TRANSPHORM

Transport related Air Pollution and Health impacts –

Integrated Methodologies for Assessing Particulate Matter

Collaborative Project, Large-scale Integrating Project

SEVENTH FRAMEWORK PROGRAMME

ENV.2009.1.2.2.1 Transport related air pollution and health impacts

Deliverable D2.1.1, type R

PM sampling and analysis in ESCAPE/TRANSPHORM cities

Due date of deliverable: project month 24

Actual submission date: project month 24

Start date of project: 1 January 2010 Duration: 48 months

Organisation name of lead contractor for this deliverable: TNO

Scientist responsible for this deliverable: Aleksandra Jedynska

Revision: [0]

Contents

PM sampling and analysis in ESCAPE/TRANSPHORM cities

Aleksandra Jedynska, Menno Keuken
TNO, Netherlands Research Organisation, P.O. Box 80015, 3508 TA Utrecht

1. Introduction

This document presents the details on the sampling and analysis of PM, which were the main activities of Task 2.1.1 in WP2.1. The results of the analysis is provided in Deliverable 2.1.4. The activities in Task 2.1.1. are elaborated in the description of work (DOW) for the project. WP2.1 activities are directed to measurements and monitoring data of PM while Task 2.1.1 is directed to collect and analyze $PM_{2.5}$ and PM_{10} samples in ESCAPE/TRANSPHORM cities. The 7th European framework project ESCAPE ("*European Study of Cohorts for Air Pollution Effects")* investigates long-term effects on human health of exposure to air pollution in Europe. As part of this research PM2.5 and PM10 samples have been collected. In collaboration with TRANSPHORM, the chemical analysis of these samples has been extended. In this document the sampling program is detailed (Eeftens et al., 2012). The results of the sampling program are presented in a database on the website of TRANSPHORM (http://www.transphorm.eu/).

2. Measurement locations

In ESCAPE (http://www.escapeproject.eu/), $PM_{2.5}$ and PM_{10} samples have been collected in 20 study areas with information on human health in fifteen countries in total. In Fig. 1 these countries and study areas are presented. In most countries, 20 sampling sites per study areas have been selected with the exception of the Netherlands/Belgium and Spain with 40 sampling sites. Descriptions of the 20 study areas and number of sites per study area are presented in the Table 1. In each study area, three types of sampling sites were selected: regional background, urban background and street location. Street locations were defined as locations at a major road with more than 10,000 vehicles per day. Urban background locations were sites with less than 3000 vehicles per day within a radius of 50 m. Regional background locations were mostly located in small villages. The partners

in the fifteen countries used identical sampling protocols and common criteria for the selection of sampling sites.

Figure 1: The 20 study areas in the fifteen participating countries in the ESCAPE project.

In all 20 study areas in the fifteen countries, the following components have been analyzed: NO, NO_2, NO_x, $PM_{2.5}$, PM_{10}, $PM_{2.5}$-absorbance, PM_{10}-absorbance and elemental composition by XRF in $PM_{2.5}$ and PM_{10}. In addition, in 10 study areas $PM_{2.5}$ samples have been analyzed for polycyclic aromatic hydrocarbons (PAH), EC/OC and oxidative potential. In four study areas (the Netherlands/Belgium, Munich, Catalonia and Oslo) the analysis of $PM_{2.5}$ samples was further extended with analysis of hopanes and

steranes, levoglucosan and an additional oxidative potential analysis by the DDT test (see: Section 5 Analytical methods).

Table1. Description of the 20 ESCAPE/TRASPHORM study areas and chemical analysis.

