaches. IP has been suggested, and tested, as a complementary method. Contaminant immobilization in mineral phases often leads to changes in pore chemistry and fluid–mineral interfacial charge properties. Given its sensitivity to capture changes in these parameters, the IP method has great potential for monitoring contaminant immobilization in mineral phases.

Another potential application of the IP method is in soil enhancement projects. A common approach to increase soil strength/stiffness is through the precipitation of a new mineral phase such as calcite. Microbially induced calcite precipitation (MICP) is taking advantage of the same ureolytic processes (Fig. 5) but with a different objective. MICP generally results in biomineralization of calcium carbonate, where the dominant stable mineral phase is calcite (Saneiyan *et al.* 2019). Calcite acts as a binding agent in the soil and could be used for crack remediation, erosion prevention, dust control and infrastructure engineering applications (e.g. supporting foundations and restoration of limestone buildings; DeJong *et al.* 2010, 2013; Dhami, Reddy and Mukherjee 2013).

Monitoring studies including SIP measurements successfully established the method's sensitivity to calcite precipitation and provided quantitative information on the precipitation rates (Wu *et al.* 2008, 2009, 2010, 2011; Saneiyan *et al.* 2018). Experiments on abiotic calcite precipitation on glass beads at the bench-scale show that SIP responses to calcite precipitation exhibit peak frequencies between 100 Hz and 1 kHz (Wu *et al.* 2010). Ottawa sand and sand-clay mixtures show SIP peak relaxation frequencies between 1 mHz and 1 Hz (depending on soil mixture type; Saneiyan *et al.* 2018). MICP processes had similar SIP signatures, however, with higher signal magnitude (higher maximum phase amplitude). This difference can be due to different precipitation patterns

and processes, the impact of microbes and/or mineral distribution style (Saneiyan *et al.* 2016). A recent field study from Saneiyan *et al.* (2019) demonstrated the benefit of IP monitoring during a field application of MICP, where IP successfully provided near real-time information on MICP progress. Further research is needed to extract quantitative information on calcite precipitation rate from field TDIP measurements.

Another benefit of MICP is the potential for atmospheric  $\text{CO}_2$  sequestration, as well as  $\text{Ca}^{2+}$  removal from wastewater, rendering the IP method a potential monitoring tool for engineering solutions to climate change (Hammes *et al.* 2003; Wanjari *et al.* 2011; Yadav *et al.* 2011).

### Organic matter and peatlands

A very promising application of the IP method is the study of soil organic matter (SOM). The creation and decomposition of SOM, catalysed by microorganisms, is a key factor in the carbon cycle, and subsequent greenhouse gas emissions and retention of nutrients and contaminants (Lehmann and Kleber 2015). Monitoring SOM properties and stocks is therefore of great importance, but also highly challenging due to the complex interactions between SOM and soil minerals, spatial heterogeneity and SOM dynamics. These challenges call for geophysical methods, from which the IP is the best candidate due to its high sensitivity to cation exchange capacity (CEC), a characteristic SOM property (Vinegar and Waxman 1984).

Early studies were successful in linking IP with organic soil depth (Slater *et al.* 2002), and the decomposition state of peat (Kettridge *et al.* 2008) in peatlands. Schwartz and Furman (2015) examined the impact of SOM on the IP signature of soil, and successfully linked the IP signals with the SOM, soil mineral interactions and the chemical composition of the soil solution. Furthermore, Walter *et al.* (2015) reported a strong correlation between peat CEC and quadrature conductivity (IP parameter), while Ponziani *et al.* (2012) could not find

any correlation between the IP response and various sample characteristics such as decomposition state, water content, organic content and CEC.

The above studies demonstrate both the great potential of the IP method to characterize and monitor SOM and peatlands, but also the need for more studies that will clarify the governing mechanisms controlling the IP signature of SOM and its dependence on biogeochemical properties and processes.

#### Direct detection of microbially mediated processes using induced polarization

As an indirect method, interpretation of IP signals is always challenged with non-uniqueness and ambiguity. For example, in the case of contaminant immobilization/sequestration monitoring, it is very challenging to directly link IP signals with the rate or concentration of metal sequestered in the host material. The same applies to hydrocarbon degradation studies: IP can be used to monitor degradation, and even biogeochemical activity, but exact interpretation of the stage of the hydrocarbon or the concentration is far from trivial. While numerous laboratory-based studies have established the sensitivity of IP signals to a variety of targets (e.g. precipitation processes that are potentially effective at immobilizing or sequestering contaminants, hydrocarbon contaminants), application of such monitoring approaches in the field has been sparse. This is largely attributed to the intertwined and interactive processes, such as changes in lithology, soil structure and moisture dynamics, each having its own IP characteristics that complicate the decoupling of those signals associated with the contaminant sequestering precipitation processes. To solve this challenge, future studies need to couple IP measurements with direct biogeochemical measurements and mechanistic models. Further research effort will need to be spent on the development of such mechanistic models for better understanding the IP signal, as well as making links with underlying reaction mechanisms. As demonstrated in Wu *et al.* (2011), while geophysical approaches provide indicator signals of system dynamics, mechanistic models, such as reactive transport modelling, are important to help understand changes that drive geophysical signatures.

