The impacts of geothermal power plant emissions on terrestrial ecosystems in contrasting bio-climatic zones

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Resources

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Abstract

Very little is known on the ecosystem impacts of emissions from geothermal power plants. The emissions, comprising mainly of non-condensable gases (NCGs) i.e. carbon dioxide, hydrogen sulphide, methane and trace elements such as arsenic, boron, antimony and mercury, have the potential to deposit and accumulate in ecosystems. At elevated levels, some NCGs can cause ecosystem stress, especially H_2S and the trace elements. The aim of this thesis is to assess the effects of these elements on terrestrial ecosystems around two geothermal areas in contrasting biomes i.e. Kenya and Iceland.

Dominant plant species around each geothermal study area, *Tarchonanthus camphoratus* shrub in Kenya and *Racomitrium lanuginosum* moss in Iceland, were used as bio-indicators and concentrations of sulphur, arsenic, boron, antimony and mercury were mapped in their tissues and soils at increasing distances from the power plants along the prevailing wind direction in field surveys. Patterns of plant growth and health along the same distances and wind direction gradients were also studied to assess any potential effects related to the power plants. Controlled experiments were thereafter carried out on the same plant species to assess in detail the effects of the most abundant phytotoxic NCG, i.e. H_2S gas, on plant growth and health.

Results of the field surveys and experiments indicated that the main geothermally emitted component, H_2S gas, deposits and accumulates in plants and soils. The measured trace element concentrations in plants and soils (from the field surveys): arsenic, boron, antimony and mercury, did not show strong patterns attributable to the geothermal power plant emissions. Further, results of the surveys in relation to geothermal power plant emissions showed weak indications of effects on *Tarchonanthus camphoratus* shrub growth and health around the Olkaria geothermal power plants in Kenya, while in Iceland, the growth of *Racomitrium lanuginosum* moss was reduced around the Hengill geothermal power plants. Additionally, the experiments showed that, 30 µg/L aqueous H_2S

(10.96 ppm in air) may be a tolerable limit for plants around geothermal power plants in Kenya and Iceland. These findings serve as important baseline data toward environmental monitoring and management around both geothermal power plant areas in Kenya and Iceland; this information is also of utmost importance in advising the public and decision makers in Kenya and Iceland on the ecosystem (terrestrial) impacts of geothermal power plant emissions.

Útdráttur

Áhrif efnalosunar frá jarðvarmavirkjunum á vistkerfi eru almennt fremur illa þekkt. Losunin samanstendur aðallega af óþéttanlegum lofttegundum, þ.e. koldíoxíði, brennisteinsvetni og metani, en einnig af snefilefnum eins og arseni, bór, antimon og kvikasilfri sem geta öll safnast fyrir í vistkerfum. Við hærri styrk geta þessi efni valdið álagi á vistkerfi, einkum brennisteinsvetni og snefilefnin. Markmið verkefnisins var að kanna áhrif þessara efna á landvistkerfi við jarðvarmavirkjanir við líffræðilega mjög ólíkar aðstæður, þ.e. í Kenýa og á Íslandi.

Ríkjandi plöntutegundir umhverfis virkjanasvæðin, *Tarchonanthus camphoratus* runni í Kenýa og *Racomitrium lanuginosum* mosi á Íslandi, voru notaðar sem líffræðilegir vísar og styrkur brennisteins, arsens, bórs, antimons og kvikasifurs mældur í þeim og í jarðvegi í mismunandi fjarlægð frá virkjununum í stefnu ríkjandi vindátta. Vöxtur og heilbrigði plantna voru einnig könnuð í sömu fjarlægðum til þess að meta möguleg áhrif er tengjast jarðvarmavirkjunum. Einnig voru gerðar staðlaðar tilraunir þar sem áhrif brennisteinsvetnis á vöxt og heilbrigði tegundanna tveggja voru könnuð.

Niðurstöður vettvangsrannsókna og tilrauna benda til þess að brennisteinsvetni, eitt meginefnið sem losað er frá jarðvarmavirkjununum, safnist fyrir í plöntum og jarðvegi. Styrkur snefilefnanna arsens, bórs, antímons og kvikasilfurs í plöntum og jarðvegi benti hins vegar ekki til þess að þau mætti rekja til losunar frá virkjununum. Við orkuverin í Olkaria í Kenýa komu fram fremur veik en neikvæð áhrif á vöxt og heilbrigði runnans *Tarchonanthus camphoratus*. Þessi áhrif voru hins vegar merkjanleg á mosann *Racomitrium lanuginosum* við íslensku orkuverin við Hengil. Tilraunir sýndu að bæði í Kenýa og á Íslandi virðist styrkur H_2S sem nemur 30 µg/l í vatnslausn (10,96 ppm í lofti) vera efri þolmörk fyrir tegundirnar tvær. Þessar niðurstöður leggja mikilvægan grunn að umhverfisvöktun og við stjórn jarðvarmaorkuvera bæði í Kenýa og á Íslandi. Að auki eru þær mjög mikilvægar bæði fyrir almenning og við alla ákvarðanatöku er snerta áhrif losunar efna frá jarðvarmavirkjunum á vistkerfi í báðum löndunum.

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For You:

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...to your unwavering support and purest form of love, and for putting me through the best education possible. I appreciate your sacrifice and wouldn't have reached this far without you!

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...You are all the determination in every page!

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List of papers

This thesis is based on three papers of which one is published, one is submitted and one is a manuscript to be submitted. The papers will be referred to in the text using their respective numbers as follows:

Paper I: Mutia, T.M., Fridriksson, T., Jónsdóttir, I.S., 2016. Concentrations of sulphur and trace elements in semi-arid soils and plants in relation to geothermal power plants at Olkaria, Kenya. Geothermics 61, 149 – 159.

Paper II: Mutia, T.M., Fridriksson, T., Magnússon, S.H., Jónsdóttir, I.S., 2016. Elevated concentrations of sulphur and trace elements in subarctic soils and moss in relation to geothermal power plants at Hengill, Iceland – ecological implications. (Submitted to Science of the Total Environment).

Paper III: Mutia, T.M., Fridriksson, T., and Jónsdóttir, I.S., 2016. Effects of experimental hydrogen sulphide deposition on dominant plants around geothermal power plants in Kenya and Iceland. (Manuscript).

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“It always seems impossible until it’s done.” ~ Nelson Mandela

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The famous words of Lupita Nyongo spurred my days; that

…...´No matter where you are from, your dreams are valid´

1. Introduction

Geothermal energy is among the energy sources that can replace fossil fuels and thereby help decrease greenhouse gas emissions (GHG) and the effects of climate change. This owes to its intrinsic stability and relatively low environmental impacts, considering its generally low CO_2 emissions and ecological effects. For this reason, growth of the installed capacity of geothermal power plants has been steady over the past ten years in countries with potential for harnessing the resource. Globally, an average increase of about 200 – 350 MW/yr between the years 2000 – 2014 has been reported, presently totaling to 12.7 GW (Bertani 2003, 2005a, 2005b, 2007, 2012 and 2016). With the successful implementation of these past projects, even further growth is projected, an additional 8 GW of project proposals is intended for transformation into real power plants by 2020 totaling 21 GW (Bertani, 2016). However, like the development of any other energy source, geothermal power plant projects are not entirely free of environmental impacts. Conventional geothermal power plants emit a range of gases into the atmosphere, several of which have been reported to deposit and accumulate into their nearby ecosystems. Some of the gases are potentially toxic to some organisms within ecosystems even at low concentrations and are a growing concern since the effects are not fully understood.

Attempts have been made to study potential ecosystem impacts such as the discharge of spent geothermal fluids (Wetangula, 2004; Were, 2007) and gaseous emissions (Wetangula, 2011; Ólafsdóttir et al., 2014) arising from geothermal wells drilling and power plants construction and operation, particularly for mitigation as most geothermal resources and power plants are located in undisturbed habitats. However, the understanding relating geothermal power plant emissions to impacts on ecosystems is still limited and mainly based on a few studies in the Mediterranean Italy (Bargagli et al., 1997; Bussotti et al., 1997; Loppi et al., 1999, 1998; Bussotti et al., 2003; Chiarucci et al., 2008).

For the entire geothermal development process i.e. drilling of wells to power plant constructions, environmental impacts arising from operations of geothermal power plants are most significant, because they are likely to have long term air quality and ecosystem effects, as the lifetime of power plants is estimated on average at 30 years (Hondo, 2005). Therefore, when compared to the other geothermal development processes, impacts arising from geothermal power plants are more critical as they involve gaseous emissions into the atmosphere for as long as the power plant operates, however minimum as assumed. Drilled wells on the other hand do not assumedly have a similar magnitude of impacts; with respect to land degradation, their surrounding areas can in most cases be reclaimed through restoration of vegetation (e.g. by re-planting) that existed before and leaving only the area occupied by an individual well (smaller compared to area occupied by power plants). Moreover, the wells are in the long run connected to power plants for steam supply and power production, therefore there is less chance of emissions from wells than from power plants, except during well maintenance.

This thesis addresses the impacts of geothermal power plant emissions on terrestrial ecosystems. Specifically, it focuses on the impacts within two contrasting climatic zones where these studies have not been conducted before, the semi-arid parts of Kenya (Figure 1.1) and subarctic Iceland (Figure 1.2). This study is thus first in provision of comprehensive and baseline knowledge on the impacts of geothermal plant emissions on terrestrial ecosystems, an important consideration in the development of sustainable geothermal power plants in Kenya and Iceland.

Figure 1.1. Olkaria II geothermal power plant in semi-arid Kenya. The visible plants are mainly the shrub Tarchonanthus camphoratus L. *that dominates the vegetation in the area (Photo: T. Mutia, 2014).*

Figure 1.2. Hellisheidi geothermal power plant in subarctic Iceland. The area is dominated by the Racomitrium lanuginosum (Hedw.) Brid., *moss plants (Photo: Reykjavik Energy, 2016).*

To evaluate these impacts on terrestrial ecosystems in each area, plants and soils were used as ecosystem indicators in field surveys. Deposition and accumulation of geothermally emitted elements in ecosystems was assessed by establishing concentration patterns of the elements in plants (dominant within the area) and soils at varying distances from the power plants and in relation to the prevailing wind direction. In addition, the impacts were assessed by measuring selected plant characteristics related to growth and damage. To investigate causal relationships, experiments were performed to assess the effect of one of the major components of geothermal emissions i.e. hydrogen sulphide (H_2S) gas, on the dominating plants in each area.

1.1. Geothermal power plant emissions and ecosystem implications

Despite having considerably lower GHG emissions in comparison to conventional fossil fuel plants (Axtmann, 1975), geothermal power plant emissions can still be significant.

Geothermal fluids contain non-condensable gases (NCGs) at various amounts, that are exhausted into the atmosphere after power production. The reported exhaust amount of NCGs per weight of steam is mostly resource dependent and varies across conventional geothermal power plants, mainly because the gas fractions depend on the underground reservoir geochemistry (Axtmann, 1975). Generally, and depending on the resource, the NCGs range between 0.2% and over 25% weight of steam, in rare cases (Ozcan and Gokcen, 2009). Similarly, the NCGs composition (and amounts of specific gases) may differ between fields and power plants. In common cases, the gases comprise 78 – 98% w/w carbondioxide (CO_2), 1 - 24% w/w hydrogen sulphide (H_2S), 0.02 - 0.65% w/w methane (CH_4), 0.1 - 8% w/w hydrogen (H_2), 0.3 - 16% w/w nitrogen (N_2), 0.1 - 3% w/w argon (Ar), and traces (<0.001% w/w) of radon (Rn), boron (B), mercury (Hg), arsenic (As), antimony (Sb), and ammonia (NH_4) in gaseous and dissolved form (Axtmann, 1975; Baldi, 1988; Loppi et al., 1998; Gunerhan, 1999; Loppi, 2001; Bussotti et al., 2003; Ozcan and Gokcen, 2009; Rodríguez, 2014). The emitted gases, depending on quantities and prevailing wind, will disperse, deposit and accumulate into ecosystems with possible consequences.

Until recently, not much attention had been paid to the consequences, especially to plant species. Yet the effects of H_2S emissions from geothermal power plants are fairly well examined on human health for occupational health and safety reasons (Guidotti, 1996; WHO, 2000; Hansell and Oppenheimer, 2004; Finnbjornsdóttir et al., 2015). However, with increasing interest on geothermal energy development, the public, policy makers and scientists are concerned and motivated to question the effects of these gases, especially hydrogen sulphide and trace elements, on plants species and ecosystems in general due to their potential toxicity, even at low concentrations.

Since the emitted amounts of hydrogen sulphide gas and trace elements exceed those in the ambient environment, they pose concern due to their toxicity, even at low concentrations, and their possibility to bio-accumulate causing deleterious effects. The effects are described in the next sub sections.

1.1.1. Hydrogen sulphide gas emissions and effects on plants

Monitoring of H_2S gas emissions from geothermal power plants is increasingly becoming important for many geothermal power project developers as a national and international requirement to mitigate against the likely gas effects on human health and wellbeing. In that case, several air quality guidelines such as those for WHO, (2000) have been prepared for occupational health and safety reasons. However, for ecosystems, H_2S gas emission guidelines or limits have not yet been developed, probably because of the low awareness of potential impacts of H_2S gas on ecosystems.

The H_2S gas once emitted into the atmosphere undergoes a series of reactions depending on the environmental conditions, and due to its instability it may in some cases oxidise to sulphur dioxide gas or sulphuric acid (in case of precipitation) and be deposited subsequently in ecosystems. Kellogg et al., (1972) explain the chemical reactions of H_2S gas in air. These various forms of sulphur deposit and accumulate in plants and soils. Sulphur (S) concentration in upper soil layers and above ground plant parts which are the major receptors of atmospheric pollution, has been used to assess sulphur accumulation from emitted hydrogen sulphide gas in the ecosystems (Gonzales, 1984; Bussotti et al., 1997;

Bargagli et al., 1997; Loppi et al., 1998; Bussotti et al., 2003; Bargagli et al., 2003). Other plant growth related traits such as plant height, leaf/shoot injury and abundance and leaf physiological measurements have also been used in such assessments (Clarke and Murray, 1990; Bussotti et al., 2003; Rajput and Agrawal, 2005; Tuyor et al., 2005; Wali et al., 2007; Chiarucci et al., 2008; Zvereva et al., 2010). Despite any anticipated negative effects on plant health, sulphur, up to a certain optimum level of concentration, is a plant macronutrient and therefore hydrogen sulphide gas is a potential nutrient (sulphur) source for plants.

As pointed out earlier, field studies relating effects of excess sulphur (from H_2S emissions) from geothermal power plants on plants are mainly limited to the Mediterranean. These studies report increasing sulphur concentrations in plant leaves with decreasing distances away from geothermal power plants. Furthermore, higher sulphur concentrations have been found in the upper soil layers than below in the vicinity of the power plants (Bussotti et al., 2003). These findings imply atmospheric input of sulphur from the nearby geothermal power plants. In addition, a few sulphur fumigation experiments have been conducted to assess plant responses (Thompson and Kats, 1978; Gonzales, 1984; Maas et al., 1987). The findings from both the field surveys and experiments indicate various effects on plant leaves and the experiments especially showed varied plant responses across species at different H_2S gas dosages. Most commonly, excess sulphur is reported to affect plant growth and metabolism. Effects are visible on leaves and include foliar injuries manifested as necrosis, defoliation and in the long term as reduced growth, early senescence and chlorosis (Thompson and Kats, 1978; Varshney et al., 1979; Maas et al., 1987; Bargagli et al., 1997; Bussotti et al., 1997; Bussotti et al., 2003).

In non-vascular plants such as mosses, levels of excess sulphur from geothermal power plants have also been revealed (Baldi, 1988; Bargagli et al., 2002; Bragason and Yngvadóttir, 2009; Carballeira and Fernandez, 2002; Loppi et al., 1999, 1998; Loppi and Bargagli, 1996). However, the effects on growth and physiology are not well explored.

1.1.2. Trace element emissions and effects on plants

The practice of monitoring amounts of trace elements emitted from geothermal power plants is rare. Most geothermal power plant operators are not cognizant of this need, likely because of the assumption that the elements are found in trace amounts, if at all. Even so, data is insufficient to ascertain the assumption.

The deposition patterns of trace elements in the surroundings of geothermal power plants have been assessed in a similar way as for sulphur in plants. Results show trends of decreasing element concentrations in plants with increasing distance away from geothermal power plants, implying that the elements are of power plant origin (Baldi, 1988; Bargagli et al., 1997; Bussotti et al., 2003). Moreover, the relationship between the measured trace element concentrations in plants and plant health has also been assessed in the Mediterranean studies (Bargagli et al., 1997; Bussotti et al., 1997; Loppi et al., 1999; Bargagli et al., 2002; Bussotti et al., 2003) and associated consequences reported.

Similar to the elevated sulphur levels in plants, high levels of arsenic, boron, mercury, antimony and other trace elements have been measured in plants (Bargagli et al., 1997). Symptoms mainly of boron and arsenic toxicity have been observed and described in fair detail (Bussotti et al., 1997). Elevated levels of boron and arsenic in plants are associated with compromised leaf conditions such as leaf area reduction, damaged chloroplasts and reduced chlorophyll contents. Boron is also associated with leaf injury, manifested as marginal necrosis (Bussotti et al., 1997; Bussotti et al., 2003). Since most trace elements are of no nutritive value to plants, except boron, their high levels can also cause deleterious effects on plant growth and metabolism. Their effects are discussed in detail in Kabata-Pendias, (1992) and Nagajyoti et al., (2010). Nonetheless, it is important to understand that the measured trace elements (stated above) in plants around volcanic areas may also be of crustal origin and their combined effect with environmental conditions and atmospheric pollutants may exacerbate their effects on plants (Bussotti et al., 1997).

1.2. Geothermal energy development in Kenya

Geothermal energy resources in Kenya are spread over 14 prospective sites within the Rift valley (Figure 1.3) at an estimated potential of between 7,000 – 10,000 MWe (Republic of Kenya, 2013).

The history of geothermal development in Kenya began with exploration at Olkaria in the late 1950s until 1981 - 1984 when production started and the first 45 MWe Olkaria I power plant was constructed. Drilling works continued and additional power plants were later established in stages: 2003 - 70 MWe Olkaria II, 2010 - 35 MWe Olkaria II expansion, 2015 – 140 MWe Olkaria I expansion and 2015 – 140 MWe Olkaria IV (Saitet and Muchemi, 2015). Additionally, 55.6 MWe geothermal power is generated at wellheads in Olkaria (Saitet and Muchemi, 2015) as a temporary strategy to make use of idle steam directly from individual wells for earlier power generation prior to construction of the main power plants. Currently, 632.1 MWe in total of geothermal power is installed at Olkaria (including Oserian flower farms 629.6 MWe) and Eburru (2.5 MWe) geothermal fields in Kenya (Omenda et al., 2014; Kenya Power Limited, 2015; Ormat unpublished data, 2016). For direct use, opportunities are yet to be fully explored, however 22.4 MWth is used for green house heating at Oserian flower farms in Naivasha (near Olkaria), swimming pools at Lake Bogoria (North Rift) and the Blue Lagoon at Olkaria (Lagat, 2010; Omenda et al., 2014).

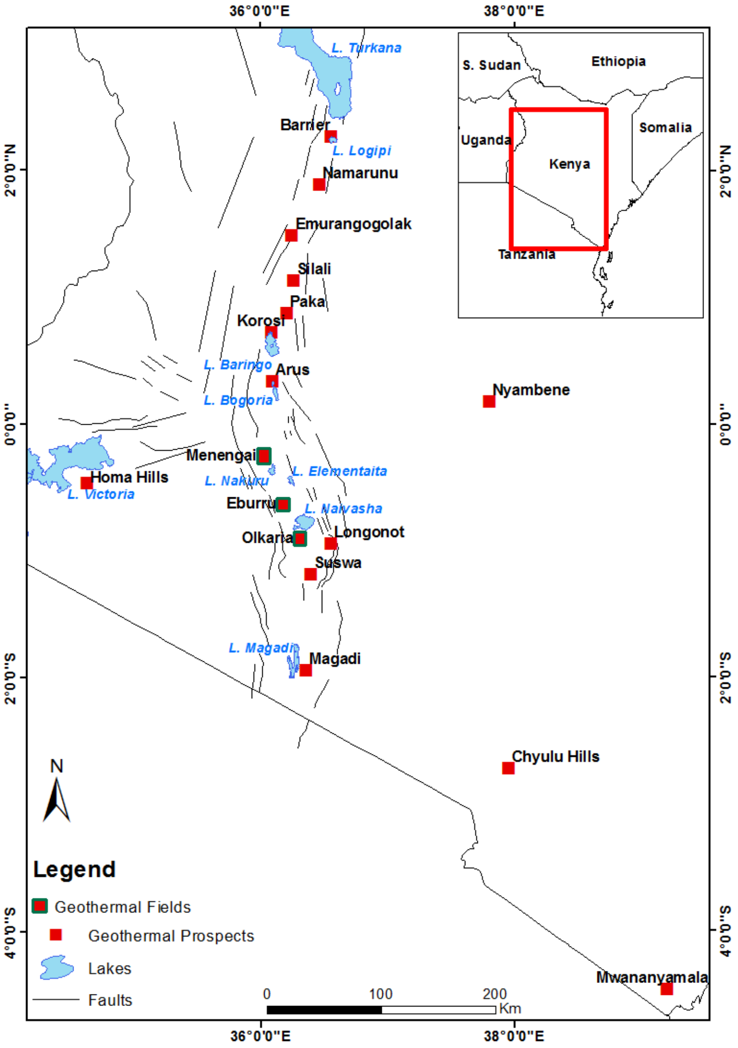

Figure 1.3. Location of geothermal areas in Kenya.

As a signatory to the Kyoto protocol and in efforts to promote a low carbon economy, the government of Kenya is putting emphasis toward the development of clean, indigenous and sustainable energy sources to meet the ever increasing and urgent power demand (Republic of Kenya, 2007, 2015). The Governments *Vision 2030* (Republic of Kenya, 2007) foresees the country to be prosperous and newly industrialised with a medium income and a high quality of life by the year 2030. The availability of adequate, reliable, and affordable electricity is a key factor in attaining this goal. Since geothermal power is environmentally benign and reliable as baseload over hydropower (affected by varying hydrology and climate) and thermal power sources that dominate the current installed electric capacity in Kenya (Omenda et al., 2014), more focus and resources are now dedicated to accelerating the deployment of geothermal energy sources. This is evident from the ambitious expansion plans set up by the government, of providing up to 1600 MWe from geothermal resources in the near future (Republic of Kenya, 2013; Omenda et al., 2014). To fast track this, several intensive geothermal development project plans and works are currently underway at different geothermal fields. The status as of now is that: production drilling is on-going at Olkaria and Menengai (100 km north of Olkaria) geothermal fields while for the northern prospects (Baringo - Silali block) surface exploration studies are finalised and exploration drilling is expected to commence immediately after completion of the ongoing infrastructural works. Likewise, for the southern geothermal prospects, exploratory works are advancing at Longonot and Suswa areas: two exploration wells have been drilled at Longonot while at Suswa surface exploratory works are complete and ground breaking preparations are on course for commencement of infrastructural development. In general, geothermal power is expected to contribute significantly to the total energy mix in Kenya by providing 5500 MWe by the year 2030 (Republic of Kenya, 2015). This contribution will be meaningful in provision of the much needed power for the population in fulfilment of Kenya’s energy needs toward socioeconomic and sustainable development.

1.3. Geothermal energy development in Iceland

Iceland is located on the Mid-Atlantic ridge and is thus well endowed with a huge geothermal potential. For a long time, the resources have been extensively harnessed for power generation and direct use; particularly heating (homes and greenhouses), swimming, spas and other industries (Ragnarsson, 2015). In the country, the resources are mapped based on temperatures at 1 km depth, either as high (>200 °C) or low (<150 °C) temperature geothermal resources (Ragnarsson, 2015). High temperature resources occur within the active volcanic belts of the country and low temperature resources in quaternary and tertiary formations, Figure 1.4 (Bodvarsson, 1961; Arnórsson, 1995).

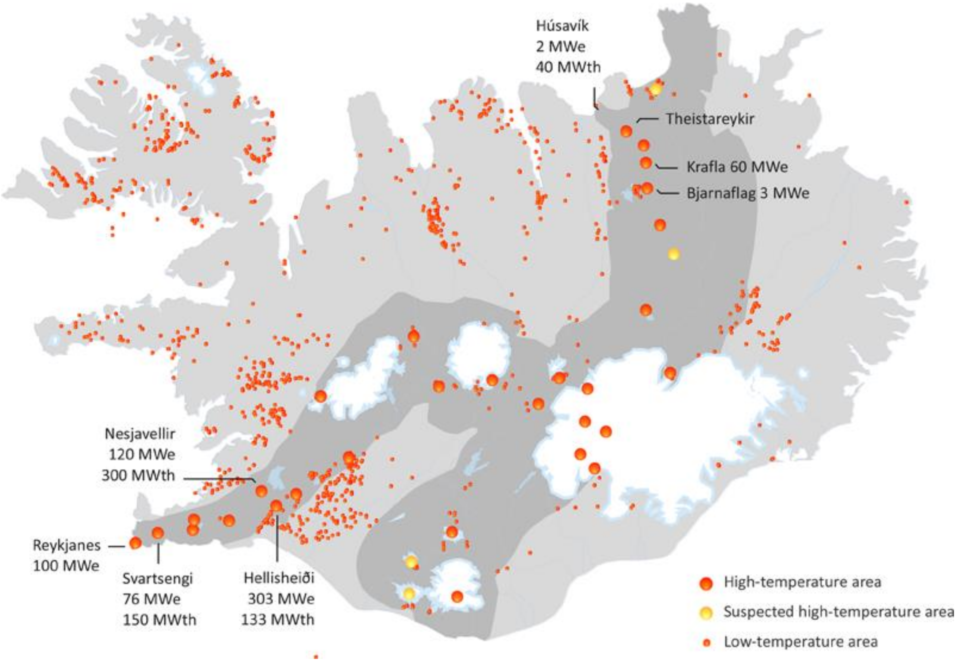

Figure 1.4. Location of geothermal areas in Iceland (Source: Iceland Geosurvey (ISOR), 2016)

Development of geothermal energy for electricity production in Iceland started in 1970 and has rapidly increased over the years to present (Ragnarsson, 2015). An overview of the individual power plants and how generation has developed between the periods, 1970 – present is well presented in Ragnarsson, (2015). However, before 1970, geothermal resources were still used for direct use and in particular district heating for a significant part of the population. To date, 663 MWe is installed for electricity generation from seven power plants and 2,040 MWth for direct uses (Lund and Boyd, 2015; Ragnarsson, 2015).

As a policy by the government of Iceland to increase the utilisation of renewable energy sources in Iceland for all industrial uses to spur economic development, development of geothermal and hydropower energy sources has been increasing over the years. About 86% of the primary energy supply in Iceland currently stems from indigenous renewable energy sources with geothermal accounting for 68% and hydro power 18% (Ragnarsson, 2015). Because of the vast amount of geothermal resources in the country, there is focus to increase development of these resources for more energy production. A 12% increase in electricity production from geothermal resources is targeted by year 2020 (Ragnarsson, 2015).

1.4. Knowledge gaps and aims of this research

As outlined above, there is an urgent global need to replace fossil fuels with renewable energy sources for sustainable development. Geothermal energy is a relatively clean and reliable option for countries endowed with the resources. For Kenya and Iceland, which are the main focus of this research, there are clear plans to increase the current energy supply with significant contributions from geothermal resources to satisfy economic needs. However, awareness on the ecosystem effects of these projects with regard to geothermal power plant emissions is limited. Moreover, few studies have been performed to assess these effects. Due to limited knowledge, there are no policy guidelines that address the control of these emissions into the ecosystems besides those relevant to Occupational Health and Safety. Yet the deposition and accumulation of geothermally emitted elements in different ecosystem components, such as plants, has been revealed. These elements are well known to cause phytotoxic stress and plant damages at elevated levels. As ecosystems are a complex interaction of biotic and abiotic components, an effect on one component can affect others directly or indirectly and may in the long run cause large and irreversible damages e.g. species loss. In addition, the relationships between geothermal power plant emissions and plant responses in ecosystems are also still unclear from the few existing studies. Our understanding of the effects is rudimentary, particularly at different element exposure limits.

Some plant damages have been reported around geothermal power plants. For instance, in 2008, *Racomitrium lanuginosum* moss damages were observed around Hellisheidi and Svartsengi geothermal power plants in Iceland (Natturufraedistofnun Islands unpublished report, 2008) (Figure 1.5). However, the causes of the damages are not known and were speculated to have arisen from excess sulphur pollution originating from the nearby geothermal power plants (Natturufraedistofnun Islands unpublished report, 2008). A year after the dying mosses were observed (2009), moss samples were collected around geothermal power plants for sulphur and trace element analysis. Higher element concentrations were found in moss tissues closer to the power plants than further away suggesting element deposition from the nearby power plants through emissions (Bragason and Yngvadóttir, 2009). However, the relationships between the element concentrations in the moss tissues and moss growth, physiology and damages had not been studied. Similarly, in Kenya around the Olkaria I power plant, leaves of plants in the immediate surroundings show some visible injuries characteristic of yellow leaves, a symptom that may be associated to chlorosis due to excess sulphur or boron, leaf marginal necrosis and brown leaves that may be indicative of early senescence (see some damage illustrations in Figure 1.5). Again, it is not known whether these damages are associated to the power plant emissions or not.

Figure 1.5. A section of plant damages around geothermal power plants in: Kenya for Tarchonanthus camphoratus *(left) and Iceland for* Racomitrium lanuginosum *moss (right) (Photos: T. Mutia, 2014 and S.H. Magnússon, 2013).*

To ensure sustainable development, this study is of importance to assess potential ecosystem effects that relate to geothermal power plant emissions. The overall aim of this study was to assess the impacts of geothermal power plant emissions on plants in two contrasting bio-climatic zones i.e. the semi-arid Kenya and the subarctic Iceland. The dominant plants in each area were identified and used as ecological indicators. The specific objectives aimed at:

- Surveying whether geothermally emitted elements, i.e. sulphur (from H_2S), and trace elements arsenic, boron, antimony and mercury, deposit and accumulate in plants and soils around geothermal power plants with consequences for plant health (assessed as plant growth related traits, damage and physiology).
- Experimentally evaluating the effects of one of the major components of geothermal emissions, H_2S gas, on plants by assessing sulphur concentration and accumulation in plants and plant health responses.

The objectives are addressed in a collection of three papers: first specific objective in **Papers I** and **II** and second specific objective in **Paper III**, which form the next chapters of this thesis.

2. Methods

2.1. Study areas

In Kenya, this research (**Paper I**) was carried out within the Olkaria geothermal field (204 km^2, Omenda, 1998) in which five geothermal power plants are situated; Olkaria I, II, III, and the recent Olkaria I additional units IV and V, and Olkaria IV power plants (Omenda et al., 2014).

The surroundings of Olkaria I (3 units · 15 MWe) and II (3 units · 35 MWe) geothermal power plants (South Rift) up to 4 km away and in the prevailing wind direction (upwind and downwind) formed the study area in Kenya (Figure 2.1). The power plants are owned and operated by the Kenya Electricity Generating Company Limited (KenGen) and are located within the Hells Gate National Park (HGNP), 120 km northwest of Nairobi.

The HGNP is a small park covering 68.25 km^2 in area with a wide variety of wildlife and striking scenery. It was gazetted in 1984 as a National Park and placed under the management of Kenya Wildlife Service (KWS), immediately after construction of the second unit of Olkaria I power plant in 1983. The first unit of Olkaria I had been commissioned earlier in 1981 while its last unit was established in 1985. The Olkaria II power plants were established in 2003 (units 1 and 2) and 2010 (unit 3). To foster a harmonious coexistence between the wildlife and geothermal operations in the area, a Memorandum of Understanding (MoU) between KenGen and KWS was prepared and adopted. The MoU emphasizes on compliance and accountability to the best environmental monitoring practices and wildlife conservation with regard to geothermal operations as stated in the project’s Environmental Impact Assessment and Audit (EIA/A) study reports. Quarterly environmental study reports and meetings covering all geothermal activities from drilling to power plant operations in the area are regularly prepared by KenGen in consult with KWS (HGNP) for collective monitoring purposes. The topography is characterized by volcanic features, mostly steep sided rhyolite and pumice domes, fault scarps, fractures, active thermal manifestations and the Ol Njorowa Gorge cutting across an inferred caldera (Omenda, 1998). Vegetation in the area is rich and diverse, mostly dominated by the shrub *Tarchonanthus camphoratus* L., see Barasa et al., (2012) and **Paper I** for detailed information.

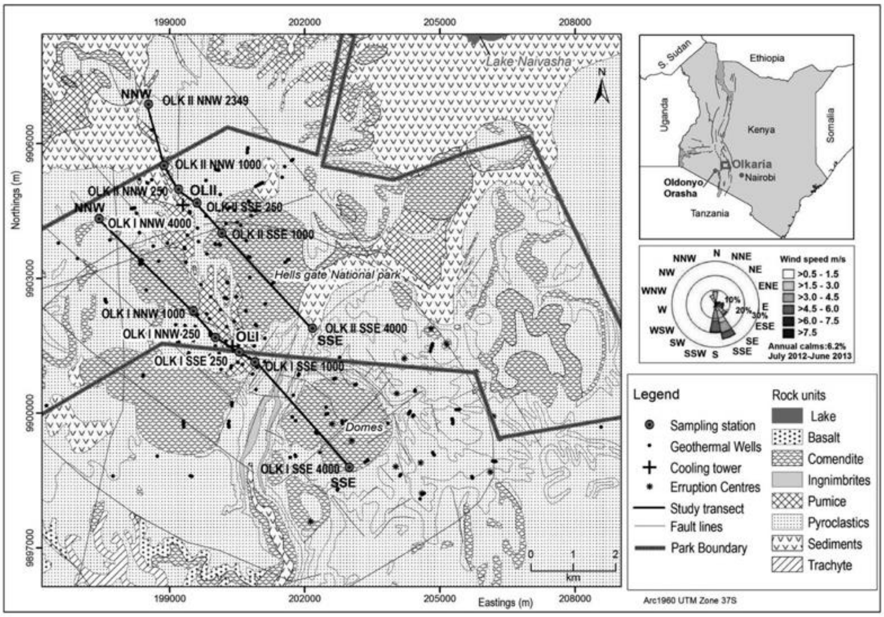

Figure 2.1. Location of Olkaria geothermal area study sites and sampling stations.

In Iceland, the research (**Paper II**) was carried out within the Hengill geothermal area (area 110 - 115 km^2, Franzson et al., 2010) where two geothermal power plants are located, the Hellisheidi and Nesjavellir geothermal power plants (Figure 2.2). Similar to the Kenya study, the surroundings of each power plant, up to 4 km upwind and downwind, formed the study area. The two power plants are owned and operated by a power company, Reykjavik Energy (Orkuveita Reykjavíkur), and are located southeast of Reykjavik, 25 km away for Hellisheidi and 38 km for Nesjavellir. The Nesjavellir geothermal power plant is older and with a lower installed capacity than Hellisheidi power plant. It was established between the years 1998 – 2005 and has a total installed capacity of 120 MWe (4 units). Hellisheidi geothermal power plant, on the other hand, was established at later years, 2006 – 2010, and has in total an installed capacity of 303 MWe (9 units). The Hengill topography is quite variable and consists of volcanic features (Björnsson et al., 1986) mainly, lava fields, eruptive fissures, faults, fractures, volcanic ridges, basaltic volcanic rocks and surface geothermal manifestations such as fumaroles (Foulger and Toomey, 1989). Extensive thick carpets of moss heaths dominate the vegetation, a description of vegetation can be found in Helgadóttir, et al., (2013) and Elmarsdóttir et al., (2015). The area in which the power plants are located and up to the extent of the study area is uninhabited by people.

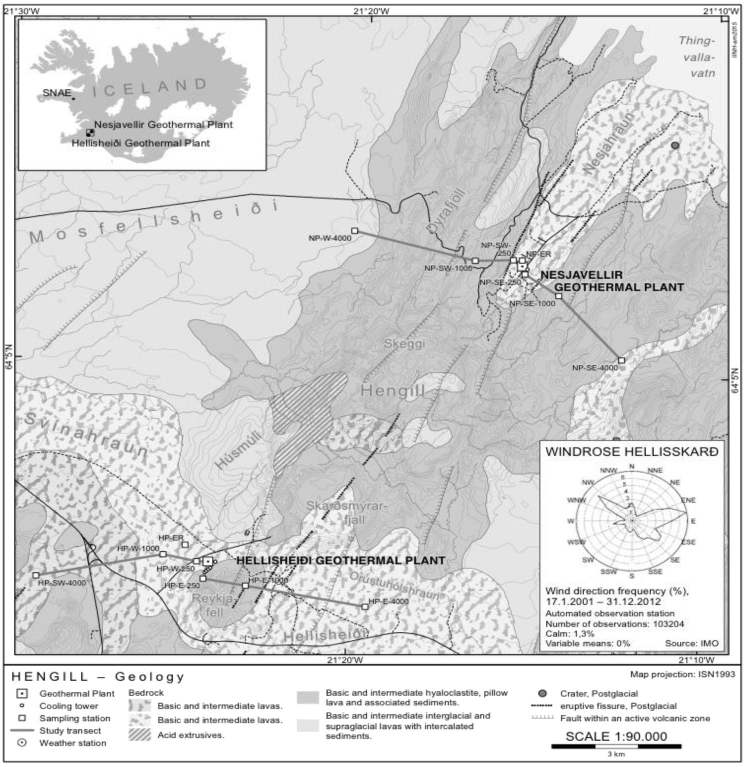

Figure 2.2. Location of the Hengill geothermal area study sites and sampling stations.

2.2. Study species

In Kenya, the shrub *Tarchonanthus camphoratus* L. was chosen as a bio-indicator and study species due to its dominance within the area. The species is quite common in semi-arid Africa and Arabia and often grows between 1500 m - 2200 m above sea level in areas with 500 - 1000 mm annual rainfall. It is a semi-deciduous shrub that grows large and dense when alone, and is often associated with *Acacia* spp. Its leaves are narrow, green-grey on the upper side and pale grey, felted with conspicuous venation on the lower side. *T. camphoratus,* is generally unpalatable to wildlife, with the exception of extreme drought when cows, giraffes, impalas and springboks browse its leaves and shoots. Traditionally, it has many medicinal applications with leaf infusions and tinctures used for, among others, indigestion, heartburn, coughs, stomach upsets, headache, toothache, asthma, bronchitis and inflammation (Orwa et al., 2009). In terms of ecosystem services, the shrub/tree is useful in protection against soil erosion, enhancement of soil fertility (through litter falls) and land reclamation due to its resistance to drought, fire and wind.

