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Primary and secondary organics in the tropical Amazonian rainforest aerosols: chiral analysis of 2-methyltetraols

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This work presents the application of a new method to facilitate the distinction between biologically produced (primary) and atmospherically produced (secondary) organic compounds in ambient aerosols based on their chirality. The compounds chosen for this analysis were the stereomers of 2-methyltetraols, (2*R*,3*S*)- and (2*S*,3*R*)-methylerythritol, (L- and D-form, respectively), and (2*S*,3*S*)- and (2*R*,3*R*)-methylthreitol (L- and D-form), shown previously to display some enantiomeric excesses in atmospheric aerosols, thus to have at least a partial biological origin. In this work PM₁₀ aerosol fractions were collected in a remote tropical rainforest environment near Manaus, Brazil, between June 2008 and June 2009 and analysed. Both 2-methylerythritol and 2-methylthreitol displayed a net excess of one enantiomer (either the L- or the D-form) in 60 to 72% of these samples. These net enantiomeric excesses corresponded to compounds entirely biological but accounted for only about 5% of the total 2-methyltetrol mass in all the samples. Further analysis showed that, in addition, a large mass of the racemic fractions (equal mixtures of D- and L-forms) was also biological. Estimating the contribution of secondary reactions from the isomeric ratios measured in the samples (=ratios 2-methylthreitol over 2-methylerythritol), the mass fraction of secondary methyltetrols in these samples was estimated to a maximum of 31% and their primary fraction to a minimum of 69%. Such large primary fractions could have been expected in PM₁₀ aerosols, largely influenced by biological emissions, and would now need to be investigated in finer aerosols. This work demonstrates the effectiveness of chiral and isomeric analyses as the first direct tool to assess the primary and secondary fractions of organic aerosols.

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Environmental impact

Chiral speciation is commonly used in geochemistry to determine if organic compounds in the environment are biologically produced or biologically processed, in which case they display an excess of one enantiomer (*i.e.* of the “right” or “left” form). We recently developed such a method for compounds present in atmospheric aerosols and applied it to the 2-methyltetrols, previously considered to be exclusively produced by the abiotic oxidation of isoprene. In this work we show that the 2-methyltetrols present in PM₁₀ aerosols above the Amazonian forest are quantitatively biological and therefore that the oxidation of isoprene contributes less to the aerosol mass in that region than previously inferred from these compounds.

Introduction

Secondary organic aerosols (SOA), which are produced directly in the atmosphere by abiotic (=non-biological) reactions, are estimated to play important roles in atmospheric chemistry.⁴ Yet most of the information currently available on them is indirect, as it is based either on strong assumptions in atmospheric observations (for instance with Aerosol Mass Spectrometers),¹ smog chamber results, or on differences with the primary organic aerosols that are emitted by non-atmospheric sources (biosphere, soils, combustion processes,...). These assumptions have contributed to the large underestimations of the SOA mass in current models.²⁻⁶ There is thus a strong need to develop methods to distinguish the secondary from the

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primary material directly in ambient aerosols, thereby avoiding assumptions on their sources and formation mechanisms. A method offering these advantages was recently developed and is based on separating the different enantiomers (*i.e.* “right” and “left” forms) of chiral compounds present in aerosols.^{7,8} Its rationale is based on the fundamental chiral principle that only biological processes or chiral media (for instance, chiral catalysts) can produce a net excess of one chiral form over another while abiotic reactions involving non-chiral components produce exclusively racemic mixtures (*i.e.* 50% of both forms), regardless of their mechanisms and conditions.^{9,10} A net chiral excess in environmental samples thus identifies with certainty a biologically produced or biologically processed compound. Racemic mixtures, however, can result either from abiotic reactions or biological processes as living organisms can produce such mixtures¹¹ or different species can emit different enantiomers with equal strength. This fundamental chiral principle bears no exception, and its robustness holds the key for the development of chirality and life on Earth.^{12,13} Chiral analysis has thus been used in environmental sciences for decades, for instance to investigate the biological degradation of anthropogenic compounds^{14,15} or to find patterns in gas phase biogenic emissions.^{16,17}

Our chiral analysis focuses on the 2-methyltetraols because these compounds have been identified in the past as tracers for the SOA produced abiotically by the atmospheric oxidation of isoprene,¹⁸ the most abundant biogenic volatile organic compound (BVOC) in the atmosphere^{19–21} and a major SOA precursor globally. Since their first identification in Amazonian aerosols,¹⁸ these compounds have been found in aerosols from many regions of North and South America^{22–25} and Europe.^{26–29}