### ROOT CHARACTERIZATION

An emerging area of research in the near-surface geophysics community is the use of induced polarization (IP) measurements to investigate plant-root systems. Root system architec-

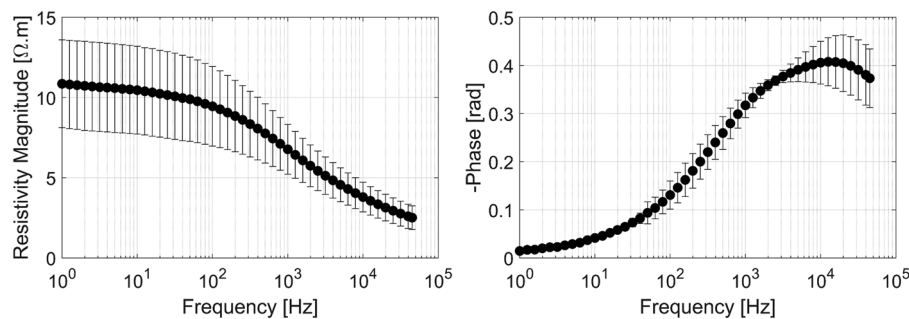
ture plays an important role for plant water uptake and thus affects both the hydrological cycle and the land-surface energy balance. In addition, root water uptake is directly related to crop growth, playing an important role in agricultural food production. Geophysical methods, and particularly IP, need to be evaluated as non-destructive methods to characterize and monitor plant-root systems. Vanderborght *et al.* (2013) extensively reviewed the use of electromagnetic methods sensitive to conduction and polarization processes for root investigations: ground-penetrating radar (GPR), electrical resistivity tomography (ERT) and IP. The magnitude of the complex electrical conductivity, through ERT, has been extensively used to image soil-water content changes associated with root water uptake (e.g. Garré *et al.* 2011; Cassiani *et al.* 2015). In the following subsections, we first describe the known electrical behaviour of a root, before reviewing studies characterizing the IP signal of single roots and plant-root systems. We then review imaging studies of plant-root systems in their environment (soil or hydroponic solution) using indirect current injection methods. We finally develop a conceptual model that summarizes our current understanding of the IP signature of root cells.

#### The root structure and its electrical behaviour

A key issue with the use of ERT for the direct detection of roots is that the expected contrast in electrical properties between soil and roots is ambiguous in the presence of soil-water content variation, since the real part of the conductivity of plant roots is expected to fall between the values for dry and saturated soil. The contrast in the imaginary part of the electrical conductivity between soil and roots is expected to be stronger because of electrical polarization processes associated with the EDLs that exist at charged interfaces associated with roots. In particular, the inside of the roots consists of cells that are known to have a plasma membrane and an associated EDL (e.g. Wang *et al.* 2011) that is expected to polarize. The outside of the root consists of an epidermis surrounding a cortex, both composed of cells and expected to provide a resistive barrier to current flow.

#### Induced polarization signature of single roots and root system

An example of the frequency-dependent electrical properties of a single root segment is shown in Fig. 6. It can be seen that the magnitude of the resistivity varies between 3 and 10 ohm.m for an 8-day-old maize root. At the same time, the measured phase increases strongly with frequency up to a



**Figure 6** Average magnitude (left) and phase (right) of the complex electrical resistivity of an 8-day-old maize root determined from four-point SIP measurement on four root segments of living seedlings. Error bars indicate the standard deviation of the four measurements. The SIP measurements were made with a custom-made sample holder where both current and potential electrodes were connected with the root segment using a conductive gel (Ehosioko *et al.* 2018).

maximum of 400 mrad at 10 kHz. This high relaxation frequency suggests a relatively small length scale of polarization. The electrical properties of root segments cannot be obtained using *in situ* measurements. Therefore, several studies have used so-called stem-based excitation methods to investigate the electrical properties of root systems. In this approach, the electrical properties of roots are determined by measuring the electrical potential that results from the application of an electrical field between two electrodes, one of them in the plant stem, and the other one in the growing medium (soil or hydroponic solution). Given the relatively low resistivity of segments of young roots (see Fig. 6), it may be expected that current flows preferentially through the roots in this type of set-up. Chloupek (1972) was one of the first to apply this method and reported a correlation between root mass and root electrical capacitance at 1 kHz (the electrical capacitance, the imaginary part and the phase of the electrical conductivity are all related to energy storage processes). Since then, a linear relationship between root mass and root electrical capacitance has been reported by several authors (Kendall, Peferson and Hill 1982; van Beem, Smith and Zobel 1998). Given the strong polarizability of root segments shown in Fig. 6, these relationships are not surprising. These results are usually interpreted using the electrical circuit model proposed by Dalton (1995). In this model, the root behaves like a cylindrical capacitor and the root system capacitance can be obtained by summing (in parallel) the capacitance of each root segment. A different circuit model was proposed by Dietrich *et al.* (2012), who suggested that the root behaves like a continuous dielectric material embedded in a medium of higher capacitance. The theoretical consequence of this alternative model is that the capacitance measured between the root and the soil/hydroponic medium is related to the root cross-sectional area (Dietrich *et al.* 2012, 2013).

In several studies, multi-frequency measurements of the electrical properties of root systems were made. For example, Cao *et al.* (2011) made two-point SIP measurements (in the Hz to kHz frequency range) to separate between the root and stem capacitances and achieved a better correlation between root traits and root electrical properties than in single-frequency measurements. The importance of multi-frequency measurements was recently also emphasized by Postic and Doussan (2016), who showed that the correlation between root traits and electrical properties is frequency dependent. Again, this is also consistent with the strong frequency-dependent phase response of root segments shown in Fig. 6.

#### Imaging of the induced polarization response of root systems in their environment

Time-lapse IP imaging is required to spatially and temporally differentiate root structure and activity. So far, distinct polarization signatures of non-woody crop roots have been reconstructed from spectral IP 2D or 3D images in controlled laboratory experiments on root systems embedded in aqueous solutions (Weigand 2017; Weigand and Kemna 2017, 2019). These studies showed that the spatial resolution of IP imaging is not sufficient to resolve single roots, but that the root extent can potentially be reconstructed. Interestingly, the frequency dependence of the electrical properties of root signals was also evident in the imaging results (see Weigand and Kemna 2017), providing potential to improve measurement results by careful selection of measurement frequencies. This will be especially helpful in cases where the electrical properties of both soil and root exhibit strong frequency dependence.

