

ISOTOPIC ANALYSIS OF SULPHUR IN THE ASSESSMENT OF SO₂ EMISSION SOURCES

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Abstract

Reported herein is the isotopic composition of sulphur in Norway spruce needles, lichens and soils from two well-defined climatic regions in Slovenia: The Šalek Valley and the Zasavje region. The samples are from distinct locations in these regions, where the impact of SO₂ on vegetation is well documented. In both regions, the main source of SO₂ pollution is from thermal power stations. The isotopic measurements of Norway spruce needles were complemented with δ³⁴S-analyses of coals (lignite and brown coal) and SO₂ in stack-gases from two thermal power plants, to correlate SO₂ pollution with a specific emission source. Reference samples of SO₂ from an unpolluted region (The Triglav National Park) were collected for comparison. To elucidate the turn over of SO₂ from source to spruce needles *via* air and/or soil, characteristic soils and lichens (*Hypogymnia physodes*) were sampled. The sulphur in the samples was converted by combustion in a small oxygen bomb (Parr Instrument) to sulphate, precipitated as BaSO₄ and the δ³⁴S values measured using an isotope ratio mass spectrometer coupled on-line to an elemental analyser. The δ³⁴S values demonstrate only a close relationship between environmental SO₂ and lichens. Correlation between a particular emission source and the apparent impact on vegetation from the δ³⁴S values is not straightforward.

Introduction

Although, sulphur is an important macro-element in the forest ecosystem, being an essential element for plant and soil microorganisms, there is now convincing evidence that forest decline is mainly caused by SO₂ pollution, O₃ and related compounds. Sulphur originates from both natural and anthropogenic sources. Of the latter, eighty-five *per cent* of the sulphur emitted - mainly as SO₂ - derives from the combustion of coal and oil, while the majority of the remaining fifteen *per cent* is from industrial sources.¹ The two main sources of sulphur in the forest ecosystem are from the atmosphere and the soil. Among the gaseous inputs of sulphur, SO₂ contributes the major part.²

Stable sulphur isotopes are widely used to study sulphur dynamics in different ecosystems.³ Naturally occurring Sulphur has four stable isotopes with masses 32, 33, 34 and 36 of which ³²S and ³⁴S are the most abundant (³²S represents 95.02 %, and ³⁴S 4.21 %). The input of sulphur into the ecosystem can cause significant changes in the S-isotopic composition of the pedo-, hydro- and atmosphere as a result of the complex mechanisms: physical, chemical and biological processes, that affect sulphur turnover during its pathway from source to its uptake. This phenomenon is known as isotopic fractionation and albeit small, these variations can be characteristic of a particular emission.⁴ Because it is easier to compare isotopic ratios in two samples than it is to measure the absolute abundances in any one sample, the expression $\delta^{34}\text{S}$ (in ‰), which is the ratio of ³⁴S/³²S relative to an international standard (the Canyon Diablo meteorite), is used to study sulphur isotope variations in nature. For many reasons, the International Atomic Energy Agency (Vienna) introduced and distributed an Ag₂S standard with a defined $\delta^{34}\text{S}$ value of -0.30 ‰. At any specific site, the differences in $\delta^{34}\text{S}$ can be measured with a precision below 0.5 per mil.

In the present work we aim to investigate the relationships between $\delta^{34}\text{S}$ values, SO₂ levels and forest decline in two polluted regions in Slovenia: the Šalek Valley and Zasavje region. In both regions, the dominant sources of SO₂ pollution are the Thermal Power Plants at Šoštanj and Trbovlje, respectively. To do this we measured the isotopic composition of sulphur in lignite, brown coal, chimney-stack gases, spruce needles (current year needles, 1-year-old-needles), soils (four layers), air (Pokljuka plateau). We then compared these values to the isotopic composition of samples collected from the Triglav National Park, which was chosen as an example of an unpolluted region. Because the variations in chemical composition and tissue alterations make lichens sensitive biological indicators of regional trends in ambient SO₂ pollution and because arboreal lichens are never in contact with soil, they are useful for assessing atmospheric SO₂ impact. For this reason we collected specimens of the lichen *Hypogymnia physodes* (L.) Nyl., and exposed them to the environments under study. Finally, since the production of glutathione (GSH) in Norway spruce needles is related to environmental

concentrations of SO₂,⁵ we decided to isolate GSH from Norway spruce needles of the same origin and measure the δ³⁴S values on a molecular level.

Experimental

Sampling and sample preparation

Soil samples: We collected soil samples in September 1996. We first separated the soil profile into two organic layers (O_{lf}, O_h) and two mineral layers (A_{0-10 cm}, A_{10-20 cm}). Each layer was dried at 30 °C and sieved (3 mm). The sample was then mixed with an Eschka mixture in a ratio 1:3 and ignited at 800 °C to convert the sulphur to sulphate,⁶ which was then dissolved in hot deionised water.