Around the Hengill area in Iceland, the moss *Racomitrium lanuginosum* (Hedw.) Brid. dominates in the vegetation. It was chosen as a study and bio-indicator species. *R. lanuginosum* is often found growing on exposed rock and boulder scree or lava fields forming a continuous ‘carpet’, that may grow between 20 - 40 cm in thickness (Bjarnason, 1991; Jónsdóttir, 1991; Jónsdóttir et al. 1995). It is commonly abundant in high northerly latitudes i.e. boreal and arctic regions, and in alpine regions of temperate and sub-tropical biomes (Tallis, 1964). The species has an elongated main stem with a variable number of lateral primary and irregularly branched shoots (Tallis, 1959a, 1959b) and the leaves usually have a long hyaline hair point (Nyholm, 1998). Its growth is rather slow, for example a mean cumulative growth of 15.8 mm during two years (July 1990 - 1992) was measured in a study within the Thingvellir national park in Iceland (Jónsdóttir et al., 1995). Like other mosses, the species plays a significant role in ecosystems, especially in the Arctic where mosses are ubiquitous components. Mosses strongly influence nutrient, carbon and water cycling in the plant-soil interface, significantly regulating ecosystem functions (Turetsky et al., 2012). Among other roles, some species are key in succession as they are the early colonizers of a disturbed site (Bjarnason, 1991; Turetsky et al., 2010, 2012). In recent years, mosses have become increasingly important in monitoring ecosystem health in relation to atmospheric pollution (Berg and Steinnes, 1997; Loppi and Bonini, 2000; Bargagli et al., 2002; Harmens et al., 2015) due to their lack of roots and interception of nutritional requirements from the atmosphere. Changes in their growth, physiology, distribution and tissue element concentrations (Magnússon and Thomas, 2007; Magnússon, 2013) serve as an early warning signal of serious effects of atmospheric pollution.

2.3. Field surveys

The first and second papers discuss and analyse the patterns of sulphur, arsenic, boron, antimony and mercury concentrations in plants and soil around geothermal power plants in Kenya (**Paper I**) and Iceland (**Paper II**), and some growth traits of the dominant plants in each area.

In both study areas, the survey was based on a nested design. Two line transects from each power plant were established along the prevailing wind direction, upwind and downwind, at increasing distances away from the power plants (Figures 2.1, 2.2) and away from any visible geothermal manifestations. The distances, 250 m, 1000 m and 4000 m were chosen as sampling stations, within which sub-transects were marked for plant and soils measurements and sampling. Selected plant variables and growth related traits i.e. abundance, stem height, number of stems, main stem circumference and leaf growth (number of leaves) and leaf damage (visible injury) for the shrub and moss shoot growth (shoot length increase, shoot turnover and biomass increase), physiology (chlorophyll concentrations) and visible moss shoot damages were assessed for plant growth and health evaluations. For leaf/shoot damages, plants leaves/shoots colourations were visually assessed and categorised into three: A) healthy green, B) yellow and C) brown dead. At the end of plant measurements, leaf samples from the shrub at the sampling stations were obtained and grouped according to the different leaf categories for determination of sulphur, arsenic, boron, antimony and mercury concentrations. For moss, samples were obtained from the sampling stations and damage assessed at sampling stations. Samples were analysed for sulphur, arsenic, boron, antimony and mercury concentrations.

A reference area well outside the range of the study areas and geothermal power plant element depositions was included in each study area for comparison. Plant leaves, moss shoots and upper soil layers were sampled in each case for analysis of sulphur, arsenic, boron, antimony and mercury concentrations. Additionally, soil characteristics: pH, moisture, total carbon and nitrogen, were also measured as co-variables to account for variation in the measured response variables that might have been caused by these environmental factors. All sample preparations and processing for chemical analyses were conducted at internationally accredited laboratories: Kenya Bureau of Standards (KEBS) for the Kenya study and ALS Scandinavia labs in Luleå, Sweden for the Iceland study.

2.4. Experiments

Following the surveys, two experiments were performed, one in Kenya and another in Iceland, using the dominating plants in each geothermal area to assess the experimental effects of H_2S deposition on plants (**Paper III**). Eight month old seedlings of the shrub *T. camphoratus* and extracted moss mats of *R. lanuginosum* that had been growing in areas well out of range of geothermal activity were used.

In Kenya, the experiment was set-up and performed outdoors at an open ground area in Nakuru and at indoor growth chambers in Iceland (See experimental set-ups in Figure 2.3).

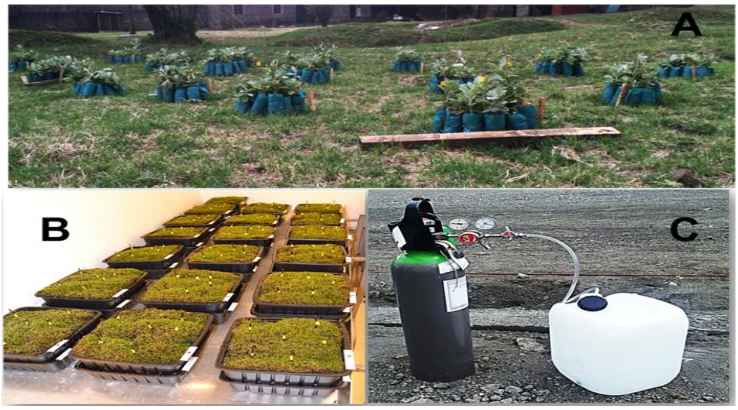

Figure 2.3. Experimental layout for A) Tarchonathus camphoratus *seedlings, B)* Racomitrium lanuginosum *moss and C) preparation of* H_2S *stock solution for the different treatments.*

Solutions of H_2S gas dissolved in distilled water were prepared at 0, µg/L, 30 µg/L, 100 µg/L and 300 µg/L concentrations and used as experimental treatments. These concentrations correspond to air saturated water with H_2S concentrations in air of 0 ppm, 10.96 ppm, 36.52 ppm, and 109.57 ppm, respectively (using a Henry's law constant of 0.001 mol/(L*atm), (Sander, 2015)). A control solution, prepared from distilled water (0 µg/L) was included for comparison.

A nested design was adopted for each experiment with four units randomly assigned to each treatment and multiple measurements conducted per unit. Treatment applications were done four times per week. The experiments were conducted for a period of 6.5 weeks in Kenya and 13 weeks in Iceland.

Plant variables related to growth, i.e. stem height, change in number of stems and change in number of healthy leaves (A) healthy green), moss shoot length increase and biomass, and foliar damages i.e. proportions of damaged leaves (based on the two categories: B) yellow and C) brown dead), were measured and assessed at the beginning and end of the experiments. Sulphur and chlorophyll concentration in plant tissues were determined in plant leaves/moss shoots at the end of the experiment i.e. at the ALS Scandinavia labs in Luleå, Sweden for sulphur analysis and Institute of Freshwater Fisheries in Iceland for chlorophyll determination.

2.5. Data analyses

Due to the nested design of most data for the field surveys and experiments, linear mixed effects models (LMM) were fitted whenever possible. For the field survey data, i.e. concentrations of sulphur, arsenic, boron, antimony and mercury in plant leaves and soil, and the plant characteristics and growth related variables, LMMs were fitted against the predictors (fixed factors): distance (250 m, 1000 m and 4000 m), direction (upwind or downwind) and location (identity of the power plant in each study area). Sampling stations were included as random factors. In cases of pooled data, simple linear models were run with the same predictor variables as the LMMs. Soil co-variates were included to account for un-explained variations in the model whenever possible.

The experimental data was analysed in a similar way as the survey data except with different predictors (fixed factors), i.e. the different H_2S treatment concentrations (0 µg/L, 30 µg/L, 100 µg/L and 300 µg/L). The plant units were included as random factors. All interpretation was based on the effect size of the treatments as a whole and comparisons between the different treatments levels.

The models were run in R 3.2.2 (R Development Team, 2010) using the functions, lmer in the lme4 packages (Bates et al., 2014) for the LMM and lm in the MASS package in R (Ripley et al., 2015) for linear models. See details in **Papers I, II** and **III**.

3. Results and Discussions

3.1 Field surveys

Different distances from geothermal power plants along the prevailing wind direction are key in assessment of gas emission dispersion for air quality monitoring studies (Wetangula, 2011; Ólafsdóttir et al., 2014), and the effects of these gases on ecosystems (Bargagli et al., 1997; Bussotti et al., 2003). Element concentrations in plants and soils in relation to these distances and wind direction (upwind and downwind) will thus provide an indication of the deposition and accumulation patterns of these elements on ecosystems.

The findings of the field surveys (**Paper I** and **II)** revealed that plants and soils around geothermal power plants are enriched with sulphur and trace elements. However, there were no clear patterns for arsenic, boron, antimony and mercury concentrations in plants and soils in relation to distance and direction from the power plants. Only sulphur in plant tissues and soils at the geothermal areas (Kenya and Iceland) showed patterns of decreasing concentrations with increasing distance (Figures 3.1, 3.2), implying atmospheric deposition of sulphur from the geothermal power plants. The patterns were stronger in plant tissues than soils, indicating that plants may have been immediate receptors of geothermally emitted sulphur rather than soils. Wind direction, on the other hand, did not show clear indications of its effects on element distribution at both study areas. Further, element deposition in relation to location did not show differences between the two power plants in Kenya, while in Iceland, there were generally higher element concentrations in plants and soils around Hellisheidi geothermal power plant than Nesjavellir. There are several reasons that may have been the cause of these variations. In Kenya, a proper evaluation of the effect of the wind and location may have been difficult due to the closeness of the two power plants (Olkaria I and II, see Figure 2.1) such that the effects of wind on the surroundings of one power plant would affect the effects in the vicinity of the other power plant. For Iceland however, the variations may have been attributed to the effects of topography on the prevailing wind between the two power plants, since the Nesjavellir geothermal power plant is located in a valley and is probably more sheltered from the effect of the prevailing east winds than Hellisheidi, which lies exposed at a higher elevation. This means that around Hellisheidi there may have been much more efficient element distribution than at Nesjavellir. The element concentration variations may have also been caused by other sources of these elements, since the study areas are volcanic/active geothermal areas. Volcanic areas are known to be quite abundant in these elements, especially in the soils, these elements are usually of crustal or magmatic gas origin (Bussotti et al., 1997; Davies, 2008). Geothermal fluids from steam sprays during geothermal well testing (on-going at the study areas), natural hot springs and fumaroles also contain these elements and may deposit them within the area.

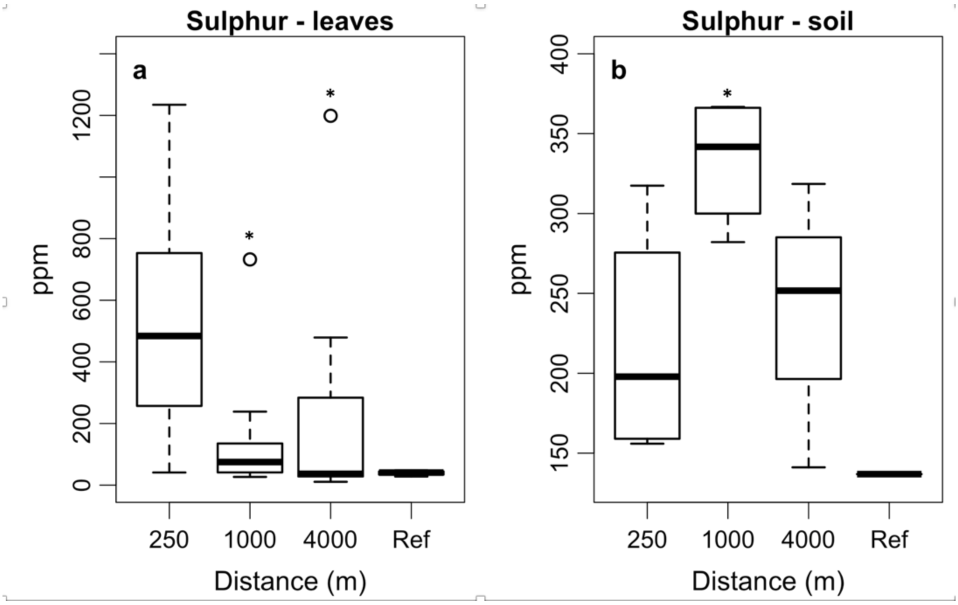

Figure 3.1. Concentrations of sulphur in T. camphoratus *leaves and soil in relation to the Olkaria I and II geothermal power plants in Kenya. Ref indicates the reference area. n = 3 leaves per station, n = 1 soil per station. * indicates significant differences (p<0.05) for sulphur concentrations in leaves and soil the different distances compared to 250 m. The reference area values were not included in statistical models for the comparisons.*

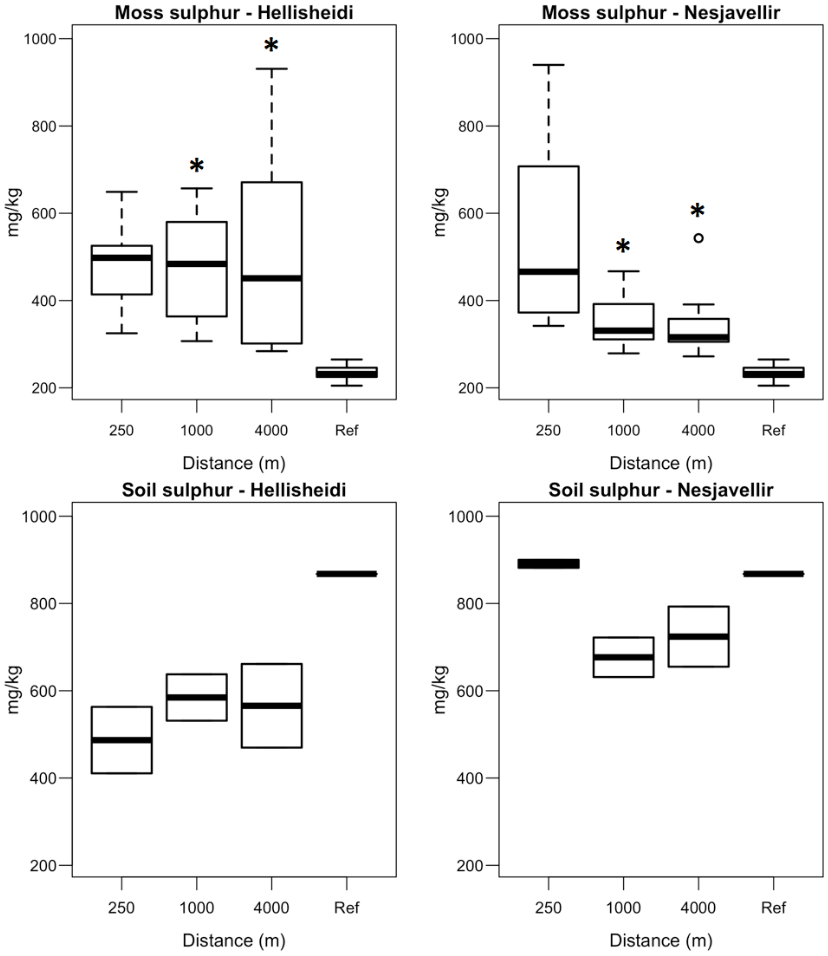

Figure 3.2. Concentrations of sulphur in the R. lanuginosum *moss and soil in relation to the Hengill geothermal power plants in Iceland. n = 10 shoots per station, n = 1 soil per station. * indicates significant differences ($p<0.05$) for sulphur concentrations in moss and soil at the different distances compared to 250 m. The reference area concentrations were not included in the statistical models for comparisons.*

The general patterns of plant health (assessed in the form of growth, foliar damage and other plant characteristics) did not also show clear patterns with the predictors (distance, direction and location) around the power plants, except for the Iceland study. In Iceland, moss showed faster growth and more positive health responses at Hellisheidi than Nesjavellir, while at Nesjavellir, plant growth was slower closer to the power plant than further away. This may partly be related to the effect of wind and topography as discussed previously.

For Kenya, sulphur concentrations were generally higher in the plants than soils around the power plants, while the opposite was true for moss versus soils in Iceland. One reason for the higher sulphur enrichment in the shrub leaves than mosses when compared to soils is probably related to the different plant mechanisms in relation to nutrient absorption; *T. camphoratus* acquires more nutrients (sulphur) from both the soil through roots and from air through leaves than *R. lanuginosum* which obtains all its nutrients (sulphur) from the air. However, at the reference areas in both Kenya and Iceland, soils had much higher sulphur concentrations than plants, which is expected in the absence of atmospheric pollutants. Further, sulphur concentrations were at higher levels in both plant tissue and soils in Iceland than Kenya**.** There could be several interacting reasons for this discrepancy between the two ecosystems; The amount of H_2S emissions from the Hengill geothermal power plants is 83% higher than that of the Olkaria power plants, see details in **Papers I** and **II**. It therefore follows that there would be much more sulphur deposition and accumulation in the Hengill (Iceland) terrestrial ecosystems than Olkaria (Kenya). In addition, the different soil processes in the two ecosystems may provide an explanation for the higher sulphur concentrations in the Hengill soils than in Olkaria soils: the shrub leaves are most likely more easily decomposed than the recalcitrant moss shoots. Therefore, decomposition and mineralisation rates are in general much faster in semi-arid Kenya ecosystems than the much colder ecosystems of Iceland. The slow decomposition means that the elements are likely to be much more enriched in the soils of Iceland than in Kenya, suggesting that the impact of geothermal emission (H_2S gas in this case) on ecosystems may be greater in Iceland than Kenya.

Overall, in comparison to the reference areas, the geothermal areas in Kenya and Iceland showed elevated element enrichment in plants and soils, except for sulphur in soil at the reference area in Iceland which was higher than at the Hengill geothermal area. Additionally, plant growth and overall health was greater at the reference areas than the geothermal areas. While the study findings show elevated trace element concentrations in plants and soils in the geothermal fields, this element enrichment cannot be attributed only to the power plants. Their patterns in relation to the power plants did not suggest the power plants as a significant factor in their distribution around the two contrasting ecosystems. Patterns of the observed plant health effects in relation to the power plants may therefore be related to excess sulphur enrichment in combination with other factors beyond the scope of this study. These findings agree with other studies especially in the Mediterranean that reveal high sulphur depositions in plants and soils closer to geothermal power plants than further away with some consequences for plant health (Bargagli et al., 1997; Bussotti et al., 1997; Bussotti et al., 2003).

3.2 Experiments

The field surveys revealed a connection between geothermal power plants and sulphur enrichment in plants and soils around the geothermal areas in Kenya and Iceland, a concern due to the potentially phyto-toxic levels of sulphur at elevated levels. However, **Paper III** findings showed no clear evidence of elevated sulphur concentrations and accumulation from experimental wet hydrogen sulphide deposition (0 µg/L, 30 µg/L, 100 µg/L and 300 µg/L) on dominant plants around geothermal power plants in Kenya (*T. camphoratus* seedlings) and Iceland (*R. lanuginosum* moss), Figure 3.3. In spite of this, both plants showed responses to the different wet H_2S treatment exposures.

The growth (increase in shoot length) of *R. lanuginosum* decreased in response to high H_2S exposure levels (300 µg/L) while there was an increase in shrub stem height growth at intermediate concentrations of H_2S (30 µg/L), Figure 3.4. Such a decrease in *R. lanuginosum* growth at high H_2S exposure levels was expected. Mosses are highly sensitive to atmospheric pollutants and elevated levels of such components have been reported to affect their growth, e.g. a similar decrease in growth has been reported for *Sphagnum* moss species in response to high sulphur exposure levels (Ferguson et al., 1978; 1980). For *T. camphoratus*, the low H_2S exposure levels seemed to have had a fertilising effect on the seedlings by stimulating shoot height. This compares with the findings of a H_2S fumigation experiment in Thompson and Kats, (1978) that showed significantly stimulated growth of lettuce, sugar beets and alfalfa at low H_2S exposure levels (30 ppb). The other plant variables for *T.*

camphoratus and *R. lanuginosum* were however not affected by the treatments over the short exposure periods.

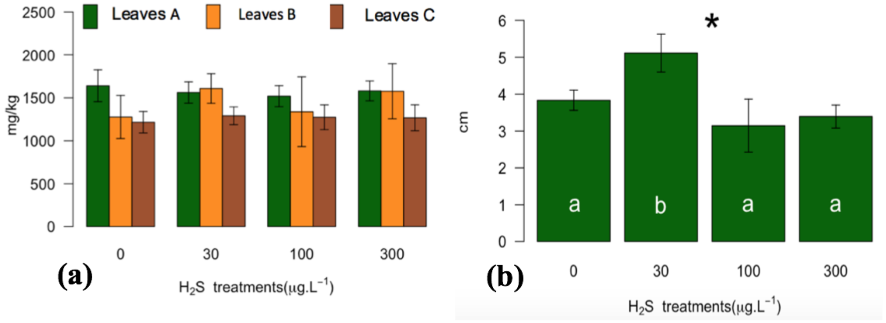

Figure 3.3. Various responses of T. camphoratus *to H_2S treatments (application of 0 µg/L, 30 µg/L, 100 µg/L and 300 µg/L):* ***(a)****. Concentrations of sulphur in different categories of* T. camphoratus *leaves and* ***(b)*** *Stem height increase, after 6.5 weeks in an outdoor experiment (mean ± SE, n = 4). Concentrations in the leaves are assigned to different damage categories based on visual assessment: healthy green leaves (leaves A), yellow leaves (leaves B) and dead brown leaves (leaves C). Asterisks (*) indicate significant effect of treatment ($p<0.05$) and smaller case letters show differences between treatments.*

In general, the leaves of *T. camphoratus* seedlings showed 75% more sulphur concentrations than the *R. lanuginosum* moss shoots. This is opposite to the findings of the field surveys (**Paper I** and **II**) where higher element concentration was found in the shoots of *R. lanuginosum* (in Iceland) than leaves of *T. camphoratus* (in Kenya). Besides the unseen effect of the different environmental/ experimental conditions for the two plants and in the absence of pollution, these sulphur concentration variations can be explained by the different plant mechanisms in relation to nutrient absorption; *T. camphoratus* acquires more nutrients (sulphur) from both the soil through roots and from air through leaves than *R. lanuginosum* which obtains all its nutrients (sulphur) from the air.

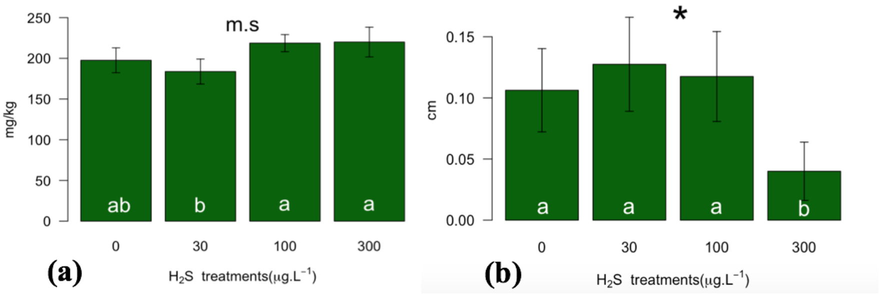

Figure 3.4. Various responses of R. lanuginosum *to H_2S treatments (application of 0 µg/L, 30 µg/L, 100 µg/L and 300 µg/L):* **(a)** *Concentrations of sulphur in* R. lanuginosum *moss and* ***(b)*** *Shoot length increase, after 13 weeks in growth chambers (mean ± SE, n = 4). 'm.s' indicates a marginally significant effect of treatment (p=0.06)). Asterix (*) indicates significant effect of treatment (p<0.05) and smaller case letters show differences between treatments.*

Overall, there were no strong responses of either plant to the treatment exposures over the short experimental period.

“The sky´s no limit” ~ Claude Munyao Mutia

4. Conclusions and future perspectives

In the field surveys presented in **Papers I** and **II**, there is evidence that sulphur (in the form of hydrogen sulphide gas) emitted from the geothermal power plants in Kenya (Olkaria) and Iceland (Hengill) deposits and accumulates in terrestrial ecosystems in the vicinity of the power plants. However, the trace element concentrations: arsenic, boron, antimony and mercury, do not show such consistent and similar patterns; according to data from this study, their levels in terrestrial ecosystems in the Kenya and Iceland geothermal study areas cannot be attributed to the geothermal power plants. Further, because trace elements are not monitored in the emissions and their concentrations are not known, it is difficult to conclude that the measured trace element concentrations in the plants and soils may to some extent have been influenced by the power plants. This conclusion is slightly distinct from the Meditteranean studies (Baldi, 1988; Bargagli et al., 1997; Bussotti et al., 1997; Bacci et al., 2000) that report high sulphur and trace element concentrations in plants and soils near geothermal power plants with patterns indicating potential enrichment from the power plant emissions. The Meditteranean geothermal systems are different. In terms of aquifer fluid phases, the Mediterranean geothermal systems are mainly dry steam dominated (Bertini et al., 2006), different from the liquid - vapour dominated geothermal systems in Kenya (Koech, 2014) and Iceland (Scott et al., 2014; Ragnarsson, 2015). The trace elements in the Kenya and Icelandic study context are not nearly as volatile as the H_2S gas and may not have been in very high concentrations in the NCGs of the geothermal power plants in Kenya and Iceland.

The plants (bio-indicators) in the field surveys compared to reference areas (**Papers I** and **II**) showed some indications of geothermal power plant effects on plant growth that corresponded to the findings of geothermally enriched sulphur in their tissues and soils. This may suggest that the effects on plants are somehow related to the excess sulphur levels in the plant tissues and soils and may be affecting plant growth. However, the influence of other environmental factors is to be considered, as for example the soil conditions were in most part significant in explaining the variations of the different element concentrations in the plants and soils.

Due to the indications of effects on plant growth noted, further field surveys are recommended for both areas for better assessments of sulphur effects on plants and especially in relation to bio-accumulation in the ecosystem. In the present study, sulphur accumulation levels in *R. lanuginosum* shoots (**Paper II**) were calculated from *R. lanuginosum* concentration data and may not have been an accurate assessment, these estimates were however not evaluated for the Kenya study (**Paper I**) due to limited leaf biomass data. The study design needs to be improved for more accurate assessment of element accumulation in the ecosystem components (with a larger sample size).

Further long term studies are recommended to properly evaluate sulphur accumulation in plants in relation to the geothermal power plants and associated plant growth/health effects, because of the moderate to slow growing nature of the plants: *T. camphoratus* is reported to grow between 600 – 800 mm/year (Orwa et al., 2009) and *R. lanugiosum* up to 5 mm/year (Tallis, 1964; Jónsdóttir et al., 1995). For the Kenya survey, it may even be more interesting to combine the shrub assessments with other additional and sensitive bio-indicators such as lichens and mosses or soil-microbes (Baldi, 1988; Loppi et al., 1998; Bargagli et al., 2002; Zouboulis et al., 2004; Storelli, 2013). Further responses of *T. camphoratus* related to growth and physiology can also be included in the studies, including recruitment of new flowers, leaf area, leaf biomass and photosynthesis.

In an improved study design, the addition of more study transects around the power plants and a large number of replicates could provide clear information on element distribution and effects around the power plant areas to complement our findings. New study transects perpendicular to the main transects of the two field surveys are interesting to explore to get an overall picture of element distribution and plant responses over the entire geothermal area. To strengthen the ecological explanations as to why the elements, accumulate to a greater extent in the soils of Iceland than in Kenya, decomposition studies such as those described in

Keuskamp et al., (2013) are recommended as important additions for future studies across the two contrasting biomes. Overall, regarding differences between the two bio-climatic zones, it can be concluded that the semi-arid ecosystems are less susceptible to the effects of geothermal power plant emissions than subarctic systems due to higher turnover rates of biomass (plant growth, decomposition).

The experiments (**Paper III**) revealed that short-term exposure to 30 µg/L, 100 µg/L and 300 µg/L wet H_2S deposition does not result in increased sulphur concentrations in plants of *T. camphoratus* dominant around geothermal power plants in Kenya. Further, there was no evidence of increased sulphur accumulation with the treatment exposures in both plants. Contrary to our predictions, plant tissues of *T. camphoratus* showed more elevated sulphur concentrations and accumulation than *R. lanuginosum*.

Furthermore, short term exposure to moderate levels of H_2S deposition (30 µg/L (ppb) approximately 10.96 ppm air concentrations) does not result in harm to the two plants. This H_2S level seemed to benefit plant growth in the shrub *T. camphoratus*, and did not reduce *R. lanuginosum* moss growth. However, high exposure concentrations of H_2S depositions (300 µg/L (ppb) – about 109.57 ppm air concentrations) reduced *R. lanuginosum* growth but did not affect *T. camphoratus* growth. The observed effects on plant health within the short duration of the experiment are indicative that if the experiment is conducted for a long duration, stronger and clear responses would be evident. A follow-up experiment over a longer period is thus recommended. It is important because these plants, within their natural set-up, are usually exposed (although not directly) to emissions (dry and wet deposition) over a long period of time, that is as long as the power plant operates. A detailed understanding of the effects on plant health is important for planning of mitigation measures.

In future experiments, the growth conditions for the *R. lanuginosum* can be improved by carrying out the experiments in the field and away from any atmospheric pollution activities rather than in a growth chamber. This is due to the atmospheric sensitivity of mosses, for instance in this experimental study, growth chambers were slightly hotter than normal and may have affected *R. lanuginosum* growth as it is susceptible to drying on exposure to high heat.

Since more ecosystem effects were noted in Iceland than in Kenya, it is recommended that the geothermal power plant developer at Hengill in Iceland fosters emission curbing mechanisms to prevent future effects of sulphur depositions in terrestrial ecosystems. Reykjavík Energy, the power developer at the Hellisheidi geothermal project, is already undertaking trials of a H_2S emission abatement strategy by testing the feasibility of H_2S re-injection back into the earth (Gunnarsson et al., 2013). This project should be fully supported at all levels to ensure sustainable geothermal power development. In addition, monitoring of trace elements from the power plant emissions is highly recommended at the Olkaria and Hengill geothermal power plants, as at present knowledge is lacking on which trace elements are emitted, their concentrations, amounts and fate from geothermal power plants emissions. Monitoring of the trace element levels in emissions is thus advised for mitigation of any likely associated effects.

Overall, these findings serve as a necessary yardstick in advising future geothermal projects. Especially because the target species are also common in some of the other earmarked geothermal fields for development in Kenya and Iceland. For example, the species *T. camphoratus* is also abundant in Suswa and Menengai geothermal areas in Kenya while the moss *R. lanuginosum* abundance is also widespread in most geothermal areas of Iceland. Furthermore, the general findings will advise policy makers, conservationists and the public on the effects of these emissions on ecosystems and the urgent need for development of air quality environmental guidelines related to geothermal power plants. This new knowledge will also increase public awareness on the effects of geothermal power plants on the environment and reduce uncertainties or ambiguities on such projects, an important aspect in increasing social confidence and possibly public acceptance and support of geothermal projects.

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Paper I

Concentrations of sulphur and trace elements in semi-arid soils and plants in relation to geothermal power plants at Olkaria.

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Authors contributions:

- Conceived and designed the study: TMM, TF, ISJ
- Performed fieldwork: TMM
- Analysed the data: TMM, ISJ
- Wrote the paper: TMM, TF, ISJ

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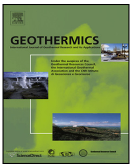

Concentrations of sulphur and trace elements in semi-arid soils and plants in relation to geothermal power plants at Olkaria, Kenya

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Plant growth trait

ABSTRACT

Exploitation of geothermal energy is considered to have minimal ecological impacts. However, this assumption has not been widely studied. We tested the hypothesis that emitted elements from geothermal power plants would be enriched in both plant tissue and soil close to the power plants with consequences for plant health. The concentrations of sulphur, arsenic, boron, antimony and mercury in the soil and leaves of the dominating shrub, *Tarchonanthus camphoratus,* were assayed and associated foliar injury and growth traits assessed at variable distances and directions from two geothermal power plants in Kenya, Olkaria I (operated since 1981) and Olkaria II (since 2003). Sulphur concentration in the leaves was elevated close to the power plants and decreased with increasing distance, implying atmospheric input of sulphur to the ecosystem from the power plants. Similar trends were not detected in soil and with the other elements. Our study design did not support the observed higher degree of leaf injury close to the power plants. Similarly, any association of growth traits with distance or location was not detected. The results were compared with data from a reference site well out of the range of element deposition from the power plants. Overall, the levels of sulphur, arsenic, boron and antimony in leaves of *T. camphoratus* and sulphur, and boron concentration in soil around the Olkaria I and Olkaria II geothermal power plants were higher than at the reference site. Furthermore, the number of healthy leaves per shrub and stem circumference were lower around the power plants than the reference site, while leaf damage and other plant growth traits did not differ. In spite of relatively weak indication of the harmful effects of the geothermal power plants on the dominating shrub species, follow-up experimental studies and studies on more sensitive ecosystem components are recommended to advise existing mitigation measures against chronic exposure from the emitted gases and associated impacts.

1. Introduction

Geothermal energy is listed among those world's renewable energy sources considered to have minimal ecological impacts with a great potential for the future (Bayer et al., 2013; Wong and Tan, 2014). However, a range of non-condensable gases (NCGs) and trace elements typically ranging from less than 0.2% to over 25% weight of steam (Rodríguez, 2014) are emitted from the power plants during the energy conversion process. Some of these components have been reported to deposit in the surrounding ecosystems (Bargagli et al., 1997; Bacci et al., 2000; Paoli and Loppi, 2008), but the consequences are still poorly known. Potentially, they can cause toxicological stress on human beings, plants, and other ecosystem components (Bayer et al., 2013). With increasing utilization of this energy source there is an urgent need for detailed studies on ecological responses to geothermal power plant emissions.

Commonly, the NCG fraction comprises 73–98% w/w carbon dioxide (CO_2), 1–24% w/w hydrogen sulfide (H_2S), 0.02–0.65% w/w methane (CH_4), 0.1–8% w/w hydrogen (H_2), 0.3–16% w/w nitrogen (N_2), 0.1–3% argon (Ar), and traces (<0.001% w/w) of radon, boron, mercury, arsenic, antimony, and ammonia in gaseous and dissolved form (Baldi, 1988; Bargagli et al., 1997; Loppi et al., 1998; Gunerhan, 1999; Loppi, 2001; Bussotti et al., 2003; Rodríguez, 2014). Of these gases, H_2S poses a major concern due to its odour and potential toxicity even at low concentration. The trace elements are also widely

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understood to bio-accumulate in ecosystems causing deleterious consequences (e.g. Kabata-Pendias, 1992).

Our knowledge of the effects of geothermal power plant emission on terrestrial ecosystems is limited, and mainly based on a few studies in the Mediterranean Italy. All these studies indicate increased levels of the emitted elements including sulphur, arsenic, boron, mercury, and antimony in tissues of vascular plants, mosses and epiphytic lichen close to the power plants with an apparent trend of decreased concentrations over increasing distances (Baldi, 1988; Bargagli and Barghigiani, 1991; Panichi and Orlando, 1992; Edner et al., 1993; Ferrara et al., 1994; Loppi and Bargagli, 1996; Bargagli et al., 1997; Loppi et al., 1998; Loppi, 2001; Bussotti et al., 2003). These trends strongly imply atmospheric deposition of sulphur (in H_2S gas form) and trace elements into terrestrial ecosystems from the nearby geothermal power plants.

High concentrations of H_2S gas may have harmful, acute and chronic effects on ecosystems. The H_2S gas molecule is quite unstable in air, and may be oxidized to SO_2 (Kellogg et al., 1972). Mobile sulphur is available to plants primarily in the form of anionic sulphate ($SO_4{}^{2-}$) from the soil or as gaseous SO_2 or H_2S which is readily absorbed and assimilated by leaves (Leustek and Saito, 1999). As an essential macro-nutrient for plant metabolism and growth, both deficiency and excess of sulphur will lead to foliar necrosis, leaf lesions and defoliation. Consequently, the long term effects manifest as reduced plant growth, early senescence and chlorosis (Thompson and Kats, 1978; Varshney et al., 1979; WHO, 2000). According to WHO (2000), SO_2 can also alter plant responses to other environmental stresses often intensifying their impacts.

The effect of H_2S on crop and forest plants was experimentally studied in a greenhouse (Thompson and Kats, 1978). Continuous fumigation with 30–100 ppb H_2S gas stimulated plant growth whilst 300–3000 ppb caused patches of dead cells on leaves (leaf lesions), defoliation and reduced or stunted growth. The effect was more noticeable on fast growing species such as grapes, alfalfa, and lettuce than slow growing species such as buckeye and ponderosa pine. Symptoms similar to those observed during the experimental fumigation may be seen in natural ecosystems, such as forests, close to geothermal power plants, indicating stressed environmental conditions (Bussotti et al., 1997).

Increased concentrations of other emitted elements may also be harmful to plants. In Bussotti et al. (1997), high boron and arsenic concentrations in *Quercus cerris* L. leaves were associated with higher crown defoliation around geothermal power plants in Travale, Southern Tuscany. From the same area, Bussotti et al. (2003) reported widespread leaf damage in *Quercus pubescens* Willd., including necrosis and decreased leaf area, which they related to elevated boron and sulphur concentration of geothermal power plant origin. The predominant role of foliar uptake was suggested by, higher boron and sulphur levels in *Q. pubescens* leaves than in soil close to the power plants. Higher boron and sulphur concentrations in superficial soil layers (0–20 cm) than in deeper layers (20–40 cm), indicated atmospheric deposition as the primary origin of these elements (Bussotti et al., 2003).

Very little is known about accumulations of emitted elements from geothermal power plants in semi-arid terrestrial ecosystems in the tropics and their potential impacts. In Kenya, the production of geothermal energy began three decades ago with the development of geothermal resources at the Olkaria geothermal field in the Great Rift Valley and accounts for 37% gross national electricity production today (Omenda et al., 2014). Due to the reliability and assumed minimal ecological and climate impacts of geothermal power compared to other sources, expansion plans are underway in other geothermal fields to meet the current power demand. So far, only a few studies have addressed the environmental impacts of the power plants, all focusing on trace elements in spent geothermal waters and bioaccumulation in aquatic plants (Simiyu and Tole, 2000; Were, 2007). However, solid knowledge of the environmental impacts on the surrounding terrestrial ecosystems is needed to strengthen existing mitigation measures against pollution and to ensure sustainable development of geothermal power plants in Kenya. This study contributes to that knowledge by investigating the ecosystem accumulation of elements emitted from geothermal power plants in Kenya.

We studied the patterns of sulphur and trace element concentrations in plants and soil around two power plants at the Olkaria field, Olkaria I and Olkaria II, and some growth traits of the shrub *Tarchonanthus camphoratus* L. We chose this species as a bio-indicator due to its widespread distribution and dominance in the vegetation at Olkaria. We hypothesized that the concentration of the elements emitted would be enriched in both plant tissue and the soil around the power plants with consequences for plant health. We expected stronger responses around Olkaria II than Olkaria I, because it is a higher output power plant with a higher emission rate of H_2S and Hg (Table 1). To test the hypothesis, we assessed the soil and leaf chemical compositions at different distances along transects along the prevailing wind direction, a key factor in dispersion of atmospheric contaminants around the power plants (Olafsdottir et al., 2014; Wetang'ula, 2011). Further, we assessed the frequency of different leaf damage categories, and measured growth related morphological traits of the shrub. A reference site well out of range of all geothermal activity was also established for comparison.