The existence of abiotic “secondary” formation pathways for these compounds has been established by smog chamber and laboratory experiments, although their exact mechanisms are still unclear.^{26,30–33} But the fundamental chiral principle implies that, regardless of their mechanisms and conditions, these secondary reactions produce exclusively racemic mixtures. This was recently confirmed by obtaining racemic mixtures of the 2-methyltetraols both from the gas-phase oxidation of isoprene in the smog chamber and from its liquid-phase oxidation in the laboratory.⁸

On the other hand, 2-methyl-D-erythritol (2*S*,3*R*) is a known metabolite of the Methyl Erythritol Phosphate (MEP) biosynthesis pathway,^{34–36} which is employed by a very large number of living species.³⁷ This compound was thus isolated in numerous plants, such as *Convolvulus glomeratus*,^{38,39} *Liriodendron tulipifera*,⁴⁰ *Ferula sinaica*,⁴¹ and *Phlox subulata*,⁴² and in bacteria such as *Corynebacterium ammoniagenes*.³⁴ More recently, glycosides of 2-methyl-L-erythritol (2*R*,3*S*) were isolated in *Gardenia jasminoides*, suggesting that L-forms might also be involved in biosynthetic pathways.⁴³ In addition, the oxidation of isoprene was shown to be biologically triggered within leaves⁴⁴ thus also potentially producing non-racemic 2-methyltetraols. The 2-methyltetraols present in atmospheric aerosols can therefore have either abiotic (secondary) or biological (primary) origins, or a combination of both.

The first application of the new chiral method showed the presence of non-racemic 2-methyltetraols in aerosols from Aspvreten, Sweden, indicating at least a partial biological origin.^{7,8} However, the primary mass fractions estimated for these compounds were highly variable from one sample to another. In order to determine the importance of their secondary and primary sources in different regions of the atmosphere and further explore the information obtained from chiral speciation, this method was applied in this work to PM₁₀ aerosol fractions collected in Central Amazonia. The enantiomeric distribution for the 2-methyltetraols was investigated in the wet and dry seasons, and their primary and secondary fractions estimated with the help of their absolute concentrations, isomeric fractions, modelled isoprene emissions and backward air mass trajectories.

Methods

Aerosol sampling

Aerosol samples were collected between June 2008 and May 2009. The sampling site is located in Central Amazonia, at the INPA (Brazilian National Institute for Research in Amazonia) ecological reserve of “Reserva Ecológica do Cuieiras”, also called ZF2 Ecological Reservation. The measurements took place at the TT34 tower (2°35.66' S, 60°12.56' W, 110 m a.s.l.). The site is located about 60 km north of Manaus and rarely affected by air pollution plumes from the city or any other anthropogenic sources. The forest canopy height near the tower varied between 30 and 35 m. Electrical power is provided by a 60 kW diesel generator located 0.72 km downwind of the TT34 tower. The generator was separated from TT34 by a 50 m deep valley. Fast response, on-line instruments (a CPC, a CO monitor and a nephelometer) indicate negligible contamination from the generator on the filters collected.⁴⁵

Samples were collected using a stacked filter unit equipped with a PM₁₀ inlet. Details of the stacked filter unit have been published elsewhere.⁴⁶ Nine filters collected during the dry season (Jun–Aug 2008), and nine during the wet season (Feb–Mar 2009) were analyzed. The average sampling time was 72 h at a flow rate of 18 L min⁻¹. Aerosols were collected on quartz filters (SKC, 47 mm, 1.2 μm) using Nuclepore polycarbonate filter holders. The filters were prebaked at 500 °C for 12 h to remove any possible organic material that could contaminate the samples. After collection, the filters were placed in Petri dishes and stored in a freezer until analysis. Four field blanks were collected during the measurement period. These were obtained in the same way as aerosol samples with the exception of activating the air pump.

Materials

Authentic racemic mixtures of 2-methylerythritol and 2-methylthreitol were synthesized by Innochemie GmbH (Germany) with a purity >95%. These standards were used to calibrate the instrument and quantify the concentrations of these compounds in the ambient aerosol samples. *Meso*-erythritol (purity >99%) was used as a recovery internal standard and was

purchased from Sigma Aldrich. The elution order and retention times of each enantiomer were determined by using authentic enantiomerically pure standards, synthesized according to the method of ref. 47. All other chemicals were purchased from Fischer Scientific unless specified. These standards, as well as commercial standards of L- and D-threitol and erythritol, were stored for the same duration or longer than the aerosol samples and at higher temperature without displaying any change in their enantiomeric or isomeric composition. This indicated that storing the aerosol samples would not affect their enantiomeric or isomeric composition. This is because transforming one enantiomer or one isomer into another requires swapping the position of different chemical groups, thus breaking some bonds and forming new ones, which can only occur in actual chemical reactions.

