Comparison of different extraction techniques to analyze polycyclic aromatic hydrocarbons in tire particle with different sizes combined with bioassay characterization

Ivan A. Titaley[1], Anna Larsson[2], Nikita Burton[2], Felicia Johansson[2], Anna Karlsson[2], Katarina Kristoffersson[2], Ida Markström[2], Ulrika Eriksson[1], Maria Larsson[1]

[1]Man-Technology-Environment (MTM) Research Centre, School of Science and Technology, Örebro University, Örebro 701 82, Sweden
[2]School of Science and Technology, Örebro University, Örebro 701 82, Sweden

Introduction

Artificial turf contains granulates made of recycled car tires, which contain polycyclic aromatic hydrocarbons (PAHs). Current characterization of PAHs in car tires are limited to a list of 8 PAHs as defined by REACH (PAH_8): Chrysene, Benzo[*a*]anthracene, Benzo[*b*]fluoranthene, Benzo[*k*]fluoranthene, Benzo[j]fluoranthene, Benzo[*e*]pyrene, Benzo[*a*]pyrene, and Dibenzo[*a,h*]anthracene. However, other PAHs and polycyclic aromatic compounds (PACs), such as alkylated PAHs, oxygenated PAHs, and azaarenes are often neglected in the analysis of car tire particles. In addition, there is currently limited knowledge on the bioactivity of PAHs found in tire particles. Currently, there is also a lack of replicable standard extraction method of PAHs from car tire particles.

Study Objectives

In this preliminary study, we investigated three aspects relating to the presence of 38 target PAHs in tire particles. **First**, we investigated the effect of tire particle sizes on the measured PAH concentrations. We tested three different particle sizes: fine, medium, and granular (Figure 1). Fine and medium tire particles originated from the same batch, while the granular sample was from a different batch. **Second**, we compared the efficiency of two extraction methods to analyze PAHs from tire particles. The two methods that we tested were: a modified sonication method (GS2014:01 PAK) and pressurized liquid extraction (Figure 2). **Third**, we explored the bioactivity of PAHs in the tire particles using the H4IIE-luc bioassay for aryl hydrocarbon receptor (AhR). We compared the Bioassay-derived-Toxic Equivalency Quotient (Bio-TEQ) of the whole tire particle extracts, calculated at EC_{25}, with the chemical-TEQ (Chem-TEQ) based on the relative potency (REP) value, also at EC_{25}, for 18 PAHs: 16 U.S. EPA PAHs (PAH_{16}) and two PAHs that are included in PAH_8, but not in PAH_{16}. Bio-TEQ (pg/g) = $\dfrac{\text{TCDD } EC_{25}\left({}^{pg}/_{mL}\right)}{\text{extract } EC_{25TCDD}\left({}^{g}/_{mL}\right)}$

Figure 1. Three different particle sizes tested in this study (from left to right): granular, medium, and fine