2. Materials and methods

2.1. Study area and species

The study area is within the Olkaria geothermal field (area, 204 km^2), situated on the floor of the Great Rift Valley of Kenya (Fig. 1), at an average elevation of 2000 m above sea (Omenda, 1998). The topography is dominated by volcanic features mainly steep sided rhyolite and pumice domes, fault scarps, fractures, and the Ol Njorowa Gorge cutting across a purported buried caldera (KenGen, 2004; Omenda, 1998). Annual rainfall is low recording a mean of 634 mm (2000–2013) with a bi-modal pattern. The average minimum and maximum monthly temperatures ranged from 15.9 to 17.8 °C, and 24.6 to 28.3 °C, respectively, for 2001–2012 (Barasa et al., 2012). Annual predominant wind direction is from south and south–south east (Fig. 1) (KenGen 2013, unpublished; Kollikho and Kubo, 2001). The area is classified as semi-arid due to its porous soils coupled with a high evaporation rate of 1000–1700 mm per year (KenGen, 2004). Soils are of volcanic origin containing a mixture of sands, clays and air fall pyroclastics with pumice. The study focused on the surroundings of Olkaria I and II geothermal power plants, which are approximately 3.7 km apart and located within the precincts of the Hells Gate National Park (HGNP). Table 1 shows the main features of the power plants based on available data. Human settlement within the field is minimal due to its location within the HGNP (area, 68.25 km^2). The vegetation is mainly diverse types of grassland and shrub land (Ogola, 2004) with *Tarchonanthus camphoratus* L. covering extensive areas, occasionally interspersed with *Vachellia drepanolobium* (Harms ex Sjöstedt) P.J.H.Hurter (Syn. *Acacia drepanolobium* Harms ex Sjöstedt) and *Vachellia xanthophloea* (Benth.) P.J.H.Hurter (Syn. *Acacia xanthophloea* Benth.) (KenGen, 2004). The shrub *T. camphoratus* is semi deciduous, usually multi-stemmed (Young and Francombe, 1991) reaching 2–6 m height. The stem group of the multi-stem forms is known as a clump and the stem with the largest stem circumference and height is termed main stem.

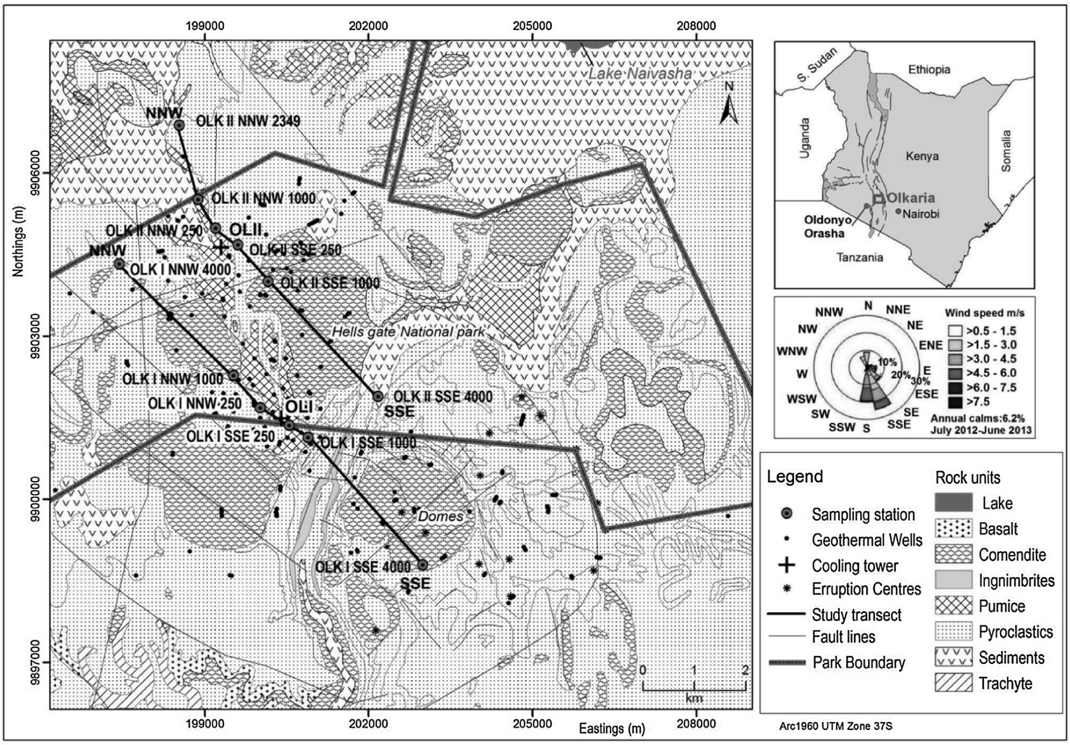

Fig. 1. Location of the Olkaria geothermal area showing the study sites and sampling stations. Annual (July 2012–June 2013) wind rose of the area is also shown.

Table 1
Summary data of the two power plants in the immediate vicinity of the study area. NL—Not in literature. There is no data on the emission rates of arsenic, boron and antimony in the air.

Power plant	Total installed capacity (Mwe)	Year of commisioning	No. of connected wells	H_2S tonnes/year[a]	Arsenic tonnes/year[b]	Boron tonnes/year[b]	Mercury grams/year[c]
Olkaria I	45	1981, 1983 & 1985	19	426	13.1	10.3	4,027
Olkaria II	105	2003 & 2010	21	1,323	NL	NL	19,212

[a] KenGen unpublished data, 2013.
[b] Simiyu and Tole (2000). The concentrations were assessed in condensed steam.
[c] Wetangula (2011).

2.2. Study design, sampling and field measurements

After a preliminary survey, two line transects running South South East (SSE), upwind and North North West (NNW), downwind from the cooling towers were established around each power plant (Fig. 1). Along each transect, sampling stations were set up at 250 m, 1000 m and 4000 m distance from the cooling towers, twelve in total (Fig. 1). In addition, one station was set up at a reference site at Oldonyo Orasha in Narok at similar environmental conditions 68.3 km away South West (SW) of the study area. At each sampling station, four 10 × 10 m plots with equal spacing (10 m) were established for sampling along a 70 m sub-transect, perpendicular to the main transect. At the reference site, a similar sampling design was adopted. All field measurements and sampling were conducted in February and March 2014.

In each plot, *T. camphoratus* abundance was determined by counting clumps. Assessment of morphological traits and leaf sampling was then performed on three clumps of *T. camphoratus* in each plot, i.e. at the mid central point and at the midpoint of each of the two sides of the plot perpendicular to the sub transect. The total number of stems in each clump was counted. On the main stem of each clump, stem height and circumference were measured. Stem height was determined using a demarcated rod while stem circumference was measured 30 cm off the ground (Oszlányi, 1997; West and West, 2009; Young and Francombe, 1991). To assess leaf damage, three leaf categories were established: (A) healthy green leaves, (B) yellow deteriorating leaves, and (C) brown dry leaves (dead). We chose the three leaf categories based on previous studies on the effects of the emitted elements on leaves and own observations. For example, the symptoms of elevated boron are manifest as yellowing (leaf category B), spotting or drying of leaf tissues in older leaves at the tips and margins and as leaf drying and chlorosis with increased exposure that advances toward the centre of the leaf (Ayers and Westcot, 1985; Tuyor et al., 2005). We categorized the early senescence as leaf category C and all these foliar symptoms as chronic effects of leaf damage influenced by the emitted geothermal elements. A leaf was categorized as B or C if more than 50% of the leaf colour was yellow or brown, respectively (Fig. S1). The number of leaves on the third upper branch of the main stem in each of three leaf categories was counted per clump.

Wearing polyethylene gloves, ten leaves per category per clump were randomly sampled from the same branch in each plot for the determination of sulphur, arsenic, boron, mercury and antimony concentrations. This branch was chosen, as it was high enough to be exposed to atmospheric pollution. Due to limited chemical analytical funds, all collected leaf samples for all plots per station were pooled within each leaf category and thoroughly mixed. Ten leaves were randomly sub-sampled to represent a single sample for the station for each category. In total, 39 leaf samples for all categories (18 samples per transect around each Olkaria power plant and three samples at the reference site) were packed in pre-cleaned polythene bags for laboratory analysis.

The upper 0–10 cm layers of soil were sampled to determine accumulation of airborne substances (modified from Bussotti et al., 2003). A soil sample was taken at each of the three clumps within a plot. Soil samples were pooled for each station and thoroughly mixed. Three sub-samples were drawn out of the station pool for analyses of sulphur, arsenic, boron, mercury and antimony concentrations. Total soil nitrogen, soil pH, and soil moisture, were also determined in each of the three soil sub-samples to account for possible influence of soil conditions on the variance of the plant data.

2.3. Sample treatment and laboratory analysis

At the laboratory, leaf sub-samples were rinsed with distilled water (three times), dried in an oven at 40 °C for 48 h to constant weight and pulverized using agate mortars. All analyses were carried out according to standard analytical procedures at the internationally accredited Kenya Bureau of Standards (KEBs) chemistry laboratory. The concentration of sulphur in leaves was determined according to standard methods of the Bureau of Indian Standards (1986) reaffirmed in 2003 using a Shimadzu 1700 series pharmaspec UV/VIS spectrophotometer. Concentrations of other elements, i.e. arsenic, mercury, boron and antimony, were determined according to procedures described and modified from Hettipathirana (2011), and Vummiti (2015), using the Agilent 4100 MP-AES (Microwave Plasma Atomic Emission spectrometer). Element concentrations were expressed as weight per dry weight of sample in μg/g or μg/kg. Procedural blanks were below the minimum detection level. Accuracy was checked through analysis of standard reference materials for soil (Montana soil) and leaves (Tomato leaves) (National Institute of Standards and Technology, Gaithersburg, MD, USA), and better than 95% recoveries obtained.

The soil samples were dried at 40 °C for 48 h to constant weight, and sieved through 2 mm and analysed for sulphur, arsenic, mercury, boron, and antimony using the same protocols as for the leaves. Total soil nitrogen was determined according to the Kjedahl method described in Bureau of Indian Standards (2007) and ISO (2012) using a FOSS Kjeltec 8400 series analyser. For soil pH, 5 g of 96 h air-dried and sieved soil were weighed into 25 ml de-ionized water, shaken for two hours and allowed to settle for 8 h before measuring pH (Blakemore et al., 1987). Soil moisture was measured as percentage weight reduction after oven drying 10 g fresh soil at 105 °C for 24 h to constant weight.

2.4. Statistical analyses

We used the R software version 3.2.2 for all statistical analyses (R Development Team, 2010). For visual assessment and exploration of patterns in our plant concentration, soil concentration, and plant trait data, respectively, we performed a Non-Metric Multidimensional Scaling (NMDS) ordination in R (Oksanen et al., 2015). Euclidean dissimilarity was used as a distance metric.

For each of the elements sulphur, arsenic, boron, and antimony, we tested whether concentrations were different between the different categories of leaves using a simple linear model. For mercury, the concentrations in the plant tissues and soil were below detection for all samples. Because the concentrations of sulphur and arsenic did not significantly differ between leaf categories, we treated them as replicates for the stations in further statistical analyses. However, for boron and antimony concentration, the leaf categories A–C were significantly different and each category was separately analysed statistically. The concentrations of sulphur, arsenic, boron, and antimony in plant leaves and soil were analysed using linear models with distances from the power plants (250 m, 1000 m and 4000 m), location of the sampling stations (Olkaria I versus Olkaria II) and direction from the power plants (i.e. SSE upwind and NNW downwind) as additive predictor variables. To meet the model assumptions, sulphur and boron concentrations in leaves and sulphur, and arsenic concentrations in soil were $\log_e$ transformed. The model that had boron concentrations in leaves as a response variable against the predictor leaf category was $\log_e (x + v)$ transformed, with (v) representing the smallest value of boron concentration in leaves of the sampled data in order to avoid negative values for samples that had concentrations below the detection limit. For analysis of leaf and soil concentrations, the soil variables, soil pH, soil nitrogen, and soil chemistry (sulphur, arsenic, boron, and antimony concentrations), were included as co-variates, one at a time, and then the best fitted model based on the lowest AIC (Akaike's Information Criterion) selected.

For the continuous morphological response variables (stem height, stem circumference), we fitted linear mixed effect models with the same predictor variables as in the linear models above (distance, location, direction) and their interaction as fixed factors, soil variables as co-variates, and sampling station and plots within station as random factors (to account for spatial auto-correlation). For the count data (shrub abundance, number of leaves per category, and number of stems per shrub), generalized linear mixed effect models with a Poisson distribution were fitted. The effect of the interactions did not yield clear trends and were therefore not included in the final models.

The Variance Inflation Factors (VIF) for all models was calculated to ensure no collinearity and according to Zuur et al. (2010), only variables of VIF below 3 were preserved for the final models. Further standard model diagnostics were used to check the residuals (for normality) and control for outliers. The optimal model for each response variable was identified by selecting the lowest AIC after running a series of model sub-sets. The final models chosen were therefore reduced. The models were run using the functions lm in the MASS package in R (Ripley et al., 2015) for linear models, lme in the nlme package (Pinheiro et al., 2015) for linear mixed effect models and lmer in the lme4 packages (Bates et al., 2014) for the generalized linear mixed effect models. Lists of models and test statistics used are presented in Tables S1a, b, S2, S3 and S4.

3. Results

Soil nitrogen content and soil pH were similar among the sampling stations (Table S5). As expected, soil moisture was more variable as it is easily affected by recent rain events.

3.1. Element concentrations in T. camphoratus leaves

The leaf categories A–C were not significantly different for sulphur and arsenic concentrations (Table S1a, Fig. 2a, b), while boron and antimony concentrations differed significantly. In the leaf category B, the concentrations of boron and antimony were significantly lower than leaf category A and C (Tables 2 and S1a). We could not understand the implication as we had expected increased element

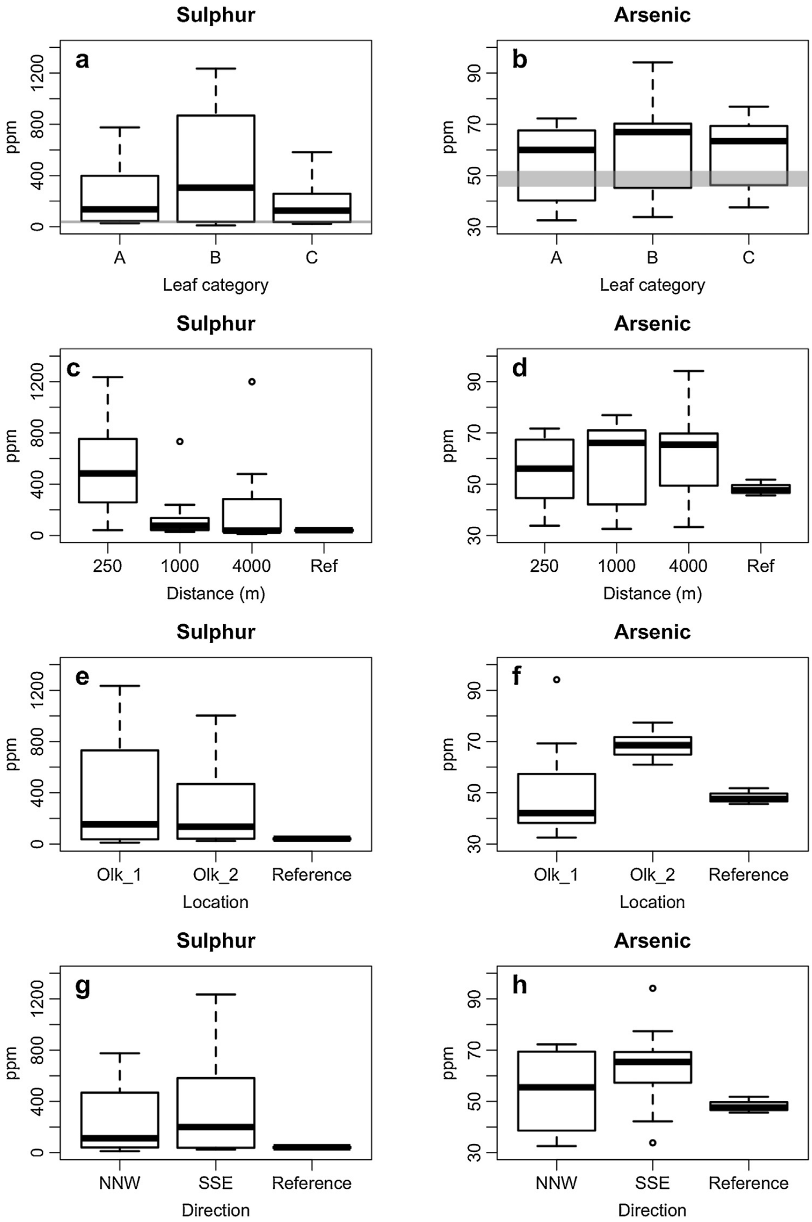

Fig. 2. Concentration of sulphur and arsenic in *T. camphoratus* leaves at Olkaria compared to a reference site at 68300 m away (Ref.); (2a and b) in the leaf categories A–C (horizontal grey bar represents the concentration range across the leaf categories at the reference site), (2c and d) at different distances from Olkaria I and II power plants, (2e and f) around the Olkaria I and II power plants, (2g and h) at different directions from Olkaria I and II power plants. Sulphur concentration: significantly different for the different distances compared to 250 m ($t(32) = -3.68$ $p = 0.001$ for 1000 m and $t(32) = -3.71$ $p = 0.001$ for 4000 m). Arsenic concentration: significantly different between Olkaria I and II ($t(28) = 5.91$ $p = 0.000$ for Olkaria II), and between direction NNW and SSE ($t(28) = 2.40$ $p = 0.023$ for SSE). Detection limits, 0.83 mg/kg sulphur and 0.6 mg/kg arsenic. $n = 39$.

concentrations in the leaf categories B and C over the leaf category A.

The NMDS ordination revealed large variation in the overall element concentrations in the leaves of *T. camphoratus* among the sampling stations around Olkaria I and II (Table 2, Fig. 3a–c). However, the elements responded differently to the predictors (distance, direction or location) and therefore the ordination pattern was not clearly related to any of them (Table S1b).

The concentration of sulphur in leaves decreased with distance from the power plants and was significantly higher at 250 m away than at 1000–4000 m away (Table S1b, Fig. 2c). However, it did not differ between the two locations or directions (Fig. 2e, g). Arsenic

Table 2
Concentrations of boron, antimony and mercury in *T. camphoratus* leaves and soil (dry weight), collected along transects at two power plants in the Olkaria geothermal area in Kenya (leaves $n = 39$, soil $n = 13$). Detection limits: boron 0.05 mg/kg, antimony 0.6 mg/kg and mercury 8.8 μg/g.

			Olkaria I				Olkaria II			
Element	Direction	Distance (m)	Leaf A	Leaf B	Leaf C	Soil	Leaf A	Leaf B	Leaf C	Soil
Boron (mg/kg)	NNW	250	0.80	0.11	<0.05	2.27	2.93	<0.05	3.40	1.50
		1000	2.08	0.29	4.78	0.67	0.18	0.30	2.34	2.27
		4000	4.00	0.10	1.40	1.51	1.46	2.49	1.75	0.45
	SSE	250	2.27	<0.05	2.75	2.29	<0.05	3.49	<0.05	0.94
		1000	2.08	0.37	5.49	3.43	1.82	2.03	0.19	1.34
		4000	2.77	0.00	2.17	2.15	2.11	2.49	1.75	1.71
		Reference site Leaf A <0.05, Leaf B 0.10, Leaf C 0.09, Soil 1.01								
Antimony (mg/kg)	NNW	250	<0.6	<0.6	<0.6	12.25	19.70	<0.6	22.93	5.05
		1000	20.92	<0.6	21.04	5.16	<0.6	8.14	21.84	4.74
		4000	20.71	<0.6	20.78	8.66	21.35	19.10	17.50	<0.6
	SSE	250	22.55	5.16	22.94	5.13	<0.6	19.01	<0.6	7.66
		1000	17.57	13.48	18.11	9.08	18.78	19.78	<0.6	11.01
		4000	14.81	<0.6	22.92	4.46	18.44	<0.6	17.70	5.32
		Reference site Leaf A <0.6, Leaf B <0.6, Leaf C <0.6, Soil 11.24								
Mercury (μg/kg)			<8.8				<8.8			
		Reference site Leaf A <8.8, Leaf B <8.8, Leaf C <8.8, Soil <8.8								

concentrations on the other hand were not affected by distance (Fig. 2d), but were higher around Olkaria II than I (Fig. 2f) and higher in SSE direction (upwind) than NNW (Table S1b, Fig. 2h). For boron and antimony, their concentrations did not vary with our predictors. The soil characteristics (co-variates) improved the models by explaining significant variations for sulphur, arsenic, boron, and antimony concentration in the leaves.

Across all sampling stations around the two Olkaria power plants, the concentrations of sulphur, arsenic, antimony and boron in *T. camphoratus* leaves were generally higher than at the reference site (Table 2, Fig. 2a, b). This was also evident in the NMDS plot that showed a clear separation of the reference site from the power plant stations along the first ordination axis (Fig. 3a–c).

3.2. Element concentrations in soil

In general, the element concentrations were at lower levels in the soil compared to leaves, indicating a bio-accumulation (Figs. 2, 4 and Table 2). However, similar to the leaves the elements responded differently to the predictors, which was reflected in the ordination of the overall element soil concentrations (Fig. 3d–3f).

Distance had a significant effect on soil concentration of sulphur, although in a non-linear way: it was highest at 1000 m distance (Fig. 4a, Table S2). The co-variate soil pH was significant and accounted for some of the unexplained variation in sulphur concentrations, improving the model (Table S2). The other two predictors, direction and location did not significantly affect sulphur concentration (Fig. 4c, e, Table S2). Arsenic concentration in soil was affected by location as in leaves, but was at higher levels at Olkaria I than II, opposite to what was found in leaves (Fig. 4d, Table S2). We could not determine the causes for the differences as arsenic concentrations in the leaves around Olkaria II did not vary with distance to indicate an influence of the power plant. Distance and direction did not affect arsenic concentration (Fig. 4b, f). The concentrations of the other elements in soil were not significantly affected by any of the predictors (Table S2).

Compared to the reference site, the highest soil concentrations of sulphur, arsenic, and boron occurred at the Olkaria sampling stations, except for antimony (Fig. 4a, b, Table 2). Similar to the leaves, this was reflected in the ordination of the overall element concentration in soil where the reference site had lower scores along the first axis than at all the transect stations (Fig. 3d–f).

3.3. *T. camphoratus* growth related morphometrics

Overall, the growth related plant traits varied considerably around Olkaria I and Olkaria II, although with no clear dissimilarity patterns along the ordination axes of the NMDS plot (Fig. 3g–i). Abundance was the only plant trait that showed significant response to the any of the predictors and was higher upwind (SSE) than downwind (NNW) from the power plants (Fig. 5, Tables S3 and S4) and it even exceeded the abundance at the reference site.

Further comparisons to the reference site showed a generally smaller number of category A leaves (healthy) by 43% and 24% smaller stem circumferences around the power plants (Table 3, Fig. 5).

4. Discussion

Within a geothermal area, trends in element concentrations over an increasing distance from the source (power plants), will provide the strongest indication of potential emission input to the surrounding ecosystems. Our most significant findings provided some support for the overall hypothesis. The relatively high levels of sulphur in leaves close to the power plants that decreased with distance away, and the generally higher concentrations of some of the elements in leaves and soil around the power plants than at the reference site in combination with lower number of healthy leaves and smaller main stem circumference, all provide an indication of an influence of geothermal power plant emissions on the ecosystems in their vicinity.

The strong pattern of accumulated sulphur concentration in leaves suggests atmospheric input of this element with the nearby geothermal power plants as a source (in H_2S gas form). Studies in the Mediterranean in which lichens, mosses, and forest trees were used as bio-monitors of geothermal air pollution (Baldi, 1988; Bargagli et al., 1997; Bussotti et al., 2003; Loppi et al., 1998; Paoli and Loppi, 2008) showed similar trends of decreasing sulphur concentrations in plant tissues with distance away from power plants. Similar to the findings of Bussotti et al. (2003), the higher concentrations of most elements in plant leaves than in the soil in this study indicate foliar accumulation from atmospheric deposition. On the other hand, the higher arsenic concentration in soil than in the leaves points to its potential origin from other sources besides atmospheric deposition. The same argument may apply for the other elements which did not show clear trends and var-

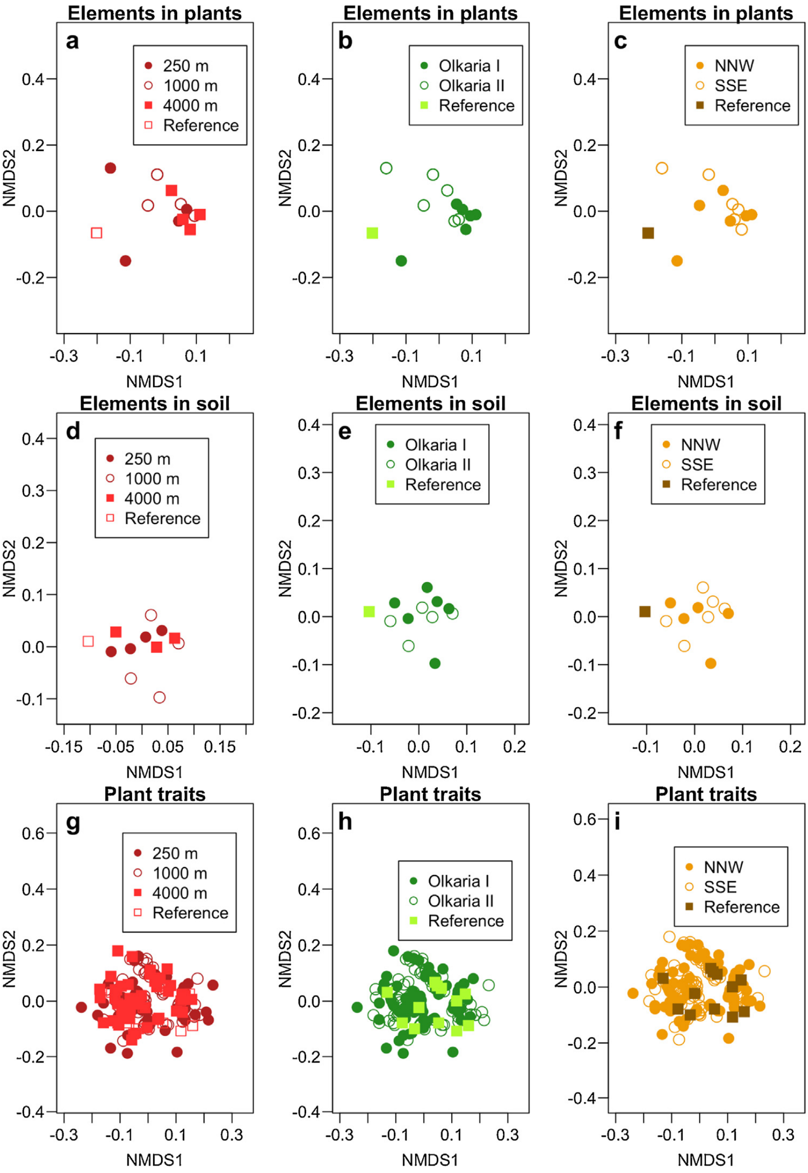

Fig. 3. Nonmetric multidimensional scaling of (3a–c) sulphur, arsenic, boron and antimony concentrations in plant leaves, (3d–f) sulphur, arsenic, boron and antimony concentrations in soil and (3g–i) the various plant traits measured.

ied widely between locations and directions. These other sources may include surface discharge of geothermal fluids (either from natural discharges, drilled and well tests fluids or a combination of all) which contain various geothermal elements. The elements interact with the recipient ecosystem components such as soil and plants and may bio-accumulate, e.g. as shown in Simiyu and Tole (2000), where elevated element concentrations occurred in soil and plants in contact with geothermal waters (both natural discharges and drilled fluids) at the Olkaria geothermal field. Other environmental factors such as the varying bedrock in the volcanic active surroundings (see Fig. 1) may contribute to the high and varying concentrations of elements of volcanic origin in the soil which can have chronic effects on plants (Davies, 2008).

There were no clear indications of the effect of the prevailing wind in element deposition. A proper evaluation of the effect of the wind may, however, be difficult owing to the closeness of the power plants. The effect of different output capacities of geothermal power plant emissions on the plants and soil in the immediate vicinity was not clear. Such a question can better be explored using experimental studies such as those in Thompson and Kats (1978) and Maas et al. (1987). With regard to the effect of these emissions on the plant traits, our findings showed a weak indication of harmful effects of the geothermal power plants on the dominating shrub

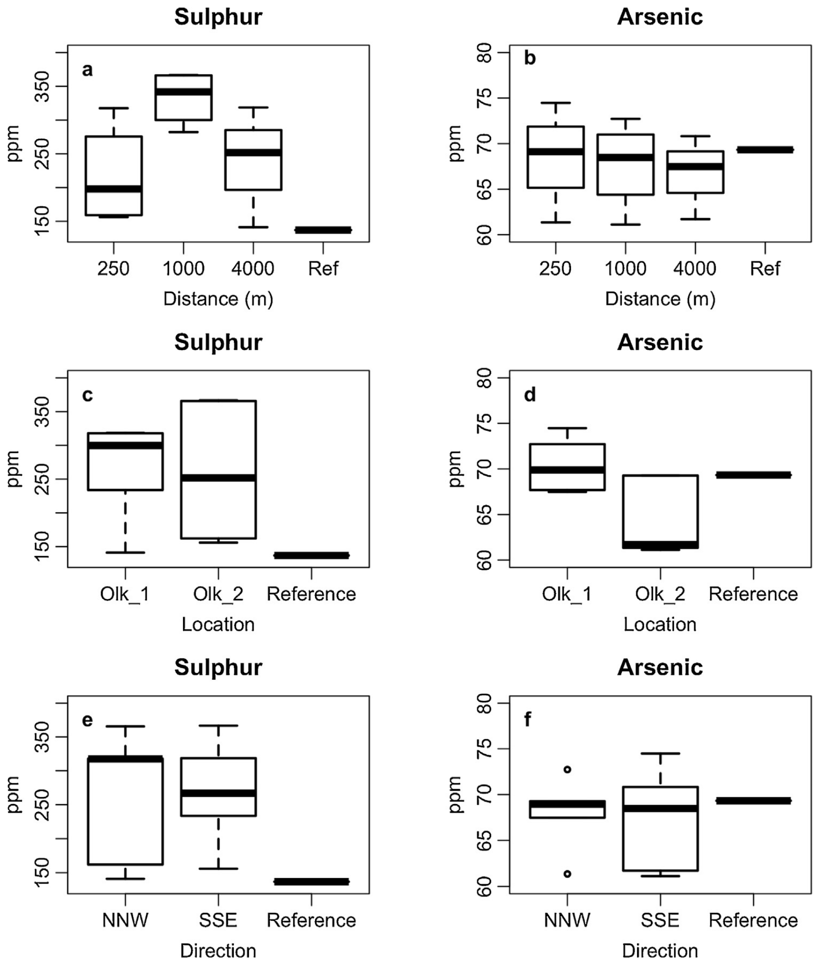

Fig. 4. Concentration of sulphur and arsenic in soil at Olkaria compared to a reference site at 68300 m away (Ref.); (4a and b) at different distances from the power plants, (4c and d) around the Olkaria I and II and (4e and 4f) at different directions from Olkaria I and II power plants. Sulphur concentration: significantly different between the distances 250 m and 1000 m ($t_{(7)} = 2.17$ $p = 0.052$ for 1000 m). Arsenic concentration: significantly different between the locations Olkaria I and II ($t_{(9)} = -2.70$ $p = 0.024$ for Olkaria II). Detection limits, 0.83 mg/kg sulphur and 0.6 mg/kg arsenic. $n = 13$.

Table 3
Various plant traits (mean ± SE, $n = 156$) of *T camphoratus* around Olkaria I and Olkaria II geothermal power plants in Kenya. The units for each leaf category count is number of leaves per branch (as defined in methods).

Location	Direction	Distance (m)	Main stem height (m)	Main stem circumference (cm)	No. Leaf A	No. Leaf B	No. Leaf C
Olkaria I	NNW	250	2.50 ± 0.19	12.20 ± 1.54	120.00 ± 33.94	1.00 ± 0.66	2.00 ± 0.97
		1000	4.00 ± 0.31	18.20 ± 2.11	96.00 ± 18.79	2.00 ± 0.71	3.00 ± 0.59
		4000	4.60 ± 0.24	26.00 ± 2.56	56.00 ± 12.39	1.00 ± 0.38	3.00 ± 1.29
	SSE	250	4.90 ± 0.30	29.00 ± 2.13	54.00 ± 8.45	2.00 ± 0.65	2.00 ± 0.66
		1000	4.60 ± 0.29	28.80 ± 2.99	59.00 ± 8.33	1.00 ± 0.51	2.00 ± 0.92
		4000	3.50 ± 0.27	15.60 ± 1.47	71.00 ± 11.19	2.00 ± 0.34	4.00 ± 1.22
Olkaria II	NNW	250	5.40 ± 0.39	36.30 ± 3.13	63.00 ± 8.86	1.00 ± 0.58	4.00 ± 1.30
		1000	5.20 ± 0.48	31.70 ± 3.71	52.00 ± 11.04	1.00 ± 0.23	3.00 ± 1.44
		4000	3.50 ± 0.29	18.90 ± 2.53	92.00 ± 15.66	3.00 ± 0.64	2.00 ± 0.78
	SSE	250	4.30 ± 0.36	26.50 ± 3.59	70.00 ± 20.67	1.00 ± 0.65	2.00 ± 0.91
		1000	4.00 ± 0.22	18.60 ± 1.00	61.00 ± 9.67	1.00 ± 0.26	3.00 ± 0.90
		4000	4.60 ± 0.17	24.90 ± 1.00	70.00 ± 7.95	2.00 ± 0.56	6.00 ± 1.37
Reference site		68300	4.40 ± 0.25	31.00 ± 3.13	127.00 ± 22.93	1.00 ± 0.44	2.00 ± 0.89

species when compared to a reference site. Although plant traits may not only be affected by the geothermal power plant emissions but also other unforeseen environmental conditions such as soil characteristics, which accounted for some variation in our models.

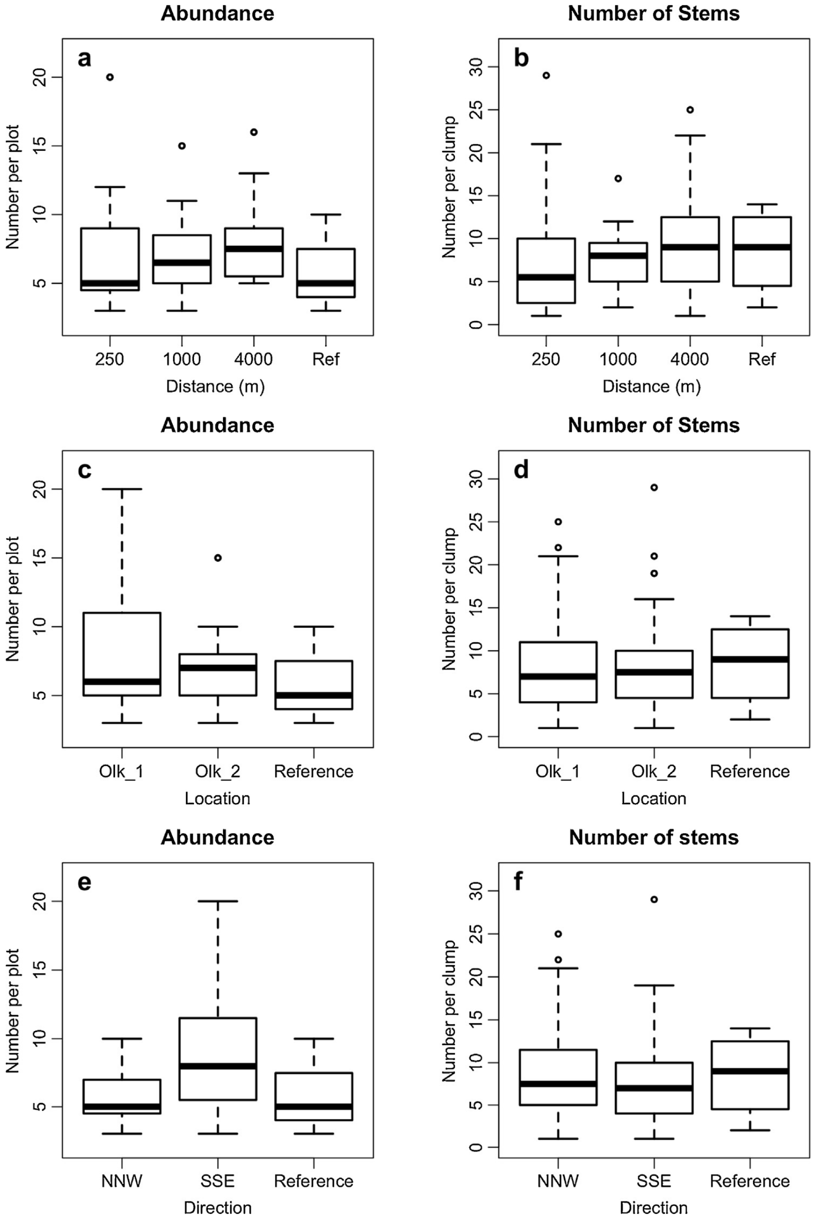

Fig. 5. The abundance (number of shrubs per plot) and number of stems per clump of *T. camphoratus* compared to a reference site at 68300 m away (Ref.); (5a and b) at different distances from the power plants, (5c and d) around the Olkaria I and II power plants, (5e and f) at different directions from Olkaria I and II power plants. Abundance: significantly different between the directions NNW and SSE ($z_{(139)} = 2.58$ $p = 0.009$ for SSE). $n = 156$.

Bussotti et al. (1997) explain that these natural fluctuations can to a larger extent conceal the effects caused by the power plants.

We can imply that *T. camphoratus* is not affected by the measured element levels. The sulphur levels in the plant leaf tissues seem to be within its nutrition benefits and the concentrations of the other elements within its toxi-tolerance limits. However, it is important to be aware that plants can accumulate components in their foliage and not instantly result in plant health deterioration (Bussotti et al., 2003). In forest ecosystems for example, the effects may be manifested through foliage shedding and accumulation on the forest floor consequently affecting soil biota, decomposition rates and nutrient cycling (Ferretti, 1997). Various tree species, however, adopt toxi-tolerance by regulation and physiological adaption mechanisms. Nonetheless, such tolerance responses are costly for the plants and long-term stress leads to their weakening, resulting in the destruction of their mechanisms (Oszlányi, 1997).