Aerosol analysis

All the samples and standards were analysed following the method described in ref. 7, which included an extraction step, a derivatisation, and the separation of the enantiomers on two different GC columns: 2-methylerythritol on a Varian Chirasil Dex and 2-methylthreitol on a FS Lipodex E column. This implied two separate sets of analysis for each isomer. The full area of the filter was extracted in 50 : 50 v/v solution of dichloromethane in methanol (Sigma Aldrich). The derivatisation followed exactly the procedure described in ref. 7. The instrument used for the analysis was a Varian 3400 GC coupled to a Finnigan SSQ 7000 MS, operated in electron ionization (70 eV) and single ion monitoring modes. The GC set up was slightly modified compared to ref. 7 by setting the carrier gas flow to 102.7 kPa, the injector temperature to 175 °C and the auxiliary temperature to 185 °C. All the other set-ups and temperature program for the Varian Chirasil Dex column were the same as in ref. 7 with the exception of the final oven temperature hold time, which was 10 min. The total analysis time with this column was 97 min. With the FS Lipodex E column two changes were made compared to ref. 7: the column used was 50 m, 0.25 mm ID and the temperature program was consequently changed. The initial oven temperature was 60 °C for 2.5 min. The first ramp was 20 °C min⁻¹ up to 100 °C held for 20 min. The second ramp was 5 °C min⁻¹ up to 160 °C held for 25 min. The third ramp was 20 °C min⁻¹ up to 180 °C held for 5 min. Altogether, the analysis time on this column was 117 min.

Blank filters were extracted and analysed by exactly the same procedures as the regular samples and used to verify the absence of handling or analysis artefacts that could have affected the results.

Determination of the enantiomeric fractions, concentrations and uncertainties

The separation of the enantiomers of each pair and their quantification by chiral GC-MS analyses provided two independent types of information: the enantiomeric (or chiral) information, defining the spatial structure of each enantiomer relative to a single carbon center, and the isomeric information defining the relative abundance of the two isomers

(2-methylerythritol and 2-methylthreitol) relative to two carbon centers. It will be shown in this work that combining these two quantities provides valuable information.

The enantiomeric fraction, E_f , measuring the relative abundance of the enantiomers in a pair, was determined as the ratio of the peak area for one enantiomer in the chromatogram over the sum of the peak areas for both enantiomers. Thus for instance for (2*R*,3*S*)-methylerythritol:

$$E_f = A_{2R,3S} / [A_{2R,3S} + A_{2S,3R}], \quad (1)$$

where " $A_{X,Y}$ " refers to the peak area of the corresponding enantiomer. As discussed below, it was first verified that the detection sensitivities of both enantiomers were identical. E_f thus varies between 0 and 1, 0.5 corresponding to racemic mixtures and 0 or 1 to a pure enantiomer. Eqn (1) also implies that if one enantiomer has a fraction E_f , the other has a fraction $1 - E_f$.

Any significant deviation of E_f from the racemic value in the samples thus identified with certainty the presence of compounds of biological origin. Determining the uncertainties on E_f was therefore crucial. Those were determined as the 99% confidence interval obtained when repeating the analysis of a reference racemic mixture.⁷ For 2-methylerythritols 20 such analyses gave an average value for racemic mixtures of $E_f = 0.50 \pm 0.006$ (relative standard deviation, 1.2%) and for 2-methylthreitols 9 analyses gave 0.5 ± 0.008 (relative standard deviation, 1.7%). The relative uncertainties on E_f were thus about 2%. Uncertainties on chiral quantities are always much smaller than those on absolute quantities such as concentrations because operating steps (collection, extraction, derivatization,...) affect both enantiomers identically and not their relative abundances.¹⁵ The same series of tests gave as 99% confidence intervals for racemic E_f of 0.49–0.50 and 0.50–0.51 for (2*R*,3*S*) and (2*S*,3*R*)-methylerythritol and 0.50–0.51 and 0.49–0.50 for (2*S*,3*S*) and (2*R*,3*R*)-methylthreitol, respectively. The compounds found in the samples and having E_f outside these intervals, thus non-racemic, were identified as being at least partly biological. Since, as explained above, operating steps such as sampling and extraction did not affect the enantiomeric ratios, the uncertainties on the E_f values obtained for the samples were identical to those established with the reference compounds, thus about 2%.

Because 2-methylerythritol and 2-methylthreitol were separated on different columns, their comparison and the investigation of potential correlations between them required determining their absolute concentrations. This was done by comparing the peak areas for each enantiomer in the chromatograms with calibration curves established with known solutions of the racemic standards. The atmospheric concentrations, in ng m⁻³, were then obtained by dividing the concentrations in the extracts by the volume of air collected for each sample.

In addition to E_f and the absolute concentrations, the isomeric ratio, IR, quantifying the relative abundance of the isomers, 2-methylthreitol and 2-methylerythritol, in racemic mixtures was also determined for each sample as:

$$IR = [2\text{-methylthreitol}]_{\text{racemic}}/[2\text{-methylerythritol}]_{\text{racemic}} \quad (2)$$

where the quantities between brackets are concentrations (in ng m^{-3}). The uncertainties on IR were estimated as $\pm 20\%$, resulting from the uncertainties on the concentrations of each isomer.