Country	Study area: 1-20	Sampling period	Analyses	Sites	Sites types		
					RB	UB	S
Norway	Oslo (1)	05-02-2009–29-01-2010	XRF_PM10, PAH, EC/OC, DTT, H&S	19	3	9	8
Sweden	Stockholm (2)	03-12-2008–01-12-2009	XRF_PM10	20	3	6	11
Finland	Helsinki (3)	27-01-2010–26-01-2011	XRF_PM10, PAH, EC/OC, DTT	20	2	10	8
Denmark	Copenhagen (4)	19-11-2009–17-11-2010	XRF_PM10, PAH, EC/OC, DTT	20	3	6	11
Lithuania	Kaunas (5)	20-01-2010–19-01-2011	XRF_PM10	20	4	6	10
United Kingdom	Manchester (6)	27-01-2009–20-01-2010	XRF_PM10	20	0	8	12
	London (7)	26-01-2010–18-01-2011	XRF_PM10, PAH, EC/OC, DTT	20	1	12	7
The Netherlands/ Belgium	Rotterdam, Amsterdam (8) Groningen, Amersfoort	17-02-2009–19-02-2010	XRF_PM10, PAH, EC/OC, DTT, H&S	16	4	4	8
	Utrecht, Groningen Maastricht, Doetinchem, Antwerp		XRF_PM10	40	10	12	18
Germany	Ruhr area (9)	15-10-2008–12-10-2009	XRF_PM10	20	4	8	8
	Munich/Augsburg (10)	27-10-2008–05-11-2009	XRF_PM10, PAH, EC/OC, DTT	20	5	6	9
Austria	Vorarlberg Cities (11)	27-10-2008–05-11-2009	XRF_PM10	20	3	7	10
France	Paris (12)	04-01-2010–04-01-2011	XRF_PM10, PAH, EC/OC, DTT, H&S	20	4	9	7
Hungary	Gyor (13)	22-02-2010–24-02-2011	XRF_PM10	20	1	9	10
Switzerland	Lugano (14)	02-03-2009–10-03-2010	XRF_PM10	19	3	6	10
Italy	Turin (15)	01-02-2010–25-01-2011	XRF_PM10	20	1	8	11
	Rome (16)	27-01-2010–26-01-2011	XRF_PM10, PAH, EC/OC, DTT, H&S	20	2	8	10
Spain	Catalunya (Barcelona, Girona, Sabadell) (17)	14-01-2009–14-01-2010	XRF_PM10, PAH, EC/OC, DTT	40	4	13	23
	Barcelona City (18)	14-01-2009–14-01-2010	XRF_PM10, PAH, EC/OC, DTT, H&S	20	1	8	11
Greece	Athens (19)	21-04-2010–27-04-2011	XRF_PM10, PAH, EC/OC, DTT	20	1	12	7
	Heraklion (20)	18-02-2009–16-02-2010	XRF_PM10	20	0	12	8

3. Sampling strategy

At each sampling site of the 20 study areas, three two-weekly samples were collected during one year and in different seasons, e.g. the first two weeks of March (winter), the first two weeks of August (summer), and the first two weeks of June or December (spring or Autumn). At each sampling site, two parallel samples were collected with size-

selective inlets on Teflon filters to determine the mass of $PM_{2.5}$, PM_{10} and elemental composition of these PM fractions. In addition, in ten study areas two extra $PM_{2.5}$ samples were collected: one on glass Teflon coated filter (T60A20) and one on quartz filter. The former for analysis of organic components (PAH, hopanes/steranes) and the latter for analysis of EC/OC, oxidative potential (DTT assay) and levoglucosan.

4. Adjustment for seasonal variability

The three sampling periods of two weeks during a year were used to estimate the annual average of a pollutant. Annual averagess are suitable for assessing the health impacts related to long-term exposure to air pollution. For practical reasons, it was not possible to collect samples simultaneously at all sites at each of the 20 study areas. Hence, due to temporal variation in air quality, the estimated annual average from different sampling periods at the 20 sampling sites would result in different annual averages, as a result of the sampling strategy. In order to correct for the temporal variation, a reference site was introduced at each of the 20 study areas.

At this reference site, two-weekly samples were collected over the year and analyzed for similar components as at the other 20 sampling sites. The results for each pollutant at each of the 20 sampling sites in a specific sampling period were compared with the same period at the reference site. Subsequently, the annual average at a specific site was corrected for the temporal variation as measured at the reference site. In this way, the annual averages at all of the 20 sampling sites have been obtained in a comparable way.

The organic components (e.g. PAH, and hopanes and steranes), EC/OC and oxidative potential by DTT method were not analyzed at the reference sites. To adjust the results of these pollutants for temporal variation, the following procedure was used. Firstly, the temporal correlation was calculated at a specific site between organic components and the "standard" pollutants: $PM_{2.5}$, $PM_{2.5}$-absorbance and NO_x. Secondly, the standard component with the highest median correlation with the organic components was used to correct the annual average of the organic components, EC/OC and oxidative potential. Thirdly, this was performed for each of the three sampling periods at a specific site and finally, the average of these three periods was used to calculate the annual average.