5. Conclusions and recommendations

The emissions from the geothermal power plants at Olkaria, which have been in operation for over a decade, have not had an apparent effect on the dominating plant *T. camphoratus.* However we do recommend improved studies on other organisms to assess if effects do emerge, preferably using sensitive ecosystem components, such as lichens, mosses, grasses, herbs or soil microbes (Baldi, 1988; Bargagli et al., 2002; Loppi et al., 1998; Storelli, 2013; Zouboulis et al., 2004). Additional plant responses such as flowering and recruitment of new plants, photosynthesis, growth and leaf production can be assessed and related to variables associated with the power plant emissions. Concentrations of the elements sulphur, arsenic, boron, antimony and mercury in the steam, air and precipitation around the power plants need to be determined to support these studies. Moreover, a pattern study like this needs to be supported by experimental studies especially to closely assess the effects on plant growth. The findings of this study are important in informing such future studies and will advise the public, policy/decision makers and guide mitigation of environmental impacts related to geothermal power plants.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.geothermics.2016.01.017.

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Paper II

Concentrations of sulphur and trace elements in subarctic soils and mosses in relation to geothermal power plants at Hengill, Iceland – ecological implications

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- Performed fieldwork: TMM, ISJ
- Performed laboratory work: TMM
- Analysed the data: TMM, ISJ
- Wrote the paper: TMM, TF, SHM, ISJ

Concentrations of sulphur and trace elements in subarctic soils and mosses in relation to geothermal power plants at Hengill, Iceland – ecological implications

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Abstract

Amid the globally accelerated plans to increase geothermal energy utilization, knowledge on ecological responses to power plant emissions is limited. We attempted to answer the question whether emitted elements from geothermal power plants in Iceland are deposited in both plants and soil with consequences for plant health. The moss *Racomitrium lanuginosum* was used as a bio-indicator, a dominating plant in our study areas. Concentrations of sulphur, arsenic, boron, antimony, and mercury in soil and shoots of *R. lanuginosum*, were analysed, and growth and other moss characteristics (moss damage, physiology and mat depth) assessed at different distances and directions from two geothermal power plants, Hellisheidi (303 MWe, operated since 2006) and Nesjavellir (120 MWe, operated since 1996).

Higher concentrations of these elements were detected around Hellisheidi than Nesjavellir. Sulphur, antimony, and mercury concentrations in moss decreased with increasing distance from the power plant around Nesjavellir. Similar trends for sulphur and antimony followed in soils. At Hellisheidi, element concentrations in moss and soil generally increased with distance, but their patterns with direction in relation to prevailing winds were not clear. Moss growth and characteristics showed stronger positive growth responses at Hellisheidi than Nesjavellir. At Nesjavellir, values of moss response variables increased with increasing distance away while at Hellisheidi trends were non-linear. Frequency of moss damage was quite low around both power plants. The results, compared to a reference area away from geothermal activity, showed much higher element concentrations in moss and soil at the study area than the reference area, except for sulphur and mercury in soil. Biomass gain was higher at the reference area than at the study areas. We conclude that sulphur emissions from the geothermal power plants are depositing in moss within the surroundings which may affect moss growth at high levels. The pattern of trace element (As, B, Sb and Hg) concentrations in plants and soils around the power plants did not show a significant contribution of the elements from the power plants. However, monitoring of potential environmental effects is advised. We recommend future experimental studies to assess effects of sulphur in detail for appropriate mitigation.

Keywords: *Racomitrium lanuginosum*, Nesjavellir, Hellisheidi, moss growth, geothermal power plants, emissions.

Introduction

Today, exploitation of geothermal energy, where available, is increasingly preferred over nuclear and fossil fuels for power production. This is owing to its relatively clean and reliable nature. However, like the development of any other energy source, some environmental impacts are expected. These include among others, surface disturbance and atmospheric pollution (Kristmannsdóttir and Ármannsson, 2003). Mitigation efforts need to be based on solid research, but so far only very few studies have been performed to address environmental impacts of geothermal power plants in general and impacts of geothermal emission in particular (e.g. Bargagli et al., 1997; Bussotti et al., 1997; Kristmannsdóttir and Ármannsson, 2003; Rybach, 2003; Kömürcü and Akpınar, 2009; Bayer et al., 2013).

Emissions from geothermal power plants involve the release of a range of gaseous compounds and elements that are not condensed at operating temperatures and pressures, i.e. non-condensable gases (NCGs). The NCGs content varies extensively between power plants depending on the resource, often in the range of 0.2% to 2% by weight, and up to over 25%, in rare cases (Ozcan and Gokcen, 2009b; Rodríguez, 2014). Generally, the NCGs comprise 73 - 98% w/w carbon dioxide, 1 - 24% w/w hydrogen sulfide, 0.02 - 0.65% w/w methane, 0.1 - 8% w/w hydrogen, 0.3 - 16% w/w nitrogen, 0.1 - 3% argon, and traces (<0.001% w/w) of radon, boron, mercury, arsenic, antimony, and ammonia in gaseous and dissolved form (Axtmann, 1975; Baldi, 1988; Bargagli et al., 1997; Loppi et al., 1998; Gunerhan, 1999; Loppi, 2001; Bussotti et al., 2003; Rodríguez, 2014). Some of these gases and elements have been reported to deposit and accumulate in the immediate environment of geothermal power plants with several ecological implications (Loppi and Bonini, 2000; Mutia et al., 2016).

The effects on human health of hydrogen sulphide and trace elements that may be found in geothermal power plant emissions (Bargagli et al., 1997; Bussotti et al., 1997) and volcanic activities (Hansell and Oppenheimer, 2004; Davies, 2008) i.e. arsenic, boron, antimony and mercury in gaseous and dissolved form, are fairly well documented, with indications of potential harm at elevated concentrations (Hansell and Oppenheimer, 2004; Davies, 2008; Finnbjornsdóttir et al., 2015). It is likely that the effect of high levels of geothermally emitted elements may also be hazardous to the natural ecosystems, cause toxicological stress on plant and animal life and affect ecosystem functions (Bargagli et al., 1997; Kristmannsdóttir and Ármannsson, 2003). However, data on the ecosystem impacts are still scanty and limited to a few studies in Italy and Kenya (Bargagli et al., 1997; Bussotti et al., 2003; Mutia et al., 2016).

Elevated levels of the emitted elements in plants (including mosses) and lichens have been reported near geothermal power plants, that decrease with increasing distance away both in Italy (Baldi, 1988; Bargagli et al., 1997; Loppi et al., 1998; Loppi, 2001; Bargagli et al., 2002; Bussotti et al., 2003) and Kenya (Mutia et al., 2016), suggesting geothermal power plant origin. These studies suggest that elements originating from geothermal power plants can accumulate in plants. Studies on the effects of the emitted elements have mainly focused on vascular plants, while knowledge of their effects on non-vascular plants, such as mosses, is more limited.

In plants, H_2S gas can be an important contributor of the macro-nutrient sulphur, which is essential for plant growth and metabolism. However, in excess, sulphur can cause detrimental effects to plants (Linzon et al., 1979; Bussotti et al., 2003). In vascular plants, excess sulphur levels, for example from H_2S gas emissions, have been reported to cause foliar injury manifested as necrosis, lesions, loss of leaf area and defoliation (Thompson and Kats, 1978; Bussotti et al., 2003). Further, long term exposure to H_2S gas leads to leaf chlorosis, decreased plant growth and increased senescence (Thompson and Kats, 1978; Varshney et al., 1979; WHO, 2000).

Similarly, higher concentrations of trace elements affect growth and metabolism in plants (Kabata-Pendias, 1992; Nagajyoti et al., 2010). For instance, elevated concentrations of arsenic and mercury can induce physiological disorders in plants. Higher concentrations of arsenic can affect metal sensitive enzymes in plants and lead to growth inhibition and death (Nagajyoti et al., 2010). The ionic form of mercury (mercuric ion: Hg^{2+}) can bind to water channel proteins, thus causing leaf stomata to close resulting in physical obstruction of water flow in plants (Nagajyoti et al., 2010). Further, high levels of Hg^{2+} can also disrupt bio-membrane lipids and cellular metabolism in plants (Nagajyoti et al., 2010). For high boron concentrations, typical macroscopic symptoms include leaf burn and chlorosis and/or necrotic patches mostly at the margins and tips of older leaves that lead to reduced plant growth, loss of leaf area and decreased carbon dioxide gas fixation amongst a wide variety of plant species (Eaton, 1944; Bergmann, 1992; Bennett, 1993; Nable et al., 1997). Antimony as well has deleterious effects to plants at higher concentrations. For example, in a study by Vaculík et al. (2015), high antimony concentrations (in the oxidation state: antimonite) in young sunflower plants (*Helianthus annuus* L.) related to reduced plant growth and photosynthesis.

Since mosses are more sensitive to atmospherically deposited pollutants than vascular plants, we can expect them to be more severely affected. For example, studies by Tallis, (1964a) suggested atmospheric pollution as the probable cause of the virtual disappearance of the *Sphagnum* moss species from the bog vegetation of the southern Pennines, Northern England. A later study (Ferguson et al., 1978) that assessed vegetation changes for the same area through laboratory experiments revealed the sensitivity of *Sphagnum* moss species to Sulphur pollutants; growth of *Sphagnum recurvum* was reduced up to 35% on exposure to 40.51 mg/l (0.5mM) bisulphite. Previous studies on *R. lanuginosum* also indicate that it is affected by increased deposition of atmospheric pollutants such as sulphur (Woodin and Farmer, 1993) and nitrogen (Pearce and Van der Wal, 2008). Although growth may be initially stimulated, high loads may damage the moss. There are however limited studies that have addressed the impact of geothermal emissions on growth and functioning of mosses.

In subarctic Iceland, the contribution of geothermal energy has since the twentieth century been important in the primary energy supply. Today 2,040 MWth is used directly by a greater part of the population in various ways, such as space heating (Ragnarsson, 2015). The use of geothermal energy for electricity production began 45 years ago and rapidly increased the last 15 years to reach a 29% share in electricity production, with a total installed capacity of 663 MWe (Ragnarsson, 2015). An additional 12% increase of the total installed geothermal electric power is targeted by the year 2020 (Ragnarsson, 2015). With all the planned developments, it is consequently important to fill the knowledge gap on the ecosystem impacts of the geothermal power plants.

The Hengill geothermal power plants: Hellisheidi and Nesjavellir (Table 1), located in Southwest Iceland, contribute a larger share of geothermal power to the economy of Iceland than other geothermal power plants in the country (Ragnarsson, 2015). Average emissions from the Hengill geothermal power plants NCG content have been reported as 73% carbon dioxide w/w, 25% hydrogen sulphide w/w, 1% hydrogen w/w and 0.2% methane w/w respectively for the period 2011 – 2013 (Sigurdardóttir and Thorgeirsson, 2015). However, only the emissions of hydrogen sulphide gas have been measured from the power plants, while those of trace elements i.e. arsenic, boron, antimony and mercury, either in gaseous and dissolved form, from the power plants are not yet determined. To assess the impact of Hengill geothermal power plants on terrestrial ecosystems, we refered to the Italian geothermal power plants emission studies that have revealed evidence of trace element emissions i.e. arsenic, boron, antimony and mercury, from geothermal power plants and deposition in plants within the vicinity of the power plants (e.g. in Bargagli et al., 1997). As there is no data for trace elements emissions from the Hengill geothermal power plants, we predicted similar signatures of trace elements emissions and deposition from the Hengill geothermal power plant as those of the Italian studies.

Extensive and thick carpets of the moss *Racomitrium lanuginosum* dominate the landscape in the vicinity of the geothermal power plants in Southwest Iceland. In some areas around the power plants in Iceland, damage has been observed on the moss carpet (Bragason and Yngvadóttir, 2009; Helgadóttir, et al., 2013). So far the cause of these damages has not been established, but elevated concentration of sulphur, boron and arsenic has been reported in moss in the area that may be related to the power plant emissions (Bragason and Yngvadóttir, 2009; Magnússon, 2013). To ensure sustainable development of geothermal power plants in Iceland, the relationships between elevated element concentrations on one hand and moss growth and observed moss damage on the other needs to be established.

The aim of this study is to investigate these relationships by providing information on the ecosystem accumulation of elements emitted from two geothermal power plants at the Hengill geothermal field in Southwest Iceland, Hellisheidi and Nesjavellir. We hypothesized that the elements emitted would be deposited and enriched in both plant tissue and soil around the power plants with consequences for plant health. To test the hypothesis, we assessed chemical compositions of the soil and the dominating moss species, *Racomitrium lanuginosum* (Hedw.) Brid., at different distances along transects in the prevailing wind direction, an important factor in the dispersion of atmospheric pollutants around the power plants (Ólafsdóttir et al., 2014a). We assessed plant health by measuring moss growth, moss mat depth and chlorophyll concentration.

We targeted this plant species due to its widespread distribution, dominance and sensitivity to atmospheric contaminants. The results were compared with a reference area away from any geothermal activity.

Materials and Methods

Study area and species

The Hengill geothermal area (110 - 115 km^2 (Franzson et al., 2010)) is located in the southern part of the western volcanic zone of Iceland and has no human settlement (Fig. 1). Two geothermal power plants are located 10 km apart on either side of Mt. Hengill, i.e. Nesjavellir (180 m a.s.l.) to the north in a small valley surrounded by low ridges, and Hellisheidi (260 m a.s.l.) to the south with Mosfellsheidi heath sloping toward the capital area, Reykjavik, in the west. Table 1 shows the main features of the power plants. The study area extends 4000 m from each of the two power plants (Fig. 1). The topography consists of variable volcanic features and surface geothermal manifestations; primarily pillow lavas and hyaloclastites, eruptive fissures, faults, fractures, basaltic volcanic rocks of various kinds, and fumaroles (Björnsson et al., 1986). The mean annual temperature (2011–2015) is 2.4°C at Hellisheidi and 2.1 °C at Nesjavellir (as measured at Hellisskard, approximately 3.76 km NW of Hellisheidi and 9.99 km SE of Nesjavellir geothermal power plants), with an average annual (2011–2015) precipitation of 2,400 mm at Hellisheidi and 2,800 mm at Nesjavellir (Icelandic Meteorological Office, unpublished 2016; Aradóttir, 2012). Prevailing winds are from the east (Icelandic Meteorological Office, unpublished data 2015), thus the potentially main receptor areas of the power plant emissions are to the west of the power plants. H_2S emission data from each power plant is presented in Table 1. We however point out that the average annual H_2S gas emission from each of the two power plants is similar for the years 2007 - 2015 (Table 1). Despite the differences in the power plants electricity output, we had anticipated the Hellisheidi geothermal power plant to emit much more H_2S gas (due to its big plant size) than Nesjavellir. However, the comparable H_2S gas emission levels between the two power plants may be due to the ongoing efforts of the geothermal power developer (Reykjavik Energy) that have contributed a 25% decrease in H_2S gas emissions from the Hellisheidi power plant (Juliusson, 2016). This is being done through gas re-injection (H_2S and CO_2) and sequestration as minerals in nearby, subsurface basaltic formations. Technical details of the process are explained in Gunnarsson et al., (2013); Juliusson et al., (2015); Aradóttir et al., 2015).

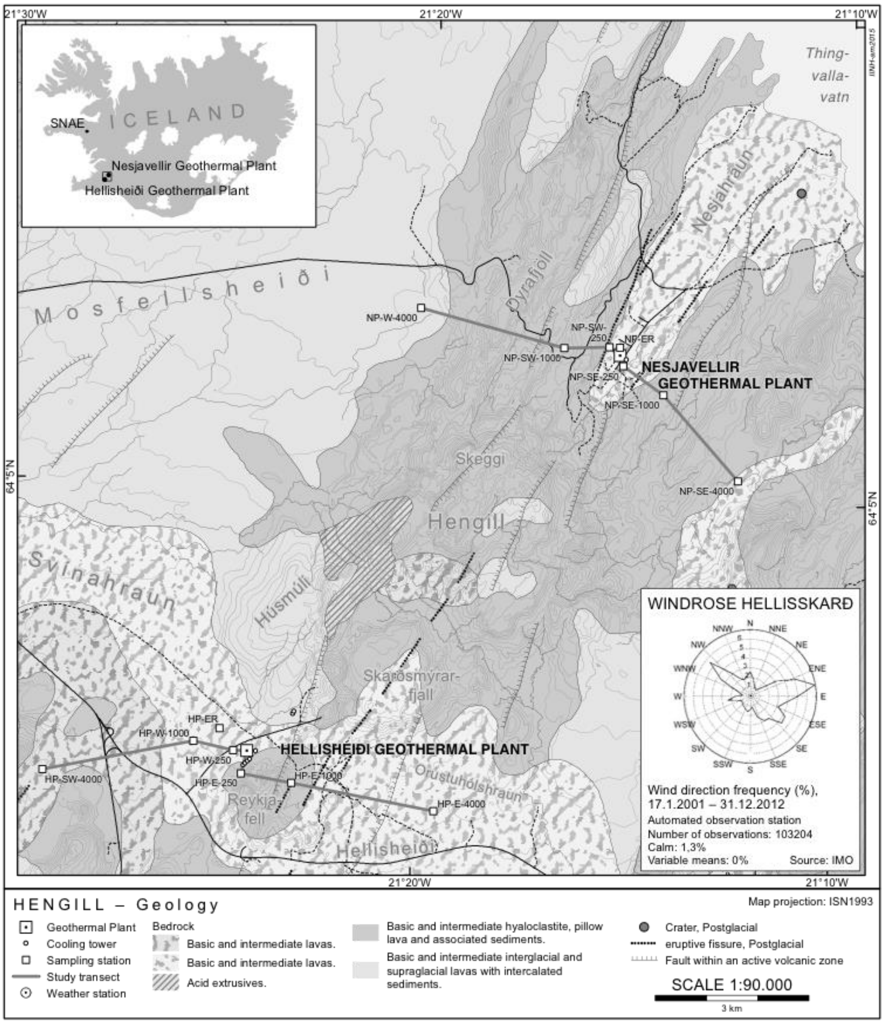
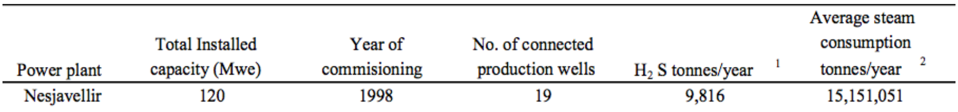


Figure 1. Location of the Hellisheidi and Nesjavellir geothermal power plants and the sampling stations along the east and west transects. Annual average (January 2001–December 2012) wind rose of the area is also shown (Icelandic Meteorological Office, Unpublished 2015). The extensively damaged areas around the power plants are shown as HP-ER for Hellisheidi and NP-ER for Nesjavellir.

Table 1. Summary data of the two power plants within the study areas. There is no data available on the emission rates of arsenic, boron, antimony and mercury in the air.



Power plant	Total Installed capacity (Mwe)	Year of commisioning	No. of connected production wells	H_2 S tonnes/year [1]	Average steam consumption tonnes/year [2]
Nesjavellir	120	1998	19	9,816	15,151,051
Hellisheidi	303	2006	35	10,072	33,554,886

[1] Averages for the years 2007 – 2015, (Sigurdardóttir and Thorgeirsson, 2015).

[2] Averages for the years 2013 – 2015, (Reykjavik Energy unpublished data, 2016).

Soils in the area are volcanic and commonly Brown and Gleyic Andosols and Leptosols (Arnalds, 2015). The vegetation mainly comprises moss heaths dominated by the moss *Racomitrium lanuginosum* on the lava fields, intersected by grassland and dwarf shrub heath vegetation in depressions and along rivers and ponds (Aradóttir, 2012).

Like most other moss species, *R. lanuginosum* lacks roots and a vascular system, it highly relies on atmospheric sources for nutrients and does not shed plant parts as easily as vascular plants (Dilks and Proctor, 1979; Nash and Wirth, 1988; Loppi and Bonini, 2000; Proctor, 2001). Mosses are therefore good indicators in assessment of atmospheric pollution (Loppi and Bonini, 2000) and are used as bio-monitors in documenting relative spatial and temporal deposition patterns of contaminants (Nash and Wirth, 1988; Harmens et al., 2015). The growth of *R. lanuginosum* is slow but variable, ranging between 5 and 15 mm per year (Tallis, 1964b; Jónsdóttir et al., 1995), and highly depends on the prevailing environmental factors (Armitage et al., 2012).

Study design, sampling and field measurements

The study design was based on a long-term on-going monitoring program by the Icelandic Institute of Natural History (IINH) of the impact of emissions from the Hellisheidi and Nesjavellir geothermal power plants on the surrounding moss heath (Helgadóttir, et al., 2013). Our study thus provides baseline information on ecosystem accumulation of emitted compounds and elements. To test our main hypothesis two transects were chosen in September 2013, based on the prevailing wind direction, i.e. one upwind (E) and the other downwind (W) from the cooling towers of the power plants, slightly modified by topographical features and future geothermal expansion plans (Fig. 1). Sampling stations were chosen along each transect at increasing distances from the geothermal power plants, at 250 m, 1000 m and 4000 m from the cooling towers, 6 stations per power plant, 12 in total (Fig. 1). The stations were located on convex lava ridges, exposed to wind and dominated by thick (at least 10 cm) and dense moss mats of *R. lanuginosum*. As such, they provide favorable growing conditions for *R. lanuginosum* because this species does not favour deep snow. Due to the topography of the area, the sampling stations varied in altitudes, between 138 m and 420 m above sea level. At each sampling station we systematically sampled along a 20 m sub-transect perpendicular to the main transect. Ten 10x10 cm squares were marked at 2 m intervals along these sub-transects.

At each 10x10 cm square moss damage was scored (brown or black colour) to obtain the frequency of damage for each station and the depth of the moss mat was measured to the nearest mm at the four corners and the mid-point of the square, from the moss surface to the moss-soil interface and the mean depth calculated. The growth rates i.e. shoot length increase and biomass increase were measured for the moss in three of the squares (at 0 m, 10 m and 18 m, in total 36 sampling points) for ten months (method description below). Upon completing all the *in situ* moss measurements in June 2014, moss samples were collected at each square for analysis of the concentrations of chlorophyll, sulphur, arsenic, boron, antimony and mercury. The moss samples were immediately stored in dark bags to preserve the chlorophyll content. The uppermost 10 cm layer of soil was also sampled beneath each moss sample, in order to determine primary accumulation of airborne substances (modified from Bussotti et al. (2003)) and for determination of some soil characteristics: pH, moisture, total carbon, and total nitrogen. These characteristics were considered as other environmental factors that could affect soils and ultimately plant health and were included in our statistical models as co-variables.

Additional sampling areas

To investigate if damaged moss patches (brown or black in colour) had higher concentrations of the emitted elements than healthy looking moss (a potential cause of their damage), we conducted a targeted systematic sampling at each of the sampling stations, where the first five damaged moss patches encountered along the sub-transect were sampled. In those patches we measured moss mat depth and chlorophyll concentrations, and took moss and soil samples to analyse for sulphur, arsenic, boron, antimony and mercury concentrations (60 sampling points in total).

Two additional areas close to the power plants (approximately 100 m^2 each) were surveyed (at 267 m and 830 m NNW of the cooling towers at Nesjavellir and Hellisheidi, respectively), characterised by mats of extensively damaged moss (mainly brown and black moss). At Nesjavellir the damage may originate from the early operational years of the power plant, when hot emissions were ejected using lower chimneys than cooling towers (Einar Gunnlaugsson- Reykjavik Energy pers. comm. 2016), and is therefore probably related to combined thermal and emission pollution. At Hellisheidi, overflows of spent geothermal fluid were suspected to have been the cause in combination with thermal and emission pollution (Einar Gunnlaugsson- Reykjavik Energy pers. comm. 2016). Moss mat depth measurements and moss and soil samples were collected at 10 points along 20 m transects as described above.

A reference area outside the influence of the power plant emissions was chosen for comparison at Raudhalsahraun (22.2640° W 64.8483°N, 331 m a.s.l), 99 km away NNW of the study area. The area is well outside the influence of the power plant emission. The mean annual precipitation and temperature is 1,181 mm (measured at Hítardalur, approximately 11.5 km SE of Raudhalsahraun) and 5°C (measured at Hafursfell, 12.9 km WSW of Raudhalsahraun) for the years 2011 – 2015 (Icelandic Meteorological Office, unpublished data 2016). In a similar way to the sampling design above, measurements were collected along a 20 m transect (ten 10x10 cm squares at 2 m intervals, moss growth in 3 squares).

Finally, in July 2014 we had the opportunity to sample moss and soil in the high Arctic Svalbard well out of range of both geothermal activity and volcanism. We selected an area (165 m^2) at Skansenbukta (N 78.51881° E 016.00052°), central Svalbard, that was dominated by *R. lanuginosum*. The mean annual precipitation and temperature at the closest meteorological station (Svalbard Lufthavn) for 2011 – 2015 were 499.34 mm and -2.6 °C, respectively. Six moss and soil samples were collected following the same protocol described above. All samples were transported to the laboratory for processing.

For ease of reference, we grouped our data sets into five, i.e. systematic sampling area (for the overall hypothesis), damaged moss patches areas, reference area, the extensively damaged areas and Svalbard.

Moss growth measurements

Moss growth was measured at three sampling points within the systematic sampling stations and at the reference area by transplanting trimmed shoots into the moss mats (Jónsdóttir et al., 1999; Armitage et al., 2012). Fresh moss shoots longer than 30 mm were removed from mats of *R. lanuginosum* collected at the Raudhalsahraun area (reference area). For each sampling point, 39 in total (three at each sampling station along each transect and power plant plus three at the reference area), twenty moss shoots were trimmed to 30 mm length. Ten of these shoots were weighed (fresh weight) and placed in a tagged open-ended netlon bag as described in Armitage et al. (2012) and carefully inserted into the moss mat at the sampling point in September 2013 (t_0), held in place using plastic coated wire prongs. The other ten shoots were used to determine the ratio between fresh and dry weight (after drying to a constant weight at 70°C) and calculate the dry weight of the transplanted shoots (Jónsdóttir et al., 1999). The transplanted shoots were left *in situ* for 10 months, until June 2014 (t_1). Shoot growth was measured as shoot length increase in excess of the original 30 mm and as biomass increase. The transplanted shoots were dried at 70°C to obtain the dry weight at time t_1. Biomass increase was calculated by subtracting the calculated dry weight at time t_0 from the dry weight at time t_1. Shoot length increase was estimated from each of the ten shoots per bag and biomass increase was calculated collectively for the ten shoots per bag. 'Shoot turnover' for the ten months was estimated for three squares along the 20 m transect at each station (at 0 m, 10 m and 18 m: same position as for the shoot length increase measurements) as the proportion of moss mat depth comprising of the last ten months' growth: [Mean growth (in mm) / mean moss depth (mm)] × 100 = shoot turnover (%). This measure gives an indication of the relative rate of shoot biomass turnover, on the assumption that the moss mat remains in a steady state with uniform rate of compression as moss material decomposes and buries within the mat (Armitage et al., 2012).

Sample treatment and laboratory analysis

Each moss sample was split into two, one for chlorophyll analysis and the other for sulphur and trace element analysis. For all samples, up to 3 cm of the shoot apices were removed, washed in distilled water and dried at room temperature in the dark.

For the chlorophyll determination, each sample was milled, weighed and split into two sub samples. One for chlorophyll content analysis and the other sample for dry weight determination (after oven drying to a constant weight at 70°C for 24 hours), so that samples used for chlorophyll analysis were not exposed to high temperature and possible break down of chlorophyll. Then, 10 ml of 96% ethanol was added to 0.5 g of each sample and the mixture hand shaken for 15 seconds. The samples were then covered by aluminum foil to prevent light exposure and allowed to stand for 24 hours at 6°C in darkness and centrifuged for 10 min at 1000 revolutions per min. 3.5 ml samples were extracted and transferred to 4 ml cuvettes for analysis (modified from Sumanta et al., 2014). Light absorbance at wavelengths of 750 nm, 663 nm and 652 nm was measured by a spectrophotometer (HACH LANGE UV Visible Spectrophotometer, DR 5000) at the Institute of Freshwater Fisheries in Iceland. Chlorophyll content in mg/g dry weight was calculated according to Arnon, (1949).

For sulphur and trace element analyses moss samples were oven dried at 40°C for 48 hours to constant weight and pulverized using agate mortars. The concentrations of sulphur, arsenic, boron, antimony and mercury were analysed using standard analytical procedures at the internationally accredited ALS Scandinavia labs in Luleå, Sweden. Prior to analysis, samples were acid digested (in 5 ml conc. HNO_3 + 0.5 ml 30% H_2O_2) in closed teflon containers in a microwave. Element analyses were conducted using an Element 2 ICP MS. The analyses were carried out according to (modified) USEPA methods 200.8 (U.S.EPA, 1994) and SS EN ISO 17294 parts 1 (ISO, 2005) and 2 (ISO, 2003).

Soil analysis was done on samples that were pooled and thoroughly mixed for each sampling station; three sub-samples were drawn out of each pooled sample. Each soil sample was then split into two, one for the analysis of sulphur, arsenic, antimony, boron and mercury, and the other for analysis of soil characteristics i.e. total carbon (% C), total nitrogen (% N), pH and moisture. The soil samples were dried at 40°C for 48 hours to constant weight, sieved through 2 mm and analysed for sulphur, arsenic, antimony, boron and mercury using the same protocols as for the moss shoots. Total elemental carbon and nitrogen was determined on ball-milled soil samples (<0.1 mm), dried at 50°C for 24 hours, using the Flash 2000 Elemental Analyser (Thermo Scientific, Italy). For soil pH, soil solution was extracted from 5 g (<2 mm) of 96 hours air-dried soil in 25 ml de-ionized water, by shaking it for two hours and allowed to settle for 8 hours before measuring pH (Blakemore et al., 1987). Soil moisture (%) by mass was obtained after oven drying 10 g of fresh soil at 105°C for 24 hours to constant weight.

Moss shoot samples were analysed at the ALS Scandinavia labs in Luleå, Sweden. Analyses of soils characteristics were conducted at the University of Iceland, except for total carbon and nitrogen for the Svalbard samples, which were performed at the University Centre in Svalbard (UNIS), Longyearbyen, Norway. The concentrations of sulphur, arsenic, boron, antimony and mercury in moss and soil were expressed as mg/kg on dry weight basis and in% by dry weight for total carbon and total nitrogen. Procedural blanks were below the minimum detection level. Accuracy was checked through analysis of standard in house reference materials for soil (ALS Labs, Sweden; University of Iceland, Iceland; UNIS, Longyearbyen) and peach leaves (NIST 1547) (National Institute of Standards and Technology, Gaithersburg, MD, USA; (Rodushkin et al., 2008) and obtained more than 95% recoveries. Because of the high sulphur proportions reported from power plant emissions (as H_2S gas, see Table 1), estimates of sulphur accumulation for moss shoots were calculated for the systematic sampling areas based on the biomass of the ten moss shoots at the three measurement squares per sampling station at the end of the survey and multiplied by the corresponding shoot concentrations of sulphur per measurement square.

Data analyses

For visual assessment of our data, we performed a Non-Metric Multidimensional Scaling (NMDS) ordination (Oksanen et al., 2015), using Euclidean dissimilarity as a distance metric for the three matrices of all response data i.e. element concentrations in moss, element concentrations in soil, and moss growth and characteristics on the predictor variables (distance, direction and location).

Element concentrations in moss shoot tissues (sulphur, arsenic, boron, antimony and mercury), sulphur accumulation and moss measurements (i.e. moss depth, shoot length increase, shoot turnover, biomass increase, and chlorophyll) were separately analysed as response variables using Linear Mixed effects Models (LMM). Distance from the power plant (250 m, 1000 m, 4000 m), location (Nesjavellir and Hellisheidi), direction from the power plants (upwind, E and downwind, W) and all possible interactions were included as fixed factors. These interactions made sense from a biological point of view, as we may expect to see different element accumulation patterns and effects on plant growth with increasing distances from the power plant depending on the position relative to prevailing winds (direction), and influence of the power plant (location); similarly, responses upwind and downwind may differ depending on the influence of the power plant (location). The elevation of the sampling stations (altitude) and soil characteristics i.e. soil pH,% soil nitrogen,% soil carbon,% soil moisture and soil chemistry (sulphur, arsenic, boron, antimony and mercury concentration), were included as co-variates; and ‘sampling station’ as a random factor to account for the sampling design. For shoot length increase, ‘sampling bag’ was included as random factor nested within ‘sampling station’. The co-variates were included one at a time; the best-fitted model was selected based on the lowest AIC (Akaike´s Information Criterion) value, provided that inclusion of an additional parameter in the model reduced the AIC value by more than 2.0. Due to less complete design of soil sampling (sample pooling per station), we separately fitted Linear Models (LM) for the concentrations of sulphur, arsenic, boron, antimony and mercury in soil, with the same predictor variables as in the LMMs above (distance, location and direction, and their interactions) and covariates. To satisfy the model assumptions, sulphur, arsenic, boron, antimony and mercury concentrations in moss shoots and sulphur in soil were log-transformed. To avoid negative values for models with moss and soil element concentrations of values below detection (whose data value had been assumed as zero for statistics), data were log (x+v) transformed with v representing the minimum value of the sampled data.

The Variance Inflation Factor (VIF) was calculated for all variables included in the models to assess multicollinearity. Highly collinear co-variables were not included in the models, so that all VIF values were below 3 in the final models (Zuur et al., 2010). Standard model diagnostics were used to check the residuals (for normality and homoscedasticity). The optimal model for each response variable was identified by selecting the lowest AIC after a series of model sub-sets were run. The final models chosen were reduced with our predictors preserved as the minimum. Non-significant interactions were dropped from the models to allow interpretation of the independent terms. We used the R software version 3.2.2 for all data analyses (R Development Team, 2010). The models were run using the functions, lmer in the lme4 packages (Bates et al., 2014) for the LMM and lm in the MASS package in R (Ripley et al., 2015) for linear models. Pairwise correlations were performed for the following variables: moss growth, characteristics and element concentrations in moss and in soil, and tested for significance using Pearson's product moment correlations. Lists of models and test statistics used, and summary soil characteristics, elevation data and correlation matrices are presented in Tables S1 – S9 and appendix S1. The reference area and Svalbard data were not included in any of the statistical analyses.

Results

The NMDS ordination revealed that element concentrations in soil and moss and the moss traits differed in relation to location (Fig. 2); there was a good separation between Nesjavellir, Hellisheidi and reference sampling stations indicating dissimilarities (Figs. 2e and h). In relation to the different distances, the x-axis (Fig. 2) of the ordination plot reflected an overall concentration gradient in moss and soil from left (250 m) to right (through 1000 m to 4000 m). Such overall pattern related to direction was not apparent in the ordination plots, but the stations seem to be mainly ordinated along the x-axis.

The frequency of moss shoot damages around the power plants was low in our assessment and could not be related to any of the predictors. Relative to mosses from the reference area in Raudhalsahraun (Table 4), moss from the Hellisheidi and Nesjavellir geothermal power plant areas (Tables 2) had considerably higher concentrations of sulphur (Fig. 3), arsenic and mercury. Concentrations of all other elements in moss were low at all sites. The element concentrations in mosses were generally higher around Hellisheidi than Nesjavellir (Fig. 3ai-ii, Tables 2, S1). Opposite to what we predicted, element concentrations increased with distance around Hellisheidi but were, as expected, higher downwind (W) than upwind (Fig. 3a-iii, Table S1). At Nesjavellir, however, the concentration patterns agreed with our expectations in response to distance, but not in response to direction (no strong overall trend), Fig. 3a-ii, Table S1. Sulphur accumulation in moss shoots tended to be higher closer to the power plants than further away (marginally significant effect of distance; Table S1, Fig. 3b-i). Direction and location did not affect sulphur accumulation in *R. lanuginosum* shoots (Table S1, Fig. 3bii-iv).