MEGAN estimates

To help determine the fraction of 2-methyltetraols resulting from the oxidation of isoprene, thus SOA, potential correlations between isoprene and the concentrations of 2-methyltetraols were investigated. But as measurements of isoprene or other relevant compounds were not available for this site isoprene emissions were estimated using the Model of Emissions of Gases and Aerosols from Nature, MEGAN version 2.1.⁴⁸ This is a flexible framework that estimates biogenic VOC emissions as a function of emission factors and variations driven by changes in land cover and environmental conditions. The emission factors ($\mu\text{g compound m}^{-2}$ ground area h^{-1}) for these estimates were based on direct eddy covariance observations of above canopy fluxes⁴⁹ at a nearby site and include the value of 4500 for isoprene. Thirty minute average BVOC emissions estimated with MEGAN were driven by changes in land cover and environmental conditions. Land cover inputs consisted of monthly average Leaf Area Index (LAI) based on MODIS satellite observations. LAI variations were used to characterize changes in total foliage and leaf age according to procedures described in ref. 19. The model was constrained for the specific conditions of the sampling site. Above canopy temperature, photosynthetically active radiation, relative humidity, and wind speed were measured with a Campbell meteorological station (located at the top of the K34 tower, close to the TT34 tower within the ZF2 ecological reservation) and used as inputs to the MEGAN canopy environment model. Leaf temperature and solar radiation incident on sun and shade leaves were calculated at five canopy depths and used to drive the short-term and long-term components of ref. 20 algorithms for simulating the emission response to light and temperature. Based on observations,⁴⁹ isoprene emissions were assumed to be dominated by light-dependent emissions.

Air mass back trajectories analysis

Air mass back trajectories were generated using the HYSPLIT model (Hybrid Single Particle Lagrangian Integrated Trajectory).^{50,51} The meteorological dataset used was Global Data Assimilation System, GDAS (2006–present). The back trajectories were set to originate at the latitude and longitude of the measurement site. Each individual trajectory was calculated every 12 hours for every sample collected. The isentropic vertical motion calculation method was used.

Results and discussion

Enantiomer fractions

The enantiomeric fractions, E_f , obtained for 2-methylerythritol and 2-methylthreitol in the samples are presented in Fig. 1.

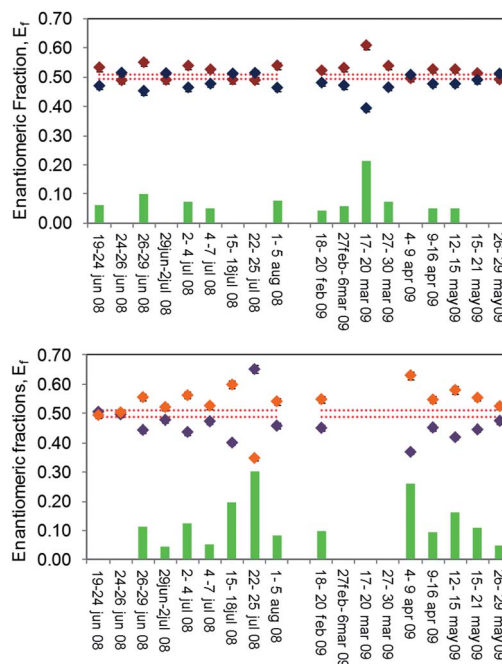


Fig. 1 Enantiomeric fractions, E_f , for 2-methylerythritol (top, (2*R*,3*S*), dark red; (2*S*,3*R*), dark blue) and 2-methylthreitol (bottom, (2*R*,3*R*), orange; (2*S*,3*S*), purple). The dashed lines represent the 99% confidence intervals, thus the uncertainties, on the racemic value for E_f . The green bars are the enantiomeric excesses, ee , for each sample.

Fig. 1 shows that the 2-methyltetraols were outside the confidence interval for racemic mixtures in most samples, 61% of them for 2-methylerythritol and 72% for 2-methylthreitol, evidencing the presence of biologically produced compounds. The average E_f value for 2-methylerythritol was 0.52 ± 0.03 (standard deviation) with little variability between samples and (2*R*,3*S*) (*L*-form) being generally in excess over (2*S*,3*R*) (*D*-form). By contrast (2*S*,3*R*) was generally found in excess over (2*R*,3*S*) in aerosols from Aspöreten, Sweden.¹⁷ The 2-methylthreitol were below the detection limit in three samples of the wet season, Feb. 27 to March 30. In the other samples the average value for E_f was 0.55 ± 0.04 with more variability between samples than with 2-methylerythritol, and (2*R*,3*R*) (*D*-form) being in excess over (2*S*,3*S*) (*L*-form) except in one sample. The wide variability in E_f between the samples of this work and between the Amazonian and boreal forest aerosols suggested compounds of biological origin and resulting from multiple emitters. A similar variability was reported for chiral BVOCs such as (–)- α - and (+)- α -pinene between boreal (Hyttiälä, Finland) and tropical forests (French Guyana, Suriname, and Guyana).⁵² Such a biological origin will be further investigated below by quantifying the primary and secondary mass fractions for these compounds.