Spruce needles: Samples were harvested from Norway spruce trees (*Picea abies* (L.) karsten) in September 1996 from all three sites. Twigs were cut from the light crown of five trees at each location. The needles were frozen in liquid nitrogen, lyophilised and ground to a powder. The sulphur was converted by combustion in a small Parr 1108 oxygen bomb (Parr Instrument) to sulphate following the procedure described in the literature.⁷

Coal: Samples of lignite and brown coal were collected from the Thermal Power Plants at Šoštanj and Trbovlje, respectively. The sulphur, like that for spruce needles, was converted by combustion to sulphate.

Chimney-stack-gases: Samples from both Thermal Power Plants were obtained by trapping the SO₂ in a hydrogen peroxide solution.

Air samples: The sampling site for air at the Triglav National Park was the Pokljuka plateau. In this instance SO_x in the ambient air was trapped on alkaline impregnated glass fibre filters.⁸

Lichens: We collected specimens of the Lichen *Hypogymnia physodes* (L.) Nyl., from the reference sampling sites for SO₂ monitoring (Spodnja Komisija, Table 1) and exposed them for six months to the environments at the given sampling sites. We then followed the same sample preparation procedure as for the spruce needles.

S-isotopic analysis

Isotopic analysis involved precipitating the sulphur (sulphate) in the samples, as BaSO₄, which was then burned in an elemental analyser to SO₂, separated using a packed gas column and the ³⁴S and ³²S concentration determined using a Finnigan isotope ratio mass spectrometer.⁹ From these values, we calculated δ³⁴S (relative to the Canon Diablo meteorite international standard) using the expression:

$$\delta^{34}\text{S} = \frac{(\delta^{34}\text{S} / \delta^{32}\text{S})_{\text{sample}} - (\delta^{34}\text{S} / \delta^{32}\text{S})_{\text{standard}}}{(\delta^{34}\text{S} / \delta^{32}\text{S})_{\text{standard}}} * 10^3 (\text{‰})$$

To isolate glutathione (GSH), 150 g of lyophilised spruce needles were dissolved in 500 ml of distilled water. The solution was injected onto a preparative HPLC column (Spherisorb ODS2, 5µm, 150 x 20 mm) and separated using a gradient elution of 0.1 % TFA in H₂O and 0.1 % TFA in H₂O: MeCN (1: 1). Detection was performed with an UV-Diode array detector and a Finnigan TSQ mass spectrometer under ESI conditions.

Results

Figures 1 - 3 show the average δ³⁴S values in the samples from the three sampling sites. The δ³⁴S for lichens are given in Table 1. The standard deviation (reproducibility) is ± 0.3. ‰

Sampling site	Area	δ ³⁴ S (‰)
Zavodnje	Affected by TPP Šoštanj	7.2
Veliki vrh	Affected by TPP Šoštanj	7.7
	Affected by TPP Šoštanj	7.0
Podvolovjek	1	5.6
Učeja	2	3.2
Spodnja Komisija	3	5.6

Table 1: Mean δ³⁴S values in the lichen *Hypogymnia physodes* (L.) Nyl., exposed for 6 months in different environments (TTP = Thermal Power Plant)

- 1 ... Reference site on the border of the Šalek Valley
- 2 ... Reference site near the Triglav National Park
- 3 ... Lichens from an unpolluted area (Spodnja Komisija) transferred to the given sites for a period of 6 months

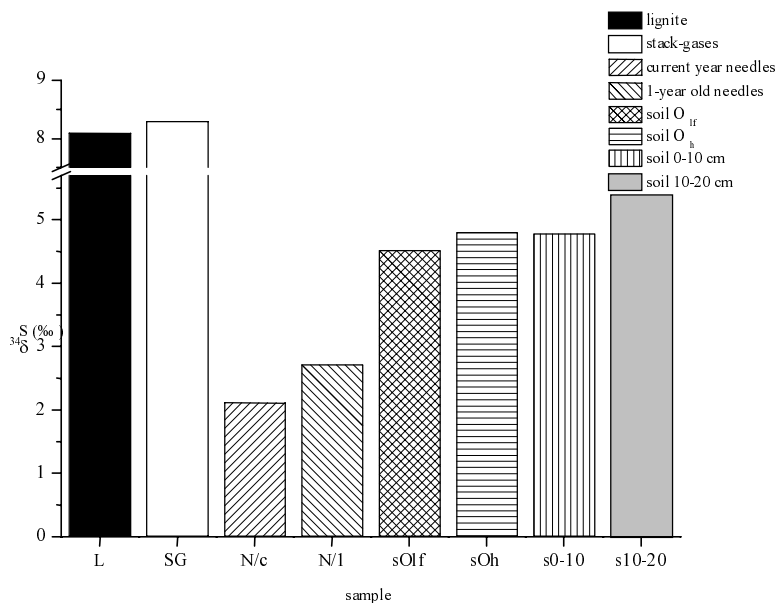


Fig. 1: Mean $\delta^{34}\text{S}$ values in the area under the influence of the Šoštanj Power Plant (1996)