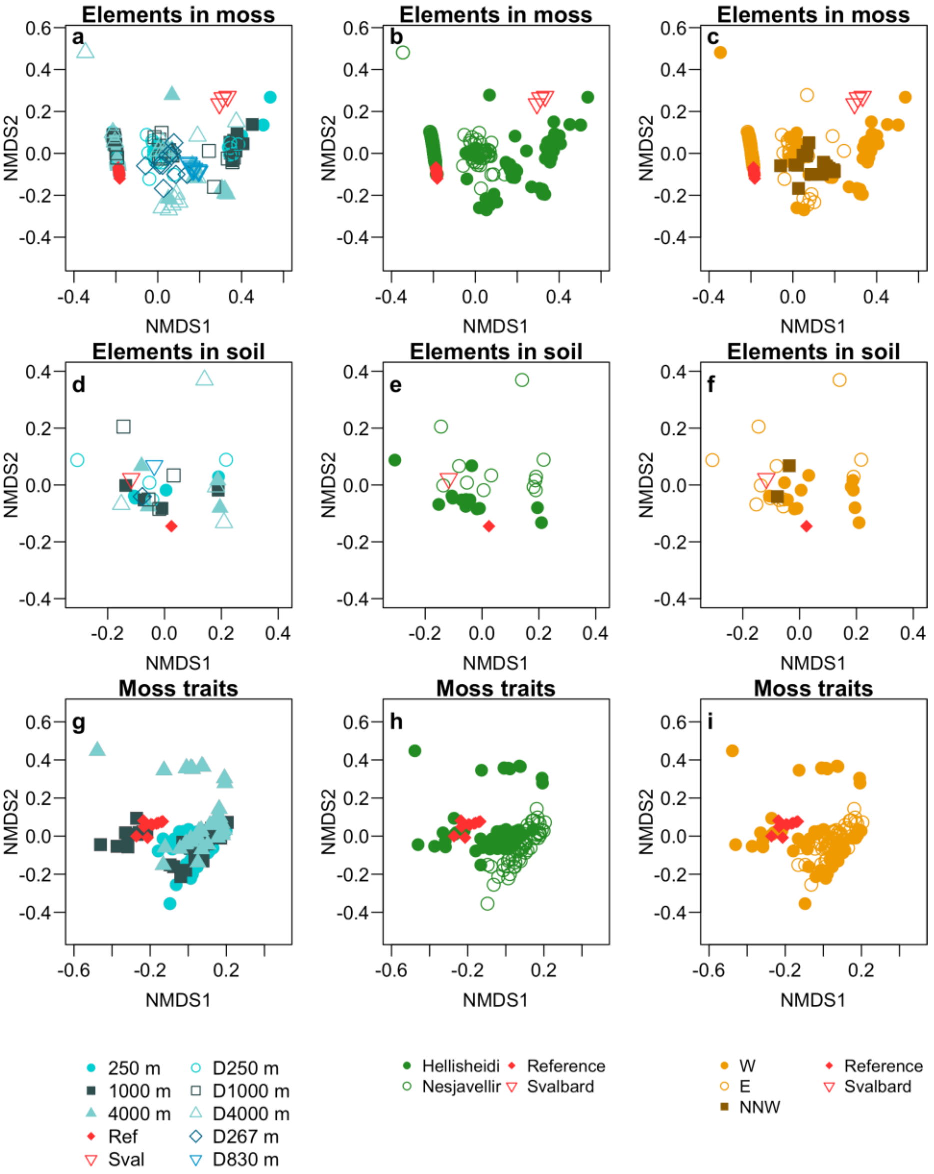
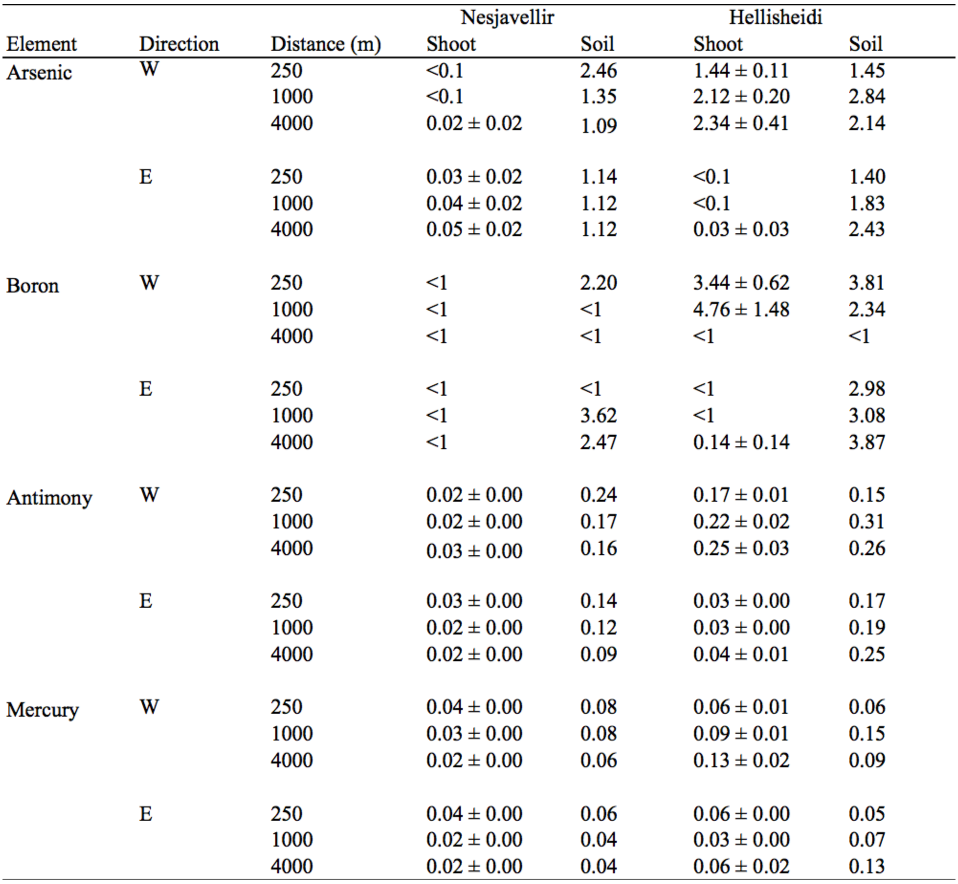


Figure 2. Nonmetric Multidimensional Scaling (NMDS) ordinations of (2a – c) sulphur, arsenic, boron, antimony and mercury concentrations in moss shoots, (2d-f) sulphur, arsenic, boron, antimony and mercury concentrations in soil and 2g – i) the various moss traits measured. Moss growth and characteristics data from the damage patch targeted sampling and extensively damaged sampling areas were not included in the NMDS plot as they were few. NNW in the direction represents samples from the extensively damaged sampling areas.

In soils, sulphur and mercury concentrations were much higher at the reference area (Fig. 4a, Table 4) than around Hellisheidi and Nesjavellir (Tables 2), while the other elements, i.e. arsenic, boron and antimony showed the opposite pattern, with higher soil concentrations around power plants. Sulphur concentrations in soils were significantly higher at Nesjavellir than Hellisheidi (Table S2, Fig, 4a). On the contrary, the other elements were at higher concentrations in soils around Hellisheidi than Nesjavellir, except for boron, the concentrations of which did not differ between locations (Table S2). Around Hellisheidi, element concentration patterns did not vary in response to distance and direction (Fig. 4b i, iii). However, for Nesjavellir, most of the elements responded differently to the predictors. Sulphur concentrations in soil decreased with distance (Fig. 4b ii), while the effect of direction was not significant. Additionally, antimony concentrations in soil at Nesjavellir decreased with distance and tended to be higher to the west of the power plant than east. Distance and direction did not affect arsenic, boron and mercury concentrations (Table S2).

The growth of moss shoots (biomass increase, Fig. 5) was overall higher at the reference area than at Hellisheidi and Nesjavellir, while the other moss response variables were comparable (Tables 3, 5, Fig. 5i-ii). However, the mosses at the reference area had lower chlorophyll concentrations and the moss mats were thinner than those around the power plants (Tables 3, 5). In general, moss biomass and shoot length increase, moss mat depth, chlorophyll concentrations and moss shoot turnover values were lower around Nesjavellir than at Hellisheidi (Table 3, Fig. 5i-ii, S5). Around Nesjavellir, moss mat was thinner close to the power plant than far away, and as expected its depth increased with distance from the power plant. Deeper moss mats were measured to the east of Nesjavellir power plant (upwind) than west (Table S5). Moss biomass also increased with distance at Nesjavellir and was also higher in the direction east than west (Fig. 5b). Moss shoot length increase did not vary with distance from the power plant but showed a decrease in the direction west of the Nesjavellir power plant than east. Moss shoot turnover and chlorophyll concentrations showed opposite patterns with distance, they decreased away from the power plant and did not vary with direction. These growth trends imply thinner moss shoots close to the power plants and thicker shoots further away.

Table 2. Concentrations of arsenic, boron, antimony and mercury in Racomitrium lanuginosum *shoots and soil (mg/kg dry weight), collected at the systematic sampling stations at different distances along transects to the east and west of the two power plants, Hellisheidi and Nesjavellir, in the Hengill geothermal area in Iceland (n = 10 shoots per station, n = 1 soil per station). Some element concentrations were below the detection limits: 0.1 mg/kg for arsenic concentrations in shoots and 1 mg/kg for boron concentrations in shoots and soil. Elements concentrations for* R. lanuginosum *are given as mean ± SE and only as mean for soils due to sample pooling per station (see methods).*



			Nesjavellir		Hellisheidi	
Element	Direction	Distance (m)	Shoot	Soil	Shoot	Soil
Arsenic	W	250	<0.1	2.46	1.44 ± 0.11	1.45
		1000	<0.1	1.35	2.12 ± 0.20	2.84
		4000	0.02 ± 0.02	1.09	2.34 ± 0.41	2.14
	E	250	0.03 ± 0.02	1.14	<0.1	1.40
		1000	0.04 ± 0.02	1.12	<0.1	1.83
		4000	0.05 ± 0.02	1.12	0.03 ± 0.03	2.43
Boron	W	250	<1	2.20	3.44 ± 0.62	3.81
		1000	<1	<1	4.76 ± 1.48	2.34
		4000	<1	<1	<1	<1
	E	250	<1	<1	<1	2.98
		1000	<1	3.62	<1	3.08
		4000	<1	2.47	0.14 ± 0.14	3.87
Antimony	W	250	0.02 ± 0.00	0.24	0.17 ± 0.01	0.15
		1000	0.02 ± 0.00	0.17	0.22 ± 0.02	0.31
		4000	0.03 ± 0.00	0.16	0.25 ± 0.03	0.26
	E	250	0.03 ± 0.00	0.14	0.03 ± 0.00	0.17
		1000	0.02 ± 0.00	0.12	0.03 ± 0.00	0.19
		4000	0.02 ± 0.00	0.09	0.04 ± 0.01	0.25
Mercury	W	250	0.04 ± 0.00	0.08	0.06 ± 0.01	0.06
		1000	0.03 ± 0.00	0.08	0.09 ± 0.01	0.15
		4000	0.02 ± 0.00	0.06	0.13 ± 0.02	0.09
	E	250	0.04 ± 0.00	0.06	0.06 ± 0.00	0.05
		1000	0.02 ± 0.00	0.04	0.03 ± 0.00	0.07
		4000	0.02 ± 0.00	0.04	0.06 ± 0.02	0.13

Around Hellisheidi, the effects of distance on moss characteristics were not always clear (Table S5). For instance, moss mat depth was significantly deeper at 250 m than at 1000 m but became deeper again at 4000 m away from the power plant (Table 3, S5). Chlorophyll concentrations increased from 250 m to 1000 m and decreased again at

4000 m away from the power plant (Table 3, S5). Shoot turnover significantly increased with distance, while shoot length increase and biomass increase did not vary with distance (Tables 3, S5, Fig. 5). However, biomass increase was significantly higher in the direction W than E while shoot length increase did not vary with direction. Contrary to our expectations and Nesjavellir results, the findings at Hellisheidi were different. Higher values for moss growth and the other characteristics were measured to the west (downwind) of the Hellisheidi power plant than east (Table S5, Fig. 5b).

Element concentrations in moss tissues and soils, and the moss response variables at the geothermal area varied differently with the predictors (location, distance, direction and their interactions) and there were not always clear trends with predictors as hypothesized. Addition of the co-variates in general did not affect the strength and direction of the effects of our predictors (Tables S1, S2 and S5), and improved most models by explaining parts of the un-accounted variations.

Element concentrations were generally much higher in damaged moss patches than in healthy-looking mosses. Overall, trends in element concentration in mosses and soils associated to damaged moss patches were similar to the healthy mosses (systematic sampling), for details see Appendix S1. There was not any marked difference in moss mat depth between the healthy and damaged moss patch sampling areas, but chlorophyll concentrations were much lower in the damaged moss as expected (Appendix S1: Table A3, A3a).

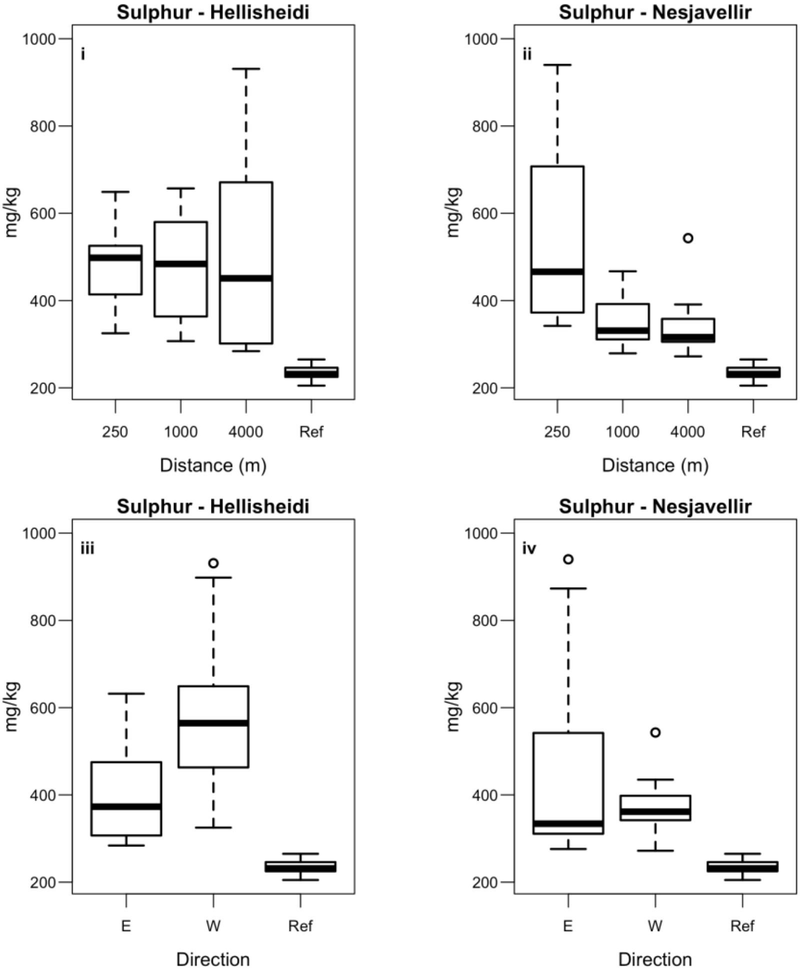


Figure 3a. Concentrations of sulphur in Racomitrium lanuginosum *shoots at Hengill compared to a reference area at 99 km away (Ref) at the systematically sampled areas around Hellisheidi and Nesjavellir power plants, (3i - ii) at different distances, and (3iii -iv) different directions from the power plants. n = 10 per station. Sulphur concentration: significantly different for Hellisheidi compared to Nesjavellir ($t_{(120)}$ = -8.74 p = <0.001), for the different distances at Hellisheidi compared to 250 m ($t_{(60)=}$ -4.96 p = <0.001 for 1000 m and $t_{(60)}$ = -6.40 p = <0.001 for 4000 m), for direction E compared to W ($t_{(60)}$ = -2.17 p = 0.034) at Hellisheidi, for the different distances at Nesjavellir compared to 250 m ($t_{(60)=}$ -11.60 p <0.001 for 1000 m and $t_{(60)}$ = -12.42 p = <0.001 for 4000 m) and for direction E compared to W ($t_{(60)}$ = -9.53) p <0.001) at Nesjavellir.*

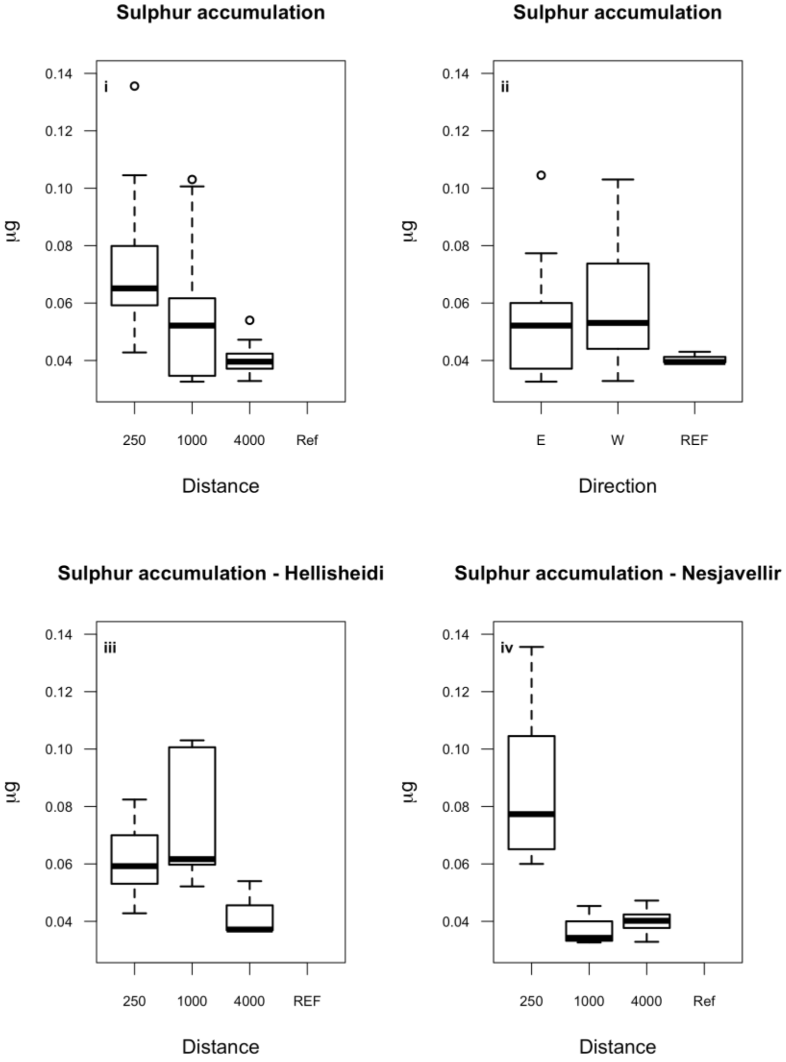


Figure 3b. Sulphur accumulation in R. lanuginosum *shoots at Hengill at the systematically sampled areas compared to a reference area (REF) at 99 km away around both Hellisheidi and Nesjavellir power plants in relation to: (3b-i) distance from the power plants, (3b-ii) direction from the power plants and (3b-iii, iv) distance from the power plants. n = 10 per station.*

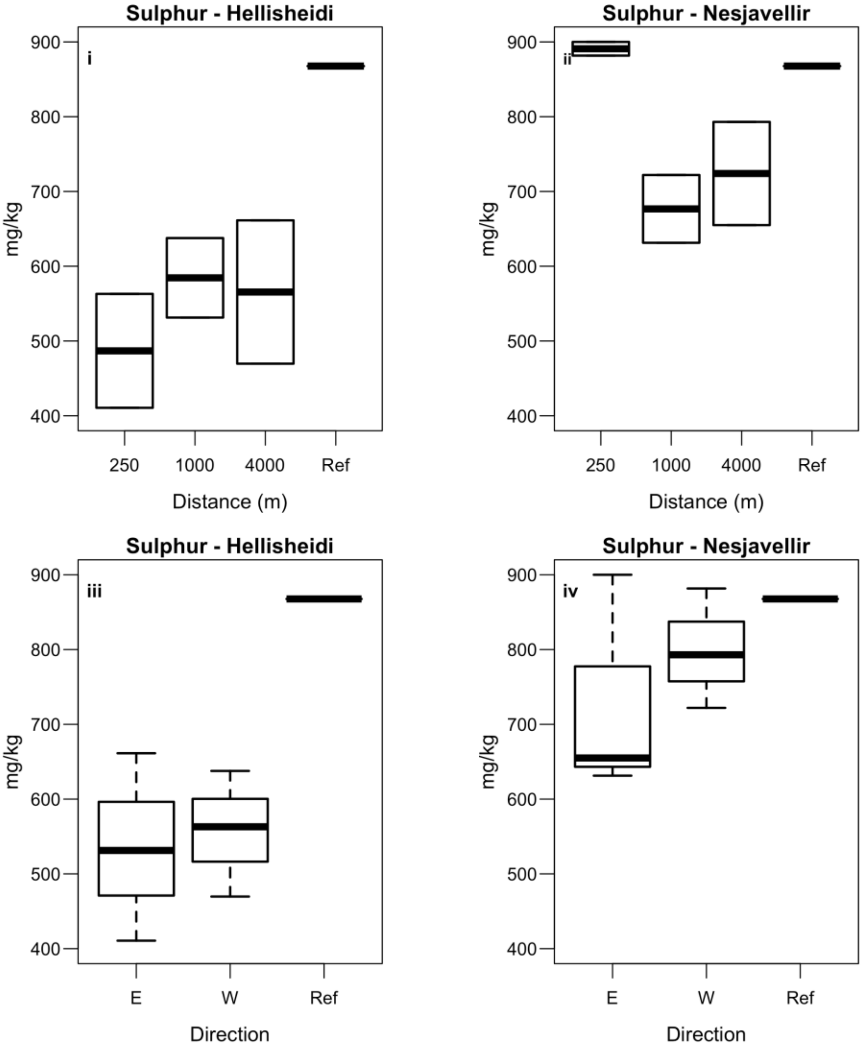


Figure 4. Concentrations of sulphur in soil at Hengill compared to a reference area at 99 km away (Ref) at the systematically sampled areas around Hellisheidi and Nesjavellir power plants, (4i-ii) at different distances, and (4iii-iv) different directions from the power plants. n = 1 per station. Sulphur concentration: significantly different for Hellisheidi compared to Nesjavellir ($t_{(6)} = 8.36$ $p = <0.001$.

Table 3. Characteristics (mean ± SE) of Racomitrium lanuginosum *at the systematic sampling stations around Hellisheidi and Nesjavellir power plants in Iceland. n = 6 per station. For shoot length increase, n = 3 per station.*

Moss characteristics	Direction	Distance (m)	Nesjavellir	Hellisheidi
Moss mat depth (cm)	W	250	19.27 ± 2.47	28.17 ± 2.05
		1000	22.96 ± 1.87	17.50 ± 1.37
		4000	25.59 ± 1.87	28.63 ± 2.56
	E	250	20.03 ± 1.72	30.23 ± 0.76
		1000	27.56 ± 2.36	27.52 ± 0.93
		4000	33.20 ± 2.36	26.90 ± 2.53
Shoot length increase (cm)	W	250	0.46 ± 0.25	0.63 ± 0.09
		1000	0.50 ± 0.06	0.60 ± 0.06
		4000	0.37 ± 0.03	0.10 ± 0.00
	E	250	0.43 ± 0.03	0.50 ± 0.06
		1000	0.40 ± 0.06	0.53 ± 0.03
		4000	0.40 ± 0.06	0.53 ± 0.03
Shoot turnover (%)	W	250	3.00 ± 1.71	2.41 ± 0.29
		1000	2.39 ± 0.61	3.56 ± 0.63
		4000	1.64 ± 0.19	0.52 ± 0.16
	E	250	2.08 ± 0.22	1.70 ± 0.31
		1000	1.63 ± 0.35	1.82 ± 0.16
		4000	1.13 ± 0.20	2.14 ± 0.42
Chlorophyll concentration mg/g	W	250	0.04 ± 0.01	0.06 ± 0.01
		1000	0.05 ± 0.02	0.67 ± 0.12
		4000	0.02 ± 0.01	0.06 ± 0.01
	E	250	0.18 ± 0.03	0.04 ± 0.05
		1000	0.11 ± 0.02	0.04 ± 0.00
		4000	0.04 ± 0.01	0.04 ± 0.01

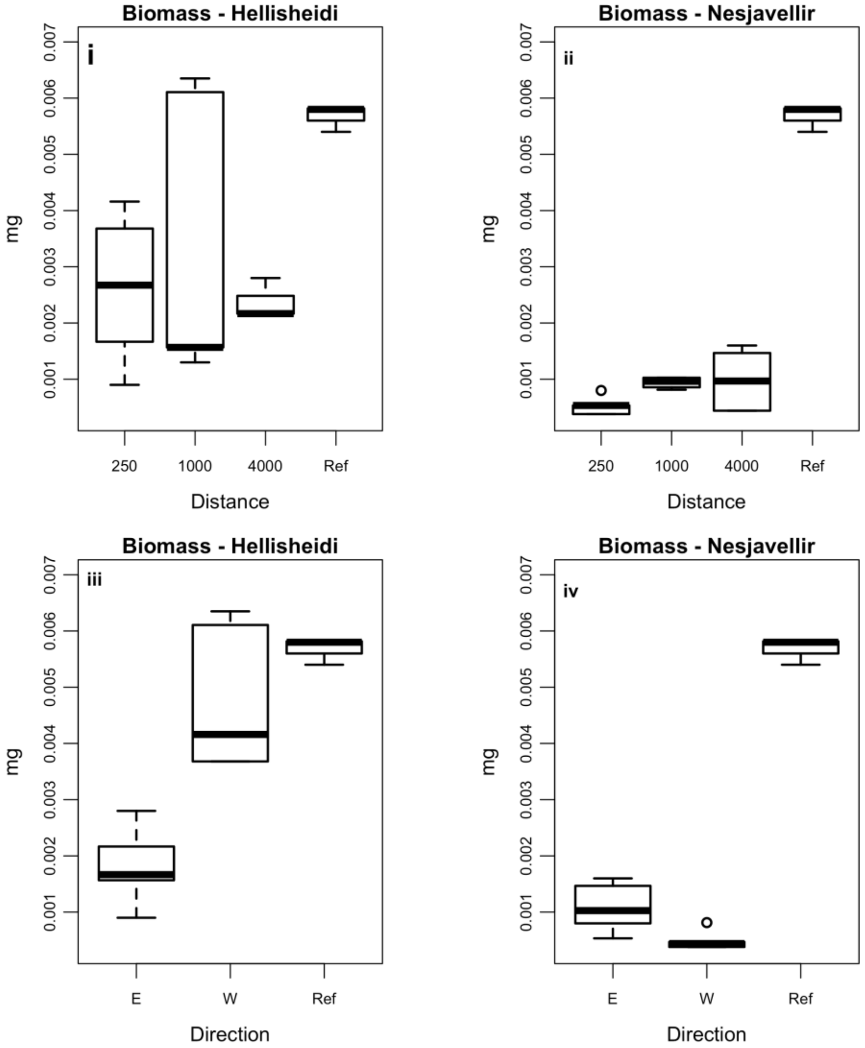
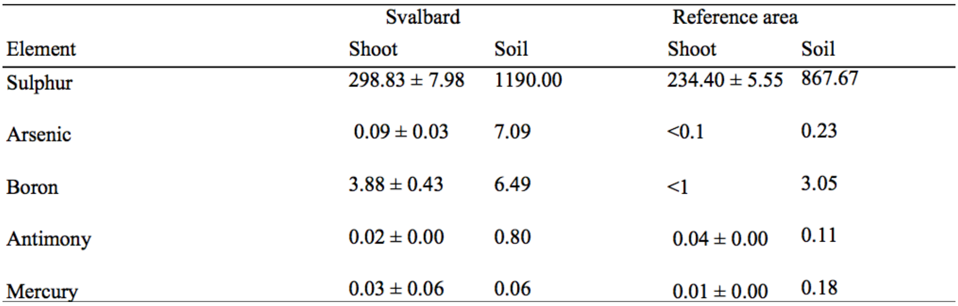


Figure 5. The biomass increase of Racomitrium lanuginosum *shoots at Hengill at the systematically sampled areas compared to a reference area (Ref) at 99 km away around Hellisheidi and Nesjavellir power plants, (5i - ii) at different distances, and (5iii -iv) different directions from the power plants. n = 3 per station. Biomass increase: significantly different for Hellisheidi compared to Nesjavellir (*$t_{(11)} = -4.40$ *p = 0.001), for the different distances at Nesjavellir compared to 250 m (*$t_{(6)} = 3.13$ *p = 0.020 for 1000 m and* $t_{(6)} = 2.84$ *p = 0.030 for 4000 m), for direction E compared to W (*$t_{(6)} = -3.53$ *p = 0.012) at Nesjavellir and direction E compared to W (*$t_{(6)} = -6.96$ *p <0.001) at Hellisheidi.*

Table 4. Concentrations of arsenic, boron, antimony and mercury in Racomitrium lanuginosum *shoots and soil (mg/kg dry weight), collected along transects in Skansenbukta, Svalbard (n = 6 shoots, n= 1 soil) and the Raudhalsahraun Reference area in Iceland (n = 10 shoots, n = 1 soil). Some concentrations were below the detection limits in shoots: 0.1 mg/kg arsenic and 1 mg/kg boron. Elements concentrations for* R. lanuginosum *are given as mean ± SE and only as mean for soils due to sample pooling per station (see methods).*



	Svalbard		Reference area	
Element	Shoot	Soil	Shoot	Soil
Sulphur	298.83 ± 7.98	1190.00	234.40 ± 5.55	867.67
Arsenic	0.09 ± 0.03	7.09	<0.1	0.23
Boron	3.88 ± 0.43	6.49	<1	3.05
Antimony	0.02 ± 0.00	0.80	0.04 ± 0.00	0.11
Mercury	0.03 ± 0.06	0.06	0.01 ± 0.00	0.18

Table 5. Characteristics (mean ± SE) of Racomitrium lanuginosum *at the extensively damaged moss areas around Hellisheidi (HP-ER) and Nesjavellir (NP-ER) power plants and the Raudhalsahraun reference area in Iceland. n = 10 per location. For shoot length increase at the reference area, n = 3. NA- Not Available.*

Moss characteristics	Nesjavellir	Hellisheidi	Reference area
Moss mat depth (cm)	11.39 ± 1.22	23.18 ± 3.36	21.76 ± 1.45
Chlorophyll concentration (mg/g)	0.01 ± 0.00	0.03 ± 0.08	0.01 ± 0.02
Shoot length increase (cm)	NA	NA	0.57 ± 0.03
Shoot turnover (%)	NA	NA	2.94 ± 0.18

In the extensively damaged areas, sulphur concentration in moss tissues was overall much higher than at our regular systematic sampling areas indicating input from the geothermal power plants (Appendix S1: Table A4, Fig. 3). For element concentrations in soil, sulphur and boron were elevated at the systematic sampling areas more than at the extensively damaged areas; all other element concentrations in soil between the two areas did not differ (Tables 2, A4). As expected, moss mat depth and chlorophyll concentrations showed generally lower values at the extensively damaged areas than the systematically sampled areas around the power plants (Tables 3, 5). For Svalbard mosses, element concentrations were distinct to all other sampled mosses in Iceland (NMDS plot; Fig. 2a – c). Mosses from Svalbard showed 20% lower sulphur concentrations than those at the systematic sampling areas (Table 4 and Fig. 3). In soil, element concentrations were much lower at the systematically sampled areas than in Svalbard (Tables 2, 4, Fig. 4), although this difference was not apparent in the NMDS plot (Fig. 2d - f).

Comparison of element concentration in moss and soils

In general, all measured element concentrations were higher in soils than moss tissues at all sampling areas around the two power plants and at the reference area and Svalbard, probably due to the recalcitrant nature of moss shoots, inferring slow decomposition and mineralization rates and resulting in element enrichment in the soils. However, in the damaged moss patches (mostly 250 m away from power plants) and the extensively damaged areas, considerably higher sulphur was found in moss tissues than soil (Appendix S1: Tables A1 and A4).

Correlations between element concentrations in moss, soil and moss characteristics

The element concentrations in moss at the systematic sampling and damaged moss patches did not reveal any significant correlations with element concentrations in soil (correlation values were generally below 0.5, details in Tables S7 –S8). This indicated that element concentrations in moss were not related to concentration of those particular elements in the soil and vice versa. Biomass increase was positively correlated to arsenic and antimony concentrations in moss tissues, which corresponds to our previous finding of high element concentrations and high biomass increase around Hellisheidi, 4000 m away from the power plant. Other moss characteristics were not correlated with element concentrations in either moss tissue or soil. (Table S7).

At the extensively damaged areas close to the power stations, most elements in plants were negatively correlated with soil element concentrations (correlation values were generally above - 0.5, Table S9). This may imply partially different sources of the elements in the soil (bedrock) and the moss (geothermal emission). Furthermore, moss mat depth and chlorophyll concentrations were all negatively correlated with arsenic, boron, antimony and mercury concentrations in the soil, but positively correlated with sulphur concentrations in moss (Table S9). The negative correlations may probably indicate harmful effects of the elements to moss characteristics while the positive sulphur correlations may be linked to some plant benefits.

Discussion

Evaluating the concentration patterns of geothermally emitted elements, using bio-indices such as plants and soils, in relation to distances from the power plants (Bargagli et al., 1997; Bussotti et al., 1997; Loppi, 2001; Mutia et al., 2016) and prevailing wind direction (Ólafsdóttir et al., 2014a) provides a robust indication of their deposition and accumulation into ecosystems; an important aspect in assessing their potential phytotoxicity at elevated concentrations. In relation to distance and direction from the Hengill geothermal power plants in Iceland, our study findings reveal some patterns of element concentrations in the dominant moss and soils, and affected moss characteristics and growth providing some support for the main hypothesis, that the power plant emissions deposit and accumulate in ecosystems within their vicinity with some impacts.

The trends of sulphur, arsenic, boron, antimony and mercury concentrations in moss and soil with our predictors (distance, direction and location) were in many cases variable but often followed our expectations, particularly around the Nesjavellir geothermal power plant. The patterns of moss sulphur accumulations with distance and direction (although only marginally significant) in the vicinity of power plants provide some indirect indication of sulphur accumulation in moss from the power plants. Further, the trend of high sulphur concentrations in Nesjavellir soils may be explained as a transfer of sulphur from the decaying moss to soil (bio-accumulation) over the longer period that

Nesjavellir power plant (since 1998) has been operating than Hellisheidi (since 2006). Besides, the trace element variations (generally higher at Hellisheidi than Nesjavellir) may be explained by the different steam consumption rates and subsequent emission rates for each power plant. We base this argument on the understanding that the Hellisheidi geothermal power plant has a higher steam consumption rate (see Table 1 for data) than Nesjavellir due to its capacity. Since the concentrations of arsenic, boron, antimony and mercury in emissions from the power plants are not known, we assume that the trace element concentrations, if any, would be the same for each power plant, and because there is more steam needed for the power plant at Hellisheidi, larger amounts of these elements are emitted. However, we caution that this interpretation is based on an assumption and warrants further studies for confirmation or rebuttal.

Further, the effect of wind and topography in the area may also affect the dispersion of geothermal power plant emissions, as has been reported in Ólafsdóttir et al. (2014a, b) for geothermally emitted H_2S gas. According to the wind rose for the Hengill area, the prevailing wind direction is from the east (Fig. 1) and the Nesjavellir power plant is situated in a valley, well sheltered from the easterly wind, and probably the other wind direction as well (south). The Hellisheidi geothermal power plant on the other hand lies exposed at a high elevation. These topographical variations in location, and effects of the prevailing wind, may mean that elements disperse more efficiently around Hellisheidi than at Nesjavellir. This, in combination with the emission rates, may explain the more variable element concentration trends with distance and direction, i.e. element distribution not following the predicted patterns at Hellisheidi as well as at Nesjavellir; although our arguments are limited to wind direction predictions from a single wind rose. Nonetheless, the Nesjavellir findings compare well with other studies, in Italy and Kenya, where several species of trees, shrubs, mosses and lichens have been used as bio-indicators, together with soil, to monitor the impacts of geothermal power plant emissions on terrestrial ecosystems (Baldi, 1988; Bargagli et al., 1997, 2003; Bargagli and Barghigiani, 1991; Loppi and Bonini, 2000; Mutia et al., 2016). Their results showed similar trends as in our study, i.e. with relatively higher sulphur and trace element concentrations in plant tissues (including mosses) and soils closer to the power plants than further away, indicating an input of elements from the nearby geothermal power plants.

Even though there is evidence of sulphur deposition and accumulation in moss and sulphur deposition in soil around the Nesjavellir power plant, suggesting its origin to be the geothermal power plant, the variability in concentrations of the trace elements may imply that the power plants are not a significant contributor in their distribution within the area, due to the effect of other sources of these elements in the Hengill ecosystem. The Hengill geothermal area lies within the active volcanic belt of Iceland (Arnórsson, 1995). Being a volcanic area with active and on-going geothermal activities, there is a high chance that occurrence of these elements into the ecosystem is also derived from these activities, since the elements form a significant proportion of volcanic and geothermal gases. However, it is known that trace elements may be found in power plant emissions at smaller proportions than sulphur (from H_2S gas) (Axtmann, 1975). Accordingly, in this study, trace elements occurred at much lower concentration levels in both soil and moss than sulphur; these low concentrations may have made it more difficult to detect any trends. We therefore cannot rule out the contribution of the power plants to the ecosystem content of these elements. In addition, concentrations of arsenic ranging 0.008 – 0.093 mg/l and boron 1.33 – 6.79 mg/l have been detected in geothermal waters of the Nesjavellir geothermal wells at the Hengill area (Giround, 2008), though it has not been determined as to whether these elements are present in the power plant emissions. Data on the concentrations and emission rates of the other elements, i.e. antimony and mercury, in geothermal fluids and power plant emissions at the Hengill area is still limited. Further studies are thus recommended to assess the trace element levels in the emissions from the power plants.

Other sources of these elements in the geothermal terrestrial ecosystems may include steam from natural surface geothermal manifestations, such as hot springs and fumaroles, and steam sprays from geothermal well tests (on-going at Hengill) that contain these elements and distribute them into the atmosphere. The bedrock (Fig. 1) could also be a source of these variations especially in soils. At the Olkaria geothermal field in Kenya, where a similar study was conducted, findings also showed the same unclear distribution patterns of trace element concentrations in plant tissues and soil against distance and direction gradients and were related to influence from other sources; although the effect of the power plants could not be ruled out as the trace element levels in the Olkaria geothermal power plant emissions are also not determined (Mutia et al., 2016).

Around the power plants, sulphur concentrations were overall higher in soils than in moss tissues. This results contradict the findings of a recent and similar study in Kenya where a vascular plant (a shrub: *Tarchonanthus camphoratus*) was used as a bio-indicator in evaluating the effects of the Olkaria geothermal power plant emissions on terrestrial ecosystems. As opposed to this study, the Olkaria study findings revealed higher sulphur concentrations in the shrub leaves than in soil (Mutia et al., 2016). A possible reason for the higher nutrient enrichment in the shrub leaves than mosses when compared to soils in each study area is probably related to both the active root and leaf uptake of nutrients/elements for the shrub, whereas in mosses, due to their ectohydric nature, nutrient/element uptake is mainly through the atmosphere. This suggests that elements would be more enriched in the shrub leaves than soil, while for moss, the opposite is true. However, at the reference areas for both study areas i.e. Iceland and Kenya, sulphur concentration was on overall higher in soils than plants, which was expected due to the absence of atmospheric pollution. Further, sulphur concentrations in plant tissues and soils were in general at higher levels in Iceland than Kenya. There could be several interacting reasons for this discrepancy between the two ecosystems; The amount of H_2S emissions from the Hengill geothermal power plants is 83% higher than that of the Olkaria power plants, see Table 1 and Mutia et al., (2016). It therefore follows that there would be more sulphur deposition and accumulation in the Hengill (Iceland) terrestrial ecosystems than Olkaria (Kenya). In addition, the different soil proceses in the two ecosystems may provide an explanation for the higher sulphur concentrations in the Hengill soils than Olkaria soils: the shrub leaves are most likely more easily decomposed than the recalcitrant moss shoots. Therefore, decomposition and mineralisation rates are in general much faster in semi-arid Kenya ecosystems than the much colder ecosystems of Iceland. The slow decomposition means that the elements are likely to be much more enriched in the soils of Iceland than in Kenya, suggesting that the impact of geothermal emission (H_2S gas in this case) on ecosystems may be greater in Iceland than Kenya.

The response of the moss growth and characteristics was only partly consistent with our predictions. The apparent indications of decreased biomass growth mats in relation to the Nesjavellir power plant with even lower growth downwind than upwind imply some probable effects to plants. Because sulphur in moss at Nesjavellir was also high close to the power plants, it could be that the mosses are receiving excessive nutrient enrichment which is affecting their biomass growth such that they don’t grow as healthily (low biomass) as the mosses further away (high biomass). The high biomass increase at 4000 m away may partially explain deeper moss mat at 4000 m than 250 m away from the power plant. We could not however explain the trend of the other variables from the Nesjavellir power plant i.e. shoot length increase, shoot turnover and chlorophyll concentrations. Although the high chlorophyll concentrations close to the power plant, corresponding decreased moss growth and high element concentrations were not surprising, because previous studies have reported high chlorophyll levels in plants affected by pollution (Carreras et al., 1996; Saarinen, 1993; Gratani et al., 2000; Bussotti et al., 2003). The causes have been attributed to probably the triggering of metabolic compensation mechanisms as an adaptation by affected plants (Bussotti et al., 2003).