5. Analytical methods

The "standard" components: NO, NO_2, NO_x, $PM_{2.5}$, PM_{10}, $PM_{2.5}$-absorbance and PM_{10}-absorbance were analyzed at IRAS (University of Utrecht, the Netherlands) [SOP-NO2; SOP-PM sampling; SOP-weighing; SOP-Absorbance]. The analysis of the elemental composition of $PM_{2.5}$ and PM_{10} by XRF were conducted at Cooper Environmental Services laboratory in the USA [SOP-XRF]. The series of more than 2400 PM samples generated in the campaigns was too large for the laboratories at the Universities or Institutes involved in ESCAPE or TRANSPHORM to analyze. No commercial laboratory had been identified in Europe to perform the XRF at competing costs and hence, the choice of Coopers in the USA. All organic components, EC/OC and oxidative potential analysis were measured in the laboratory of TNO Applied Environmental Chemistry in the Netherlands.

5.1 Quality of the analytical methods.

All methods used at TNO are validated according to national norm NEN-7777 Environment - Performance characteristics of measurement methods. Reproducibly of the methods are based on 8 measurements conducted on different days. Total uncertainty is twice uncertainty calculated based on reproducibility, recovery of the method and uncertainty of the calibration standard.

5.2 EC/OC with thermal-optical analysis

From each quartz filter, a $1cm^2$ patch is sampled for elemental (EC) and organic (OC) carbon analyses. The analysis of EC/OC is based on the thermal optical method as described in the American Standard Method NIOSH 5040. The EUSAAR 2 protocol was used for the temperature settings (Cavalli et al. 2010, TNO-AEC: ORG-225). OC is removed from the filter in the temperature range of 200-650 °C in a non-oxidising carrier gas (Helium). EC is then removed in the temperature range of 500-850 °C in? a mixture of helium and oxygen. The resulting CO_2 is then converted to methane and detected by flame ionisation detection (FID). Correction for pyrolysis of OC to EC is carried out by measurement of light transmission. Sucrose is used as a control standard based on a gas mixture of 5% methane in helium.

5.3 Levoglucosan

About 2.5 cm^2 of each quartz filter is sampled for measurements of levoglucosan with gas chromatography in combination with mass spectrometric detection in electron impact mode (GC/MS EI). The analyses are conducted on an Agilent 6890/5973N GC/MS. The reproducibility of the method lies around 30%. Levoglucosan quantification is based on component identification by retention time, specific ion ratios and an internal standard (TNO-AEC: ORG-223).

5.4 Oxidative potential

After EC/OC and levoglucosan analysis, the remaining quartz filter was used for oxidative potential measurements following two procedures: "DTT" and "CRAT-ROS" (Chemiluminescent reductive acridinium triggering):

- DDT; the principle of the DTT assay is catalysis of the sample by redox-active compounds reduction of oxygen to superoxide by DTT, which is oxidized to its disulfide. The remaining thiol is reacted with DTNB, generating the mixed disulfide and 5-mercapto-2-nitrobenzoic acid which is determined by its absorption at 412nm (Cho et al., 2005).
- CRAT-ROS; the chemiluminescence reaction of acridinium ester under basic conditions forms the basis of the CRAT-ROS assay to measure production of reactive oxygen species from the interaction of reducer and oxidant. The reducer used in the assay (DTT) delivers the electron to particulate matter. The produced hydrogen peroxide reacts with acridinium ester after addition of the buffer (pH 9.2). The light emitted during this reaction is measured for about 1 second with a luminescence meter (Zomer et al., 2011).

Both assays are sensitive to organic components. Additionally, the CRAT-ROS assay is responsive to metals e.g. Fe, Cu. (Yang et al., to be submitted).

Firstly, the filter is extracted in ethanol for 1.5 hour and subsequently, dried. Next, 100µl ethanol and 900µl MiliQ water is added. The extracts were used for both oxidative potential assays:

