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The Fuel Cell Model of Abiogenesis: A New Approach to Origin-of-Life Simulations

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Running title: The Fuel Cell Model of Abiogenesis

Abstract:

In this paper we discuss how prebiotic geo-electrochemical systems can be modeled as a fuel cell and how laboratory simulations of the origin of life in general can benefit from this systems-led approach. As a specific example, we detail the components of what we have termed the “prebiotic fuel cell” (PFC) operating at a putative Hadean hydrothermal vent, and we present preliminary results utilizing electrochemical analysis techniques and proton exchange membrane (PEM) fuel cell components to test the properties of this and other geo-electrochemical systems. The modular nature of fuel cells makes them ideal for creating geo-electrochemical reactors to simulate hydrothermal systems on wet rocky planets and characterize the energetic properties of the seafloor / hydrothermal interface. That electrochemical techniques should be applied to simulating the origin of life follows from the recognition of the fuel cell-like properties of prebiotic chemical systems and the earliest metabolisms. Conducting this type of laboratory simulation of the emergence of bioenergetics will not only be informative in the context of the origin of life on Earth, but may help us understand whether it is possible for life to have emerged in similar environments on other worlds.

1. Introduction:

Identifying the specific processes responsible for the emergence of life on Earth and then convincingly simulating these same processes in the laboratory are amongst the most intriguing challenges for contemporary biogeochemists. A large part of the difficulty is that plausible scenarios for the origin of life must not only account for the fundamental properties of life today but also for the geochemical conditions on the early Earth through which life arose; conditions of which we may have only patchy knowledge and which raise thorny engineering problems during attempts at simulation.

A primary process common to all extant life is the conversion of a pH gradient into energy-storing phosphate anhydride bonds via chemiosmosis across a semi-permeable membrane. In prokaryotes, as well as in mitochondria, the "power plant" of eukaryotes, electric potentials across the membrane that induce a pH gradient between the membrane interior and exterior (i.e. the proton motive force) drive the fundamental energy generation mechanism for life (Mitchell 1961; Jagendorf and Uribe, 1966; Harold, 1986). As in life today, the potential energy contained within these electron and proton gradients is transduced into useful chemical energy and stored in meta-stable energy currency molecules such as adenosine triphosphate (ATP) or inorganic pyrophosphate (PP_i). It has been argued that the very first microorganisms were autotrophic: that is, they gained and assembled their building constituents from the simple molecules available to them on the early Earth (e.g. H_2, CO_2, CH_4, NH_3, HPO_4^{2-} and HS^- / H_2S) (Fuchs 1989, 2011; Berg et al., 2010; Say and Fuchs, 2010). The electron transfer proteins involved in the generation of the proton motive force – many of which host inorganic metal sulfide-containing active centers – have been suggested to be amongst the most ancient catalysts (Eck and Dayhoff, 1966; Baymann et al. 2003; Schoepp-Cothenet et al., 2013; Volbeda and Fontecilla-Camps, 2006; Vignais and Billoud, 2007; Lill and Siegbahn, 2009; McGlynn et al., 2009; Nitschke et al., 2013).

Microbes are, therefore, essentially performing the same chemical redox processes as those in ion-exchange membrane fuel cells (Mitchell, 1977; Barbir, 2005), and, like fuel cells, biology uses proton and ion gradients to generate energy. In fact, recognition of this similarity within the fuel cell community has seen microbes, their redox-active enzymes, and even mitochondria themselves, being successfully used as components of electrodes for biofuel cells due to their excellent catalytic ability to transfer electrons and promote environmentally significant redox reactions (Arnold and Rechnitz, 1980; Heller, 1992; Chang et al., 2006; Arechederra and Minteer, 2008; Tran and Barber, 2012; Huang et al., 2012). Certain geochemical environments also constitute fuel-cell-like systems, for example, at hydrothermal vents where electrical and pH potentials are generated at the interface between reduced hydrothermal fluid and oxidizing seawater (Yamamoto et al. 2013; Russell and Hall 1997, 2006; Baross and Hoffman 1985; Martin and Russell 2007). In the hydrothermal vent example, the ambient geochemical potentials can be maintained and mediated across physical boundaries, e.g. electrically conductive biofilms, sediments or an inorganic chimney precipitate (Nakamura et al., 2010a, b; Yamamoto et al., 2013; El-Naggar et al. 2010; Ludwig et al. 2006). A second geological example of fuel-cell-like behavior believed to be of significance within early Earth environments is Galvanic corrosion of iron-nickel alloys (associated with meteoritic in-fall) from contact with seawater, which can in principle establish pH / E_h gradients (Bryant et al. 2013).

Since life universally depends on pH / E_h gradients to drive metabolism, geological environments that already generate similar electrochemical energy gradients are of interest for origin of life studies. One such example is hydrothermal vents, which can be produced by magmatic activity (e.g. black smokers) or water-rock chemistry (e.g. serpentinite-hosted alkaline vents). Alkaline hydrothermal vents produced by serpentinization (the process of olivine / pyroxene oxidation via seawater interaction in a series of exothermic reactions) are of special interest to the origin of life, since they can generate a very alkaline hydrothermal effluent and thus an ambient pH gradient as well as electrical potential at the fluid interface (Russell et al. 2010; Russell and Hall 2006; Lane and Martin 2012; Baross and Hoffman 1985; Martin et al. 2008). Serpentinization is likely to have progressed on the early Earth (Russell et al. 1989; Sleep et al. 2011; Arndt and Nisbet 2013), and may have also been active elsewhere in the Solar System, e.g. on early Mars or Europa, or on other icy moons with a water-rock interface (Ehlmann et al. 2010; Wray and Ehlmann 2011; Vance et al. 2007; Michalski et al. 2013) – possibly providing an environment for emergent prebiotic chemistry on other worlds.

The operations of extant life are analogous to those of a fuel cell, and some version of the fundamental components that make the biological fuel cell function (ATP-synthase; ion-selective membranes maintaining pH / electrical gradients; the electron transport chain) were also likely present in LUCA, the last universal common ancestor (Gogarten et al., 1989; Martin and Russell, 2007; Mulkidjanian et al. 2007; Schoepp-Cothenet et al., 2013). Because we consider that LUCA – the last unified biochemical system arising from geochemistry – also had these basic fuel cell properties (Russell and Hall 1997; Nitschke and Russell 2009; cf. Huang et al., 2012, Lane and Martin, 2012), we suggest that some key steps at the origin of metabolism may then be simulated experimentally within the context of the electrochemical gradient architecture of a fuel cell. As life is an example of a far-from-equilibrium biological system, which presumably emerged from a far-from-equilibrium geological and geochemical system, it seems logical that a far-from-equilibrium chemical system such as a fuel cell may provide an effective link between the two.

We set out to examine whether the fuel-cell-like properties of certain geochemical environments, such as seafloor interfaces and hydrothermal vent systems, could be simulated in out-of-equilibrium electrochemical experiments and ultimately modeled in the laboratory as a fuel cell. In a geological / geochemical fuel cell scenario, some mineral or other inorganic component would be required to act as electrocatalyst as well as ion transfer ‘membrane’ separating contrasting chemical reservoirs. Depending on the scenario this component could be conductive minerals, gels, or other porous material. For our preliminary experiments we chose to focus on (i) Iron sulfides and (ii) Fe-Ni phases within iron meteorites. Both of these are viewed as being potential electron transfer catalysts considered relevant for proto-metabolic reactions and are considered to have been readily available and accessible on the early Earth (Williams 1961, 1965**;** Eck and Dayhoff, 1966**;** Russell et al., 1994; Nitschke and Russell 2011; Bryant et al. 2009, 2013). Metal sulfides in particular constitute an important part of the environmental fuel cell in modern hydrothermal vent systems as they facilitate transfer of electrons and catalyze redox reactions (Nakamura et al. 2010b). The nature of mineral precipitation in a gradient is also significant, as the chemical disequilibrium can result in complex precipitate structures such as hydrothermal “chemical garden” chimneys (Ludwig et al. 2006; Haymon et al. 1983; Russell et al. 1989, 1994) that may exhibit electrochemical and ion-transfer capabilities (Ayalon 1984; van Oss 1984). Thus, we synthesized iron sulfide electrocatalyst material by interfacing contrasting acidic, Fe^{+2}-containing solution and alkaline, sulfide-containing solution across a permeable barrier to form a self-assembling precipitate membrane in a gradient. This allowed for in situ experiments where the simulated geological precipitate actually separated two contrasting solutions and could mediate gradients, as might occur in a natural far-of-equilibrium system such as chimney growth at a hydrothermal vent. We also conducted electrochemical experiments using geological analog materials (both synthetic and field samples) as catalysts, and constructed a PEM fuel cell apparatus to simulate an electrochemically active geochemical interface. The fuel cell represents a holistic chemical system which is well understood, amenable to computational modeling, and open to sophisticated analytical diagnostics. It is a systems approach that has not yet been exploited experimentally within the sphere of abiogenesis yet offers a potentially powerful theoretical and experimental model through which to explore such emergent physicochemical systems. These experimental techniques could be applied to a variety of geochemical systems or analog materials, and could even be used to simulate energetic processes at seafloor interfaces on other planets as well, to determine whether prebiotic chemistry would be possible on other wet, rocky worlds.

2. Examples of Fuel Cell-like Systems in Biology and Geology

In order to envision experimental ways to test prebiotic chemistry within an electrochemical framework, it is useful to define and constrain the functional parts of the natural fuel cells of geology and biology, as well as a proposed prebiotic fuel cell (PFC) that may have powered inorganic and organic redox reactions on the early Earth.