Minimum primary fraction

As explained above, the net excess of the most abundant enantiomers in each sample corresponded to compounds that were with certainty entirely biological. This was quantified with the enantiomeric excess, ee , which is defined as the absolute

value of the difference between the mole fractions of both enantiomers in a pair, or the difference between their respective E_f values. For instance, for 2-methylerythritol:

$$ee = |E_{f(2R,3S)} - E_{f(2S,3R)}|. \quad (3)$$

In addition, to account for the 99% confidence interval for racemic fractions, ee values equal to or lower than 0.02 were set to 0. The ee values thus obtained in the samples are shown in Fig. 1. In individual samples, ee was as high as 0.1 for 2-methylerythritol and 0.21 for 2-methylthreitol, confirming the non-racemic character of these compounds. The mass concentrations of these net enantiomeric excesses, obtained from the difference between the mass concentrations of each enantiomer (see the next section), represented the absolute minimum fraction of biological tetraols (Table 1). Table 1 also provides the contributions (in %) of these excesses to the total tetraol mass in each sample. These minimum primary mass fractions accounted for up to 20% of the total 2-methyltetrol mass in individual samples but for only 5.5% of this mass on average for all the samples. However, it will be shown below that the total primary fraction of 2-methyltetrols in these samples was much larger as, in addition to these enantiomeric excesses, some of the racemic compounds were also biological.

Concentrations

The absolute concentrations of the different enantiomers in the samples, in ng m^{-3} , are given in Table 2. Clearly, the dry season aerosols contained much more 2-methyltetrols than wet season ones, representing 91% of their total mass. This was consistent with both larger biogenic emissions and larger emissions of SOA precursors (isoprene) with high temperature and light intensity.⁵³ In the dry season the 2-methylerythritols ranged

Table 2 2-methyltetraols enantiomer concentrations in the samples (in ng m^{-3}) and isomeric ratios, IRs

Sampling period		2-Methylerythritol		2-Methylthreitol		IR
Season	Date	(2R,3S)-	(2S,3R)-	(2S,3S)-	(2R,3R)-	
Dry	19–24 Jun 08	51.48	45.41	2.32	2.29	0.05
	24–26 Jun 08	97.76	102.99	4.51	4.55	0.05
	26–29 Jun 08	166.20	136.69	3.29	3.76	0.02
	29 Jun–2 Jul 08	160.31	167.88	2.72	2.83	0.02
	2–4 Jul 08	80.77	69.68	4.44	5.17	0.06
	4–7 Jul 08	30.77	27.96	5.26	5.69	0.19
	15–18 Jul 08	11.58	12.04	1.52	1.64	0.13
	22–25 Jul 08	13.98	14.66	1.61	1.47	0.11
	1–5 Aug 08	90.81	78.05	1.86	2.00	0.02
	Wet	18–20 Feb 09	14.43	13.40	4.53	4.90
27 Feb–6 Mar 09		1.67	1.53	— ^a	— ^a	
17–20 Mar 09		9.03	6.15	— ^a	— ^a	
27–30 Mar 09		8.84	7.72	— ^a	— ^a	
4–9 Apr 09		7.66	7.83	0.98	1.03	0.13
9–16 Apr 09		4.55	4.15	0.77	0.80	0.19
12–15 May 09		5.88	5.40	2.62	3.02	0.49
15–21 May 09		4.15	3.98	0.93	0.97	0.23
26–29 May 09		7.82	8.11	2.20	2.27	0.28

^a 2-Methylthreitol below the detection limit.

between 23.7 and 168.9 ng m^{-3} and the 2-methylthreitol between 3.1 and 11.0 ng m^{-3} whereas in the wet season they varied between 3.2 and 27.8 ng m^{-3} and 1.0 and 9.4 ng m^{-3} , respectively. The total concentrations for each isomer were thus within the ranges reported in other works.⁴

For the dry season (July) these results are consistent with the total concentrations of 2-methyltetraols reported previously at a nearby site.¹⁷ These absolute concentrations allowed the investigation of potential correlations between the compounds. In the