Apart from providing information on the root system extension, it has also been shown that spectral IP imaging can capture information on functional processes within the root

system. For example, Weigand and Kemna (2017) showed a temporal and spatial decrease of polarization strength during prolonged nutrient deprivation of oilseed plants, indicating a sensitivity to nutrient uptake and translocation mechanisms. This is further supported by the observation that decapitated plants, which cannot take up water and nutrients, did not show a decrease in electrical polarization under nutrient deprivation (Weigand and Kemna 2019).

A remaining challenge is the direct imaging of root systems in soil. In such systems, interpretation is complicated by the superposition of root signatures and soil polarization signatures and associated processes, such as water dynamics, salinity and temperature changes (e.g. Mary *et al.* 2017; Weigand 2017). In particular, the SIP response of unsaturated soil is still poorly understood (e.g. Breede *et al.* 2012; Mainault, Jougnot and Revil 2018). Early attempts to image coarse root systems under controlled field conditions using simple mapping approaches showed promising results, but highlighted the importance of incorporating anisotropy into the measurement and analysis procedures (Mary *et al.* 2017). Possible solutions might also be found in the combination of imaging with stem-injection set-ups, as recently done by Mary *et al.* (2018) for grapevine root systems using resistance measurements. The approach combines both increased current flow (information content) through the root system with the spatial differentiation of imaging methods. Finally, multi-frequency measurements also seem essential for root investigations in soils and substrates. Such measurements not only help in differentiating between soil and root systems (Mary *et al.* 2017) but also allow to extract characteristic relaxation times from the imaging results, which may provide information on the length scales of the polarization processes (e.g. Weigand and Kemna 2019).

### Conceptual model of induced polarization response of plant-root systems

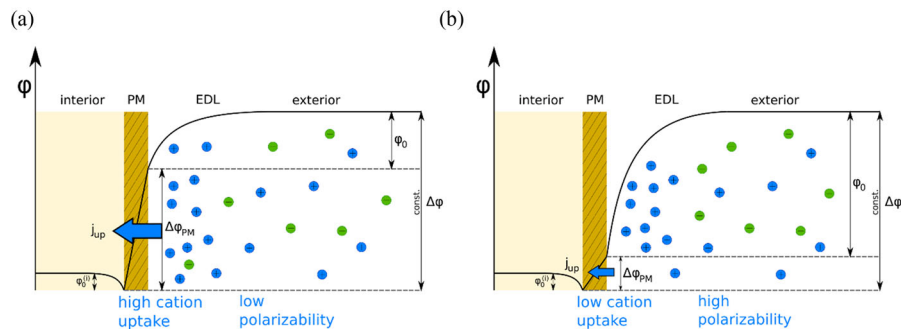
In order to make progress in the study of roots with IP measurements, a ‘phytophysical’ model that connects root electrical signatures to root properties will need to be developed. On the one hand, this will require a deeper experimental understanding of energy storage processes within roots (measured as electrical capacity or polarization) and their relation to root anatomy and physiology. This can best be achieved with dedicated laboratory experiments that aim to link the frequency-dependent electrical properties of roots to root properties (root diameter, surface area, biomass etc.) and root activity (ion transport, membrane potential etc.). On the other hand,

there is a clear need to support the experimental work with a theoretical framework that goes beyond the currently used electrical circuit models and links electrophysiological models of cells to electrochemical models that simulate electrical polarization (e.g. Bücker and Hördt 2013).

Recently, Weigand and Kemna (2019) proposed a first step towards such a theoretical framework in order to explain diurnal variations in IP response associated with variations in nutrient uptake. They proposed a conceptual electrochemical model at the root cell scale that is detailed for the first time below and illustrated in Fig. 7. It integrates established relationships between cell-membrane electrical characteristics and cross-membrane ion transport (Kinraide 2001; Wang *et al.* 2011) with the mechanistic understanding of electrochemical polarization around charged, electrically conductive particles (Wong 1979; Bücker *et al.* 2018, 2019). In particular, Kinraide (2001) proposed a model to describe the ion flux across the cell membrane as a function of the ion activities and electric potentials at the inner and outer surfaces of the membrane. In this model, a Gouy–Chapman–Stern model of the electrical double layer is used to describe the electrical potentials at inner and outer surface of the membrane. Changes in the typically negative surface electric potential on the outside of the membrane ( $\varphi_0$  in Fig. 7) are offset by changes in the transmembrane potential difference that controls the gating of ion channels and ion fluxes across the membrane ( $\Delta\varphi_{PM}$  in Fig. 7). Thus, a decreased negativity of  $\varphi_0$  corresponds with an increased cation flux ( $j_{up}$  in Fig. 7; and decreased anion flux) into the cell, and vice versa (see Fig. 7; Kinraide 2001; Wang *et al.* 2011). A decreased negativity of  $\varphi_0$ , which is directly related to the surface charge density of the membrane (Kinraide and Wang 2010), results in a weaker EDL at the outer membrane surface and thus a decreased polarizability of the EDL in an external electric field (Fig. 7). As such, the proposed polarization mechanism is similar to the electrochemical polarization around metallic particles, where charge transfer reactions at the electrolyte–metal interface cause a weakening of the EDL and consequently a reduced polarizability (Wong 1979; Bücker *et al.* 2018, 2019).