Fuel cells are primarily composed of an anode (the surface at which oxidation occurs), a cathode (the surface at which reduction occurs), and an ion-conducting electrolyte that physically separates reservoirs containing the fuel and oxidant (Surampudi et al., 1994; Haile 2003a,b). The physical set up of the fuel cell in which two chemically distinct reservoirs are separated is essential, for it is this chemical / redox disequilibrium – chemical gradients maintained over some distance – from which electricity is ultimately produced. The essential components include: The **electrolyte,** which physically separates the fuels and oxidants and allows a voltage gradient to be maintained; the **electrodes,** which facilitate oxidation / reduction and contain catalytic materials; and the **fuel** (electron donor) and **oxidant** (electron acceptor), which transfer electrons to and from the electrodes.

Mitochondria, the free energy factories of animal cells, are very efficient electrochemically and they and their associated enzymes have been successfully used as electrodes in manmade fuel cells (e.g. Arechederra and Minteer, 2008). As in a PEM fuel cell, the flow of protons and electrons are spatially separated in mitochondria and life has evolved precise chemical means of harnessing the energy present in these potential gradients. The lipid bilayer membrane is the capacitor separating contrasting chemical environments, and the electrodes of mitochondria are the ends of a series of electrochemically connected enzyme complexes within the inner membrane, together known as the electron transport chain. The iron-nickel-sulfide-containing centers of the enzymes act as anodes and cathodes (Berg and Holm, 1982; Baymann et al. 2003; Vignais and Billoud 2007); meanwhile, the membranes – capable as they are of selective ion transport – maintain chemical disequilibrium (Lane and Martin 2012). Derived from proteobacteria, the mitochondria bear certain similarities to chemiosmotic LUCA (Yang et al., 1985; Martin and Müller, 1998; de Paula et al. 2013). LUCA's biological battery would have had some similar functional parts, such as the Complex I (referred to as the 'steam engine of the cell' by Efremov and Sazanov (2011)) that, through redox disequilibria, drives protons to the periplasm. These protons, now forming a pH gradient due to the disequilibrium between membrane exterior and interior, drive ATP synthase – the rotary biomolecular motor – to make ATP (Yoshida et al. 2001, Branscomb and Russell, 2013).

A geological fuel cell phenomenon can also emerge (without biological mediation) at seafloor interfaces, for example, in hydrothermal vents produced either by magmatic activity or by water-rock chemistry. High-temperature hydrothermal alteration on oceanic spreading ridges can produce a hot (>350°C) acidic hydrothermal fluid driven by magmatic intrusion that fuels black smokers, named after the black particles that form as the rapid temperature / pH change induces precipitation of metal sulfides at the fluid interface. In the absence of magmatic heating, reducing hydrothermal fluids can also be produced by serpentinization, a process in which seawater permeates through fractures in the Fe/Mg-silicate ocean crust and produces an alkaline, H_2- and CH_4-enriched, moderately hot (~100-200°C) hydrothermal fluid (Russell et al., 1989; Kelley et al., 2001, 2005; Bradley and Summons 2010; Klein et al., 2013; Charlou et al. 2010) and even in low temperature exhalations (Neal and Stanger, 1983; Coveney et al. 1987; Etiope and Sherwood-Lollar, 2013; Etiope et al. 2013). Both black smoker and alkaline vents can produce chimney precipitates where hydrothermal fluids feed back into the ocean, and these naturally occurring mineral precipitates are significant in that they provide a semi-permeable and semi-conducting barrier between the two contrasting fluids. The redox-active fluid interface of a hydrothermal vent, with fuel / oxidant reservoirs separated by an inorganic mineral precipitate, constitutes a naturally occurring fuel cell that does not depend on biological processes to function. The inside and outside surfaces of the hydrothermal chimney could be considered as anode and cathode, respectively, and, depending on composition / environmental conditions, the chimney itself may be able to catalyze the oxidation of hydrothermal fuels and conduct resulting electrons to seawater oxidants. The fuel-cell-like properties of hydrothermal vents have been directly demonstrated in field studies of black smoker vents, both in situ by harnessing the electrical current produced by the vent to power a small device on the seafloor (Yamamoto et al., 2013), as well as in the laboratory where it has been demonstrated that black smoker chimney material is electrically conductive and capable of catalyzing redox reactions (Nakamura et al., 2010b).

It is also perhaps pertinent to the PFC model that, as pure metals and metal alloys are frequently found to be highly efficient electrocatalysts (Chen et al 2011), such materials were probably geologically present within the Hadean period through the reducing power of serpentinizing vents (e.g. awaruite, Ni_3Fe; Ulrich 1890; Frost 1985; Klein and Bach 2009) and as Fe-Ni meteoritic deposits during the late heavy bombardment (Buchwald, 1977; Cockell, 2006). The corrosion of metals and metal alloys by an electrolyte such as seawater can give rise to a localized fuel-cell-like system: anodic oxidation of Fe to Fe^{2+} with concomitant cathodic reduction of, for example, SO_4^{2-} to elemental S or S^{2-}; H^+ to H_2 or NO_3^- to NO_2^- (Bryant et al 2013) - each of which could, in principle, establish localized pH / E_h gradients.

3. Creating A Fuel Cell Model of A Prebiotic Hydrogeological System

One could consider the energetic ‘essence’ of extant anaerobic life to be quite similar to a fuel cell, in that ion gradients and electron potentials maintained across a membrane produce energy so long as fuels and oxidants are supplied. The sediment / water or hydrothermal / ocean interfaces in modern marine systems also can constitute naturally occurring fuel cells, deriving from the electrochemical cell of the Earth and its atmosphere and mediated by microbes acting as redox catalysts. This has led to the proposal that ion / electron potentials were also the driving forces for life’s origin, and that these gradients were initially provided by the naturally occurring pH / E_h potentials in alkaline hydrothermal vents (Russell and Hall, 1997; Russell et al., 2010). This alkaline hydrothermal origin-of-life model can be generalized to any wet rocky world with a chemically similar water-rock interface (Russell et al. 2014, in review), and so, we chose to develop our electrochemical / fuel cell model using alkaline vents as one example of a hydrogeological origin-of-life scenario that could benefit from this concept (similar fuel cell models could also be applied to simulating electrochemically active water-rock interfaces on early Mars, early Venus, or the icy moons of Jupiter and Saturn).

Earth's oceans four billion years ago are thought to have been anoxic and mildly acidic (pH ~ 5-6 due to dissolved atmospheric CO_2, NO and ephemeral SO_2), rich in Fe^{2+}, and also could have contained Ni^{2+}, Mn^{2+}, phosphates, nitrate, nitrite and some ferric and manganese ions (Macleod et al. 1994; Russell and Hall 1997; Hagan et al., 2007; Martin et al. 2007; Ducluzeau et al., 2009; Mloszewska et al., 2012; Nitschke et al., 2013). The hydrothermal fluid produced by serpentinization would likely have been similar to modern-day hydrothermal fluids: alkaline, containing some silicate, trace Mo and W, and dissolved H_2 and CH_4 along with formate and a range of hydrocarbons (Proskurowski et al. 2008; Mielke et al. 2010; Lang et al. 2010, 2012; Konn et al. 2009; Charlou et al. 2010). It is also thought that ancient hydrothermal springs in serpentinizing systems would have contained millimolar sulfide from dissolution of crustal sulfide minerals (as determined from laboratory reactor experiments simulating serpentinization in a Hadean system; Mielke et al. 2010, 2011). Vents driven by serpentinization produce relatively low-temperature fluids compared to the scalding environment of black smokers, but between them, hydrothermal vent systems on the early Earth could have contributed many of the fuels and materials that are thought to be relevant for an autogenic/autotrophic emergence of life. The interfacing of alkaline hydrothermal fluids and seawater on the early Earth might have produced a variety of inorganic precipitates within the ambient geochemical / electrochemical gradients, among them, silica gel, ferrous/ferric oxyhydroxides, and transition metal sulfides (Russell and Hall, 1997). Hydrothermal precipitates formed by serpentinization on the early Earth might therefore have had compositions (and electrochemical properties) in some ways similar to modern black smoker chimneys, for example, conductive iron sulfide minerals that could catalyze redox reactions. Thus, electrochemical studies of modern black smokers (e.g. Yamamoto et al. 2013; Nakamura et al. 2010b) may also be relevant for understanding the energetics of hydrothermal vent fuel cells on the early Earth.

A putative Hadean hydrothermal vent with a chimney structure would have operated somewhat like a flow-through fuel cell. The contrasting reservoirs of reduced, H_2 / CH_4-containing hydrothermal fluid and the relatively oxidized (containing minor concentrations of Fe^{III} and dissolved NO), CO_2-rich ocean would have been separated by a precipitate of mixed composition, containing material such as carbonates, silicates, and iron and other transition metal sulfides and mixed valence layered iron 'double' oxyhydroxides **(**Russell et al. 2014 in review**; Figure 1**). Transition metal sulfides are but one possible component of a prebiotic hydrothermal chimney, but as one of the more electrochemically active materials likely to be present, it is useful to characterize iron sulfide chimney growth and electrochemistry in the laboratory, which various studies have pursued (Russell et al., 1989; Mielke et al. 2010, 2011; Barge et al. 2014; McGlynn et al. 2012). Many properties of iron sulfide precipitates relevant to a far-from-equilibrium geological system have been experimentally determined. For example, when initially precipitated between contrasting solutions, self-assembling iron sulfide membranes are capable of generating an electrical potential of ~0.6 to 0.7 V, and of maintaining the pH gradient between the hydrothermal and ocean solutions (Filtness et al., 2003; Russell and Hall, 2006; Barge et al. 2014). The precipitates produced in out-of-equilibrium experiments have a complex structure at the micro scale; membranes vary in thickness from ~5 to ~100 μm, exhibit and comprise a mix of iron sulfides, iron oxyhydroxides / silicates, and can also incorporate dissolved ions such as other transition metals and phosphates (Mielke et al. 2011; McGlynn et al. 2012; Barge et al. 2012; Barge et al. 2014; Barge and Kanik unpublished data).