- *DTT;* 100µl of sample with known concentrations was incubated with 200 µl of 0.5mM DTT in 0.1M potassium phosphate buffer (pH 7.4) at 37°C . The incubation time for the assays are: 0, 10, 20, 30, 40, 50 minutes. After the incubation time, 300 µl of 10% trichloroacetic acid is added. Next, 1 ml of 0.4M Tris–HCl, pH 8.9 with 20 mM EDTA is mixed with 0,5 ml of incubation mixture. Finally 30 µl of 10 mM DTNB is added. Absorption of the formed 5-mercapto-2-nitrobenzoic acid was measured at 412 nm. A soot sample is used as quality control.
- *CRAT-ROS*; The antioxidant depletion assay is performed as follows. On a 96 well micro plate, where 50 µl of sample is incubated with 50 µl antioxidant (35 µM DTT) for 10 minutes at 438 rpm in the plate reader. On the same plate calibration measurements are performed: 50 µl of MilliQ water is incubated with 50 µl of DTT solution with the concentrations ranging from 0 - 35µM. After incubation time, 50 µl of TAOC reagent is added, composed of PPHO and acridan ester in PBS with 0.01% Tween20. Immediately after the addition of 50 µl trigger solution (0.003% H_2O_2, 10 ng/ml HRP in PBS, 0.01% Tween20), the luminescence is measured every minute for 90 minutes. The results are calculated by specially developed software on the basis of calibration curves. The standard 1,2-naphthoquinone is used as a quality control.

5.5 PAHs, Nitro-,Oxy-PAHs, hopanes and steranes

To measure PAHs, nitro- and oxy-PAHs, and hopanes and steranes, the T60A20 filters were extracted using the accelerated solvent extraction method (ASE) with toluene. Further, the toluene extracts were fractioned into three fractions using a silica column. This method separates the hopanes/steranes from the PAHs and PAHs' derivates.

5.5.1 Analytical procedure PAHs

The 16 EPA (US Environmental Protection Agency) priority pollutant PAHs (as defined in EPA Method TO-13A), were analysed by means of gas chromatography in combination with mass spectrometric detection in electron impact mode (GC/MS EI).

Prior to extraction the following internal standards were applied to the sample: a mixture of the 16 deuterated EPA-PAH components, d50 -n-tetracosane a mixture of the deuterated oxy/nitro-PAH components: d6-1,4 naphthoquinone, d8-9-fluorenone, d8-9,10-anthaquinon, d9-2-nitrofluoreen, d9-9-nitroanthracene, d9-3-nitrofluoranthene, d8-1-nitropyrene and d11-6-nitro chrysene. The identification of all components was based on retention time and qualifier ion ratios. Components' quantification was conducted based on the highest characteristic qualifier ion using the internal standard technique (TNO-AEC: ORG-217). The analyses were conducted on an Agilent 6890/5973N GC/MS. The total uncertainty of the method varies per component from 11 to 35%. The following eight non-volatile PAH components were quantified: benzo[*a*]anthracene, chrysene, benzo[*b*]fluoranthene, benzo[*k*]fluoranthene benzo[*a*]pyrene, indeno[*1,2,3-CD*]pyrene, dibenzo[*a,h*]anthracene and benzo[*g,h,i*]perylene.

5.5.2 Analytical procedure PAH's derivates

The PAH's derivates were analysed using a gas chromatography method with mass spectrometric detection combined with negative ion chemical ionisation mode (GC/MS NICI) (TNO-060-UT-2011-00953). The components are measured with a GC/MS Shimadzu QP2010. The expanded uncertainty of the method was determined at the concentration level of 50 ng/ml and varies between 38 and 77% for different components. The following PAH's derivates are analyzed : 1,4-naphthoquinone, 1-naphthaleen carboxaldehyde, 9-fluorenone, 9,10-anthraquinone, benzanthrone, 1-pyreencarboxaldehyde, benz[a]anthracene-7,12-quinone, 1-nitronaphthalene, 2-nitronaphthalene,4-nitrobiphenyl, 2-nitrofluorene, 3-nitrofluorantene, 1-nitropyrene en 6-nitrochrysene.

5.5.3 Analytical procedure Hopanes and Steranes

The hopanes and steranes (TNO-AEC: ORG-224) were analysed by gas chromatography in combination with mass spectrometric detection in electron impact mode (GC/MS EI). The analyses were conducted on an Agilent 6890/5973N GC/MS. The reproducibility of the method is determined with three concentration levels between 20 and 800 ng/ml. The

expanded uncertainty of the method varies between 26% and 63% for the components 17α (H),21α(H)-30-Norhopane/ 17α(H),21α(H)-30-norhopane (H29aa/ba) and 22R-17α(H),21β(H)-homohopane (H31R).