This conceptual model implies an inverse relationship between the magnitude of the IP response of root systems and the strength of cation uptake and is able to qualitatively explain the diurnal variations of the IP magnitude recently observed by Weigand and Kemna (2019). However, it is still unclear how the model can be appropriately scaled up to an entire root system. Also, it is yet unclear whether the internal structure of roots may give rise to effective polarization at scales





**Figure 7** Conceptual model at the root cell scale as proposed by Weigand and Kemna (2019) relating ion uptake fluxes to the strength of electrochemical (EDL) polarizability based on the electric potential distribution across a root cell membrane (Kinraide 2001; Kinraide and Wang 2010; Wang *et al.* 2011). Lower negativity of outer surface potential ( $\phi_0$ ) at the plasma membrane (PM) ((a) left scenario versus (b) right scenario) corresponds with, on the one hand, increased surface-to-surface transmembrane potential difference ( $\Delta\phi_{PM}$ ) and enhanced cation flux ( $j_{up}$ ) into the cell and, on the other hand, decreased surface charge density of the membrane and decreased polarizability of the EDL at the outer surface of the membrane due to an external electric field, similar to the behaviour of electrochemical polarization around metallic particles.

larger than the cell scale. The analysis of the spectral IP imaging results of various experiments by Weigand and Kemna (2019) yielded relaxation time estimates in the microsecond range, suggesting length scales of the underlying polarization processes of several micrometres (Weigand and Kemna 2019). This is in agreement with the proposed cell-based conceptual model, but the experimental basis is not broad enough to draw definite conclusions yet.

### Research directions and applications

Based on the first studies on plant-root systems, the IP method represents an interesting tool to map the root distribution and monitor soil-root interactions. Indeed, the IP method could complement or even partly substitute rhizotube or trenching methods. Given that those established methods also have large associated uncertainties (due to being either point measurements, or manually counted/drawn), IP uncertainties linked with the stacking of multiple polarization mechanisms might not be too disruptive.

In addition, time-lapse IP measurements could be used as an affordable environmental monitoring tool and, for example, indicate when the physiological equilibrium of a plant-root system is reached. This envisioned application is based on a qualitative analysis of the IP data, and thus could be used, even when a full understanding of the underlying (polarization) mechanisms associated with roots is lacking.

In the near future, IP may also be used to support the conceptualization and calibration of physiological models, such as root water uptake models, which can then be easily connected to various kinds of local to global mod-

elling/forecasting frameworks. The observed sensitivity of the IP signal to nutrient uptake is particularly exciting, since it suggests a potential to separate root structure and function using non-invasive measurement.

### WOOD AND TREE CHARACTERIZATION

Besides the description of microbial processes and root systems, induced polarization (IP) can also be used to characterize wood and tree structures. The electrical properties of wood have been studied for almost a century. Indeed, Stamm (1930) used an electrical instrument to determine the moisture content of wood. Shigo and Shigo (1974) developed an electrical instrument to detect discoloration and decay in living trees and creosoted utility poles. In the following years, further instrumental development allowed increasing the range of applications such as tree root detection (e.g. Piirto and Wilcox 1978; Zürcher 1988; Brandt and Rinn 1989). Later, electrical measurements in tree trunks showed that tomographic examinations are suitable for reproducing the internal structure of a living tree, for observing possible fluid transfer, for detecting decay and infections or for separating different wood sections (e.g. Weihs *et al.* 1999; Nicolotti *et al.* 2003; Al Hagrey 2006; Bieker and Rust 2010; Bieker *et al.* 2010; Guyot *et al.* 2013). Schleifer *et al.* (2002) showed that wood is a polarizable material and were able to detect historically important wood structures in the subsurface using the IP method. In addition to information extracted from electrical resistivity, the phase shift is sensitive to additional wood parameters such as wood cell structure, and can thus be used for various applications such as the assessment of fungal infection and thus

tree stability (Martin 2012; Martin and Günther 2013) or the classification of different tree species (Martin *et al.* 2015). Furthermore, phase shift evaluation has the potential to detect the maturation status of trees and therefore estimate the best time for tree felling (Martin *et al.* 2015).

### Factors affecting the induced polarization signal of tree wood

The interpretation of the IP signature in wood and trees is very much affected by the heterogeneity and the strong anisotropy of wooden material. Martin (2012) measured the laboratory IP signature of healthy oak wood samples and observed that both resistivity and phase shift of the wood samples vary with their position within the tree. The lowest resistivities with the highest phase effects are measured for samples extracted from regions where all the fluid transport in the tree trunk occurs.

Besides anisotropy, different tree species also influence the IP signals due to their different wood structure. For example, Martin *et al.* (2015) showed that the polarization of different European tree species (e.g. beech, oak and lime) differs significantly in terms of both chargeability and peak frequency. Tropical wood can be separated and classified from other wood (e.g. European) due to an often-higher polarization effect (up to 140 mrad) and a peak frequency at very low frequencies ( $< 0.01$  Hz). In particular, sandalwood trees can be monitored using IP (Martin *et al.* 2015). These trees react to injuries by producing oil and oil-bearing ingredients within the tree, which affect the IP response: the phase shift increases significantly with oil content. The IP method can thus be used to determine the best time to cut down the tree and collect the highest (very valuable) oil concentration (Martin *et al.* 2015).

Additionally, fungal infection of trees has a measurable impact on the IP signature. Martin (2012) showed that fungal infections destroy polarizable wood cells in the laboratory. With increasing fungal infection, the IP phase signal decreases continuously, whereas the resistivity mainly reflects the wood moisture and dissolved ions. These laboratory findings were verified by tomographic (2D) field applications on tree trunks at different heights (Martin and Günther 2013). The infected parts of an oak tree wood showed a decrease in resistivity (due to the moisture of the fungi) and decrease in phase.