In a hydrothermal system with a chimney structure, the precipitate could facilitate diffusion of ions and electron conduction (Nakamura et al 2010b); thus it could function as a selectively permeable electrolyte membrane between hydrothermal fuels and seawater oxidants. In a fuel cell concept of prebiotic alkaline hydrothermal vents, the anode and cathode would be the interior and exterior chimney mineral surfaces, respectively. The exterior and interior of a chimney could have different mineral compositions, since chemical garden-like structures tend to exhibit compositional variations across the membrane reflecting the chemical gradients in which they precipitate (Russell and Hall 2006; Pagano et al. 2006; Cartwright et al. 2011; Barge et al. 2012). Though a hydrothermal chimney would likely also contain components such as silica gel or carbonate, the ability of the “electrodes” to drive redox reactions would probably be dependent on the presence of electrocatalytic minerals such as metal sulfides and oxyhydroxides (cf. Arrhenius 2003; Antony et al. 2008). Minerals such as mackinawite (FeS) and greigite (Fe_3S_4) are electrically conductive and can also behave as capacitors, and are thought to be able to function as catalysts for organic reactions (Rickard et al. 2001; Huber and Wächtershäuser 1997). These iron sulfide minerals are also structurally similar to the Fe_4S_4 and FeS groups in metalloenzymes which catalyze redox reactions in biology, a similarity that has led to the suggestion that these metalloenzyme centers originally derived from inorganic hydrothermal minerals (Russell and Hall 1997, 2006; Rothery et al. 2008; Nitschke et al. 2013). In a putative prebiotic hydrothermal fuel cell, transport of electrons from fuel to oxidant could be mediated by a series of smaller redox steps, as in biological systems. Within this scenario, the fuels and oxidants would be indefinitely replenished as long as serpentinization remained active, which in modern alkaline hydrothermal systems is envisaged to last for over a hundred thousand years (Ludwig et al., 2011). The chimney membranes themselves could be continuously renewed as the hydrothermal fluids produced by serpentinization continue to rise and interface with the ocean, feeding through and possibly disaggregating older membranes.

4. Experimental Studies: Exploiting Electrochemical Techniques and Fuel Cells as Planetary Geology Test-Beds

The large body of fuel cell work using enzymes, mitochondria, or microbes as electrocatalysts is testament to the importance of charge and concentration gradients in biology. Since both hydrogeological environments (e.g. vents) and biological cells (at least, in their free energy generation mechanisms) can be conceptualized as fuel cells, it makes sense to pursue hydrothermal origin-of-life experiments in a setup that preserves these fundamental aspects of pH and electrochemical gradients across a membrane. To this end, we have conducted some preliminary experiments using electrochemical techniques to simulate hydrogeological systems with electrochemical gradients using geological materials as electrode catalysts. We illustrate how these methods can be utilized to simulate geochemical free energy interfaces and test hypotheses regarding the transition to bioenergetics. It is our intention to broaden these initial studies that simulate seafloor / hydrothermal interfaces to explore the simulation of other hydrogeological environments, such as Galvanic corrosion of iron meteorites, or water / rock interfaces on other planets, within the framework of a fuel cell reactor. We envisage the results presented here to be preliminary indications of possible value in the model we propose.

4.1. Electrochemical Studies of Simulated Hydrothermal Precipitates

Though an ancient hydrothermal chimney would have had a heterogeneous composition, some of the most electrochemically relevant minerals thought to be formed in this system are iron and iron/nickel sulfides. To investigate the electrochemical properties of metal sulfide minerals that could be formed at alkaline vents on the early Earth, we have precipitated inorganic membranes in out-of-equilibrium solution interface experiments (**Figure 2**; Filtness et al. 2003; Barge et al. 2014). We interfaced contrasting solutions representing the dissolved Fe^{2+} in the acidic primordial ocean (added as dissolved $FeCl_2•4H_2O$) and dissolved sulfide in the alkaline hydrothermal fluid released from crustal sulfide minerals (added as dissolved Na_2S). When interfaced, these solutions formed self-assembling mineral precipitates in the same manner as the injection chemical garden experiments that have previously been used to simulate Hadean hydrothermal chimney precipitates (Mielke et al. 2011; Barge et al. 2012, 2014). However we have found that a glass fuel cell setup yields a more structurally stable membrane precipitate that can be used in subsequent electrochemical experiments (Barge et al., 2013; 2014). We have previously formed iron sulfide membranes (and chimneys) using various Fe^{2+} and sulfide concentrations, and have found that although membranes can be formed using geologically realistic mM concentrations over 3 to 4 days, more robust membranes form over shorter timescales using higher (~50 mM) concentrations and these still preserve the out-of-equilibrium nature of the precipitates. Therefore, we have used higher reactant concentrations in this study to form precipitates on shorter timescales reasonable for laboratory experimentation and to generate more precipitate material for use as an electrocatalyst. We recognize that in a natural system, similar precipitates might have taken a much longer time to form from the dilute Fe^{2+}- and sulfide-containing fluids.

Formation of membranes simulating hydrothermal chimney walls: A synthetic porous membrane template was clamped between two fluid reservoirs in a two-chamber glass membrane fuel cell apparatus (Adams & Chittenden glassware, with high-temperature clamp stable up to 150°C). Most experiments were performed using dialysis tubing (Fisher, 3500 MCW) as the precipitation template, but conductive carbon cloth (Sigracet, hydrophilic, no microporous layer) as a template material was also tested. The iron and sulfide solutions were added to the half-cells respectively, and the cell was kept anoxic throughout with constant N_2 purging and allowed to react for 24 hours at room temperature. High-resolution imaging and chemical analysis were carried out on the iron sulfide membranes using an environmental scanning electron microscope (ESEM) with an attached energy-dispersive X-ray tube (EDX). The electrical potential generated across the inorganic membrane as it precipitated was recorded over the course of experiments using an Agilent LXI Data Acquisition / Data Logger Switch Unit with electrodes placed in each cell. pH measurements were recorded with an Excel XL20 pH / conductivity meter (Fisher Scientific) with temperature calibration. The membranes formed were used in subsequent electrochemical experiments, described below.

Membrane potential tests: We conducted membrane potential tests in which an iron sulfide membrane was allowed to precipitate for 24 hours, and then the iron and sulfide solutions were removed. The membrane and both half-cells were carefully rinsed with ddH_2O, then NaCl or KCl salt solutions of varying concentration were added to both sides of the precipitated membrane. (Salt concentrations were 10x greater on the "formerly alkaline" side than on the "formerly acidic" side, and concentrations between 10 mM and 1 M were tested for both NaCl and KCl.) Electrodes were placed several millimeters from either side of the membrane surface to record the membrane potential. In previous experiments this technique has been shown to generate concentration-dependent membrane potentials across inorganic precipitate membranes in many different chemical systems, including some sulfides (e.g. of Hg, Co, Ni, Sn) though Fe-sulfide measurements are not reported (Ayalon 1984; Beg et al. 1977, 1978, 1979; Beg and Matin 2002; Malik et al. 1980; Malik and Siddiqi 1963; Kushwaha et al. 2001; Siddiqi and Alvi 1989; Sakashito and Sato 1977).

Membrane galvanostatic tests: We conducted galvanostatic tests in which electrical current was applied across an iron sulfide membrane after it had formed (Honig and Hengst 1970). The half-cells were filled with $FeCl_2$-containing acidic solution and Na_2S-containing alkaline solution and a dialysis membrane was clamped between the two reservoirs. Two electrodes were placed in each cell: one in the bulk solution to apply current, and one very close to the membrane to measure membrane potential. After the membrane had precipitated for 24 hours, a current of +/- 0.75 mA/cm^2 was applied across the membrane and the resulting membrane potential (relative to the rest potential) was measured. The measurement was also repeated after both cells had been emptied and refilled with 0.1 M NaCl solution.

Voltammetry studies: To test the ability of iron sulfide chimney precipitates to facilitate electron transfer from hydrothermal reductants to seawater oxidants in a prebiotic hydrothermal system, we applied simulated hydrothermal iron sulfide-containing precipitate membrane material as a catalyst on a glassy carbon electrode (GCE). Cyclic voltammetry half-cell experiments were conducted in which the GCE+catalyst was characterized in a Hadean ocean simulant containing oxidants of interest to the origin of life in serpentinizing systems. In this experiment, a Fe/S precipitate membrane was formed in the fuel cell apparatus shown in **Figure 2**, and after 24 hours the membrane was removed, rinsed with ddH_2O, and dried under N_2. The dried precipitate was removed from the dialysis membrane template, attached to a GCE with conductive carbon tape, and potential from +/-1 V was applied (relative to Ag/AgCl, using a Pt counter electrode) in a solution representing bicarbonate as an electron acceptor in the Hadean ocean (20 mM $NaHCO_3$ + 0.6 M NaCl, titrated to pH ~5). A control was performed in which there was no FeS catalyst applied to the working electrode, but instead ground FeS particles (Sigma) were stirred into the simulated ocean while potential was applied (as well as controls where there was no FeS in the system). Experiments were also performed in which carbon cloth was used as the membrane separator in the two-cell precipitation apparatus described above, so that Fe/S membranes were precipitated directly onto the carbon cloth and pieces of the Fe/S-covered cloth (5 mm x 50 mm) could be used directly as working electrodes in voltammetry studies. Controls were also performed using plain carbon cloth (with no Fe/S) as working electrode.