Our results could not support the lack of relationships for moss growth and characteristics with predictors around the Hellisheidi geothermal power plant, especially because the measured element concentrations were elevated in the moss around Hellisheidi and thus we expected a related harm to them. Instead, moss around Hellisheidi showed more growth and higher values to the measured moss characteristics with even higher values downwind than upwind compared to Nesjavellir. Mosses seemed to be growing better (higher shoot length increase, biomass increase and chlorophyll) around Hellisheidi than at Nesjavellir suggesting that the elements dispersing and depositing downwind did not correspond to harmful effects on the moss at Hellisheidi. These elements seem to be within the moss nutrient beneficial limits as there is evidence of greater biomass increase and higher chlorophyll content downwind than upwind, where these elements are expected to deposit. In addition, our study was limited to three distances and two wind directions to calculate trends/ gradients. It is thus important to understand that other differences between sites, other than those related to our predictors, may have potentially influenced the effects of the Hengill geothermal power plant emissions on their terrestrial ecosystems, for instance other environmental factors such as snow cover and differences in precipitation can affect moss growth and physiology.

Our study design did not show any relationship between the frequency of moss shoot damages and the effect of each power plant, distance and direction from the power plants. This noted, element concentration in the damaged moss patches also showed similar trends as the healthy systematically sampled mosses, implying that we cannot rule out the possible role of the power plants in the damages. The comparisons between element concentrations both in soil and moss at the systematically sampled and damaged moss patches in the otherwise healthy moss mats was interesting. The cause of the damaged moss patches is not known, and has been associated with several speculations, including damages related to deposited sulphur emissions from the power plants (Natturufraedistofnun Islands unpublished report, 2008). Sulphur and mercury concentration in moss at the damaged patches generally decreased with distance away, and corresponded to decreasing moss mat depths and chlorophyll levels and higher measured element levels, than in the systematically sampled healthy moss. The extensively damaged moss areas (known to be associated with the geothermal power plants element depositions and possibly thermal pollution) also showed generally similar trends for element concentrations in plants and soils and effects on moss response variables as the healthy moss (systematic sampling). This suggests some relationship of element concentrations (particularly sulphur) in moss and soil, and moss growth and other characteristics from the systematic sampling moss areas with the power plant emissions.

Overall, the variation of element concentrations in moss and soil and moss response variables with our predictors around the Hengill geothermal power plants, provided evidence to infer a sulphur deposition into the ecosystem from the power plants, along with other potential sources, while power plant origin of trace elements was less evident. The evidence was strengthened when comparing our findings to the Raudhalsahraun reference area in Iceland. We found that the concentrations of these elements in moss and soil (except sulphur in soil) around the geothermal area are much higher than at the reference area, providing some indication of high element concentrations in our bio-indicators at the Hengill ecosystem. The general slower moss growth and low moss characteristic values around the geothermal power plant areas, compared to the reference area, also provide a general implication of negative effects on moss health, however with no visible signs of moss damage at our systematic sampling areas.

Further general comparisons of each element concentration in moss between the Hengill geothermal area and the reference area revealed a 30% higher sulphur concentration at the geothermal area. In addition, the moss at the geothermal area accumulated about 18% more sulphur than at the reference area. The concentrations of the other elements were in most cases comparable. On the contrary, soils at the reference area had the highest sulphur concentrations, or 13% more than at the Hengill geothermal area. This could be explained as being related to the different concentrations of sulphur in the two lavas i.e. Raudhalsahraun lava and Hengill lava. Another possibility is that if the Raudhalsahraun area is more exposed to the ocean spray, marine SO_4 may be deposited there in more abundance than at Hellisheidi. Further, the thinner moss mat depths at Raudhalsahraun than the geothermal power plant areas may be associated to the different lava ages. The youngest lava in the Hengill geothermal area is around 2000 years old, found to the East of the Hellisheidi geothermal power plant and around the Nesjavellir power plant (Franzson et al., 2010). However, Raudhalsahraun may be approximately 1000 years younger (Jóhannesson, 1978) than the Hellisheidi and Nesjavellir lava. It is therefore possible that the younger Raudhalsahraun lava has a thinner moss mat formation, compared to Hengill.

An additional reference area, well out of range of geothermal activity was in central Svalbard. The concentration levels of sulphur in *R. lanuginosum* was similar to the reference area at Raudhalsahraun while other element concentrations in moss were comparable with those at the geothermal area. However, all other elements were higher in soils at Svalbard than the study areas in Iceland. This was surprising because the bedrock of central Spitsbergen is predominantly composed of various sedimentary rocks, in contrast to the igneous origin of most of Icelandic bedrock. Previous studies from Svalbard show that vascular plants (Askaer et al., 2008) and mosses (Klos et al., 2015) accumulate high levels of sulphur and arsenic and other metals resulting from coal mining activities. Although our sampling area in Svalbard was at least 17 km away from the closest coal mine, one that stopped operating in 1998, these elements may have dispersed widely and accumulated in the ecosystems.

Since we detect a possible deposition of sulphur, emitted as H_2S gas from the geothermal power plants in the Hengill area and some indications of plant effects, we recommend a follow-up experimental study of, the effect of sulphur enrichment on *R. lanuginosum*, the dominating plant in the moss-rich plant communities of the area, in terms of growth and physiology. We also encourage the start-up of routine monitoring of the concentration of sulphur, arsenic, boron, antimony, mercury, and all other toxic elements of geothermal origin in steam, air, precipitation and ecosystems (using bio-indicators as in this study) as a part of the environmental management plan for the Hengill power plants. Clear information on the origin of these elements can be established using isotopic studies. Furthermore, the efforts undertaken by the developer of the Hengill geothermal power plants to reduce power plant emissions through re-injection of gases, should be highly supported and implemented as abatement strategies to minimize ecosystem effects of the emissions that may emerge later. Overall, this knowledge will aid in the development of appropriate mitigation measures to avoid future irreversible effects that may be costly to ecosystems. These findings also serve as important background in assessment of potential pollution from geothermal power plant emissions. However, we recommend further similar but long term studies (due to the slow growing nature of mosses), with an improved study design to provide a more accurate ecosystem assessment. For example, additional transects parallel to the prevailing wind direction with increased sampling stations and larger sample sizes can be considered to get a clear evaluation of the element distributions and effects within the geothermal area . Other sensitive bio-indicators such as lichens may also be included in the assessments.

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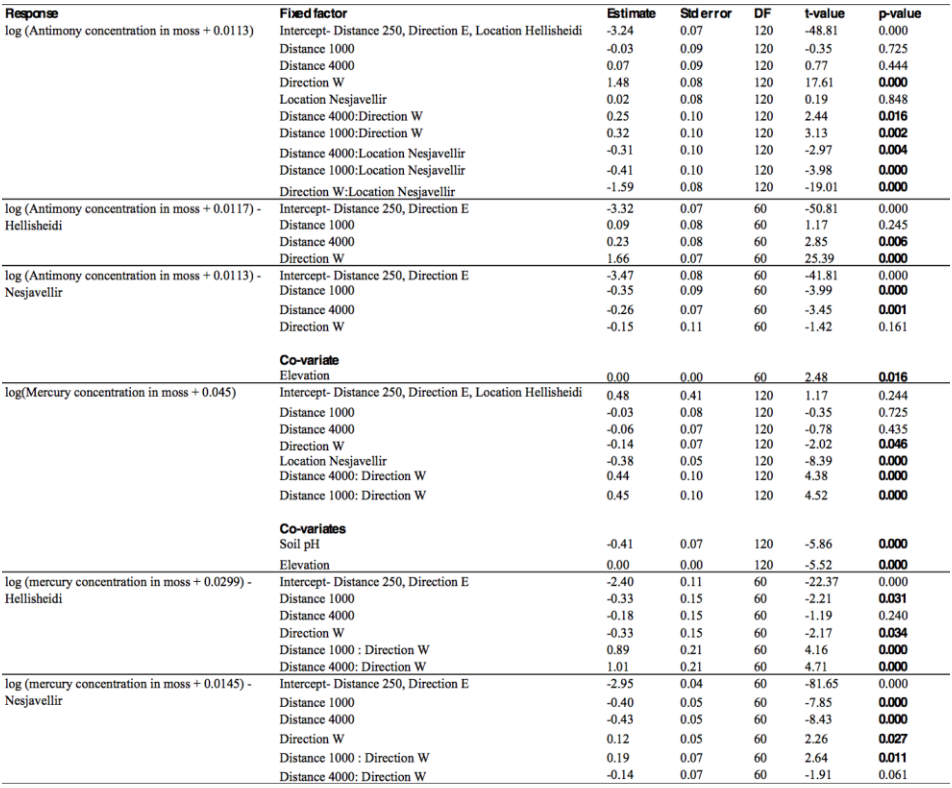
Supplementary Material

Table S1. Linear mixed models for sulphur, arsenic, boron, antimony and mercury concentrations in Racomitrium lanuginosum *shoots at the systematically sampled areas. Estimates indicate the effects of fixed factors and co-variates compared to the intercept and followed by a test statistic. Bold p values indicate significant effects.*

Response	**Fixed factor**	**Estimate**	**Std error**	**DF**	**t-value**	**p-value**
log(sulphur concentration in moss)	Intercept - Distance 250, Direction E, Location Hellisheidi	6.22	0.04	120	140.57	0.000
	Distance 1000	-0.37	0.06	120	-6.16	**0.000**
	Distance 4000	-0.42	0.06	120	-7.13	**0.000**
	Direction W	-0.14	0.06	120	-2.42	**0.017**
	Location Nesjavellir	0.32	0.06	120	5.68	**0.000**
	Distance 1000 : Direction W	0.68	0.07	120	9.96	**0.000**
	Distance 4000 : Direction W	0.81	0.07	120	11.80	**0.000**
	Direction W : Location Nesjavellir	-0.49	0.06	120	-8.74	**0.000**
	Distance 1000 : Location Nesjavellir	-0.36	0.07	120	-5.27	**0.000**
	Distance 4000 : Location Nesjavellir	-0.40	0.07	120	-5.87	**0.000**
log(sulphur concentration in moss) - Hellisheidi	Intercept - Distance 250, Direction E	6.23	0.05	60	121.89	0.000
	Distance 1000	-0.36	0.07	60	-4.96	**0.000**
	Distance 4000	-0.46	0.07	60	-6.40	**0.000**
	Direction W	-0.16	0.07	60	-2.17	**0.034**
	Distance 1000 : Direction W	0.67	0.10	60	6.53	**0.000**
	Distance 4000 : Direction W	0.89	0.10	60	8.68	**0.000**
log(sulphur concentration in moss) - Nesjavellir	Intercept - Distance 250, Direction E	6.53	0.04	60	145.87	0.000
	Distance 1000	-0.73	0.06	60	-11.60	**0.000**
	Distance 4000	-0.79	0.06	60	-12.42	**0.000**
	Direction W	-0.60	0.06	60	-9.53	**0.000**
	Distance 1000 : Direction W	0.70	0.09	60	7.79	**0.000**
	Distance 4000 : Direction W	0.73	0.09	60	8.16	**0.000**
Sulphur accumulation in moss	Intercept - Distance 250, Direction E, Location Hellisheidi	0.06	0.01	11	7.02	0.000
	Distance 1000	-0.02	0.01	10	-2.08	0.063
	Distance 4000	0.00	0.01	11	-0.18	0.861
	Direction W	0.00	0.01	11	0.17	0.870
	Location Nesjavellir	-1.00	0.01	11	-0.52	0.611
log(Arsenic concentration in moss + 0.050)	Intercept - Distance 250, Direction E, Location Hellisheidi	-3.00	0.09	107	-33.21	0.000
	Distance 1000	0.06	0.09	107	0.61	0.547
	Distance 4000	0.14	0.09	107	1.45	0.150
	Direction W	3.34	0.12	107	27.31	**0.000**
	Location Nesjavellir	0.33	0.10	107	3.20	**0.002**
	Direction W : Location Nesjavellir	-3.68	0.16	107	-23.06	**0.000**
log(Arsenic concentration in moss + 0.050) - Hellisheidi	Intercept - Distance 250, Direction E	-3.09	0.09	60	-34.28	0.000
	Distance 1000	0.18	0.11	60	1.63	0.108
	Distance 4000	0.28	0.11	60	2.50	**0.015**
	Direction W	3.55	0.09	60	39.44	**0.000**
log(Arsenic concentration in moss + 0.050) - Nesjavellir	Intercept - Distance 250, Direction E	-2.688	0.118	60	-22.71	0.000
	Distance 1000	0.065	0.145	60	0.45	0.657
	Distance 4000	0.183	0.145	60	1.26	0.212
	Direction W	-0.340	0.118	60	-2.88	**0.006**
log (Boron concentration in moss + 1.38)	Intercept - Distance 250, Direction E, Location Hellisheidi	1.22	0.34	12	3.61	0.004
	Distance 1000	0.23	0.22	12	1.01	0.332
	Distance 4000	-0.17	0.21	12	-0.82	0.431
	Direction W	0.50	0.17	12	2.92	**0.013**
	Location Nesjavellir	-0.59	0.18	12	-3.25	**0.007**
	Co-variate					
	Elevation	0.00	0.00	12	-2.08	**0.059**
log (Boron concentration in moss + 1.38) - Hellisheidi	Intercept - Distance 250, Direction E	-0.68	0.14	60	-4.81	0.000
	Distance 1000	-0.54	0.10	60	-5.22	**0.000**
	Distance 4000	-1.00	0.09	60	-10.76	**0.000**
	Direction W	0.71	0.07	60	10.54	**0.000**
	Co-variate					
	Soil nitrogen	5.74	0.61	60	9.48	**0.000**
Boron concentration in moss - Nesjavellir	**ND**					

Table S1. Continued linear mixed models for antimony and mercury concentrations in Racomitrium lanuginosum *shoots at the systematically sampling areas. Estimates indicate the effects of fixed factors and co-variates compared to the intercept and followed by a test statistic. Bold p values indicate significant effects.*



Response	**Fixed factor**	**Estimate**	**Std error**	**DF**	**t-value**	**p-value**
log (Antimony concentration in moss + 0.0113)	Intercept- Distance 250, Direction E, Location Hellisheidi	-3.24	0.07	120	-48.81	0.000
	Distance 1000	-0.03	0.09	120	-0.35	0.725
	Distance 4000	0.07	0.09	120	0.77	0.444
	Direction W	1.48	0.08	120	17.61	**0.000**
	Location Nesjavellir	0.02	0.08	120	0.19	0.848
	Distance 4000:Direction W	0.25	0.10	120	2.44	**0.016**
	Distance 1000:Direction W	0.32	0.10	120	3.13	**0.002**
	Distance 4000:Location Nesjavellir	-0.31	0.10	120	-2.97	**0.004**
	Distance 1000:Location Nesjavellir	-0.41	0.10	120	-3.98	**0.000**
	Direction W:Location Nesjavellir	-1.59	0.08	120	-19.01	**0.000**
log (Antimony concentration in moss + 0.0117) - Hellisheidi	Intercept- Distance 250, Direction E	-3.32	0.07	60	-50.81	0.000
	Distance 1000	0.09	0.08	60	1.17	0.245
	Distance 4000	0.23	0.08	60	2.85	**0.006**
	Direction W	1.66	0.07	60	25.39	**0.000**
log (Antimony concentration in moss + 0.0113) - Nesjavellir	Intercept- Distance 250, Direction E	-3.47	0.08	60	-41.81	0.000
	Distance 1000	-0.35	0.09	60	-3.99	**0.000**
	Distance 4000	-0.26	0.07	60	-3.45	**0.001**
	Direction W	-0.15	0.11	60	-1.42	0.161
	Co-variate					
	Elevation	0.00	0.00	60	2.48	**0.016**
log(Mercury concentration in moss + 0.045)	Intercept- Distance 250, Direction E, Location Hellisheidi	0.48	0.41	120	1.17	0.244
	Distance 1000	-0.03	0.08	120	-0.35	0.725
	Distance 4000	-0.06	0.07	120	-0.78	0.435
	Direction W	-0.14	0.07	120	-2.02	**0.046**
	Location Nesjavellir	-0.38	0.05	120	-8.39	**0.000**
	Distance 4000: Direction W	0.44	0.10	120	4.38	**0.000**
	Distance 1000: Direction W	0.45	0.10	120	4.52	**0.000**
	Co-variates					
	Soil pH	-0.41	0.07	120	-5.86	**0.000**
	Elevation	0.00	0.00	120	-5.52	**0.000**
log (mercury concentration in moss + 0.0299) - Hellisheidi	Intercept- Distance 250, Direction E	-2.40	0.11	60	-22.37	0.000
	Distance 1000	-0.33	0.15	60	-2.21	**0.031**
	Distance 4000	-0.18	0.15	60	-1.19	0.240
	Direction W	-0.33	0.15	60	-2.17	**0.034**
	Distance 1000 : Direction W	0.89	0.21	60	4.16	**0.000**
	Distance 4000: Direction W	1.01	0.21	60	4.71	**0.000**
log (mercury concentration in moss + 0.0145) - Nesjavellir	Intercept- Distance 250, Direction E	-2.95	0.04	60	-81.65	0.000
	Distance 1000	-0.40	0.05	60	-7.85	**0.000**
	Distance 4000	-0.43	0.05	60	-8.43	**0.000**
	Direction W	0.12	0.05	60	2.26	**0.027**
	Distance 1000 : Direction W	0.19	0.07	60	2.64	**0.011**
	Distance 4000: Direction W	-0.14	0.07	60	-1.91	0.061

Table S2. Linear models for sulphur, arsenic, boron, antimony and mercury concentrations in soils at the systematic sampling areas. Estimates indicate the effects of the predictors and co-variates compared to the intercept and followed by a test statistic. Bold p values indicate significant effects.

Response	**Predictors**	**Estimate**	**Std error**	**t-value**	**p-value**
Sulphur concentration in soil	Intercept - Distance 250, Direction E, Location Hellisheidi	339.43	51.46	6.60	**0.001**
	Distance 1000	-131.77	38.77	-3.40	**0.015**
	Distance 4000	-134.21	39.90	-3.36	**0.015**
	Direction W	-52.60	34.69	-1.52	0.180
	Location Nesjavellir	254.47	30.45	8.36	**0.000**
	Co-variate				
Residual standard error: 51.50, **DF** 6	Soil carbon	40.88	7.39	5.53	**0.001**
Sulphur concentration in soil - Hellisheidi	Intercept - Distance 250, Direction E	350.76	74.17	4.73	0.133
	Distance 1000	-67.00	93.93	-0.71	0.606
	Distance 4000	-89.78	94.96	-0.95	0.518
	Direction W	-43.92	61.45	-0.72	0.605
	Co-variate				
Residual standard error: 68.20, **DF** 1	Soil carbon	34.25	13.44	2.55	0.238
Sulphur concentration in soil - Nesjavellir	Intercept - Distance 250, Direction E	855.78	46.29	18.49	**0.003**
	Distance 1000	-214.17	56.69	-3.78	0.063
	Distance 4000	-166.83	56.69	-2.94	0.099
Residual standard error: 56.69, **DF** 2	Direction W	70.11	46.29	1.52	0.269
Arsenic concentration in soil	Intercept - Distance 250, Direction E, Location Hellisheidi	0.69	0.70	0.98	0.366
	Distance 1000	0.00	0.39	-0.01	0.995
	Distance 4000	-0.09	0.39	-0.23	0.823
	Direction W	0.06	0.36	0.16	0.881
	Location Nesjavellir	-0.91	0.35	-2.61	**0.040**
	Co-variate				
Residual standard error: 0.53, **DF** 6	Soil nitrogen	4.78	2.80	1.70	0.139
Arsenic concentration in soil -Hellisheidi	Intercept - Distance 250, Direction E	1.29	0.39	3.31	0.080
	Distance 1000	0.92	0.48	1.91	0.196
	Distance 4000	0.86	0.48	1.81	0.213
Residual standard error: 0.48, **DF** 2	Direction W	0.26	0.39	0.66	0.577
Arsenic concentration in soil - Nesjavellir	Intercept - Distance 250, Direction E	1.54	0.41	3.74	0.065
	Distance 1000	-0.57	0.51	-1.12	0.380
	Distance 4000	-0.69	0.51	-1.36	0.307
Residual standard error: 0.51, **DF** 2	Direction W	0.51	0.41	1.23	0.344
Boron concentation in soil	Intercept - Distance 250, Direction E, Location Hellisheidi	0.26	7.17	0.04	0.975
	Distance 1000	-0.24	0.92	-0.27	0.816
	Distance 4000	-0.09	1.12	-0.08	0.942
	Direction W	-0.31	0.98	-0.32	0.780
Residual standard error: 1.03, **DF 2**	Location Nesjavellir	-0.62	0.82	-0.76	0.525
Antimony concentration in soil	Intercept - Distance 250, Direction E, Location Hellisheidi	0.10	0.04	2.46	**0.049**
	Distance 1000	0.00	0.03	-0.15	0.888
	Distance 4000	-0.02	0.03	-0.70	0.511
	Direction W	0.02	0.03	0.70	0.513
	Location Nesjavellir	-0.05	0.02	-2.28	0.063
	Co-variate				
Residual standard error: 0.04, **DF** 6	Soil carbon	0.02	0.01	2.68	**0.037**
Antimony concentration in soil - Hellisheidi	Intercept - Distance 250, Direction E	0.14	0.04	3.60	0.069
	Distance 1000	0.09	0.05	1.87	0.202
	Distance 4000	0.09	0.05	1.94	0.192
Residual standard error: 0.04, **DF** 2	Direction W	0.04	0.04	0.97	0.434
Antimony concentration in soil - Nesjavellir	Intercept - Distance 250, Direction E	0.15	0.02	9.32	**0.011**
	Distance 1000	-0.04	0.02	-2.15	0.165
	Distance 4000	-0.07	0.02	-3.40	0.077
Residual standard error: 0.02, **DF** 2	Direction W	0.07	0.02	4.52	**0.046**
Mercury concentration in soil	Intercept - Distance 250, Direction E, Location Hellisheidi	0.00	0.02	-0.01	0.996
	Distance 1000	0.00	0.01	-0.07	0.949
	Distance 4000	-0.01	0.01	-1.11	0.310
	Direction W	0.00	0.01	-0.17	0.874
	Location Nesjavellir	-0.02	0.01	-2.51	**0.046**
	Co-variate				
Residual standard error: 0.02, **DF 6**	Soil carbon	0.01	0.00	5.27	**0.002**
Mercury concentration in soil - Hellisheidi	Intercept - Distance 250, Direction E	0.04	0.03	1.33	0.315
	Distance 1000	0.06	0.04	1.52	0.268
	Distance 4000	0.06	0.04	1.55	0.262
Residual standard error: 0.04, **DF 2**	Direction W	0.02	0.03	0.52	0.655
Mercury concentration in soil - Nesjavellir	Intercept - Distance 250, Direction E	0.05	0.01	4.33	**0.049**
	Distance 1000	-0.02	0.02	-1.13	0.377
	Distance 4000	-0.03	0.02	-2.28	0.151
Residual standard error: 0.02, **DF 2**	Direction W	0.04	0.01	3.21	0.085

Table S3. Soil characteristics for the systematically sampled areas around the Hengill geothermal area in Iceland, n= 1 per station. The mean elevation data for the sampling areas is also shown. Soil data from sampling points per station is pooled to represent a station. See Figure 1. for specific transect names in the given directions below.

Soil characteristics and elevation	Direction	Distance	Nesjavelllir	Hellisheidi
Carbon (%)	W	250	9.11	6.37
		1000	7.76	12.44
		4000	8.39	7.66
	E	250	7.27	2.86
		1000	4.90	6.41
		4000	5.68	11.40
Nitrogen (%)	W	250	0.39	0.25
		1000	0.36	0.39
		4000	0.43	0.23
	E	250	0.31	0.19
		1000	0.27	0.26
		4000	0.26	0.36
Soil moisture (%)	W	250	100.45	113.88
		1000	66.82	153.94
		4000	91.12	193.49
	E	250	97.93	48.50
		1000	77.78	142.26
		4000	83.37	128.14
Soil pH	W	250	5.13	5.78
		1000	5.65	5.61
		4000	6.05	5.28
	E	250	5.36	5.76
		1000	6.30	5.56
		4000	6.24	5.45
Elevation (m. a.s.l)	W	250	196.58	250.53
		1000	403.3	265.45
		4000	339.95	283.71
	E	250	174.29	285.09
		1000	159.86	388.74
		4000	138.38	351.45

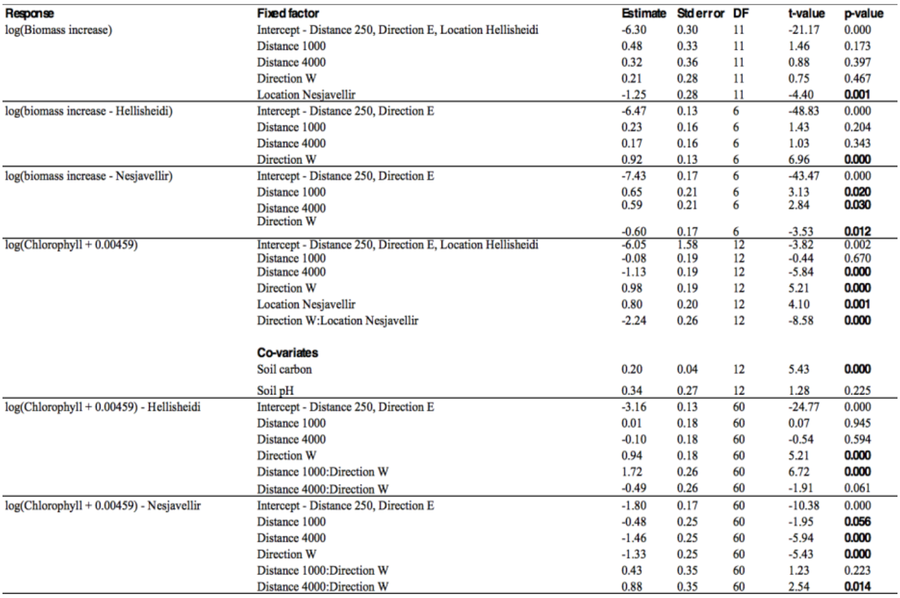
Table S4. Linear models for co-variates (soil characteristics and elevation) at the systematic sampling areas. Estimates indicate the effects of the predictors and co-variates compared to the intercept and followed by a test statistic. Bold p values indicate significant effects.

Response	**Predictors**	**Estimate**	**Std error**	**t-value**	**p-value**
Soil carbon	Intercept - Distance 250, Direction E, Location Hellisheidi	5.31	1.70	3.13	0.017
	Distance 1000	1.80	1.86	0.97	0.366
	Distance 4000	2.20	1.86	1.18	0.275
	Direction W	2.42	1.52	1.59	0.156
Residual SE: 2.63,**DF** 7	Location Nesjavellir	-0.89	1.52	-0.58	0.578
Soil nitrogen	Intercept - Distance 250, Direction E, Location Hellisheidi	0.22	0.05	4.71	0.002
	Distance 1000	0.04	0.05	0.73	0.490
	Distance 4000	0.04	0.05	0.74	0.484
	Direction W	0.07	0.04	1.64	0.145
Residual SE: 0.07,**DF** 7	Location Nesjavellir	0.06	0.04	1.36	0.215
Soil moisture	Intercept - Distance 250, Direction E, Location Hellisheidi	86.24	30.31	2.85	0.025
	Distance 1000	20.15	33.20	0.61	0.563
	Distance 4000	39.22	33.20	1.18	0.276
	Direction W	31.83	27.11	1.17	0.279
Residual SE: 46.95,**DF** 7	Location Nesjavellir	-47.17	27.11	-1.74	0.125
Soil pH	Intercept - Distance 250, Direction E, Location Hellisheidi	5.50	0.25	21.95	0.000
	Distance 1000	0.28	0.27	1.02	0.343
	Distance 4000	0.25	0.27	0.90	0.399
	Direction W	-0.20	0.22	-0.88	0.411
Residual SE: 46.95,**DF** 7	Location Nesjavellir	0.21	0.22	0.93	0.381
Elevation	Intercept - Distance 250, Direction E, Location Hellisheidi	240.86	56.61	4.25	0.004
	Distance 1000	77.72	62.02	1.25	0.250
	Distance 4000	60.79	62.02	0.98	0.360
	Direction W	46.30	50.64	0.91	0.391
Residual SE: 87.71,**DF** 7	Location Nesjavellir	-74.79	50.64	-1.48	0.183

Table S5. Linear mixed models for the moss traits at the systematically sampled areas. Estimate indicates effects of the fixed factors and co-variates compared to the intercept and followed by a test statistic. Bold p values indicate significant effects.

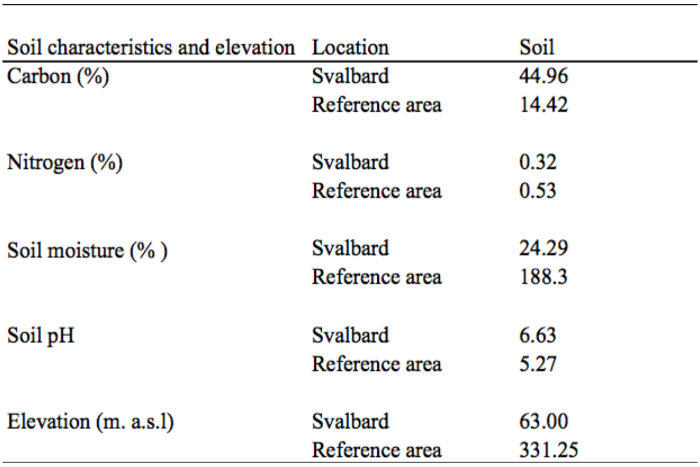
Response	**Fixed factor**	**Estimate**	**Std error**	**DF**	**t-value**	**p-value**
Moss mat depth	Intercept - Distance 250, Direction E, Location Hellisheidi	35.55	1.88	120	18.95	0.000
	Distance 1000	1.59	1.37	120	1.16	0.248
	Distance 4000	6.89	1.40	120	4.90	**0.000**
	Direction W	-0.61	1.22	120	-0.50	0.615
	Location Nesjavellir	-2.69	1.09	120	-2.46	**0.015**
	Covariate					
	Soil carbon	-1.47	0.26	120	-5.77	**0.000**
Moss mat depth- Hellisheidi	Intercept - Distance 250, Direction E	30.93	2.00	6	15.47	0.000
	Distance 1000	-6.69	2.45	6	-2.73	**0.034**
	Distance 4000	-1.43	2.45	6	-0.59	0.579
	Direction W	-3.45	2.00	6	-1.73	0.135
Moss mat depth - Nesjavellir	Intercept - Distance 250, Direction E	21.87	1.57	60	13.92	0.000
	Distance 1000	5.52	1.92	60	2.87	**0.006**
	Distance 4000	9.66	1.92	60	5.02	**0.000**
	Direction W	-4.26	1.57	60	-2.71	**0.009**
log(Shoot length increase + 0.1)	Intercept - Distance 250, Direction E, Location Hellisheidi	-0.46	0.11	9.822	-4.22	0.002
	Distance 1000	0.12	0.12	10.48	0.99	0.346
	Distance 4000	-0.20	0.12	10.48	-1.64	0.132
	Direction W	-0.13	0.10	10.226	-1.27	0.233
	Location Nesjavellir	-0.23	0.10	10.226	-2.37	**0.039**
log(Shoot length increase - Hellisheidi) + 0.1	Intercept - Distance 250, Direction E	-0.56	0.08	18	-7.09	0.000
	Distance 1000	0.02	0.10	18	0.20	0.843
	Distance 4000	-0.16	0.10	18	-1.69	0.108
	Direction W	0.12	0.08	18	1.56	0.136
log(Shoot length increase - Nesjavellir) + 0.1	Intercept - Distance 250, Direction E	-0.61	0.15	17	-4.05	0.001
	Distance 1000	0.25	0.19	17	1.32	0.204
	Distance 4000	-0.21	0.19	17	-1.10	0.285
	Direction W	-0.39	0.15	17	-2.60	**0.019**
Shoot turnover	Intercept - Distance 250, Direction E, Location Hellisheidi	1.25	0.20	29	6.21	0.000
	Distance 1000	-0.15	0.15	29	-0.99	0.329
	Distance 4000	-0.61	0.17	29	-3.61	**0.001**
	Direction W	0.08	0.14	29	0.56	0.582
	Location Nesjavellir	-0.33	0.13	29	-2.62	**0.014**
	Co-variate					
	Soil carbon	0.13	0.03	29	4.64	**0.000**
Shoot turnover - Hellisheidi	Intercept - Distance 250, Direction E	1.53	0.14	11	10.60	0.000
	Distance 1000	0.49	0.20	11	2.41	**0.035**
	Distance 4000	0.28	0.23	11	1.23	0.244
	Direction W	0.74	0.20	11	3.60	**0.004**
Shoot turnover - Nesjavellir	Intercept - Distance 250, Direction E	1.86	0.13	15	13.92	0.000
	Distance 1000	-0.41	0.18	15	-2.23	**0.041**
	Distance 4000	-0.81	0.16	15	-4.94	**0.000**
	Direction W	0.43	0.14	15	2.93	**0.010**

Table S5. Continued linear mixed models for the moss traits at the systematically sampled areas. Estimate indicates effects of the fixed factors and co-variates compared to the intercept and followed by a test statistic. Bold p values indicate significant effects.



Response	**Fixed factor**	**Estimate**	**Std error**	**DF**	**t-value**	**p-value**
log(Biomass increase)	Intercept - Distance 250, Direction E, Location Hellisheidi	-6.30	0.30	11	-21.17	0.000
	Distance 1000	0.48	0.33	11	1.46	0.173
	Distance 4000	0.32	0.36	11	0.88	0.397
	Direction W	0.21	0.28	11	0.75	0.467
	Location Nesjavellir	-1.25	0.28	11	-4.40	**0.001**
log(biomass increase - Hellisheidi)	Intercept - Distance 250, Direction E	-6.47	0.13	6	-48.83	0.000
	Distance 1000	0.23	0.16	6	1.43	0.204
	Distance 4000	0.17	0.16	6	1.03	0.343
	Direction W	0.92	0.13	6	6.96	**0.000**
log(biomass increase - Nesjavellir)	Intercept - Distance 250, Direction E	-7.43	0.17	6	-43.47	0.000
	Distance 1000	0.65	0.21	6	3.13	**0.020**
	Distance 4000	0.59	0.21	6	2.84	**0.030**
	Direction W	-0.60	0.17	6	-3.53	**0.012**
log(Chlorophyll + 0.00459)	Intercept - Distance 250, Direction E, Location Hellisheidi	-6.05	1.58	12	-3.82	0.002
	Distance 1000	-0.08	0.19	12	-0.44	0.670
	Distance 4000	-1.13	0.19	12	-5.84	**0.000**
	Direction W	0.98	0.19	12	5.21	**0.000**
	Location Nesjavellir	0.80	0.20	12	4.10	**0.001**
	Direction W:Location Nesjavellir	-2.24	0.26	12	-8.58	**0.000**
	Co-variates					
	Soil carbon	0.20	0.04	12	5.43	**0.000**
	Soil pH	0.34	0.27	12	1.28	0.225
log(Chlorophyll + 0.00459) - Hellisheidi	Intercept - Distance 250, Direction E	-3.16	0.13	60	-24.77	0.000
	Distance 1000	0.01	0.18	60	0.07	0.945
	Distance 4000	-0.10	0.18	60	-0.54	0.594
	Direction W	0.94	0.18	60	5.21	**0.000**
	Distance 1000:Direction W	1.72	0.26	60	6.72	**0.000**
	Distance 4000:Direction W	-0.49	0.26	60	-1.91	0.061
log(Chlorophyll + 0.00459) - Nesjavellir	Intercept - Distance 250, Direction E	-1.80	0.17	60	-10.38	0.000
	Distance 1000	-0.48	0.25	60	-1.95	**0.056**
	Distance 4000	-1.46	0.25	60	-5.94	**0.000**
	Direction W	-1.33	0.25	60	-5.43	**0.000**
	Distance 1000:Direction W	0.43	0.35	60	1.23	0.223
	Distance 4000:Direction W	0.88	0.35	60	2.54	**0.014**

Table S6 Soil characteristics and elevation measures (co-variates) for Skansenbukta in Svalbard (n = 1) and Raudhalsahraun Reference area (n = 1) in Iceland. Data represents means of pooled samples from sampling points per station.



Soil characteristics and elevation	Location	Soil
Carbon (%)	Svalbard	44.96
	Reference area	14.42
Nitrogen (%)	Svalbard	0.32
	Reference area	0.53
Soil moisture (%)	Svalbard	24.29
	Reference area	188.3
Soil pH	Svalbard	6.63
	Reference area	5.27
Elevation (m. a.s.l)	Svalbard	63.00
	Reference area	331.25

Table S7. Correlation coefficients (r) for element concentrations in moss, soil and moss growth traits for the systematic sampled areas at Hengill geothermal area in Iceland (n = 6 per paired data per location). Significant correlations in bold (P <0.001).

	Soil sulphur	Soil arsenic	Soil boron	Soil mercury	Soil antimony	Moss mat depth	Shoot elongation	Biomass increase	Shoot turnover	Chlorophyll	Moss arsenic	Moss boron	Moss mercury	Moss sulphur	Moss antimony
Soil sulphur	1.0000														
Soil arsenic	0.1126	1.0000													
Soil boron	-0.4595	0.3762	1.0000												
Soil mercury	0.0588	**0.7577**	-0.0059	1.0000											
Soil antimony	-0.1094	**0.7567**	0.0112	0.9051	1.0000										
Moss mat depth	-0.3928	-0.3419	0.1879	-0.3769	-0.3232	1.0000									
Shoot elongation	0.0650	0.4978	0.5416	0.1774	0.0091	-0.1837	1.0000								
Biomass increase	-0.4763	0.3658	0.2114	0.5256	0.5772	-0.1266	0.0776	1.0000							
Shoot turnover	0.2746	0.5370	0.1645	0.3778	0.2672	-0.7373	**0.6683**	0.1807	1.0000						
Chloroplyll	0.0331	0.4580	0.0625	0.4805	0.4270	-0.3356	0.2797	0.5545	0.4463	1.0000					
Moss arsenic	-0.3553	0.1640	-0.0422	0.4223	0.5227	-0.0294	-0.1746	**0.7682**	-0.0147	0.4040	1.0000				
Moss boron	-0.1090	0.3613	0.2346	0.3340	0.2944	-0.1319	0.3675	0.5458	0.3019	0.4762	0.5071	1.0000			
Moss mercury	-0.3318	0.1721	-0.0961	0.4143	0.5425	-0.0190	-0.2695	0.4627	-0.1062	0.1614	**0.7543**	0.1670	1.0000		
Moss sulphur	-0.0775	-0.0599	-0.3318	0.2545	0.3019	-0.1889	-0.2426	0.3427	0.0176	0.3324	0.4620	0.1838	0.4989	1.0000	
Moss antimony	-0.3650	0.1794	-0.0344	0.4559	0.5523	-0.0614	-0.1651	**0.7999**	-0.0065	0.4023	**0.9850**	0.4874	**0.7526**	0.4863	1.0000

Table S8. Correlation coefficients (r) for element concentrations in moss, soil and moss growth traits for the damaged moss patch areas at Hengill geothermal area in Iceland (n = 6 per paired data per location). Significant r values in bold (P<0.001). Moss boron correlations not shown as concentrations were always below detection.