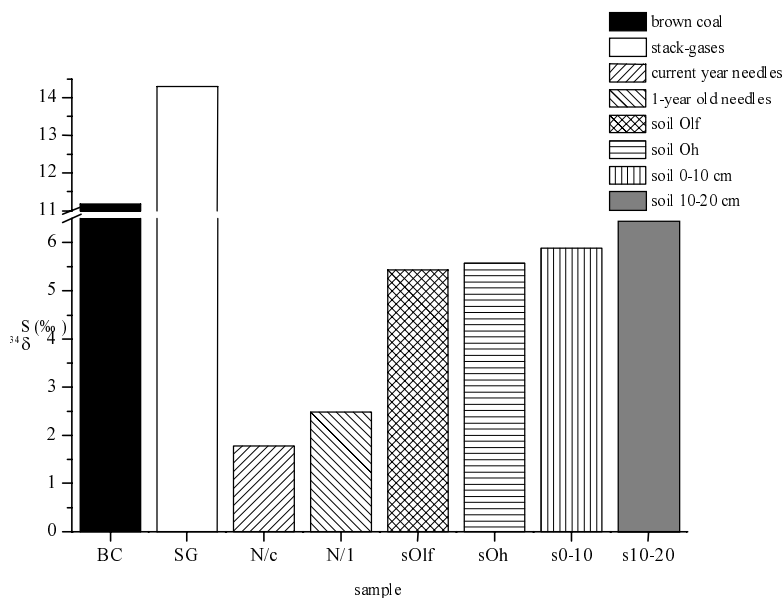


Fig. 2: Mean $\delta^{34}\text{S}$ values in the area under the influence of the Trbovlje Power Plant (1996)

From the results (Figs. 1 and 2) the isotopic fractionation of sulphur during the burning of lignite in the Šoštanj Power Plant falls within the measured uncertainty, while the isotopic fractionation from burning of brown coal in the Trbovlje Power Plant leads to an enrichment of ^{34}S . Even after repeated measurements, the same effect was observable, which means that this difference cannot be explained in terms of reaction kinetics. In both polluted areas, the $\delta^{34}\text{S}$ value in Norway spruce needles increases with age, unlike in the needles from the Triglav National Park where there is no increase (Fig. 3). The $\delta^{34}\text{S}$ value in the soil at all three sites increases with depth but is more pronounced in the polluted areas. In the Norway spruce needles the $\delta^{34}\text{S}$ value was less positive than in the ambient SO_2 and in the soil in the two polluted areas, whereas, in the Triglav National Park the $\delta^{34}\text{S}$ in spruce needles is similar to that in the soil. The $\delta^{34}\text{S}$ values in the lichen *Hypogymnia physodes* (L.) Nyl., exhibits an assumed cause-and-effect relationship, i.e., the $\delta^{34}\text{S}$ values $>7\text{‰}$ in the specimens exposed to ambient SO_2 originating from the Šoštanj Power Plant ($\delta^{34}\text{S} \approx 8.2\text{‰}$) indicate a direct impact from the emission source.