4.2. Fuel Cell Reactors Simulating Prebiotic Geo-Electrochemical Systems

Fuel Cell Design Rationale: An origin-of-life fuel cell experiment testing a hydrogeological system on the early Earth, or any wet rocky planet, might involve a combination of techniques: synthesizing simulated mineral catalysts in out-of-equilibrium systems (as in Barge et al. 2012, 2013, 2014; Mielke et al. 2010, 2011); electrochemical studies of simulated precipitate materials to characterize their utility as electrocatalysts; and using these simulated prebiotic catalysts to create catalytic electrodes for use in a fuel cell. There are various types of fuel cell experiments that could be informative in this regard. Imitating the design of microbial fuel cells (MFCs), where the catalytic electrodes are submerged in two liquid reservoirs and separated by an ion-exchange membrane, would allow for testing of different fluid chemistries and dissolved ionic as well as gaseous electron donors and acceptors. MFC-type experiments would also lend themselves well to testing other relevant biological components as catalysts, such as individual enzymes or organisms. A geo-electrochemical system could also be simulated using a PEM fuel cell (PEMFC), which could use gaseous reductants / oxidants under higher pressure or temperature, with the simulated geological catalysts embedded in the gas diffusion layers (GDLs) within the membrane electrode assembly (MEA). Either way, the modular engineering arrangement of fuel cell components makes for a convenient planetary geology test-bed system in which components, such as the ion-exchange membranes, electrodes and catalysts (or electrolyte assembly unit), and the anode / cathode reactant feeds, may be substituted by materials more closely connected to geological environments. For example, rather than a commercial GDL catalyst layer, one could substitute simulated hydrogeological precipitates similar to those described above. This type of experiment is not limited to simulating hydrothermal vent environments; other geological electrochemical processes of interest could be explored in a similar experimental set-up, for example, as mentioned in Section 1 above, the electrochemical corrosion of meteorites on the early Earth (Bryant et al. 2009). Field samples of catalytic minerals derived from reduced Fe/Ni in meteorites, previously proposed to assist with early phosphorus chemistry (Pasek and Lauretta 2005; Pasek et al. 2013), could be treated in the same way as hydrothermal precipitates in the GDL electrocatalyst layers (vide infra).

We conducted some preliminary proof-of-concept tests using a PEM fuel cell to simulate the catalytic action of geological mineral catalysts in a far-from-equilibrium system. A sample experimental arrangement for a PEM fuel cell simulating a geo-electrochemical system is shown in **Figure 3**. In our preliminary tests, a commercial proton-exchange membrane and two GDLs containing electro-catalysts were sandwiched between two composite graphite bipolar plates, and the fuel cell was fed with humidified hydrogen (to represent hydrogen produced by serpentinization) and air at the anode and cathode sides, respectively. We examined the potential for efficient carbon-black-deposited platinum electrodes to be replaced by geological materials, in this case using Fe-Ni alloys present within iron meteorites (within which taenite and kamacite are the dominant phases). Whilst these materials may at first glance appear incompatible with terrestrial minerals, there is support for them to have contributed a significant component of the early earth lithosphere (Pasek and Lauretta 2008) and they are closely related to the hydrothermal Fe-Ni mineral, awaruite (Klein and Bach 2009). But most importantly, these meteorite samples represent a proof-of-concept for fabricating geologically analogous GDLs or electrodes for a planetary geology fuel cell reactor, using field samples rather than synthetic catalysts. Such a procedure could be applied to other field samples of interest, e.g., samples of hydrothermal precipitates, serpentinite, metal sulfides, or other minerals that might be important in a prebiotic system.

<u>Preparation of Geological Fuel Cell Catalysts:</u> The Fe-Ni "geological electrocatalyst" was prepared using shavings from the course octahedrite, group IAB-sLL Toluca meteorite which has a mean composition of 91% Fe and 8.1% Ni (**Figure 4**). This geological sample was ball-milled to an average particle diameter of ca. 5 microns before being dispersed in ethanol, sonicated in an ultrasound bath for 10 minutes, mixed with Nafion polymer binder and then spray-coated onto a GDL. For both electrodes, the amount of the catalyst required was calculated based on a 1.0 mg/cm^2 catalyst loading within an 11.56 cm^2 active area. Both the cathode and anode electrodes were positioned on the faces of the catalyst-coated Nafion membrane and hot-pressed (using a hydraulic press) to form a unit of MEA. The required amount of the catalyst was first ultrasonically mixed with a few drops of deionized water for a few seconds in order to break the catalyst powder into small pieces, and prevent the catalyst from being deactivated when blending it with the

dispersion agent, ethanol. A calculated amount of 50 wt% ethanol based on 15 mL ethanol per 50 mg catalyst was added to the wet catalyst and the resulting mixture was ultrasonically blended for about 3 minutes. The resulting blend was then mixed with a 5 wt% Nafion solution (Sigma Aldrich, UK) based on a loading of 0.1 mg Nafion/cm^2. (The Nafion solution is added in order to create an ionic phase through which the protons are transported from the anode to cathode.) This final mixture was ultrasonically blended for 15 minutes to form what is normally known as an ‘electrode ink’. The electrode ink was then manually sprayed onto the surface of the GDL mounted on a PTFE plate heated to 80°C in order to evaporate the alcoholic components, until the desired catalyst loading, 1.0 mg/cm^2, was reached.

Preparation of Membrane Electrode Assembly: A mix of 5 wt% Nafion solution, the amount of which was calculated based on 0.5 mg Nafion/cm^2, and acetone (about 3-5 mL) was sprayed onto the surface of the catalysed GDL in order to enhance the contact between the catalyst layer and the Nafion membrane in the MEA being fabricated. The Nafion membranes, N115, were pre-treated before being used in the fabrication of the MEAs. Namely, they were boiled at 80°C in 2 wt% hydrogen peroxide solution for 1 hour, rinsed several times with deionised water, then boiled in 1 M sulphuric acid at 80°C for 1 hour, and again rinsed several times with deionised water. Both the cathode and anode electrodes were positioned on the faces of the Nafion membrane and hot-pressed (using a hydraulic press) at 130°C and 50 kg/cm^2 for 3 minutes to form a unit of MEA.

Assembly and Operation of the PEM fuel cell: A single PEM fuel cell was assembled by sandwiching the MEA between two composite graphite bipolar plates (Bac2, UK) with an 11-turn and single-pass serpentine flow channel (2 mm x 2 mm, with a rib of width 0.8 mm). The PEM fuel cell, after being placed between two copper end plates, was connected to an in-house fuel cell test station at room temperature and fed with hydrogen at the anode side and air at the cathode side (**Figure 5**). The flow rates of both hydrogen and air were kept constant, equivalent to a stoichiometry of 2 calculated based on a 500 mA/cm^2 current density. Dry hydrogen and air were fully humidified by flowing them through bubbler-type humidifiers. The fuel cell was operated at room temperature. The pressures of the inlet and outlet gases were 1 and 0 barg, respectively. The electrochemical tests were performed using Gamry Reference 3000.

5. Results

5.1. Electrochemical Characterization of Simulated Hydrothermal Precipitates:

When the contrasting acidic/iron (ocean simulant) and alkaline/sulfide (hydrothermal simulant) solutions were added to the two-chamber cell, they interfaced across the porous separating barrier, and immediately began to produce a self-assembling iron-sulfide-containing precipitate film in/on the template. The characteristics of this inorganic membrane precipitate varied depending on compositions of the two solutions, and on the nature of the membrane template material, but commonly a complex landscape of micron-scale crystal structures formed (**Figure 6A**) – not unlike the crystal morphologies observed in the interior membranes of chemical gardens formed in injection experiments (Barge et al. 2012; Cartwright et al. 2011, 2002). When dialysis tubing was used as the template, the membrane precipitate formed within the pores and also on the surface of the dialysis tubing. Membranes precipitated on dialysis tubing proved to be ideal for electrochemical characterization within the original precipitation apparatus, since the dialysis template is only permeable to ions and solution mixing was completely prevented, and thus precipitates only formed on the template separating the two solution reservoirs, not in either solution. Dialysis templates also yielded membranes that were structurally stable enough to enable gentle rinsing and emptying / refilling of solutions, and when removed from the cell, the precipitate was a flaky solid film that could be easily dried and removed from the dialysis tubing for later use as a catalyst. Carbon cloth was much more porous than the dialysis tubing and when used as a precipitation template it did not prevent solution mixing as effectively. Thus in the precipitation fuel cell apparatus where carbon cloth was used as the separator, the two solutions often mixed near the interface (forming Fe/S precipitates in the bulk solutions as well as on the carbon cloth) and it was not possible to do electrochemical experiments with the membrane still in situ. We also found that it was necessary to pre-treat the carbon cloth by wetting it with ddH_2O, rinsing to remove extraneous carbon material that was not contained within the woven fibers, and applying $FeCl_2$ and Na_2S solutions directly to the cloth to form a "starter" precipitate in the pores before placing it between the two solution reservoirs. In any case, using carbon cloth caused the FeS precipitate to form in / on the carbon cloth fibers (**Figure 6B**) and since carbon cloth itself is conductive, a piece of the cloth containing FeS precipitate could be directly used as a working electrode.