	Soil arsenic	Soil boron	Soil mercury	Soil sulphur	Soil antimony	Moss mat depth	Moss arsenic	Moss boron	Moss mercury	Moss sulphur	Moss antimony	Chlorophyll
Soil arsenic	1.0000											
Soil boron	0.5909	1.0000										
Soil mercury	0.3683	-0.0952	1.0000									
Soil sulphur	-0.0512	-0.3247	0.2624	1.0000								
Soil antimony	**0.7353**	0.1929	**0.8189**	-0.0683	1.0000							
Moss mat depth	-0.5255	-0.2144	-0.5654	-0.0858	-0.6420	1.0000						
Moss arsenic	0.3793	0.1002	0.4448	-0.1648	0.4612	-0.3800	1.0000					
Moss boron	0.2246	0.0350	0.3225	-0.1595	0.2993	-0.2993	0.7427	1.0000				
Moss mercury	0.5516	**0.5924**	0.4760	-0.0954	0.5756	-0.5274	0.3332	0.1130	1.0000			
Moss sulphur	0.0032	-0.0610	0.0819	0.5587	-0.0571	0.0463	-0.0002	-0.0794	0.0277	1.0000		
Moss antimony	0.3930	0.1282	0.4866	-0.1609	0.5062	-0.4057	**0.9691**	**0.6917**	0.3728	0.0394	1.0000	
Chlorophyll	0.1508	0.0082	0.2205	0.1058	0.1843	-0.1859	0.4451	0.3821	0.0812	0.0889	0.5002	1.0000

Table S9. Correlation matrix for element concentrations in moss, soil and moss growth traits for the extensively damaged sampling areas at Hengill geothermal area in Iceland (n = 20 per location). Significant correlations in bold (P<0.001).

	Soil arsenic	Soil boron	Soil mercury	Soil sulphur	Soil antimony	Moss mat depth	Moss arsenic	Moss mercury	Moss sulphur	Moss antimony	Chlorophyll
Soil arsenic	1.0000										
Soil boron	**1.0000**	1.0000									
Soil mercury	**1.0000**	**1.0000**	1.0000								
Soil sulphur	**-1.0000**	**-1.0000**	**-1.0000**	1.0000							
Soil antimony	**1.0000**	**1.0000**	**1.0000**	**-1.0000**	1.0000						
Moss mat depth	**-0.6140**	**-0.6140**	**-0.6140**	**0.6140**	**-0.6140**	1.0000					
Moss arsenic	**-0.7057**	**-0.7057**	**-0.7057**	**0.7057**	**-0.7057**	0.4009	1.0000				
Moss mercury	0.5665	0.5665	0.5665	-0.5665	0.5665	-0.3788	-0.4266	1.0000			
Moss sulphur	**0.7698**	**0.7698**	**0.7698**	**-0.7698**	**0.7698**	-0.4577	**-0.6785**	0.5841	1.0000		
Moss antimony	-0.4810	-0.4810	-0.4810	0.4810	-0.4810	0.2501	0.1572	**-0.7036**	**-0.6419**	1.0000	
Chlorophyll	-0.5770	-0.5770	-0.5770	0.5770	-0.5770	0.4581	0.4089	-0.2150	-0.3022	0.2155	1.0000

Appendix S1. Detailed results from additional sampling sites

Damaged moss patch sampling areas

Element concentrations in *R. lanuginosum* shoots

At different distances from the power plants, the NMDS plot showed clustering of damaged patch moss samples together, indicating similarity; however, samples from the systematic and extensively damaged sampling areas were separate (dissimilar), Fig. 2a. The element concentrations in damaged patches targeted sampling mosses were overall higher at Hellisheidi than Nesjavellir, except for sulphur which showed similar concentrations at both locations (Table A1). Around Hellisheidi, sulphur and boron concentrations in damaged moss showed a decrease with distance away, opposite to the trend in healthy moss (systematic sampling), while mercury increased (Table A1a). Higher element concentrations were also present in damaged mosses to the west of Hellisheidi power plant than to the east, except for sulphur and mercury that showed the opposite pattern. The damaged moss element concentrations (sulphur, antimony and mercury) around the Nesjavellir geothermal power plant generally decreased with increasing distance away, but were higher (sulphur and arsenic) to the east than west of the power plant (Table A1a).

Element concentrations in soil

The soil element concentrations were similar beneath damaged (Table A1) and healthy moss (Tables 2, Fig. 4). The NMDS plot (Fig. 2e) did not reveal any striking dissimilarity but a higher scatter among the soil samples under the damaged moss patches. There was no difference in element concentrations between the damaged patch targeted sampling areas for sulphur, boron and mercury, and just as beneath healthy moss there were higher concentrations of arsenic and antimony in soils at Hellisheidi than Nesjavellir (Table A1b).

Moss mat depth and chlorophyll concentrations

The moss variables did not reveal any marked difference in moss mat depth between the healthy (Table 3) and damaged moss patches (Table A2), but chlorophyll concentrations were much lower in the damaged moss as expected. For the damaged moss patches however, moss mat was deeper at Nesjavellir than Hellisheidi (opposite to moss depths at the systematic sampled stations) (Tables A2, A2a), while chlorophyll concentrations did not differ between locations. At both power plants, moss mat depth and chlorophyll generally decreased with distance away, although at Nesjavellir the effect of distance was not significant on moss mat depth. Direction affected both moss variables and higher moss mat depths and higher chlorophyll concentrations were found east (upwind) of the power plants than west (Table A2a).

The covariates (soil characteristics and elevation) did not vary between the damaged patch targeted sampling areas (Tables A3, A3a); they were important in improving most of the models by accounting parts of unexplained variations in the models (Tables A1a and A2a).

Extensively damaged sampling areas and Svalbard

Element concentrations in *R. lanuginosum* shoots

Moss element concentrations at the Hellisheidi systematic sampling area were similar as those of the extensively damaged sampling moss areas at both locations except for sulphur. Sulphur was overall much higher at both locations of the extensively damaged sampling moss areas than the systematic sampling areas (Table A4, Fig. 3). Mosses at the Nesjavellir systematic sampling areas, on the other hand, showed the lowest element concentrations (Tables 2, 4). The concentrations of boron were below detection for all the sampled areas except for the systematically sampled areas close to the Hellisheidi power plants. The NMDS ordination did not reveal striking dissimilarity patterns in moss element concentration samples at the systematic sampling and extensively damaged sampling moss areas (Fig. 2).

In comparison to the damaged patch targeted sampling areas at 250 m away from the power plant for Nesjavellir, all element concentrations in the extensively damaged sampling moss areas at Nesjavellir (267 m away) were generally high (Tables A1, A4). Similarly, at Hellisheidi, moss at the extensively damaged sampling areas (830 m away) showed high concentrations for only sulphur and antimony relative to mosses at the damaged patch targeted sampling area at 1000 m away; the other element concentrations were much lower than those at the damaged patch targeted sampling areas (Tables A1, A4).

For Svalbard mosses, their element concentration values were clearly dissimilar (NMDS ordination) to all sampled mosses in Iceland (Fig. 2a – c). Mosses from Svalbard showed 20% lower sulphur concentrations than those at the systematic sampling areas (Table 4 and Fig. 3). For the same comparisons, the other element concentrations for Svalbard mosses were similar to those of mosses at Hellisheidi systematic sampling but higher than those for mosses at Nesjavellir systematic sampling.

Element concentrations in soil

All element concentrations in soil at the systematic sampling areas were comparable to those at the extensively damaged sampling areas except for sulphur and boron (Tables 2, A4). Sulphur concentrations in soil were much higher at the Hellisheidi systematic sampling areas than the extensively damaged sampling moss areas, while concentrations at Nesjavallir systematic sampling areas were similar to those at the extensively damaged moss area. Boron concentrations were, on the other hand, much higher in soils at Nesjavellir systematic sampling areas than the extensively damaged sampling moss area; but concentrations at Hellisheidi were similar to those for soils at the extensively damaged sampling moss area (Tables 2, A4). There were no obvious dissimilarity patterns revealed by the NMDS ordination (Fig. 2d - f) between the systematic sampling and the extensively damaged sampling moss areas.

In comparison to Svalbard soils, element concentrations were much lower at the systematically sampled areas than in Svalbard (Tables 2, 4, Fig. 4), although this difference was not apparent in the NMDS ordination (Fig. 2d - f).

Moss depth and chlorophyll concentrations

As expected, moss mat depth and chlorophyll concentrations showed generally lower values at the extensively damaged sampling moss areas (Table A5) than the systematically sampled areas around the power plants (Table 5).

Table A1. Concentrations of sulphur, arsenic, boron, antimony and mercury in Racomitrium lanuginosum *shoots and soil (mg/kg dry weight), collected from the damaged moss patch sampling areas along transects at two power plants at Helisheidi and Nesjavellir, in the Hengill geothermal area, Iceland (n = 10 per location for moss and n = 1 per location for soil). Some boron concentrations were below the 1 mg/kg detection limit in shoots and soil. Elements concentrations for* R. lanuginosum *are given as mean ± SE and only as mean for soils due to sample pooling per station (see methods).*

			Nesjavellir		Hellisheidi	
Element	Direction	Distance (m)	Shoot	Soil	Shoot	Soil
Sulphur	W	250	853.20 ± 145.49	868.33	638.20 ± 14.89	568.00
		1000	701.20 ± 156.96	835.67	636.40 ± 4.15	636.33
		4000	415.00 ± 32.65	735.33	587.40 ± 85.02	663.67
	E	250	1424.00 ± 154.26	777.67	1116.00 ± 139.84	330.00
		1000	412.80 ± 17.43	680.33	506.00 ± 35.97	618.67
		4000	421.80 ± 13.71	431.33	469.40 ± 33.47	635.67
Arsenic	W	250	0.29 ± 0.08	2.72	2.10 ± 0.21	1.87
		1000	0.11 ± 0.05	1.12	2.50 ± 0.16	2.24
		4000	0.05 ± 0.03	1.42	0.85 ± 0.26	4.08
	E	250	0.20 ± 0.01	1.31	0.16 ± 0.06	1.04
		1000	0.02 ± 0.02	0.68	0.22 ± 0.15	1.62
		4000	0.13 ± 0.03	0.59	0.42 ± 0.05	2.82
Boron	W	250	<1	3.74	4.00 ± 0.52	2.35
		1000	<1	3.4	2.60 ± 0.98	4.05
		4000	<1	<1	1.26 ± 1.26	<1
	E	250	<1	<1	<1	4.59
		1000	<1	2.31	0.25 ± 0.25	2.95
		4000	<1	<1	0.27 ± 0.27	9.02
Antimony	W	250	0.05 ± 0.01	0.28	0.22 ± 0.02	0.19
		1000	0.03 ± 0.01	0.14	0.04 ± 0.01	0.32
		4000	0.03 ± 0.01	0.21	0.07 ± 0.01	0.36
	E	250	0.05 ± 0.01	0.17	0.06 ± 0.01	0.12
		1000	0.02 ± 0.00	0.10	0.04 ± 0.01	0.19
		4000	0.02 ± 0.00	0.06	0.07 ± 0.01	0.28
Mercury	W	250	0.10 ± 0.01	0.05	0.11 ± 0.01	0.08
		1000	0.02 ± 0.00	0.07	0.15 ± 0.00	0.09
		4000	0.02 ± 0.00	0.07	0.18 ± 0.05	0.15
	E	250	0.08 ± 0.02	0.14	0.12 ± 0.03	<0.04
		1000	0.02 ± 0.00	<0.04	0.07 ± 0.02	0.08
		4000	0.02 ± 0.00	<0.04	0.25 ± 0.04	0.10

Table A1a. Linear mixed models for sulphur, arsenic, boron, antimony and mercury concentrations in Racomitrium lanuginosum *shoots at the damaged moss patch sampling areas. Estimates indicate the effects of fixed factors and co-variates compared to the intercept and followed by a test statistic. Bold p values indicate significant effects. NA – Not Available.*

Response	**Predictors**	**Estimate**	**Std error**	**DF**	**t-value**	**p-value**
log (Sulfur concentration in moss)	Intercept- Distance 250, Direction E, Location Hellisheidi	6.92	0.10	60	72.57	**0.000**
	Distance 1000	-1.05	0.10	60	-10.53	**0.000**
	Distance 4000	-1.10	0.10	60	-10.89	**0.000**
	Direction W	-0.65	0.10	60	-6.25	**0.000**
	Location Nesjavellir	0.09	0.07	60	1.39	0.169
	Distance 1000: Direction W	0.84	0.14	60	5.98	**0.000**
	Distance 4000: Direction W	0.44	0.15	60	2.99	**0.004**
	Co-variate					
	Soil carbon	0.04	0.01	60	3.48	**0.001**
log (Arsenic concentration in moss + 0.245)	Intercept- Distance 250, Direction E, Location Hellisheidi	-0.87	0.13	60	-6.64	**0.000**
	Distance 1000	0.04	0.18	60	0.20	0.840
	Distance 4000	0.28	0.18	60	1.58	0.120
	Location Nesjavellir	-0.02	0.17	60	-0.14	0.887
	Direction W	1.62	0.17	60	9.80	**0.000**
	Distance 4000:Location Nesjavellir	-0.56	0.20	60	-2.79	**0.007**
	Distance 1000:Location Nesjavellir	-0.22	0.20	60	-1.10	0.278
	Distance 1000:Direction W	0.13	0.20	60	0.64	0.527
	Distance 4000:Direction W	-0.90	0.20	60	-4.46	**0.000**
	Location Nesjavellir:Direction W	-1.30	0.17	60	-7.90	**0.000**
log (Arsenic concentration in moss + 0.245) -	Intercept- Distance 250, Direction E	-1.41	0.11	60	-12.80	**0.000**
	Distance 1000	0.00	0.14	60	0.00	1.000
	Distance 4000	0.05	0.13	60	0.39	0.695
	Direction W	2.26	0.13	60	17.84	**0.000**
	Distance 1000: Direction W	-0.18	0.17	60	-1.04	0.301
	Distance 4000: Direction W	-0.51	0.18	60	-2.82	**0.006**
log (Arsenic concentration in moss + 0.108) -	Intercept- Distance 250, Direction E	-2.01	0.07	60	-29.84	**0.000**
	Distance 1000	0.04	0.09	60	0.41	0.682
	Distance 4000	0.11	0.10	60	1.07	0.288
	Direction W	-0.25	0.08	60	-3.02	**0.004**
log (Boron concentration in moss + 1.27)	Intercept- Distance 250, Direction E, Location Hellisheidi	-0.37	0.39	60	-0.96	0.342
	Distance 1000	-0.03	0.17	60	-0.18	0.861
	Distance 4000	-0.14	0.17	60	-0.81	0.420
	Direction W	1.35	0.21	60	6.31	**0.000**
	Location Nesjavellir	0.15	0.26	60	0.58	0.565
	Distance 1000: Direction W	-0.42	0.21	60	-1.99	**0.052**
	Distance 4000: Direction W	-0.71	0.21	60	-3.45	**0.001**
	Direction W:Location Nesjavellir	-1.31	0.30	60	-4.36	**0.000**
	Distance 1000:Location Nesjavellir	0.05	0.19	60	0.25	0.802
	Distance 4000:Location Nesjavellir	0.40	0.19	60	2.07	**0.043**
	Co-variate					
	Elevation	0.00	0.00	60	2.03	**0.047**
log (Boron concentration in moss + 1.38) - Hellisheidi	Intercept- Distance 250, Direction E	0.34	0.28	10	1.22	0.250
	Distance 1000	0.00	0.15	56	0.01	0.993
	Distance 4000	0.01	0.21	59	0.03	0.978
	Direction W	0.97	0.37	8	2.59	**0.031**
	Distance 1000: Direction W	-0.32	0.18	56	-1.74	0.088
	Distance 4000: Direction W	-0.20	0.26	59	-0.79	0.432
Boron concentration in moss - Nesjavellir	**NA**					

Table A1a. Continued linear mixed models for sulphur, arsenic, boron, antimony and mercury concentrations in Racomitrium lanuginosum *shoots at the damaged moss patch sampling areas. Estimates indicate the effects of fixed factors and co-variates compared to the intercept and followed by a test statistic. Bold p values indicate significant effects.*

Response	**Predictors**	**Estimate**	**Std error**	**DF**	**t-value**	**p-value**
log (Antimony concentration in moss)	Intercept- Distance 250, Direction E, Location Hellisheidi	-3.57	0.24	12	-14.95	0.000
	Distance 1000	-0.72	0.14	12	-5.03	**0.000**
	Distance 4000	-0.88	0.15	12	-5.98	**0.000**
	Direction W	0.89	0.15	12	5.85	**0.000**
	Location Nesjavellir	-0.81	0.13	12	-6.07	**0.000**
	Direction W: Location Nesjavellir	-1.12	0.18	12	-6.11	**0.000**
	Co-variate					
	Soil nitrogen	4.68	1.03	12	4.53	**0.001**
log (Antimony concentration in moss) - Hellisheidi	Intercept- Distance 250, Direction E	-3.65	0.13	60	-28.56	0.000
	Distance 1000	0.05	0.16	60	0.31	0.758
	Distance 4000	0.14	0.15	60	0.94	0.350
	Direction W	2.18	0.15	60	14.73	**0.000**
	Distance 1000: Direction W	-0.25	0.20	60	-1.26	0.214
	Distance 4000: Direction W	-0.57	0.21	60	-2.72	**0.009**
log (Antimony concentration in moss) - Nesjavellir	Intercept- Distance 250, Direction E	-3.89	0.17	6	-23.02	0.000
	Distance 1000	0.09	0.10	54	0.91	0.369
	Distance 4000	-0.27	0.13	60	-2.07	**0.043**
	Direction W	0.20	0.23	5	0.85	0.431
log (mercury concentration in moss + 0.0137)	Intercept- Distance 250, Direction E, Location Hellisheidi	-2.18	0.24	60	-8.96	0.000
	Distance 1000	-0.35	0.13	60	-2.58	**0.012**
	Distance 4000	-0.43	0.14	60	-3.01	**0.004**
	Direction W	-0.53	0.13	60	-4.11	**0.000**
	Location Nesjavellir	-0.78	0.11	60	-6.75	**0.000**
	Co-variates					
	Soil arsenic	0.88	0.14	60	6.40	**0.000**
	soil carbon	0.09	0.02	60	4.52	**0.000**
	soil water content	0.00	0.00	60	-2.37	**0.021**
	Elevation	0.00	0.00	60	-3.07	**0.003**
log(mercury concentration in moss + 0.0313) - Hellisheidi	Intercept- Distance 250, Direction E	0.64	0.74	60	0.87	0.390
	Distance 1000	0.69	0.18	60	3.83	**0.000**
	Distance 4000	0.61	0.15	60	4.08	**0.000**
	Direction W	-0.54	0.22	60	-2.43	**0.018**
	Co-variates					
	Soil carbon	-0.02	0.01	60	-1.76	0.083
	Elevation	-0.01	0.00	60	-4.26	**0.000**
log(mercury concentration in moss + 0.0145) - Nesjavellir	Intercept- Distance 250, Direction E	-2.70	0.06	8	-43.98	0.000
	Distance 1000	-0.25	0.06	6	-4.08	**0.006**
	Distance 4000	-0.38	0.05	6	-7.21	**0.000**
	Direction W	0.06	0.07	6	0.79	0.462
	Co-variates					
	Soil carbon	0.00	0.00	56	-0.28	0.778
	Elevation	0.00	0.00	6	0.63	0.550

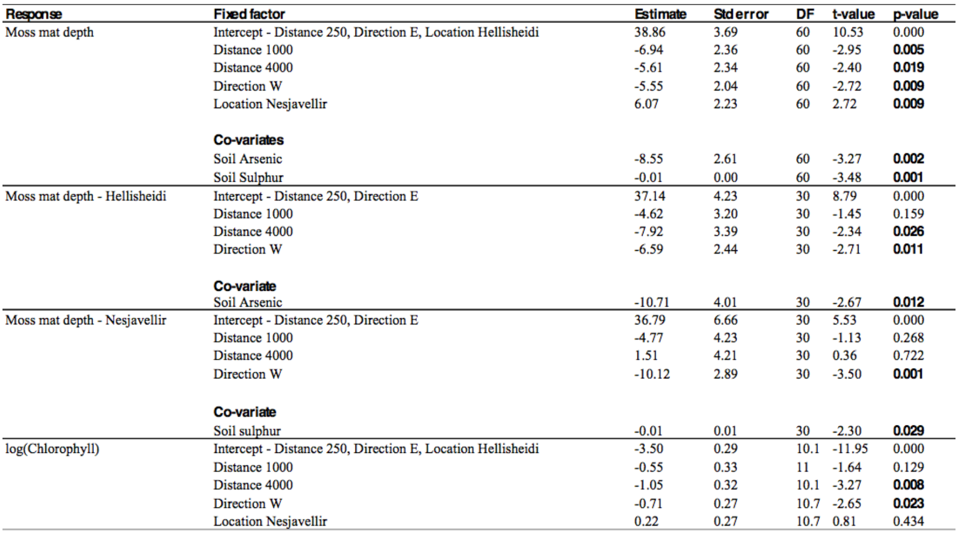
Table A1b. Linear models for sulphur, arsenic, boron, antimony and mercury concentrations in soils at the damaged moss patch sampling areas. Estimates indicate the effects of the predictors and co-variates compared to the intercept and followed by a test statistic. Bold p values indicate significant effects.

Response	**Predictors**	**Estimate**	**Std error**	**t-value**	**p-value**
log(Sulphur concentration in soil)	Intercept - Distance 250, Direction E, Location Hellisheidi	5.58	0.30	18.77	0.000
	Distance 1000	-0.10	0.19	-0.55	0.602
	Distance 4000	-0.25	0.19	-1.32	0.236
	Direction W	0.00	0.17	-0.03	0.978
	Location Nesjavellir	0.11	0.13	0.86	0.421
	Co-variate				
Residual standard error: 0.21, **DF** 6	Soil nitrogen	2.87	1.34	2.15	0.075
Arsenic concentration in soil	Intercept - Distance 250, Direction E, Location Hellisheidi	0.32	1.12	0.28	0.786
	Distance 1000	-0.94	0.70	-1.35	0.225
	Distance 4000	-0.18	0.72	-0.25	0.808
	Direction W	0.27	0.63	0.43	0.685
	Location Nesjavellir	-1.26	0.49	-2.58	**0.042**
	Co-variate				
Residual standard error: 0.80, **DF** 6	Soil nitrogen	7.30	5.03	1.45	0.197
Arsenic concentration in soil - Hellisheidi	Intercept - Distance 250, Direction E, Location Hellisheidi	0.88	1.98	0.44	0.734
	Distance 1000	-0.83	1.45	-0.57	0.669
	Distance 4000	1.26	1.42	0.89	0.537
Residual standard error: 1.10 , **DF 1**	Direction W	0.48	1.34	0.36	0.782
Arsenic concentration in soil - Nesjavellir	Intercept - Distance 250, Direction E, Location Hellisheidi	0.00	0.49	0.01	0.997
	Distance 1000	-0.42	0.23	-1.81	0.322
	Distance 4000	-0.97	0.26	-3.78	0.165
Residual standard error: 0.20, **DF** 1	Direction W	0.70	0.20	3.46	0.179
Boron concentration in soil	Intercept - Distance 250, Direction E, Location Hellisheidi	2.27	4.55	0.50	0.633
	Distance 1000	0.18	2.78	0.07	0.950
	Distance 4000	-0.77	2.87	-0.27	0.796
	Direction W	-1.22	2.48	-0.49	0.637
	Location Nesjavellir				
	Co-variate				
Residual standard error: 3.21 , **DF** 7	Soil nitrogen	3.85	19.00	0.20	0.845
Antimony concentration in soil	Intercept - Distance 250, Direction E, Location Hellisheidi	0.00	0.08	-0.01	0.995
	Distance 1000	-0.08	0.05	-1.55	0.173
	Distance 4000	-0.05	0.05	-0.88	0.411
	Direction W	0.02	0.05	0.35	0.736
	Location Nesjavellir	-0.12	0.04	-3.25	**0.017**
	Co-variate				
Residual standard error: 0.06, **DF 6**	Soil nitrogen	0.92	0.38	2.43	**0.051**
Antimony concentration in soil - Hellisheidi	Intercept - Distance 250, Direction E	0.13	0.06	2.18	0.161
	Distance 1000	0.04	0.07	0.47	0.686
	Distance 4000	0.16	0.07	2.07	0.175
Residual standard error: 0.07, **DF** 2	Direction W	0.06	0.06	1.04	0.409
Antimony concentration in soil - Nesjavellir	Intercept - Distance 250, Direction E	-0.12	0.01	-13.66	**0.047**
	Distance 1000	-0.03	0.00	-7.75	0.082
	Distance 4000	-0.13	0.00	-27.84	**0.023**
	Direction W	0.07	0.00	19.37	**0.033**
	Co-variate				
Residual standard error: 0.00, **DF** 1	Soil nitrogen	0.96	0.03	28.65	**0.022**
Mercury concentration in soil	Intercept - Distance 250, Direction E, Location Hellisheidi	0.07	0.04	1.83	0.109
	Distance 1000	-0.01	0.04	-0.19	0.854
	Distance 4000	0.01	0.04	0.32	0.760
	Direction W	0.03	0.03	0.99	0.357
Residual standard error: 0.06, **DF 7**	Location Nesjavellir	-0.03	0.03	-0.88	0.407

Table A2. Characteristics (mean ± SE) of Racomitrium lanuginosum *in the damaged moss patch sampling areas around Hellisheidi and Nesjavellir power plants in Iceland. n = 6 per location.*

Moss response variable	Direction	Distance (m)	Nesjavellir	Hellisheidi
Moss mat depth (cm)	W	250	20.07 ± 4.74	20.77 ± 3.44
		1000	18.42 ± 2.33	9.07 ± 1.13
		4000	21.10 ± 3.31	13.73 ± 3.55
	E	250	18.66 ± 2.54	28.30 ± 1.50
		1000	24.98 ± 3.19	24.65 ± 5.38
		4000	34.75 ± 1.88	10.93 ± 2.12
Chlorophyll concentration mg/g	W	250	0.02 ± 0.01	0.02 ± 0.01
		1000	0.01 ± 0.00	0.01 ± 0.00
		4000	0.02 ± 0.01	0.19 ± 0.08
	E	250	0.06 ± 0.01	0.03 ± 0.00
		1000	0.02 ± 0.00	0.04 ± 0.01
		4000	0.02 ± 0.01	0.01 ± 0.00

Table A2a. Linear mixed models for the moss plant traits at the damaged moss sampling areas. Estimate indicates effects of the fixed factors and co-variates compared to the intercept and followed by a test statistic. Bold p values indicate significant effects.



Response	**Fixed factor**	**Estimate**	**Std error**	**DF**	**t-value**	**p-value**
Moss mat depth	Intercept - Distance 250, Direction E, Location Hellisheidi	38.86	3.69	60	10.53	0.000
	Distance 1000	-6.94	2.36	60	-2.95	**0.005**
	Distance 4000	-5.61	2.34	60	-2.40	**0.019**
	Direction W	-5.55	2.04	60	-2.72	**0.009**
	Location Nesjavellir	6.07	2.23	60	2.72	**0.009**
	Co-variates					
	Soil Arsenic	-8.55	2.61	60	-3.27	**0.002**
	Soil Sulphur	-0.01	0.00	60	-3.48	**0.001**
Moss mat depth - Hellisheidi	Intercept - Distance 250, Direction E	37.14	4.23	30	8.79	0.000
	Distance 1000	-4.62	3.20	30	-1.45	0.159
	Distance 4000	-7.92	3.39	30	-2.34	**0.026**
	Direction W	-6.59	2.44	30	-2.71	**0.011**
	Co-variate					
	Soil Arsenic	-10.71	4.01	30	-2.67	**0.012**
Moss mat depth - Nesjavellir	Intercept - Distance 250, Direction E	36.79	6.66	30	5.53	0.000
	Distance 1000	-4.77	4.23	30	-1.13	0.268
	Distance 4000	1.51	4.21	30	0.36	0.722
	Direction W	-10.12	2.89	30	-3.50	**0.001**
	Co-variate					
	Soil sulphur	-0.01	0.01	30	-2.30	**0.029**
log(Chlorophyll)	Intercept - Distance 250, Direction E, Location Hellisheidi	-3.50	0.29	10.1	-11.95	0.000
	Distance 1000	-0.55	0.33	11	-1.64	0.129
	Distance 4000	-1.05	0.32	10.1	-3.27	**0.008**
	Direction W	-0.71	0.27	10.7	-2.65	**0.023**
	Location Nesjavellir	0.22	0.27	10.7	0.81	0.434

Table A3. Soil characteristics for the damaged moss patch sampling areas around the Hengill geothermal area in Iceland, n= 1 per station. Data from sampling points per station is pooled to represent a station. See Figure 1. for specific transect names in the given directions below. Elevation data for these areas is similar to that of the systematic sampling stations.

Soil characteristics	Direction	Distance	Nesjavelllir	Hellisheidi
Carbon (%)	W	250	8.30	5.80
		1000	7.36	11.23
		4000	7.70	20.10
	E	250	5.98	2.02
		1000	3.67	7.25
		4000	3.63	8.61
Nitrogen (%)	W	250	0.37	0.25
		1000	0.39	0.42
		4000	0.40	0.35
	E	250	0.30	0.13
		1000	0.26	0.31
		4000	0.32	0.35
Soil moisture (%)	W	250	93.40	89.19
		1000	76.66	117.49
		4000	82.12	162.18
	E	250	89.38	32.60
		1000	69.61	116.47
		4000	80.90	105.47
Soil pH	W	250	5.04	5.60
		1000	6.52	5.81
		4000	5.52	6.00
	E	250	5.43	5.95
		1000	5.92	6.28
		4000	5.99	6.23

Table A3a. Linear models for soil characteristics and elevation at the damaged moss patch sampling areas. Estimates indicate the effects of the predictors and co-variates compared to the intercept and followed by a test statistic. Bold p values indicate significant effects.

Response	**Predictors**	**Estimate**	**Std error**	**t-value**	**p-value**
Soil carbon	Intercept - Distance 250, Direction E, Location Hellisheidi	9.34	4.92	1.90	0.100
	Distance 1000	-0.75	3.69	-0.20	0.845
	Distance 4000	-3.59	5.22	-0.69	0.515
	Direction W	4.95	2.46	2.01	0.085
Residual SE: 4.26**DF** 7	Location Nesjavellir	-5.40	4.26	-1.27	0.246
Soil nitrogen	Intercept - Distance 250, Direction E, Location Hellisheidi	0.17	0.08	2.15	0.068
	Distance 1000	0.03	0.06	0.47	0.655
	Distance 4000	0.12	0.08	1.44	0.193
	Direction W	0.09	0.04	2.21	0.063
Residual SE: 0.07,**DF** 7	Location Nesjavellir	0.12	0.07	1.76	0.121
Soil moisture	Intercept - Distance 250, Direction E, Location Hellisheidi	102.71	40.09	2.56	0.037
	Distance 1000	-22.60	30.06	-0.75	0.477
	Distance 4000	-26.53	42.52	-0.62	0.552
	Direction W	40.73	20.04	2.03	0.082
Residual SE: 34.72,**DF** 7	Location Nesjavellir	-19.42	34.72	-0.56	0.593
Soil pH	Intercept - Distance 250, Direction E, Location Hellisheidi	6.06	0.54	11.11	0.000
	Distance 1000	0.03	0.41	0.08	0.939
	Distance 4000	0.04	0.58	0.06	0.952
	Direction W	-0.22	0.27	-0.82	0.440
Residual SE: 0.47,**DF** 7	Location Nesjavellir	-0.22	0.47	-0.47	0.650
Elevation	Intercept - Distance 250, Direction E, Location Hellisheidi	255.27	113.64	2.25	0.060
	Distance 1000	0.09	85.23	0.00	0.999
	Distance 4000	42.64	120.53	0.35	0.734
	Direction W	32.26	56.82	0.57	0.588
Residual SE: 98.41,**DF** 7	Location Nesjavellir	-32.32	98.41	-0.33	0.752

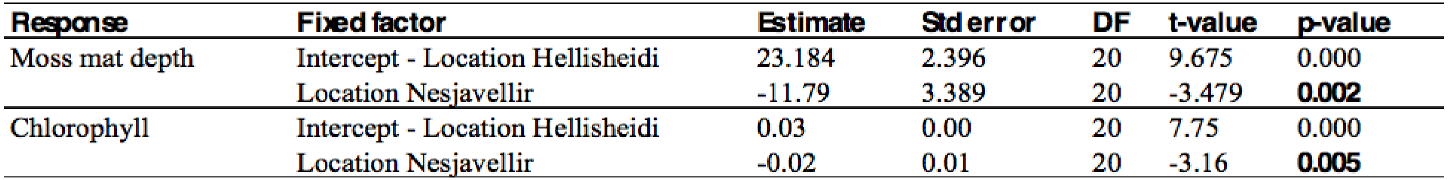
Table A4 Concentrations of sulphur, arsenic, boron, antimony and mercury in Racomitrium lanuginosum *shoots and soil (mg/kg dry weight), collected from extensively damaged areas at two power plants at Hellisheidi and Nesjavellir, in the Hengill geothermal area, Iceland (n = 10 per station for moss and n = 1 per station for soil). Some boron concentrations were below the 1 mg/kg detection limit in shoots. NA- Not Available.*

		Nesjavellir		Hellisheidi	
Element	Distance (m)	Shoot	Soil	Shoot	Soil
Sulphur	267	1785.00 ± 135.72	615.33	NA	NA
	830	NA	NA	1061.60 ± 44.12	1276.67
Arsenic	267	0.76 ± 0.12	2.87	NA	NA
	830	NA	NA	1.35 ± 0.13	0.79
Boron	267	<1	4.88	NA	NA
	830	NA	NA	<1	2.59
Antimony	267	0.08 ± 0.00	0.25	NA	NA
	830	NA	NA	0.10 ± 0.01	0.11
Mercury	267	0.24 ± 0.04	0.08	NA	NA
	830	NA	NA	0.09 ± 0.00	0.05

Table A4a. Linear mixed models for sulphur, arsenic, boron, antimony and mercury concentrations in Racomitrium lanuginosum *shoots at the extensively damaged sampling areas at Hellisheidi (Transect 830 m) and Nesjavellir (Transect 267 m). Estimates indicate the effects of fixed factors and co-variates compared to the intercept and followed by a test statistic. Bold p values indicate significant effects. NA- Not Available.*

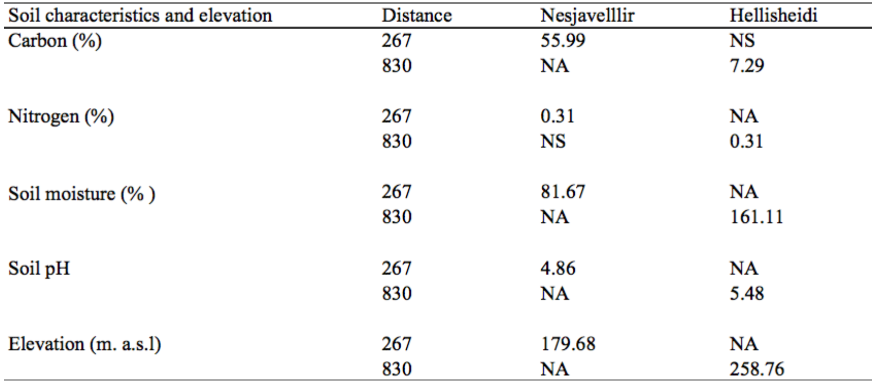
Response	**Predictors**	**Estimate**	**Std error**	**DF**	**t-value**	**p-value**
log(Sulfur concentration in moss)	Intercept - Location Hellisheidi	6.96	0.06	20	125.11	0.000
	Location Nesjavellir	0.50	0.08	20	6.36	**0.000**
log(Arsenic concentration in moss)	Intercept - Location Hellisheidi	1.35	0.09	20	14.46	0.000
	Location Nesjavellir	-0.59	0.13	20	-4.45	**0.000**
Boron concentration in moss	**NA**					
Antimony concentration in moss	Intercept - Location Hellisheidi	0.10	0.01	20	15.96	0.000
	Location Nesjavellir	-0.02	0.01	20	-2.45	**0.023**
log (mercury concentration in moss)	Intercept - Location Hellisheidi	0.09	0.03	20	2.69	0.014
	Location Nesjavellir	0.15	0.05	20	3.07	**0.006**

Table A5. Linear mixed models for the moss plant traits at the extensively damaged sampling areas at Hellisheidi (Transect 830 m) and Nesjavellir (Transect 267 m). Estimate indicates effects of the fixed factors and co-variates compared to the intercept and followed by a test statistic. Bold p values indicate significant effects.



Response	Fixed factor	Estimate	Std error	DF	t-value	p-value
Moss mat depth	Intercept - Location Hellisheidi	23.184	2.396	20	9.675	0.000
	Location Nesjavellir	-11.79	3.389	20	-3.479	**0.002**
Chlorophyll	Intercept - Location Hellisheidi	0.03	0.00	20	7.75	0.000
	Location Nesjavellir	-0.02	0.01	20	-3.16	**0.005**

Table A6. Soil characteristics for the extensively damaged sampling areas around the Hengill geothermal area in Iceland, n= 1 per location. Data shows means of pooled samples from sampling points per station. In Fig. 1., the specific transect names are given as HP-ER for Hellisheidi and NP-ER – for Nesjavellir. Elevation data for the areas is also shown. NA – Not Available.



Soil characteristics and elevation	Distance	Nesjavelllir	Hellisheidi
Carbon (%)	267	55.99	NS
	830	NA	7.29
Nitrogen (%)	267	0.31	NA
	830	NS	0.31
Soil moisture (%)	267	81.67	NA
	830	NA	161.11
Soil pH	267	4.86	NA
	830	NA	5.48
Elevation (m. a.s.l)	267	179.68	NA
	830	NA	258.76

Paper III

Effects of experimental wet hydrogen sulphide deposition on dominant plants around geothermal power plants in Kenya and Iceland.

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(*Manuscript*)

Authors contributions:

- Conceived and designed the study: TMM, ISJ
- Performed experiment: TMM, ISJ
- Performed laboratory work: TMM
- Analysed the data: TMM, ISJ
- Wrote the paper: TMM, ISJ

Effects of experimental hydrogen sulphide deposition on dominant plants around geothermal power plants in Kenya and Iceland.