Table 1 Enantiomeric excess mass in the samples

Date	2-Methylerythritol ng m^{-3}	2-Methylthreitol ng m^{-3}	Total enantiomeric excess mass fraction in sample, %
19–24 Jun 08	6.1 ± 0.1	0.0 ± 0.0	6.3
24–26 Jun 08	5.2 ± 0.1	0.0 ± 0.0	2.6
26–29 Jun 08	29.5 ± 0.6	0.5 ± 0.0	9.9
29 Jun–2 Jul 08	7.6 ± 0.2	0.1 ± 0.0	2.3
2–4 Jul 08	11.1 ± 0.2	0.7 ± 0.0	7.9
4–7 Jul 08	2.8 ± 0.1	0.4 ± 0.0	5.5
15–18 Jul 08	0.5 ± 0.0	0.1 ± 0.0	2.5
22–25 Jul 08	0.7 ± 0.0	0.1 ± 0.0	2.9
1–5 Aug 08	12.8 ± 0.3	0.1 ± 0.0	7.6
18–20 Feb 09	1.0 ± 0.0	0.4 ± 0.0	5.0
27 Feb–6 Mar 09	0.1 ± 0.0	— ^a	4.4
17–20 Mar 09	2.9 ± 0.1	— ^a	19.0
27–30 Mar 09	1.1 ± 0.0	— ^a	6.8
4–9 Apr 09	0.2 ± 0.0	0.1 ± 0.0	1.4
9–16 Apr 09	0.4 ± 0.0	0.0 ± 0.0	4.9
12–15 May 09	0.5 ± 0.0	0.4 ± 0.0	7.8
15–21 May 09	0.2 ± 0.0	0.0 ± 0.0	2.6
26–29 May 09	0.3 ± 0.0	0.1 ± 0.0	2.3
Total samples			5.5

^a 2-Methylthreitol below the detection limit.

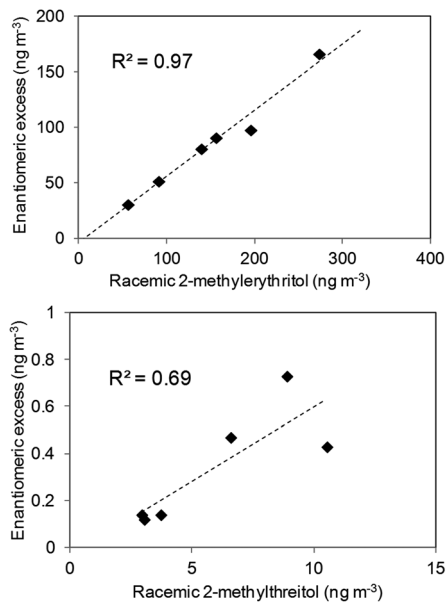


Fig. 2 Correlations between the enantiomeric excesses and racemic fractions of the compounds in the dry season.

dry season, strong correlations were found between the racemic and enantiomeric excess mass fractions of both compounds (when excluding the samples within the racemic interval), $R^2 = 0.97$ for 2-methylerythritol and $R^2 = 0.69$ for 2-methylthreitol, (Fig. 2). As the enantiomeric excesses are exclusively biological, this suggested that at least a large part of the racemic fractions was also of biological origin. In the wet season these correlations were weaker. Conversely, good correlations ($R^2 = 0.71$) were found in the wet season between the racemic fractions of both compounds (Fig. 3), hinting that they had mostly secondary origin. No such correlations were found in the dry season (Fig. 3), consistent with the potential biological origin suggested by the correlations in Fig. 2.

Isomeric ratios and determination of the primary and secondary fractions

The primary and secondary fractions of the racemic 2-methyltetrols were further investigated with the help of the isomeric

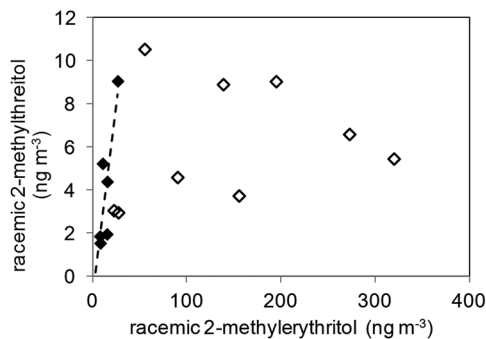


Fig. 3 Correlation between the racemic fractions of 2-methylerythritol and 2-methylthreitol in the wet (black symbols) and dry season (white symbols).

information. Based on the correlations presented above, and to determine the largest possible secondary contribution in the samples, the racemic fractions of the 2-methyltetrols in the wet season were assumed to be entirely secondary.