The following hopanes and steranes were analysed: 18α(H)-21β(H)22,29,30-Trisnorneohopane (Ts), 17α(H),21β(H)22,29,30-Trisnorhopane (Tm), 17α(H),21β(H)-30-Norhopane (H29 αβ), 17α(H),21α(H)-30-Norhopane (H29 αα), 17β(H),21α(H)-30-Norhopane (H29 βα), 17α(H),21β(H)-Hopane (H30 αβ), 17β(H),21α(H)-Hopane (H30 βα), 17β(H),21β(H)-Hopane (H30 ββ), 22S-17α(H),21β(H)-Homohopane (H31 S), 22R-17α(H),21β(H)-Homohopane(H31 R), 20R-5α(H),14β(H),17β(H)-Cholestane (C27 αββ R), 20R-5α(H),14α(H),17α(H)-Cholestane (C28 αββ R), 20R-5α(H),14β(H),17β(H)-24S-methycholestane (C27 ααα R), 20R-5α(H),14β(H),17β(H)-24R-Ethylcholestane(C29 αββ R), 20S-5α(H),14β(H),17β(H)-24RS-24Ethylcholestane(C29 αββ S). The repeatability of the method varies between 5 and 15%. The reproducibility of the method amounts to 22% and the total uncertainty of the method amounts to 33%.

6. References

1. Eeftens et al., 2012. Spatial variation of $PM_{2.5}$, PM_{10}, $PM_{2.5}$-absorbance and PMcoarse concentrations between and within 20 European study areas and the relationship with NO2: Results of the ESCAPE project. Atmospheric Environment 62, 303-317

2. SOP-NO_2 (www.transphorm.eu). Description passive sampling and analysis of NO_2.

3. SOP-PM sampling(www.transphorm.eu). Description of sampling $PM_{2.5}$ and PM_{10}.

4. SOP-weighing(www.transphorm.eu). Description weighing procedures $PM_{2.5}$ and $PM_{10.}$

5. SOP-Absorbance(www.transphorm.eu). Description Black Smoke analysis.

6. SOP-XRF(www.transphorm.eu). Description XRF analysis of PM2.5 and PM10 samples.

7. Method 5040 - Elemental Carbon, Diesel Particulate, National Institute for Occupational Safety and Health. Issue 3 (Interim); March 2003.

8. Cavalli F., Viana M., Yttri K.E., Genberg J., Putaud J.-P. Toward a standardised thermal-optical protocol for measuring atmospheric organic and elemental carbon: the EUSAAR protocol. Atmos. Meas. Tech., 3, 79-89, 2010

9. TNO-AEC: ORG-225-v4 EC/OC_Determination of organic and elemental carbon with Sunset Laboratory. TNO, Utrecht, The Netherlands.

10. TNO-AEC: ORG-223-v1.0 LEVO_Determination of levoglucosan by using isotope dilution and GCMS. TNO, Utrecht, The Netherlands.

11. Cho, A. K.; Sioutas, C.; Miguel, A. H.; Kumagai, Y.; Schmitz, D. A.; Singh, M.; Eiguren-Fernandez, A.; Froines, J. R. Redox activity of airborne particulate matter at different sites in the Los Angeles Basin. Environmental Research 2005, 99, 40–47.

12. Zomer, B.; Collé, L.; Jedyńska, A.; Pasterkamp, G.; Kooter, I.; Bloemen, H. Chemiluminescent reductive acridinium triggering (CRAT)—mechanism and applications. Analytical and Bioanalytical Chemistry 2011, 401, 2945–2954.

13. Yang A., Jedynska A., Hellack B, Kooter I.M., Hoek G., Brunekreef B., Kuhlbusch T., Cassee F.R., Janssen N.A.H. Comparison of different methods for measuring oxidative potential of PM2.5. To be submitted.

14. EPA Method TO-13A. Determination of Polycyclic Aromatic Hydrocarbons (PAHs) in Ambient Air Using Gas Chromatography/Mass Spectrometry (GC/MS)

15. TNO-AEC: ORG-217 v2.1 PAK-MS_Determination of polycyclic aromatic hydrocarbons (PAH) with i dilution and GCMS analysis. TNO, Utrecht, The Netherlands.

16. TNO-060-UT-2011-00953. Determination of nitro-and oxy-PAH"s with isotopic dilution and GC / MS analysis. TNO, Utrecht, The Netherlands.

17. TNO-AEC: ORG-224 v1, Determination of hopanes and steranes with GCMS analyses. TNO, Utrecht, The Netherlands.