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Abstract

Previous studies show that plants close to geothermal power plants can accumulate sulphur from emitted H_2S gas, but the responses in terms of growth and physiology are not well described. We carried out two separate controlled experiments on dominant plants around geothermal power plants in Kenya and Iceland, i.e. the shrub *Tarchonathus camphoratus* (using seedlings) and the moss *Racomitrium lanuginosum*, respectively. We tested the hypothesis that sulphur concentration and accumulation in plant tissues would increase with increasing concentrations of wet hydrogen sulphide deposition, with consequences for plant growth and health. We irrigated the plants with 0, 30, 100 and 300 µg /L hydrogen sulphide gas dissolved in distilled water, for 6.5 (shrub) and 13 (moss) weeks, and measured plant responses in terms of sulphur concentrations (and calculated accumulation), foliar damage, growth, chlorophyll concentrations and contents (total amount). Due to lack of roots and their sensitivity to atmospheric depositions, we expected mosses to respond more strongly than the shrub. The treatments did not affect sulphur concentrations and accumulation in shrub leaves, nor did they affect foliar damage or chlorophyll concentrations and content of seedlings. However, stem height increase was greatest at intermediate H_2S exposure. The treatments had no effect on sulphur concentration and accumulation, biomass increase or chlorophyll concentrations/contents of moss shoots, but shoot length was reduced at high H_2S concentration exposures. We thus conclude that short-term exposure to moderate levels of H_2S (watering with 30 µg/L H_2S solution) is not harmful to either of the two plants and may even stimulate shrub growth while high levels may reduce moss growth.

Keywords: Hydrogen sulphide emissions, geothermal power plants, sulphur accumulation, *Tarchonanthus camphoratus*, *Racomitrium lanuginosum*, plant growth, chlorophyll.

Introduction

Hydrogen sulphide (H_2S) gas from geothermal power plants using hot water and steam for electricity generation, is the second most abundant non condensable gas (NCG) emitted after carbon dioxide. Generally, it constitutes 1- 5% w/w of the NCGs fraction but may in rare cases reach up to 24% (Axtmann, 1975; Rodríguez, 2014). Its deposition (in various forms of sulphur) and sulphur accumulation in terrestrial ecosystems around geothermal power plants is a growing concern (Bargagli et al., 1997; Bussotti et al., 2003, 1997; Mutia et al., 2016a, b), particularly because of its potential phyto-toxicity at elevated levels (Bussotti et al., 1997).

Emissions of H_2S from geothermal power plants may have severe ecological consequences as wet or gaseous deposition (Kellogg et al. 1972). H_2S gas is relatively unstable in air and can undergo oxidation to form SO_2, and with even further reactions to produce sulphuric acid (Kellogg et al., 1972). In the presence of precipitation, the likelihood of acid precipitation around the power plants or further away is high but depends on the wind dispersion of the gas. This is of concern as acid precipitation can affect plant growth, survival and nutrient cycling in ecosystems (Likens et al., 1972).

Plants and lichens have been used as bio-monitors to map the deposition and distributions of geothermally emitted elements from geothermal power plants (Bargagli et al., 1997; Bussotti et al., 2003, 1997; Mutia et al., 2016a, b). These studies have shown clear patterns of increased sulphur concentration in plants and lichens close to the power plants. However, the associated effects of increased H_2S levels on the growth and physiology of plants remain unclear. Some indications of leaf damage including necrosis, chlorosis, reduced growth and early senescence have been reported (Varshney et al., 1979; Bussotti et al., 1997; Chiarucci et al., 2008; Bussotti et al., 2003; Mutia et al., 2016a, b), but still it is not established whether these are caused by excess sulphur or other environmental factors (Bussotti et al., 1997).

Assessment of the effects of a single pollutant on plant growth in the field is complicated by several interacting biotic and abiotic factors, including other pollutants. Only a few experimental studies have evaluated the effects of H_2S gas emissions on plants, mostly using vascular plants in fumigation experiments (Thompson and Kats, 1978; Gonzales, 1984; Maas et al., 1987, 1988). For example, Thompson and Kats, (1978) assessed the effects of continuous fumigation of H_2S on crops and forest plants in a greenhouse, and showed stimulated plant growth at 30 – 100 ppb H_2S gas and damaging effects at 300 – 3000 ppb. The damages were characterized by leaf lesions, defoliation and stunted growth (Thompson and Kats, 1978). It is worth noting that at optimum concentrations (variable between plants), sulphur serves as a macro-nutrient essential for plant metabolism and growth, but becomes phyto-toxic at high levels (Linzon et al., 1979). Other studies report decreased plant growth with H_2S fumigation at different concentrations, although the effects vary across different species (Gonzales, 1984; Maas et al., 1987). On the other hand, experimental studies on the effects of H_2S gas on non-vascular plants, such as mosses, which dominate plant communities in many subarctic ecosystems, are limited. Existing studies are mainly based on field surveys that assess sulphur levels in mosses growing around geothermal power plants (Baldi, 1988; Berg and Steinnes, 1997; Bargagli et al., 2002; Mutia et al., 2016b). There is more information, however, on sulphur effects related to experimental sulphur dioxide (SO_2) deposition on mosses. A study by LeBlanc and Rao, (1973) revealed damages on the mosses *Orthotrichum obtusifolium* and *Pylaisia polyantha* at 5 and 30 ppb SO_2 exposures: moss colour changed from green to golden brown and leaf cells were plasmolysed.

In the design and implementation of appropriate mitigation measures toward sustainable geothermal development in Kenya and Iceland, where there are on-going and future geothermal development plans (Omenda et al., 2014; Ragnarsson, 2015), it is important to establish the effects of the power plant emissions. The aim of this study is therefore to assess plant responses to excess sulphur (in H_2S form) in dominant plants around geothermal power plants in Kenya (Mutia et al., 2016a) and Iceland (Mutia et al., 2016b) by experimentally evaluating causal relationships between wet H_2S and plant growth and health. Since existing experimental studies have assessed the effect of dry H_2S deposition (fumigation experiments) on plants, we chose to study effects related to wet H_2S deposition. This is owing to limited knowledge on this, and the fact that around power plants emitted H_2S may dissolve in precipitation before deposition in ecosystems.

We studied the effect of wet hydrogen sulphide deposition at different concentrations on two plant species: seedlings of the shrub *Tarchonanthus camphoratus* L., dominant around geothermal power plants in Kenya, and the moss *Racomitrium lanuginosum* (Hedw.) Brid., dominant around the Hengill geothermal power plants in Iceland. We hypothesized that with increasing concentration of wet H_2S exposure, sulphur would be enriched and accumulate in the plants with consequences for plant growth and health. Specifically, we expected sulphur enrichment in plant tissue when exposed to high H_2S concentration (> 30 µg/L) leading to increased foliar damage, reduced plant growth and decreased chlorophyll levels. We expected stronger responses in *R. lanuginosum* than in the *T. camphoratus* seedlings due to lack of roots and general sensitivity of mosses to atmospheric pollutants (Rao, 1982).

Materials and methods

We carried out two experiments, one in each country. In Kenya, the experiment was performed on seedlings of the shrub *T. camphoratus* (Experiment 1) and in Iceland on extracted moss mats of *R. lanuginosum* (Experiment 2).

We prepared H_2S treatment solutions at 0, 30, 100 and 300 µg/L (ppb) concentrations in distilled water. These solutions correspond to air saturated water with H_2S concentrations in air of 0 ppm, 10.96 ppm, 36.52 ppm, and 109.57 ppm, respectively (using a Henry's law constant of 0.001 mol/(L*atm), (Sander, 2015)). To prepare the treatments, a H_2S stock solution was initially prepared from 5% hydrogen sulphide gas in nitrogen. The gas was first bubbled in a 20 L distilled water container for 20 minutes and pH measurements taken at 5 minute intervals until there was no change (saturation). H_2S concentration in the stock solution was then determined using mercury acetate, according to methods described in Arnórsson et al. (2006), and ranged between $3.0 \cdot 10^4$ and $4.0 \cdot 10^4$ µg/L. From the stock solution, we prepared the H_2S gas concentrations for the treatments and further confirmed their concentrations via titration (Arnórsson et al., 2006). The solutions were made immediately before each use.

For each experiment, a similar nested design was adopted with four units randomly assigned to each treatment and multiple measurements conducted per unit.

Experiment 1: set-up and design

Seeds of *T. camphoratus* were obtained from the Menengai forest in Kenya (0.2500° S, 36.0833° E, 2278 m a.s.l.). The forest grows and extends over the Menengai caldera, a trachytic volcano in the Kenya Rift Valley (Leat, 1984). The area presently has no geothermal power plants and is under exploration for geothermal development, with drilling of geothermal wells ongoing. Seeds were sown at a tree nursery within the caldera between August and September 2014 and the seedlings pricked after one month, and potted in perforated polythene bags (13 cm x 20 cm) filled with volcanic soil of sandy texture obtained from the area, and nurtured at the tree nursery for a period of seven months, to attain optimum growth (3 – 4 true leaves) for transplanting and acclimation to environmental conditions.

Potted seedlings were later transferred to an outdoor open ground in Nakuru, Kenya (0.2777° S and 36.0504° E, 1889 m a.s.l) where the experiment was conducted between 18th March and 1st May 2014. Over the course of the experiment, seedlings were exposed to three rain events totaling 88 mm. The daily temperature ranges were between 17 and 20° C as measured at the Mlima Punda automatic weather station at Menengai (Geothermal Development Company Ltd, unpublished data 2014).

Seedlings were arranged in groups of ten per experimental unit, for a total of sixteen units. Within each unit, five seedlings were randomly chosen and labelled for measurements and sampling, and the remaining five served as reserves in case of mortality, for a total of 80 seedlings across all treatments.

The H_2S solutions were applied to the *T. camphoratus* seedlings. Ten liters of the solutions were applied per group four times a week using watering cans (simulating a rainfall event). The experiment was performed for a period of 6.5 weeks (45 days). The duration was chosen based on the growth rates of seedlings (average 3.92 cm stem height increase during the experiment), and that was assumed long enough to detect effects of the treatments.

Experiment 1: growth measurements, chlorophyll and sulphur determination

Growth related variables i.e. stem height, number of stems and number of healthy green leaves per seedling were measured/counted at the beginning and end of the experiment. Foliar damage was assessed at the end of the experiment based on leaf colour and appearance, as previously described in Mutia et al. (2016a): A) healthy green leaves, B) yellow leaves and C) brown dead leaves. The proportion of damaged leaves on each seedling was calculated as the number of leaves in categories B and C over the total number of leaves. Some seedlings had damaged leaves at the beginning of the experiment, but there were no differences in the proportion of damage among treatment groups prior to the experimental manipulations (Chisq=5.168, p=0.160), so we assumed that any potential differences at the end of the experiment would be due to the H_2S exposure. Foliar damage, i.e. the proportion of damaged leaves (categories B and C) in control seedlings at the beginning and the end of the experiment, was compared using a paired t-test to assess the effect of time, independent of the experimental manipulations; results showed that the proportion of foliar damage did not significantly change over the course of the experiment (t = -2.011, p = 0.058), although marginally significant, this may indicate that foliar damage occured to the plants during the course of the experiment even though no treatment was applied.

At the end of the experiment, wearing polythene gloves, all leaves were carefully removed from each seedling and grouped according to the three damage categories. At the Geothermal Development Company Ltd (GDC) laboratory, each sample was washed in distilled water, dried at room temperature in the dark and divided into two parts, one for chlorophyll determination and the other for total sulphur analysis. Chlorophyll concentrations were determined in A and B leaves (not in the dead C leaves). For chlorophyll determination, each sample was milled, weighed and split into two sub samples, one for chlorophyll concentration analysis and the other for dry weight determination (after oven drying to a constant weight at 70°C for 24 hours). Ten ml of 96% ethanol was added to 0.5 g of each sample and the mixture hand shaken for 15 seconds. The samples were then covered by aluminum foil to prevent light exposure and allowed to stand for 24 hours at 6°C in darkness and centrifuged for 10 min at 1000 revolutions per min. 3.5 ml samples were extracted and transferred to 4 ml cuvettes for analysis at the Institute of Freshwater

Fisheries in Iceland (modified from Sumanta et al., 2014). To determine chlorophyll concentrations, light absorbance at wavelengths of 750 nm, 663 nm and 652 nm was measured using a spectrophotometer (HACH LANGE UV Visible Spectrophotometer, DR 5000). Chlorophyll concentration in mg/g dry weight was calculated according to Arnon (1949). For sulphur determination, leaf samples were analysed using standard analytical procedures at the internationally accredited ALS Scandinavia labs in Luleå, Sweden. Prior to analysis, samples were acid digested (in 5 ml conc. HNO_3 + 0.5 ml 30% H_2O_2) in closed teflon containers in a microwave. Sulphur analyses were conducted using an Element 2 ICP MS according to (modified) U.S.EPA methods 200.8 (U.S.EPA, 1994) and SS EN ISO 17294 parts 1 (ISO, 2005) and 2 (ISO, 2003). Procedural blanks were below the minimum detection level. Accuracy was checked through analysis of standard in house reference materials for soil (ALS Labs, Sweden) and peach leaves (NIST 1547) (National Institute of Standards and Technology, Gaithersburg, MD, USA; (Rodushkin et al., 2008) and obtained more than 95% recoveries. Estimates of sulphur accumulation and chlorophyll content (total amount) for each seedling were based on the number of leaves in each category, multiplied by the sulphur or chlorophyll concentration of that leaf category in that seedling, and the average leaf weight (0.015±0.002 grams, mean ± SE), and summed across all leaf categories. Concentrations of sulphur in healthy leaves (category A) for each seedling are also compared with the sulphur concentration in the healthy moss.

To account for other factors that influence plant growth and health, we measured soil sulphur concentrations and soil characteristics i.e. pH and moisture (% by weight) in the seedling pots, 80 in total. Each sample was split into three, one for the analysis of sulphur concentration and the other for analysis of soil pH and moisture (% by weight). The samples for sulphur analysis were dried at 50 °C for 48 h to a constant weight, sieved through a 2 mm sieve and analysed using the same protocols and equipment as for the leaves at the ALS Scandinavia labs in Luleå, Sweden. For soil pH, soil solution was extracted from 5 g (<2 mm) of 96 hours air-dried soil in 25 ml de-ionized water, by shaking it for two hours and allowing to settle for 8 hours before measuring pH (Blakemore et al., 1987). Soil moisture (%) by mass was obtained after oven drying 10 g of fresh soil at 105°C for 24 hours to constant weight.

Experiment 2: set-up and design

Sixteen 16 x 24 cm mats of the moss *R. lanuginosum* trimmed to a depth 5 cm were extracted from Raudhalsahraun, a lava field with no geothermal activity, located within the Snaefellsness volcanic belt (Thordarson and Larsen, 2007) in West Iceland (22.2640° W 64.8483°N) at 331 m above sea level. Plastic trays (16 x 24 x 8 cm) were filled with 3 cm tephra at the base (obtained from the same area as the moss) for use as growth substrate over which the extracted moss mats were placed. The experiment was carried out in a growth chamber at the University of Iceland in the late summer-autumn period (2nd August 2013 – 30th October 2013). We maintained constant conditions within the growth chambers with 12 hours day light exposure (Photosynthetically Active Radiation (PAR) 250 μmolm^{-2}s^{-1}) and air temperatures between 17 - 20 °C. Optimal growth temperatures for *R. lanuginousm* of 5–13 °C (Tallis, 1964; Kallio and Heinonen, 1973) could not be maintained in the chamber due to heat development from the photosynthetic light bulbs; for the same reason, we had to compromise the light period, even though *R. lanuginosum* grows under almost 24 light hours (Average PAR 170 μmolm^{-2}s^{-1}) in the Icelandic summer.

The hydrogen sulphide treatment solutions were applied to *R. lanuginosum* moss mats. 300 ml of the solutions were applied in each tray four times a week using a mist sprayer, with four replicate trays per treatment. This experiment was conducted for a longer period (90 days, August – October 2013) than experiment 1, owing to the slow growth of mosses.

Experiment 2: shoot growth, moss damage assessments, chlorophyll, and sulphur determination

Moss growth was assessed as shoot length increase and biomass increase over the experimental period using open-ended netlon bags (Jónsdóttir et al. 1999, Armitage et al. 2012). For each bag, twenty fresh moss shoots of *R. lanuginosum* were collected from the same area as the moss mats (Raudhalsahraun) and trimmed to 30 mm apical length. Ten of these shoots were weighed fresh and placed in the tagged bag and carefully inserted into the moss mats within the trays at the beginning (t_0) of the experiment; one bag was included per tray. The other ten shoots were used to determine the ratio between fresh and dry weight (after drying to constant weight at 70°C) to calculate the dry weight of the transplanted shoots at the beginning of the experiment (Jónsdóttir et al., 1999). The transplanted shoots were left in the moss mats until the end of the experiment (t_1). Shoot growth was measured as shoot length increase in excess of the original 30 mm, for each shoot. After the experiment, the same shoots were dried at 70°C to obtain the dry weight at time t_1. Biomass increase was calculated collectively for the ten shoots per netlon bag, by subtracting the calculated dry weight of the ten shoots at time t_0 from the dry weight at time t_1. Moss foliar damage was assessed on a weekly basis by inspecting the colour and appearance of the moss shoots, as described in Mutia et al., (2016b): A) healthy green shoots, B) yellow shoots and C) brown/black dead shoots.

At the end of the experiment, three moss samples were systematically extracted from each tray, at the mid-point and both ends of the tray, and each sample divided into two parts/ subsamples, one for analysis of sulphur concentrations and the other for chlorophyll determination. For all samples, only the uppermost 3 cm (most photosynthetically active) of the moss shoots were used in the analysis. Further sample preparations and analysis in the shoots were performed in the same way and in the same labs as in experiment 1. Calculations of sulphur accumulation and chlorophyll content for moss shoots were based on the biomass of the ten moss shoots in each tray at the end of experiment, multiplied by the average shoot concentrations of sulphur or chlorophyll per tray.

Data analyses

Sulphur concentration and accumulation in seedlings of *T. camphoratus*, foliar damage, growth measurements (stem height increase, change in number of stems, and change in number of healthy green leaves per seedling) and chlorophyll concentrations and content of seedlings were separately analysed using Linear Mixed effects Models (LMM) or Generalized Linear Mixed effects Models (GLMM). Count data (change in number of stems and number of green leaves) and proportional data (foliar damage) were analysed with GLMM using a poisson and binomial distribution, respectively. The experimental treatment (0, 30 µg/L, 100 µg/L and 300 µg/L) was included as a fixed factor and sampling units were included as a random factor. Soil characteristics (soil pH and% soil moisture) and soil sulphur concentrations were included as co-variates. The co-variates were included one at a time and then the best fitted model based on the lowest AIC (Akaike´s Information Criterion) value selected, provided that inclusion of an additional parameter in the model reduced the AIC value by more than 2.0. Significance of the variables in LMM and GLMM was calculated, comparing models with and without the variable of interest, so values are reported as F values and Chisq values, respectively.

Differences in sulphur and chlorophyll concentrations between leaf categories, and across the experimental treatments, were analysed with LMM, including sampling units as a random factor. As fixed factors we included the interaction between leaf category and experimental treatment. Co-variates were also included as indicated above. When the interaction was not significant, it was dropped from the final model.

Sulphur concentration and accumulation in moss shoots, growth measurements (shoot length increase and biomass increase), and chlorophyll concentration and content were analysed using Linear Models (LM) or LMM. LMs were used when one measurement was taken per sampling unit (sulphur accumulation, shoot biomass increase and chlorophyll concentration and content). The random factors included in the LMMs were ‘tray’ for sulphur concentration in *R. lanuginosum* shoots (3 measurements per tray) and sulphur accumulation and ‘sampling bag’ for shoot length increase measurements (one bag per tray, 10 shoots measured in each bag). Foliar damage of mosses was not analysed because moss colour change was only detected in one tray in the H_2S 30 µg/L treatment where the moss formed a brown colour patch (category C) after four weeks.

The models were run in R 3.2.2 (R Development Team, 2010) using the functions *lmer* in the lme4 package (Bates et al., 2014) for the LMM and *lm* in the MASS package (Ripley et al., 2015) for linear models. All plant variable and sulphur concentrations are summarised as mean ± standard error (SE).

Results

Effect of wet H_2S treatments on *T. camphoratus* seedlings

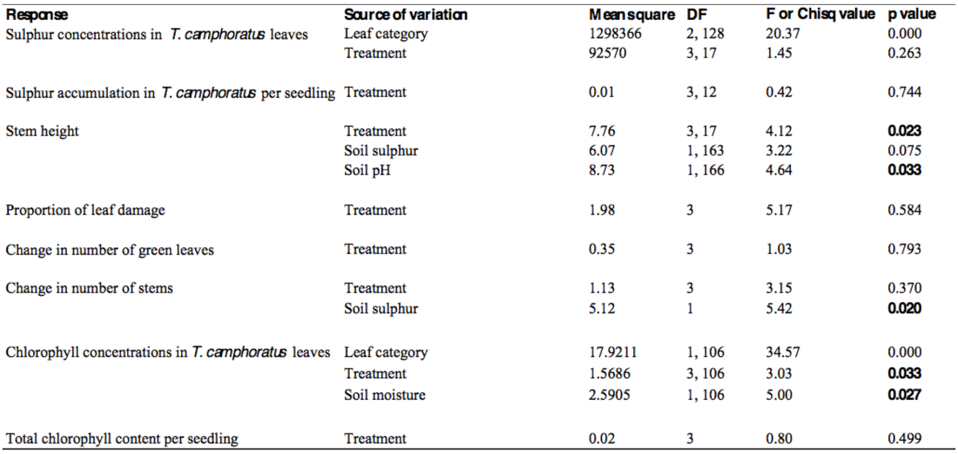
On average, the S concentration in the leaves was 1639.44±94.38 mg/kg and the seedlings had accumulated 0.342 ± 0.013 mg of sulphur in their leaves by the end of the experiment for all the treatments (average foliar dry mass per seedling was 0.233±0.050 grams). Sulphur concentration in the different leaf categories did not differ across the experimental treatments (Table 1, Figure 1a). Furthermore, the accumulation of sulphur in the seedlings was not significantly affected by the experimental exposure to increased wet H_2S depositions (Table 1, Figure 1b).

The increase in stem height was on average 3.92±0.14 cm for all treatments and was positively affected by the 30 μg/L treatment while it was not significantly different from the control at higher H_2S levels. (Figure 1c, Table 1).

By the end of the experiment the proportion of damaged leaves was on average 0.244 ± 0.013 for all treatments. Seedlings increased their number of stems by 1 ± 0.105 and on average, their number of green leaves decreased by one (-1 ± 0.477) for all treatments. Chlorophyll concentrations of the leaves averaged 1.45±0.09 mg g^{-1} for leaves A and 0.46±0.08 mg g^{-1} leaves B in all treatments. The average chlorophyll content (total amount) of seedlings was 0.240 ± 0.016 mg, ranging between 0.001 and 0.696 mg. Experimental exposure to wet H_2S deposition did not affect foliar damage (proportion of leaf damage), the number of stems and the number of green leaves (Table 1). Chlorophyll concentration was affected by the treatments, and as expected was at higher levels in the green healthy leaves than yellow leaves; leaves in the 30 μg/L and 100 μg/L treatments had lower chlorophyll levels than the control and the 300 μg/L treatment (Figure 1d). The total chlorophyll content of seedlings was, however, not affected by the experimental exposure (Table 1).

The soil co-variates did not differ across the experimental treatments (Table S1), but they improved some models by accounting for parts of un-explained variation (Table 1).

Table 1: Model results for the effects of H_2S *exposure on responses of* T. camphoratus *seedlings: sulphur concentrations and accumulation, proportion of damaged leaves, stem height increase, change in number of new green leaves, change in number of stems and chlorophyll concentrations and contents. In all models, the effect of the experimental manipulation of* H_2S *exposure ('Treatment') was assessed; covariates ('Soil moisture', 'Soil sulphur' and 'Soil pH') were retained in the final model if they improved model fit. For LMMs, the numerator and denominator degrees of freedom are indicated. F values are reported for sulphur concentrations and accumulation and chlorophyll concentrations and contents per seedling (LMM) while Chisq values are given for the other response variables (GLMM).*



Response	**Source of variation**	**Mean square**	**DF**	**F or Chisq value**	**p value**
Sulphur concentrations in *T. camphoratus* leaves	Leaf category	1298366	2, 128	20.37	0.000
	Treatment	92570	3, 17	1.45	0.263
Sulphur accumulation in *T. camphoratus* per seedling	Treatment	0.01	3, 12	0.42	0.744
Stem height	Treatment	7.76	3, 17	4.12	**0.023**
	Soil sulphur	6.07	1, 163	3.22	0.075
	Soil pH	8.73	1, 166	4.64	**0.033**
Proportion of leaf damage	Treatment	1.98	3	5.17	0.584
Change in number of green leaves	Treatment	0.35	3	1.03	0.793
Change in number of stems	Treatment	1.13	3	3.15	0.370
	Soil sulphur	5.12	1	5.42	**0.020**
Chlorophyll concentrations in *T. camphoratus* leaves	Leaf category	17.9211	1, 106	34.57	0.000
	Treatment	1.5686	3, 106	3.03	**0.033**
	Soil moisture	2.5905	1, 106	5.00	**0.027**
Total chlorophyll content per seedling	Treatment	0.02	3	0.80	0.499

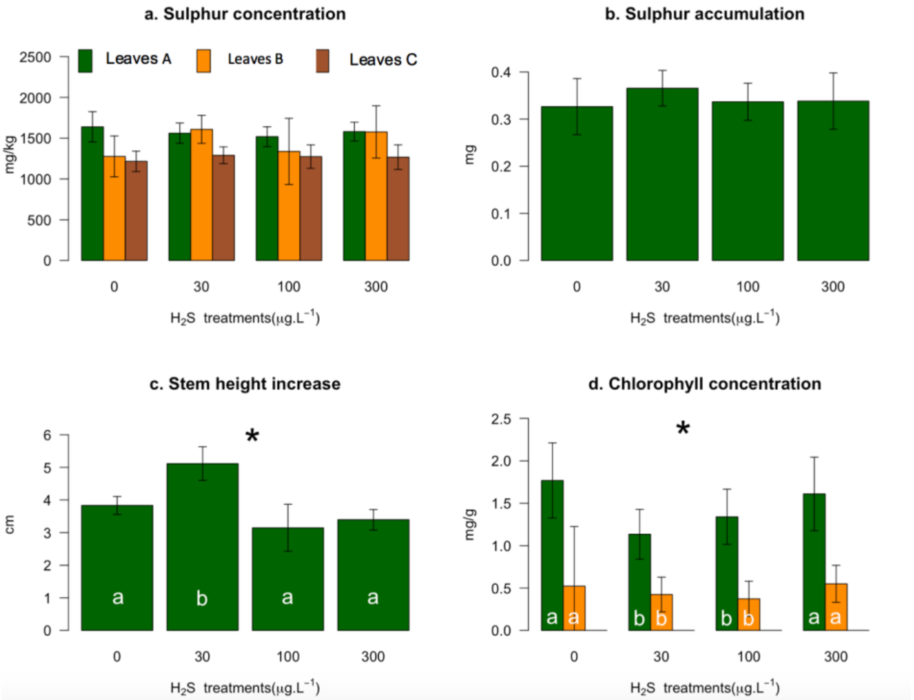


Figure 1. Various responses of T. camphoratus *to H_2S treatments (application of 0* µg/L, *30* µg/L, *100* µg/L *and 300* µg/L*): (a) Sulphur concentrations in leaves, (b) sulphur accumulation per seedling, (c) stem height increase and (d) chlorophyll concentrations after 6.5 weeks in an outdoor experiment (mean ± SE, n = 4). Sulphur (a) and chlorophyll (d) concentrations in leaves of* Tarchonanthus camphoratus *are assigned to different damage categories based on visual assessment: healthy green leaves (leaves A), yellow leaves (leaves B) and dead brown leaves (leaves C), across the different H_2S experimental treatments. Asterisks (*) indicate significant effect of treatment ($p<0.05$) and smaller case letters show differences between treatments.*

Effect of wet H_2S treatments to *R. lanuginosum* moss

The average sulphur concentration in moss was 205±4 mg/kg and the moss had accumulated 0.019±0.002 μg sulphur at the end of the experiment (average biomass weight per ten moss shoots was 0.09±0.006 mg) for all the treatments. The overall treatment effect on the concentration of sulphur in moss was marginally significant (Table 2), and was significantly lower at 30 μg/L wet H_2S deposition than at the higher exposures (Figure 2a), while the controls showed intermediate concentrations. The treatment did not affect sulphur accumulation in moss tissues (Table 2, Figure 2b).

Over the 90 days of the experiment, shoots elongated on average by 0.09±0.008 cm, biomass increased by 0.016±0.003 mg on average, and chlorophyll content of moss shoots ranged between 0.001 and 0.018 µg (average 0.008 ± 0.001 µg) for all treatments. Experimental exposure to H_2S had a significant effect on shoot length increase (Table 2; Figure 2c). Shoot length increase at the highest levels of exposure (300 μg/L H_2S) was significantly lower than at all other treatment levels. At these highest exposures, shoot length increase was reduced by 59%. The treatments did not affect moss biomass increase, or the chlorophyll concentrations and contents of moss shoots (Table 2, Figure 2d).

Comparison of sulphur concentration and accumulation between species

In general, mosses have low levels of nutrients (including sulphur) compared to vascular plant tissues, a reason why dead moss is recalcitrant. As such, the samples of *R. lanuginosum* showed much lower sulphur concentrations than seedlings of *T. camphoratus* (205 ± 4 mg/kg sulphur in moss vs 1441.407 ± 25.742 mg in the healthy leaves of *T. camphoratus* (leaves A)). However, contrary to our predictions the moss did neither become more enriched nor accumulate more sulphur in response to the treatments.

Table 2: Model results for the effects of H_2S exposure on responses of R. lanuginosum*: sulphur concentrations and accumulation, biomass increase, shoot length increase and chlorophyll concentrations and contents. In all models, the effect of the experimental manipulation of H_2S exposure ('treatment') was assessed. For LMMs, numerator and denominator degrees of freedom are indicated.*

Response	**Source of variation**	**Mean square**	**DF**	**F value**	**p value**
Sulphur concentrations in R. lanuginosum	Treatment	1502.1	1,16	3.97	0.064
Sulphur accumulation in *R. lanuginosum*	Treatment	0.00	3	0.66	0.591
Biomass increase	Treatment	0.00	1	0.34	0.570
Shoot length increase	Treatment	0.04	3, 16	3.30	**0.048**
Chlorophyll concentrations in *R. lanuginosum*	Treatment	0.00	3.00	0.61	0.621
Total chlorophyll content per 10 shoots biomass weight	Treatment	0.00	3	0.36	0.782

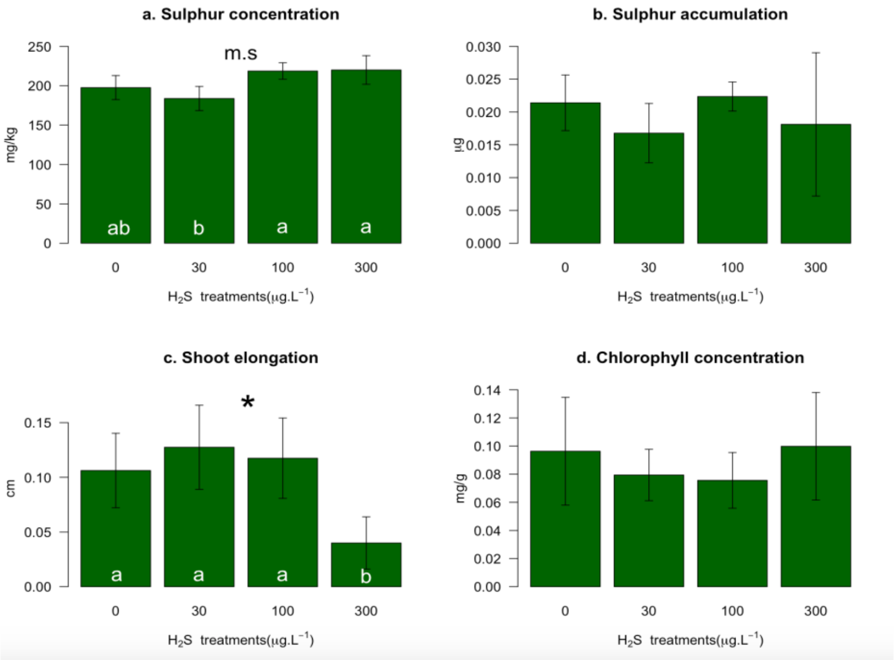


Figure 2. Various responses of R. lanuginosum *to H_2S treatments (application of 0 µg/L, 30 µg/L, 100 µg/L and 300 µg/L): (a) Sulphur concentrations in shoots, (b) sulphur accumulation in shoots, (c) shoot elongation (shoot length increase) and (d) chlorophyll concentrations after 13 weeks in growth chambers (mean ± SE, n = 4). 'm.s' indicates a marginally significant effect of treatment ($p=0.06$), asterisks (*) indicate significant effect of treatment ($p<0.05$) and smaller case letters show differences between treatments.*

Discussion

Deposition and accumulation of sulphur in terrestrial ecosystems around geothermal power plants is a growing concern, because of the potential phyto-toxicity of sulphur at high levels. However, in the present study we found no clear evidence of accumulation of sulphur in response to experimentally increased wet H_2S deposition in the plants that dominate around geothermal plants, i.e. the shrub *T. camphoratus* in Kenya, and the moss *R. lanuginosum* in Iceland. However, the plants responded to the wet H_2S experimental exposure in terms of growth. According to our expectations, growth of *R. lanuginosum* decreased in response to high exposure levels. In the case of the shrub, there was an increase in stem height growth at intermediate concentrations of H_2S (30 µg/L).

Accumulation of sulphur in plants exposed to high concentrations of H_2S has been reported from field studies (Mutia et al., 2016a, b). In the present study, sulphur concentrations in the seedlings of *T. camphoratus* were about 70% higher than those measured in mature leaves of *T. camphoratus* in the field (Mutia et al., 2016a). This is not surprising; as mature leaves are poor sulphur sinks compared to young expanding leaves. Mature leaves preferentially redistribute sulphate to young expanding leaves (Rennenberg et al., 1979; Bell et al., 1995; Hartmann et al., 2000), roots (Sunarpi and Anderson, 1996) and generative sinks (seeds) for nutrition and growth. Such redistribution of sulphur was indeed indicated in our study by the reduced suphur concentrations in the senescing and dead leaves. In contrast, *R. lanuginosum* showed sulphur values 35% higher in field samples (Mutia et al., 2016b) than in the present experiment. This may be partly explained by the duration of the experiment, with a shorter time exposure to H_2S compared to the continued exposure of mosses growing around power plants, and the environmental conditions during the experiment.

In general, there were no strong treatment effects on the responses of either plant, which was surprising, especially for the moss due to its susceptibility to air pollutants. Since mosses are slow growing, possibly a longer duration of the experiment would have shown more clearly the effects of excess sulphur in their tissues. For example, in a field experiment applying 1.0 mM (8.10×10^4 µg/L) bisulphite and 5.0 mM (4.8×10^5 µg/L) sulphate, marked sulphur accumulation and reduced shoot length increase in *Sphagnum* species which were only evident after 18 months of treatment applications (Ferguson and Lee, 1980), so a period of over a year might be recommended in future studies. Still, even in the relatively short duration of our experiment (3 months) we already detected reduced shoot length increase at high H_2S exposures. Exposure to high concentration (100 µg/L and 300 µg/L) of H_2S treatment corresponded to increasing sulphur concentrations in *R. lanuginosum* (marginally significant) that matched slow shoot growth at high treatment exposure levels (300 µg/L). This yields some indication that the high sulphur concentrations may have had a negative influence on the shoot growth, in agreement with other studies (Ferguson et al., 1978; Ferguson and Lee, 1980).

In the case of *T. camphoratus,* the 30 µg/L treatment seemed to have a positive effect (fertilising effect), through stimulated shoot height. This is comparable to the findings of a H_2S fumigation experiment in Thompson and Kats, (1978), where 30 ppb significantly stimulated the growth of lettuce, sugar beets and alfalfa.

Overall, healthy leaves of *T. camphoratus* showed higher sulphur levels than the moss shoots in response to the treatments. This was opposite to what we anticipated, since mosses are more susceptible to atmospheric deposition (either as wet or dry deposition) of pollutants, so we expected the moss to accumulate more sulphur than the shrub. In the absence of pollution, this difference can be explained by the different plant mechanisms for nutrient absorption, where *T. camphoratus* acquires more nutrients (sulphur) from both the soil through roots and leaves than *R. lanuginosum*, which obtains all its nutrients (sulphur) from the air.

Other environmental factors, like the high light intensity and temperature conditions for *R. lanuginosum* during the experiment and the rain events for *T. camphoratus,* may help explain the weak responses we found. Strong light intensities cause photo-inhibition (Murata et al., 2007) and can destroy chlorophyll and DNA structures of bryophytes in moist condition (Glime, 2007). The concentrations of chlorophyll in our *R. lanuginosum* samples were similar to values measured in field (Mutia et al. 2016b), so we cannot unequivocally infer chlorophyll damage. However, high light intensity may have affected some other physiological processes within the mosses under these conditions. Similarly, the effect of temperatures higher than optimal (due to heating from the bulbs) in the experimental growth chambers for *R. lanuginosum* could also have affected the moss responses to the treatment. To overcome these experimental limitations, we recommend that similar experiments are performed in better controlled environments or outdoors and away from geothermal activity or atmospheric pollution during their natural growth period, so that the moss grows in as close to natural conditions as possible. However, field experiments have other limitations. For example, in our outdoors experiment with shrub seedlings, the three rain events could have affected sulphur levels and other responses in the plant leaves through dilution and nutrient leaching. The measured soil variables, which improved the models, also suggest the influence of other environmental factors on the shrub growth and responses to the treatments.

We can therefore imply that short-term exposure to moderate levels of wet H_2S deposition (30 µg/L) does not harm the dominating plants around power plants in Kenya and Iceland. These levels of H_2S seemed to benefit growth in the case of the shrub, and did not reduce moss growth. However, caution needs to be taken with this interpretation and experiments assessing the long term effects of exposure should be conducted. In the case of *R. lanuginosum*, due to the high variability in the responses, experiments with larger numbers of replicates are required. Additional physiological plant responses such as photosynthetic rate (Maas et al., 1988) and changes in leaf area and dry matter for *T. camphoratus* (Maas et al., 1985; Bussotti et al., 2003) are advised. Fumigation experiments of the same H_2S concentrations are also encouraged on the plant species for comparison, especially to assess the threshold levels at which damages may be emergent.

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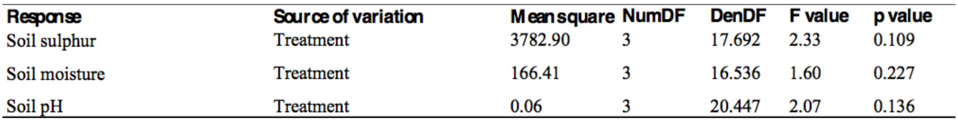
Supplementary Material

Table S1: Analysis of variance of various soil co-variables subjected to H_2S treatments.



Response	Source of variation	Mean square	NumDF	DenDF	F value	p value
Soil sulphur	Treatment	3782.90	3	17.692	2.33	0.109
Soil moisture	Treatment	166.41	3	16.536	1.60	0.227
Soil pH	Treatment	0.06	3	20.447	2.07	0.136