We observed that even in a system containing only iron and sulfide, these self-assembling inorganic membranes were able to mediate chemical / pH gradients for several days (as observed by monitoring the pH of the acidic cell). The membranes also generated a diffusion potential of ~300-600 mV as they formed depending on experimental conditions (this has also been observed in a similar experiment by Filtness et al. 2003). This potential declined over several days as ions continued to diffuse through the membrane, eventually reaching equilibrium. When the iron and sulfide solutions were removed after a membrane had precipitated, and the solution reservoirs were re-filled with concentrated salt solutions of varying concentration, the iron sulfide-containing membranes still generated a membrane potential of a few tens to a hundred millivolts. Consistent with previous studies, this membrane potential was dependent on the concentrations of salts on either side of the membrane, and is likely due to the adsorption of charged cations / anions on the charged inorganic membrane surfaces. In galvanostatic tests where current was applied across a precipitated iron sulfide membrane, the I/V characteristic curves of these Fe/S membranes showed that the electrical resistance across the membrane changed at an inflection point (**Figure 7**), indicating that the Fe-S precipitate membranes formed in these out-of-equilibrium systems are somewhat bipolar – that is, partially cation- and anion-selective. This is an important property of biological and also electrodialysis membranes (Sakashita et al. 1983; Bauer et al. 1988; Strathmann et al. 1993; Honig and Hengst 1970; Aritomi et al. 1996).

The ability of simulated prebiotic iron sulfide chimney precipitates to facilitate electron transfer to a simulated Hadean ocean, with geologically realistic applied potentials between +/- 1 Volt, is shown in **Figure 8**. The simulated hydrothermal FeS membrane catalyst (grown on dialysis template, then attached to a GCE) on the electrode facilitated ~0.4-0.8 milliamps of both anodic and cathodic current. No significant current (> tens of µA) was observed with just the GCE, GCE with carbon tape, or in the ocean simulant containing FeS particles. Similar results (though at higher current ranging to +/- 10 mA) were observed when carbon cloth containing Fe/S precipitate was directly used as an electrode. As in the dialysis tubing experiments, the carbon cloth without Fe/S did not facilitate significant current at these potentials. The higher current range of Fe/S membranes on carbon cloth, compared to Fe/S membrane material taken from dialysis tubing, is likely due to the fact that the active surface area is greater on the carbon cloth working electrodes.

5.2. PEM Fuel Cell Function using Geological Electrocatalyst Materials Derived from Iron Meteorites

Electrochemical analyses of the PEM fuel cell in operational mode revealed a stable open circuit voltage difference across the electrodes peaking close to 0.7 V (**Figure 9 upper**). In the presence of Toluca electrocatalyst impregnated GDLs, the current-voltage polarization curves (**Figure 9 lower**) show distinct behavior confirming that the fuel cell is functional, and the current flow increases with decreasing voltage as expected, approaching a load of ca. 13 mA. In the absence of the geological catalyst components, we observed no such polarization behavior.

6. Discussion:

The iron sulfide membranes formed at the interface of contrasting solutions in our experiments represent specific mineral components that could have been contained within larger, carbonate- and silica-containing hydrothermal chimney precipitates on the early Earth. Iron sulfide precipitates in a hydrothermal chimney, formed in pH / ion gradients via diffusion and self-assembly, could contribute to the electrocatalytic properties of the chimney if they were sufficiently conductive. Mackinawite (FeS) is usually the first metastable product that forms from the precipitation of aqueous Fe^{2+} and HS^-, and over time it can transform to greigite (Fe_3S_4), pyrrhotite, or pyrite (Csákberényi-Malasics et al. 2012; Sines et al. 2011, Kwon et al. 2011). Mackinawite is a layered mineral composed of Fe-S tetrahedra stabilized by van der Waals forces, and since the Fe-Fe distance is only 2.60 Å (similar to elemental α-iron: 2.48 Å), it is highly electrically conductive and even has been shown to exhibit superconducting characteristics (Kwon et al. 2011). It has been demonstrated that black smoker chimney material – also mostly composed of metal sulfides – is capable of converting the geochemical redox potential between hydrothermal fluid and seawater to electrical current and catalyzing redox reactions (Nakamura et al. 2010b). Even though a hydrothermal chimney at an alkaline vent on the early Earth may not have such a high fraction of metal sulfides as a modern black smoker chimney, still it is possible that in a prebiotic system subject to electrical potential gradients between hydrothermal fuels (e.g. H_2, CH_4) and seawater oxidants (e.g. CO_2, NO_3^-, NO_2^-, Fe^{III}), deposits of conductive Fe/S precipitates – aided by their large active surface area as seen in **Figure 6** – might have been able to facilitate electron conduction and catalyze redox reactions on the mineral surfaces.

Our preliminary experiments reveal that, due to their far-from-equilibrium nature, inorganic precipitate membranes analogous to hydrothermal chimney walls have many unusual properties (van Oss 1984) that may be biologically and/or prebiotically relevant. The growth of a hydrothermal chimney precipitate separating contrasting solutions could generate and maintain an electrical potential for long periods of time, yet still facilitate selective ion transfer through the membrane. The precipitates formed would likely exhibit a compositional gradient from exterior to interior (as is seen in real hydrothermal chimneys, as well as chemical garden lab experiments (Haymon 1983; Barge et al. 2012; Cartwright et al. 2002;)) and the charged mineral surfaces could adsorb and concentrate ions from either ocean or hydrothermal solution. Metal sulfide-containing chimneys formed within a temperature / pH gradient could potentially host a range of mineral phases with various electrochemical properties. It is likely that these inorganic membranes could also incorporate silicates, phosphates, Ni, and Mo into the precipitate (Barge et al. 2014; Barge and Kanik, unpublished data). More experimental work is needed to fully characterize the properties of geological mineral precipitates in far-from-equilibrium systems, in order to understand the reactions that may be possible in hydrothermal chimney environments.

In some ways, the inorganic Fe/S precipitates formed on carbon cloth in our interface experiments are similar to the ion-exchange membranes and gas diffusion layers in commercial as well as experimental fuel cells (Kim et al. 2009; Paulo and Tavares 2011). Our preliminary success of using commercially available fuel cell electrode/GDL material (carbon cloth) as a template to precipitate simulated hydrogeological mineral catalysts is promising, as these fabricated electrodes can be directly used in PEM fuel cell assemblies to catalyze redox reactions in a pressurized out-of-equilibrium geology test-bed reactor. Our preliminary PEM fuel cell tests show that geological minerals applied to the GDLs as catalysts have the potential to facilitate current flow and possibly catalyze redox reactions in putative geological fuel cells. Our methods proved successful for creating PEM fuel cell MEAs using geological materials (Toluca meteorite taenite & kamacite material) as electrode catalysts, and should work for any field sample or synthetic material of similar composition.

Combined, these electrochemical techniques and fuel cell reactor designs may allow for testing of specific reactions that are proposed to be relevant to the emergence of metabolism. For example, some metal sulfide minerals that form under hydrothermal conditions are structurally similar to the inorganic active centers of certain fundamental redox enzymes (Rothery et al. 2008; Baradaran et al., 2011; Russell and Hall 1997; Nitschke et al. 2013), and so synthetic hydrothermal precipitates formed in the laboratory could be tested as "proto-metalloenzymes" in electrochemical and fuel cell studies. Experimentally testing the effects of trace chemical components on the electron transfer capabilities of a hydrothermal chimney could also be instructive. To give one example of this, Nitschke and Russell (2013) have proposed a denitrifying methanotrophic acetogenic pathway for the origin of metabolism, in which the energy barriers (from CH_4 to methanol, and CO_2 to CO) could be solved by electron bifurcation by hydrothermally-produced Mo precipitating in trace amounts within a chimney wall (Helz et al., 2011; Nitschke and Russell 2009, 2011) – an interesting proposition, since, in biology, energy barriers are also surmounted through redox-bifurcating enzymes such as quinones, flavins and molybdenum and tungsten pterins (Staniek et al., 2002; Darrouzet and Daldal, 2002; Herrmann et al., 2008; Li et al. 2008; Nitschke and Russell, 2009, 2011, 2013; Kaster et al., 2011; Schuchmann and Müller, 2012; Buckel and Thauer, 2012; Poehlein et al., 2012). The end result of a fuel cell experiment simulating a geo-electrochemical system would be to reveal redox reactions in either half-cell as a function of the composition of electrodes and membrane. In a simulation of a prebiotic alkaline hydrothermal vent, for example, electrical current could be produced by oxidation of hydrothermal H_2 to electrons and protons, thus driving the reduction of oceanic CO_2 to formate and possibly further to formaldehyde; meanwhile hydrothermal CH_4 could be oxidized to methanol and formaldehyde perhaps using nitrate or nitrite as an electron acceptor (Nitschke and Russell, 2013; Russell et al., 2013; Ducluzeau et al. 2009). Organic redox cofactors such as quinones, flavins, NADP, FAD, GMP and AMP which participate in electron transfer in modern bioenergetics and were likely also present in LUCA (Buckel and Thauer 2012; Schoepp-Cothenet et al., 2013) could also be incorporated into fuel cell electrodes for origin-of-life simulations.