At the field scale, the IP signal not only is sensitive to the presence of fungi but will also vary with the environment. Indeed, a living tree is affected by the seasons, the weather and the tree location. In contrast to 'dead' wood used for laboratory measurements, the influence of the environment must also be considered for field data interpretation of living trees

(Fig. 8). For example, the tree stops the water and nutrient transport in autumn and winter, which affects the electrical properties.

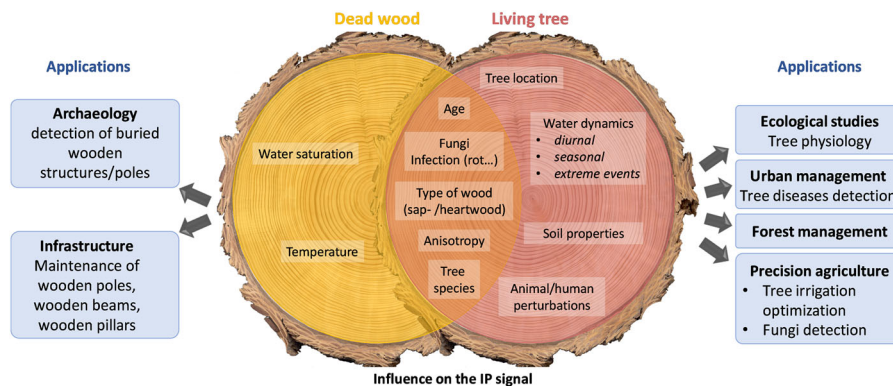
### Research directions

In the last 10 years, many successful and promising experiments have shown that the IP method is sensitive to changes in wood and tree structures. However, for a deeper understanding of the polarization processes and before any commercial application, further research is still needed. Future laboratory experiments should be geared towards understanding the effects of different kinds of fungi under varying environmental conditions and tree species. Tomographic field investigations for different tree species are also required to explain the IP signature of different tree species. In addition to the applications mentioned herein, experiments could be made in the laboratory and the field to test IP's sensitivity to estimate the hydraulic conductivity in wood or trees and to separate the influence of water salinity. The field measurement set-ups also need to be adapted (e.g. electrode material, configuration, instruments etc.) for IP measurement on wood. Finally, a better understanding of the physiological processes involved in the polarization mechanisms must be achieved to be able to better interpret the IP signals collected from this living material. This requires further efforts for developing mechanistic models linking the tree physiological responses to their IP signatures.

The IP method can provide additional information, such as the localization of hidden wooden structures (e.g. Schleifer *et al.* 2002), the condition of wood or the stability status of the tree (e.g. Martin 2012) that are hard to evaluate non-destructively with other methods. Many additional applications of the IP method for tree and wood can thus be envisioned. IP measurements can be particularly interesting for archaeological and forestry applications, as well as for stability studies of road trees and wood poles and/or to understand tree physiology in agricultural and ecological studies (see Fig. 8).

### FROM THE LABORATORY TO THE FIELD SCALE

In the previous sections, it was clearly shown that induced polarization (IP) signals are sensitive to biological changes and processes, at both the microscopic and macroscopic scales. The IP method can indeed be used at various scales, measuring the same parameters, but scaling (up or down) the



**Figure 8** Known causes of the IP signature of dead wood and living trees and application domains for both materials. Causes at the junction of both circles are common to both dead wood and living trees.

observations, is a non-trivial issue that needs further research efforts. In this section, we summarize the state-of-the-art up- and down-scaling methods, the current challenges and point out at future research directions.

At the laboratory scale, it is possible to conduct experiments in a controlled environment, and to consider macroscopically homogeneous samples to individually investigate the different parameters and processes that affect the IP response. In addition, complementary analyses can be used to better understand the associated biogeochemical mechanisms. The combination of IP measurements and complementary physical, chemical and biological analyses can even lead to the development of petrophysical relationships that can be used by biogeochemists or plant/crop scientists as a proxy to monitor their experiments at various scales due to the multi-scale adaptability of the IP method. Laboratory investigations are also strongly needed to interpret field-scale experiments. Thus far, the transferability of laboratory observations to field applications is challenging due to (i) the change in the investigated representative volume; (ii) the higher complexity of the system in the field in terms of heterogeneity of soil properties (water content, soil composition, salinity, pH, temperature) and interactions between different processes; (iii) the change in measuring equipment, methodology and measurement sensitivity.

At the field scale, IP measurements are affected by a variety of influences such as soil properties, water chemistry or environmental changes, resulting in a superposition of different IP signals. For example, Weigand (2017) showed that the IP response from non-woody crop roots is hard to measure due to the small resistivity contrast with the surrounding subsoil and the small volume of root biomass. Changes in the environment can not only mask the microbial, root

or tree signatures, but also change them as they react to their environment. Weigand and Kemna (2019) observed changes in the chargeability of the roots that reacted physiologically to varying sunlight. The interpretation of field IP signals thus requires an understanding of the biogeochemical dynamics.