In the dry season, the maximum possible secondary fraction of 2-methyltetrols was calculated by assuming racemic 2-methylthreitol to be entirely secondary and secondary reactions to produce both isomers in a constant isomeric ratio, IR. Such a constant IR value, independent of the atmospheric conditions, was reported for methyltetrols in atmospheric aerosols from a polluted urban region²⁹ and a tropical forest in China⁵⁴ and presented as the main argument to conclude on their secondary origin.⁵⁴ In this work the secondary fraction of 2-methylerythritol in each sample was thus calculated from the racemic 2-methylthreitol and IR:

$$[2\text{-Methylerythritol}]_{\text{2ary}} = [2\text{-Methylthreitol}]_{\text{racemic}}/\text{IR}, \quad (4)$$

and its primary racemic fraction by the difference to the total racemic 2-methylerythritol. The total secondary fraction of 2-methyltetrols in each sample was thus calculated as the sum of racemic 2-methylthreitol and of secondary 2-methylerythritol given by eqn (4), and the total primary fraction by the sum of the primary racemic fraction of 2-methylerythritol and of the net enantiomeric excesses for both compounds.

One unknown in these calculations was the value of IR resulting from the secondary reactions. The IRs obtained for the samples are shown in Table 2, exhibiting a wide variability. According to the definition of IR (eqn (2)) and to eqn (4), the larger the value of IR assumed for secondary reactions, the smaller the quantities of 2-methylerythritol resulting from these reactions. Thus, to determine the maximum possible secondary fractions in these samples, the smallest value of IR observed in the dry season was used, which was 0.19 (85% of 2-methylerythritol and 15% of 2-methylthreitol). Smaller values could not be used since, for this value of IR, the entire racemic fraction in some samples (for instance 4–7 July 08) has already been

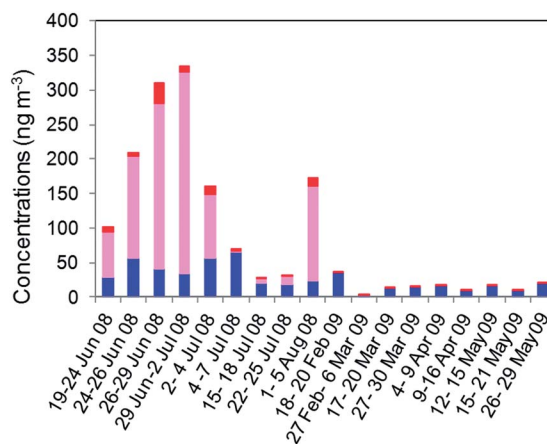


Fig. 4 Upper limit of the secondary fractions (blue) for the 2-methyltetrols in the samples and lower limits for their primary fraction, comprising of the racemic primary fractions (pink) and of the enantiomeric excesses (red).

accounted for as entirely secondary. The primary and secondary fractions for the 2-methyltetrols thus calculated as described above and assuming $IR = 0.19$ are shown in Fig. 4. It can be seen in Fig. 4 that, while the secondary fractions accounted for most of the 2-methyltetrols mass in some samples (mostly in the wet season), the primary fraction was largely dominating in the dry season samples, containing most of the 2-methyltetrol mass. Thus, over all the samples, the maximum possible secondary fraction of 2-methyltetrols represented only 31% of their total mass, while a minimum of 69% was estimated to be biological, thus primary.

Fig. 4 also presents the distribution of the primary fraction between racemic fractions (in pink) and enantiomeric excesses (in red) and clearly shows that the racemic fractions dominate, representing 92% of the primary mass in total. This demonstrates that biological sources can produce large amounts of racemic compounds. The large primary fractions of 2-methyltetrols in the dry season are consistent with the correlations in Fig. 2, which reinforces the confidence in these calculations.

But the secondary fractions of 2-methyltetrols calculated above being upper limits, it cannot be excluded that they are, in reality, even lower and the primary fractions even larger. In particular, the large variability in the IR values shown in Table 2 suggests compounds that were almost entirely primary as it is in strong contrast with the constant IR value inferring photochemical (secondary) sources.^{29,54} But using the IR values reported by these other works, 0.35–0.40, to estimate the contribution of secondary reactions would lead to even smaller estimates for the secondary fractions of 2-methyltetrols in the samples of this work. This large fraction of primary compounds was probably to be expected in PM_{10} aerosol fractions, as they are known to be mostly influenced by primary sources. It would now be interesting to perform the same analysis on fine aerosols at the same site.

Correlations with isoprene emissions

The emission estimates of isoprene obtained from the MEGAN model for the sampling period are presented in Table 3.