Regardless of the specific parameters, approaching origin-of-life experiments within a fuel cell / electrochemical framework could allow experimenters to take advantage of the many rigorous analytical methods and diagnostics that are routinely employed in fuel cell tests. Moreover, there are various computational models that can be used to model fuel cell systems, none of which (to the best of our knowledge) have been employed with an abiogenesis perspective in mind. There are a great many reactions of importance to the origin of life aside from the proto-metabolic redox reactions discussed above, such as RNA synthesis or formation of peptides or other complex organics, or polymerization of phosphates to create energy currency. Any of these could be incorporated into a fuel-cell-like experiment and could benefit from electrochemical techniques. For example, it has been recently shown that Fe^{2+} can cause some biological RNAs to become redox active (Athavale et al. 2012; Hsaio et al. 2013), and thus in an early Earth hydrothermal system it is possible that these redox-active RNAs might participate in the electrocatalytic function of a hydrothermal chimney system along with the inorganic components (cf. McGlynn et al. 2012). Another example could be studying the generation of condensed and/or activated phosphates, which are necessary at the origin of life to function as energy currency molecules similar to ATP (Baltscheffsky 1996). It has been proposed that green rust, a redox-active iron oxy-hydroxide mineral that efficiently absorbs phosphate (Barthelemy et al. 2012; Antony et al. 2008), might be capable of forming pyrophosphate bonds via regulation of proton flow through mineral layers (Arrhenius 2003; Russell et al., 2013); alternately, pyrophosphate could be formed via substrate phosphorylation within a hydrothermally precipitated membrane (Barge et al. 2014; Martin and Russell 2007). Incorporating components like these into electrodes in a fuel cell experiment might yield new prebiotically plausible ways to synthesize necessary high-energy molecules.

7. Conclusions:

There are many proven engineering design techniques and analytical methods for building membrane fuel cells and characterizing the electrochemical properties of different organic and inorganic catalysts. These techniques and methods can offer much insight into the workings of bioenergetics and metabolism if applied to biological & geological early Earth catalysis. Considering that 1) microbes and mitochondria may be viewed as highly evolved biological fuel cells, and 2) that the earliest life on Earth was likely chemiosmotic, chemosynthetic and utilized energy sources such as those commonly found in modern alkaline hydrothermal vents, we believe that the prebiotic-fuel cell modeling approach has potential. We argue that the electrochemical methods which have been so successful in the fields of fuel cell development and bioelectrochemistry should be applied to simulating the origin of life because of the conspicuous fuel cell-like properties of metabolic and geo-electrochemical systems. Our preliminary experiments presented here were successful in forming putative electro-catalytic minerals in out-of-equilibrium chemical systems, mimicking the formation of metal sulfide precipitates at the interface between ocean and hydrothermal fluids on the early Earth. These inorganic membranes simulating prebiotic submarine hydrothermal precipitates are shown to be structurally complex, are capable of generating and maintaining membrane potentials and pH gradients, and are electrically conductive. We also successfully formed simulated hydrothermal chimney precipitates on commercial fuel cell electrode / GDL material, a technique which can be utilized to create electrode components for various types of fuel cell experiments. Finally, we have developed a PEMFC for simulating geo-electrochemical processes, and have demonstrated that the PEMFC is operational when field samples of geological material were used as the electrode catalysts. The methods utilized in this work are easily generalized to other geochemical interfaces of interest, such as water-rock reactions on early Mars or the icy moons. Moving toward this type of laboratory simulation of the emergence of bioenergetics will not only be informative in the context of the origin of life on Earth, but may help us understand whether it is possible for life to have emerged in similar environments on other planets.

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Figure Captions:

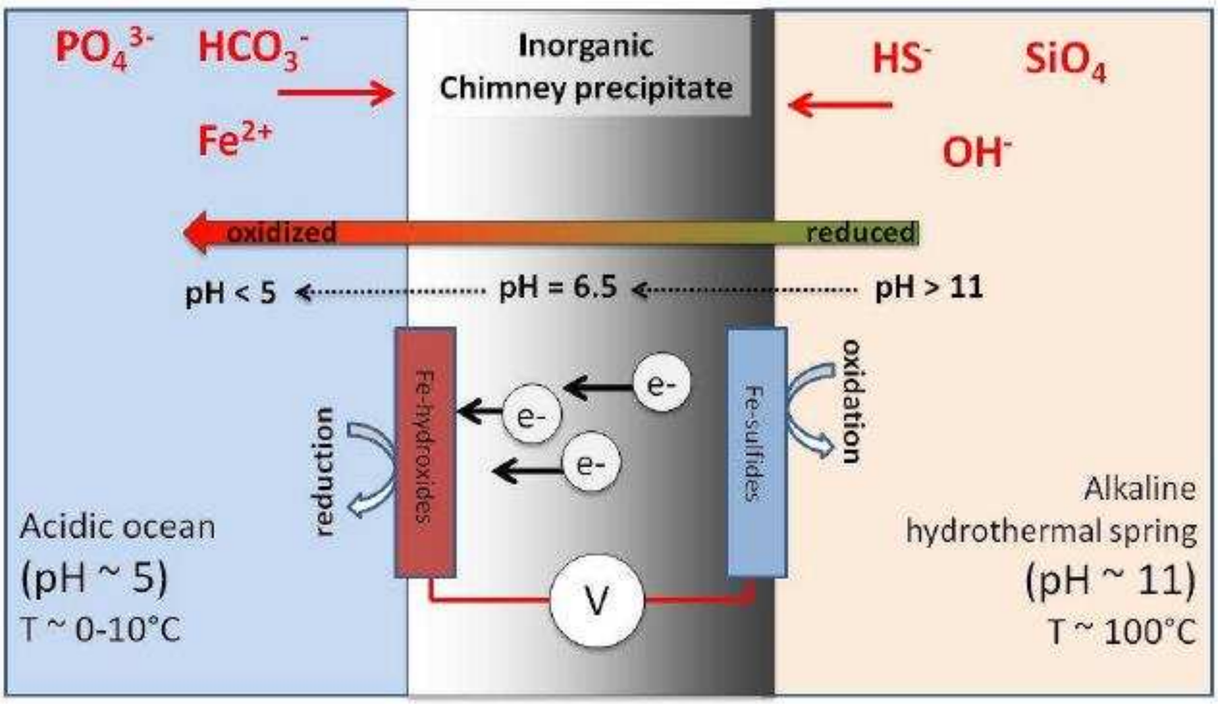

Figure 1: Alkaline hydrothermal vent on the early Earth modeled as a membrane fuel cell. The reservoirs of seawater (relatively oxidizing, acidic) and hydrothermal fluid (reducing, alkaline) are physically separated across a hydrothermal chimney precipitate, which functions as part electrolyte and part capacitor. The chimney precipitate would have contained (among other things) iron sulfide minerals, due to the solubility of Fe^{+2} in the early anoxic oceans and the influx of soluble bisulfide (HS-) from the hydrothermal fluid. The anode and cathode are the inside and outside surfaces of the chimney, which would have variable composition reflecting the chemical gradients in which they precipitated. The chimney walls would have been somewhat conductive to electrons, which would pass from fuels in the hydrothermal reservoir to oxidants in the ocean reservoir, catalyzed by electrochemically active minerals in the chimney wall.

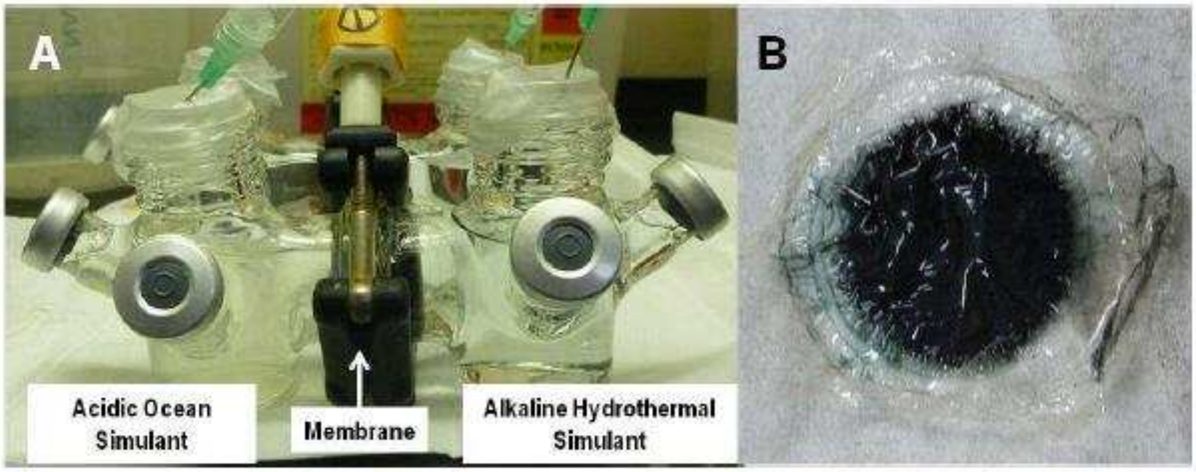

Figure 2: (A) Precipitation of Fe/S membrane between two interfacing solutions in a glass fuel cell apparatus. The reservoirs represent the Hadean ocean and alkaline hydrothermal fluid, and they are separated by a synthetic porous separator that allows ion contact but not solution mixing. (B) Fe/S membrane precipitated on dialysis membrane (precipitate area ~ 4.9 cm^2).