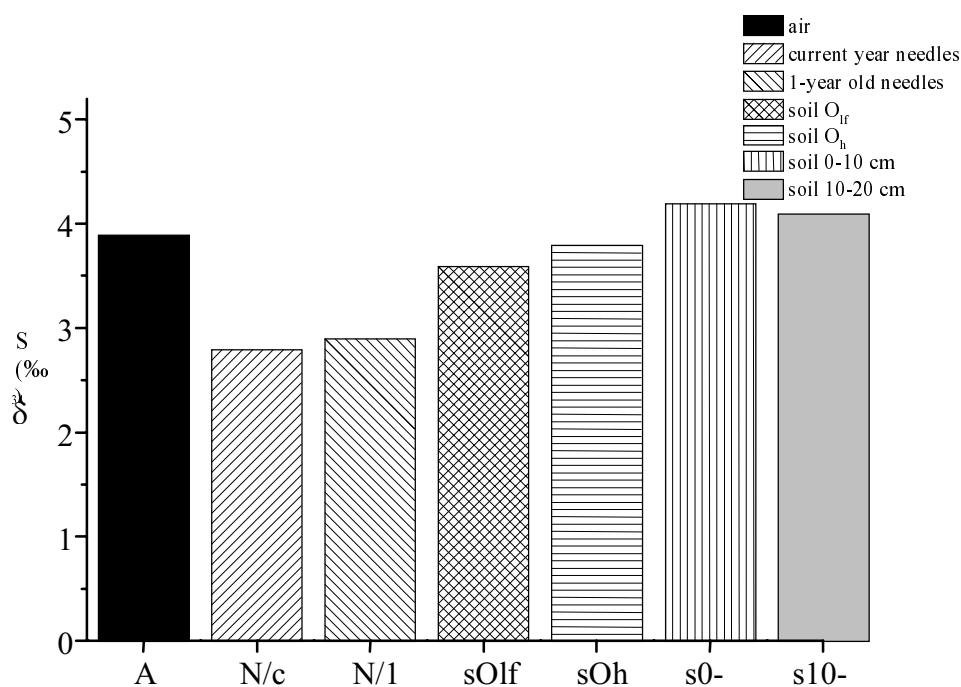


Fig. 3: Mean $\delta^{34}\text{S}$ values in the Triglav National Park (1996)

The content of GSH in the Norway spruce needles is much lower than that stated in the literature, which for a selected sample is about 1 µg/ml. This means that to isolate 500 µg of GSH (50 µg S) for isotopic analysis using preparative HPLC, approximately 150 h working time and more than 70 litres of MeCN is required.

Discussion

In the presence of higher SO₂ concentrations in air, for example in the areas affected by the Šoštanj and Trbovlje Power Plants, plants are expected to use both atmospheric and soil sulphur for their metabolism. An assumed cause and effect relationship between the average SO₂ levels and its isotopic composition should be reflected in alterations of S-containing compound in plants and their isotopic composition. Nevertheless, spruce needles show less positive δ³⁴S values than both potential sources. This might be due to S-isotope fractionation during S-uptake and/or metabolism, leading to compounds enriched in ³²S. Unlike lichens whose δ³⁴S values reflect those of ambient SO₂ in the air, spruce needles exhibit values lower than air and soil at the same site, indicating the mixing of sulphur from two distinct sources (bedrock and atmosphere) and/or pronounced isotope fractionation in soil. As expected, the δ³⁴S in the lichens exposed to different environmental conditions show a higher correlation between environmental SO₂ and a particular emission source (Power Plant at Šoštanj).

All sites exhibit a small decrease in δ³⁴S from topsoil to mineral soil. This can be explained by the fact that in nonsaline upper soil horizons, more than 95% of sulphur is in organic form.¹⁰ Even at extremely high SO₂ deposition, sulphur appears to be cycled through an organic pool in the soil. In addition, unless the mean residence time of sulphur in the soil is greater than the time scale of the SO₂ pollution, the positive δ³⁴S in soil may reflect natural sulphur.

Conclusions

Establishing a cause and effect relationship between a particular SO₂ emission source and the apparent impact on vegetation from the δ³⁴S value is not straightforward. The accumulation of sulphur in vegetation is complex and we can find no direct

correlation between the $\delta^{34}\text{S}$ value in spruce needles and stack-gases, not even in the surrounding of a given emission source. Despite this, the differences in $\delta^{34}\text{S}$ values between chimney-stack SO_2 and sulphur in spruce needles and in soils is significant for both regions. The $\delta^{34}\text{S}$ values demonstrate only a close relationship between environmental SO_2 and lichens (uptake of SO_2 from air).

In this instance, the assumption that determining $\delta^{34}\text{S}$ values on a molecular level in spruce needles reflect the isotopic composition of the surrounding SO_2 ¹¹ can not be proved by measuring the $\delta^{34}\text{S}$ of GSH. The low concentration of GSH in spruce needles makes the isolation of this compound for $\delta^{34}\text{S}$ measurements impractical. Generally, future advances in applying stable isotope techniques on a molecular level will largely depend on the ability of instrumentation to measure microgram amounts of organic sulphur.

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Povzetek

V vzorcih premogov, dimnih plinov, smrekovih iglic in tal iz dveh onesnaženih področjih (Šaleška dolina in Zasavje) smo merili izotopsko sestavo žvepla ($\delta^{34}\text{S}$). Za primerjavo smo merili vzorce zraka in tal iz področja Triglavskega narodnega parka. Da bi pojasnili prenos SO_2 v iglice smreke preko zraka in/ali tal, smo merili $\delta^{34}\text{S}$ vrednosti v značilnih drevesnih lišajih in tleh. Žveplo v trdnih vzorcih smo s sežigom v kisiku, iz zraka pa neposredno pretvorili v BaSO_4 in nato v SO_2 ter vrednost $\delta^{34}\text{S}$ izmerili z masnim spektrometrom. Medtem ko je pri lišajih opazna korelacija med $\delta^{34}\text{S}$ vrednostmi za okoliški SO_2 pa je vpliv na $\delta^{34}\text{S}$ vrednosti v iglicah smreke manj izrazit.