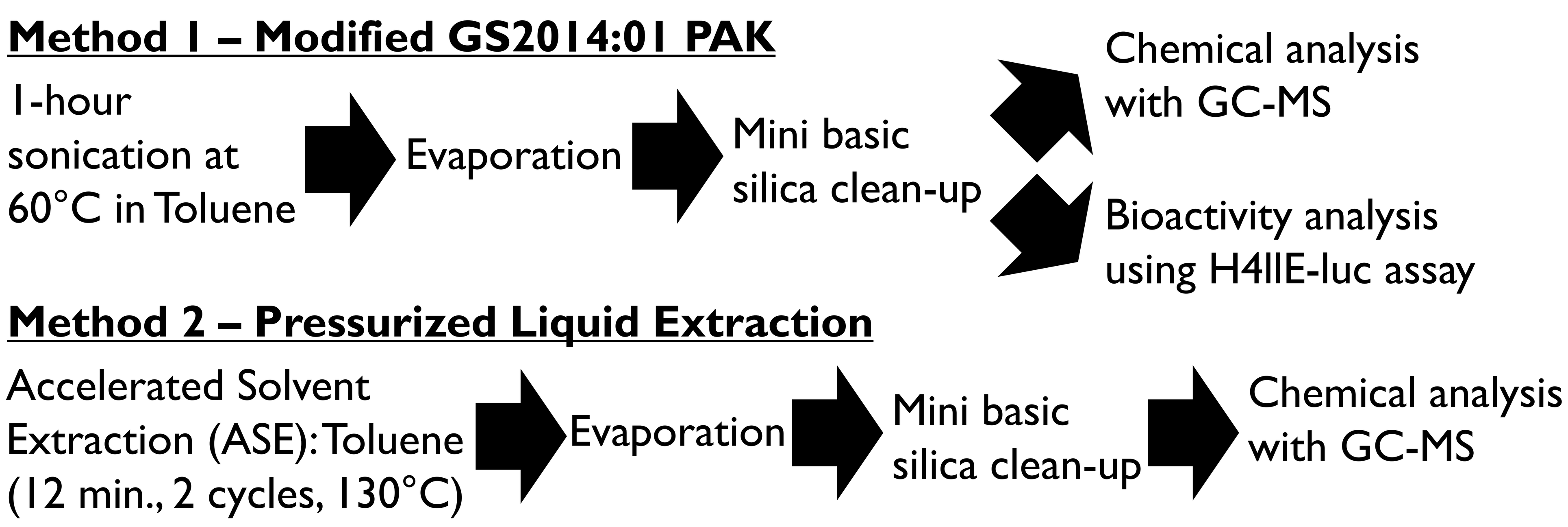

Figure 2. Comparison of the two different extraction methods used in this study

Results and Discussions

- 26 and 28 of the 38 PAHs were measured in extracts from sonication and ASE methods, respectively (Table 1). Coronene was measured in sonication extracts, but not in ASE extracts. We measured higher concentrations of lower molecular weight PAHs in the granular sample than in the fine or medium samples.
- Concentrations of higher molecular weight PAHs were similar between the three different car tire particle sizes. Based on the ASE extracts, we observed higher variations (*e.g.*, larger standard errors) of PAH concentrations in the granular size, suggesting that extractions of tire particle should rely on fine or medium sizes.
- ASE resulted in significantly higher concentrations of PAHs for all samples compared to sonication. The result suggested that the sonication method was not enough to extract all PAHs from the tire particles. ASE offered a more complete extraction step, which may be due to the higher temperature and pressure.
- Chem-TEQ calculation of the sonication extracts only explained some of the measured Bio-TEQ (0,2-0,8%) (Figure 3), which indicated the presence of other bioactive compounds in the samples. The bioactivity may be due to other PAHs of which there are no REP values, and other PACs not analyzed in this study.
- In order to further validate which particle size should be used in future studies, which extraction technique is the most reliable, and which other bioactive compounds are present in the samples, this study should be repeated, but with the aim of analyzing for alkylated PAHs, oxygenated PAHs, and azaarenes.

Table 1. Profile of average concentrations of PAHs (ng/g) found in different car tire particles based on sonication and ASE

Particle size, Extraction	Fine (ng/g)		Medium (ng/g)		Granular (ng/g)	
Compounds	Sonication	ASE	Sonication	ASE	Sonication	ASE
Naphtalene (PAH_{16})	170	7800	170	9200	200	10900
Biphenyl	110	780	50	600	1300	29900
Acenaphthylene (PAH_{16})	310	2900	300	5500	240	6200
Acenaphthene (PAH_{16})	110	2400	80	2100	1200	31900
Fluorene (PAH_{16})	300	3300	210	3300	1100	14600
Phenanthrene (PAH_{16})	2100	26200	1700	22600	5700	66100
Anthracene (PAH_{16})	350	4400	260	3300	660	7400
4H-Cyclopenta[def]phenanthrene	<LOQ	4200	<LOQ	3700	<LOQ	12500
Fluoranthene (PAH_{16})	3100	29100	2800	25900	6700	48000
Pyrene (PAH_{16})	10900	96800	11200	91500	13300	99300
Benzo[*a*]fluorene	30	2100	20	1400	50	2600
Benzo[*c*]fluorene	180	3400	110	1900	350	4100
Benzo[*c*]phenanthrene	30	1300	30	1200	40	1700
Benzo[*ghi*]fluoranthene	560	8200	500	7000	520	7100
Cyclopenta[*cd*]pyrene	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Benzo[*a*]anthracene (PAH_8 and PAH_{16})	200	2200	100	1000	120	1100
Triphenylene	150	3700	160	4600	250	6300
Chrysene (PAH_8 and PAH_{16})	310	2400	230	1900	390	2900
Benzo[*b*]fluoranthene (PAH_8 and PAH_{16})	370	2600	300	1800	420	2400
Benzo[*k+j*]fluoranthene (PAH_8 and PAH_{16})	250	2100	180	1200	160	1400
Benzo[*a*]fluoranthene	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Benzo[e]pyrene (PAH_8)	930	8700	880	7300	1000	7600
Benzo[*a*]pyrene (PAH_8 and PAH_{16})	840	5100	620	4200	650	4200
Perylene	140	1500	110	1200	120	1300
Dibenzo[*a,j*]anthracene	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Dibenzo[*a,c*]anthracene	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Indeno[1,2,3-*cd*]pyrene (PAH_{16})	450	4400	410	3900	490	3300
Dibenzo[*a,h*]anthracene (PAH_8 and PAH_{16})	<LOQ	187	<LOQ	0	<LOQ	152
Benzo[*b*]chrysene	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Picene	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Benzo[*g,h,i*]perylene (PAH_{16})	2600	19500	2900	19400	3300	14800
Anthanthrene	700	6800	600	8800	700	6000
Dibenzo[*b,k*]fluoranthene	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Dibenzo[*a,e*]pyrene	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
Coronene	2200	<LOQ	1900	<LOQ	2200	<LOQ
Dibenzo[*a,l*]pyrene	<LOQ	9100	<LOQ	10100	<LOQ	6500
Naphtho[2,3-*a*]pyrene	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ

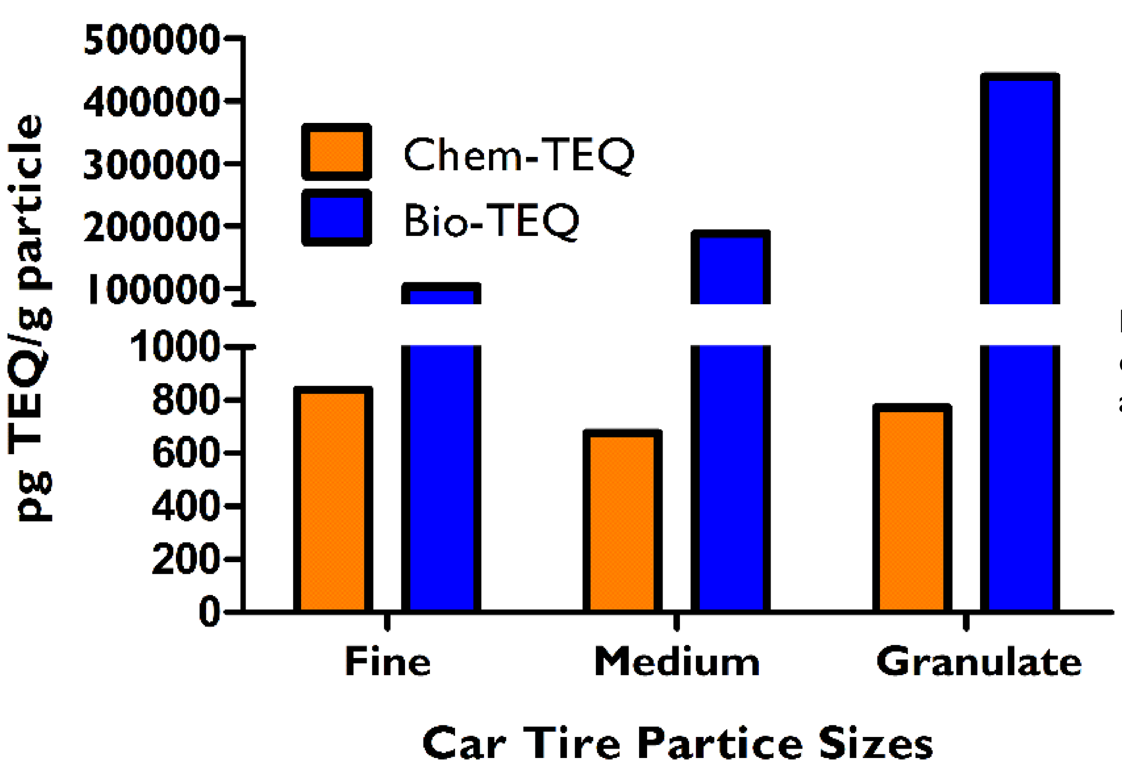

Figure 3. Comparison of Chem- and Bio-TEQ of the extracts of car tire particles. Note the difference in y-axis before and after the break.

Conclusions

1. *Smaller than granular car tire particle size should be used for chemical analysis*
2. *ASE offers a more complete extraction procedure of PAHs than sonication*
3. *Car tire particle extracts contain bioactive compounds, which may suggest potential human and environmental risks*

References

Geiss, O.; Senaldi, C.; Bianchi, I.; Lucene, A.; Tirendi, S.; Barrero-Moreno, J. A fast and selective method for the determination of 8 carcinogenic polycyclic aromatic hydrocarbons in rubber and plastic materials. *J. Chrom. A.* **2018**, *1566*, 13-22.
Larsson, M.; Hagberg, J.; Rotander, A.; van Bavel, B.; Engwall, M. Chemical and bioanalytical characterisation of PAHs in risk assessment of remediated PAH-contaminated soils. *Enviro. Sci. Poll. Res.* **2013**, *12*, 8511-8520.
Perraudin, E.; Budzinski, H.; Villenave, E. Analysis of polycyclic aromatic hydrocarbons adsorbed on particles of atmospheric interest using pressurised fluid extraction. *Anal. Bioanal. Chem.* **2005**, *383*, 122-131.
Sadiktsis, I.; Bergvall, C.; Johansson, C.; Westerholm, R. Automobile tires—A potential source of highly carcinogenic dibenzopyrenes to the environment. *Environ. Sci. Technol.* **2012**, *46*, 3326-3334.

This work was partially funded by the KK Foundation (KK-stiftelsen).