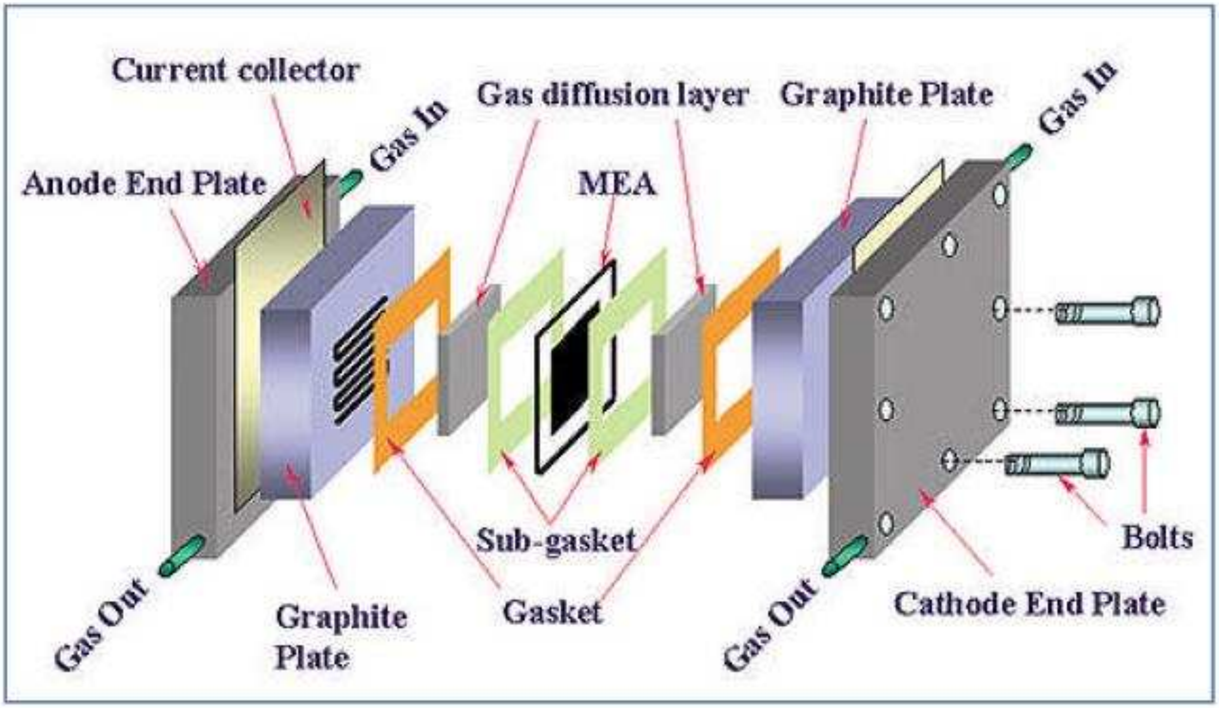

Figure 3: Outline of PEM fuel cell sub-system construction.

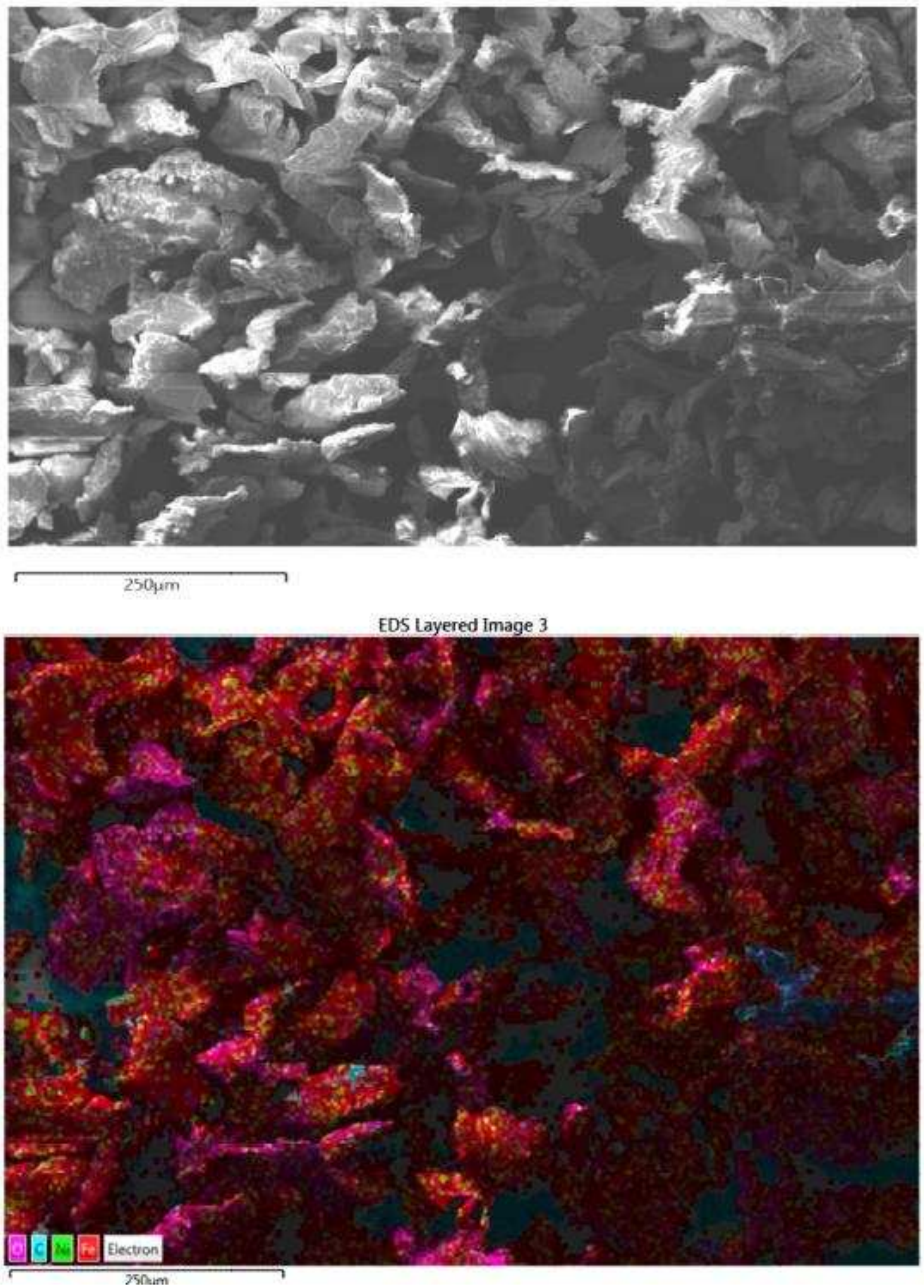

Figure 4. (A) SEM image (secondary electron) of shavings from the type IAB-sLL Toluca meteorite with mean composition of 91% Fe and 8.1% Ni. (B) Energy dispersive mapping spectrum (EDS) on the same course-grained meteorite sample with elemental color identification showing a small degree of surface oxidation of the meteorite shavings.

Figure 5: (A) PEM fuel cell test-rig at University of Leeds. (B) Close-up of complete MEA fuel cell assembly with the anodic surface in the foreground connected to dihydrogen inlet and outlet supply. The cathodic reverse side is supplied with air, within which dioxygen acts as ultimate electron acceptor.

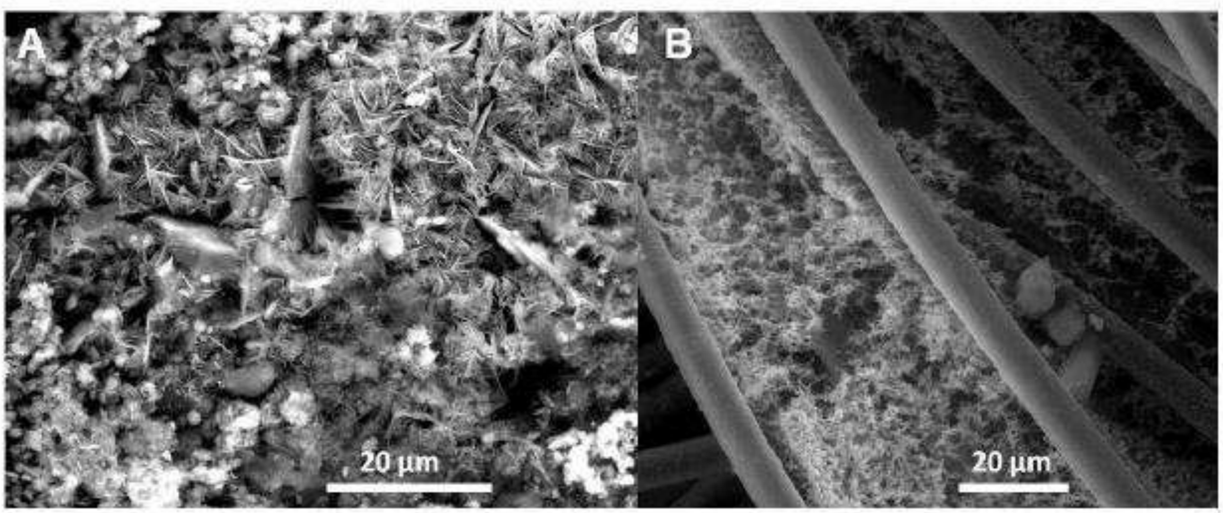

Figure 6: ESEM images of iron sulfide membrane precipitates on different membrane templates: (A) dialysis tubing and (B) carbon cloth (the cylinders are carbon fibers). ESEM images were obtained using a voltage of 20 kV and a working distance of 10 mm.