Table 3 MEGAN daytime averaged isoprene emissions

Date	Isoprene ($\mu\text{g m}^{-2} \text{h}^{-1}$)
19–24 June 2008	1228
24–26 June 2008	1200
26–29 June 2008	1194
29 June–2 July 2008	1409
2–4 July 2008	1378
4–7 July 2008	1170
15–18 July 2008	1541
22–25 July 2008	1769
1–5 Aug 2008	3933
18–20 Feb 2009	2083
27 Feb–6 Mar 2009	1928
17–20 Mar 2009	2183
27–30 Mar 2009	2396
4–9 April 2009	3134
9–16 April 2009	2360
12–15 May 2009	1554
15–21 May 2009	2134
26–29 May 2009	1754

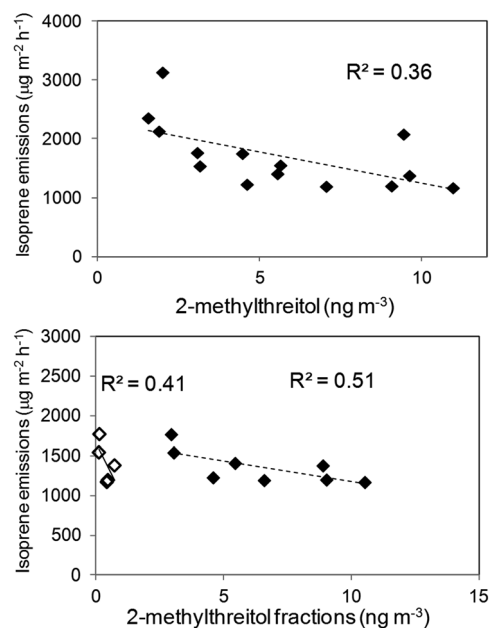


Fig. 5 Correlations between isoprene emissions and total 2-methylthreitol (top) and its racemic (black symbols) and enantiomeric fractions (white symbols) in the dry season (bottom).

Potential correlations between these emissions and the 2-methyltetrol concentrations were investigated. No correlation was found between the isoprene emissions and 2-methylerythritols or total 2-methyltetrols. But a weak anti-correlation ($R^2 = 0.36$) was observed with 2-methylthreitol, which was even more pronounced ($R^2 = 0.51$) for its racemic fraction in the dry season (Fig. 5).

The racemic fraction of 2-methylthreitol was shown above (Fig. 3) to be likely of secondary origin. In that case, the anti-correlations would suggest a kinetic competition between the secondary formation pathways for the 2-methyltetrols and other atmospheric oxidation pathways for isoprene. For instance if the 2-methyltetrols are produced by a reaction involving more than one stoichiometric equivalent of isoprene and in competition with the other oxidation pathways of isoprene, their formation would be favoured by large isoprene concentrations while at low isoprene concentrations the competing reactions will be favoured and less 2-methyltetrols would be produced. Alternatively, if some of the racemic 2-methylthreitol is biological, the anti-correlations would indicate a competition between its biological production and the one of isoprene, similar to the de-coupling recently reported for the primary emission of isoprene and of its oxidation products.⁵³

Origin of the air masses

The analysis of the trajectories showed that all the air masses collected on filters originated from a sector northeast to southeast of the measurement site. No other region seemed to have influenced the collected samples. No specific pattern or correlation could be found between the regions of origin and the different fractions of the samples, in particular their non-racemic fractions. This suggested a uniform distribution of

both the secondary and the primary sources of 2-methyltetrols in the region studied.

Conclusions

The enantiomeric speciation of the 2-methyltetrols in PM₁₀ samples from the Amazonian forest in Brazil evidenced a majority of samples containing non-racemic compounds, both in the dry and wet season, identifying with certainty the presence of compounds of biological origin. These non-racemic compounds accounted for a significant fraction of the 2-methyltetrol mass in some samples but only for about 5% of their total mass for all the samples. By complementing the chiral analysis with information on the absolute concentrations and isomeric ratios the analysis could be extended to the racemic fractions. First, correlations between the racemic and non-racemic fractions in the dry season suggested a large primary component in the racemic fractions in that season. By contrast, a correlation between the racemic fractions of both compounds in the wet season suggested mostly a secondary origin. Using the isomeric ratios observed in the samples as an estimate for the secondary reactions, an upper limit for the secondary fraction of these compounds in the samples could be calculated and was found to represent only 31% of their total mass, while a minimum of 69% of this mass would be primary. The results also show that the large majority of this primary fraction (92%) was racemic, thus demonstrating that biological sources can have a major contribution to racemic compounds. Comparisons with estimated isoprene emissions at the site did not provide more information on the primary or secondary fractions but indicated a potential competition between the secondary formation pathways for 2-methyltetrols and other consumption pathways for isoprene. The back trajectory analysis could not identify any specific source for the primary or secondary material, suggesting that these sources were uniformly distributed over the region studied.

The large biological fraction of the 2-methyltetrols estimated in these samples might have been expected for PM₁₀ aerosols and would need now to be investigated in fine aerosols at the same site. Note, however, that non-racemic 2-methyltetrols were reported for PM_{2.5} aerosols previously.⁸ This work clearly demonstrates the ability of chiral and isomeric analysis to provide essential information on the primary and secondary origin of organic compounds in ambient aerosols.

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