Furthermore, some biological processes that are observed in the laboratory show IP signatures at very low frequencies, which are hard to measure in the field due to the large amount of time such field surveys would take, compared with the reaction time scale of the monitored biological phenomena. For instance, Martin and Günther (2013) could differentiate fungal-infected wood and healthy wood through their phase variations only below 0.1 Hz. However, full-scale tomography on actual tree trunks was not possible with commercially available equipment at such low frequency because of time constraints. High spatiotemporal resolution and information at a broad frequency range in the field is also important for the characterization of geochemical processes accompanying microbial activity using SIP. For example, extensive monitoring investigations at the Rifle study area (Colorado, USA) were performed to assess the stimulation of iron-reducing bacteria for the immobilization of uranium in groundwater. Initial campaigns performed at a single frequency (1 Hz) demonstrated the applicability of the IP method to assess the precipitation of iron sulphides (Williams *et al.* 2009). The extension to a broader frequency range (0.06–10 Hz) demonstrated the possibility to also gain information about changes in subsurface redox status (Flores-Orozco *et al.* 2011). An even broader frequency range (0.06–256 Hz) allowed Flores-Orozco *et al.* (2013) to determine a characteristic relaxation frequency that provided information regarding the dominant grain size controlling the polarization mechanism. Although other studies

have demonstrated the possibility to perform SIP measurements in a broad frequency range (e.g. Flores-Orozco *et al.* 2012, 2018; Gallistl *et al.* 2018), applications are still rare due to the extensive acquisition times (for measurements at frequencies below 1 Hz), and the contamination of the data due to parasitic electromagnetic effects (specially at frequencies above 50 Hz). This can be improved by adapting existing field measurement set-ups and by developing further FDIP field measurement equipment (e.g. Dahlin and Zhou 2006; Flores-Orozco *et al.* 2013; Kelter *et al.* 2018). Correction methods for capacitive and inductive coupling effects have also been developed for borehole (Zhao *et al.* 2013, 2015), and surface FDIP measurements (Zimmermann *et al.*, 2019), but have not yet been transferred to commercially available measurement equipment.

Another issue for upscaling laboratory results to field applications is that the IP methodology in laboratory and field measurements is not always practically equivalent. TDIP and FDIP measurements may provide different resolution and spectral ranges due to instrumentation and configuration. In laboratory experiments, where time constraints are smaller, mostly SIP measurements are used due to their better spectral resolution and signal-to-noise ratio. To obtain the same high-quality wide-range data as in the laboratory, more effort and time are required for field measurements in the case of SIP measurements compared with TDIP measurements. Therefore, in the field, TDIP measurements may be more convenient and time saving when using multi-core shielded cables and considering short acquisition times (Olsson 2018). However, spectral information is lost using TDIP, especially at early (high frequencies) and late times (lowest frequencies). To improve TDIP field data quality, full waveform acquisition is a powerful tool that will help bridging the gap between SIP laboratory measurements and TDIP field acquisition. A second type of challenges with field applications is associated with the difficulty of acquiring high-quality data as well as the reduced resolution at depth. The IP signature of the targeted biogeochemical process is then often smeared by large lithological contrasts. Using different instruments, electrode material, cables (shielded or not) and so on can also cause different parasitic signals, noise levels, spectral range as well as resolution and must be considered when trying to upscale laboratory results (Dahlin, Leroux and Nissen 2002; Dahlin and Leroux 2012; Zarif, Kessouri and Slater 2017). Due to the significant but often small phase variations in the laboratory IP signature of biological material (Ntarlagiannis *et al.* 2005), it is even more necessary to ensure suitable system performance and thus good-enough

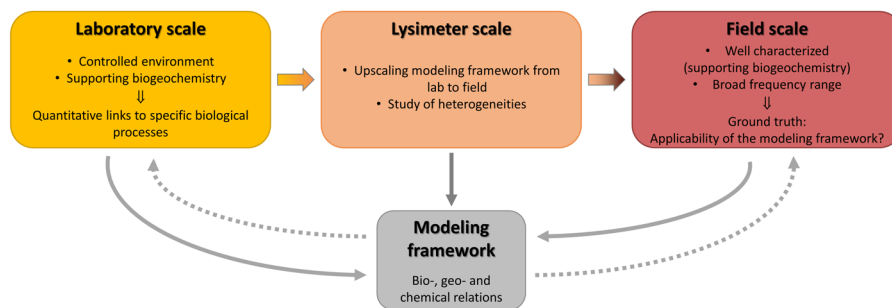
measurement sensitivity to small biological signatures in the field.

Despite the incomplete understanding of IP signal sources and the different measurement techniques, field measurements are very useful, especially to study temporal processes (see above-mentioned time-lapse field monitoring studies by Flores-Orozco *et al.* 2013) or biological processes in a living system such as standing trees that cannot be transferred to the laboratory. Indeed, Martin and Günther (2013) observed different biological behaviour in living trees compared with (dead) wood samples. Moreover, significant progress has been made in recent years to compare laboratory and field-scale IP results linked with biogeochemical processes. For instance, Saneiyan *et al.* (2019) demonstrated, at the field scale, that the imaginary conductivity signal tracks subsurface microbial induced calcite precipitation changes more clearly than just resistivity, which was in good agreement with their laboratory observations (Saneiyan *et al.* 2018).

To further improve our understanding and interpretation of biogeochemical processes at the laboratory and field scale, deduced from IP signal changes, we propose the following bridging framework (see Fig. 9). Laboratory experiments need to be conducted to quantitatively separate the effects of different biogeochemical processes and to build a modelling framework that can be used for direct laboratory applications or upscaling to field observations. Field campaigns also need to be led on well-characterized field sites (access to temporally and spatially dense biogeochemical information), using a broad frequency range. They can then be used to assess the applicability and/or improve the modelling framework. Finally, the development of experiments at an intermediate lysimeter scale can be the answer to (i) upscaling the modelling framework from laboratory observations to uncontrolled field measurements and (ii) studying the effect of heterogeneities on the IP signal.