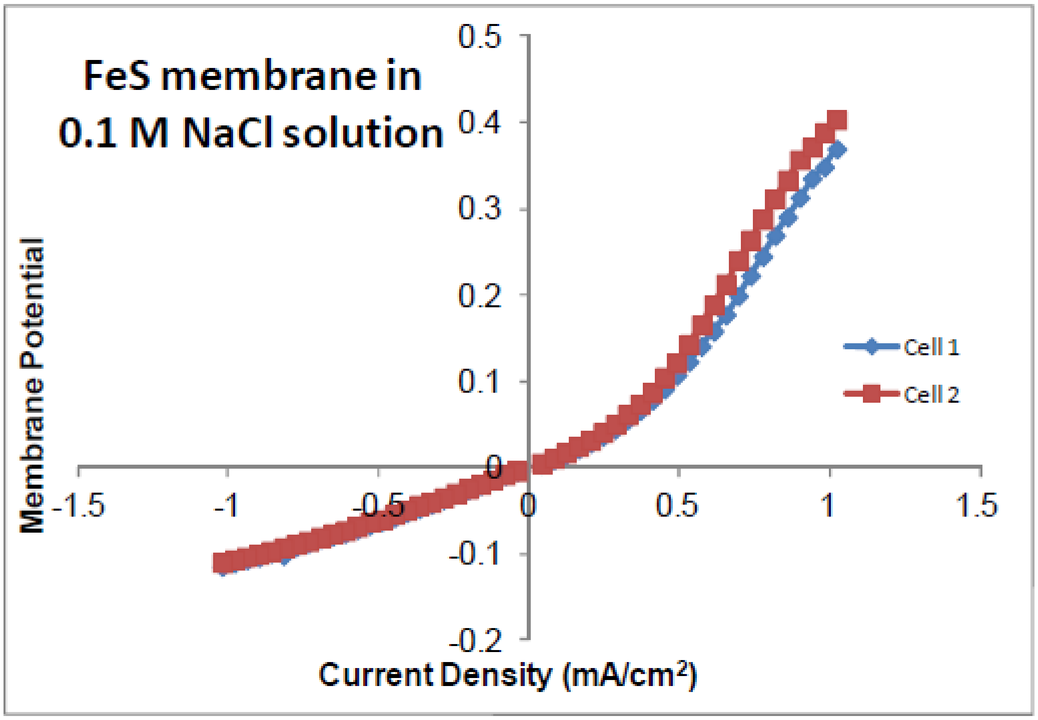

Figure 7: Current/voltage characteristics of an iron sulfide precipitate membrane. The membrane was formed at the interface between simulated Hadean ocean and alkaline hydrothermal solutions, then these solutions were removed and replaced with 0.1 M NaCl. Voltages are measured relative to a rest potential.

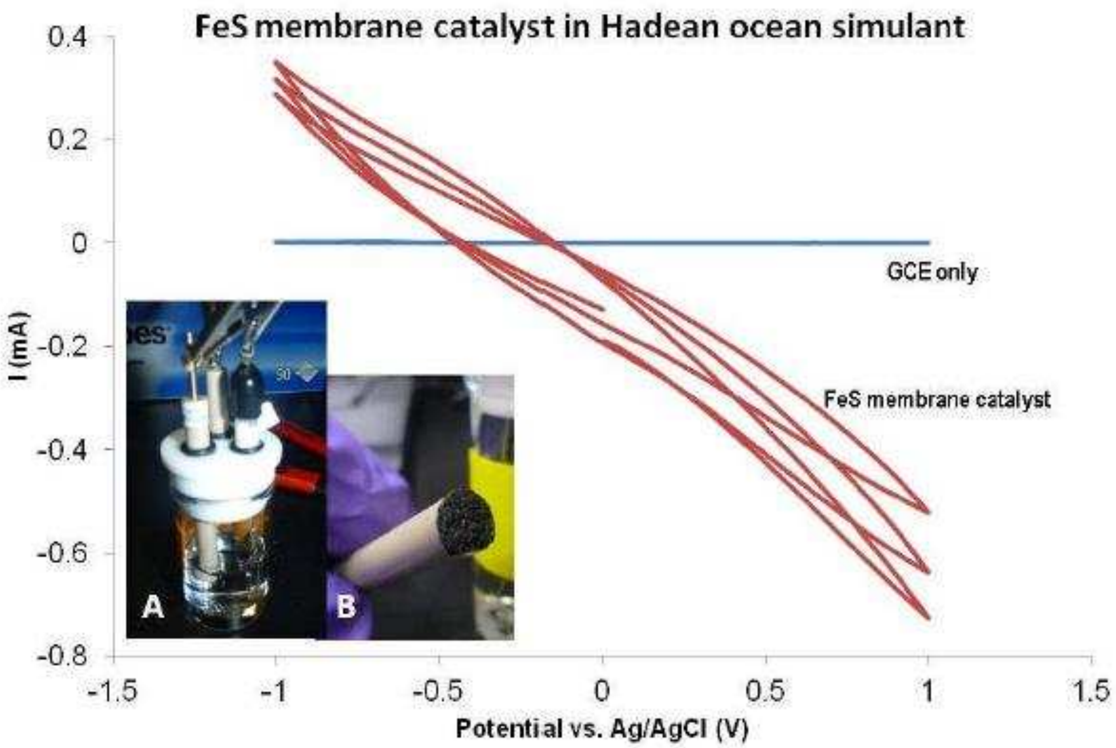

Figure 8: Cyclic voltammograms of GCEs in N_2-purged Hadean ocean simulant (20 mM $NaHCO_3$ + 0.6 M NaCl, pH 5), with and without simulated Fe/S hydrothermal membrane as an electro-catalyst. Inset A: anoxic voltammetry cell. Inset B: Fe/S membrane precipitate dried and applied to GCE.

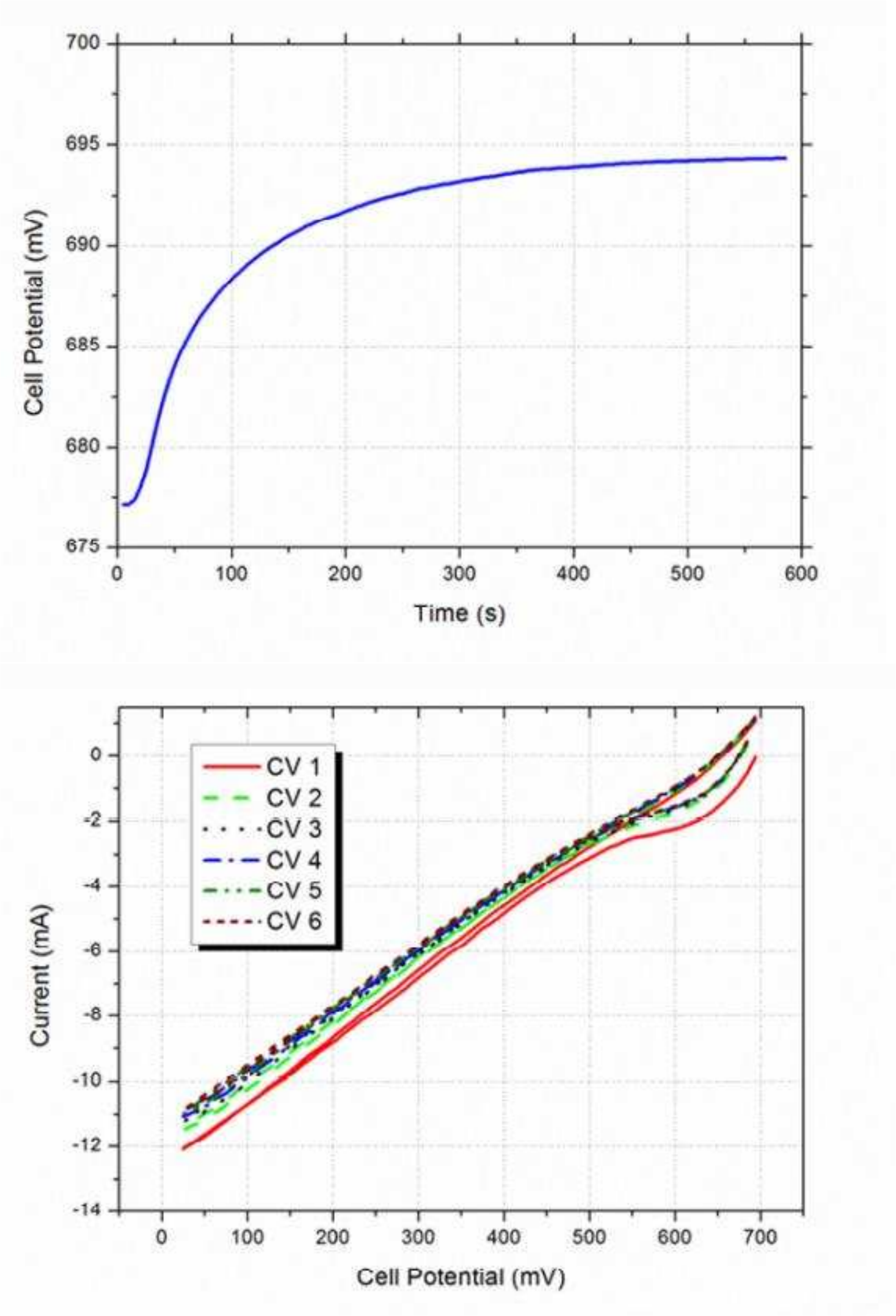

Figure 9. Open circuit voltage (upper) and polarization (I/V) curves of the Toluca MEA fuel cell

(lower). The negative sign in the I/V graph indicates that the current is being taken from the functional fuel cell.