## JOINT INTERPRETATION

Joint interpretation is the concurrent use of (i) different geophysical data sets or (ii) induced polarization (IP) and non-geophysical data sets to assess the biological, chemical and physical states of the subsurface. Joint interpretation of different geophysical data sets will not be discussed here, as it is not specific to IP nor to biogeophysics. Rather, we focus on the use of IP data to calibrate a mechanistic model that contains the general understanding of a biogeochemical process. As biogeophysics is relatively new, there are very few examples of such joint interpretation in the literature. Hence, this



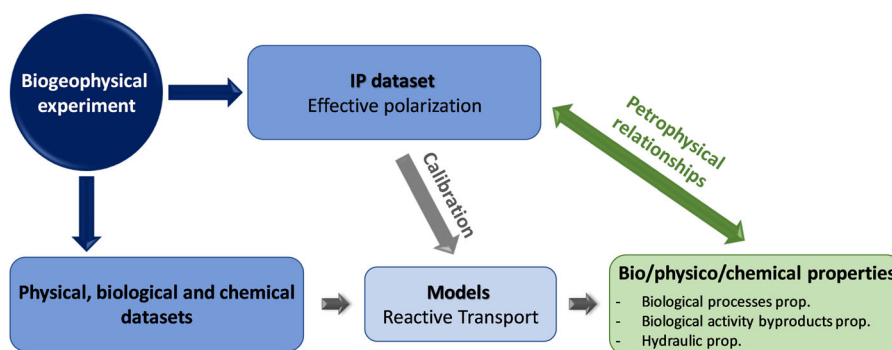
**Figure 9** Conceptual model of the next steps needed in research projects on IP and biogeophysics to be able to use the IP method as a tool to characterize biological processes at various scales from the laboratory to the field scale. Solid arrows represent the primary target for the study: for example, laboratory studies are aiming to build a modelling framework linking the IP signals with the biological processes. Dashed arrows are representing the secondary goals that could be achieved as well; for example, modelling framework could be re-applied in laboratory biogeochemical experimentations where IP is used as a laboratory tool.

section is intended as a discussion of potential directions, rather than a critical review. It must be said upfront that soil systems in general, and soil biogeochemical systems in particular, are inherently complex and characteristically generate many different dynamic signals, natural or induced. This makes the interpretation of biogeophysical signatures more challenging than conventional geophysical signatures. Even in a controlled laboratory environment, it is difficult to isolate a sub-system, and the interpretation almost always needs additional sources of data. Incorporation of additional data is often done in a process modelling framework (Fig. 10).

### Joint interpretation of biological processes

Recently, (bio)geochemically induced changes in IP responses have been coupled to reactive transport models (RTMs), a combination which has resulted in the improvement of IP data interpretation, model formulation and biogeochemical

system understanding (Wu *et al.* 2011, 2014; Mellage *et al.* 2018a, b). In abiotic experiments, Mellage *et al.* 2018a developed a coupled SIP-sensing and transport simulation framework to fit the breakthrough behaviour of iron-oxide nanoparticles in natural aquifer sand. This simulation framework yielded direct quantitative relationships between nanoparticle concentration in the porous medium and measured SIP responses. The nanoparticle concentrations could only be inferred from outflow samples, but could be directly computed within the experimental domain by an RTM. In biotic sand column incubation experiments, Mellage *et al.* (2018b) coupled a diffusion–reaction model to SIP measurements. The integration of geophysically derived parameters (e.g. relaxation time) improved the process-based conceptual understanding of their experiments, in turn improving the numerical model calibration. In similar systems, IP data can be either used to replace conventional breakthrough data, or better, used jointly with breakthrough data to better



**Figure 10** Conceptual joint interpretation scheme. Models such as RTMs use biological, chemical and physical data sets available (low spatial and temporal resolutions) to estimate biological, physical or chemical properties with higher resolutions. Comparison with IP measurements can help develop petrophysical relationships between these two types of data sets. As IP data have better spatial and temporal resolutions than the other data sets used in the RTMs, they can be used to calibrate these models.



understand a system. A joint approach is inherently superior for non-conservative transport, where data regarding accumulation typically are available only at the end of a conventional breakthrough experiment. The integration of RTMs with SIP data sets therefore provides a framework for upscaling the approach to the field, where large-scale modelling approaches could benefit from more spatially and temporally comprehensive data sets. The challenge lies in the integration of methods, such as SIP, with existing modelling frameworks. Biogeophysical data do not relate directly to RTM-generated outputs (e.g. geochemical speciation data, reaction rates and solute concentrations). Therefore, the establishment of relationships between model parameters and method-specific outputs (i.e. conductivity) is required (Li *et al.* 2017). The development of an integration framework would require bench-scale experiments to develop the relationships between the techniques applied and the targeted information we are trying to gather. For example, high-resolution biogeophysical signal changes can be related to specific processes via computed reaction rates. This would allow for the development of system-specific semi-quantitative relationships that can be applied to inform and set-up a larger-scale model at an analogue field site (Fig. 10).

#### Joint interpretation of biological activity by-products

While many challenges exist for direct coupling between SIP signals and RTMs, perhaps a more developed application of IP is its utilization to monitor by-products and end-products of reactive transport processes. Such products include (i) emergence or disappearance of minerals, (ii) changes in pH or ionic strength and speciation or (iii) changes of the physical structure of the system (e.g. permeability) due to dissolution or deformation. Among these studies, the application of IP to monitor mineral precipitation during biogeochemical processes has been a major topic of interest. For example, studies conducted by Williams *et al.* (2005) and Ntarlagiannis *et al.* (2005) investigated the SIP signals associated with microbially induced sulphide precipitation, and Wu *et al.* (2010) studied the SIP signature of calcite precipitation. In relevant studies, Personna *et al.* (2008), Zhang *et al.* (2012), Hubbard *et al.* (2004) and Placencia-Gómez *et al.* (2013) studied processes relevant to mineral surface dynamics, including mineral transformation, surface absorption/desorption, as well as mineral transformation during redox reactions. While such studies have collectively demonstrated the sensitivity of SIP signals to the by-products of biogeochemical processes, the way SIP signals can be better related to reactive processes in the mineral-water system is still poorly explored. Wu *et al.* (2011)

developed a joint interpretation concept, relating between SIP signals that originate with calcite deposition, and a reactive transport model. Yet this is only a conceptual approach that needs further improvements in the context of calcite and also requires generalization.

#### Remaining challenges

The potential of SIP signals to help understand fundamental biogeochemical processes has been adequately demonstrated in the laboratory. However, IP data sets are difficult to self-interpret as they result from the addition of different IP signatures (e.g. the soil–water system, the biological components and interaction with the soil system). A modelling framework that looks at the mechanics of the studied process (remediation, precipitation etc.) and is calibrated using both IP and non-geophysical data is a promising way to integrate such data sets at various scales from laboratory to field studies (Fig. 10).

Because the geophysical field and the biogeochemical field communities are not traditionally overlapping, communication and interactions are critical to better understand the strength and needs of each community and facilitate the joint development of biogeophysics. For example, whether IP signals contain rates and speciation information that can help to better validate biogeochemical process modelling could be a question of interest to both communities. Outputs from biogeochemical process modelling may help constraining IP data interpretation, or can directly feed into SIP modelling. This can help better understand the underlying mechanisms driving IP signals. As suggested above with respect to the work of Mella *et al.* (2018a, b) or Wu *et al.* (2011), using the IP data to calibrate a mechanistic model not only makes use of signals that are more directly related to the investigated process, but also provides *in situ* and time-continuous data that are normally hard to collect without sacrificial experiments.

#### SUMMARY AND CONCLUSIONS

Over the past decade, the induced polarization (IP) method has been used to monitor a variety of microbially mediated environmental processes and tested to directly characterize microorganisms and biofilms. In addition, IP was found to be sensitive to plant systems, both in underground root systems and in aboveground stems.

Although the qualitative use of IP in biogeophysical studies is now established, several key points need to be researched in the near future to bring the IP method to its full potential. In the next 10 years, we envision research to focus on

gaining a better understanding of the polarization mechanisms associated with the above IP signatures, and synthesize those findings into modelling frameworks for practical use. Particularly, more experimental and modelling work is necessary to understand the polarization mechanisms of single bacterial cells and even more complex biofilms. In order for laboratory experiments to be significant and help build comprehensive mechanistic models, more efforts need to be put in evaluating measurement uncertainty. Indeed, many experimental observations today are made on set-ups that are not replicated. It is thus hard to use such results as a basis for model development, as well as for convincing neighbouring disciplines of the benefits of using the IP method. We believe that future laboratory experiments will systematically need to be replicated (measurement of at least triplicates) to better evaluate uncertainties in IP measurements. This applies to repetitions of measurements on the same set-up as well as repetitions of the entire preparation protocol.

Recent modelling efforts suggest that bacterial cells are polarizable because of the presence of a polymeric brush layer that allows the development of an electrical double layer (EDL) around the bacterial cells. The polarization of biofilms is attributed to (i) bacterial cells with EDL, (ii) extracellular polymeric substances (EPS) that may behave like a polymer electrolyte and (iii) noncellular materials such as clay grains that increase the surface conduction. Those explanations remain highly speculative, and careful experimental studies are needed to verify the models. Regarding root structures, the polarization mechanisms are even more complex because the plant-root systems are composed of multiple cell layers. The observations thus far have been explained by the presence of polarizable root cells inside the roots, with the development of an EDL outside their plasma membrane and cross-membrane ion transport. More work is needed to understand how to scale-up these findings at a root system level and whether the interactions between the cells are playing an important role in the IP signal. The role of the porous medium should also be better considered when investigating both microbe-related and plant-related processes.

A major drawback is that IP is sensitive to multiple biological, physical and chemical processes in complex environments. Thus, a key challenge is the non-unique interpretation of the IP signal, due to detection of superimposed signatures of processes influencing the effective polarization. This can be overcome by several strategies that need to be tested in combination. Ideally, IP measurements should be coupled with direct biogeochemical measurements and interpreted jointly in mechanistic approaches. Additionally, for plant-root systems,

indirect current injection could be combined with stem-based excitation methods to distinguish root polarization from the other soil polarization sources. Another possible strategy is to use multi-frequency (in frequency-domain IP) or full waveform (in time-domain IP) measurements to help differentiate the processes that have different relaxation frequencies. Besides, as biological processes are much more dynamic than other processes (e.g. soil textural changes or groundwater-level variations), time-lapse measurements could help to distinguish between different contributions. Lastly, the development of experiments at an intermediate scale (lysimeter scale) is needed to study the IP sensitivity to heterogeneities in a controlled environment.

Apart from the direct development of the IP method to biogeophysical studies in the future, both in the laboratory and in the field, we believe IP could be very useful if integrated in the experimental or modelling protocol of neighbouring disciplines. IP could especially bridge the methodological gap in geomicrobiological studies, as it is non-destructive and possesses a good spatial and temporal resolution. It could also be used in studies of extracellular electron transfer to understand the fundamental mechanisms. Besides, IP could become a tool for plant physiologists to study plant-root systems at various scales from the laboratory to the field, including the lysimeter scale. In addition, IP can be used to monitor tree stem activity in fully grown trees, which could not be studied at smaller scales or by using destructive methods. To understand the methodological needs of these different neighbouring research communities, it is crucial to encourage communication and interactions between the different scientific communities through research collaborations and joint symposiums and conferences. We believe that only such close partnerships will reveal the full potential of the IP method related to the biosphere.

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